2022

Characterization of a Novel Sensing Mechanism Governing Antigenic Variation in P. Falciparum

Victoria M. Schneider

CHARACTERIZATION OF A NOVEL SENSING MECHANISM GOVERNING ANTIGENIC VARIATION IN *P. FALCIPARUM*

A Thesis Presented to the Faculty of
The Rockefeller University
in Partial Fulfillment of the Requirements for
the degree of Doctor of Philosophy

by
Victoria M. Schneider
June 2022

Characterization of a Novel Sensing Mechanism Governing Antigenic Variation in *P. falciparum*

Victoria M. Schneider, Ph.D.
The Rockefeller University 2022

Plasmodium falciparum expresses a multi-copy gene family called *var* in the intraerythroyctic stages of its life cycle in a mutually exclusive manner. *var* genes encode the chief antigenic and virulence determinant of *P*. *falciparum*, PfEMP1, and switching between active genes results in antigenic variation, allowing the parasite to evade the human immune system and cause chronic infections. The molecular mechanisms that control activation and silencing of individual *var* genes, as well as coordination of the switching process, presently remain incompletely defined. *P. falciparum* contains only ~60 *var* gene family members in its genome. Consequently, the question remains as to how this parasite can maintain an antigen-switch rate that allows for the emergence of a new variant when necessary, without rapidly exhausting all 60 members, to sustain chronic infections. The currently held paradigm proposes that antigenic variation follows an intrinsic, programed switching rate, operating independently of any external stimuli. In the following thesis I will present results suggesting the novel possibility that *P*. *falciparum* possesses cellular machinery capable of sensing changes in the environment of its host and is able to respond by altering antigen expression.

It has been shown that changes in the transcription state of a *var* gene are controlled epigenetically. The methylation state of histone marks, deposited at active and silent *var* genes by histone methyltransferases (HMTs), play prominent roles in *var* gene regulation. Previously, Ukaegbu et al., 2015 showed that manipulating deposition of these marks had a striking impact on *var* gene expression. Metabolism and epigenetic control of gene expression are linked, as HMT activity is dependent on the intracellular concentrations of methyl donors, which can fluctuate based on nutrient availability. Various studies in other organisms have shown that there is a direct link

between the level of intracellular S-adenosylmethionine (SAM), the principle methyl donor in biological methylation modifications, and histone methylation.

I explored this connection between metabolism and *var* gene expression in *P. falciparum.* Parasites were cultured in growth media containing altered concentrations of nutrients involved in SAM metabolism. Bulk RNA was extracted from cultures, used as a template to synthesize cDNA, and analyzed by qPCR to determine the *var* gene expression at the population level. Conditions believed to increase SAM pools induced a coordinated switch to one particular *var* gene, *var2csa*, over time, phenocopying the results from Ukaegbu et al., 2015. This hypothesis was further tested by modifying expression of key enzymes involved in SAM metabolism. Once again, modifications thought to increase the intracellular level of SAM were found to induce a coordinated switch at the population level to *var2csa.* Conversely, modifications that lower the level of SAM did not induce expression of *var2csa,* but instead activated many *var*s at once across the population. These observations directly challenge the stochastic *var* switching paradigm by instead suggesting *P. falciparum* possesses the ability to sense environmental changes.

After recognition of a pathogen, activated macrophages modify their microenvironments in various ways. I next tested the effect of two of these immune responses, depletion of amino acids and release of polyamines, on *var* expression of parasites *in vitro.* Both perturbations altered *var* expression, again specifically inducing *var2csa.*

Taking these results together, I propose and discuss two possible models of antigenic variation in *P. falciparum*. The first centers on intracellular SAM metabolism in describing a promoter competition model governing *var* switching through *var2csa.* The second suggests that *P. falciparum* can sense when the host immune system first begins to recognize it via environmental cues resulting from antibody recognition, and respond by switching *var* gene expression. This would allow parasites to switch expression of *var* genes exactly when needed, allowing the most efficient utilization of their limited *var* gene repertoire.

For my Grandparents,

It has been such a blessing to have you all in my life.

Thank you for all of your unwavering love and encouragement.

Sorry about the tattoos.

Acknowledgements

I have many people to thank for making the past six years the intellectual and emotional journey it has been. First and foremost, I would like to thank my mentor, Kirk Deitsch. Working in your lab has been an absolute pleasure. Thank you for the scientific and professional support, and, above all, your kindness and patience. Thank you for your willingness to allow an outside student into the lab and for dealing with the resulting bureaucracy. Thank you for paying attention to the disappointing results as much as to the exciting ones. You encouraged me to explore my interests and find what inspires me outside of the lab and I cannot express how much that has meant to me. One thing I will take with me beyond graduate school is Kirk's mindset and ability to think about experiments and questions in a larger biological context. This has helped shape the way I think about science, particularly in the context of infectious disease. As a result of my time in the Deitsch Laboratory, I know my abilities as a scientist have greatly improved, and I am forever grateful.

Second, I would like to thank my thesis committee members, Howard Hang, Luciano Marraffini, and Charles Rice. I will always cherish your guidance, insight, and thoughtful feedback. Many thanks specifically to Howard for graciously offering to serve as my co-mentor at Rockefeller, which allowed me to work in Kirk's lab off-campus. Thank you in advance to my external committee member, Manoj Duraisingh, for your time and feedback. Thank you as well to our collaborator Choukri Ben Mamoun, whose research and sharing of materials ultimately inspired and started this project.

I would like to thank the entirety of Whitney floor 7, the Deitsch, Kafsack, Kirkman labs, past and present. Thank you all for making the lab a place I have wanted to come to, day after day, year after year. I have learned something from each of you. Thank you to Bjorn Kafsack for your mentorship and guidance, especially with regards to the metabolism-side of the research in this thesis. Your help with that area of this project was absolutely indispensable. Thank you to Xu Zhang for serving as my first and continued mentor during my rotation and beyond, as well as a cherished friend and shoulder to lean on over the difficulty of working on *var* genes. To Chantal Harris, for graciously sharing your PfSAMS knockdown line and data with me, in addition to your guidance on lab protocols. To Joe Visone, first for your help as a technician when I started and knew nothing about the lab, and now as a fellow student: your help around the lab and experience in setting up drug assays was irreplaceable. Thanks to Francesca, Chantal, and Riward for your help with the Westerns. There are truly so many people in lab I could thank one-by-one for their kindness and feedback, I deeply appreciate each and every one of you as colleagues and as friends.

Very special thanks to our lab technicians, Mackensie, Calvin, and Annie, as well as past technicians, Amanda and Xinran. You all have made the research-side easier by ensuring our lab functions like a well-oiled machine behind the scenes. Calvin and Annie, you both have made coming in on the weekends significantly less painful, which is truly something worth acknowledging. Thank you both for your generous help and friendship.

Mackensie, you deserve your own paragraph! Thank you so much for all of your hard work and help around the lab, as well as your friendship. It has been a pleasure to work alongside you the past few years. Thanks for putting up with me on the good days, as well as the bad. You have grown so much as an independent scientist and I know as you move beyond the lab with your attitude and work ethic, you will no doubt continue to have a positive impact on those around you. Much of the data in this thesis was the direct result of her work.

To my SEPA, NSPN, science policy and diplomacy colleagues, thank you all for your professional and personal friendship. Special thanks to Jesse Ausubel and Mande Holford for your work in this area on campus – your class and presence have been absolutely crucial to my career development.

Lastly on the work-side, but definitely not least, the Rockefeller Dean's office has been incredibly supportive throughout my time here. Thank you all for removing some of the stress from the PhD process. You all were so helpful when I first got here, and somehow even more helpful now as the end approaches. Thank you to the Deans, Emily and Sid, for ultimately signing off on my work in Kirk's lab at Weill Cornell. Thank you to Cris for your time, help, and above all patience, with my many questions and asked favors. All of your time and support mean a lot to me. It was an absolute pleasure and privilege to be a Graduate Fellow at Rockefeller—largely due to all of the admin's tireless commitment to the students.

I would like to give a special shout out to my undergraduate mentors, Graham Hatfull, Deborah Jacobs-Sera, and Carlos Guerrero, as well as the entire Hatfull lab and Phagehunting Program. Not being hyperbolic in the least, the lab environment there and your mentorship changed my life and pushed me to pursue a PhD. Thank you to my friends from the Hatfull lab, who I remain in touch with to this day for your continued guidance. Especially Emilee - you are still such a treasured friend and I truly do not know where I would be without you!

Thank you to my NYC and DC friends, who are largely "not a Science" as we all joke. You all have been the best possible escape and stress relief from the lab. To Emma, TD, Cameron, Lauren, Hannah, Anna, Josh, Wynonna, Andy, Joseph, Paige, Rachel, Laura, Becca, and Francine. Further thanks to all of my "shutopian" friends who are always "in my pocket" and there when I need to just shoot things in a round of Fortnite.

To my family for their presence, love, support, and encouragement over the past 28 years. I really won the lottery in this department - I love you all more than I can say in words. Above all, thank you to my grandparents. This thesis is dedicated to you all, as your life-long hard work has ultimately made it possible for all of your grandchildren to pursue their passions (like doing this PhD). You all have always been a constant source of love and support in my life that I do not take for granted for a second. I thank Pap Pap Schneider for his tenacity, Grandma Schneider for her strength, Pap Pap Kaminski for his sense of humor, and Grammy Kaminski for her "all heart, Bern" (and especially all of the surprise greeting cards – I keep them all).

To my newly minted parents-in-law. Thank you both for welcoming me into your family. Thank you for giving us a home – both your apartment that we now live in, as well as your home to always stay in. I am always happy to watch Antiques Roadshow with you both, even if your daughter won't. Love you both!

To my parents, whom it is hard to thank enough. Thank you for ensuring everything was always so easy. Dad, you are the hardest worker I know – thanks for all your sacrifices and for instilling a similar work ethic in me. Mom, I have always been able to talk to you about literally anything – thanks for always being there to listen and convincing me to keep going when things get hard. Thank you both also for giving me a life-long best friend, my brother Eric. Eric, you are as genuinely kind as you are dependable and talented – thanks for being YOU.

To my fuzzy comrades, Dotty, Josie, and Jay, for the snuggles and company. Dotty, thank you for a lifetime of puppy licks and always remembering me when I come home. Josie, you have been my "PhD cat" and truly there through it all - what an absolute trooper. I never thought I would connect with a cat on a deep spiritual level, but here we are together. Jay, you are truly the strangest creature I have ever met, but I love you all the more for it.

Lastly, and most importantly, to my wife, Elyse. I never in a million years dreamed of finding someone I would consider a soulmate, but our relationship truly feels that way. You have made a lot of sacrifices for me to finish this degree, and for that I'm eternally thankful; for giving up weekends, for living in a studio apartment so my commute to lab was manageable, for your cooking, for the copyediting, for our stupid little inside jokes, for every seemingly boring night watching silly reality TV, and for everything else in-between. Your love and encouragement are largely the reason this thesis exists. Thank you for being my Home. I can't wait for what's next - our "Great, big, beautiful tomorrow!"

Table of Contents

List of Figures

List of Tables

Abbreviations

RBC	Red blood cell
iRBC	Infected red blood cell
PfEMP1	*Plasmodium falciparum* erythrocyte membrane protein 1
HMT	Histone Methyltransferase
Kb	Kilobase
ncRNA	non-coding RNA
uORF	Upstream open reading frame
CTD	C-terminal domain
RNA Pol II	RNA polymerase II
SAM	S-adenosylmethionine
SAH	S-adenosylhomocysteine
LysoPC	Lysophosphatidylcholine
PtdCho	Phosphatidylcholine
PtdEtn	Phosphatidylethanolamine
p-Etn	Phosphoethanolamine
p-Cho	Phosphocholine
PfPMT	*Plasmodium falciparum* phosphoethanolamine N-methyltransferase
MIM	Malaria Incomplete Media
MCM	Malaria Complete Media (RPMI 1640)
qPCR	Quantitative reverse transcriptase polymerase chain reaction
(PfSAMS/PfSAHH) OE	Overexpresser
(PfSAMS) KD	Knockdown
ISR	Integrated stress response
mTOR	mechanistic target of rapamycin
eIF2α	eukaryotic initiation factor 2
eIF2α-p	phosphorylated eukaryotic initiation factor 2
AA	amino acid
Ile	Isoleucine
IDO	Indoleamine 2,3 dioxygenase
Trp	Tryptophan
Kyn	Kynurenine
Arg	Arginine
Put	Putrescine
Spd	Spermidine
Orn	Ornithine
ODC	Ornithine Decarboxylase
SpdSyn	Spermidine Synthase

DCAdoMet	decarboxylated SAM
AdoMetDC	SAM decarboxylase
DHS	deoxyhypusine synthase
DOHH	deoxyhypusine hydroxylase
h-eIF5/eIF5-h	hypusinated eIF5A
NMD	nonsense-mediated decay
dORF	downstream open reading frame
TI	4-Thiaisoleucine
DFMO	DL-α-difluoromethylornithine

Chapter 1
Introduction

A. Introduction to Antigenic Variation in *Plasmodium falciparum*

1.1 *Why study* Plasmodium*? The Global Health Burden of Malaria*

Parasite species of the genus *Plasmodium* are global pathogens capable of infecting vertebrate hosts, and are the causative agent of malaria. Despite remarkable progress in mosquito vector control and other interventions, malaria remains a significant global threat due to mosquitoes' insecticide resistance, parasite drug resistance to antimalarials, and the lack of an effective vaccine. [1] Over half of the world's population is considered at risk for *Plasmodium* infection; in 2018 alone it was responsible for an estimated 228 million cases and 405,000 deaths .[1,2] The vast majority of these cases (93%) and fatalities (94%) occur in Africa with children under five years of age being the most vulnerable, accounting for 67% of deaths worldwide. [1]

Of the over 200 *Plasmodium* species known to infect vertebrates, five cause malaria in humans: *falciparum, vivax, knowlesi, ovale,* and *malariae.* [2,3] Infections by *P. falciparum* and *P. vivax* are the most prevalent, with *falciparum* accounting for the highest number of deaths and thus considered to cause the most severe form of malaria [1].

As of the writing of this document, the world is not on track to reach its targets in reducing malaria morbidity and mortality outlined in both the World Health Organization's Global Technical Strategy for Malaria 2016–2030, as well as the United Nations' Sustainable Development Goals [4]. To successfully battle any infectious disease, it is imperative to first understand the biology of the pathogen. *P. falciparum* has a set of unique features that lend to its remarkable pathogenicity, my contribution to our understanding of which will serve as the subject of this dissertation.

1.2 Plasmodium *Life Cycle and Malaria Disease*

The life cycle of *Plasmodium* is complex, with sexual and asexual reproductive stages in both female *Anopheles* mosquito vectors and vertebrate hosts shown in **Figure 1**.

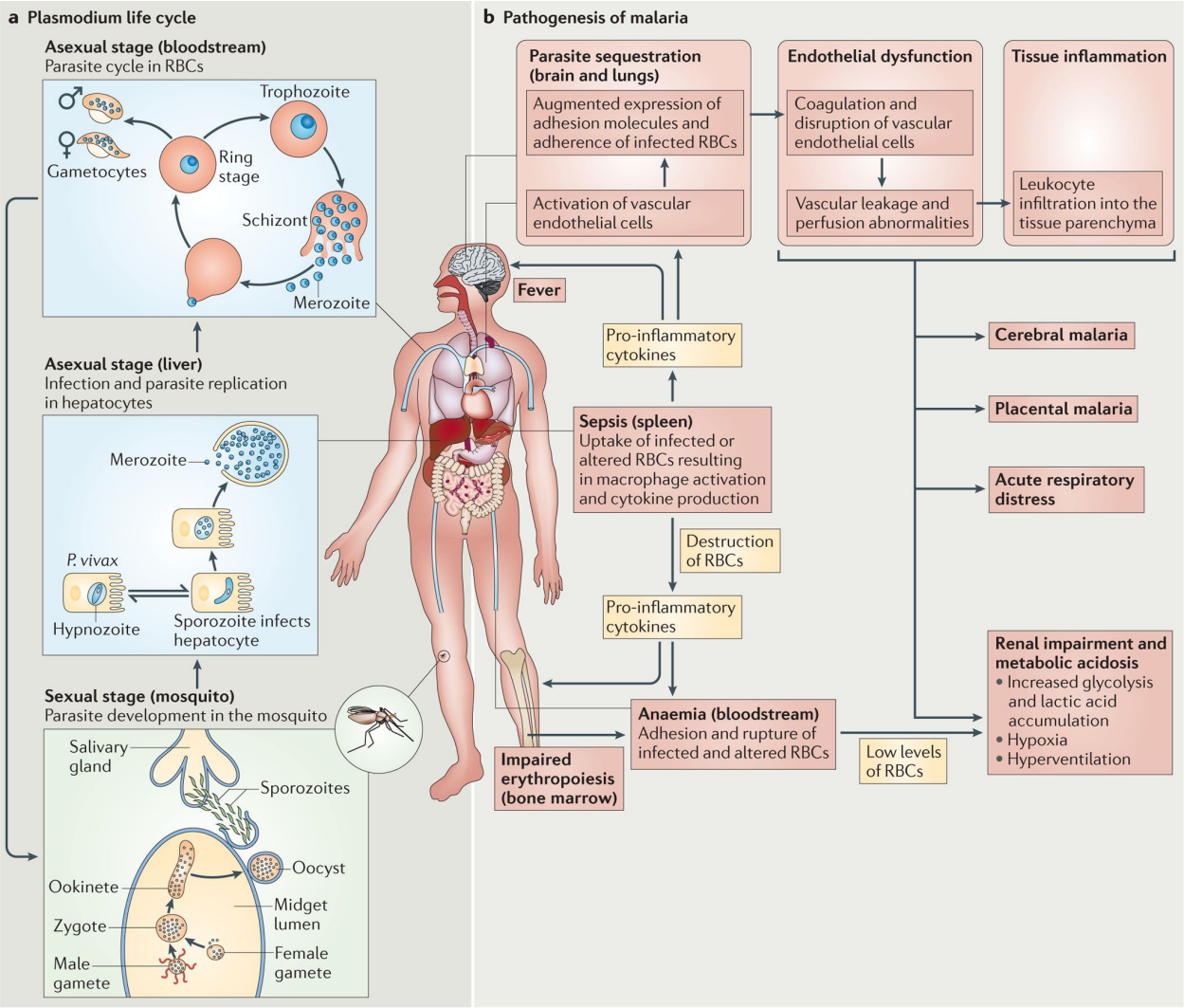


Figure 1. *Plasmodium* life cycle and the pathogenesis of malaria A. After a mosquito bite, sporozoites travel to the liver to infect hepatocytes and develop into merozoites that are released in the bloodstream. Repeated cycles of RBC invasion, replication and merozoite release result in the exponential growth of the parasite population and lead to disease. Infected RBCs circulate containing ring-stage parasites, and a small proportion of merozoites develop into male and female gametocytes that infect mosquitoes, completing the parasite life cycle. **B.** The removal of infected RBCs by splenic macrophages or the uptake of free haemozoin results in the activation of innate immune receptors and cytokine storm. The circulating cytokines cause paroxysms and induce the expression of adhesion molecules by endothelial cells, which mediate parasite sequestration. The sequestration of infected RBCs disrupts blood flow, promotes blood clots, injures endothelial cells and ruptures vascular walls, leading to the extravasation of vascular content and local tissue inflammation. These mechanisms contribute to acute respiratory distress, cerebral malaria or placental malaria. Figure and adapted caption reprinted by permission from Springer Nature: Nature Reviews Immunology, Gazzinelli, R. T., Kalantari, P., Fitzgerald, K. A. & Golenbock, D. T. Innate sensing of malaria parasites. *Nature Reviews Immunology* **14**, 744–757 (2014).[5] Copyright 2014.

The *Plasmodium* life cycle (**Figure 1A**) begins as female *Anopheles* mosquitoes inject sporozoites through its host's skin, from which they disseminate into the bloodstream. They first travel via the circulatory system to the liver to begin asexual replication, expanding to a population of around 30,000 merozoites before invading red blood cell (RBCs). [5] This initial replication within hepatocytes lasts about a week, during which the host experiences no symptoms of infection. Once within RBCs, merozoites mature within a parasitophorous vacuole through three additional, morphologically distinct stages: mononucleated ring stages develop into trophozoites, then schizonts. Schizonts are the product of schizogony, asexually dividing their nucleus and expanding into 16-30 daughter merozoites that are released to find a new RBC to invade and begin the cycle again. [5] [6] In *P. falciparum*, the cycle of asexual replication occurs over 48 hours and can be observed under a microscope on thin blood smears stained for parasite DNA as pictured in **Figure 2**. It is the repeated rounds of asexual replication and infection within RBCs that leads to the manifestation of clinical symptoms associated with malaria disease (**Figure 1B**).

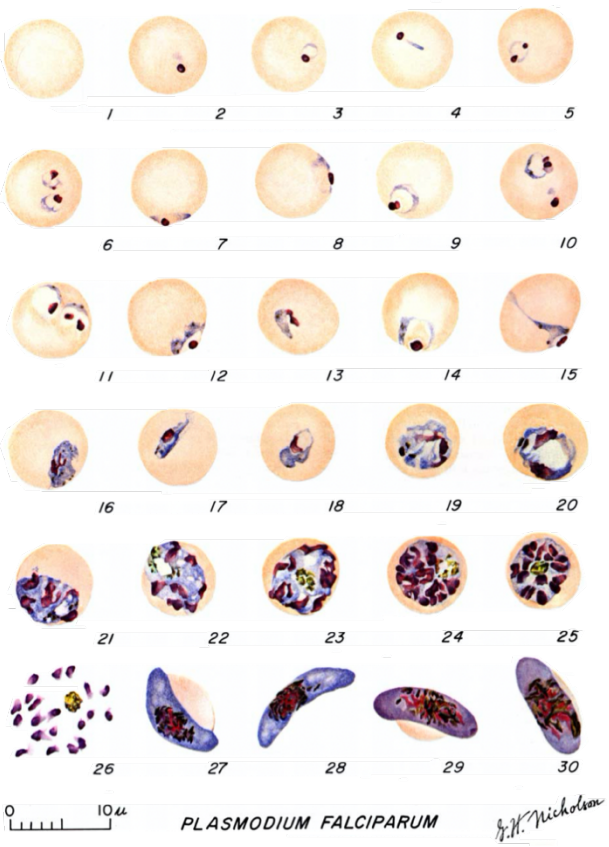


Figure 2. *P. falciparum* blood stage parasites, thin blood smears (1) uninfected RBC; (2-18) Trophozoites (among these, 2-10 correspond to ring-stage trophozoites); (19-26) Schizonts (26 is a ruptured schizont); (27, 28) Mature macrogametocytes (female); (29, 30) Mature microgametocytes (male). Illustration by Gertrude H. Nicholson, published in GR, C., WE, C., M, W. & PG, C. *The Primate Malarias*. (U.S. Department of Health, Education and Welfare, Bethesda, 1971)[150]. Accessed and reprinted with permission from the Centers for Disease Control.

Over the course of the asexual cycle, a small population of parasites will commit to sexual stage development, differentiating into gametocytes that concentrate in skin capillaries (**Figure 1A**). Male and female gametocytes are then taken up by the mosquito vector when they feed upon an infected human. In the mosquito midgut, fertilization and sexual reproduction occurs, producing motile ookinetes that exit the gut and form oocysts. Oocysts undergo rounds of replication, forming sporozoites, which travel from the mosquito abdomen to the salivary glands. There, about 7-10 days after the mosquito picked them up from a blood meal as gametocytes, they can be transmitted again to a human host. [7]

Parasite proliferation alters the infected RBC (iRBC)'s physiology and morphology. Exported proteins bind the cytoskeleton of iRBCs and dramatically decrease their elasticity, changing them from flexible and disk-shaped to rigid and spherical (pictured in **Figure 3**). [8] Upon filtration through the spleen, these abnormal RBCs will be filtered out of circulation and targeted by immune cells for destruction. However, a defining feature contributing to the pathogenicity of *P. falciparum* is its ability to express a series of cytoadhering proteins on the surface of the iRBC. *Plasmodium falciparum* erythrocyte membrane protein 1 (PfEMP1) is expressed in knob-like structures on the RBC surface, facilitating cytoadherence to vascular endothelial receptors, and thereby allowing the parasite to leave peripheral circulation and sequester away in the microvasculature of various organs. This method of cytoadherence and sequestration has evolved to enable *P. falciparum* to escape filtration and mechanical clearance in the spleen, but secondarily contributes to the overall manifestation of malaria symptoms through the binding of endothelial receptors and obstruction of blood vessels, resulting in a local increase in inflammation (**Figure 1B**). [2] For example, binding to endothelial protein C receptor (EPCR), a receptor which functions in anticoagulation and endothelial cytoprotective pathways, has been implicated in cases of severe malaria, most notably in instances of cerebral malaria. [9]

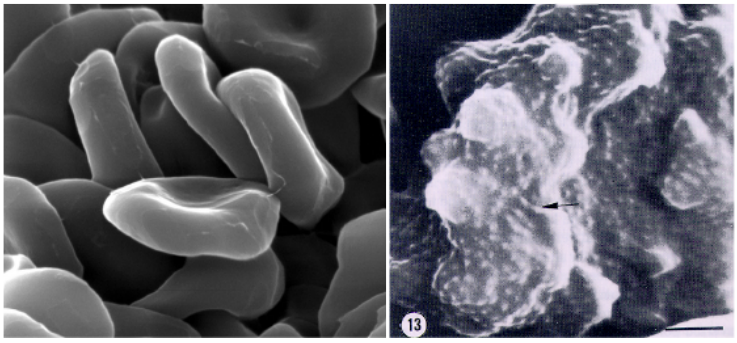


Figure 3. *P. falciparum* alters RBC morphology
Uninfected RBC (left) compared to an iRBC (right). Black arrow highlights knob-like structures on the RBC surface where parasite proteins, like PfEMP1, are displayed. Left picture reprinted by permission from Springer Nature: Nature Nanotechnology, Toumey, C. Compare and contrast as microscopes get up close and personal. *Nat Nanotechnol* **6**, 191–193 (2011). [146] Copyright 2011.

Experimental *P. falciparum* infection studies conducted on prisoners through the early 1960s revealed waves of increasing and decreasing levels of parasitemia in the blood over the course of chronic disease progression, but for many years the precise molecular basis for this phenomenon was unknown. [10] An example of this from a chronically infected child is pictured in **Figure 4**[151].

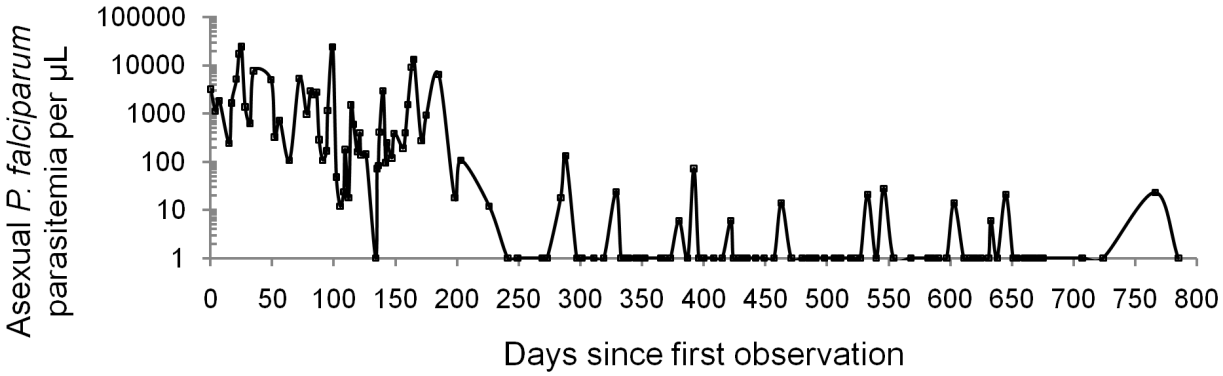


Figure 4. Waves of parasitemia over time in human *P. falciparum* infection Asexual *P. falciparum* parasitemia followed over time in a naturally infected Puerto Rican child. The parasitemia (y-axis) follows a pattern of recurrent peaks that decline in amplitude with time (x-axis). The parasitemia in this child, believed to be a clone, lasted nearly 800 days. Figure and adapted caption from Chen, D. S. *et al.* A Molecular Epidemiological Study of var Gene Diversity to Characterize the Reservoir of Plasmodium falciparum in Humans in Africa. *Plos One* **6**, e16629 (2011)[151]. (Open access).

1.3 *The Genome of* Plasmodium falciparum

The complete haploid genome sequence of *P. falciparum* was published in 2002, facilitating further study into its biology and the unique pathogenesis of the parasite. At 23.3 megabases, it encodes about 5,500 genes spread across 14 chromosomes. [7,11] The full genomic sequence is profoundly AT-rich, roughly 80%. [11]

Prior to the publication of the complete genome sequence, studies began to elucidate clues into the parasite's pathogenesis. Several groups identified that variant forms of PfEMP1 are encoded by a large, multicopy gene family called *var* (for "variant"). [12–14] Using the fully sequenced genome, it was found that each *P. falciparum* genome possesses about 60 *var* genes, primarily in subtelomeric domains, though more recent study of field isolates has shown that number can vary. [15] [11]

var gene expression operates in a mutually exclusive manner, such that in each parasite only one gene expressing PfEMP1 is active over many cycles of asexual replication, while the other 59 remain transcriptionally silent. [16] Each *var* gene has a similar structure **(see Figure 6)**, where the PfEMP1 coding region is comprised of two exons separated by an intron. One promoter sits upstream of the coding region, containing the transcription start site about one kilobase (kb) upstream of the open reading frame. This promoter drives expression of the mRNA and is governed by mutually exclusive expression. [14,17] A second, bi-directional promoter is found within the intron, transcribing sense and antisense noncoding RNAs (ncRNAs). The sense ncRNA is transcribed in both active and silent *var* genes, while the antisense transcript is only found for the single active *var*. [2,18]

The subtelomeric domains of *P. falciparum* also contain several other variant, multi-copy gene families, of whose proteins are similarly displayed on the RBC surface. In addition to *var*, there are *rif, stevor, Pfmc-2TM,* and *surf* clusters whose functions are presently uncharacterized. [19]

1.4 *Antigenic Variation and Immune Evasion by* Plasmodium falciparum

The evolution of mammalian species has resulted in the development of large, multi-cellular organisms that not only themselves replicate, but can also serve as an environment capable of supporting the proliferation of other, primarily single-celled microorganisms. Although mammals have developed a highly sophisticated, multifaceted immune system to identify, control, and ultimately dispose of harmful microorganisms, these pathogens have conversely evolved mechanisms for circumventing immune recognition, thereby establishing an evolutionary “arms race” between host and pathogen. Upon infection of a susceptible host, all pathogens face similar challenges in the colonization of their preferred niche: avoiding both mechanical clearance and recognition by the host’s immune system. Therefore, diverse microbial lineages have evolved similar strategies to survive within their respective host(s). One such strategy - antigenic variation - is the coordinated expression of variant surface-exposed antigenic determinants, which is often achieved by switching from one

member of a multigene family to another. In so doing, pathogens can establish and maintain chronic infection, despite the presence of constant immune pressure exerted by their hosts, by utilizing hypervariable surface molecules that allow them to spread undetected and, once recognized by the immune system, avoid destruction by varying expression of alternative forms of the particular antigen. [20]

In many ways the RBC serves as the perfect host cell for *P. falciparum*: since RBCs are essentially metabolically inactive and lack a nucleus, their membrane is able to hide the parasite from immune recognition, as they are the only cells to not express major histocompatibility complexes (MHCs) on their surface, while the parasite digests hemoglobin and replicates inside. Yet as the parasite's growth distorts the morphology of the RBC, the necessitated expression of PfEMP1 on the RBC surface can be seen by the host's immune system, thus serving as the primary antigenic determinant. [21] Over the course of infection, the host's immune system will mount an antibody response to the predominately-expressed variant of PfEMP1 in the growing parasite population. This response will overwhelmingly reduce the circulating parasite population, often to undetectable levels. Nonetheless, the cycle will repeat, as a sub-population of parasites that were expressing or switched to a novel *var* gene and thus far "unseen" variant of PfEMP1 will clonally expand to high parasitemia until another antibody response is mounted. Therefore, by varying the expression of their displayed antigen, *P. falciparum* can evade the immune response and maintain chronic infections by moving through the *var* repertoire. This phenomena of immune evasion by antigenic variation is believed to be the basis for the waves of parasitemia seen in chronic human infections (**Figure 5**). [2,10]

The tightly-regulated, mutually exclusive expression of *var* genes makes this pathogenic feat possible. Since each parasite only displays one variant of PfEMP1 extracellularly at any given time, it can avoid premature exposure of its full antigenic inventory. [16,22,23] The switch in expression from one *var* gene to the next has evolved to occur at a low rate, allowing for the expansion of clonal *var* populations over many generations (seen as the discrete waves of parasitemia in **Figure 5**). [2,24,25] While all *Plasmodium* species invade and replicate within RBCs, *P. falciparum* is the only

human-infecting parasite to express PfEMP1, allowing them to tightly bind host receptors and efficiently sequester out of circulation[26]. Different variants of PfEMP1 bind to different endothelial receptors. Thus, the organs in which the parasites sequester depends on which *var* gene is expressed. This can lead to additional complications of infection such as cerebral (as with EPCR, mentioned earlier) or placental malaria (**Figure 1B**). [10,28]

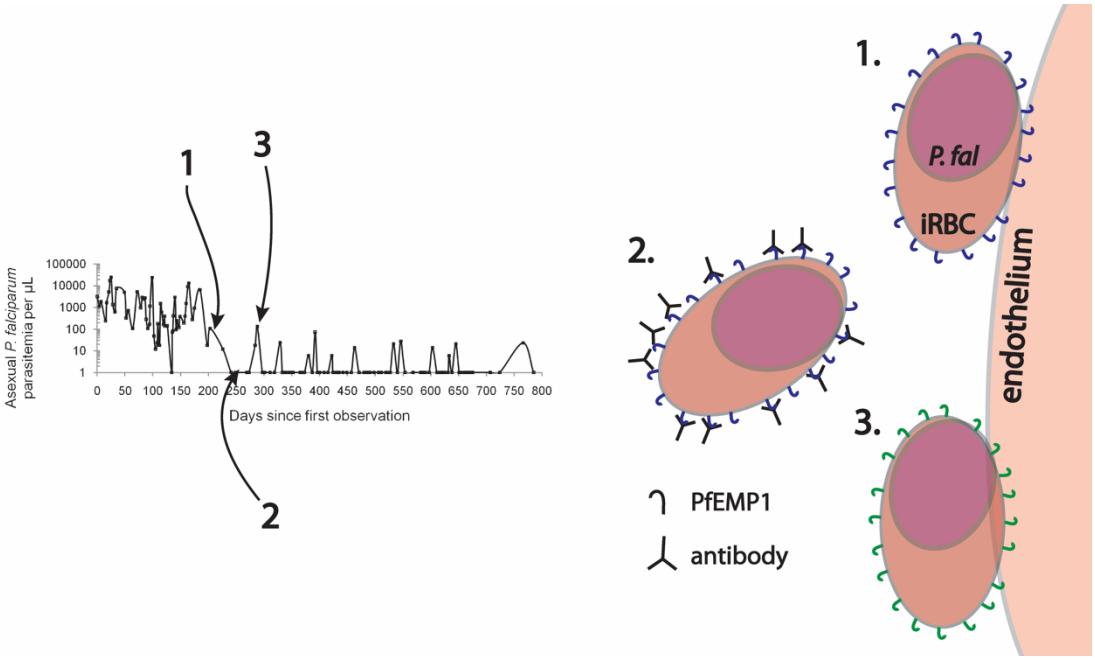


Figure 5. Model for the maintenance of chronic *P. falciparum* infection through clonal antigenic variation numbered peaks on the waves of parasitemia graph (left) over the course of natural human infection correspond to the cartoon model (right): 1. Clonal parasite population arises expressing a variant of PfEMP1 allowing them to cytoadhere to the host endothelium and avoid immune system clearance. 2. Over time individuals with a healthy immune system can mount an antibody response to this form of PfEMP1, corresponding to a sharp decrease in parasitemia. 3. A subpopulation of parasites expressing a new variant of PfEMP1, for which there are not yet antibodies against, proliferates and maintains the infection. Parasitemia graph (left) taken from Chen, D. S. *et al.* A Molecular Epidemiological Study of var Gene Diversity to Characterize the Reservoir of Plasmodium falciparum in Humans in Africa. *Plos One* **6**, e16629 (2011)[151]. (Open access).

1.5 *Epigenetics of* var *gene regulation*

Epigenetics refers to the heritable alterations of cellular phenotypes that are independent of changes in the organism's primary DNA sequence; or, it is otherwise classified as the study of alternative modes of genetic inheritance. [27,28] The activation and silencing of *var* genes is controlled epigenetically, as the transcription state of *var* genes has not been observed to involve any alterations at the DNA sequence level, or to require changes in the presence or absence of typical transcription factors. *var* genes that are transcriptionally silent are found in a state of condensed heterochromatin, whereas the single active *var* gene in any given cell is maintained in a relaxed, transcriptionally permissive euchromatic state. [29–31] Furthermore, common histone marks found in higher eukaryotes responsible for regulating gene expression were found to be uniquely distributed around clonally variant, multi-copy gene families in *P. falciparum.* [32–34] Several studies demonstrated that deposition of these marks directly influences *var* gene expression [29,35,36].

Three marks have been found to be of primary importance in *var* gene regulation. The single active *var* gene is acetylated at the H3K9 position (Histone 3, Lysine 9; H3K9ac), while the remaining silent *var*s are tri-methylated (H3K9me3). Conversely, the H3K36me3 mark is found at both active and silent *var* genes. [37] Di- and tri-methylation of the H3K4 site are enriched at actively transcribed genes in many organisms, and similarly, in addition to H3K9ac, H3K4me3 levels are found around the single *var* promoter marked for activation. [38] [31] Methyl groups at each of these positions are deposited by specific histone methyltransferases (HMTs), which in *P. falciparum* are largely SET-domain-containing proteins. PfSET2 and PfSET3 deposit the H3K36me3 and H3K9me3 marks, respectively. [37,39,40] More recently, PfSET10 was found to deposit the H3K4me3 mark. [38] Several experiments have illustrated the importance of these marks in regulating *var* expression. When PfSET2 was knocked out, leaky expression of all *var* genes across a population was observed[37]. *P. falciparum* heterochromatin protein 1 (PfHP1) and *P. falciparum* histone deacetylase 2 (PfHda2) are both silencers of gene expression, through the deposition of methyl marks and formation of heterochromatin, or removal of acetyl marks to allow for methylation, respectively.

Independent knockdowns of either protein similarly led to leaky expression of all *var* genes in the population[41,42].

DNA methylation is another method used by most eukaryotic cells to epigenetically regulate gene expression. In contrast to histone methylation, however, DNA methylation in *P. falciparum* appears to be exceptionally rare, and has not been linked to gene expression. [43] Although the identification of these epigenetic marks associated with the *var* gene family has been critical to understanding their regulation, the molecular mechanisms governing the activation and silencing of individual genes, as well as the coordination of the switching process, remain incompletely defined.

1.6 var *gene diversity*

While most *var* genes are found in the subtelomeric regions of chromosomes, some can be found in central chromosome regions. [11] The chromosomal location and gene orientation of *var* genes are linked with types of 5'-flanking regions called *ups,* sorting them into *upsA, upsB*, and *upsC* types (also known as A, B, and C type *var* genes, or *var* subfamilies). Subtelomeric *var* genes whose orientation faces the telomeric end are *upsA*, while those facing towards the centromere are *upsB*. *var*s found in internal chromosomal clusters are mostly *upsC* type, though some are *upsB*. [19] Severity of disease has been linked to *var* gene type, as this determines PfEMP1 expression and iRBC cytoadhering properties, with more severe cases manifesting from type A *var*s (notably cerebral malaria). [7,26] However, more research is needed to fully elucidate the connection between *var* expression and parasite virulence.

Given that the foundation of *P. falciparum* immune evasion is antigenic diversity, increased rates of recombination between *var* genes evolved as a strategy to drive diversity in variant, multi-copy gene families. Consequently, the 60-member *var* gene family shows remarkable hypervariability; even when comparing parasite isolates from the same geographic region, there is minimal overlap in *var* gene repertoires. [19,21] The degree of *var* diversity varies by location within the genome, with higher rates of diversity found in those located in subtelomeric regions. As seen in other eukaryotes, the telomeres of *P. falciparum* are tethered to the nuclear periphery and arranged in

clusters, or “bouquets,” that bring members of each *var* subfamily into alignment and facilitate recombination. [21,43,44] Notably, several unique *var* members are universally conversed across *P. falciparum* isolates: *var1csa* (type D)*, var2csa* (type E), and type 3 *var* genes. [19,44,45]

1.7 var2csa: *A unique* var *gene*

var2csa is an intriguing member of the *var* family. It encodes a form of PfEMP1 that exclusively binds to a placental-specific ligand, chondroitin sulfate A (CSA), meaning that the PfEMP1 protein encoded by this gene is only useful to the parasite when found in women who are infected for the first time while pregnant, as antibodies will be produced to it. [46,47] Given how small a portion of the host population this is, it is interesting to speculate as to why this gene has been so widely conserved across *P. falciparum* isolates. [45] In contrast to most other *var* genes, *var2csa* can be translationally repressed even when transcriptionally active, due to the presence of a unique upstream open reading frame (uORF) **Figure 6**. [48,49] Surprisingly, *var2csa* gene transcripts can be detected in non-pregnant individuals. [50-52] Therefore, *var2csa* can be transiently expressed without immune recognition. These observations suggest that *var2csa* might serve a separate function within the parasite apart from encoding PfEMP1 protein.

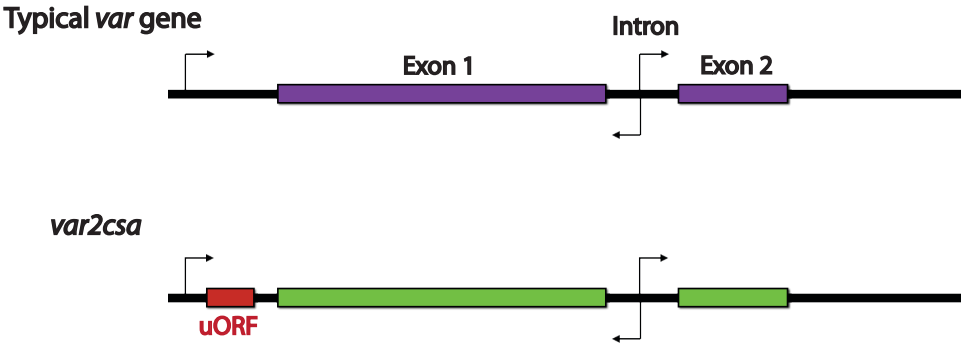


Figure 6. *var* gene structure All *var* gene family members contain two exons that encode for PfEMP1 protein, separated by an intron. The upstream 3’ promoter drives expression of the mRNA and is governed by mutually exclusive expression. A bi-directional promoter is found within the intron, transcribing sense and antisense ncRNAs. The sense ncRNA is transcribed in both active and silent *var* genes. *var2csa* has a unique uORF that allows for selective transcription with translational repression.

Several independent studies have hinted that *var2csa* might play a role in coordinating *var* switching. Clonal parasite cultures grown without selection for over approximately 200 generations (more than one year) were found to display a high activation rate of *var2csa*, suggesting that expression of *var2csa* might represent a "default" state. [50] Altering certain epigenetic processes governing mutually exclusive expression of *var* genes has also resulted in activation of *var2csa*. PfSET2, the HMT that deposits the H3K36me3 mark, is recruited to specific regions of the genome by binding directly to the C-terminal domain (CTD) of RNA polymerase II (RNA pol II) during the transcription of ncRNAs from both active and silent *var* genes. [40] Over expression of a PfSET2 dominant-negative construct, designed to knock down but not completely knock out SET2 HMT activity by competing with the endogenous protein, results in activation of *var2csa* at the population level over time. [40,51] Treatment of cultures with subIC_{50} levels of chaetocin, a proposed inhibitor of PfSET3 HMT activity, similarly results in selective activation of *var2csa.* [51] Controlled human infection studies found evidence of *var2csa* transcription over the course of monitored *in vivo* switching. [52] Lastly, deletion of *var2csa* from the parasite's genome disrupts *var* switching. (Zhang et al. personal communication)

1.8 *The paradigm of programmed, stochastic* var *switching*

African trypanosomes similarly rely on antigenic variation to cause persistent infection in a host through mutually exclusive expression of variant surface glycoprotein (*vsg*) genes. In these organisms switching appears to be stochastic, as the population found in each "wave" of parasitemia expresses many different *vsg* genes at a time. [53] While trypanosomes contain thousands of variable *vsg* genes, *P. falciparum* contains only ~60 *var* gene family members. [54] Therefore, if *P. falciparum* followed the trypanosome model of switching it would quickly exhaust all of the antigens in its repertoire in the first wave (**Figure 5**). Consequently, the question remains how *P. falciparum* can maintain an antigen-switch rate that allows for the emergence of a new variant when necessary, without rapidly exhausting all 60

members, to sustain long-term infections. Two possible mechanisms have been proposed[19]:

1. *P. falciparum* parasites have developed an inherent switch rate that, over evolutionary time, has selectively adjusted to the time it takes for the mammalian immune system to mount a productive antibody response against PfEMP1. Thus, parasites are programmed to switch *var* expression at a set interval of time, independent of any external stimuli.
2. *P. falciparum* utilizes a flexible strategy that incorporates sensing machinery to allow for changes in *var* switch rates in response to external factors, such as antibodies on the surface of the iRBC.

The currently held paradigm proposes the first model, but is assumed from characterization of antigenic variation mechanisms in trypanosomes and bacteria, as well as the observation that parasites grown in the absence of any selective pressures still show changes in *var* gene expression. [55] There is little to no evidence, however, that disproves the idea that these parasites have some sort of sensing mechanism enabling them to respond to outside stimuli, such as an immune response, and this proposition has not been investigated further.

A second question is whether a set hierarchy in switching exists (ordered *var* switching), or whether the gene that becomes active in the event of a switch is completely random (stochastic *var* switching). Numerous studies have provided evidence in support of the former. Different *var* genes have been shown to have different intrinsic switch rates: *in vitro* those located in subtelomeric chromosomal regions (Type A and some B *var*s) switch between active and silent states frequently, whereas those more centrally located (Type C and some B *var*s), once active, are stable and rarely switch. [56,142] The waves of parasitemia seen over the course of chronic infection (**Figure 5**) also support this idea: if switching was entirely random, resulting in multiple, dominantly expressed variants of PfEMP1 in the parasite population within the host at any given point in time, the peaks witnessed would not be nearly as discrete as those observed. Furthermore, each of the peaks in parasitemia encompasses an incredibly high number of parasites, measured in patients at highs of $>10^{12}$ [7]. Without

some method of coordination to *var* switching and PfEMP1 exposure, it would seem inevitable that the entire repertoire would be exposed in the first wave alone. Thus, while switching at random times would presumably exhaust the 60-gene repertoire rapidly, switching in response to the host would more efficiently make use of the limited number of genes.

Considering the observations mentioned above, from *in vitro* culture systems lacking selective pressure as well as experimental infection systems undergoing selective pressure, two mathematical models have been proposed that independently support non-random switching. Both models suggest that *var* gene switching occurs through the use of transiently active genes, or "switch intermediates", which add a level of uniformity and coordination to the process. [55] In this way, switch intermediates serve as "sink nodes": any initially dominant *var* transcript switches first to the intermediate, and then away from it to either to a new dominant transcript, or back to the original. Use of a switching intermediate thus serves to prevent bias in the switching process and ensures that genes closely linked to the previously active gene aren't preferentially activated by virtue of genomic position. [51,55] While the identity of any "switch intermediate" is presently undefined, it is hypothesized that *var2csa* could serve as the sink node, due its unique properties listed above. [2]

B. Introduction to One-carbon and SAM Metabolism

1.9 *What is One Carbon and SAM Metabolism?*

While the studies presented in Chapter 1 support the proposition that antigenic switching in *P. falciparum* is not random, they do not address what exactly triggers a parasite to switch from expressing one *var* gene to the next: is there a set genetic program that controls switching independent of any outside stimuli, or does the parasite respond to outside stimuli by switching? Recent studies highlighted in this chapter on metabolism in cancer cells and yeast have shown that nutrient availability affects intracellular metabolism, which in turn can have a profound impact on histone methylation and, consequently, gene expression.

One-carbon metabolism is a metabolic network that utilizes nutrients, such as glucose, vitamins, and amino acids, from the environment to fuel multiple biological functions: nucleotide metabolism, maintenance of cellular redox status, lipid biosynthesis, and methylation. [57] This network features two major metabolic cycles, folate and methionine, whose functions are to transfer single-carbon units to acceptor substrates. [57] The methionine arm of this network links it to histone methylation, and thus the epigenetic wing of the cell, through intermediate metabolites S-adenosylmethionine (SAM) and S-adenosylhomocysteine (SAH). SAM, generated through the folate and methionine cycles, is the universal methyl donor for many cellular functions, yielding SAH after the loss of its methyl group. SAM facilitates histone methylation by donating its methyl group to HMT enzymes, SAH is then hydrolyzed to homocysteine and recycled through **Figure 7**. [57,58]

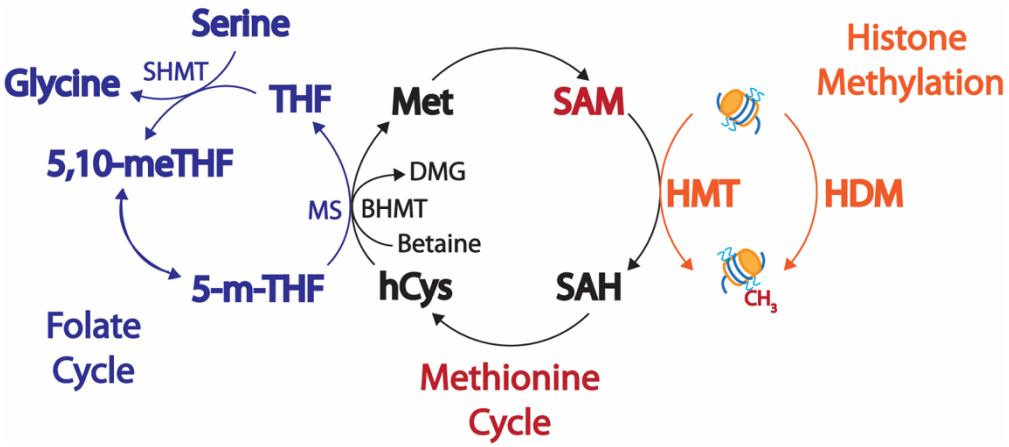


Figure 7. One-carbon metabolism and histone methylation S-Adenosylmethionine (SAM) is produced from methionine (MET) by methionine adenosyltransferase (MAT). Histone methyltransferases (HMTs) utilize SAM to donate a methyl group to their histone substrates, producing S-adenosylhomocysteine (SAH). Histone demethylases (HDMs) remove the methyl group from histones, returning them to an unmethylated state. SAH is converted to homocysteine (hCys) via S-adenosylhomocysteine hydrolase (SAHH). To complete the cycle, SAH can be remethylated to regenerate MET by donation of a methyl group from 5-methyltetrahydrafolate (5-mTHF) via methionine synthase (MS) or from betaine via betaine–homocysteine S-methyltransferase (BHMT). Figure and caption adapted from Mentch and Locscale 2015.

SAM metabolism and its role in epigenetic changes has become a popular subject in the study of chronic diseases: notably, cancer, diabetes, and obesity. Differences in the nutritional requirements of cancer cells have been noted as a hallmark of oncogenesis for almost a century, but only more recently have they been characterized as both the result of and leading to changes in the epigenome. [59] Accordingly, metabolic reprogramming of cancer cells has been found to involve many of the nutrients associated with one-carbon and SAM metabolism, specifically serine, glycine, and methionine. [28] It is from closer study of cancer cell metabolism that recent evidence for a direct link between nutrient, SAM, and histone methylation levels has been drawn.

1.10 *Changes in SAM metabolism influences gene expression*

Following canonical enzyme kinetics, HMT activity depends on the concentration of its substrate, SAM. Evidence proposes that the SAM/SAH ratio plays a role in regulating HMT activity, as SAH is a known inhibitor of HMTs. [60,61] In contrast to protein phosphorylation kinetics, where the concentrations of ATP (adenosine triphosphate) substrate far exceed the Michaelis constant (K_m), HMT K_m values lie in the range of intracellular SAM concentrations. This further suggests that even minor fluctuations in SAM levels could significantly affect HMT activity, increasing or decreasing histone methylation and providing a direct link between it and cellular metabolism through SAM. [62,28,57]

Recent work in different cell systems has provided evidence for this link. Altering the availability of methionine leads to changes in histone methylation following observed fluctuations in SAM levels. [58,63,64] All of these studies see changes specifically at the H3K4 mark, possibly owing to the higher K_m value of its corresponding HMT. [28] Decreased histone methylation at this mark was also observed in response to lowering SAM levels by reducing intracellular glycine concentrations via threonine input. [65] Overexpression of SAM-consuming enzyme nicotinamide n-methyltransferase (NNMT) increased SAH concentrations relative to SAM (or decreased the overall SAM/SAH ratio) leading to a decrease in histone methylation. [66] NNMT is overexpressed in certain

cancer cells with this work showing its potential to serve as a “methyl sink”, or consumer of methyl units from SAM, altering the cell’s overall epigenetic state. Similarly, work demonstrated that yeast cells lacking key enzymes in pathways that are major consumers of SAM accumulate SAM and exhibit hypermethylation of histones, resulting in changes in gene expression. [67] Thus, under conditions of high SAM/SAH, histones can serve as a “methyl sink” altering the epigenome and leading to downstream changes in gene expression.

1.11 *The effect of environmental nutrient fluctuations on* P. falciparum

To successfully move between sexual stages in the mosquito vector to asexual stages in a human host, *P. falciparum* must alter its phenotype to developmentally adapt to each new niche. It elegantly accomplishes this by altering its transcriptional programming in a cyclic manner, with peaks of expression of certain genes at different growth stages. [68,69] Further, each new host brings a new environment and set of selective pressures the parasite needs to respond to. In order to sufficiently respond to shifting environments, *P. falciparum* has evolved the ability to vary its transcriptional programming from parasite-to-parasite through the epigenetic plasticity of clonally variant gene expression. As touched on in **Chapter 1A**, this includes antigenic variation of PfEMP1 orchestrated by *var* genes as well as other parasite proteins expressed on the RBC surface. Excluding *var* genes, classes of these molecules have been observed to have modest but direct transcriptional responses to temperature, metabolite fluctuations, and drug pressure. [27,69–71]

Several recent reports exemplify this capacity in *Plasmodium* species, specifically in response to changes in local environment. In one, the murine parasite, *Plasmodium berghei*, demonstrated a nutrient sensing mechanism *in vitro* and *in vivo*. Under conditions of nutrient restriction, signaling through a protein kinase activates transcriptional programming to reduce multiplication rates in response. [72] The authors also found potential for *P. falciparum* to possess similar machinery *in vitro*. A second study exhibited that *P. falciparum* can sense the depletion of lysophosphatidylcholine (lysoPC), a component of human serum, and respond by activating the gene *pfap2-g.*

[73] This genetic switch commits the parasite to the sexual differentiation pathway, beginning their development into gametocytes - the stage that is picked up by mosquitoes for further transmission (**Figure 1A**). This work draws a line connecting host nutrient conditions with resulting epigenetic and gene expression changes in *P. falciparum*, as the *pfap2-g* locus is silenced by a heterochromatin-based mechanism in asexually growing parasites. [74] LysoPC depletion also resulted in changes in the expression of other metabolism-related genes, demonstrating the capacity of the organism to modify its metabolic state in response to that of its host. [73] Additionally, a separate study elucidated a mechanism by which hyperhomocysteinemia, a long-known characteristic of malaria disease, induces gametocytogenesis in *P. falciparum.* [75] It was found that the parasites lack key enzymes in a redox pathway, leading to increased local concentrations of homocysteine that results in downstream activation of an *ap2-g* transcription factor. [75] These three reports illustrate the ability of *P. falciparum* to sense changes in its environment and respond by altering its transcriptome. None, however, identify the master regulator(s) orchestrating these transcriptional changes, and additional work is needed to determine other nutrient variabilities the parasite encounters, and is capable of adapting to, in its host. [71]

1.12 *Lipid metabolic pathways in* P. falciparum *intersect with SAM metabolism*

Throughout its intraerythrocytic life cycle *P. falciparum* replicates and divides into as many as 36 merozoites, requiring large amounts of new membranes. Tantamount to this, over the course of one 48-hour replication cycle the phospholipid content of the iRBC is noted to increase approximately 5 to 6-fold. [76] Similar to most eukaryotes, the primary component of their membrane is phospholipids, with two, phosphatidylcholine (PtdCho) and phosphatidylethanolamine (PtdEtn), comprising 40-55% and 30-40%, respectively, of the parasite's total plasma membrane phospholipid content. [78-81] *P. falciparum* possesses the machinery to synthesize these compounds from host nutrient precursors, such as choline, serine, inositol, glycerol, and fatty acids. The synthesis of primary membrane lipid PtdCho can take place via two metabolic pathways: the cytidine diphosphate (CDP)-choline pathway (otherwise known as the

Kennedy pathway) and the serine-decarboxylase-phosphoethanolamine methyltransferase (SDPM) pathway (**Figure 8**). [77–79] The de novo CDP-choline pathway uses choline and fatty acids from host serum as precursors, while the SDPM pathway uses serine and fatty acids. In running the SDPM pathway, serine must first be decarboxylated to ethanolamine, then converted to phosphoethanolamine (p-Etn), before joining the CDP pathway as phosphocholine (p-Cho). Serine is in abundant supply in the parasite cytosol resulting from the digestion of hemoglobin; thus the SDPM pathway serves as an additional, reliable pathway to produce PtdCho. [79,80]

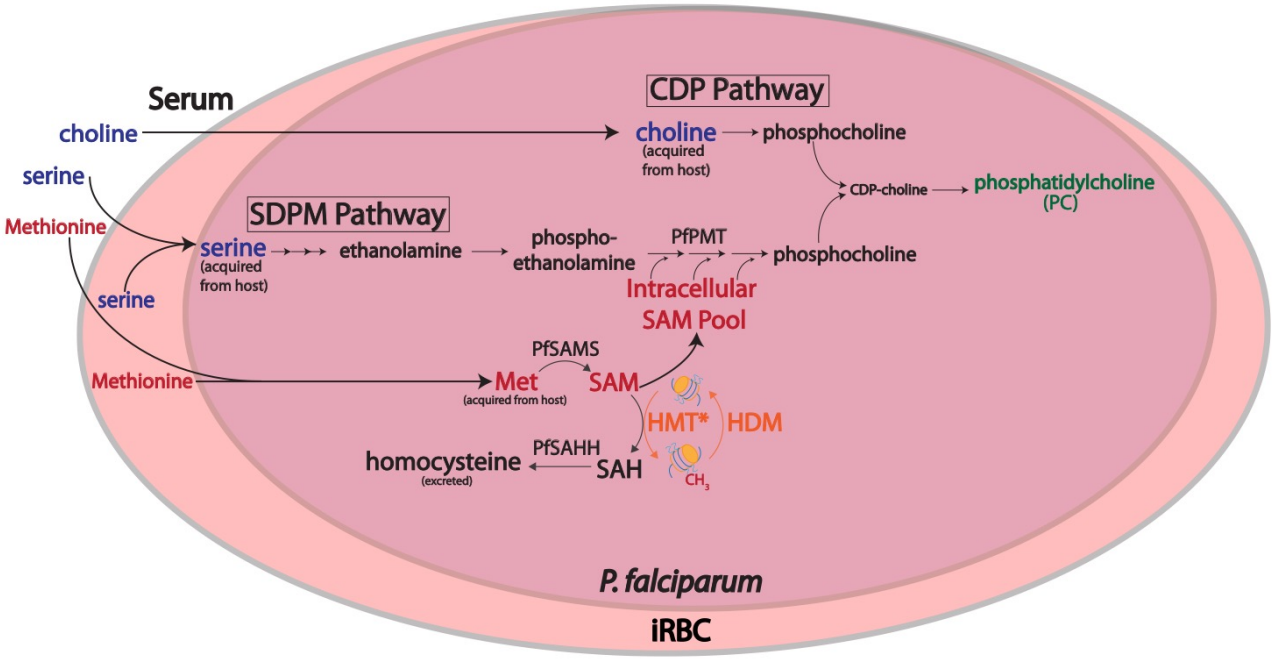


Figure 8. Pathways for the biosynthesis of phosphatidylcholine in *P. falciparum* The parasite must synthesize a large and continuous amount of phospholipid membrane components as it grows within the erythrocyte. Choline taken in from human serum is used in the cytidine diphosphate (CDP)-choline pathway to make PtdCho the primary lipid found in the membrane of *P. falciparum*. Parasites can also synthesize PtdCho from non-choline sources using the serine decarboxylase–phosphoethanolamine methyltransferase (SDPM) pathway, which joins the CDP pathway after the methylation of p-Etn to p-Cho by PfPMT. This reaction requires three SAM, thus depleting SAM pools. SAM synthetase (PfSAMs) catalyzes the formation of SAM from methionine and ATP, while SAH hydrolase (PfSAHH) catalyzes the reversible hydrolysis of SAH to adenosine and homocysteine

A key enzyme in the SDPM pathway is phosphoethanolamine N-methyltransferase (PfPMT), which specifically methylates p-Etn to p-Cho (**Figure 8**). This three-step methylation reaction is dependent on SAM as a methyl donor, linking the synthesis of these key membrane lipids to intracellular SAM pools. A *pfpmtΔ* null mutant lacking PfPMT activity was found to display delayed growth, altered DNA replication, reduced multiplication rate, and increased cell death. [79] The mutant, unable to use serine as a source of PtdCho biogenesis, is able to survive on residual choline in human RBCs. Surprisingly, the addition of excess choline in culture did not rescue growth, suggesting that the SDPM and CDP pathways are not completely redundant. [78,79] Further work found that chemical inhibition of PfPMT activity *in vitro* blocks gametocyte development, in addition to drastically slowing asexual replication. Correspondingly, mosquitoes fed on blood infected with the *pfpmtΔ* mutant and maintained for eight days were found to contain no oocysts in their gut in contrast to those fed on blood infected with wildtype parasites. [80] Thus, PfPMT is considered essential for gametocytogenesis and malaria transmission.

PfPMT is a member of the PEAMT family of phosphoethanolamine methyltransferases also found in worms, plants, and other protozoa, but is notably absent from rodent malaria parasites, *P. berghei* and *P. yoelii,* and human and mammalian databases. [79] This evidence, along with its restricted phylogenic distribution and lack of homology to human protein databases, makes PfPMT an attractive drug target. [78]

Experiments demonstrating the inhibitory effect of lysoPC on gametocyte development further demonstrate the link between PfPMT and gametocytogenesis. LysoPC restriction results in reduced numbers of progeny along with transcriptional changes, notably increased expression of PfPMT and activation of *ap2-g*. [73] When combined, these independent bodies of work intriguingly point to an overall importance of nutrient uptake and phospholipid metabolism, necessary for intraerythrocytic asexual replication and sexual differentiation, to the control of parasite gene expression, expansion, and survival in the human host.

Thesis overview

The molecular mechanisms that control activation and silencing of individual *var* genes, as well as how the entire switching process is coordinated, presently remain incompletely defined*. P. falciparum* is capable of causing long-term, chronic human infections (**Figure 5**) while containing only ~60 *var* gene family members in its genome. Each wave of parasitemia (**Figure 5**) encompasses an incredibly high number of parasites, measured in patients at highs of $>10^{12}$ [7]. Without some method of coordination to *var* switching and PfEMP1 exposure, it would seem inevitable that the entire repertoire would be exposed in just the first wave. How does this parasite maintain an antigen-switch rate that allows for the emergence of a new variant when necessary, without rapidly exhausting all 60 members? In the following thesis I will investigate one facet of this question, by suggesting the novel possibility that *P. falciparum* possesses cellular machinery capable of sensing changes in the environment of its host and responding by altering *var* gene expression.

In **Chapter 2** I will describe preliminary experiments exploring the connection between metabolism and *var* gene expression in *P. falciparum.* Here, parasites were cultured in growth media containing altered concentrations of nutrients involved in SAM metabolism. These conditions ultimately tied to key lipid synthesis pathways within the parasite that will be discussed in detail. Those believed to increase SAM pools induced a coordinated switch to one particular *var* gene, *var2csa*, over time, phenocopying the results from Ukaegbu et al., 2015. Taking these and past results together, the data suggest a model where conditions that increase intracellular SAM pools induce a coordinated population-level switch to *var2csa*.

This hypothesis will be tested further in **Chapter 3**. Here, I will describe a set of experiments where the expression of key enzymes involved in SAM metabolism were altered. Again, modifications thought to increase the intracellular level of SAM were found to induce a coordinated switch at the population level to *var2csa.* Conversely, modifications that lower the level of SAM did not induce expression of *var2csa,* but instead many *var*s at once across the population.

In **Chapter 4**, I will examine the physiological relevance of the experiments in the previous two chapters by testing the effects of possible environmental signals on *var* expression. After recognition of a pathogen, activated macrophages modify their microenvironments in various ways. I looked at two of these immune responses, depletion of amino acids and release of polyamines, on *var* expression of parasites *in vitro.* Both environmental perturbations altered *var* expression, again specifically inducing *var2csa.*

Taking all described results together, I will propose and examine two possible models of antigenic variation in *P. falciparum* in **Chapter 5**. The first will center intracellular SAM metabolism in describing a promoter competition model governing *var* switching through *var2csa.* The second will suggest that *P. falciparum* can sense when the host immune system first begins to recognize it, via environmental cues resulting from antibody recognition, and respond by switching *var* gene expression. This novel hypothesis would explain how parasites are able to switch expression of *var* genes exactly when needed, allowing them to utilize their limited repertoire of *var* genes with the upmost efficiency. Lastly, future experiments and directions of interest to explore in further support of both models will be discussed.

Chapter 2

Altering Environmental Levels of Lipid Metabolic Precursors Induces *var* Switching

Background

Many eukaryotic organisms employ antigenic variation as a strategy to avoid antibody recognition and cause persistent, chronic infections. [20] Antigenic variation is the coordinated expression of variant surface-exposed antigenic determinants, often achieved by switching from one member of a multigene family to another. This coordination is executed in a mutually exclusive manner, ensuring that only a small portion of the organism's repertoire of surface antigens is exposed at any time and thereby limiting its vulnerability to its host's antibody response. [51] *Plasmodium falciparum* has a multi-copy gene family called *var* that encodes variant forms of a cytoadhering protein, PfEMP1. PfEMP1 is expressed on the surface of *P. falciparum's* housing RBC, where it can tightly bind various host cell receptors and sequester away from peripheral circulation and mechanical clearance in the spleen. Because of its external display on the RBC surface and interaction with host proteins, PfEMP1 serves as the primary antigenic determinant. Therefore, by varying its antigen display through mutually exclusive expression of *var* genes and transcriptional switching, *P. falciparum* can cause persistent infections in humans, characterized by waves of parasitemia over time. This process is illustrated in **Figure 5**. [2,51]

P. falciparum has evolved the ability to vary its transcriptional programming from parasite-to-parasite through epigenetic control of clonally variant gene expression. *var* genes that are transcriptionally silent are found in a state of condensed heterochromatin, whereas the single active *var* gene in any given cell is maintained in a relaxed, transcriptionally permissive euchromatic state. [29–31] Common histone marks found in higher eukaryotes responsible for regulating gene expression were found to be uniquely distributed around clonally variant, multi-copy gene families in *P. falciparum.* [32–34] The single active *var* gene is acetylated at the H3K9 (H3K9ac) position and tri-methylated at H3K4, while the remaining silent *var*s are tri-methylated

(H3K9me3); The H3K36me3 mark is found at both active and silent *var* genes. [37] [38] [31] Methyl groups at each of these positions are deposited by specific HMTs: PfSET2 and PfSET3 deposit the H3K36me3 and H3K9me3 marks, respectively. [37,39,40] More recently, PfSET10 was found to deposit the H3K4me3 mark.[38]

Interfering with this epigenetic regulatory pathway believed to govern mutually exclusive expression of the *var* gene family has uncovered clues as to how the process of switching is coordinated. PfSET2 is recruited to specific regions of the genome by binding directly to the CTD of RNA pol II during the transcription of ncRNAs from both active and silent *var* genes. [40] Knocking out PfSET2 leads to leaky expression of all *var* genes, consistent with the idea that the H3K36me3 mark plays some role in maintaining mutually exclusive expression of *var* gene family members. [37] More recently, a dominant-negative genetic construct for PfSET2, designed to down-regulate, but not completely knock-out, its HMT activity, was engineered. This truncated version of PfSET2 competes with the endogenous protein for binding to RNA pol II, thereby reducing recruitment to regions of the genome surrounding *var* genes without affecting enzyme levels. [51] Over-expression of this construct in parasites resulted in changes in *var* gene expression at the population level over time. Strikingly, these changes were not random and did not lead to leaky expression of numerous *var* genes simultaneously as seen before, but instead to specific, singular activation of *var2csa*. [51]

In human cells, chaetocin is a known inhibitor of H3K9 di- and trimethylation; bioinformatic analyses identified *P. falciparum* HMT PfSET3 as its most likely target. Given that the H3K9 mark is similarly found in regions of the genome occupied by *var* genes, a second set of experiments treated cultures with sub-IC_{50} concentrations of chaetocin, to, again, down-regulate but not completely inhibit HMT activity. This chemical manipulation of the presumed activity of PfSET3 in deposition of the H3K9me3 mark, similarly resulted in induction of *var2csa* expression at the population level over time. [51] These three studies show that altering activity of PfSET2 and PfSET3 *in vitro* induces dramatic *var* expression changes at the population level.

In contrast to African Trypanosomes that have thousands of *vsg* genes in their antigenic repertoire to switch between, *P. falciparum* has only 60, begging the question as to how they manage to cause persistent, long-term infections. [54] Each wave of parasitemia seen in trypanosome infections contains a parasite population expressing many different *vsg* genes, suggesting that they employ a stochastic switching dynamic. [53] If *P. falciparum* were to follow a similar mechanism it would express all of the antigens in its repertoire in the first wave, and infections would not last as long as is documented. [7] This in turn suggests that *P. falciparum* must utilize a more elegant and precise switching mechanism, which so far has yet to be described.

Based on antigenic variation mechanisms characterized in trypanosomes and other organisms, the commonly held paradigm for *P. falciparum* suggests that the parasites evolved an internal clock programmed to time a switch in *var* expression with the time it takes the host to mount an antibody response, independent of any external stimuli. [19] In this chapter I will provide the first evidence of an observed switch in *var* expression at the population level over time in response to altered nutrient conditions. The preliminary experiments described here found that specific modification of lipid precursors in the culture media induced *var* switching. Notably, the changes in *var* expression in the population phenocopied those seen in the earlier experiments mentioned above, where HMT activity was downregulated. Based on this similarity and the connection of lipid metabolic pathways in *P. falciparum* to SAM metabolism, I will propose a model wherein the parasite regulates *var* switching through SAM pools.

Results

2.1 Preliminary Work and Rationale

The inspiration for this project arose from work conducted in Dr. Choukri Ben Mamoun's lab, studying lipid metabolic pathways in *P. falciparum.* As the parasite replicates and divides into as many as 36 merozoite progeny per cell, it requires large amounts of new membrane, increasing the total phospholipid concentration of the iRBC by as much as 6-fold. [76] The rapid growth and multiplication found within the iRBC is made possible by nutrients supplied by the host, with fatty acids, choline, serine, and ethanolamine of primary importance in phospholipid synthesis. [78] In addition to membrane biogenesis, lipid-dependent pathways play other important roles in the organism's development by facilitating intracellular signaling, protein trafficking, and hemoglobin degradation. Therefore, it is not surprising that the uptake, transport, and metabolism of lipids are essential for the asexual viability of *P. falciparum.* [78,81]

The essential and unique nature of these pathways have made them an area of interest in antimalarial drug development. [76] One such feature is the enzyme PfPMT, which allows for the input of serine (in instances of limiting exogenous choline) in synthesizing key membrane component PtdCho through the SDPM pathway (**Figure 8**). PfPMT catalyzes a three-step methylation reaction that is dependent on SAM as a methyl donor, linking lipid membrane biogenesis to intracellular SAM pools. SAM is the primary methyl donor to HMTs, which, as described above, deposit epigenetic marks controlling the mutually exclusive expression of *var* genes. Intriguingly, the addition of excess choline in culture, allowing parasites to use the CDP-choline pathway to synthesize PtdCho, was observed to lower PfPMT transcript and protein levels, suggesting the capability of *P. falciparum* to adjust its use of these pathways based on the nutrient conditions of its host. [82] Therefore, I hypothesized that altering the levels of lipid precursors *in vitro* may have an effect on *var* expression through their connection to SAM metabolism.

2.2 Prolonged conditions of excess choline to depleted serine induces *var* gene switching

To test this hypothesis, a clone of 3D7 parental line *P. falciparum* parasites was cultured in differing concentrations of serine and choline, with bulk RNA extracted for total *var* expression analysis at different time points (two and four weeks of growth). In order to carry out the nutrient alteration experiments that will be described throughout this thesis, a special dropout RPMI powder had to be developed. I called this Malaria Incomplete Media (MIM), as opposed to the standard RPMI 1640 which was denoted Malaria Complete Media (MCM). The production and composition of MIM is described in detail in **Methods**. Experimental parasites were cultured in MIM lacking choline and serine, where differing amounts of either were supplemented based on the condition being tested as shown in **Table 1**. These were grown alongside control parasites cultured in MCM. Serine is present in RBC hemoglobin, therefore conditions are labeled only as "depleted" serine referencing its absence from the media but not the RBCs used in culture. As 3D7 is the parent strain and its genome has been fully sequenced, it serves as the primary *P. falciparum* reference genome. [11] A full set of unique primers to each *var* gene in the 3D7 genome was generated, allowing for quantification of total *var* expression in the culture population by quantitative reverse transcriptase polymerase chain reaction (Q-RT-PCR, qPCR). [46] ,[Table 2] Total *var* expression was normalized to seryl-tRNA synthetase expression, a housekeeping gene in *P. falciparum*.

Table 1. ChoMetSer Conditions Tested in Chapter 2 Each altered nutrient condition tested is presented along with the compound name supplemented in parentheses. When a compound was depleted, as denoted in later figures, that nutrient was present at 0uM in growth medium. Levels of nutrients in human serum were taken from the Human Metabolome Database, https://hmdb.ca.

Condition	Amount Relative to MCM	uM tested	uM in MCM (RPMI)	uM in Human Serum	Sigma Catalog Number
Excess Choline (Choline Chloride)	20x	420	21	10-20	C7017
Choline (Choline Chloride)	1x	21	21	10-20	C7017
Excess Serine (L-Serine)	20x	5.7mM	285	56-140	S4500
Excess Methionine (L-Methionine)	20x	2mM	101	16-30	M5308

As hypothesized, altering the levels of choline and serine in the culture medium accelerated *var* switching in comparison to parasites cultured under standard nutrient conditions. Representative results of these experiments are presented in **Figure 9** with additional replicates shown in **Appendix**. Parasites cultured under conditions of excess choline (20x the amount present in MCM, **Table 1**) to depleted serine in MIM showed an accelerated and more pronounced switch in *var* expression at the population level than those grown in MCM (or under standard *P. falciparum* culturing conditions). Notably, conditions of excess choline are observed to singularly activate *var2csa,* phenocopying our lab's earlier results when HMT activity was modified genetically or chemically[51]. Parasites cultured in MIM with no supplemented choline or serine (or depleted of both choline and serine) did not see this same, coordinated switch in *var* expression and often mimicked background switching that was observed in those cultured in MCM. This supports the conclusion that the nutrient differences in the composition of the MIM, as compared to MCM, were not the sole cause of the observed switch to *var2csa* in excess choline.

Culturing parasites under conditions of excess (20x) choline utilized a much higher amount than is found in human serum (~20x higher than the upper limit found **Table 1**). In order to test the physiological relevance of the observed phenotype,

parasites were also cultured under 1x choline (the amount present in MCM and the upper limit found in human serum **Table 1**) in a background of depleted serine. The same *var* switch at the population level was also seen under these conditions (**Figure 9**, replicates in **Appendix**).

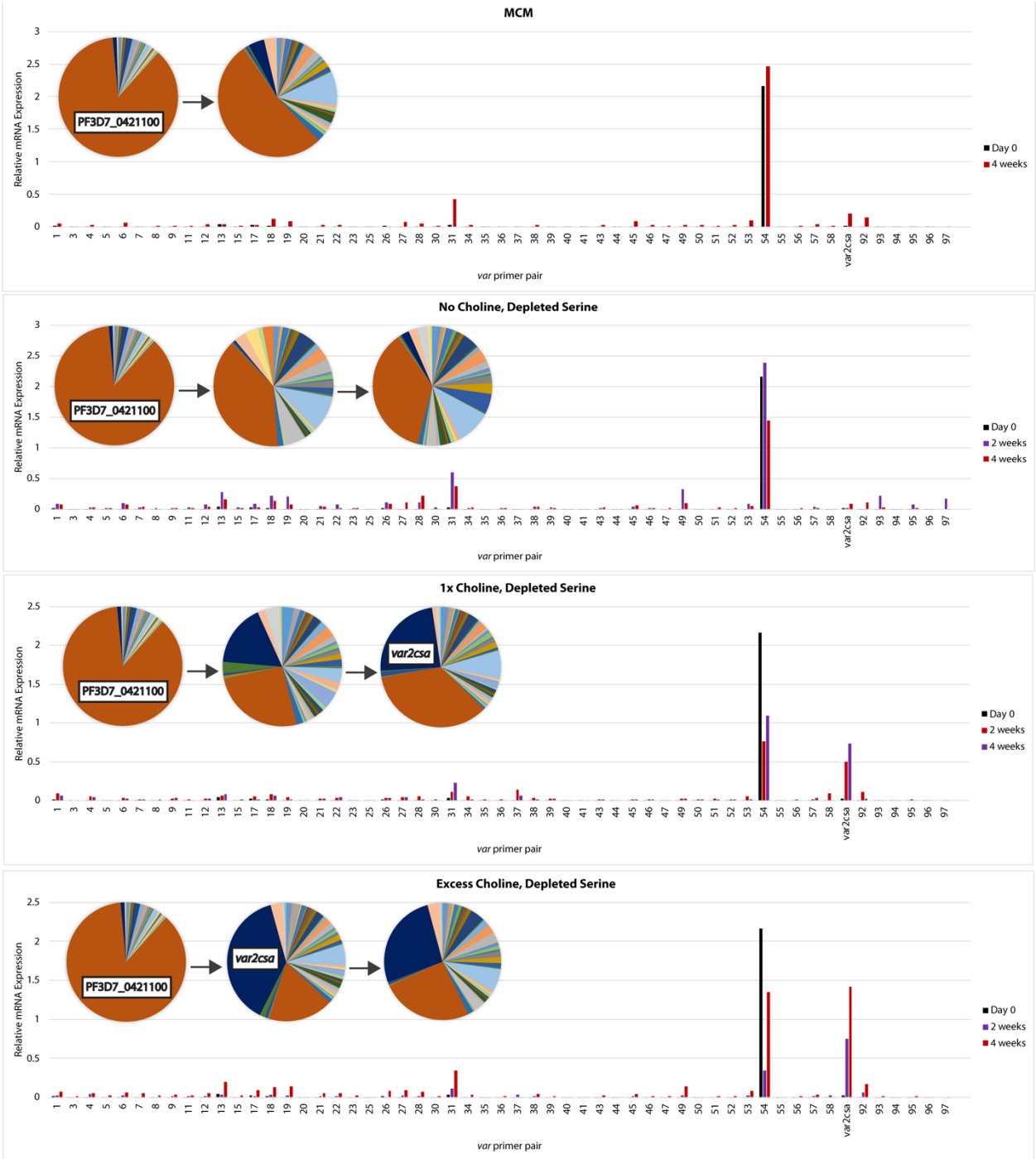
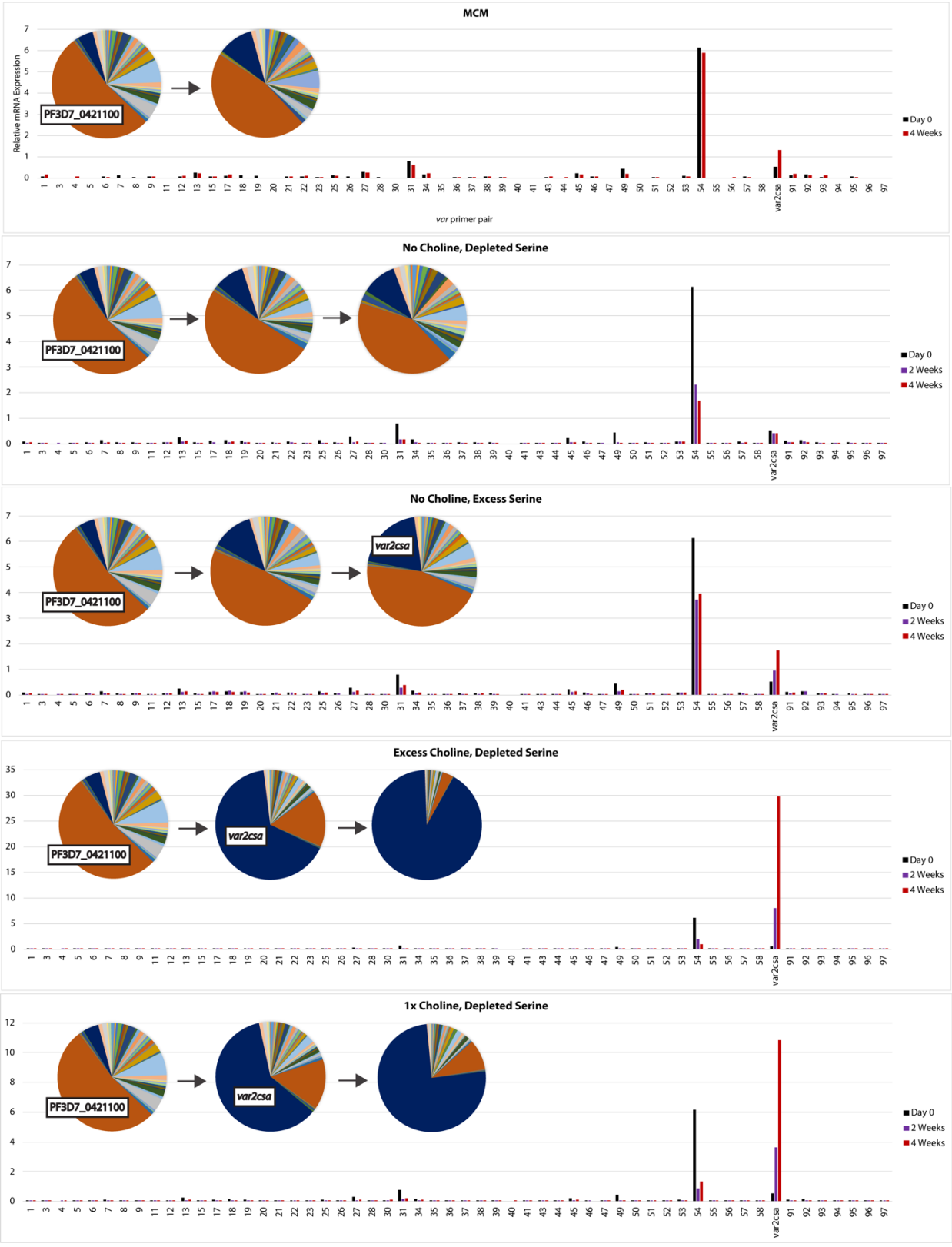


Figure 9. Elevated levels of choline in the growth media of *P. falciparum* induces a switch in *var* gene expression at the population level over time *var* gene expression as assayed for each condition is shown as a pie-chart and bar graph, with each slice of the pie and each numbered primer pair from Salanti et al. 2013 on the x-axis representing the fraction of the total *var* mRNA pool transcribed from each *var* gene. Black bars represent Day 0 timepoint, or the *var* expression of the parent culture before it was split across different nutrient conditions. Purple and red bars are the *var* expression at 2 and 4 week timepoints, respectively.

2.3 Prolonged conditions of excess serine to depleted choline do not induce *var* gene switching

Both choline and serine can be used by *P. falciparum* to synthesize membrane lipids through the CDP or SDPM pathway, respectively (**Figure 8**). While use of the CDP pathway does not require input of SAM, the SDPM pathway requires three SAM, thus both pathways have different consequences for intracellular SAM pools. Elevated levels of choline to serine in the growth media induced a *var* switch at the population level over a period of four weeks. To test if opposite conditions would have a similar effect on *var* expression, a set of parasites were cultured in the same MIM lacking choline and serine with excess serine supplemented back in following **Table 1**. These parasites were cultured alongside a set grown with supplemented choline to allow for direct comparison. RNA was extracted and bulk *var* expression analyzed by qPCR at two and four-week timepoints and results are shown in **Figure 10**. While the parasites grown in excess choline again saw a robust switch at the population level, those grown in excess serine did not see a switch above that of background (parasites grown in MCM). These results are representative of replicate experiments where parasites grown in conditions of excess serine do not see *var* switching over time (see **Appendix**). This experiment suggests that conditions where SAM pools are depleted, such as when the parasite must rely on serine to synthesize its membrane lipids, do not induce *var* switching at the population level over time.

Figure 10. Elevated levels of serine in the growth media of *P. falciparum* does not induce a switch in *var* gene expression at the population level over time *var* gene expression as assayed for each condition is shown as a pie-chart and bar graph, with each slice of the pie and each numbered primer pair from Salanti et al. 2013 on the x-axis representing the fraction of the total *var* mRNA pool transcribed from each *var* gene. Black bars represent Day 0 timepoint, or the *var* expression of the parent culture before it was split across different nutrient conditions. Purple and red bars are the *var* expression at 2 and 4 week timepoints, respectively.


MCM
Relative mRNA Expression
PF3D7_0421100
var primer pair
var2csa
Day 0
4 Weeks
No Choline, Depleted Serine
PF3D7_0421100
var2csa
Day 0
2 Weeks
4 Weeks
No Choline, Excess Serine
PF3D7_0421100
var2csa
Day 0
2 Weeks
4 Weeks
Excess Choline, Depleted Serine
PF3D7_0421100
var2csa
Day 0
2 Weeks
4 Weeks
1x Choline, Depleted Serine
PF3D7_0421100
var2csa
Day 0
2 Weeks
4 Weeks

2.4 Prolonged conditions of excess methionine induces *var* gene switching

Lipid synthesis pathways, utilizing choline or serine, intersect with SAM metabolism indirectly. Methionine, taken in by *P. falciparum* either from human serum or the digestion of hemoglobin, is used directly by the parasite to synthesize SAM (**Figure 8**). Earlier experiments in which parasites were cultured in conditions of excess choline, thought to elevate SAM pools relative to conditions of excess serine, induced a coordinated *var* switch at the population level over time (**Figures 9 & 10**). Subsequently, conditions of excess methionine and depleted methionine were tested to see if similar effects on *var* expression would be observed. For these experiments, MIM was made up so only methionine was depleted. As with serine, methionine is present in hemoglobin; therefore, it can only be depleted in the growth medium, and cannot be completely left out of the culture. Bulk *var* expression was analyzed at two and four weeks by qPCR and results are shown in **Figure 11**. Under conditions of depleted methionine, no *var* switching was observed apart from background (that observed in control conditions of MCM). When excess methionine was present (supplemented following **Table 1**) a coordinated *var* switch was again observed at the population level over time, similar to the conditions of excess choline. The results for excess methionine presented in **Figure 11** are representative of one replicate. This entire assay was repeated in a different clone with similar results (shown in **Appendix**). These experiments, coupled with those altering levels of choline and serine, suggest that conditions where SAM pools are thought to be elevated are capable of accelerating a switch in *var* expression at the population level.

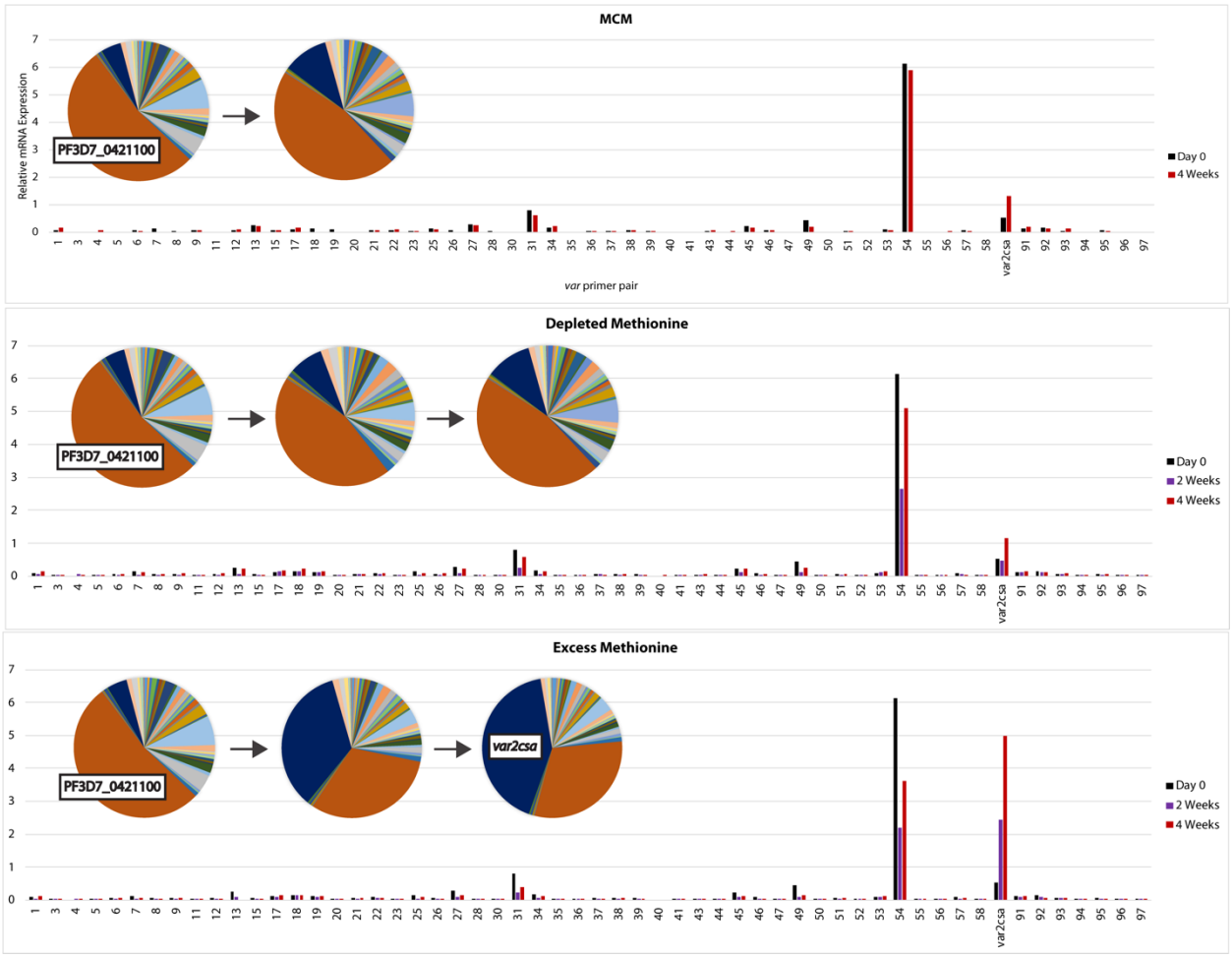


Figure 11. Elevated levels of methionine in the growth media of *P. falciparum* induces a switch in *var* gene expression at the population level over time *var* gene expression as assayed for each condition is shown as a pie-chart and bar graph, with each slice of the pie and each numbered primer pair from Salanti et al. 2013 on the x-axis representing the fraction of the total *var* mRNA pool transcribed from each *var* gene. Black bars represent Day 0 timepoint, or the *var* expression of the parent culture before it was split across different nutrient conditions. Purple and red bars are the *var* expression at 2 and 4 week timepoints, respectively.

2.5 Culturing *P. falciparum* parasites in altered amounts of choline, serine, and methionine does not significantly affect growth rate

Given the key role of choline in membrane biogenesis (**Figure 8**), I investigated whether culturing parasites in excess levels of choline or serine could impact overall growth rate. Following the growth assay procedure detailed in **Methods**, parasites were grown in the conditions presented in **Figures 9 and 10** for at least 14 days. Plotting growth on a log scale in **Figure 12,** no measurable growth phenotype was observed. Similarly, for the altered methionine conditions tested in **Figure 11**, no growth phenotype was observed as shown in **Figure 12**. **Figure 12** shows representative results for one assay; replicates and log growth numbers are shown in **Appendix**.

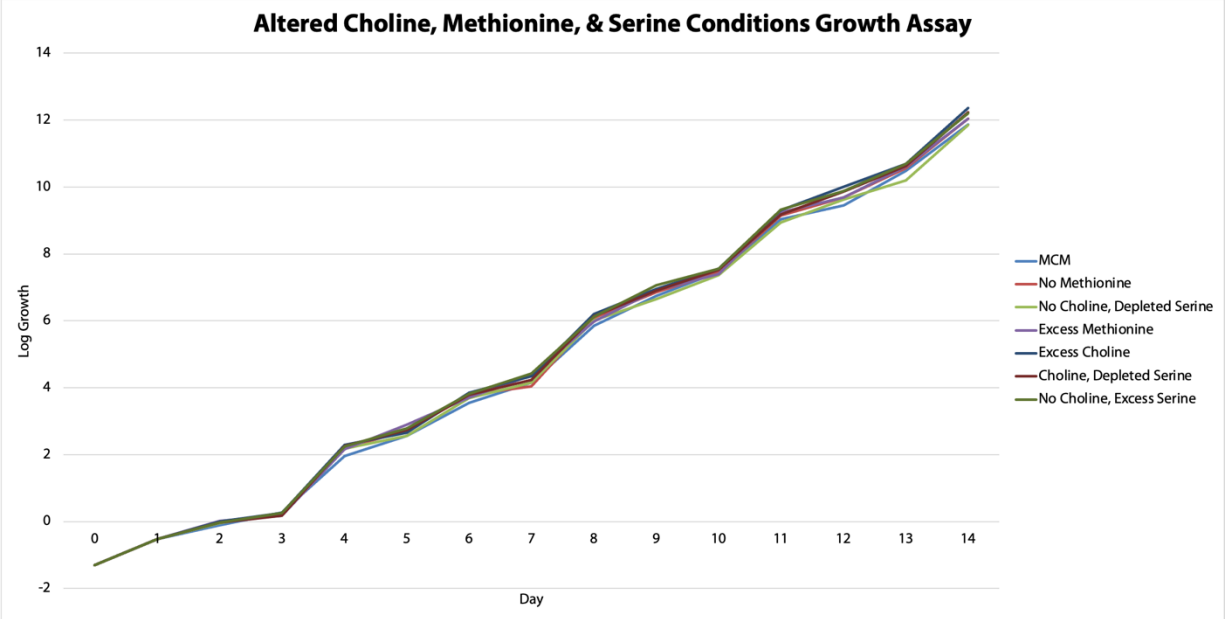


Figure 12. Culturing *P. falciparum* in differing amounts of choline, methionine, or serine does not significantly affect their growth rate Parasites were cultured in the conditions tested over the course of 14 days (x-axis). Parasitemia was measured by flow cytometry and log growth (y-axis) was calculated following the protocol in **Methods.**

Discussion

These experiments provide the first examples of a coordinated *var* switch in a parasite population in response to altered nutrient conditions. Two conclusions can be drawn from these initial experiments: first, it is clear that growing cultures in a serine deprived background with supplemented choline or under conditions of excess methionine lead to *var* switching in this parasite line. Second, at the population level, a significant number of parasites across these cultures independently switched expression from one *var* gene to *var2csa*. (**Figures 9-11**) Thus, in separate experiments where epigenetic regulation of the *var* gene family was destabilized (earlier in Ukaegbu et al., 2015[51], and here where environmental conditions were altered), *var* gene switching was observed at the population level and, in these instances, the switch was to *var2csa*. One important distinction, however, is that the modifications applied in Ukaegbu et al.,2015 lead to increasing and "permanent" expression of *var2csa* in culture; or, so long as the treatments are applied, the parasites are "stuck" expressing *var2csa* and, even over long periods of time, do not switch away from it (**Figure 13**). In contrast, varying environmental levels of choline, serine, or methionine lead to a more transient expression of *var2csa* within the parasite population, as over time levels of *var2csa* expression increase and decrease. (**Figures 9-11**) This transient phenotype is consistent with the idea that metabolic pathways are highly plastic; in response to nutrient depravation, organisms can often compensate for the missing metabolite(s) using alternative metabolic pathways. [62,78,83] A second important distinction is that in Ukaegbu et al., 2015, interference with epigenetic regulation was achieved using a dominant negative HMT construct and chaetocin, conditions that do not naturally occur and are never present in clinical *P. falciparum* cases. Conversely, fluctuations in environmental conditions, like nutrient availability, are common occurrences in biological systems. Thus, these experiments phenocopied the changes in *var* gene expression shown in Ukaegbu et al., 2015 under more biologically relevant conditions. (**Figure 13**). Using a growth assay, I verified that the altered nutrient conditions used in these long-term experiments do not meaningfully affect parasite growth rate (**Figure**

12). Therefore, it can be ruled out that the observed changes in *var* expression were the result of a global stress response to nutrient deprivation or accelerated growth.

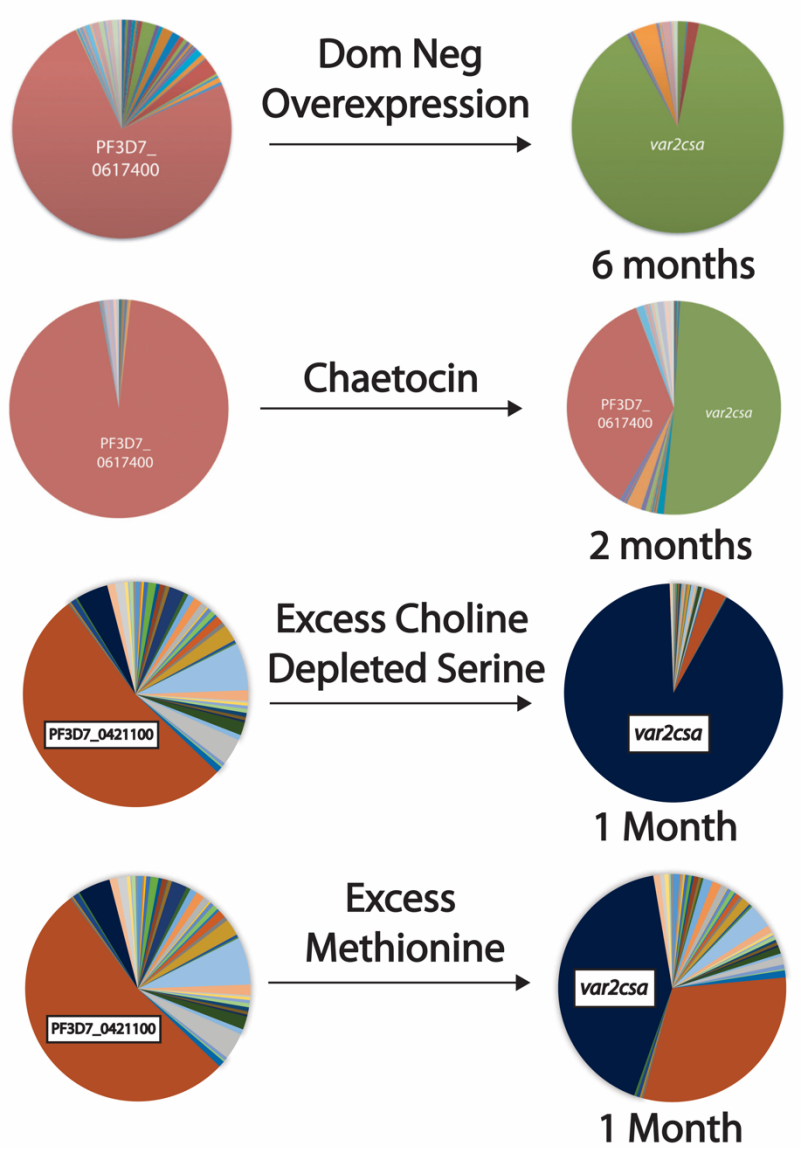


Figure 13. Altered nutrient conditions phenocopied earlier changes in *var* expression growing parasites in conditions of excess choline and depleted serine or excess methionine led to a coordinated switch in *var* expression at the population level to *var2csa***.** This result phenocopies those seen in Ukaegbu et al., 2015 in response to genetic or chemical modifications of HMT activity. First two rows of *var* panels taken from Ukaegbu et al., 2015.

The effect of these specific conditions ties back into the essential and highly used phospholipid metabolic pathways of *P. falciparum*. Choline taken in by the parasite is used in the CDP-choline pathway to make PtdCho, the primary lipid found in the membranes of most eukaryotes including *P. falciparum*. Studies using radiolabeled serine and ethanolamine have demonstrated that parasites can also synthesize PtdCho from non-choline sources using the SDPM pathway, which forms PtdCho from the methylation of p-Etn requiring three SAM (**Figure 8**). [77,78] In yeast, Ye et al. 2017 have shown that there is direct link between SAM and histone methylation by demonstrating that histones can serve as a "methyl sink", or become hypermethylated, in cases where SAM accumulates in the cell (such as when key methylation pathways requiring SAM as the methyl donor are not active). [67] A second study, Mentch et al. 2015, identified that methionine metabolism is sufficient to determine levels of histone methylation by modulating levels of SAM and SAH *in vitro* and *in vivo* mice studies. [58] Taken together, histone methylation dynamics appear to be sensitive to fluctuations in the intracellular ratio of SAM to SAH. Interpreting these observations with the results presented in this chapter, I hypothesize that altering the levels of choline, serine, and methionine in parasite culture led to fluctuations in the parasite's intracellular SAM pool, resulting in changes in the level of histone methylation and, consequently, changes in *var* gene expression. Specifically, the changes that induced a switch in *var* expression (excess choline and excess methionine) would be hypothesized to increase SAM levels following **Figure 8**. In both cases, this presumed relative increase in SAM/SAH induced a coordinated, population-level switch to *var2csa.* A possible model from these results is shown in **Figure 14.**

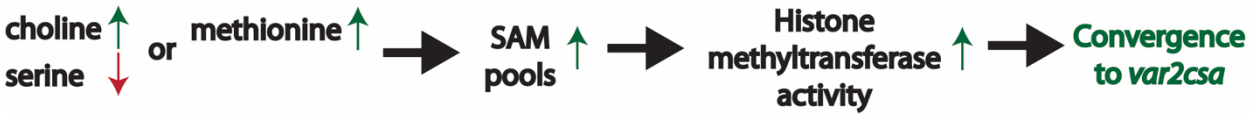


Figure 14. Hypothesized model for the link between intracellular SAM pools and *var* gene expression in response to varying levels of choline, methionine, and serine Increasing choline availability to *P. falciparum* allows for synthesis of key membrane components without consuming SAM through the CDP pathway (Figure 8), this would be hypothesized to elevate levels of SAM/SAH. Increasing methionine availability directly would be hypothesized to increase SAM/SAH, as it is used to synthesize SAM. This, in turn, allows for greater methyl donation to HMTs and increased deposition of epigenetic marks. This destabilization of the epigenetic regulatory network increases rates of *var* switching specifically to *var2csa*.

Across independent parasite cultures in the three experimental approaches presented in **Figure 13**, *var2csa* expression was induced. As detailed in **Chapter 1**, *var2csa* is a unique member of the *var* family for several reasons. In particular, *var2csa* is universally conserved across parasite isolates, while the majority of *vars* are not - despite encoding a variant of PfEMP1 that exclusively binds a placental ligand. While the niche its encoded protein can be used in is small, its genomic organization allows it to be transcriptionally active while translationally repressed, and transcripts have been detected in non-pregnant individuals. [84,85] Furthermore, cultures grown without selection for long periods of time were found to display a high activation rate of *var2csa*, suggesting that expression of *var2csa* might represent a "default" state. [50] These properties of *var2csa* coupled with the results of these three experiments support the idea that *var2csa* might serve a separate function within *P. falciparum* apart from encoding a form of PfEMP1. Notably, studies have found elevated levels of choline in the placenta[115,116]. Mathematical models of *var* gene switching predict the presence of transiently active genes, or "switch intermediates" that add a level of uniformity and coordination to the process. [55] These results make it tempting to imagine that *var2csa* might indeed have a separate function within the parasite, possibly serving as the presently unidentified "switch intermediate," though this (or the identity of any sink node) has not yet been established. [55] In the control lines (MCM or depleted serine, no choline and depleted methionine) shown in **Figures 9-11,** *var2csa*

can be observed at lower levels than supplemented choline and methionine cultures. Previously, Mok et al. 2008 observed that *P. falciparum* parasites grown in culture have a tendency to activate *var2csa* over time in the absence of any known selective pressures[50]. These experiments show that conditions of excess choline or excess methionine in the growth media are capable of accelerating this convergence above this "background" level of *var2csa* induction over many independent experiments (**Figures 9-11, Appendix**). Precisely why parasites will slowly converge to *var2csa* in culture under standard growing conditions remains to be described. However, if *var2csa* is indeed serving as the "sink node" in a *var* switching network, one could imagine that parasites may drift into *var2csa* over time in an environment with no immune pressure. This thought will be expanded upon in **Chapter 5: Perspectives**.

The results presented in this chapter directly challenge the commonly held paradigm that antigenic variation follows an intrinsic, programed switch rate by instead suggesting that this organism possesses the ability to switch in response to environmental changes. Fluctuations in environmental nutrient availability are likely very common. However, it is unlikely that these fluctuations are constant over long periods of time, as were the conditions in these experiments. Instead, which external stimuli capable of triggering *P. falciparum* to switch antigen expression could these conditions be mimicking? The results in this chapter add to a growing body of research on the topic of environmental signals altering gene expression in Plasmodium species, as previously described in **Chapter 1.11**.

Taking the model in **Figure 14** a step further, I hypothesize that *P. falciparum* can sense when the host immune system first begins to recognize it via environmental cues resulting from antibody recognition, and respond by switching *var* gene expression. If supported, this would describe a mechanism by which parasites can switch expression of *var* genes exactly when needed, with perfect timing, thus allowing them to utilize their limited repertoire of *var* genes with the upmost efficiency. These results would also suggest a possible molecular mechanism that underlies the clinically observed events illustrated in **Figures 4 and 5**. This novel possibility will be investigated in **Chapter 4** of this thesis.

Chapter 3

Altering Expression of PfSAMS and PfSAHH Affects *var* Switching and Expression

Background

Cellular decision-making occurs through signal transduction, wherein information from and about the cell's environment is propagated through intracellular networks and conveyed as reversible modifications on proteins. These networks rely on numerous regulatory mechanisms specific to each pathway, such as cascades and feedback loops, that interconnect outside conditions with the intracellular metabolic state. Metabolite sensing allows cells to appropriately respond to changes in their environment through cell signaling and adjustments in gene expression. [62,86] Recent technological and analytical advances have invigorated efforts to study the intricacies of cellular metabolism. These developments have been of particular importance in the field of cancer research, as defects in the regulation of mammalian cell growth and homeostasis are known features of rapidly proliferating cancer cells. [62] Notably, recent work has demonstrated that changes in nutrient uptake can alter gene expression through one-carbon and SAM metabolic pathways.

Epigenetic regulation of gene expression refers to heritable changes in transcription that occur not through alterations to the DNA sequence, but through alterations in chromatin structure from the methylation or acetylation of histones. [31] Given that metabolites are the substrates used in generating epigenetic modifications, an association exists between epigenetics and metabolism. [59] One-carbon metabolism is a metabolic network that utilizes nutrients, such as glucose, vitamins, and amino acids from the environment to fuel multiple biological functions, including the methylation of SAM through the methionine cycle. [57] SAM is the universal methyl donor for HMT enzymes, which ultimately deposit the methyl groups on histones (**Figure 7**). Changes at specific sites, or marks, on histones alter the chromatic state and subsequently cause changes in gene expression; repressive chromatin, or heterochromatin, is compact, whereas relaxed chromatin, or euchromatin, is open allowing for easy transcription.

Several studies in eukaryotic systems have provided evidence for a direct link between nutrient uptake, SAM metabolism, histone methylation, and gene regulation. In cases where the availability of environmental methionine was altered, changes in histone methylation at the H3K4 mark were observed alongside changes in the level of SAM. [58,63,64] Additionally, under conditions of excess cellular SAM (or a high SAM/SAH ratio), histones can serve as "methyl sinks" where they accept the extra methyl groups. [66,67] Taken together, the results of all of the aforementioned experimental conditions suggest that histone methylation dynamics appear to be sensitive to fluctuations in the intracellular ratio of SAM to SAH.

Canonical enzyme kinetics apply to those controlling SAM metabolism. HMT activity depends on the concentration of its substrate, SAM; evidence suggests that the SAM/SAH ratio plays a role in regulating HMT activity, as SAH is a known inhibitor of HMTs. [60,61] In contrast with protein phosphorylation kinetics, where the concentrations of ATP (adenosine triphosphate) substrate far exceed the Michaelis constant (K_m), HMT K_m values lie in the range of intracellular SAM concentrations. This further suggests that even minor fluctuations in SAM levels could significantly affect HMT activity, increasing or decreasing histone methylation and providing a direct link between it and cellular metabolism through SAM. [62] [28,57]

In response to changing growth environments, *P. falciparum* has evolved the ability to vary its transcriptional programming from parasite-to-parasite through the epigenetic plasticity of clonally variant gene expression. This includes the *var* gene family that orchestrates antigenic variation by encoding different variants of a cytoadhering protein on the RBC surface. While other classes of clonally variant gene families have been observed to have modest but direct transcriptional responses to temperature, metabolite fluctuations, and drug pressure, this has not yet been documented in *var*s. [27,69–71] Consistent with their role in immune evasion, *var* genes are expressed in an epigenetically regulated, mutually exclusive manner. Three HMTs govern the deposition of histone marks at *var* genes. PfSET2 and PfSET3 deposit the H3K36me3 and H3K9me3 marks, respectively. [37,39,40] H3K9me3 is devoted specifically to regulating genes that display variable expression in *P. falciparum,* as it was found to

be distributed within the narrow regions of the genome where these genes reside. Therefore, this mark is found at all the silenced *var* genes. [29,33] The H3K36me3 mark is similarly found in regions of clonally variant gene families, but at both active and silent *var* genes. [37] [38] [31] PfSET10 was found to deposit the H3K4me3 mark specifically around *var* genes[38] The key role these HMTs play in *var* gene regulation, and thus the parasite's pathogenicity, ties antigenic variation to SAM metabolism.

Over the course of its 48-hour intraerythrocytic life cycle, *P. falciparum* replicates and divides into as many as 36 daughter merozoites. This process, requiring large amounts of new membranes, increases the total phospholipid content of the iRBC approximately 6-fold. [76] The rapid growth and multiplication within the iRBC is made possible by nutrients supplied by the host, with fatty acids, choline, serine, and ethanolamine of primary importance in phospholipid synthesis. [78] Lipid metabolic pathways fuel not just the membrane biogenesis required for growth and expansion, but also facilitate various intracellular signaling, protein trafficking, and hemoglobin degradation functions. Therefore, it is not surprising that the uptake, transport, and metabolism of lipids are essential for the asexual viability of *P. falciparum.* [78,81] As detailed in **Chapters 1 and 2**, these pathways intersect with intracellular SAM pools that power HMT reactions. The enzyme PfPMT, which allows for the input of serine (in instances of limiting exogenous choline) in synthesizing key membrane component PtdCho through the SDPM pathway, catalyzes a three-step methylation reaction that is dependent on SAM as a methyl donor (**Figure 8**). [77,78] In the previous chapter, I demonstrated that altering the levels of choline and serine precursors in the culture medium within physiologically relevant concentration ranges leads *P. falciparum* parasites to switch *var* expression at the population level over time. These results phenocopied those of earlier experiments where the activity of PfSET2 and PfSET3 were genetically or chemically modified. [51] Considering these results alongside what is known about lipid metabolism in *P. falciparum*, I proposed a model where the level of SAM pools affects *var* switching by modulating HMT activity (**Figure 14**). While both pathways run simultaneously, conditions of excess choline to depleted serine may allow the parasites to use the CDP-choline pathway, rather than the SDPM pathway,

as a way to synthesize PtdCho without depleting SAM pools (**Figure 8**). [77] Supporting this is the finding that increasing amounts of choline in the culture medium decreases the amount of PfPMT transcript and protein in a dose-dependent manner, and that PfPMT activity is inhibited by its product, p-Cho. [77,82] Conversely, when lipid compound LysoPC, which is immediately metabolized to p-Cho upon uptake, was depleted, PfPMT transcription was highly induced. [73] Thus, in circumstances of excess choline, the parasite may have an overall higher ratio of SAM/SAH by decreasing PfPMT activity. As mentioned above, studies related to other organisms have found that when the ratio of SAM/SAH is high, SAM tends to offload methyl groups to "methyl sinks" such as histones through HMT. [58,67] Taken together, I hypothesize that conditions of excess choline to depleted serine, and excess methionine, specifically lead to *var* switching by increasing SAM pools, which in turn donate their extra methyl groups to HMTs, depositing them on histones and epigenetically modifying (*var*) gene expression. (**Figure 14**)

In *P. falciparum,* SAM synthetase (PfSAMs) catalyzes the formation of SAM from methionine and ATP, while SAH hydrolase (PfSAHH) catalyzes the reversible hydrolysis of SAH to adenosine and homocysteine (**Figure 8**). In this chapter, I will describe a set of experiments designed to test the role SAM and SAH play in this model, by genetically varying the expression levels of these enzymes in live parasites.

Results

3.1 Rationale and generation of transfected lines

To more directly test the hypothesized model that *var* switching is affected by conditions thought to alter the levels of SAM/SAH, I generated overexpression constructs applying the RNA aptamer system adapted for *Plasmodium* by Ganesan et al. 2016[87] to selectively overexpress PfSAMS or PfSAHH from an episome (or in addition to that from the endogenous, chromosomal locus). This aptamer system employs Tet repressor protein (TetR) aptamers placed in the 3' untranslated region (UTR) of the transcript, allowing for the regulation of protein expression at the mRNA level. *P. falciparum* translational regulation protein DOZI (PF3D7_0320800) is fused to

TetR, allowing for regulatory control from the aptamers beyond that attainable with TetR alone. In the presence of anhydrotetracycline (ATc), molecules competitively bind to TetR-DOZI, inhibiting its binding to the aptamers and allowing translation to proceed[87]. (**Figure 15A**). The specific vector used here (pMG68 supplied by Dr Jacquin Niles) expresses an epitope-tagged (HA-tagged) gene of interest under control of a calmodulin (PF3D7_1434200) promoter while also expressing the *blasticidin S deaminase (bsd)* gene to enable selection of transfected parasites. The calmodulin gene promoter is active throughout the asexual replication cycle, ensuring that the expression of the genes of interest from the episome are not limited by parasite growth stage.

Episomal expression, as derepressed by the addition of 0.5 mM ATc following Ganesan et al. 2016[87], of PfSAMS-HA was verified by western blot (**Figure 15B**). Constructs were transfected into the same 3D7 parent clone from the experiments described in **Chapter 2**. A faint band seen for those parasites off ATc was taken to be leaky expression coming from the plasmid. I was unable to visualize PfSAHH-HA by western, so overexpression of SAHH was confirmed by qPCR on cDNA (**Figure 15C**). Again, leaky expression was observed from the episome as represented by a higher level of total SAHH mRNA expression in the ATc “off” transfected lines relative to wildtype (untransfected) 3D7 parasites. These lines will be referred to as the SAMS and SAHH OE (overexpression constructs).

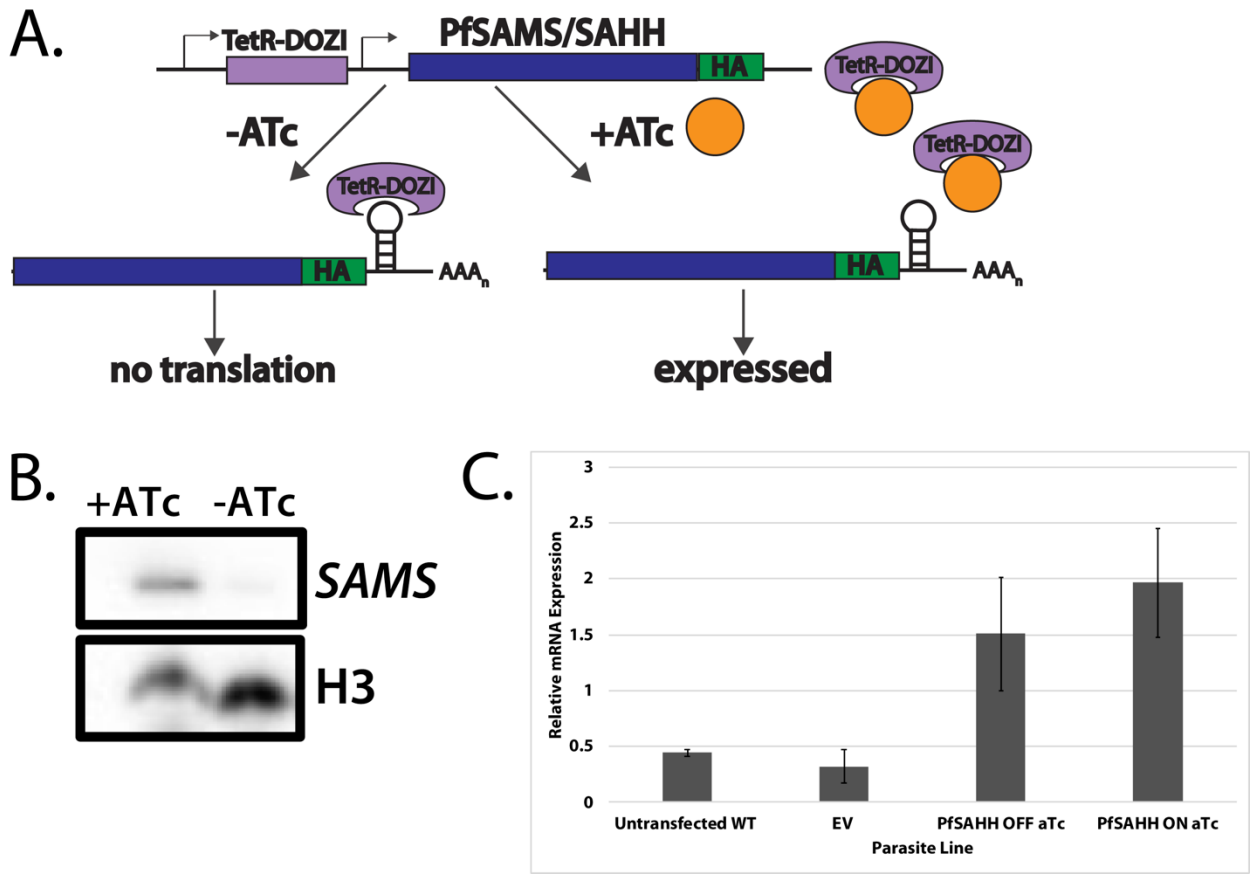


Figure 15. Overexpression lines for PfSAMS and PfSAHH were generated using the The TetR-DOZI aptamer system A. The aptamer system regulates expression at the mRNA level using Tetrepressor protein (TetR) aptamers placed in 3'UTR. In the presence of anhydrotetracycline (ATc), molecules competitively bind to TetR-DOZI fusion protein, inhibiting its binding to the aptamers and allowing translation to proceed. **B.** Western blot showing overexpression of HA tagged PfSAMS in the presence ("on"/+) or absence ("off" or -) of 0.5 mM ATc after one full growth cycle. Leaky expression occurs in the absence of ATc. **C.** PfSAHH overexpression as confirmed by qPCR. Primers (listed in **Methods**) recognizing SAHH were used on cDNA from untransfected WT, EV, and PfSAHH OE lines. Each bar represents the average SAHH mRNA expression level, normalized to seryl-tRNA synthetase expression, across n=4 samples for WT and EV controls and n=6 samples for both PfSAHH ON and OFF ATc. Error bars represent the standard deviation.

The experiments presented in **Chapter 2** support a model wherein the continuous application of conditions hypothesized to increase the intracellular ratio of SAM/SAH (or an increase the methylation index) within *P. falciparum* parasites drives a switch in *var* expression at the population level over time, specifically to *var2csa.* To test if decreasing the methylation index (through a lowering of the SAM/SAH ratio) would have the opposite effect, I also assayed the *var* profiles of three clones of a PfSAMS knockdown (KD) line, which was developed by Chantal Harris in NF54 *P. falciparum* parasites. In brief, this knockdown line was achieved through integration of the glmS ribozyme and neomycin selectable marker between the coding region of PfSAMS and the 3' UTR, allowing for post-transcriptional regulation of gene expression as previously described in Prommana et al. 2013[88]. When glucosamine (GlcN, 2.5mM) is added, the ribozyme is cleaved leading to degradation of the mRNA (**Figure 16A**). Across three clones (n=3) expression of PfSAMS was found to be reduced by about half after application of GlcN (2.5mM) relative to those off GlcN (0 mM) (**Figure 16B**).

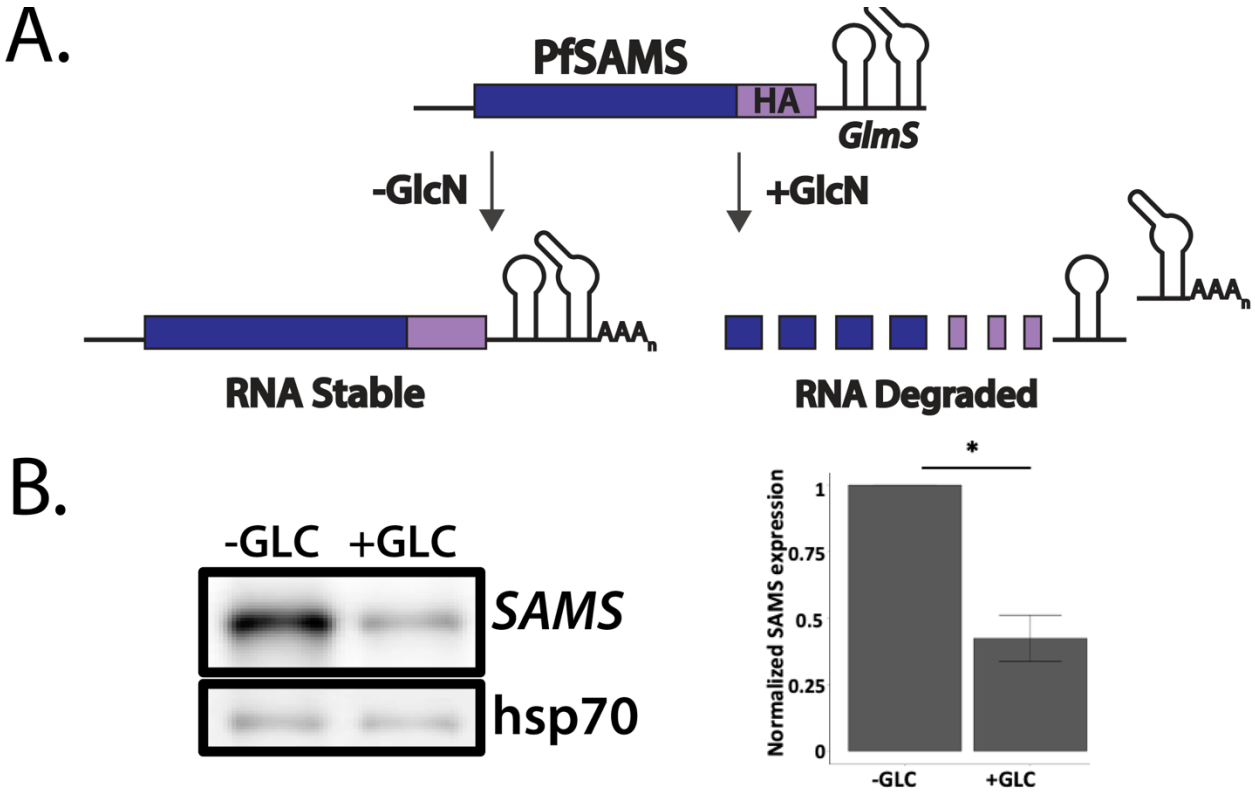


Figure 16. Integration of the GlmS ribozyme allows for a PfSAMS knockdown A. PfSAMS knockdown line was achieved through integration of the glmS ribozyme and neomycin selectable marker between the coding region of PfSAMS and 3'UTR, allowing for post-transcriptional regulation of gene expression as previously described in Prommana et al. 2013. When glucosamine (GlcN) is added, the ribozyme is cleaved leading to degradation of the mRNA. **B.** Western blot showing expression of HA tagged PfSAMS in the presence ("on"/+) or absence ("off" or -) of 2.5mM GlcN after one full growth cycle (Left). Quantification of expression level from blot (Right). Both done by Chantal Harris.

3.2 Increasing expression of PfSAMS and PfSAHH induces a coordinated *var* switch

PfSAMS and PfSAHH transfected lines were assayed for *var* expression “on” and “off” ATc, alongside an empty vector (EV) line serving as a control. Both PfSAMS and PfSAHH lines had switched *var* expression markedly from the parent population at the first time point taken as soon as cultures grew to a sufficient parasitemia (“Day 0” timepoint, **Figure 17**, likely a result of the leaky expression coming from the episome **Figure 15B**). HMT K_m values suggest that even minor fluctuations in SAM levels can significantly affect HMT activity[28,57,62], supporting that the leaky level of episomal expression was enough to robustly alter the epigenetic control of *var* genes. Notably, as seen in the experiments from **Chapter 2**, this switch was coordinated at the population level to *var2csa.* The EV control line showed no difference in *var* expression after application of ATc (**Figure 17**). Results presented in **Figure 17** are representative of two replicates shown in full in **Appendix**.

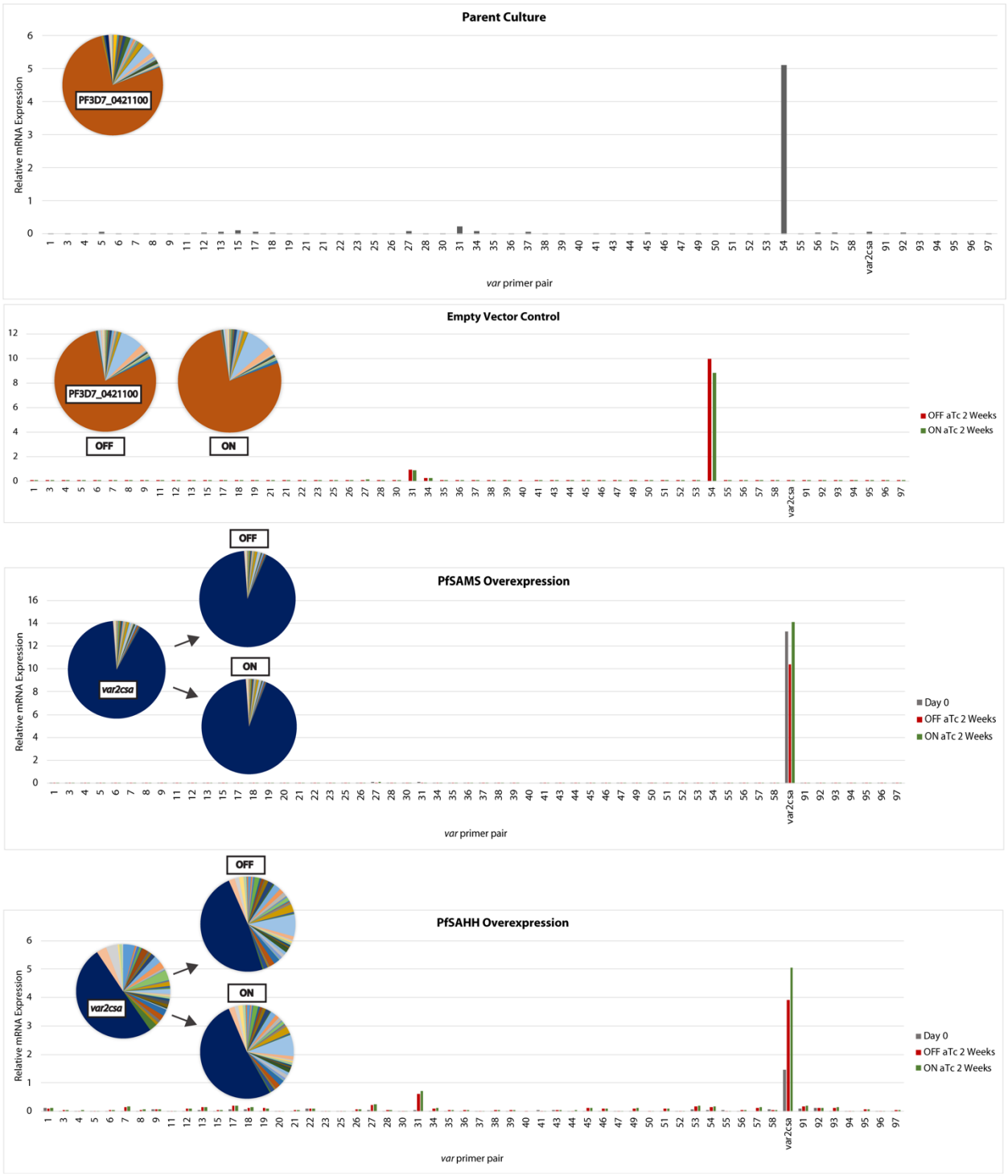


Figure 17. Increasing the Expression of PfSAMS or PfSAHH induces a *var* gene switch at the population level Representative results of two independent experiments are presented. Each pie chart and bar graph illustrates *var* gene expression across the parasite population (3D7 parent line) at two weeks growth ON (green bars) or OFF (red bars) ATc. Empty vector plasmid was transfected as a control. Parasite population before transfection shown as parent culture. Day 0 (gray bars and left pie chart) represents the earliest timepoint at which RNA could be extracted after transfection.

3.3 Decreasing expression of PfSAMS does not induce coordinated *var* switching

All three PfSAMS KD clones showed the same non-clonal *var* expression pattern (high expression levels of six *var* genes: PF3D7_1000100, PF3D7_0412400, PF3D7_0412700, PF3D7_063250, PF3D7_0809100, and PF3D7_0600200) at the first time point taken as soon as cultures grew to a sufficient parasitemia ("Day 0" timepoints, **Appendix**).

Representative results for one PfSAMS KD clone are pictured in **Figure 18,** with the others in **Appendix.** Growth over a four-week period either on or off 2.5mM GlcN did not induce a switch in *var* expression across any of the three clones. In contrast to the SAMS and SAHH OEs, the SAMS KDs do not converge to *var2csa* over time, but do maintain high expression of the six *var* genes on Day 0. Interestingly, low-level expression of multiple other *vars* can be seen coming up in the background over the course of the experiment for the cultures on 2.5mM GlcN.

To ensure that the addition of GlcN doesn't alter *var* expression on its own, a wildtype (untransfected) line with a similarly heterogenous *var* expression profile was grown in parallel, on and off the same concentration of GlcN (**Figure 18, bottom**). No observable change in *var* expression at the population level was observed.

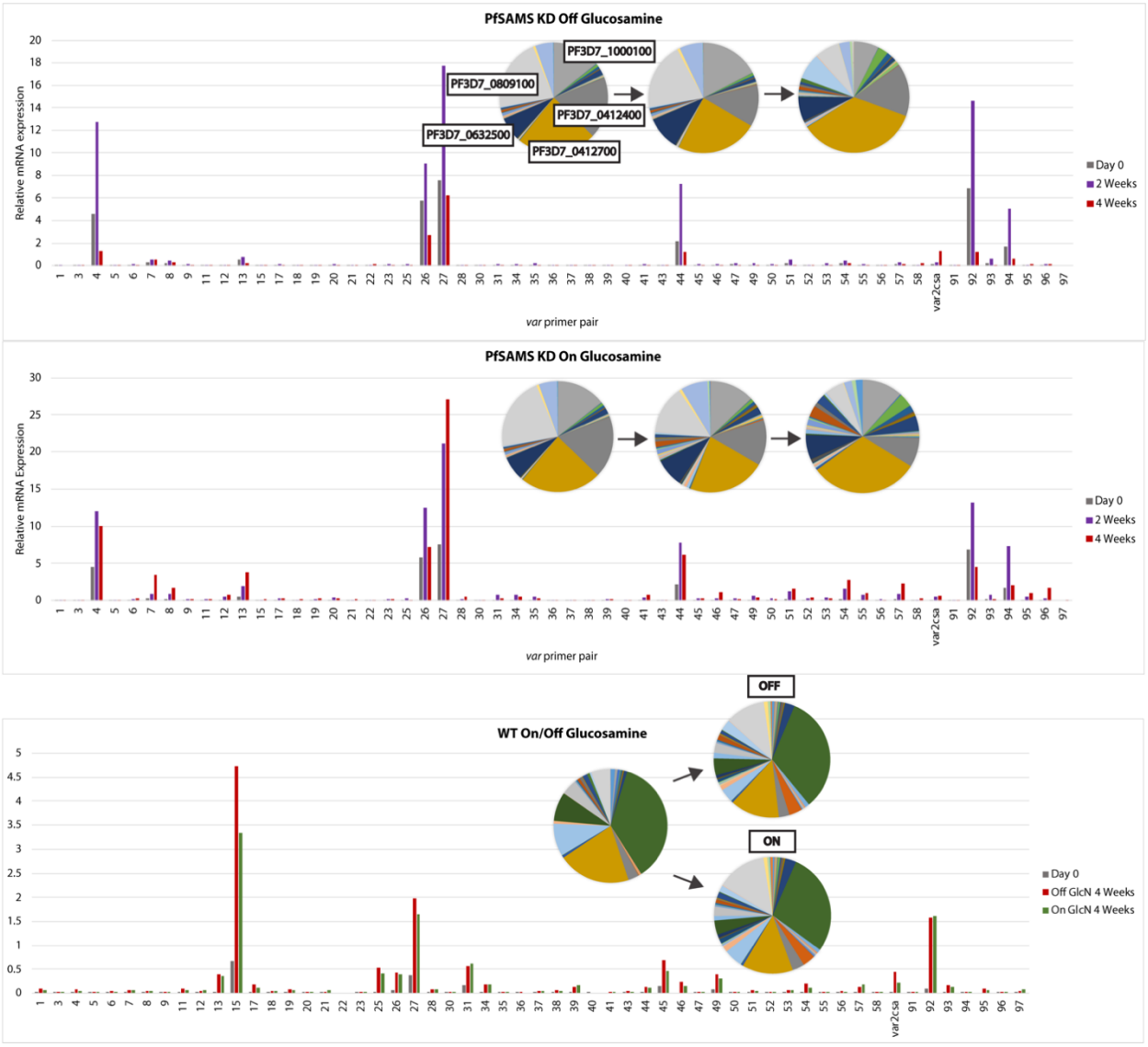


Figure 18. Decreasing expression of PfSAMS does not induce a coordinated *var* switch at the population level Representative results of three clones are presented. Each pie chart and bar graph illustrates *var* gene expression across the parasite population (NF54 parent line) at 2 and 4 weeks growth ON (top chart) or OFF (middle chart) 2.5 mM GlcN. Day 0 (gray bars and left pie chart) represents the earliest timepoint at which RNA could be extracted after transfection. 3D7 WT line (bottom chart) was grown on GlcN as a control with the last timepoint (4 weeks) shown.

3.4 Altering expression of PfSAMS and PfSAHH slightly affects growth

Given the key roles SAMS and SAHH play in cellular metabolism, and, ultimately, gene expression, I tested whether altering the expression of these enzymes in *P. falciparum* would alter its growth. Following the protocol outlined in **Methods**, I ran a series of growth assays with the PfSAMS/SAHH OEs and the PfSAMS KD on and off ATc and GlcN, respectively. The results pictured in **Figure 19A** are representative of three independent replicate assays (shown in full in **Appendix**). Across the three replicates, the PfSAHH OE, either on or off ATc, has a slight negative growth phenotype, while the PfSAMS OE does not grow at a visibly different rate than untransfected WT parasites or the EV transfected line. Adding 0.5 mM of ATc does not seem to significantly alter growth on its own.

The results for two PfSAMS KD clones are shown in **Figure 19B.** Interestingly, the KDs have a positive growth phenotype, growing at a faster rate than an untransfected WT line. Once the KD is induced through the addition of 2.5mM GlcN, the clones grow similarly to the WT. Putting the WT on GlcN causes a slight negative growth phenotype. Result pictured is representative of two replicate assays, shown in full in **Appendix**.

3.5 A rescue phenotype is observed after growing the PfSAMS and PfSAHH overexpression lines off drug selection

The PfSAMS and PfSAHH plasmid carries the *Blasticidin S deaminase* gene, allowing for selection on blasticidin antibiotic in *P. falciparum* parasites. At the Day 0 timepoint after transfection of both OE constructs, the respective populations were already largely expressing *var2csa* (**Figure 17**), likely due to leaky expression from the episome, even when off ATc (**Figure 15**). Transfection of *P. falciparum* parasites introduces a severe selective bottleneck, as the vast majority of parasites are killed off when the culture is first placed under blasticidin selection pressure. Both lines were grown off blasticidin selective pressure for two months, further supporting that this phenotype was from the OE of either enzyme and not the effect of a major selective bottleneck.

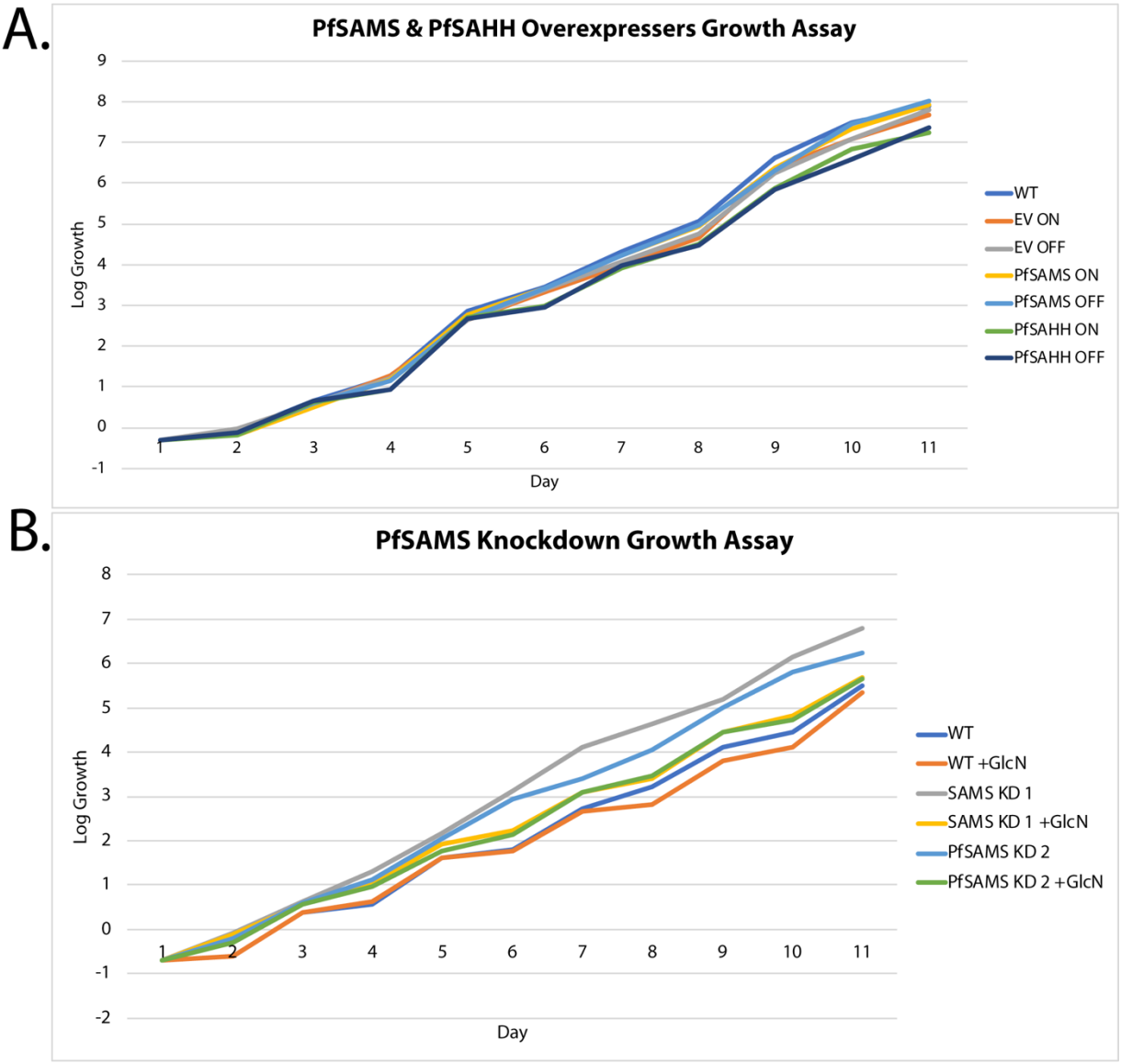


Figure 19. Altering expression of PfSAMS or PfSAHH affects growth PfSAMS and PfSAHH OE lines (**A**) and PfSAMS KD lines (**B**) were cultured in the conditions tested over the course of 11 days (x-axis). Parasitemia was measured by flow cytometry and log growth was calculated following the protocol in **Methods.**

Representative results for two technical replicates are shown in **Figure 20** (the second replicate is shown in **Appendix**). Both lines saw a shift in *var* expression away from *var2csa* when drug selection was removed, suggesting that the stable *var2csa* phenotype was a result of the increased SAMS or SAHH expression from the episomal construct. By growing parasites in the absence of blasticidin selection, parasites shed the episome, thereby reverting to wildtype levels of SAMS or SAHH expression.

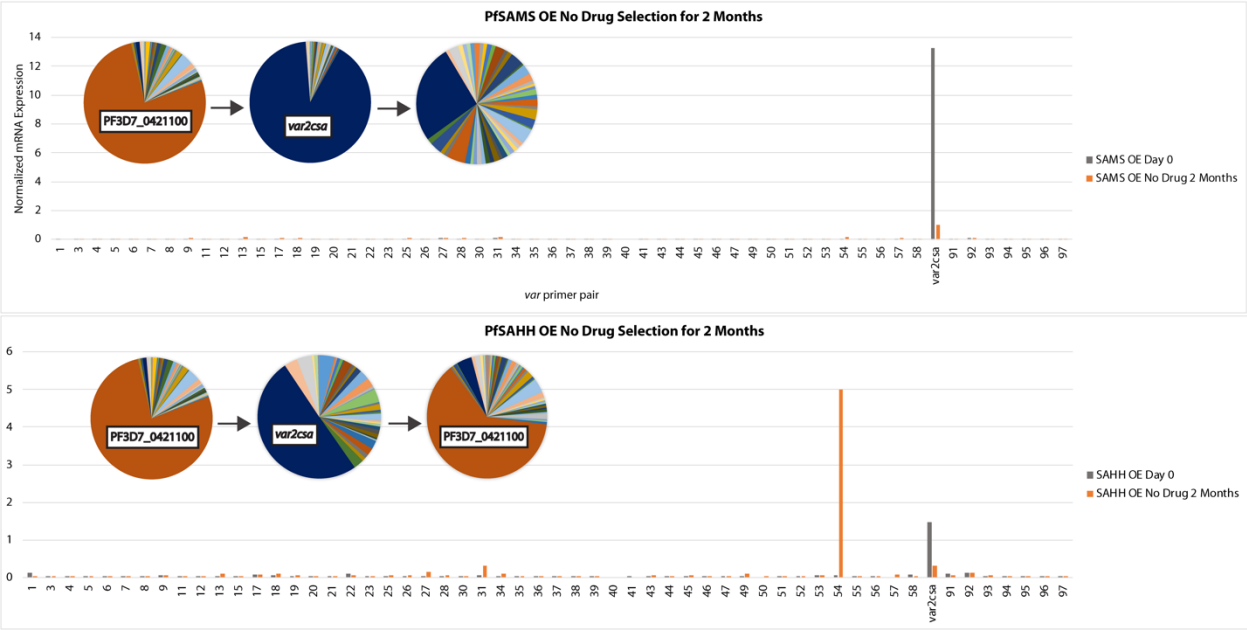


Figure 20. PfSAMS and PfSAHH overexpression lines switch *var* expression when selective drug pressure is removed PfSAMS (top) and PfSAHH (bottom) OE transfected lines were grown in the absence of blasticidin drug for 2 months and their *var* expression observed by qPCR. Day 0 timepoints for the assay are shown as gray bars and as pie charts (middle). Population at the time of transfection shown only as a pie chart (left). 2 month timepoints shown as orange bars and pie charts (right). Representative results from across 2 replicates.

3.6 Rescue of PfSAMS expression in the knockdown line induced a coordinated switch to *var2csa* but it was not replicated

All three independent PfSAMS KD clones were found to have a heterogenous *var* expression profile, displaying high levels of expression for seven *var*s. Inducing the KD with GlcN did not alter this expression profile (**Figure 18**), and did not induce a coordinated switch to *var2csa* (**Figure 17**). For the PfSAMS OE line, robust activation of *var2csa* was observed at the population level at the earliest possible timepoint (**Figure 17**). To see if a switch to *var2csa* could be induced if PfSAMS expression was restored or increased in the KD line, I transfected the PfSAMS OE construct into two of the PfSAMS KD lines. These will be referred to as PfSAMS R(escue)1 and R2.

RNA was extracted for analysis at the earliest feasible timepoint. At this Day 0 timepoint, R1 showed activation of *var2csa* at a visible, robust level, diverging from that of the parental KD line **Figure 21**, top. However, for R2 the *var* expression profile had not noticeably changed from that of the background level seen in the parent KD line **Figure 21**, bottom. Notably lacking the robust activation of *var2csa* seen in R1. Full panels are shown in **Appendix**.

This rescue phenotype, while observed in one line (R1), was not replicated and further work will be required to investigate the differences between these two lines. In order to verify SAMS OE in R2, enzyme expression levels could be quantified by qPCR. Further, investigation into SAM levels resulting from the rescue could be quantified by mass spectrometry. Indeed, one possibility is that the SAMS expression from the endogenous locus and the expression from the episome are not equal, meaning this is not a true rescue. First, the episome uses a different promoter from the endogenous SAMS locus. In addition, the parent line used here (the KD) is modified at the endogenous locus with the GlmS ribozyme and an epitope tag. For a true rescue, one could revert the SAMS locus in the KD lines to WT using CRISPR-cas9.

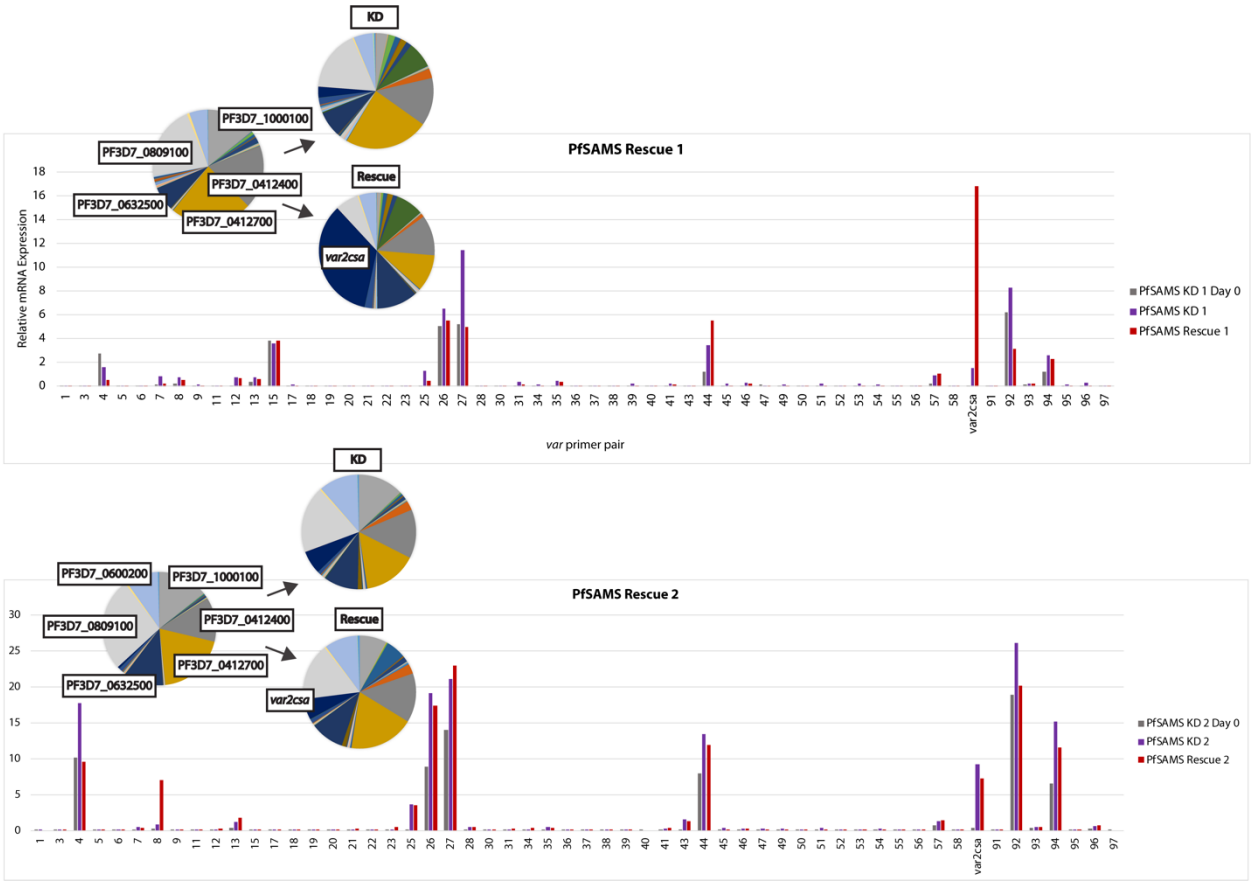


Figure 21. Rescue of PfSAMS expression in the knockdown line induced a coordinated switch to *var2csa* in one transfected line Transfection of the PfSAMS OE episome into KD line 1 (top plot) and KD line 2 (bottom plot) induced *var2csa* expression only in Rescue line 1 (top). Day 0 timepoints for the assay, or the parent KD line before transfection, are shown as gray bars and as pie charts (left). Population post-transfection with the OE episome shown as red bars and pie charts (bottom). Untransfected KD parent lines were extracted for comparison at the same time point and are shown as purple bars and pie charts (top). Full panels are shown in Appendix.

3.7 Cloning out populations of transfected parasite lines gives additional insight into their *var* switching patterns

Changes in *var* switching propensities should be particularly visible when the number of parasites within a population is small, and thus the effect of individual switching events on the total *var* expression profile is amplified. Therefore, by examining populations soon after cloning, it is possible to distinguish the switching dynamic that was in place at or near the moment that the individual parasites were isolated. Colleagues in the lab have extensively experimented with cloning WT, untransfected *P. falciparum* lines. They repeatedly observe that ~30-40% of the clones obtained by limiting dilution (see **Methods**) express a completely different *var* gene from the original parent population, thus reflecting the underlying switching rate of wildtype parasites (Dr. Francesca Florini and Joe Visone, Personal Communication). In addition, they typically recover clonal populations either expressing a single dominant *var* gene or alternatively expressing a very heterogenous mix of *var* genes, with the latter parasites presumably reflecting a population that was actively undergoing *var* gene switching at or near the moment that the founder parasite was isolated.

I applied this method of cloning by limiting dilution followed by *var* expression profiling, examining *var* gene switching patterns for parasites in which I had altered SAMS expression. Here, I cloned populations of the EV, PfSAMS OE, and PfSAMS KD lines in accordance with the cloning protocol outlined in **Methods**. I hypothesized that the EV transfected line would behave similarly to wildtype parasites, as it is simply a control line. In regards to the PfSAMS OE line, I hypothesized, based on the sole and high level of *var2csa* expression, together with my overarching hypothesis from **Chapter 2** (**Figure 14**), that as long as elevated SAMS expression was maintained in the clones, all would be found to continue expressing *var2csa* at very high levels. In contrast, the PfSAMS KD line displayed a heterogenous *var* gene expression profile (**Figure 18**). If this pattern reflects an accelerated switching rate, I would expect all of the clones to similarly display heterogeneous patterns, varying pattern between clones. In contrast, if the PfSAMS KD line has normal *var* gene switching rates, I would expect

to recover a mix of clones: some displaying a single dominant *var,* others displaying a heterogenous profile.

For each population, I analyzed eight individual clones. The clones obtained from the EV line are pictured as pie charts in **Figures 22**. Out of the eight screened EV clones, one had a unique *var* expression profile from the parent clone, manifesting as a heterogenous expression profile (full clone profiles shown in **Appendix**). This rate of switching is somewhat lower than the 30-40% previously observed for WT clones (Dr. Francesca Florini and Joe Visone, Personal Communication); however, it is notable that each of the seven remaining clones had maintained high expression of the dominant "on" *var*, and none had switched to *var2csa* **Figure 22**.

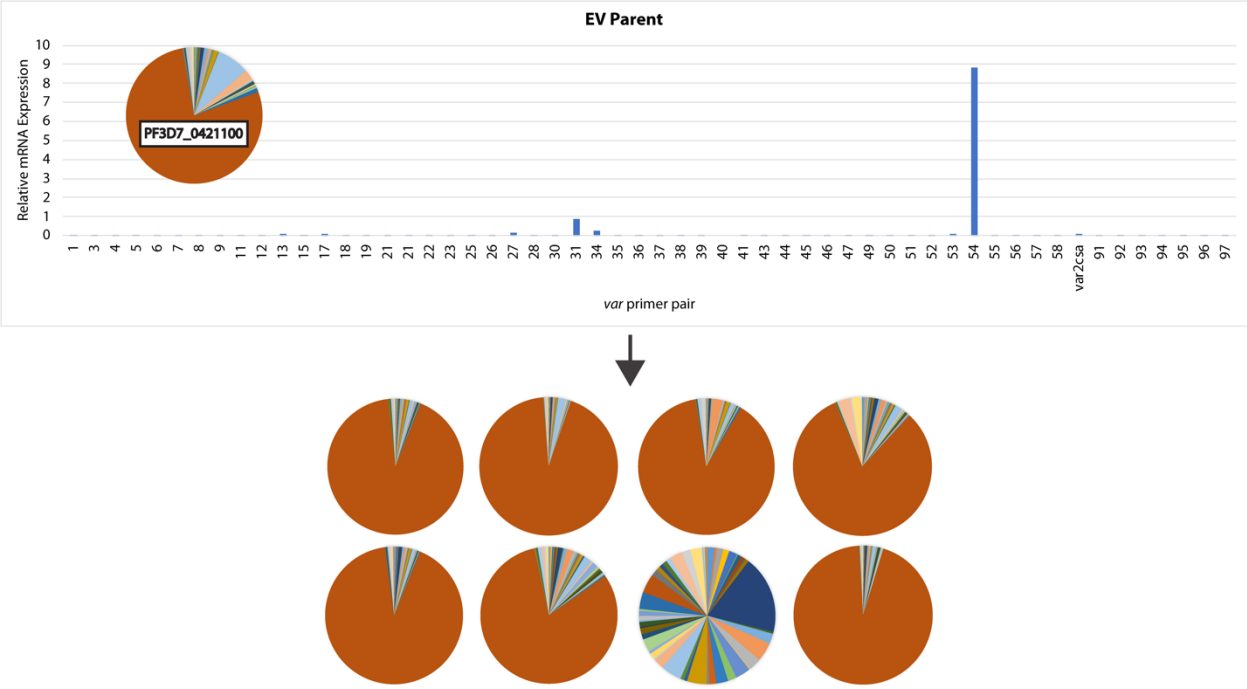


Figure 22. The majority of EV clones retain a homogeneous *var* expression profile matching the parent EV parent line (top) was cloned. The *var* expression for 8 individual clones are shown as pie charts. Within each pie chart, an individual slice represents the relative level of expression for a particular *var* in the population. The clone on the bottom row, second from the right showed a low, heterogenous *var* expression profile. The remaining seven clones all retained high expression of *var* gene PF3D7_0421100.

Analysis of the PfSAMS OE line showed that seven out of eight clones continued to express *var2csa* as the dominant *var* gene, as expected **Figure 23** (full panels shown in **Appendix**). One clone, however, had switched to expressing a heterogenous mix of *var* genes at low levels, indicative of active switching. Thus, I screened an additional six clones (shown in **Appendix**). Of these additional six clones, five continued to express *var2csa* as the dominant *var* gene, while, similarly, one expressed a heterogenous mix of *var* genes at low levels. This was unexpected, and, therefore, I more closely examined PfSAMS expression by qPCR in these two lines. Surprisingly, PfSAMS expression was greatly reduced, as shown in **Figure 24** (clones H9 and C7), indicating that this population was no longer over-expressing PfSAMS. This also provides an explanation as to why the parasites had stopped exclusively expressing *var2csa*. Expression of PfSAMS in these two clones was found to be even lower than an untransfected WT line. The reason for this loss of PfSAMS over-expression is not known, but likely is the result of an alteration in the activity of the episomal promoter driving expression of the PfSAMS construct. Additional investigation will be required to determine the cause of this change in expression.

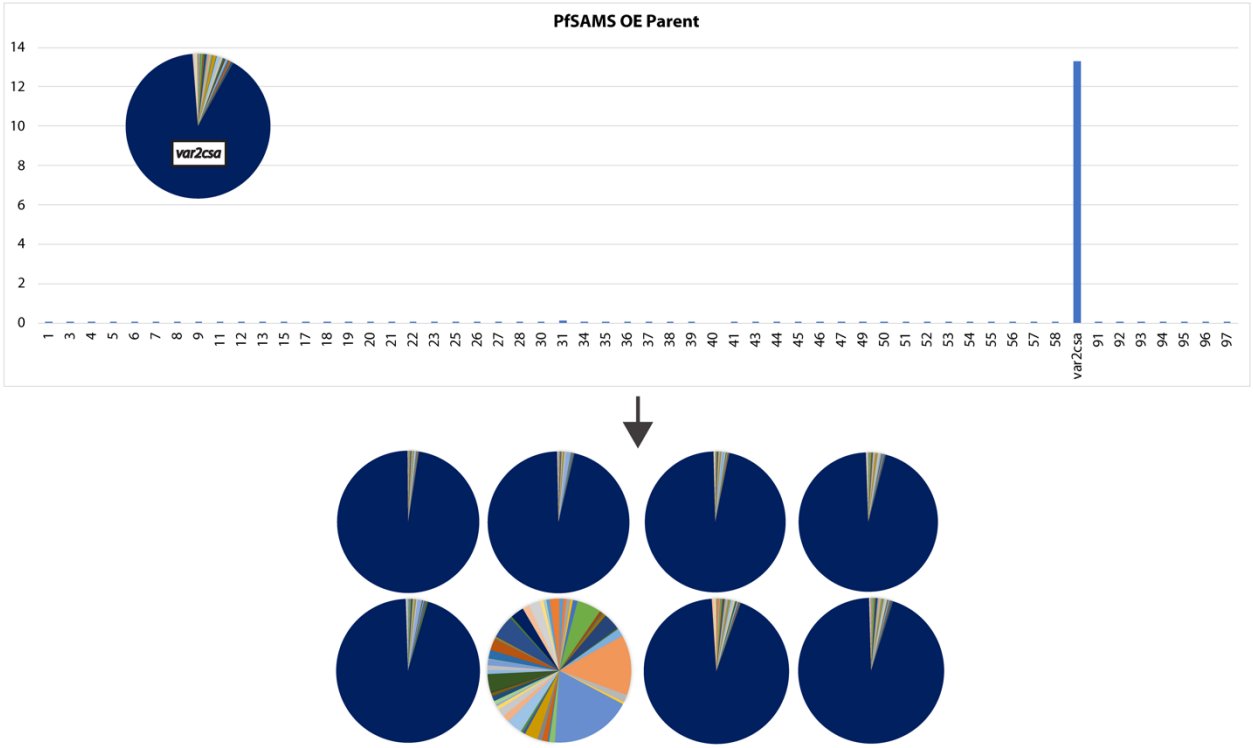


Figure 23. The majority of PFSAMS OE clones retain a homogeneous *var2csa* expression profile matching the parent PfSAMS OE parent line (top) was cloned. The *var* expression for 8 individual clones are shown as pie charts. Within each pie chart, an individual slice represents the relative level of expression for a particular *var* in the population. The clone on the bottom row, second from the left showed a low, heterogenous *var* expression profile. The remaining seven clones all retained high expression of *var2csa.*

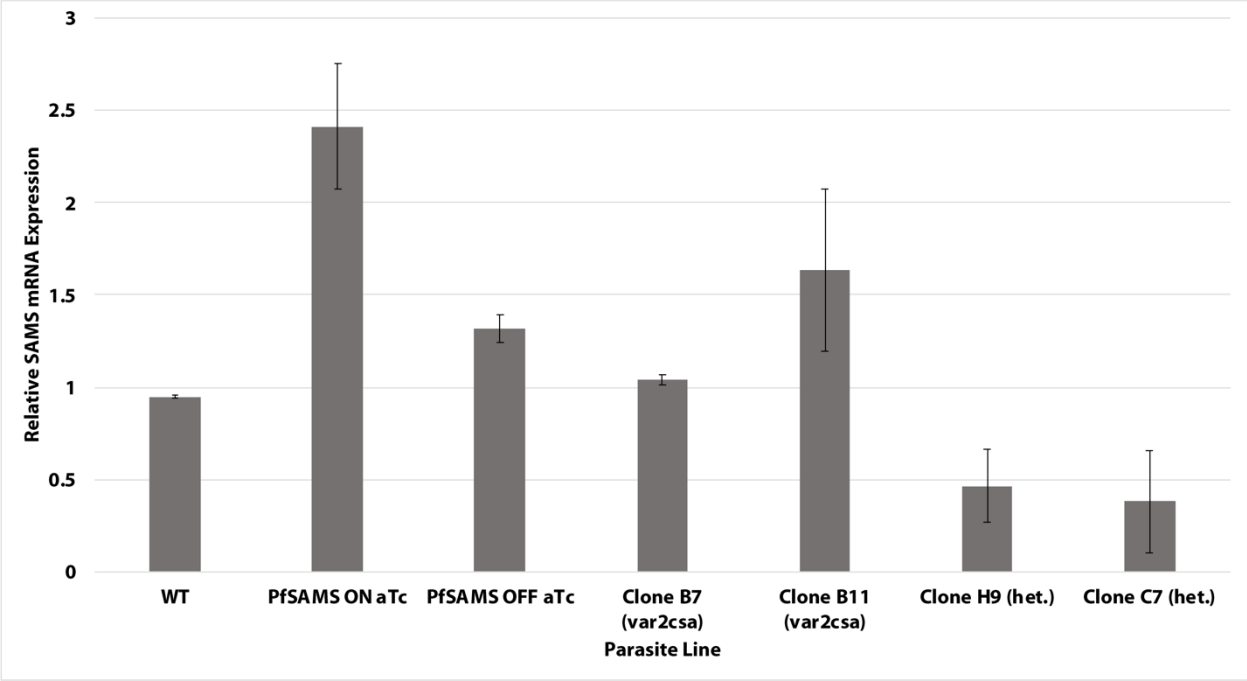


Figure 24. PfSAMS OE clones with heterogeneous *var* expression have a lower level of SAMS expression PfSAMS expression measured for each line (x-axis) by qPCR. Primers (listed in **Methods**) recognizing SAMS were used on cDNA from an untransfected WT, PfSAMS transfected lines on and off ATc, PfSAMS OE clones B7 and B11 that were on *var2csa*, and PfSAMS OE clones H9 and C7 that showing low-level, heterogenous (het.) *var* expression. Each bar represents the average SAMS mRNA expression level, normalized to seryl-tRNA synthetase expression, across n=4 reaction wells for each parasite line. Error bars represent the standard deviation.

The clones obtained from the PfSAMS KD line are pictured as pie charts in **Figure 25**. Full panels are shown in **Appendix**. Interestingly, all clones maintained generally the same heterogenous *var* expression pattern as the parent population. If these parasites were undergoing normal or rapid switching, I would expect the heterogenous profiles to differ between each clone. However, the patterns were very similar, with the most highly expressed genes shared between all eight clones. This is surprising, and indicates either that switching has become skewed toward this subset of highly expressed genes, or, alternatively, that mutually exclusive expression has been altered and that individual parasites are now expressing all of these genes. Interestingly, some of the genes that are expressed at a low level differ between the clones (for example the top right, bottom left, bottom right pie charts in **Figure 25**), suggesting that the original population maintains the ability to switch *var* genes to some extent under conditions of lower PfSAMS expression.

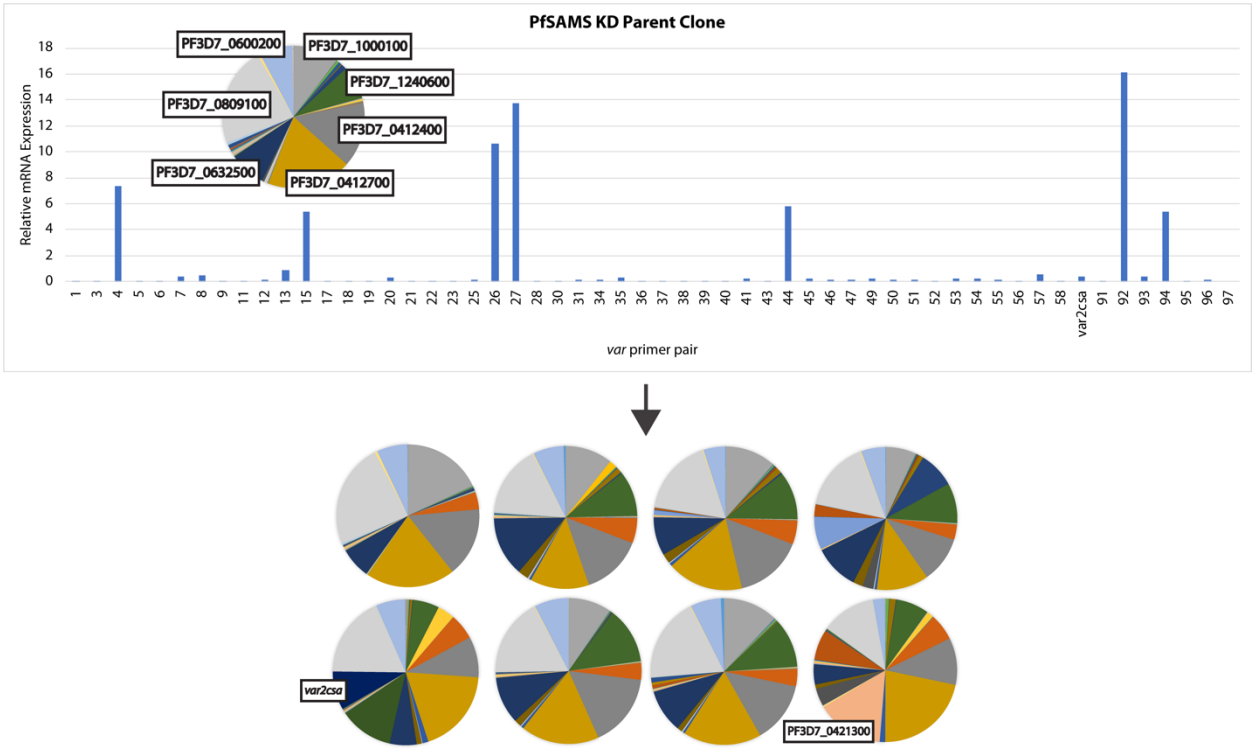


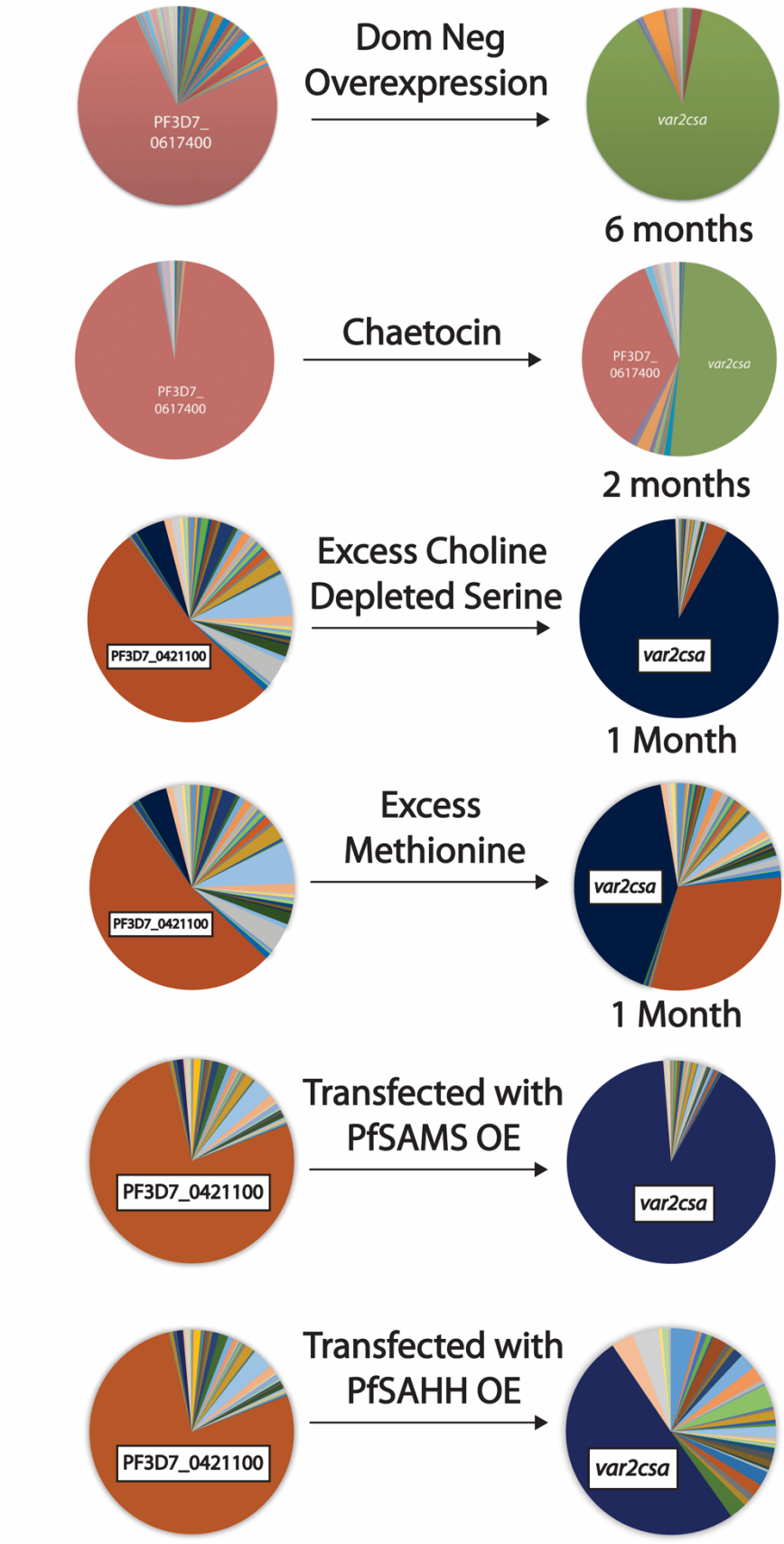
Figure 25. PfSAMS KD clones retain a heterogeneous *var* expression profile PfSAMS KD line (top) was cloned. The *var* expression for 8 individual clones are shown as pie charts. Within each pie chart, an individual slice represents the relative level of expression for a particular *var* in the population.

Discussion

Studies in other cell systems have found that when the ratio of SAM/SAH is high, SAM tends to offload methyl groups to "methyl sinks" such as histones through HMT. [58,67] Based on the results of **Chapter 2**, I hypothesized that conditions of excess choline to depleted serine, or excess methionine, lead to *var* switching through an increase in SAM pools, which in turn donate their extra methyl groups to HMTs, depositing them on histones and epigenetically modifying (*var*) gene expression. (**Figure 14**). In this chapter I took a genetic approach to test this more directly, by modifying the expression of two key enzymes in SAM metabolism: PfSAMS and PfSAHH.

Following **Figure 8**, I hypothesized that increasing the expression of either PfSAMS or PfSAHH would induce a *var* switch, specifically to *var2csa* following the results of **Chapter 2**. SAM synthetase (PfSAMs) catalyzes the formation of SAM from methionine and ATP. SAH hydrolase (PfSAHH) catalyzes the reversible hydrolysis of SAH to adenosine and homocysteine, turnover that allows the cell to make more SAM (**Figure 8**). Therefore, both would hypothetically increase the steady state levels of SAM/SAH over time. In further support of the hypothesis expressed in **Figure 14**, both OE lines were found to have robustly activated *var2csa* at the population level even at the earliest possible timepoint analyzed (Day 0 timepoints, **Figure 17 & Appendix**). HMT K_m values lie in the range of intracellular SAM concentrations, suggesting that even minor fluctuations in SAM levels could significantly affect HMT activity, increasing or decreasing histone methylation and providing a direct link between it and cellular metabolism through SAM. [62] [28,57] For both constructs, leaky expression from the episome was noted in the absence of ATc (**Figure 15**). This may explain why, even without the addition of ATc, parasites had already induced *var2csa* expression. The coordinated, stable switch at the population level to *var2csa* again phenocopies the results of Ukaegbu et al., 2015, along with the results of **Chapter 2**. These are shown together in **Figure 26**.

Figure 26. Conditions hypothesized to increase SAM pools phenocopied earlier changes in *var* expression growing parasites in conditions of excess choline and depleted serine or excess methionine led to a coordinated switch in *var* expression at the population level to *var2csa***.** Similarly, increasing the expression of SAMS or SAHH led to a coordinated switch in var expression at the population level to *var2csa.* These results phenocopy those seen in Ukaegbu et al., 2015 in response to genetic or chemical modifications of HMT activity. First two rows of *var* panels taken from Ukaegbu et al., 2015.


PF3D7_
0617400
Dom Neg
Overexpression
var2csa
6 months
PF3D7_
0617400
Chaetocin
PF3D7_
0617400
var2csa
2 months
PF3D7_0421100
Excess Choline
Depleted Serine
var2csa
1 Month
PF3D7_0421100
Excess
Methionine
var2csa
1 Month
PF3D7_0421100
Transfected with
PfSAMS OE
var2csa
PF3D7_0421100
Transfected with
PfSAHH OE
var2csa

Transfection of *P. falciparum* parasites introduces a severe selective bottleneck, as the vast majority of parasites are killed when the culture is initially placed under blasticidin selection. In order to rule out the possibility that the phenotype seen for the OE lines was simply the result of a major selective bottleneck, I grew both lines off of blasticidin selective pressure for two months. Allowing the parasites to shed the episome (and presumably lower SAM/SAH levels as increased enzyme expression was diminished) induced a switch in *var* expression away from *var2csa* (**Figure 20**). In the case of the PfSAMS OE, the population's *var* expression became more heterogenous. The PfSAHH OE largely switched back to the gene that was on in the parent clone at the time of transfection (**Figure 17**). The *var2csa* phenotype was less homogenous than that in the PfSAMS OE (**Figure 17 & 26**), possibly owing to its less direct role on SAM levels than SAMS itself in the PfSAMS OE. Mathematical models of *var* switching suggesting the existence of a "switch intermediate" or "sink node" (postulated to be *var2csa*) also support a switching network where most parasites that switch into the sink node will actually switch back to a previous *var* that was "on" instead of a previously silent one[55]. This is likely due to an epigenetic memory, which predisposes the *var* which was already on for the easiest re-activation, and would provide an explanation for the phenotype seen when the PfSAHH OE was de-selected[36 89].

Changes in *var* switching propensities should be particularly visible when the number of parasites within a population is small, and thus the effect of individual switching events on the total *var* expression profile are amplified. To examine underlying switching patterns within the population of the homogenous PfSAMS OE, I generated clonal lines through the isolation of individual iRBCs. Most of the clones screened maintained high, single expression of *var2csa* as was seen in the parent line, following my hypothesis (**Figure 23**). However, two out of a total of 14 clones analyzed had unexpectedly heterogeneous *var* expression profiles, in which many *var*s were expressed at very low levels. While investigating, I found that the clones had an unusually low level of PfSAMS expression (**Figure 24**), lower even than an untransfected WT clone. The explanation for this loss of PfSAMS over-expression is not known, but is likely the result of an alteration in the activity of the episomal

promoter driving expression of the PfSAMS construct. Further investigation of these lines will be required to determine the precise cause of this change in expression. Nonetheless, the fact that both of these clones have markedly lower SAMS expression than clones that maintained *var2csa* expression again supports my overall hypothesis that increased SAM pools induces *var2csa* activation **Figure 27**.

My colleague Chantal Harris generously provided me with her PfSAMS KD line, allowing for a comparison of phenotypes with the OE. This line proved intriguing within the scope of my model, as multiple independent PfSAMS KD lines all had a similar heterogenous *var* expression pattern at Day 0 (**Figure 18 & Appendix**). Induction of the KD through the addition of GlcN did not noticeably alter this expression profile, or induce a coordinated switch to *var2csa*. Integration of the GlmS ribozyme at the endogenous PfSAMS locus was found to lower SAM levels, even without the presence of GlcN (Chantal Harris, personal communication). Therefore, the PfSAMS KD lines hypothetically contain a decreased baseline level of SAM/SAH relative to a WT line. Taking this important piece of data in the context of the broader model, increased SAM pools seemingly push parasites into *var2csa*, while decreased SAM pools do not, but, instead, appear to dysregulate *var* expression. This dysregulation manifests as populations with heterogenous *var* expression profiles. Another instance of this can be found in the de-selected PfSAMS OE line, which, after two months off of drug pressure, saw a marked decrease in *var2csa* expression and instead low-level expression of many *var*s (**Figure 20**, top). An updated hypothetical model taking these results altogether is shown in **Figure 27**.

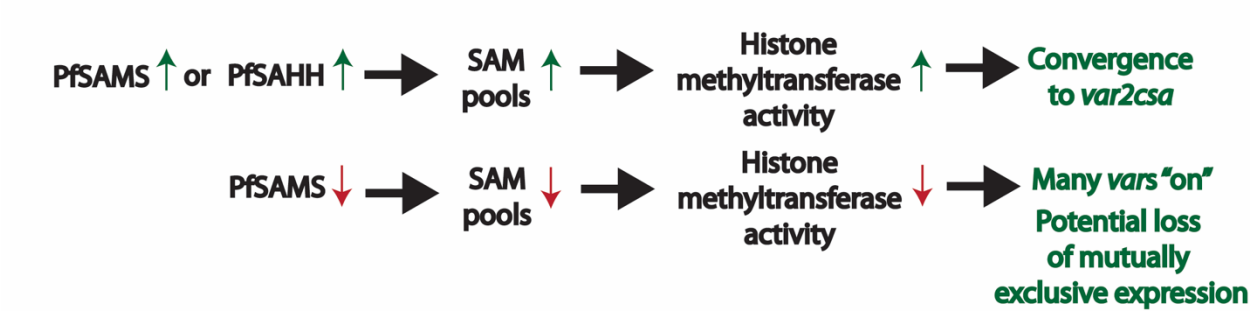


Figure 27. Hypothesized model for the link between intracellular SAM pools and *var* gene expression in response to varying expression of PfSAMS or PfSAHH Directly increasing expression of SAMS would be hypothesized to increase SAM pools (Figure 8), this would be hypothesized to elevate levels of SAM/SAH. Similarly, increasing levels of SAHH would allow for recycling of components used to synthesize SAM (Figure 8), hypothetically increasing the steady state SAM/SAH ratio. This, in turn, would allow for greater methyl donation to HMTs and increased deposition of epigenetic marks. This destabilization of the epigenetic regulatory network increases rates of *var* switching specifically to *var2csa*. Conversely, decreasing SAMS expression would decreasing SAM levels, hypothetically decreasing methyl donation to HMTs and deposition of epigenetic marks. This destabilization of the epigenetic regulatory network increases rates of *var* switching to *var* genes but notably not *var2csa.* The stable and high expression of multiple *var* genes across the populations of these parasites could be indicative of a loss of mutually exclusive expression.

In the literature, there is precedent for a heterogenous *var* expression profile in response to conditions that would be thought to lower methylation within *P. falciparum*. A knock out of PfSET2 resulted in leaky expression of all *var* genes across a population [37]. *P. falciparum* heterochromatin protein 1 (PfHP1) and *P. falciparum* histone deacetylase 2 (PfHda2) are both silencers of gene expression: through the deposition of methyl marks and formation of heterochromatin, or through the removal of acetyl marks to allow for methylation, respectively. Independent knockdowns of each similarly led to leaky expression of all *var* genes in the population[41,42]. If decreasing SAMS expression does indeed decrease SAM pools, the population-level *var* phenotype would fit with these results. Remarkably, this phenotype was consistently maintained across clones of the PfSAMS KD line (**Figure 22**). One possible, intriguing explanation for the heterogenous *var* profile seen in these lines is that a global decrease in methylation of histones leads to a loss of mutually exclusive *var* expression. Tightly-regulated, mutually exclusive expression of *var* genes avoids premature exposure of

their full antigenic inventory. [16,22,23] The switch in expression from one *var* gene to the next has evolved to occur at a low rate, allowing for the expansion of clonal *var* populations over many generations (seen as the discrete waves of parasitemia in **Figure 5**). [2,24,25] Therefore, a loss of control over mutually exclusive *var* expression in response to decreased methylation would have profound implications for the parasite's *var* switching network. The results of cloning the PfSAMS KD line (**Figure 25**) strongly support the hypothesis that these parasites are expressing more than one *var*, at high levels, per cell, as each clonal population should reflect the *var* expression of an individual founding parasite. The most definitive way to determine if any of these lines contain parasites that are expressing more than one *var* simultaneously is through single cell RNA sequencing[117]. Methods to run single cell RNA sequencing on ring stage parasites (when *var* expression peaks) are currently being developed.

In contrast to the PfSAMS OE, and since the PfSAMS KD is a modification of the endogenous SAMS locus, I was not able to definitively determine whether the heterogenous *var* phenotype present across clones is truly the results of decreased SAMS expression, or is an artifact of transfection. The numerous *var*s observed are not only on at a very high level of relative expression, but are also remarkably consistent across independently generated lines (**Appendix**) and the clones in **Figure 25**. This phenotype could very well be the result of a structural change resulting from integration of the GlmS construct at the chromosomal locus.

My attempts to rescue SAMS expression through transfection of the PfSAMS OE episome into the PfSAMS KD only altered this heterogenous *var* expression pattern in one replicate: the R1 line **Figure 21**, top plot. Therefore, while this line offers some support of my overall hypothesis and against the possibility of the PfSAMS KD phenotype simply being structural artifact, this phenotype could not be replicated. Nevertheless, these results are preliminary, and further work will need to be conducted on these two rescue lines to determine why R2 did not show activation of *var2csa*. Given the results observed when the PfSAMS OE line was cloned (**Figure 23 & 24**), it is possible the R2 line does not have increased SAMS expression from the episome. With regards to my hypothesized model (**Figures 14 & 27**), the more important piece of data

is the inability of the PfSAMS KD to induce stable *var2csa* expression unless PfSAMS expression is rescued.

The results of this chapter further support the model first proposed in **Figure 14** and expanded upon in **Figure 27**, by evidencing a role for SAM pools in influencing *var* switching. Specifically, conditions where the level of SAM/SAH is high induce a robust switch into the theorized "sink node" *var2csa*. **Chapter 2** presented the novel possibility that *P. falciparum* possesses the machinery to orchestrate this switch in response to environmental changes. **Chapter 4** will expand upon this "sensing" idea by looking specifically at immune recognition signals. If future work points to the phenotype of the PfSAMS KD simply being a structural artifact, the overall model with regards to *var2csa* will continue to be supported. However, if a loss of mutually exclusive *var* expression in response to decreased SAM pools is supported by future work, this would have profound implications for the overall regulation of the *var* switching network.

The results of this and that of **Chapter 2** suggest a model for *var* switching through SAM pools where individual *var* promoters "compete" for activation. This model of *var* "promoter competition", as it is supported by the results of this thesis, will be presented in **Chapter 5**.

Chapter 4

Searching for an Environmental Signal – The Immune Response to *P. falciparum* Intersects with Amino Acid Starvation and Polyamine Synthesis

Background

Alongside a rapidly growing interest in the study of cellular metabolism, the past decades have seen the advent of research into the metabolic control of immune responses. The transition of immune cells from quiescence to activation, and further differentiation to perform their specific functions, is regulated by and dependent on nutrient sensing to downstream metabolic intermediates and cellular responses. [90] [91] The activation of immune cells by environmental signals leads to dramatic reprogramming of their cellular metabolism, providing them with abundant energy and the metabolic intermediates needed to perform their assigned functions. Furthermore, the sensitivity of metabolic pathways to changing environmental conditions helps to maintain tight regulation over immune cell function and prevent potentially lethal mis- or overactivation. [92] Although study surrounding the precise networks joining metabolism to immune regulation is presently in its infancy, preliminary findings have demonstrated that this area of research fits into a larger biological paradigm, in which the fundamental properties of cells are dictated by their underlying metabolic state. [91]

Nutrient sensing and metabolic stress pathways play a particularly critical role in the frontline, innate immune response to pathogens. Two canonical cellular sensing systems principally control this response: the mTOR (mechanistic target of rapamycin) - and eIF2α (eukaryotic initiation factor 2) -dependent signal transduction cascades. [93,94] Both pathways independently monitor nutrient levels (with foremost sensitivity to amino acid (AA) starvation) and modulate the cellular response downstream by regulating key processes requiring nutrients and energy (such as growth and division and gene expression). [90] The mTOR complex, conserved across yeast to mammals, is the master regulator of cell growth. Under conditions of AA starvation, mTOR activity is repressed, and protein synthesis is largely shut down. [95] In the eIF2α pathway, conditions where AAs are limiting activate the AA-sensing eIF2α kinase, GCN2, which

phosphorylates, and in doing so deactivates, eIF2α (eIF2α-p). Part of the Integrated Stress Response (ISR), this cascade mediates a global decrease in translation, metabolic readjustments, and the activation of transcription factors corresponding to genes responsible for AA synthesis. [96] Given their essential role in propagating the immune response, it has been noted that several species of viruses, bacteria, and parasites possess the ability to hijack and even inhibit components of these pathways in the cells of their host. [90]

The pathogens themselves also use these pathways to respond to changing nutrient conditions over the course of infection. *Plasmodium* species lack the mTOR complex, but the *P. falciparum* genome contains an ortholog to eIF2α (PfeIF2α) and three putative eIF2α kinases. [97] Of them PfeIK1 is an ortholog of GCN2, phosphorylating PfeIF2α in the parasite's intraerythrocytic stages. [98] However, the parasite lacks the downstream transcription factors and biosynthetic pathways, including those to synthesize many of its own AAs, that mediate GCN2 action, leaving any other biological role(s) presently undefined. [97,99]

While *P. falciparum* is auxotrophic for most AAs, it is able to obtain them from the digestion of hemoglobin. However, one AA, isoleucine (Ile), is absent from hemoglobin. Therefore Ile must be obtained from human serum, as the parasite cannot synthesize it *de novo,* and over 99% of proteins encoded by the organism incorporate it. [97,99] To this point, while an earlier study found that *P. falciparum* could grow in culture medium containing only five AAs (cysteine, glutamate, glutamine, methionine, and isoleucine) [100], more recent work has found that it can survive with Ile as the sole exogenously supplied AA[108]. Babbitt et al., 2012 showed that when starved of Ile, *P. falciparum* enters a "hibernatory" state of slowed growth that can be immediately reversed upon re-supplementation of Ile. This is facilitated by rapid phosphorylation of PfeIF2α by PfeIK1. However, in contrast to a canonical eIF2α-p signal cascade, there was no activation of alternative transcriptional programs consistent with *Plasmodium*'s lack of homologs to known downstream starvation-response regulators present in other eukaryotes. Interestingly, mass spectrometry analysis on parasites under Ile starvation found disrupted levels of metabolites involved in one-carbon and SAM metabolic

pathways. Furthermore, this response appears to be Ile-specific, as glucose starvation resulted in parasite death. [97] This phenotype was recently replicated in McLean et al., 2020, in which they additionally demonstrated that parasites only enter a state of Ile-deprived hibernation *in vitro* if starved pre-S phase, or before DNA replication largely commences. [101] These findings are physiologically relevant in light of clinical readings in malaria endemic regions where malnourished individuals, predominantly children, contain low or undetectable levels of certain AAs in their plasma. Ile, specifically, can drop in malnourished patients to as little as one-tenth the concentration found in healthy individuals. [97]

While the parasite's dependence on exogenous Ile leads to a dramatic response in its absence, would a drop in the level of any other AAs trigger a more subtle response? One possibility, the IDO pathway, ties back into the idea of metabolic immune regulation. In overview, Indoleamine 2,3 dioxygenase (IDO) is an enzyme expressed by many human cell types and some antigen-presenting immune cells in response to inflammatory stimuli. [102] When IDO is expressed it affects the host immune response by depleting essential AA tryptophan (Trp), and then producing kynurenine (Kyn) as a byproduct. IDO enzymes function intracellularly and are not secreted, depleting environmental Trp and secreting Kyn byproduct. Trp depletion in the local microenvironment triggers the mTOR and eIF2α AA-sensing cascades mentioned above. Trp is an essential AA to T cell function and proliferation, and, thus, its depletion suppresses T cell activity. Kyn is a natural ligand for aryl hydrocarbon receptor (AhR) found on immune cells, and its effect on cells is also largely immunosuppressive. Hence, IDO is theorized to contribute to acquired peripheral tolerance, or the prevention of excessive immunity to non-self antigens, as in cases of transplants or chronic inflammatory diseases. [102] Notably, IDO has been found to have particularly high activity in the human placenta, and therefore has been proposed as a mechanism in preventing immunogenic rejection of the fetus. [103,104] T cell function can also be modulated in a GCN2-dependent fashion through the depletion of local arginine (Arg) by the arginase-I enzyme, expressed by activated immune cells.[105]

Present thought generally places macrophages, part of the innate immune response, into two classes: pro-inflammatory (M1 macrophages) and anti-inflammatory (M2 macrophages), with distinct metabolic and molecular signatures[106]. These two classes derive from the method by which a macrophage is activated, or polarized, by stimuli in the local microenvironment. M2, anti-inflammatory macrophages specifically deplete local concentrations of Arg, through expression of arginase-1, and Trp, through IDO-mediated degradation[106]. In line with this, M2 macrophages have been found to be largely present at the maternal-fetal interface[118].

Antigenic variation by *P. falciparum* and its effect on the adaptive immune response (T and B cells) has been researched more thoroughly than its effect on the innate immune response [119,120]. Recent research in mammalian systems has suggested that immune perturbations can epigenetically and metabolically "train" innate immune cells to respond in a certain way to subsequent infections. [121] This training can either enhance or diminish the response, and thus render it more tolerant. In malaria endemic regions, it has long been observed that individuals tolerate *Plasmodium* parasites at levels that would otherwise trigger a fever in naïve individuals [122]. One recent report tied this observation to Mali, where an increased amount of M2 macrophages in adults relative to the amount found in children suggests that repeated and/or chronic exposure to *P. falciparum* drives a more tolerant and less inflammatory innate immune response [120]. This report underscores the importance of further research into the innate immune response to *P. falciparum* and its ultimate effect on the parasite's pathogenesis.

In addition to amino acid depletion, M2 macrophages increase local concentrations of polyamines [106]. Polyamines are polycationic organic amines synthesized from amino acid precursors that bind to different cellular molecules -most notably, nucleic acids [123]. Polyamines and their synthesis enzymes are required for growth and protein translation, demonstrated by their exceptionally high presence in rapidly proliferating cell types, such as cancer cells and protozoan parasites [109]. *Plasmodium* species are the only parasitic apicomplexa to possess the complete enzymatic polyamine biosynthetic pathway, pictured in **Figure 28** [124]. As *P. falciparum*

parasites progress from ring (early) stages to schizont (late) stage there is a 10-20 fold increase in the level of intracellular polyamines [125]. Total polyamine levels in iRBCs are two orders of magnitude higher than uRBCs [124]. Inhibition of polyamine synthesis results in translational arrest, with specific compounds found to have potent antimalarial activity *in vivo* [125,126]. Taken together, polyamine synthesis is of utmost importance to the metabolism of *P. falciparum*.

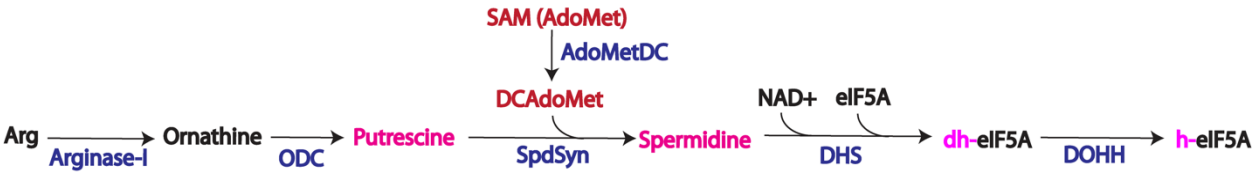


Figure 28. Polyamine synthesis pathway in *P. falciparum* *P. falciparum* possesses the complete polyamine synthesis core. Polyamines putrescine and spermidine are shown in pink text. Enzymes are shown in blue text. Adapted from Philips, 2018.

In *P. falciparum,* two polyamines are of chief importance: putrescine (Put) and spermidine (Spd) **Figure 28**. Put is produced from decarboxylation of ornithine (Orn) by Ornithine Decarboxylase (ODC). Put is then used by Spermidine Synthase (SpdSyn) to synthesize Spd using decarboxylated SAM (DCAdoMet, synthesized by SAM decarboxylase or AdoMetDC). Importantly, *P. falciparum* encodes a bifunctional ODC/AdoMetDC enzyme, and is the only known organism to do so [124,127]. One hypothesis for the evolution of this bifunctional enzyme is that it enables to *P. falciparum* to regulate polyamine synthesis through only one enzyme [128]. Interestingly, put has been found to inhibit ODC activity ~10-fold greater than the mammalian ODC activity [128]. This extra level of regulation further underscores the importance of this pathway to the parasite's biology.

Spd has been measured at about twice the level of Put in mature trophozoites (late stage parasites) [109]. This is most likely due to Spd's important role in stabilizing translation. In all eukaryotes, Spd serves as a substrate for deoxyhypusine synthase (DHS), which, together with deoxyhypusine hydroxylase (DOHH), modifies the

translation elongation factor eIF5A with the amino acid hypusine **Figure 28** [124]. eIF5A is the only known protein that receives the hypusine modification. Hypusinated eIF5A (h-eIF5A) is essential in eukaryotes to maintain translational fidelity, relieving ribosome stalling to promote elongation and facilitating termination [123,124]. Depletion of polyamines, disruption of the polyamine synthesis pathway, and disruption of hypusination of eIF5A all trigger the ISR and phosphorylation of eIF2α.

While they are distinct pathways, amino acid starvation and polyamine synthesis can be compared by their similar ties to the innate immune response and downstream effects on translational fidelity. Both pathways are illustrated in full side-by-side in **Figure 29**. In **Chapters 2 and 3** I demonstrated that either by growing *P. falciparum* in conditions of excess choline to depleted serine, or by directly modifying the levels of SAM and SAH using overexpression constructs for SAMS and SAHH, a switch in *var* genes occurs at the population level. In both cases, this switch was pointedly to the same *var* gene, *var2csa*, phenocopying an earlier set of experiments where the activity of associated HMTs was altered (**Figure 26**). In **Chapter 1.7** I detailed why *var2csa* is a unique member of the *var* family. Notably, it encodes a form of PfEMP1 that exclusively binds to a placental-specific ligand, chondroitin sulfate A (CSA), meaning that the PfEMP1 protein encoded by this gene is only useful to the parasite when found in women who are infected for the first time while pregnant, as antibodies will be produced against it. [46,47] Additionally, in contrast to most other *var* genes, *var2csa* can be translationally repressed, even when transcriptionally active, due to the presence of a unique uORF **Figure 6**. [48,49] Mathematical models suggest that *var* gene switching occurs through use of transiently active genes, or "switch intermediates", which add a level of uniformity and coordination to the process[55]. While the identity of any "switch intermediate" is presently undefined, it is hypothesized that *var2csa* could be the sink node due its unique properties (**Chapter 1.7 and 1.8**), further supported by the results of **Chapters 2 and 3**.

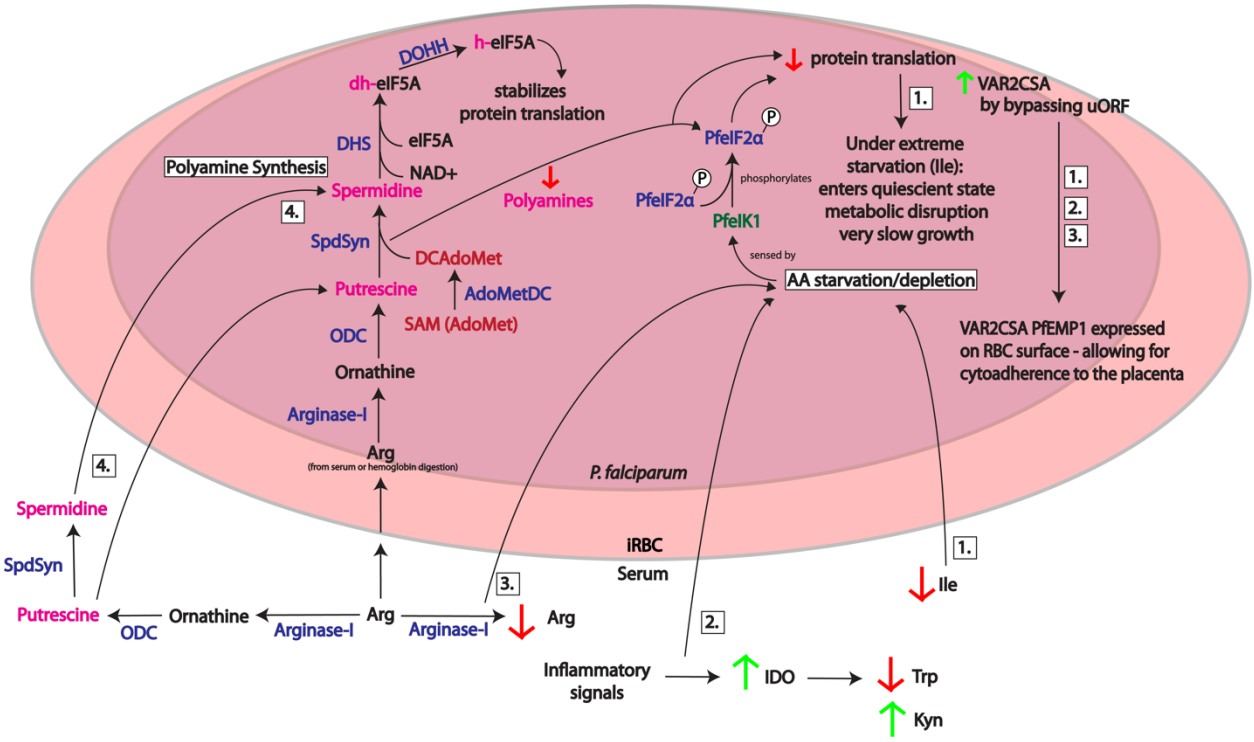


Figure 29. The innate immune response intersects with amino acid starvation and polyamine synthesis pathways in *P.falciparum* Babbitt et al., 2012 showed that Ile starvation (1) induces phosphorylation of an ortholog of eIF2α, PfeIF2α, by GCN2 ortholog, PfeIK1. This shuts down global protein translation, cellular metabolism, and drastically slows growth putting the parasite into a quiescent state that can be reversed upon re-supplementation of Ile. Following this, depletion of Trp and/or Arg (2 & 3) via IDO or arginase-I activity, respectively, may have the same effect. Phosphorylation of eIF2α in other systems has been shown to selectively upregulate translation of mRNAs with 5' uORFs. One hallmark of *var2csa* is its unique 5' uORF, possibly allowing its upregulation in response to AA stress. Spd (4) serves as a substrate for DHS, which together with DOHH modifies the translation elongation factor eIF5A with the amino acid hypusine. Spd can be synthesized from Put by the parasite or may be taken in from the external environment. The level of eIF5A-h could play a role in whether *var2csa* is translated through its interaction with ribosomes.

Although phosphorylation of eIF2α initiates a global decrease in translation, it also results in the selective upregulation of certain genes. The canonical example is activating transcription factor (ATF) 4, present in yeast and higher eukaryotes, which is only translated once eIF2α is phosphorylated and is responsible for activating the genes of the ISR. The regulation of ATF4 translation is controlled by two 5' uORFs. High levels of eIF2α-p favors ribosome re-initiation at the ATF4 coding region, rather than the inhibitory second uORF, allowing for translation. [107] Other mRNAs found to be upregulated under conditions of eIF2α-p similarly possess 5' uORFs that would ordinarily flag them for degradation via nonsense-mediated decay (NMD) pathways.[90,102] The presence of a 5' uORF at the *var2csa* locus, coupled with its function in the placenta, make it a possible target for upregulated translation upon IDO-mediated eIF2α-p.

eIF5A has similarly been found to impact the translation of mRNAs with uORFs. Whereas eIF2α-p increases translation of downstream open reading frames (dORFs), eIF5A-h decreases translation past the uORF. Thus, in conditions of lower polyamines, ribosomes will more frequently bypass the uORF stop codon and translate dORFs, while, in conditions of higher polyamines, ribosomes will drop off at the uORF stop codon [123,129]. In the placenta, polyamines are in high demand to support developing fetal cells [130,131]. Taken together, two possible models could explain how eIF5A would impact translation of *var2csa,* depending on steady state polyamine levels in the placenta*.* In one, placental cells would consume most of the local polyamines, resulting in a microenvironment where the steady state of polyamines would be lower. Following the effect on uORFs described above, circulating iRBCs would translate the full *var2csa* ORF, resulting in VAR2CSA PfEMP1 protein that can adhere to placental ligands. Macrophages recognizing PfEMP1 would increase arginase-1 activity, increasing environmental levels of Orn and polyamines. The higher level of environmental polyamines and h-eIF5A would promote ribosome stalling at the uORF stop codon, flagging *var2csa* mRNA for NMD and degradation, and promoting *var* switching away from this hypothesized switch intermediate.

The environment in the placenta should induce VAR2CSA protein translation. Thus, if the opposite is true (that localized steady state polyamine levels are high in the

placenta), a second model could postulate that higher levels of eIF5A-h may increase translation of *var2csa* past the uORF. The aforementioned studies linking translation of dORF to lower polyamine levels are based on work in mammalian cell systems [129], not *P. falciparum*, and, currently, the precise mechanism of action of eIF5A-h in *P. falciparum* parasites remains to be elucidated [132].

In contrast to African Trypanosomes, which have thousands of *vsg* genes in their antigenic repertoire to switch between, *P. falciparum* has only 60, begging the question as to how these parasites manage to cause persistent, long-term infections. [54] Each wave of parasitemia seen in trypanosome infections contains a parasite population expressing many different *vsg* genes, suggesting that they employ a stochastic switching dynamic. [53] If *P. falciparum* were to follow a similar mechanism, it would express all of the antigens in its repertoire in the first wave, and infections would not last as long as is documented[7]. This in turn suggests that *P. falciparum* must utilize a more elegant and precise switching mechanism, which has yet to be described.

The results discussed in **Chapters 2 and 3** directly challenge the commonly held paradigm that antigenic variation follows an intrinsic, programed switch rate by instead suggesting that this organism possesses the ability to switch in response to environmental changes. These experiments, however, are limited in their level of physiological relevance. My overarching hypothesis is that *P. falciparum* can sense when the host immune system first begins to recognize it (via environmental cues resulting from antibody recognition), and respond by switching *var* gene expression. If supported, it would describe a mechanism by which parasites can switch expression of *var* genes exactly when needed and with perfect timing, thus allowing them to utilize their limited repertoire of *var* genes with the utmost efficiency. This would also suggest a possible molecular mechanism that underlies the clinically observed events illustrated in **Figures 4 and 5**. In this chapter I will test the role AA starvation or depletion and polyamine supplementation could play as environmental signals capable of inducing *var* switching.

Results

4.1 Induced cell cycle arrest by isoleucine starvation does not induce *var* switching

In Babbitt et al., 2012 the dramatic effect of Ile starvation, mediated by PfeIF2α, on *P. falciparum* was demonstrated. Their preliminary MS results showed dysregulated levels of SAM metabolism-related compounds, suggesting that, while in their Ile-starved quiescent state, *P. falciparum* parasites may experience variable levels of SAM/SAH. [97] In light of my hypothesis, which ties *var* switching patterns to SAM levels, I tested whether parasites show a switch in *var* expression after being held in and then released from an Ile-starved quiescent state.

4-Thiaisoleucine (TI) is an analog of Ile that competes with Ile. When added to *P. falciparum* culture, it completely arrests parasite growth at late stages, during which the majority of DNA replication and protein synthesis takes place. In order to ensure the concentration added would arrest growth in culture, I ran a drug assay (**Methods**) on WT parasites. The drug curve obtained (**Figure 30A**) suggested the EC_{50} was ~930uM. After testing both 500uM and 1mM concentrations, I decided to use 1mM for the following experiments, as it more completely arrested parasites at late stages without killing them. In brief, the parent culture was synchronized with 5% sorbitol twice over the course of two growth cycles. RNA was extracted for an initial Day 0 timepoint, and the culture was then split into three cultures: one control off TI (grown in MCM), and two technical replicates that were put on 1mM TI (T6259 DL-4-Thiaisoleucine, Sigma) at high ring stage and left for approximately 36-48 hours, at which point the TI-treated media was washed off and each culture resuspended in MCM. The amount of time kept in arrest on TI was taken from Babbitt et al. 2012, where they showed recovery of parasites kept in an Ile-starved state for up to 72hr[97]. RNA from TI treated cultures was taken at the earliest possible timepoint after parasites were brought out of arrest, typically within the following two cycles.

Representative results across three biological replicates (**shown in Appendix**) are pictured in **Figure 30B**. Arrest induced by TI did not lead to any visible *var* switching at the population level; importantly, no switch to *var2csa* was seen beyond expression present at background level in the control line.

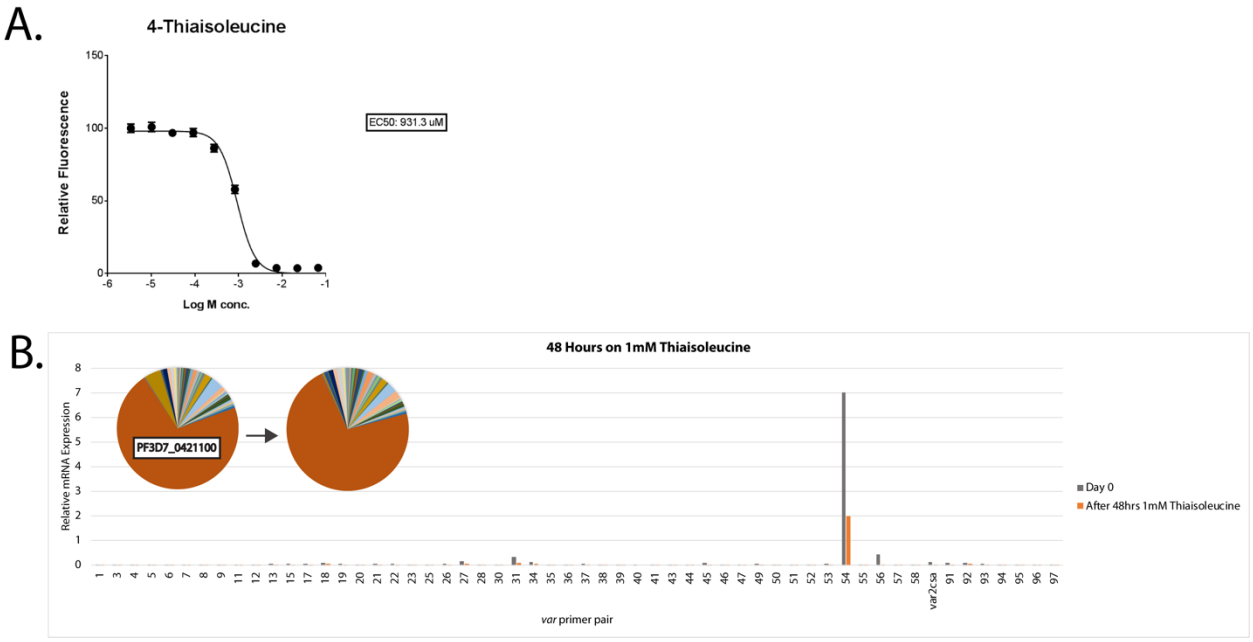


Figure 30. Induced growth arrest with 4-thiaisoleucine does not induce *var* switching A. Drug curve for 4-thiaisoleucine (TI) **B.** Parasite line was split and grown in the presence or absence of TI. Culture pictured was arrested in the presence of TI for 48 hours before it was washed off. Day 0 timepoint for the assay shown as gray bars and as pie chart (left). Culture arrested with 1mM TI shown as orange bars and pie chart (right). Representative results from across 4 replicates.

4.2 Prolonged depletion of tryptophan or arginine from growth media induces a coordinated *var* switch

Next, I decided to look at more subtle modes of AA depletion over time. Given the pronounced role of IDO in the placenta, the parasite's sensitivity to AA depletion through phosphorylation of PfeIF2α, and selective upregulation of genes with 5' uORFs by eIF2α-p, I tested whether the AA-depleted conditions resulting from IDO activity would induce a switch in *var* gene expression, specifically to *var2csa,* as was seen under previous conditions tested in **Chapters 2 and 3** (**Figure 26**)*. P. falciparum* depends on human serum, or, in culture, its growth medium, for its supply of Ile. It is able to obtain all other AAs from the digestion of hemoglobin. Therefore, testing the depletion of AAs other than Ile will not put the parasite into growth arrest, and can be applied over longer periods of time.

In order to test the effects of AA depletion, I made a new MIM powder. This powder was made exactly as detailed in **Methods** for the MIM used in Chapter 2; however, this batch only lacked Arg and Trp. This MIM will be referred to as AA MIM. I assayed *var* expression in response to Arg and/or Trp depletion, meant to mimic arginase-I or IDO activity, respectively, in a fashion comparable to the choline, methionine, and serine experiments in **Chapter 2.** Conditions tested for this chapter are shown in **Table 2.**

Table 2. AA and Polyamine conditions tested in Chapter 4 Each altered nutrient condition tested is presented along with the compound name supplemented in parentheses. When a compound was depleted, that nutrient was present at 0uM in growth medium. Levels of nutrients in human serum were taken from the Human Metabolome Database, https://hmdb.ca.

Condition	Amount Relative to MCM	uM tested	uM in MCM (RPMI)	uM in Human Serum	Product Used
Depleted Arg (L-Arginine)	0	0	1148	130-160	A8094 Sigma
Depleted Trp (L-Tryptophan)	0	0	24	54-64	T8941 Sigma
Supplemented Kyn (L-Kynurenine)	1:1 with Trp	24	0	1.6 - 1.7	L-Kynurenine from InvivoGen
Spd (Spermidine)	N/A	30nM, 10 & 100 uM	0	8.5-10.5	S0266 Sigma
Put (Putrescine dihydrochloride)	N/A	2mM	0	0.214-0.294	P5780 Sigma

Results are pictured in **Figure 31.** Depletion of Arg, Trp, or both simultaneously resulted in a coordinated switch at the population level to *var2csa.* Results shown are one of three biological replicates, shown in full in **Appendix**.

In order to more accurately mimic conditions of IDO, I also tested supplementing Trp depleted cultures with the by-product of its degradation, Kyn (**Figure 29**). This was added at the amount of Trp in MCM, following the a 1:1 degradation pathway reaction (KEGG). Addition of Kyn where Trp was depleted did not affect *var* switching in a different manner than when Trp was depleted alone; therefore, Trp depletion alone was enough to observe the *var* switching phenotype (induction of *var2csa*) pictured in **Figure 31**. Panels for cultures where Kyn was supplemented can be viewed in **Appendix**.

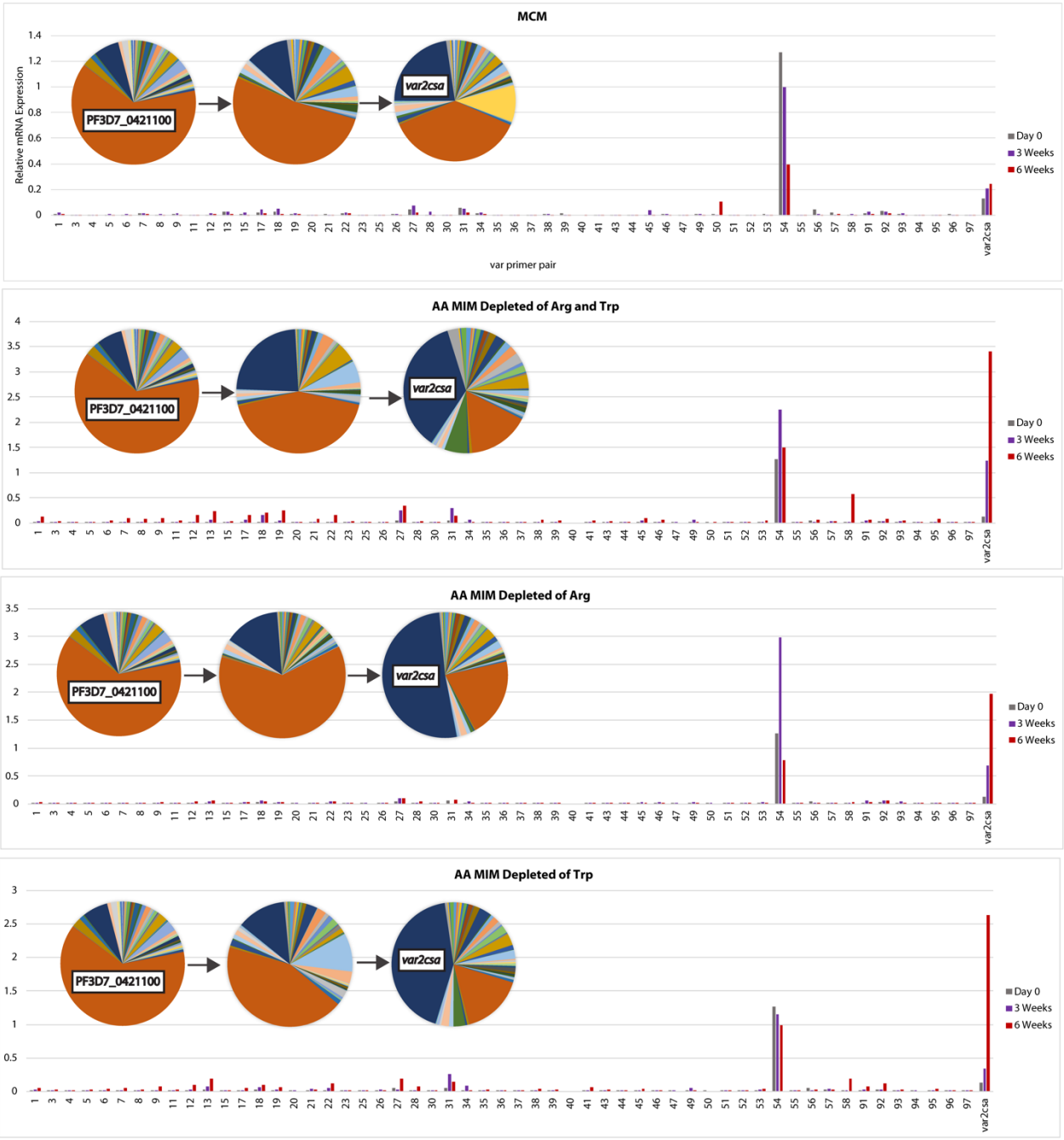


Figure 31. Depletion of amino acids in the growth media of *P. falciparum* induces a switch to *var2csa* at the population level over time *var* gene expression as assayed for each condition is shown as a pie-chart and bar graph, with each slice of the pie and each numbered primer pair from Salanti et al. 2013 on the x-axis representing the fraction of the total *var* mRNA pool transcribed from each *var* gene. Gray bars represent Day 0 timepoint, or the *var* expression of the parent culture before it was split across different nutrient conditions. Purple and red bars are the *var* expression at 3 and 6 week timepoints, respectively.

4.3 Depletion of tryptophan or arginine from growth media does not significantly affect growth

Liu et al. 2006 demonstrated that *P. falciparum* parasites will grow in medium depleted of any AA except Ile[108], the only AA the parasite cannot scavenge from hemoglobin. Thus, I tested if the depletion of Arg or Trp would notably alter growth rate compared to those grown in MCM. Following the growth assay procedure detailed in **Methods**, parasites were grown in the conditions presented in **Figure 31 & Table 2** for two weeks. Plotting growth on a log scale in **Figure 32**, no considerable growth phenotype was observed. **Figure 32** shows representative results for one assay, while replicates and log growth numbers are shown in **Appendix**. Kyn supplementation also did not meaningfully affect growth (**Appendix**).

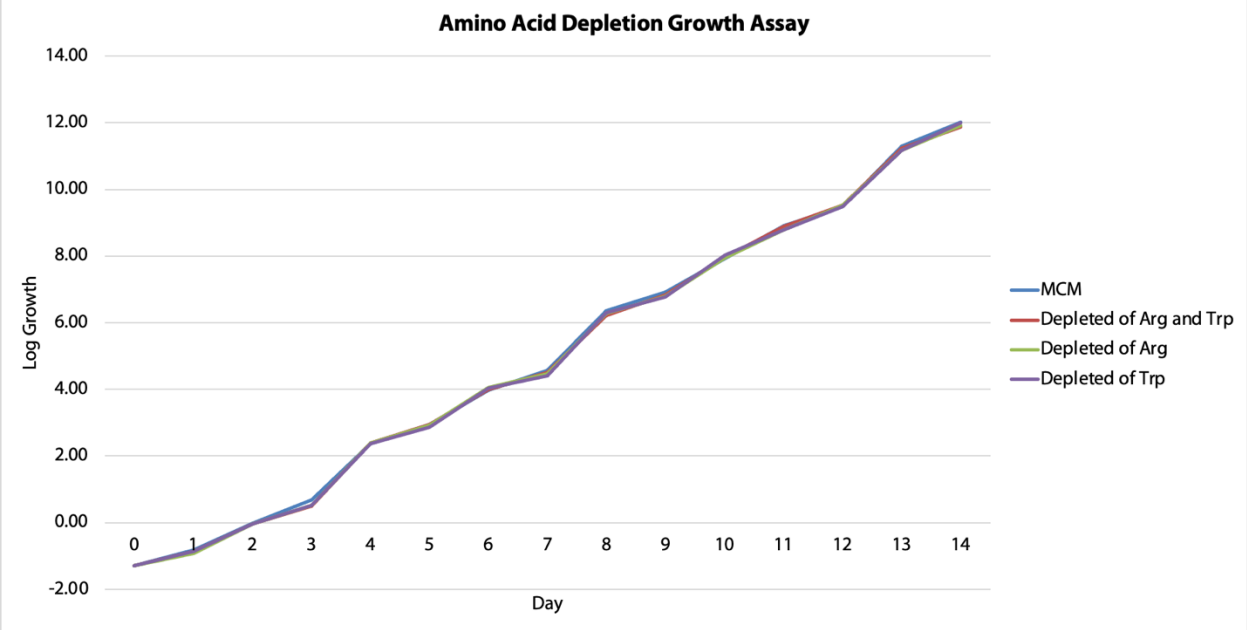


Figure 32. Depletion of Arg and Trp in the growth media of *P. falciparum* does not significantly alter its growth rate Parasites were cultured in the conditions tested over the course of 14 days (x-axis). Parasitemia was measured by flow cytometry and log growth (y-axis) was calculated following the protocol in **Methods.**

4.4 Prolonged supplementation with spermidine induces a coordinated *var* switch

In addition to amino acid depletion, macrophages increase local concentrations of polyamines [106]. Intracellularly, *P. falciparum* synthesizes large amounts of polyamines, with a 10-20 fold increase in the level of polyamines observed in late stage parasites compared to early rings [125]. Spd has been measured at about twice the level of Put in mature trophozoites, likely due to Spd's important role in stabilizing translation through the hypusination of eIF5A[109]. The synthesis of Spd from Put by SpdSyn requires consumption of SAM **Figure 29.** The results of both **Chapters 2 & 3** put forth a model where conditions thought to increase SAM pools led to a coordinated *var* switch at the population level to *var2csa*. Therefore, I hypothesized that if parasites were grown in culture in the presence of supplemented Spd, *var2csa* expression would be induced, as they would need to take less Put to Spd, consuming less SAM.

To test this, I cultured parasites in MCM with varying concentrations of exogenous Spd added following **Table 2**. Niemand et al. 2012 demonstrated that *P. falciparum* parasites will take up polyamines added to their growth medium, validating this experimental approach[109]. After testing varying concentrations, 100uM was the upper-most limit of Spd I found I could culture parasites in long-term without a significant loss of viability.

Results of growth over a period of six weeks are shown in **Figure 33A**. Following my hypothesis, parasites grown in the presence of 100uM exogenous Spd show a coordinated population switch to *var2csa*. To further test the physiological relevance of this phenotype, I also tested 10uM Spd – the upper limit found in human serum **Table 2.** Similarly, growth over a period of six weeks in the presence of 10uM Spd induced a switch to *var2csa,* faster and more robustly than that seen as background in the MCM control **Figure 33B**. Both are representative results across a series of four assays shown in full in **Appendix**. An even lower concentration of Spd, 30nM, was also tested, as this was the amount tested by Niemand et al. 2012. This amount also induced a switch to *var2csa* faster and more robustly than that seen as background in the MCM control*,* though it was less dramatic than 100 or 10uM (**Appendix**).

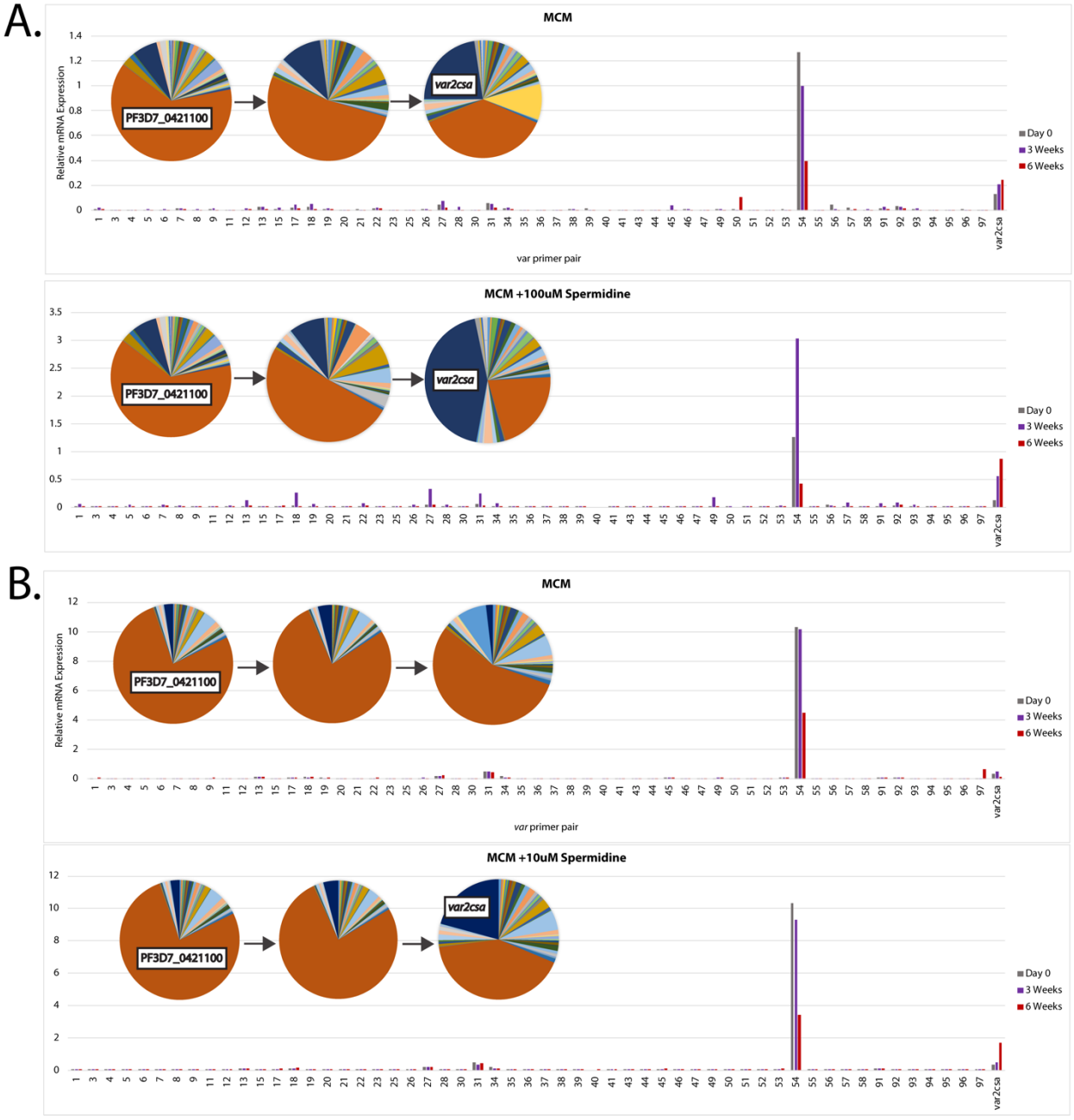


Figure 33. Supplementation of spermidine in the growth media of *P. falciparum* induces a switch to *var2csa* at the population level over time A. 100uM of Spd added exogenously **B.** 10uM of Spd. *var* gene expression as assayed for each condition is shown as a pie-chart and bar graph, with each slice of the pie and each numbered primer pair from Salanti et al. 2013 on the x-axis representing the fraction of the total *var* mRNA pool transcribed from each *var* gene. Gray bars represent Day 0 timepoint, or the *var* expression of the parent culture before it was split across different nutrient conditions. Purple and red bars are the *var* expression at 3 and 6 week timepoints, respectively

4.5 Prolonged supplementation with spermidine induces a more robust switch to *var2csa* than putrescine

SpdSyn takes Put to Spd, consuming one SAM in the process **Figure 29.** Spd concentrations are found to be twice as high in the parasite than Put, likely due to Spd's role in hypusinating eIF5A[109]. Together, both points suggest that it benefits the parasite to readily convert Put to Spd. Conditions hypothesized to increase SAM pools were earlier demonstrated to induce a switch to *var2csa* (**Figure 26**), whereas conditions hypothesized to decrease SAM pools either induced no switch in *var* expression (**excess Ser conditions, Chapter 2**) or the heterogenous expression of many *var*s across the population (**PfSAMS KD, Chapter 3**). Having learned that Spd supplementation induces a switch to *var2csa* (**Figure 33**), I tested how supplementation with Put would affect *var* expression.

Previously, van Biljon et al. 2018 found that they were able to culture *P. falciparum* in the presence of 2mM Put, ~10 fold higher than the molar concentration that I was able to culture with Spd and ~1000 fold higher than the amount in human serum **Table 2**[110]. As has been demonstrated, parasites are able to take up exogenously added Put [109,110,111]. I confirmed the viability of parasites at this concentration. Following my hypothesis regarding SAM pools, exogenously added Put should lead to increased SAM consumption. As I wanted to test the ability of added Put to hypothetically lower SAM pools, I decided to use this exaggerated concentration in assaying *var* expression, theorizing that any resulting phenotype would be easily observable by qPCR at the population level. I directly compared supplementation with Put alongside cultures where 100uM Spd was added.

Results of one representative set of cultures are shown in **Figure 34**, with replicates in **Appendix**. Overall, the Put-treated cultures did not see a dramatic shift in *var* expression at the population level. There was some activation of *var2csa;* however, this was relatively similar to that seen in the MCM control in which no Put was added. Comparatively, 100uM added Spd induced a large shift to *var2csa* at the population level in the time frame tested (three weeks).

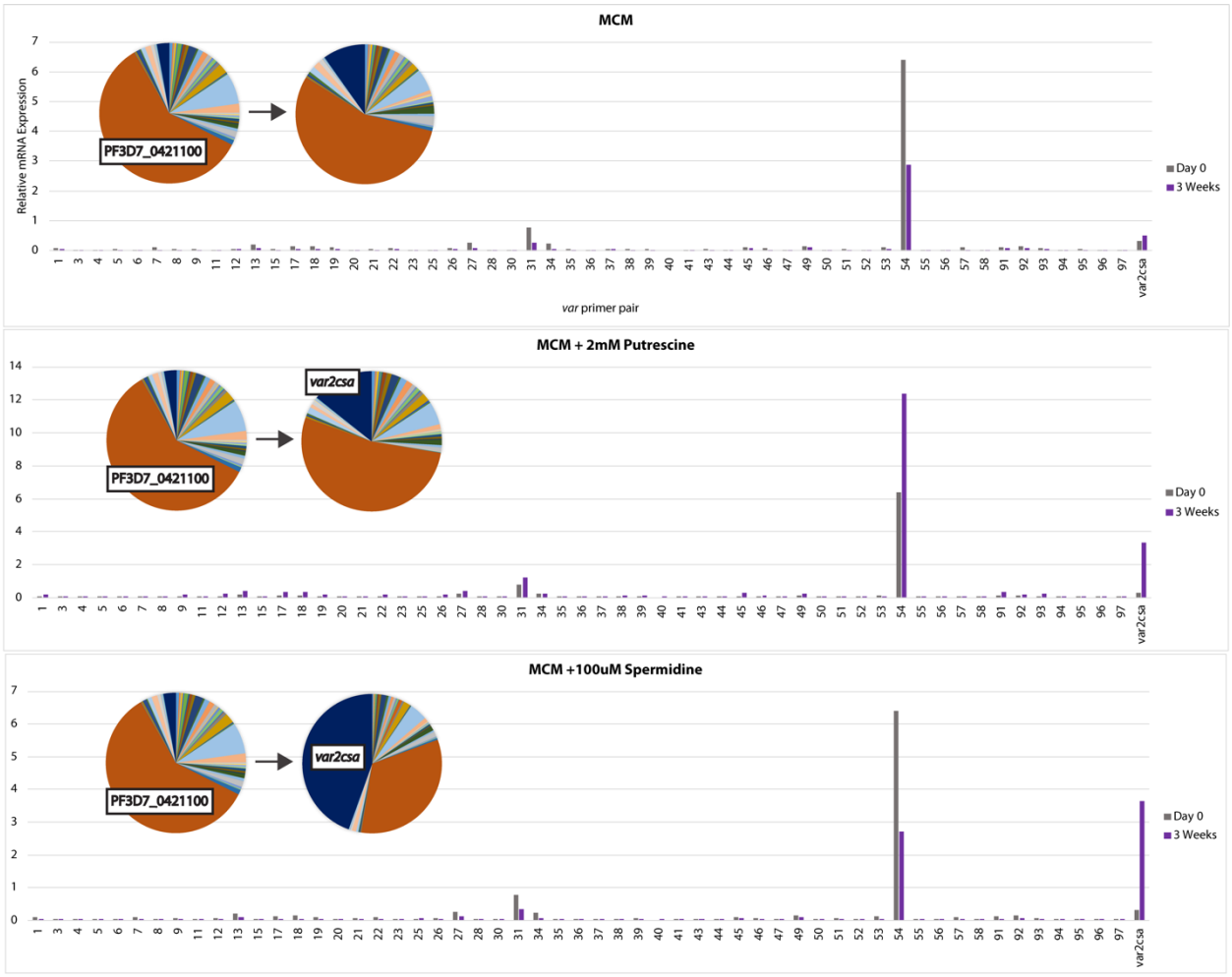


Figure 34. Supplementation with spermidine induces a more robust switch to *var2csa* than putrescine *var* gene expression as assayed for each condition is shown as a pie-chart and bar graph, with each slice of the pie and each numbered primer pair from Salanti et al. 2013 on the x-axis representing the fraction of the total *var* mRNA pool transcribed from each *var* gene. Gray bars represent Day 0 timepoint, or the *var* expression of the parent culture before it was split across different nutrient conditions. Purple bars are the *var* expression at the 3 week timepoint.

4.6 Supplementation with spermidine and putrescine does not significantly affect parasite growth

I found that *P. falciparum* could be cultured in a significantly higher concentration of Put than Spd. Parasites grown at mM concentrations of Spd were observed to die off after only a few growth cycles. To test the potential effect of concentrations that were used long-term for the above experiments on growth rate, I ran growth assays following the protocol outlined in **Methods**. Results are pictured for (**A**) 10uM Spd, (**B**) 100uM Spd, and (**C**) 2mM Put in **Figure 35**. No apparent growth phenotype was observed for any of these conditions relative to untreated controls (MCM) over a period of at least one-week growth. Log growth numbers used to generate plots are shown in **Appendix**.

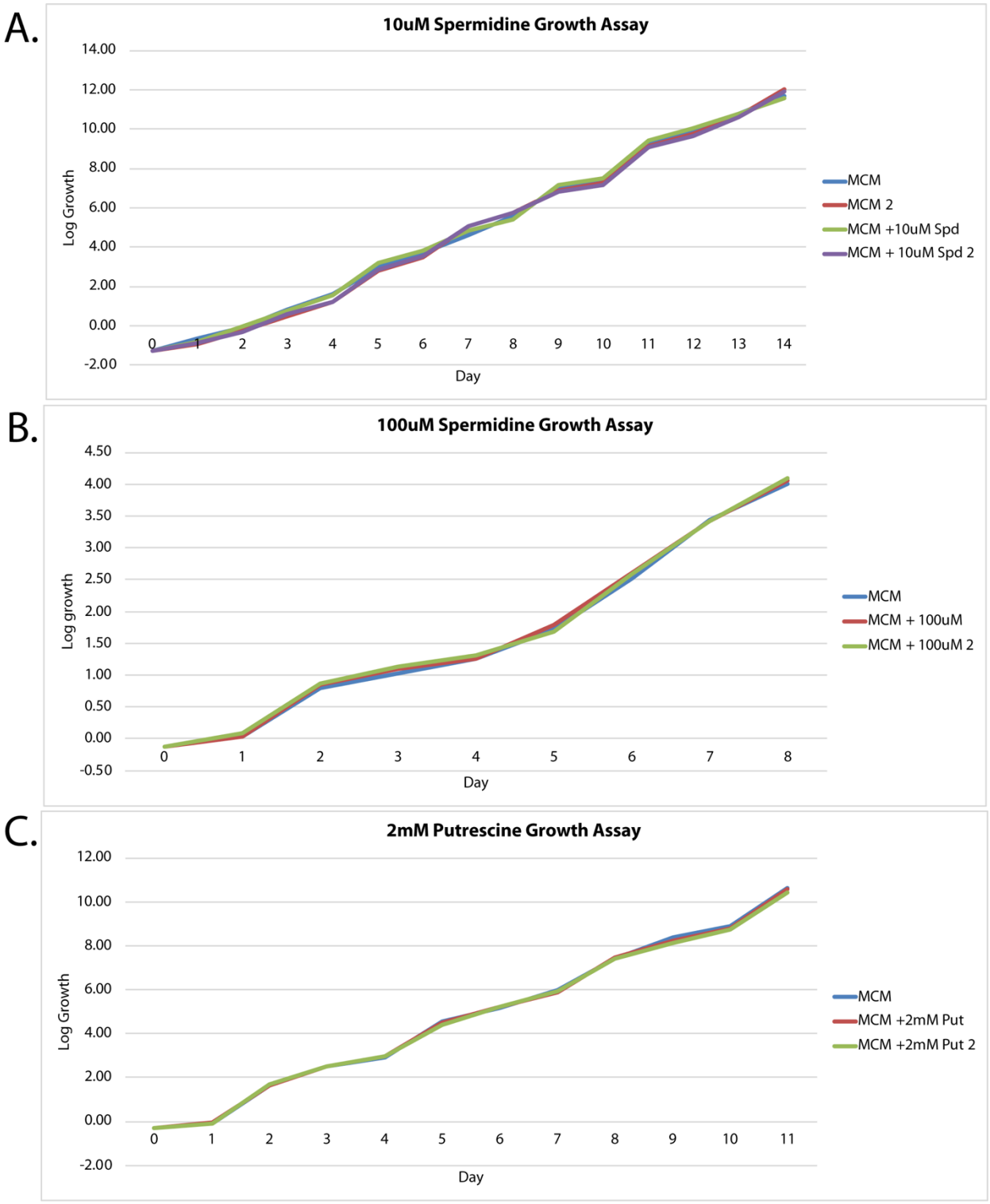


Figure 35. Supplementation of spermidine and putrescine in the growth media of *P. falciparum* does not significantly alter its growth rate Parasites were cultured in the conditions tested **A.** 10uM Spd **B.** 100uM Spd and **C.** 2mM Put over the course of 8-14 days (x-axis). Parasitemia was measured by flow cytometry and log growth (y-axis) was calculated following the protocol in **Methods.**

4.7 The effects of amino acid depletion and spermidine supplementation on *var* expression are not additive

While distinct pathways, amino acid starvation and polyamine synthesis can be compared by their similar ties to the innate immune response and downstream effects on translational fidelity **Figure 29**. Given that depletion of Arg and Trp (**Figure 31**) and exposure to exogenously added Spd (**Figure 33 & Figure 34**) induced a coordinated switch to *var2csa* at the population level, I wanted to test if applying both conditions simultaneously would exaggerate the phenotype. I hypothesized that this combination would serve as one possible way of mimicking localized environmental conditions resulting from M2 macrophage recognition. I postulated this could either push the parasites to switch into the *var2csa* sink node further from the effects of the conditions of eIF2α-p and eIF5A-h on translation of mRNAs with 5' uORFs (**Background**), or induce a switch in *var* expression at the population level away from *var2csa,* as would occur if the parasite were to switch its PfEMP1 presentation to avoid antibody detection.

To test this hypothesis, I cultured parasites in AA MIM with 100uM supplemented Spd, alongside controls in MCM and AA MIM untreated over the course of six weeks. Results are shown in **Figure 36.** A more profound switch at the population level to *var2csa* was not seen for the cultures exposed to both AA MIM and Spd supplementation relative to that seen for parasites cultured in AA MIM alone. A profound switch in *var* expression apart from *var2csa* was also not observed. Nevertheless, parasites cultured under both conditions have a more subtlety heterogeneous *var* expression pattern at the latest timepoint (six weeks) in comparison to those grown in AA MIM (**Figure 31**) and 10uM or 100uM (**Figure 33**) added Spd alone (visualized as the higher red bars on the bottom plot of **Figure 36**). This was true across two replicates, shown in **Appendix**. Importantly, while intriguing and seemingly reproducible, this phenotype is subtle enough to be attributed to artifact. Further experimentation is needed to confirm its significance.

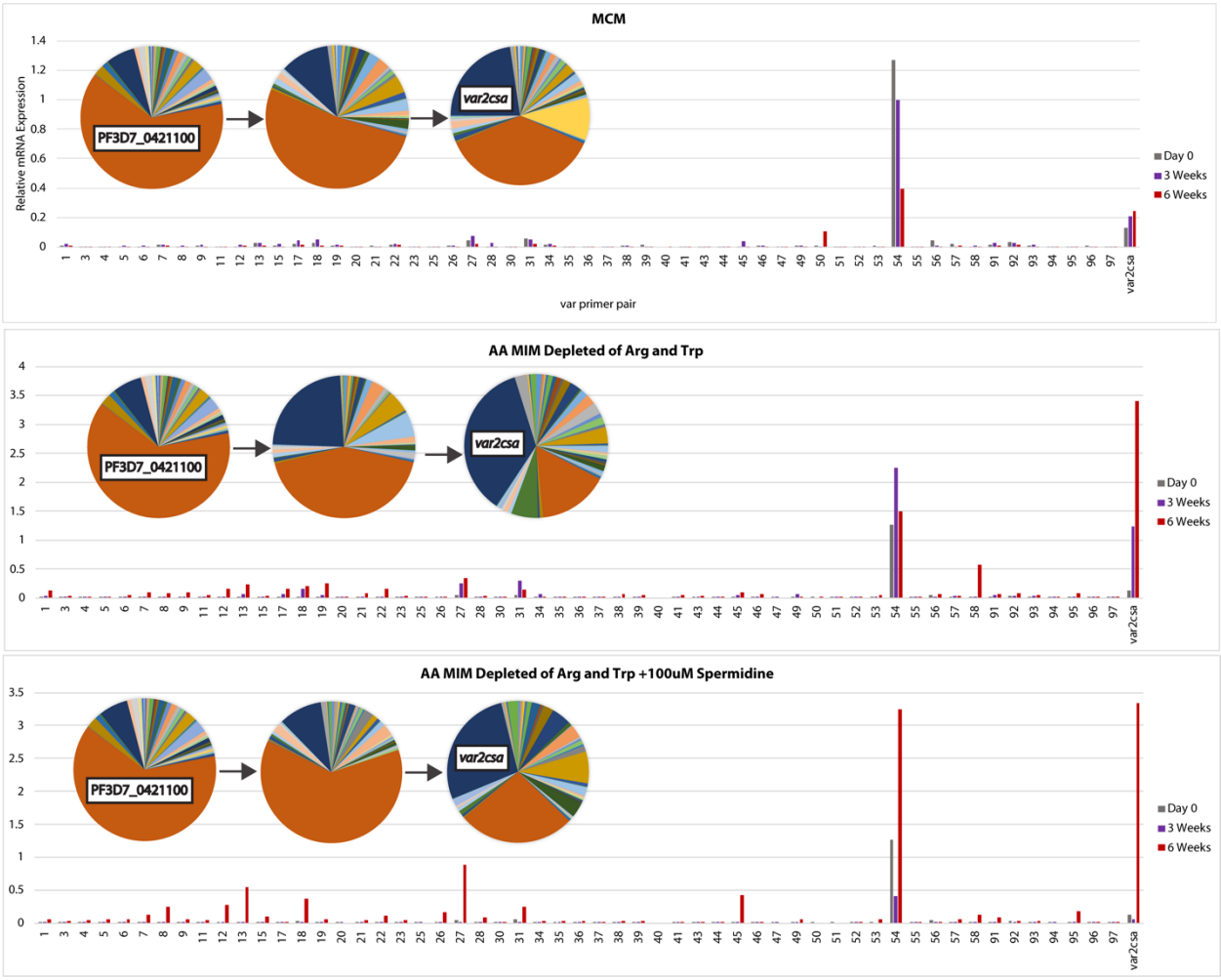


Figure 36. The effects of amino acid depletion and spermidine supplementation on *var* expression are not additive *var* gene expression as assayed for each condition is shown as a pie-chart and bar graph, with each slice of the pie and each numbered primer pair from Salanti et al. 2013 on the x-axis representing the fraction of the total *var* mRNA pool transcribed from each *var* gene. Gray bars represent Day 0 timepoint, or the *var* expression of the parent culture before it was split across different nutrient conditions. Purple and red bars are the *var* expression at 3 and 6 week timepoints, respectively.

Discussion

Early clinical reports have suggested that M2, anti-inflammatory macrophages play an important role in the innate immune response to *P. falciparum* infection[6]. This macrophage subset influences their local microenvironment through the depletion of AAs such as Arg and Trp through arginase-1 and IDO activity, respectively, alongside an increase of polyamines and polyamine-precursors (**Figure 29**)[106]. Recent metabolomics studies have demonstrated significant Arg and Trp depletion, accompanied by an increase in Kyn, in the blood of mice and human patients with *P. falciparum* infection[133]. Further supporting the clinical significance of these pathways in understanding the pathogenesis of *P. falciparum.*

In **Chapters 2 and 3** I demonstrated that growing *P. falciparum* either in conditions of excess choline to depleted serine, or by hypothetically modifying the levels of SAM and SAH using overexpression constructs for SAMS and SAHH, causes a switch in *var* genes to occur at the population level. In both cases, this switch was pointedly to the same *var* gene, *var2csa*, phenocopying an earlier set of experiments where the activity of associated HMTs was altered **Figure 26**. Taken together, the results of these experiments suggests a model of *var* switching that operates through the intracellular levels of SAM/SAH and their subsequent effect on HMT activity **Figure 27**. The results are also profound on a larger scale, supporting the novel idea that *P. falciparum* can alter its antigen expression in response to environmental nutrient fluctuations. The capacity of *P. falciparum* to alter expression of other genes in response to its environment has been previously demonstrated (See **Chapter 1.11**). Further, I have hypothesized that *P. falciparum* can sense when the host immune system first begins to recognize it via environmental cues resulting from antibody recognition, and respond by switching *var* gene expression. This would ultimately describe a mechanism that allows this parasite to most efficiently utilize its limited repertoire of *var* genes and suggests a possible molecular mechanism underlying clinically observed waves of parasitemia over the course of chronic infections, illustrated in **Figures 4 and 5**. The experiments presented in this chapter were designed to test this hypothesis, specifically with regards to macrophage recognition.

Depletion of both Arg and Trp (**Figure 31**) and supplementation with Spd (**Figure 33**) all induced a coordinated switch in *var* expression at the population level to *var2csa*. This further phenocopies the original results of altering HMT activity in Ukaegbu et al., 2015, and those presented in **Chapters 2 and 3**. Thus, independently, all of the conditions tested long-term in this thesis (apart from knocking down SAMS expression (PfSAMS KD) and supplementing with Put) were found to induce a coordinated switch at the population level to *var2csa.* These are presented altogether in **Figure 37**.

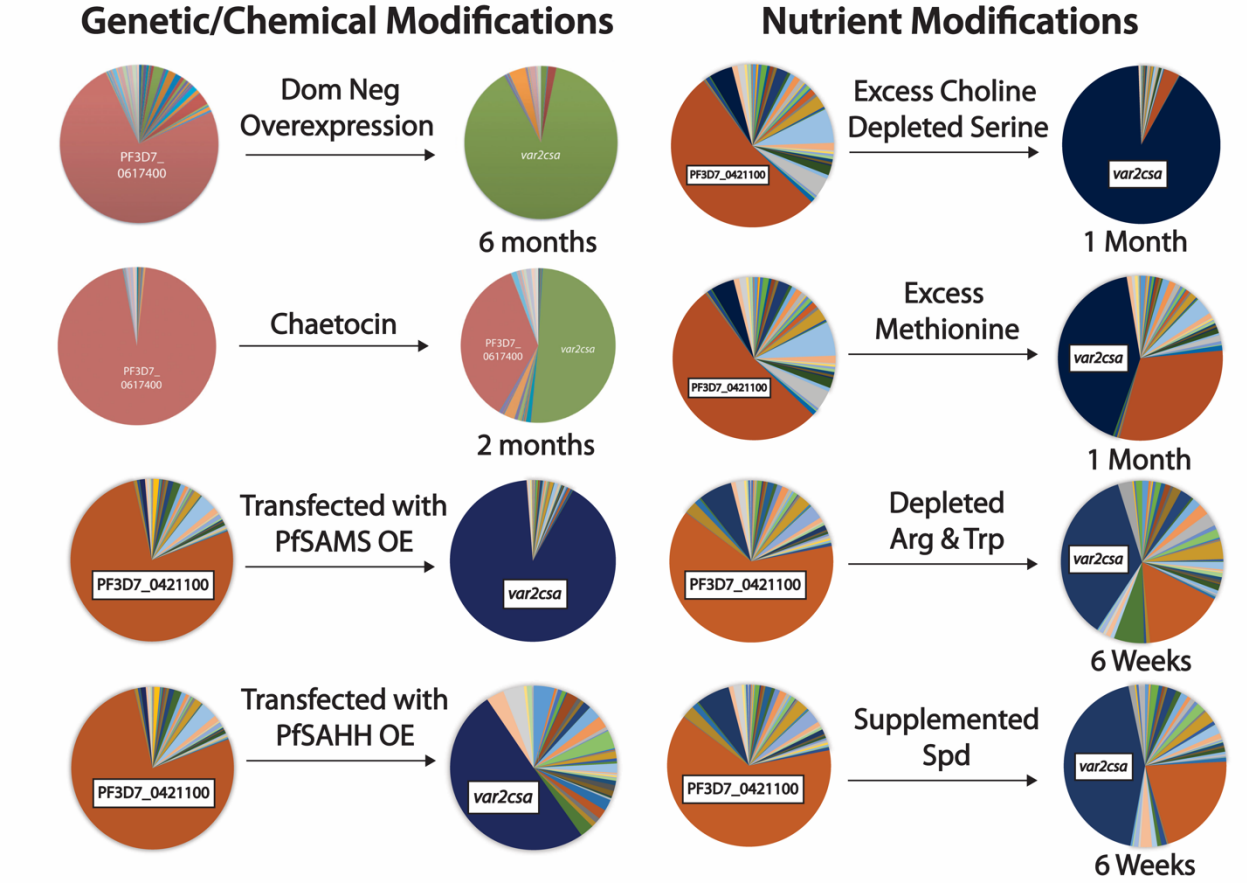


Figure 37. Conditions that induce a coordinated switch at the parasite population level to *var2csa* Presented is a list of both genetic and chemical modifications (left) and nutrient modifications (right) that led to a coordinated switch in *var* expression at the population level to *var2csa***.** All conditions listed except the dom/neg and chaetocin (originally from Ukaegbu et al., 2015) were tested by the author and are reported in this thesis. These results phenocopy those seen in Ukaegbu et al., 2015 in response to genetic (dom/neg) or chemical (chaetocin) modifications of HMT activity. First two rows of *var* panels in the left column taken from Ukaegbu et al., 2015.

The sets of conditions tested in **Chapters 2 and 3** each have direct connections to SAM metabolism, revisiting **Figures 14 & 27**. To this author's knowledge, there is no direct connection between the depletion of Arg and Trp and SAM metabolism. On a molecular basis, the specific induction of *var2csa* could tie back to the phosphorylation of eIF2α in response to the drop in AA levels. *var2csa* is unique, in that its gene transcribes an mRNA with a 5' uORF, allowing for transcriptional activation without translation. Laboratory colleagues have seen increased eIF2α-p in response to Arg and Trp depletion in *P. falciparum* parasites (Dr. Francesca Florini and Joe Visone, Personal Communication). Conditions of eIF2α-p have been shown to upregulate translation of mRNAs with 5' uORFs that would ordinarily be flagged for degradation via NMD pathways.[90,102] If eIF2α-p, in response to Arg and Trp depletion, would indeed select for VAR2CSA translation, it would explain the phenotype in **Figure 31,** as the levels of steady state *var2csa* mRNA (measured by qPCR) would be more consistent and stable (as they would not be subject to NMD). Environmentally, M2 macrophages and IDO activity have largely been observed at the maternal-fetal interface[118]. Therefore, the presence of a 5' uORF at the *var2csa* locus, coupled with its function in the placenta, make it a possible target for upregulated translation upon IDO-mediated eIF2α-p. However, the effects of Arg or Trp depletion and eIF2α-p on VAR2CSA translation remain to be definitively assessed, and further work is needed to determine if this model would explain the results presented in this thesis.

Babbitt et al., 2012 demonstrated that, when starved of Ile, *P. falciparum* parasites enter a "hibernatory" state of slowed growth, accompanied by rapid phosphorylation of eIF2α[97]. Mass spectrometry analysis of parasites under Ile starvation found disrupted levels of metabolites involved in one-carbon and SAM metabolic pathways. These findings have additional physiological relevance in light of clinical readings in malaria endemic regions, where malnourished individuals, predominantly children, contain low or undetectable levels of Ile.[97] My experiments attempting to mimic Ile starvation for up to 48 hours using an analog, 4-thiaisoleucine, did not yield parasites that had visibly switched *var* expression (**Figure 30**). This may further suggest that the IDO-like depletion of Arg and Trp was uniquely able to induce

var2csa, as it is similar to conditions in a placenta. This experiment, however, was technically limited in its inability to analyze *var* expression at the moment parasites were released from a hibernatory state by removal of TI. Mathematical models suggesting the existence of a "switch intermediate" or "sink node", postulated to be *var2csa*, also support a switching network, in which the majority of parasites that switch into the sink node will actually switch back to the previous *var* that was "on" instead of a previously silent one[55]. This is likely due to an epigenetic memory, predisposing the *var* which was already on for the easiest re-activation[36, 89]. Therefore, very few (if any) parasites that may have switched to *var2csa* or a different *var* under conditions of Ile starvation will have been outgrown by parasites that may have switched into the sink node, *var2csa*, and away from it by the time RNA could be extracted for analysis – here up to a week post-TI exposure. This may also serve as an explanation as to why my nutrient growth experiments take long periods of time (2-6 weeks) to observe the *var2csa*-activation phenotype, as it takes weeks to select for enough parasites that have switched *var2csa* on to be able to see meaningful change in expression at the population level by qPCR.

Spd supplementation has a connection to both SAM metabolism and immune recognition. SpdSyn takes Put to Spd, with input of one SAM via AdoMetDC **Figure 29**. *P. falciparum* can take up Spd from its external environment[109]. Spd is present at twice the concentration of Put in mature trophozoites, likely as it is required to hypusinate eIF5A. Supplementing the growth media with Spd would allow the parasite to hypusinate eIF5A without consumption of SAM, putatively increasing intracellular SAM pools. Activated M2 macrophages increase local concentrations of polyamines and may indirectly increase polyamines through the degradation of Arg by arginase-1, leading to increases in Orn, a polyamine precursor (**Figure 29**) [106].

The role of eIF5a-h must also to be considered. eIF5A has similarly been found to impact the translation of mRNAs with uORFs. However, while eIF2α-p increases translation of downstream open reading frames (dORFs), eIF5A-h decreases translation past the uORF. Thus, in conditions of lower polyamines, ribosomes will more frequently bypass the uORF stop codon and translate dORFs, while in conditions of higher

polyamines, ribosomes will drop off at the uORF stop codon [123,129]. In the placenta, polyamines are in high demand in support of developing fetal cells [130,131]. The results of this chapter do not rule out either possible model for eIF5A's role in *var2csa* activation, mentioned in the **Background** section, as additional work is needed in order to determine the effects of Spd supplementation and increased levels of eIF5A-h on VAR2CSA translation. Here, the supplementation with Spd appeared to induce a stable induction of *var2csa* mRNA, as measured by qPCR. This could be indicative of the conditions being selective for translation of VAR2CSA, but could also be the impact of a higher SAM/SAH ratio.

Supplementation with Put in these preliminary experiments did not induce the same robust switch to *var2csa* (**Figure 34**). This suggests that the consumption of SAM required to synthesize Spd may lower SAM pools and fit the overall suggested model **Figures 14 & 27**. Induction of many *var*s across the population as was observed with the PfSAMS KD lines was not observed in regards to Put. This phenotype more resembled the lack of *var* switching seen under growth conditions of excess serine (**Figure 10**). It is important to note that the conditions tested may not have been optimal for observing a phenotype at the population level. I tested 2mM Put, which is far above the upper limit found in human serum (**Table 2**). I used this exaggerated concentration as a way to better examine the role of the SAM-consuming synthesis of Spd. By using an excessive amount of Put, I sought to push the further synthesis of Spd, thereby consuming larger amounts of SAM, and, theoretically, lowering overall SAM pools. In support of my hypothesis, a switch to *var2csa* was not observed using 2mM Put; future experiments should look at the effect of more physiologically relevant concentrations of Put, as might be observed in a placental environment. Furthermore, high levels of Put have been shown to inhibit hypusination of eIF5A, which, if in keeping with the mammalian model described in **Background**, may actually increase translation of VAR2CSA[134]. Future work will be needed to fully elucidate the roles of Put and Spd on *var2csa* at the level of translation.

Preliminary experiments testing the combined effect of both treatments, meant to mimic immune recognition, did not induce a more robust switch to *var2csa* than

either treatment individually (**Figure 36**). However, further experiments could attempt growth over longer durations. Cultures that were grown in AA MIM with supplemented Spd did show some evidence of a more heterogenous *var* expression profile emerging at a low level at the latest timepoint. This may be indicative of parasites that are in the process of switching away from the *var2csa* sink node into previously silent *var*s as would occur in an immune recognition model (revisit **Figure 5**), but further experimentation is needed to confirm this phenotype.

In addition to those already mentioned, several future experiments could be done to further investigate an immune recognition model of antigenic variation in *P. falciparum*. First would be a set to more directly assess the impact of SAM levels (in relation to polyamine synthesis) in inducing a switch to *var2csa.* DL-α-difluoromethylornithine (DFMO) is a compound that blocks ODC activity, functionally shutting down the polyamine synthesis pathway **Figure 29**. If DFMO is added alone, parasites will arrest and die, as they can no longer synthesize the required amount of polyamines *de novo*. However, if enough Put or Spd is added exogenously in the presence of DFMO, parasites will grow normally[110]. Here, DFMO could be used to block all *de novo* polyamine synthesis, and, through polyamine rescue, might allow for control at the SAM-consuming step by addition of either Put or Spd. In this experiment, DFMO-Put treated parasites would need to consume large amounts of SAM to produce Spd for eIF5A-h; conversely, DFMO-Spd treated parasites would conserve SAM from the addition of Spd. In contrast with earlier experiments, shutting down the pathway at the ODC step (**Figure 29**) would more definitively implicate the SAM-consuming SpdSyn reaction in resulting *var* phenotypes, as background synthesis and build-up of Put would be more limited. I ran preliminary attempts at this experimental setup, testing 2mM DFMO with 2mM Put or Spd rescue following van Biljon et al. 2018, but the results on *var* expression were inconclusive. This may largely be attributed to a growth phenotype I observed while attempting to scale the cultures up to a volume large enough for RNA extraction. This phenotype could have interesting implications with regards to this immune recognition model, and is being explored further by our lab.

A second possible experiment would be to co-culture parasites with monocytes and assay *var* expression in response to a localized immune response. Using an Antibody-Dependent Cell-Mediated Inhibition (ADCI) assay developed by Bouharoun-Tayoun and Druilhe, 2015, *P. falciparum* can be cultured in the presence of blood monocytes and immunoglobulins (IgG)[112,113]. This assay is designed to mimic *in vivo* immune recognition; culturing iRBCs with antibodies for a selected variant of PfEMP1 will trigger activation of monocytes. If my proposed hypothesis is correct, this should induce *var* switching to variants other than *var2csa*.

The results of this chapter add additional evidence against the paradigm of stochastic switching in *P. falciparum*, by further showing that environmental changes can impact antigenic gene expression. The results also show preliminary support for the idea that antigenic variation in *P. falciparum* follows an immune recognition model, in which parasites can sense environmental cues resulting from antibody recognition and respond by switching *var* gene expression. However, most of these experiments were only able to push parasites into the hypothetical sink node, *var2csa*, not to a previously silent *var*, which would be indicative of a complete switch in *var* and PfEMP1 expression.

In the final Chapter (**5**) of this thesis, I will take the entirety of the results presented in **Chapters 2-4** together in consideration of two models, the first of which will center on intracellular SAM metabolism in describing a promoter competition model, governing *var* switching through *var2csa* as the hypothetical “sink node” proposed by mathematical models. The second will elaborate on this immune recognition model. Both models will, from differing vantage points, attempt to address the question as to how *P. falciparum* parasites manage to cause persistent, long-term infections with a repertoire of only ~60 *var* genes per genome, by proposing precise switching mechanisms reliant on external signals.

Chapter 5

Perspectives

Thesis Summary

Despite remarkable progress in mosquito vector control and other interventions, malaria remains a significant global threat, in part due to insecticide resistance by mosquitoes, drug resistance to antimalarials, and the lack of an effective vaccine. [1] Of the five *Plasmodium* species that cause malaria in humans, infections by *P. falciparum* account for the highest number of deaths [1]. A defining feature contributing to the pathogenicity of *P. falciparum* is its ability to express a series of cytoadhering proteins on the surface of the iRBC. PfEMP1 is expressed in knob-like structures on the RBC surface, facilitating cytoadherence to vascular endothelial receptors, and thereby allowing the parasite to leave peripheral circulation and sequester away in the microvasculature of various organs. Variant forms of PfEMP1 are encoded by a large, multicopy gene family called *var*[12–14]. Each *P. falciparum* genome possesses ~60 *var* genes [11 15]. Their expression operates in a mutually exclusive manner, such that, in each parasite, only one gene expressing PfEMP1 is active over many cycles of asexual replication, while the other 59 remain transcriptionally silent. [16] Common histone marks found in higher eukaryotes responsible for regulating gene expression were found to be uniquely distributed around *var* genes in *P. falciparum* [32–34].

Measured human *P. falciparum* infection shows waves of increasing and decreasing levels of parasitemia in the blood over the course of chronic disease progression (**Figure 4**). Over the course of infection, the host's immune system will mount an antibody response to the predominately-expressed variant of PfEMP1 in the growing parasite population. This response will overwhelmingly reduce the circulating parasite population, often to undetectable levels. The cycle will repeat, as a sub-population of parasites that were expressing or switched to a novel *var* gene, and thus far "unseen" variant of PfEMP1, will clonally expand to high parasitemia until another antibody response is mounted. Thus, by varying the expression of their displayed

antigen, *P. falciparum* can evade the immune response and maintain chronic infections by moving through the *var* repertoire (**Figure 5**). [2,10]

The molecular mechanisms that control activation and silencing of individual *var* genes, as well as the means by which the entire switching process is coordinated, presently remain incompletely defined. In contrast with African Trypanosomes that have thousands of *vsg* genes in their antigenic repertoire to switch between, *P. falciparum* has only ~60. This begs the question as to how this species manages to cause persistent, long-term infections, and suggests that it must utilize a more elegant and precise switching mechanism. The current paradigm proposes that these parasites evolved an internal clock programmed to time a switch in *var* expression with the time it takes the host to mount an antibody response, independent of any external stimuli. [19] In **Chapters 2 and 4** I illustrated observed switches in *var* expression at the population level over time in response to altered nutrient conditions, directly challenging this paradigm by instead suggesting that this organism possesses the ability to switch in response to environmental changes. In all cases, the switch was coordinated at the population level to *var2csa*. This phenocopied (**Figure 37**) previous work in our lab, reported in Ukaegbu et al., 2015, where interference with the epigenetic regulation of *var* activation and silencing induced expression of *var2csa*. *var2csa* is a unique member of the *var* family for several reasons. Namely, it is universally conserved across parasite isolates while the majority of *vars* are not, despite encoding a variant of PfEMP1 that exclusively binds a placental ligand. While the niche its encoded protein can be used in is small, *var2csa*'s genomic organization allows it to be transcriptionally active while translationally repressed, and transcripts have been detected in non-pregnant individuals. [84,85] The aforementioned properties of *var2csa,* coupled with these experimental results, support the idea that *var2csa* may serve a separate function within *P. falciparum* apart from encoding a form of PfEMP1. Mathematical models of *var* gene switching predict the presence of transiently active genes, or "switch intermediates," that add a level of uniformity and coordination to the process. [55] These points all suggest that *var2csa* serves as the presently unidentified "switch

intermediate," though this (or the identity of any sink node) has not yet been firmly established. [55]

Several studies in eukaryotic systems have provided evidence for a direct link between nutrient uptake, SAM metabolism, histone methylation, and gene regulation[58,63,64,66,67], suggesting histone methylation dynamics appear to be sensitive to fluctuations in the intracellular ratio of SAM to SAH. Conditions tested in **Chapters 2 and 4** that induced *var2csa* all intersected with SAM metabolism, and would be hypothesized to increase the level of SAM/SAH (**Figure 37**). In **Chapter 3,** I took a genetic approach to test this by modifying the expression of two key enzymes in SAM metabolism: PfSAMS and PfSAHH. My results suggest a role for SAM pools in influencing *var* switching. Specifically, conditions where the level of SAM/SAH is high induce a robust switch into the theorized "sink node" *var2csa*. Conditions where PfSAMS expression was knocked down led to a distinct phenotype, where multiple *var*s were robustly being at expressed at the population level, none of which were *var2csa*.

A complete summary of my findings in this thesis are presented in **Figure 38**. These illustrate a model where *P. falciparum* possesses sensing machinery, capable of altering antigen expression in response to environmental fluctuations through SAM pools. Conditions where SAM pools are hypothetically high induce a convergence to *var2csa* at the population level. Conditions thought to lower SAM pools, however slightly, did not induce any visible *var* switching phenotype. In the extreme case of the PfSAMS KD line, dramatically decreased SAM pools led to the activation of multiple *var*s, possibly indicating a loss of mutually exclusive *var* expression.

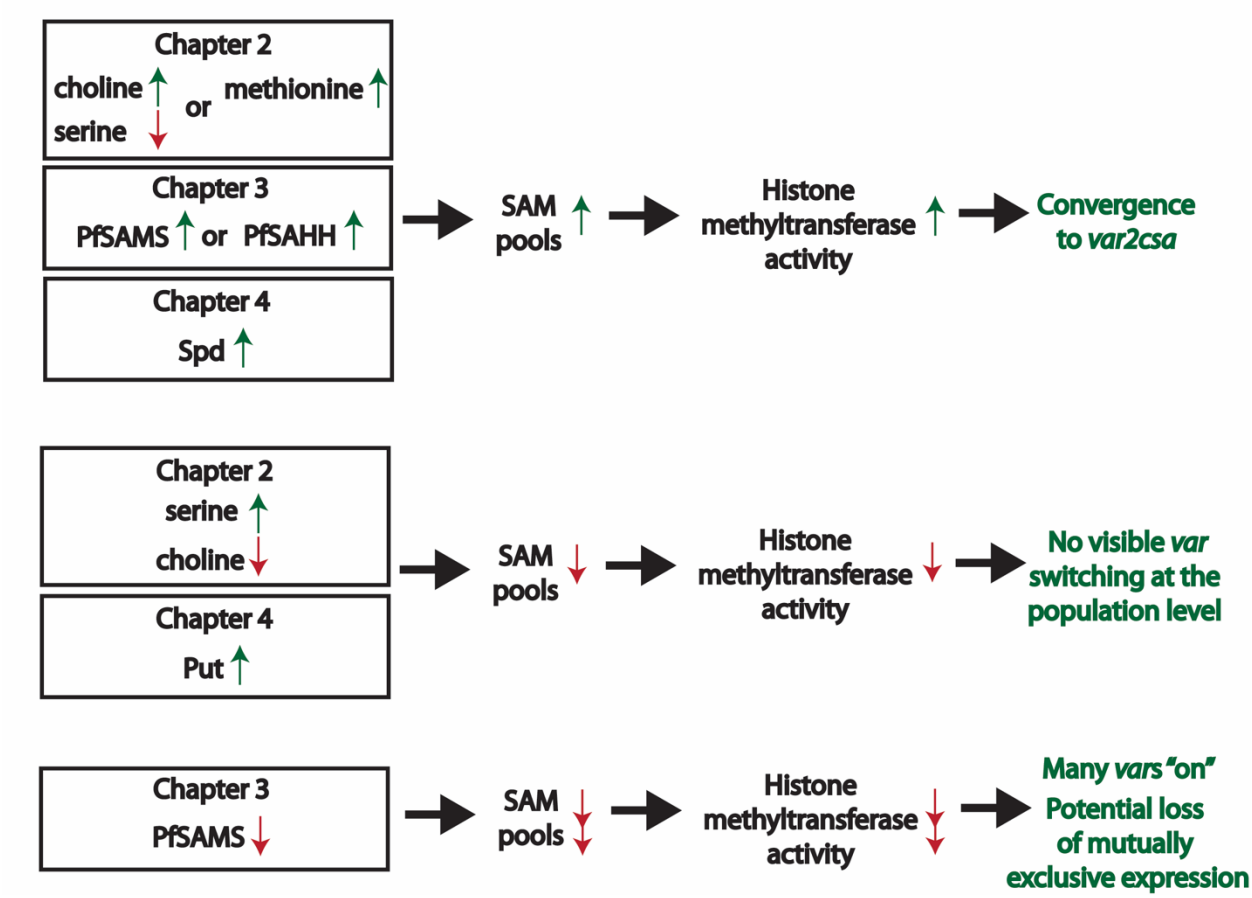


Figure 38. The results in this thesis suggest a model where *P. falciparum* possesses sensing machinery, capable of altering antigen expression in response to environmental fluctuations through SAM pools Conditions where SAM pools are hypothetically high (Top) induce a convergence to *var2csa* at the population level. Conditions thought to lower SAM pools (Middle), even if slightly, did not induce any visible *var* switching phenotype at the population level. Dramatically decreased SAM pools from a KD of PfSAMS (Bottom) led to the activation of multiple *var*s, possibly indicating a loss of mutually exclusive *var* expression.

A promoter competition model of *var* switching in *P. falciparum*

Analogous to the mutually exclusive expression of *var* genes is the olfactory receptor choice in sensory neurons. Each sensory neuron only expresses one of over 1000 odorant receptor genes, an essential feature that ultimately determines its functional identity and site of projection in the brain[135]. The current model governing olfactory receptor choice posits either competition among the many unique genes for a single expression site, or a single enhancer element that assures only one member is chosen for expression[135]. More generally referred to as a phase-separation model for transcriptional control, invoking the specific idea of compartmentalization, this theory describes a competition amongst gene families for "super-enhancer" sites of transcription. Here, super-enhancers are considered to be cooperative, high density assemblies of transcription factors, transcriptional co-factors, chromatin regulators, non-coding RNA, and RNA Pol II[136,137]. As reviewed in Lomvardas et al. 2006, *vsg* expression in trypanosomes, as well as antibodies and T cell receptors in lymphocytes, regulate expression of a single gene through DNA rearrangements that place a given coding sequence into a single expression site[143-145]. In support of this model's application in *P. falciparum* are reports evidencing that *var* gene activation involves translocation of the locus to a specific region at the nuclear periphery[138–141]. However, the identity of specific super-enhancers acting in a phase-separated *var* gene transcriptional compartment are presently unverified.

The results described in this thesis suggest a role for SAM/SAH levels in driving the deposition of epigenetic marks (revisit **Chapter 1.5**), governing *var* gene activation and silencing, and thereby influencing *var* expression at the parasite population level. The overview of my experimental results presented in **Figure 38** suggests a broader model of "promoter competition" governing the activation and silencing of *var*s, wherein the deposition of epigenetic marks makes *var* promoters more or less competitive for a super-enhancing site of transcription. Factoring into the model are two previously characterized aspects of *var* expression. First, the switch in expression from one *var* gene to the next has evolved to occur at a low rate, allowing for the expansion of clonal *var* populations over many generations (seen as the discrete waves

of parasitemia in **Figures 4 & 5**). [2,24,25] Second, epigenetic memory predisposes the single *var* which is already active for the re-activation in the next replication cycle[36,89]. Furthermore, this framework will align with mathematical models that suggest the presence of a transiently active gene, or "sink node", in adding a level of uniformity and coordination to *var* switching.[55] Previous research outlined in **Chapter 1.7 and 1.8,** as well as the results of this thesis (**Figure 38**), strongly suggest that *var2csa* is serving as the "sink node".

An illustration of this model is presented in **Figure 39**, where in each box the arrow on the left represents a scale of less competitive promoters (bottom) to more competitive promoters (top) based on methylation levels. *var* genes are split into three sets: the active *var (varA)*, the sink node (*var2csa)*, and all remaining silent *var*s (*varB-n)*. **Figure 39A** conceives how a full switch from *varA,* through *var2csa*, to a previously silent *varB* would look within this model, alongside *var* panels (shown as pie charts) from an experiment (**Chapter 3.5, Figure 20**) in which this was observed. Here the active marks on *varA* maintain its position as most competitive, and will thus maintain its "on" state until conditions change. Following mathematical models, *var2csa*, as the sink node, is always second-most competitive, being the gene parasites will induce first when a switch is initiated. All the remaining silent *var*s, lacking marks for activation and kept in a state of condensed chromatin, will remain in a state of low competitiveness, as activation will require major epigenetic modifications. A switch is initiated once either the competitiveness of *varA* begins to lower, here speculated as a loss of active marks and/or increase in silencing marks at the active locus, from changing environmental conditions or HMT activity; or, the competitiveness of *var2csa* begins to rise from added epigenetic activation marks. As *varA* and *var2csa* reach a similar competitive level, as the sink node, *var2csa*, will now be activated by a portion of parasites in the population (**Figure 39A, middle**). By competing with the active *var*, *var2csa* lowers the threshold for activation of silent *var*s. Following my observations, as conditions of increased SAM/SAH persist, *var2csa* will become increasingly competitive, becoming active in more parasites across the population and remaining active once induced. This insinuates that conditions of decreased SAM/SAH, back to

steady state levels, would then subsequently push parasites out of the node and enable expression of a previously silent *var* (**Figure 39A, bottom**). In the figure this is visualized as the activation of many *var*s across the population, as that was what was observed in this thesis when SAMS expression levels were decreased (growing the PfSAMS OE off drug (pictured) or the PfSAMS KD). As this pie chart represents expression across the entire population, and not single parasites, this phenotype does not definitively represent a loss of mutually exclusive *var* expression, although this is a possibility that will be expanded upon further below. This bulk heterogenous population most likely represents small numbers of parasites, each expressing a singular, different *var* gene. As these heterogeneous *var* expression phenotypes were the result of genetic modifications (not nutrient levels) that would not occur naturally, it is possible these examples represent an exaggerated phenotype that is able to be observed by qPCR.

Given a selective enough bottleneck, such as immune recognition, an extremely small number of parasites will make the switch from *var2csa* to a previously silenced *var*. Considering the discrete waves of parasitemia seen over the course of human infection (**Figure 4**), it follows that the chance per parasite of switching *var*s, and PfEMP1 expression, is extremely low. The smaller the number of parasites that survive immune recognition, the more time they will have to expand until the cycle is repeated (**Figure 5**). In this first instance, any environmental changes would be very quick and transitory, pushing just enough parasites through a full *var* switch and past immune recognition.

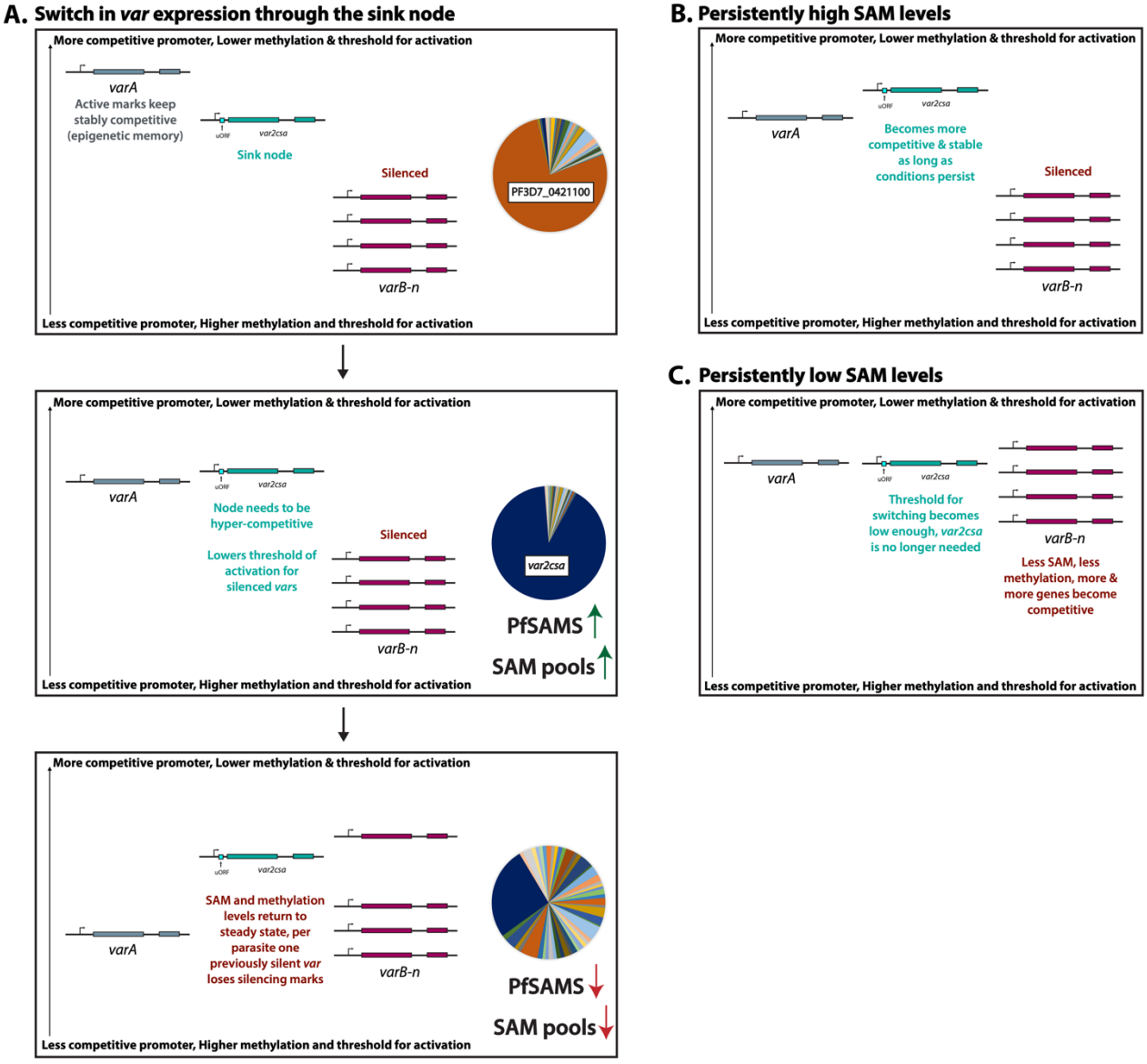


Figure 39. A model of promoter competition governing antigenic variation in *P. falciparum* A. A switch from the active *var, varA*, through the hypothetical switch node, *var2csa. var2csa* is hyper-competitive relative to the silenced *vars.* It facilitates switching by lowering the threshold to activation of silent *var*s. Pie chart *var* panels taken from Figure 20, top panel is the parent culture pre-transfection, middle is PfSAMS OE transfected line, bottom is the bulk heterogeneous population after 2 months off drug pressure **B.** Conditions where SAM and methylation levels are high, make *var2csa* more competitive and stably active as conditions persist. **C**. Lower SAM and methylation levels would not increase the competitiveness of the *var2csa* promoter. Drastically lower SAM and methylation levels are capable of lowering the threshold for *var* activation across the board, possibly eliminating mutually exclusive expression.

The rest of **Figure 39** considers other phenotypes observed in this thesis within this model of promoter competition. Experimental conditions thought to increase SAM levels and HMT activity all induced stable expression of *var2csa* (**Figure 37**). Therefore, in this model, environmental conditions that would elevate SAM levels would push parasites into *var2csa*, the sink node, as shown in **Figure 39B**. Following my observations, as these conditions persist, *var2csa* becomes increasingly competitive, becoming active in more parasites across the population and remaining active once induced. This is evidenced by the clones of the PfSAMS OE under blasticidin selection (shown as a pie chart in the middle box of **Figure 39A**), where, as long as SAMS expression was elevated, *var2csa* maintained its "on" state (**Figures 23 & 24**). Additional conditions matching **Figure 39B** are those of excess Choline to depleted Serine (**Chapter 2.2**), excess Methionine (**Chapter 2.4**), the PfSAMS and PfSAHH OEs (**Chapter 3.2**), and supplementation with Spd (**Chapter 4.4-4.5**). Further, this indicates that environmental conditions conducive to a complete switch in *var* expression would not maintain high levels of SAM. Not represented in **Figure 39** are the conditions tested where no visible switch was observed (**Figure 38, middle**). Here, theoretically, SAM levels were too low for *var2csa* to increase in competitiveness and be activated.

In contrast to these conditions was the PfSAMS KD line, where, as a result of the KD, the SAM levels were, theoretically, drastically lowered (**Figure 38**). Here, high expression of multiple *var*s was seen. This is fit into the model in **Figure 39C**. Decreased SAM and methylation levels would even the competitiveness of all promoters, and thus the threshold for activation for all *var*s, as deposition of regulatory epigenetic marks would be lowered. Owing to the lack of observable *var2csa* activation in these lines, if the threshold of activation is lowered enough that all promoters become equally competitive, parasites would not need to switch to *var2csa* before switching to previously silent *var*s. This also introduces the hypothesis that low enough SAM and methylation levels are capable of destabilizing *var* regulation, eliminating mutually exclusive expression. Additionally, there is precedent for this *var* expression phenotype elsewhere in the literature, in which a PfSET2 HMT knock out line was found to have leaky expression of all *var* genes across a population[37]. *P. falciparum*

heterochromatin protein 1 (PfHP1) and *P. falciparum* histone deacetylase 2 (PfHda2) are both silencers of gene expression, through the deposition of methyl marks and formation of heterochromatin, or through the removal of acetyl marks to allow for methylation, respectively. Independent knockdowns of either similarly led to leaky expression of all *var* genes within the population[41,42]. All are conditions wherein levels of methylation on histones would be decreased. Additionally, when the PfSAMS KD lines were cloned, maintenance of the heterogenous *var* expression pattern was observed across all clones that were screened (**Figure 25**). Rescue of PfSAMS expression in the KD lines further followed this model by pushing one replicate into the *var2csa* node **Figure 21**, top plot. However, this phenotype was not replicated, and will require further investigation.

This model proposes a key role for *var2csa*, acting as the sink node, in mediating switching between all other *var*s. Consistently, a colleague from our lab has successfully deleted *var2csa* in parasites and has observed them stuck expressing a single *var* gene for many months at a time (Zhang et al., personal communication). Her results imply that without *var2csa*, parasites lose the ability to switch *var* genes.

Many of the experiments presented in previous chapters singly induced *var2csa* **Figure 37**, as illustrated in **Figure 39B**. However, two specific examples followed the full switching model as presented in **Figure 39A**. First was the deselection of the PfSAMS and PfSAHH OEs, described in both **Chapter 3.5** and *var* panels shown for the PfSAMS OE in **Figure 39A**. The parent culture before transfection of the episomal construct was expressing one *var*. Once transfected, both constructs hypothetically increased SAM levels following the metabolic pathways outlined in **Figure 8** and, when selected on blasticidin, induced robust levels of *var2csa* expression at the population level. When I stopped adding selective drug pressure, parasites theoretically ceased increased expression of each enzyme, which hypothetically lowered SAM levels and allowed for *var* switching away from *var2csa*. Second, when the PfSAMS OE line was cloned, as described in **Chapter 3.7**, two out of 14 total clones were found to have a heterogeneous *var* pattern, in contrast with the majority that maintained singular *var2csa* expression **Figure 23**. Upon analyzing their PfSAMS expression by qPCR,

both clones were found to have meaningfully lower levels of expression than clones that had maintained *var2csa* expression **Figure 24**. This result mirrors that of the deselection experiments **Figure 39A**.

In summary, the promoter competition model postulates that the "on" *var* is most competitive, as it possesses active marks. It will remain stably "on" over many replication cycles due to epigenetic memory. The switch node, *var2csa,* is hyper-competitive relative to the silenced *vars.* It facilitates antigenic variation by lowering the threshold to activation of silent *var*s. Conditions where SAM and methylation levels are high make *var2csa* more competitive and stably active as conditions persist. Lowering SAM and methylation levels would not increase the competitiveness of the *var2csa* promoter. Drastically lower SAM and methylation levels are capable of lowering the threshold for *var* activation across the board, possibly eliminating mutually exclusive expression.

This possible method of regulating *var* switching is currently theoretical, but various experiments could be conducted to add further weight to the model. This thesis took a genetic approach to test this hypothesis. To confirm the effects on SAM pools, mass spectrometry could be done on parasite extracts grown in each condition, as well as the OE and KD lines. The results of cloning the PfSAMS KD line (**Figure 25**) strongly support the hypothesis that these parasites are expressing more than one *var*, at high levels, per cell, as each clonal population should reflect the *var* expression of an individual founding parasite. However, in order to determine if any of these lines contain parasites that are expressing more than one *var* simultaneously, single cell RNA sequencing could be performed on parasite extracts [117]. Methods to run single cell RNA sequencing on ring stage parasites, when *var* expression peaks, are currently being developed by our lab.

The experiments testing altered nutrient conditions in this thesis were preliminary in the sense that they were primarily testing what would happen to *var* expression under each condition. As listed in **Figure 38**, many of these induced stable activation of *var2csa,* that, over periods up to six weeks, did not change when the conditions applied were constant. In regards to the overall model, it would be

interesting to grow parasite populations under each condition, induce *var2csa*, and then remove it to see if *var* expression will change, similar to what was observed when the OE lines were grown off drug. If the model, as presented, is correct, maintenance of conditions that induce *var2csa* should keep parasites on *var2csa*. If the conditions are removed, parasites may switch *var* expression away from *var2csa*. What may factor in, however, is the observation by Mok et al. 2008 that *P. falciparum* parasites grown in culture have a tendency to activate *var2csa* over time in the absence of any known selective pressures[50]. This in and of itself (why parasites will activate *var2csa* in culture independent of selective pressure), is also an additional avenue for future investigation.

An immune recognition model of *var* switching in *P. falciparum*

As the growth and replication of *P. falciparum* distorts the morphology of the RBC, the necessitated expression of PfEMP1 on its surface can be seen by the host's immune system, thus serving as the primary antigenic determinant. [21] Over the course of infection, the host's immune system will mount an antibody response to the predominately-expressed variant of PfEMP1 in the growing parasite population. This response will overwhelmingly reduce the circulating parasite population, often to undetectable levels. The cycle will repeat, as a sub-population of parasites that were expressing or switched to a novel *var* gene and thus far "unseen" variant of PfEMP1 will clonally expand to high parasitemia until another antibody response is mounted. Thus, by varying the expression of their displayed antigen, *P. falciparum* can evade the immune response and maintain chronic infections by moving through the *var* repertoire. This phenomena of immune evasion by antigenic variation is believed to serve as the basis of the waves of parasitemia seen in chronic human infections that can persist for a year or more, as illustrated in **Figure 5**. [2,10]

In this model of immune recognition, a very small number of parasites make a full *var* switch, from the presently active *var*, *varA*, to a previously silent *var*, *varB,* by passing first through the sink node, *var2csa*. This was first presented in **Figure 39**, and is shown more simply in **Figure 40A**. The broad, overarching hypothesis I explored in this thesis was that *P. falciparum* can sense when the host immune system first begins

to recognize it via environmental cues resulting from antibody recognition, and respond by switching *var* gene expression. This would describe a mechanism by which parasites can switch expression of *var* genes exactly when needed, and with perfect timing, thus allowing them to utilize their limited repertoire of *var* genes with the utmost efficiency. This in turn would suggest that *P. falciparum* utilizes a more elegant and precise switching mechanism underlying antigenic variation, so far undescribed across other organisms.

Preliminary experiments testing altered nutrient conditions meant to mimic an immune recognition event, as described in **Chapter 4** of this thesis, were able to push parasites into the hypothetical sink node, *var2csa*, but not to a previously silent *var*. This is illustrated in **Figure 40B**. Only one instance, growing parasites in AA depleted media while supplementing Spd, induced a phenotype that may be indicative of *var* switching (**Chapter 4.7**). Further work is needed, however, to confirm that this is not just background noise detected by qPCR.

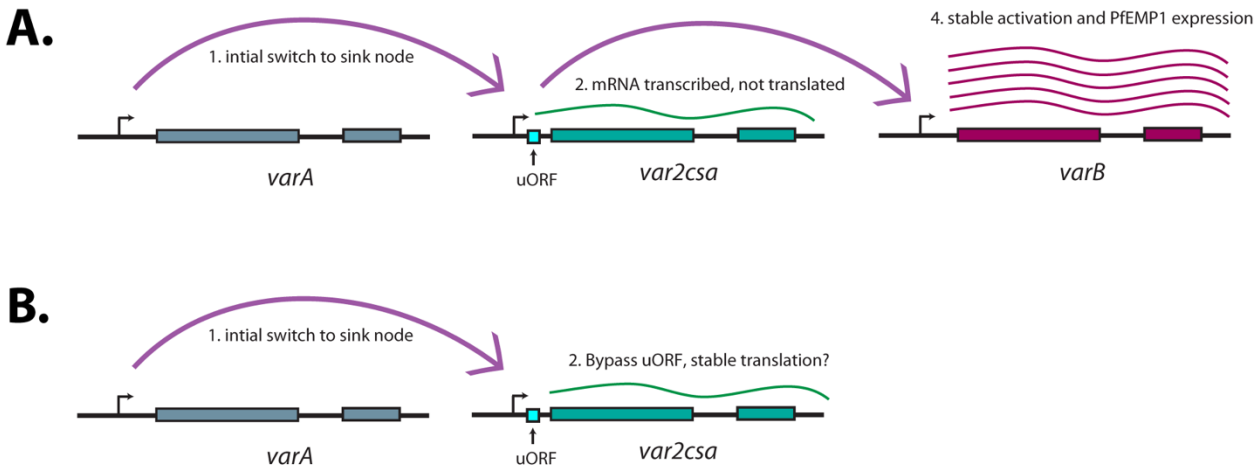


Figure 40. A model for *var* switching in *P. falciparum* A. a full switch in *var* genes, from the presently active *var*, *varA*, to a previously silent *var*, *varB,* by passing first through the sink node, *var2csa.* Current work in our lab suggests that, unless a placenta is around, *var2csa* is acting as the hypothetical sink node, being transcriptionally active but not translated. As environmental conditions change, following the model in Figure 39, parasites would switch on a previously silent *var*. **B.** phenotype observed for most of the environmental conditions tested in this thesis. I was able to induce stable expression of *var2csa*, with no induction of previously silent *var*s. Perhaps translation of *var2csa* was being activated, suggesting the conditions tested here are similar to those in a placenta.

In contrast with other *var* genes, *var2csa* can be translationally repressed even when transcriptionally active, due to the presence of a unique upstream open reading frame (uORF) (revisit **Figure 6**). [48,49] The dORF encodes a form of PfEMP1 that exclusively binds to a placental-specific ligand, chondroitin sulfate A (CSA), meaning that the PfEMP1 protein encoded by this gene is only useful to the parasite in women who are infected for the first time while pregnant, as antibodies will be produced to it. [46,47] This may suggest that the conditions tested that induced its activation (**Figure 37 & 38**) might be mimicking those of a placenta, as VAR2CSA protein only has a selective advantage in the presence of a placenta. M2 macrophages that activate IDO have been found to be present largely at the maternal-fetal interface[118,103,104] These cells deplete AAs such as Arg and Trp, and produce large amounts of polyamines, like Spd – conditions that were tested in **Chapter 4**. Additionally, studies have found elevated levels of choline in the placenta[115,116], as was tested in **Chapter 2**. Following this model, immune recognition would not induce VAR2CSA translation, as *var2csa* only serves as the sink node. Conversely, the conditions in the placenta would induce VAR2CSA translation. Future work is needed to determine how this choice is made. The specific induction of VAR2CSA translation in culture is being investigated further by our lab.

As described in the previous section, two experiments with the PfSAMS OE line were able to observe switching as the model proposes in **Figure 40A**. These evidenced a capacity for parasites to switch *var* genes in response to genetically modified SAM levels. In further support of this hypothesis, however, one might ask how a full switch in *var* expression in response to immune recognition could be studied in culture. In the discussion section of **Chapter 4**, I described a set of experiments in relation to polyamine synthesis using DFMO, a compound that blocks ODC enzymatic activity, functionally shutting down the polyamine synthesis pathway **Figure 29**. I ran preliminary attempts at this experimental setup, testing 2mM DFMO with 2mM Put or Spd rescue following van Biljon et al. 2018, but the results on *var* expression were inconclusive. Rather intriguingly, however, I observed a considerable growth phenotype when attempting to scale the cultures up to a volume large enough for RNA extraction. Here, a large majority of parasites grown in the presence of DFMO with a

Spd rescue seemingly died off at the reinvasion step, never making it to early ring stage (revisit **Figures 1 & 2**). Following this immune recognition model, a majority of parasites would die in later growth stages as immune cells recognize PfEMP1 on the iRBC surface. Therefore, this phenotype was of interest to our lab in regards to possibly mimicking localized immune recognition conditions. By nature of the population being largely killed off, however, visualization of *var* expression after inducing this phenotype takes up to at least one-week growth, thus lacking the ability to visualize expression among the remaining population of parasites. To attempt to bypass this technical limitation, this phenotype, among others, is being explored further by our lab by using reporter lines for *var* expression.

A second possible, more direct experiment would be to co-culture parasites with monocytes and assay *var* expression in response to a localized immune response. Using an Antibody-Dependent Cell-Mediated Inhibition (ADCI) assay developed by Bouharoun-Tayoun and Druilhe, 2015, *P. falciparum* can be cultured in the presence of blood monocytes and immunoglobulins (IgG) [112, 113]. This assay is designed to mimic *in vivo* immune recognition; culturing iRBCs with antibodies for a selected variant of PfEMP1 will trigger activation of monocytes. If this proposed model is correct, this would induce *var* switching to variants other than *var2csa*.

To successfully battle any infectious disease, it is imperative to first understand the biology of the pathogen. Over half of the world's population is considered at risk for *Plasmodium* infection, and the world is not on track to reach targets in reducing malaria morbidity and mortality. [1,2,4] *P. falciparum* has a unique set of features that lend to its remarkable pathogenicity. In this thesis I presented results that shift the paradigm in our understanding of antigenic variation in this unique organism. As of the finishing of this document, the United States is emerging from the COVID-19 pandemic largely as a result of vaccines produced on the backbone of basic scientific research. It is my hope that, in however small a way, the research conducted here may aid in the development of better treatments for malaria disease, and, perhaps one day, lead to its eradication.

Chapter 6

Materials and Methods

Ethics statement

Human blood was used for the study and was purchased from the New York City Blood Center (NYBC) for use in parasite culture. Contact of blood donors was not attempted and was not necessary for the livelihood of the study. Informed consent was not required (other than NYBC in-house protocol). The identity of the donors cannot be readily attained by the researchers and was unknown throughout the study. The blood was used for research purposes only-solely for *in vitro* culture of *Plasmodium falciparum* and not for transfusion into humans or animals. NYBC policy states that only surplus blood will be made available for research purposes and thus this study did not compromise blood supplies. The blood purchased from NYBC was only used as a resource for propagation of malaria parasites and no data was collected with regard to the blood itself. Therefore the inclusion of women, minorities or children is not applicable. These conditions indicate (and the NIH has concluded) that the study does not qualify as human subjects research.

Generation of Malaria Incomplete Media (MIM) Powder

Individual media compounds were purchased as listed in **Table 3**. Compounds were measured following **Table 3** for 100L. For generation of AA MIM (**Chapter 4**) recipe was followed as shown but with only arginine and tryptophan left out of the base powder. Compounds were ground and mixed together using a 6lb. capacity ball mill and ½' stainless steel grinding media purchased from United Nuclear Scientific Supplies, LLC. To make 1L MIM, 10.18g of MIM powder was measured and made up in 1L ddH2O, with HEPES, hypoxanthine, sodium bicarbonate, gentamicin (0.1 mg/ml), and Albumax II added in following the measurements in **Table 3.**

Table 3. Components and recipe for Malaria Incomplete Media (MIM)

	Per 1L			Per 100L			Product Number
Inorganic	**mg/L**	**g/L**		**mg/100l**	**g/100L**		
Ca(NO3)2*4H2O	100	0.1		10000	10		C2786 SIGMA
KCl	400	0.4		40000	40		P5405 SIGMA
MgSO4	58.8	0.0588		5880	5.88		M2643 SIGMA
NaCl	6000	6		600000	600		S3014 SIGMA
Na2HPO4	800	0.8		80000	80		S5136 SIGMA
NaH2CO3	2000	2		200000	200		S5761 SIGMA
Amino Acids							
L-Arginine	200	0.2		20000	20		A8094 SIGMA
L-Asparagine*H2O	56	0.056		5600	5.6		A4284 SIGMA
L-Aspartic Acid	20	0.02		2000	2		A7219 SIGMA
L-Cystine*2HCl	65.2	0.0652		6520	6.52		C6727 SIGMA
L-Glutamic Acid	20	0.02		2000	2		G8415 SIGMA
L-Glutamine	200	0.2		20000	20		G8540 SIGMA
Glycine	10	0.01		1000	1		VWRV0167 VWR
L-Histidine	15	0.015		1500	1.5		H6034 SIGMA
Hydroxy-L-proline	20	0.02		2000	2		H54409 SIGMA
L-Isoleucine	50	0.05		5000	5		I2752 SIGMA
L-Leucine	50	0.05		5000	5		L8912 SIGMA
L-Lysine* HCl	40	0.04		4000	4		L8662 SIGMA
L-Methionine	15	0.015		1500	1.5		M5308 SIGMA
L-Phenylalanine	15	0.015		1500	1.5		P5482 SIGMA
L-Proline	20	0.02		2000	2		P5607 SIGMA
L-Serine	30	0.03		3000	3		S4500 SIGMA
L-Threonine	20	0.02		2000	2		T8441 SIGMA
L-Tryptophan	5	0.005		500	0.5		T8941 SIGMA
L-Tyrosine*2Na*2H2O	28.83	0.02883		2883	2.883		T1145 SIGMA
L-Valine	20	0.02		2000	2		V0513 SIGMA
Vitamins							
Biotin	0.2	0.0002		20	0.02		B4639 SIGMA
D-Calcium pantothenate	0.25	0.00025		25	0.025		2194721 MP Biomedicals
Choline Chloride	3	0.003		300	0.3		C7017 SIGMA
Folic Acid	1	0.001		100	0.1		F8758 SIGMA
i-Inositol	35	0.035		3500	3.5		I5125 SIGMA
Nicotinamide	1	0.001		100	0.1		N0636 SIGMA
Para-Aminobenzoic Acid	1	0.001		100	0.1		A9878 SIGMA
Pyridoxine	1	0.001		100	0.1		P6280 SIGMA
Riboflavin	0.2	0.0002		20	0.02		R9504 SIGMA
Thiamine	1	0.001		100	0.1		T4625 SIGMA
Vitamin B12	0.005	5E-06		0.5	0.0005		V2876 SIGMA
Other							
D-Glucose	2000	2		200000	200		G7021 SIGMA
Glutathione	1	0.001		100	0.1		G6013 SIGMA
Hypoxanthine	50	0.05		5000	5		H9377 SIGMA
HEPES	5958	5.958		595800	595.8		391338 EMD Millipore
Phenol Red*Na	5	0.005		500	0.5		P5530 SIGMA
Gentamycin Antibiotic	1mL/1L						
Albumax II	5g/1L						
Minimal RPMI 10.18g/L							
Left out of base MIM powder, compounds not desired to be tested were added in when making up 1L media							
Not included in base MIM powder, added when making 1L aqueous media							

Parasite culture and transfection

Both 3D7 and NF54 parasite lines were cultivated following standard procedures at 5% hematocrit in RPMI 1640 medium or MIM (see above), 0.5% Albumax II (Invitrogen), 0.25% sodium bicarbonate and 0.1 mg/ml gentamicin. All parasites were incubated in an atmosphere containing 5% oxygen, 5% carbon dioxide, and 90% nitrogen at 37°C. Leukocytes were removed from volunteer whole blood (New York Blood Center) by leukocyte-reduction filters (Haemonetics) and by washing with MCM RPMI-1640. When testing the nutrient conditions listed in **Tables 1 or 2**, stocks were made up in either ddH2O or growth medium to be added exogenously to cultures at the final concentration listed for each condition tested as listed in **Tables 1 and 2**.

Subclones of 3D7 or NF54 were transfected using DNA-loaded RBCs as described previously [114]. Briefly, parasites were loaded with 200 µg of plasmid DNA in incomplete cytomix and pulsed with a gene pulser (bio-rad). PfSAMS and PfSAHH aptamer OE constructs were induced following protocols outlined in Ganesan et al. 2016[87]: Ring-stage parasites were tightly synchronized using 5% sorbitol and were induced using 0.5 mM ATc (Cayman Chemical product #:10009542) at ring-stage. PfSAMS GlmS constructs were grown in the presence of 2.5mM GlcN for PfSAMS knockdown. OE lines were selected on 1mg/ml Blasticidin (Blasticidin S HCL from Invitrogen, catalog # R210-01) after transfection and brought up to 2mg/ml for experimentation. KD lines were selected on 100mg/mL Neomycin (G418 Disulfate Salt, Sigma, catalog # A1720).

RNA extraction and cDNA synthesis

RNA from all parasite lines was extracted from synchronized late ring stage parasites as described previously [22]. Briefly, RNA was extracted with TRiZol (Invitrogen) and purified on PureLink (Invitrogen) columns following manufacturer's protocol. Isolated RNA was treated with Deoxyribonuclease I (DNase I) (Invitrogen) to degrade contaminating genomic DNA. cDNA was synthesized from approximately 800 ng of RNA in a total reaction volume that consisted of Superscript II RNase H reverse transcriptase (Invitrogen) as described by the manufacturer.

Quantitative RT-PCR

We employed the Q-RT-PCR *var* primer set of Salanti et al to detect transcription from all *var* genes [46]. Sequences of qPCR primers for PfSAMS and PfSAHH expression are shown in **Table 4**.

All reactions were performed in 10µl volumes using ITAQ SYBR supermix (Bio-Rad) in a 7900HT RT-PCR System (ABI) or QuantStudio 6 Flex (ThermoFisher). ΔCT for each individual primer pair was determined by subtracting the individual CT value from the CT value of the control seryl-tRNA synthetase (primer pair 60 from [46])(User bulletin 2, Applied Biosystems, http://www. appliedbiosystems.com). ΔCTs were further converted to relative copy numbers with the formula 2ΔCT. All Q-RT-PCR assays were run in single or duplicate wells with no apparent differences between runs. Relative copy numbers for each *var* gene were determined in Microsoft Excel and transcriptional profiles of individual genes are presented as pie graphs or as bar graphs. The *var* gene with the highest copy number was designated as the dominant gene.

Plasmid Construction

PfSAMS and PfSAHH OEs were constructed by inserting each gene's coding region into the pMG68 plasmid backbone supplied by Dr Jacquin Niles. Insertion was accomplished by In-Fusion using the In-Fusion® HD Cloning Kit from Takara Bio USA, Inc. Primers used for assembly are listed in **Table 4**. Constructs were transformed into Steller competent *E. coli* cells from Takara.

Table 4. Primers Used

For In-Fusion Assembly in PMG68		
VS82	SAMS Fwd	AGAAATATATCTTAAGCAAAAGATTTTTAGCATACCGTGA
VS83	SAMS Rev	ACGTCATAAGGATAGACGTCATTTTTTAATGCATTTTTTTCGTGAGA
VS84	SAHH Fwd	AGAAATATATCTTAAGATGGTTGAAAATAAGAGTAAGGTCAAAG
VS85	SAHH Rev	ACGTCATAAGGATAGACGTCATATCTGTATTCGTTACTCTTAAAGGGAC
PfSAMS and PfSAHH QPCR		
VS36	SAMS Fwd	ATGGTACCGTAAGCACAGGA
VS37	SAMS Rev	CACGTGAACGTATCTCCTTCTC
VS40	SAHH Fwd	TCTAGGATGTGCAACAGGACA
VS41	SAHH Rev	TTCTGTCAAGGAAGCGTTCA

Immunoblotting

For SDS-PAGE, total protein lysates were prepared using saponin-lysed parasites resuspended with 1x Laemmli loading buffer diluted in 1x PBS supplemented with 1x Roche Complete protease inhibitors cocktail. Protein samples were separated in 12% polyacrylamide gels and transferred to 0.2 µm Immobilion-PSQ transfer membrane (Millipore, Cat. No ISEQ00010) using a Bio-Rad transfer system. Membranes were blocked in 5% skim milk/1x TBS-Tween20 for 1 hour at RT. Primary and secondary antibodies were prepared in 3% skim milk/1x TBS-Tween20 and
389 incubated for 1 hour at room temperature. Membranes were washed three times with 1x TBS-Tween20 for 10min, after primary and secondary antibody incubations. For the PfSAMS OE, parasites grown for two cycles and protein extracted at trophozoite stage of cycle 2 (0.5uM ATc). Blot was 1st probed using anti HArat(1:5000) (2nd* anti rat-HRP 1:5000) and anti H3-Rab (1:3000**)** (2nd* and anti Rb-HRP 1:2500). Then probed with anti-flag Rb(1:2500) and anti Rb (1:2500). Immunoblots were incubated with the chemiluminescent substrate SuperSignal West Pico PLUS (ThermoFisher, Cat. No 34578) following manufacturer directions. Images were obtained using Azure c300 digital imaging system (Azure Biosystems).

Growth Assays

The starting culture was adjusted to 0.05% or 0.5% (for the PfSAMS and PfSAHH OEs only) parasitemia, 5% hematocrit, and at 5ml total volume. The parasites were diluted to 1/10th their volume (500µl) with blood and media upon reaching greater than or equal to 0.5% parasitemia. For the PfSAMS and PfSAHH OEs: parasites were diluted to 1/10th their volume (500µl) with blood and media upon reaching greater than or equal to 1% parasitemia. Parasitemia was obtained daily by flow cytometry and verified by thin smear stained with Giemsa (Sigma). The relative parasitemia was calculated by multiplying the daily parasitemia by the exponential dilution factor each day they were diluted and the data was graphed on a log scale over time.

Cloning of Parasites

Parasitemia of cultures to be cloned were obtained by flow cytometry. Hematocrit was obtained either by hemocytometer or centrifugation. Starting culture parasitemia and hematocrit were diluted into a 96-well plate such that each well was calculated to contain 0.5 parasites/well at a final well volume was 0.2mL with 3% hematocrit. 96-well plates were incubated in an airtight chamber at an atmosphere containing 5% oxygen, 5% carbon dioxide, and 90% nitrogen at 37°C until growth was observed either by thin blood smear or flow cytometry (typically ~2-3 weeks). Media in wells was changed once or twice per week. Once wells were positive for parasites, they were picked at random and grown up in culture flasks.

Drug Assay

Parasite growth inhibition assays were performed on parasites cultured in sterile 96-well plates at a total 200µL volume per well and a 0.5% initial parasitemia and 2% hematocrit. Plates were incubated in an airtight chamber at an atmosphere containing 5% oxygen, 5% carbon dioxide, and 90% nitrogen, 37°C for 72 hours. Plates were then placed in the –80°C freezer for lysis upon thawing. SYBR Green solution (100µL of 0.2µL SYBR Green per mL lysis buffer) was then added to each well and the plates were shaken in the dark at room temperature for 1 hour. Fluorescence was then

recorded in a SpectraMax Gemini plate reader with excitation 490 nm and emission 530 nm. Fluorescent counts were normalized and plotted by non-linear least square regression to yield EC_{50} values (PRISM). This was done two times independently and run in triplicate each time.

Appendix

Appendix A

Full *var* panels and replicates

Panels are organized by corresponding chapter and section. Culture and/or condition tested are listed at the top of each plot. Timepoint is listed either for each plot or next to corresponding chapter and section. Each plot has five to six columns: Primer pair number from Salanti, A. *et al.* 2003, CT value (one or two replicate wells, Average CT (if two replicate wells), Average CT value subtracted from p60 expression (relative to p60), and then relative copy number (2^relative copy number). One to two separate panels are shown per page. Each panel represents bulk cDNA from one culture.

Chapter 2.2 *var* panels

MCM Day 0 (Rep 1)						20xCho, Dep Ser 2Wks (Rep 1)					
Primer pair	CT		CT Average	relative to p60	realtive copy number	Primer pair	CT		CT Average	relative to p60	realtive copy number
1	24.878239	25.376583	25.127411	-5.707411	0.01913807	1	25.72532	25.085554	25.405437	-5.205437	0.02710237
3	27.238873	27.950066	27.5944695	-8.1744695	0.0034613	3	27.8309	28.003649	27.9172745	-7.7172745	0.00475192
4	27.004942	28.077688	27.541315	-8.121315	0.00359121	4	24.734316	24.617952	24.676134	-4.476134	0.04493134
5	26.781912	29.16699	27.974451	-8.554451	0.00265983	5	27.218584	27.480974	27.349779	-7.149779	0.0070421
6	26.41398	31.114567	28.7642735	-9.3442735	0.00153849	6	25.982023	25.063086	25.5225545	-5.3225545	0.02498915
7	26.895315	28.196453	27.545884	-8.125884	0.00357985	7	27.760513	27.910275	27.835394	-7.635394	0.00502941
8	27.230766	28.36899	27.799878	-8.379878	0.00300196	8	27.60465	27.462317	27.5334835	-7.3334835	0.00620014
9	25.953802	28.552435	27.2531185	-7.8331185	0.00438527	9	25.994858	26.271141	26.1329995	-5.9329995	0.01636776
11	27.026237	28.9049	27.9655685	-8.5455685	0.00267626	11	26.390902	26.42438	26.407641	-6.207641	0.0135305
12	25.781681	27.296232	26.5389565	-7.1189565	0.00719417	12	25.727282	25.83992	25.783601	-5.583601	0.020853
13	23.486622	24.3648	23.925711	-4.505711	0.04401957	13	24.854343	24.967484	24.9109135	-4.7109135	0.03818332
15	29.110735	33.215233	31.162984	-11.742984	0.00029175	15	27.467522	27.456656	27.462089	-7.262089	0.00651468
17	24.250158	24.790396	24.520277	-5.100277	0.02915168	17	26.402481	26.607868	26.5051745	-6.3051745	0.012646
18	24.595482	26.472828	25.534155	-6.114155	0.0144363	18	24.98492	24.996687	24.9908035	-4.7908035	0.03612638
19	25.867865	26.87216	26.3700125	-6.9500125	0.00808794	19	25.18165	25.433752	25.307701	-5.107701	0.02900206
20	31.769949	Undetermined	31.769949	-12.349949	0.00019156	20	29.123713	28.958452	29.0410825	-8.8410825	0.00218056
21	25.414068	27.42215	26.418109	-6.998109	0.00782275	21	26.287428	26.483187	26.3853075	-6.1853075	0.01374159
22	26.175093	27.797037	26.986065	-7.566065	0.005277	22	26.207611	25.89986	26.0537355	-5.8537355	0.01729219
23	27.355692	28.682688	28.01919	-8.59919	0.00257861	23	27.793385	27.851786	27.8225855	-7.6225855	0.00507426
25	32.45085	33.629524	33.040187	-13.620187	7.9417E-05	25	29.290316	29.141176	29.215746	-9.015746	0.00193192
26	24.385386	26.7663	25.575843	-6.155843	0.01402512	26	27.19843	26.879839	27.0391345	-6.8391345	0.00873404
27	26.253544	27.812426	27.032985	-7.612985	0.00510814	27	25.457434	25.21646	25.336947	-5.136947	0.02842005
28	25.802982	29.834446	27.818714	-8.398714	0.00296302	28	25.858355	25.862373	25.860364	-5.660364	0.01977246
30	27.166964	26.697134	26.932049	-7.512049	0.00547833	30	28.204832	27.91604	28.060436	-7.860436	0.00430302
31	22.934992	25.48601	24.210501	-4.790501	0.03613396	31	23.213945	23.450924	23.3324345	-3.1324345	0.11403634
34	29.539488	28.99086	29.265174	-9.845174	0.00108719	34	26.42725	27.135805	26.7815275	-6.5815275	0.0104415
35	28.757473	30.556005	29.656739	-10.236739	0.00082877	35	28.121042	28.26879	28.194916	-7.994916	0.00392004
36	26.473577	26.223442	26.3485095	-6.9285095	0.00820939	36	27.39512	26.468355	26.9317375	-6.7317375	0.00940904
37	26.913721	29.22072	28.0672205	-8.6472205	0.00249418	37	25.041103	24.991592	25.0163475	-4.8163475	0.03549236
38	27.837763	26.76473	27.3012465	-7.8812465	0.00424139	38	26.15459	25.982018	26.068304	-5.868304	0.01711845
39	26.217207	28.760664	27.4889355	-8.0689355	0.00372399	39	26.889952	27.00065	26.945301	-6.745301	0.00932099
40	29.642115	28.580927	29.111521	-9.691521	0.00120938	40	27.963566	27.947731	27.9556485	-7.7556485	0.00462719
41	31.612438	Undetermined	31.612438	-12.192438	0.00021365	41	28.122696	28.12324	28.122968	-7.922968	0.00412049
43	29.077538	27.857454	28.467496	-9.047496	0.00188987	43	26.943682	26.843706	26.893694	-6.693694	0.00966045
44	30.282833	Undetermined	30.282833	-10.862833	0.00053698	44	28.449142	28.560465	28.5048035	-8.3048035	0.00316231
45	31.45227	30.89152	31.171895	-11.751895	0.00028995	45	25.95497	25.92388	25.939425	-5.739425	0.01871806
46	25.891478	30.00578	27.948629	-8.528629	0.00270786	46	26.709501	26.799788	26.7546445	-6.5546445	0.01063789
47	27.175127	26.712933	26.94403	-7.52403	0.00543302	47	27.226854	27.287214	27.257034	-7.057034	0.00750967
49	25.605413	28.017921	26.811667	-7.391667	0.00595507	49	25.555399	25.459806	25.5076025	-5.3076025	0.02524948
50	26.142452	26.12042	26.131436	-6.711436	0.00954237	50	27.108051	26.824495	26.966273	-6.766273	0.00918647
51	25.793669	27.47985	26.6367595	-7.2167595	0.00672263	51	26.618551	26.786156	26.7023535	-6.5023535	0.01103053
52	26.490784	26.29952	26.395152	-6.975152	0.00794822	52	27.418486	27.514212	27.466349	-7.266349	0.00649548
53	25.729969	33.443104	29.5865365	-10.166537	0.0008701	53	25.813345	25.72136	25.7673525	-5.5673525	0.02108919
54	18.145279	18.475	18.3101395	1.1098605	2.15824777	54	21.763275	21.678547	21.720911	-1.520911	0.34846581
55	Undetermine	33.515247	33.515247	-14.095247	5.7136E-05	55	27.8711	27.448277	27.6596885	-7.4596885	0.00568081
56	29.739899	Undetermined	29.739899	-10.319899	0.00078235	56	28.241528	28.494467	28.3679975	-8.1679975	0.00347686
57	28.469976	27.529186	27.999581	-8.579581	0.0026139	57	26.106752	26.196003	26.1513775	-5.9513775	0.01616058
58	30.76554	Undetermined	30.76554	-11.34554	0.00038428	58	25.140232	25.465303	25.3027675	-5.1027675	0.0291014
var2csa	23.78223	25.675133	24.7286815	-5.3086815	0.0252306	*var2csa*	20.49234	20.725925	20.6091325	-0.4091325	0.75307607
91	25.061666	28.665134	26.8634	-7.4434	0.00574531	91	24.962519	24.927156	24.9448375	-4.7448375	0.03729594
92	26.04976	Undetermined	26.04976	-6.62976	0.01009819	92	24.197638	24.220804	24.209221	-4.009221	0.0621018
93	33.06892	Undetermined	33.06892	-13.64892	7.7851E-05	93	27.323349	27.859404	27.5913765	-7.3913765	0.00595626
94	30.260933	Undetermined	30.260933	-10.840933	0.0005452	94	28.364779	28.541199	28.452989	-8.252989	0.00327795
95	29.285955	34.13851	31.7122325	-12.292233	0.00019937	95	26.848314	26.887724	26.868019	-6.668019	0.00983391
96	32.47438	Undetermined	32.47438	-13.05438	0.00011755	96	29.156122	29.880383	29.5182525	-9.3182525	0.00156649
97	Undetermine	Undetermined			0	97	29.478811	29.8408	29.6598055	-9.4598055	0.00142009
60	26.189377	26.597664	26.3935205			60	20.203348	20.198565	20.2009565		

20xCho, Dep Ser 4Wks (Rep 1)							1xCho, Dep Ser 2Wks (Rep 1)						
Primer pair	CT			CT Average	relative to p60	realtive copy number	Primer pair	CT			CT Average	relative to p60	realtive copy number
1	25.016582	25.441942		25.229262	-3.729262	0.07540155	1	25.47907	25.302551		25.3908105	-3.3908105	0.09533763
3	27.275087	27.320904		27.2979955	-5.7979955	0.01797337	3	Undetermine	Undetermined		#DIV/0!	#DIV/0!	0
4	25.54619	25.645891		25.5960405	-4.0960405	0.05847483	4	26.367182	26.199772		26.283477	-4.283477	0.05135053
5	26.750832	27.317078		27.033955	-5.533955	0.02158309	5	32.22665	33.199306		32.712978	-10.712978	0.00059576
6	25.473473	25.5645		25.5189865	-4.0189865	0.06168286	6	26.74118	26.832287		26.7867335	-4.7867335	0.03622844
7	25.62154	25.929125		25.7753325	-4.2753325	0.05164124	7	28.220772	28.289938		28.255355	-6.255355	0.01309033
8	26.47259	27.114336		26.793463	-5.293463	0.02549816	8	29.154396	29.402962		29.278679	-7.278679	0.0064402
9	26.035095	26.46731		26.2512025	-4.7512025	0.03713176	9	27.351458	27.223356		27.287407	-5.287407	0.02560542
11	26.456326	26.79207		26.624198	-5.124198	0.02867231	11	28.023384	28.129602		28.076493	-6.076493	0.01481813
12	25.487656	25.709007		25.5983315	-4.0983315	0.05838204	12	27.501255	27.120932		27.3110935	-5.3110935	0.02518846
13	23.755463	23.872078		23.8137705	-2.3137705	0.20113409	13	26.023764	25.93416		25.978962	-3.978962	0.06341808
15	26.965635	27.045809		27.005722	-5.505722	0.02200962	15	28.640495	28.595776		28.6181355	-6.6181355	0.01017988
17	24.805197	24.89474		24.8499685	-3.3499685	0.09807515	17	26.05175	26.084297		26.0680235	-4.0680235	0.0596215
18	24.382652	24.369709		24.3761805	-2.8761805	0.13620197	18	25.752733	25.266457		25.509595	-3.509595	0.08780245
19	24.220121	24.504368		24.3622445	-2.8622445	0.13752402	19	26.803017	26.318354		26.5606855	-4.5606855	0.04237375
20	28.083511	28.479528		28.2815195	-6.7815195	0.0090899	20	30.032438	29.959986		29.996212	-7.996212	0.00391652
21	25.4326	25.965502		25.699051	-4.199051	0.05444521	21	27.967106	27.0002		27.483653	-5.483653	0.02234889
22	25.68435	25.822184		25.753267	-4.253267	0.05243715	22	26.926105	26.582952		26.7545285	-4.7545285	0.03704625
23	26.689474	26.929848		26.809661	-5.309661	0.02521348	23	28.46194	28.875477		28.6687085	-6.6687085	0.00982921
25	28.225908	28.638525		28.4322165	-6.9322165	0.00818832	25	30.27383	29.64618		29.960005	-7.960005	0.00401606
26	24.982182	25.298454		25.140318	-3.640318	0.08019644	26	26.534115	27.102783		26.818449	-4.818449	0.0354407
27	24.491621	25.407373		24.949497	-3.449497	0.09153727	27	26.821058	26.199009		26.5100335	-4.5100335	0.04388788
28	25.018225	25.582588		25.3004065	-3.8004065	0.07177342	28	26.292513	25.99509		26.1438015	-4.1438015	0.05657069
30	27.441526	27.556885		27.4992055	-5.9992055	0.01563361	30	27.903826	27.801298		27.852562	-5.852562	0.01730626
31	22.926912	23.166025		23.0464685	-1.5464685	0.34234705	31	Undetermine	25.08905		25.08905	-3.08905	0.1175177
34	26.058338	26.517452		26.287895	-4.787895	0.03619928	34	26.18491	26.265642		26.225276	-4.225276	0.05346446
35	28.509216	28.867254		28.688235	-7.188235	0.00685687	35	28.018555	28.324791		28.171673	-6.171673	0.01387207
36	27.192213	28.032297		27.612255	-6.112255	0.01445533	36	28.405432	27.914768		28.1601	-6.1601	0.0139838
37	28.790014	29.00982		28.899917	-7.399917	0.00592111	37	24.793566	24.792883		24.7932245	-2.7932245	0.14426323
38	25.808157	26.34982		26.0789885	-4.5789885	0.04183956	38	26.925686	27.057232		26.991459	-4.991459	0.03143555
39	27.482264	27.76679		27.624527	-6.124527	0.01433289	39	27.275364	26.980883		27.1281235	-5.1281235	0.0285944
40	28.186989	28.697662		28.4423255	-6.9423255	0.00813115	40	29.455505	28.89896		29.1772325	-7.1772325	0.00690936
41	30.757038	30.776964		30.767001	-9.267001	0.00162314	41	28.967815	28.461731		28.714773	-6.714773	0.00952033
43	26.54794	27.46122		27.00458	-5.50458	0.02202705	43	28.032095	27.6769		27.8544975	-5.8544975	0.01728306
44	28.404633	28.564466		28.4845495	-6.9845495	0.00789662	44	29.449575	29.686548		29.5680615	-7.5680615	0.00526971
45	25.614895	26.632305		26.1236	-4.6236	0.04056558	45	27.559519	27.668362		27.6139405	-5.6139405	0.02041905
46	26.969173	27.82558		27.3973765	-5.8973765	0.01677694	46	28.287767	28.300789		28.294278	-6.294278	0.01274188
47	27.548588	28.386711		27.9676495	-6.4676495	0.01129909	47	28.289152	28.828587		28.5588695	-6.5588695	0.01060678
49	24.23271	24.314354		24.273532	-2.773532	0.14624589	49	27.033602	27.189615		27.1116085	-5.1116085	0.02892361
50	28.265099	28.735233		28.500166	-7.000166	0.0078116	50	28.164541	28.4957		28.3301205	-6.3301205	0.01242922
51	27.65886	27.964388		27.811624	-6.311624	0.0125896	51	27.533072	27.257305		27.3951885	-5.3951885	0.02376219
52	27.361628	28.134102		27.747865	-6.247865	0.01315846	52	28.481985	28.436508		28.4592465	-6.4592465	0.01136509
53	25.01672	25.138193		25.0774565	-3.5774565	0.08376803	53	26.245714	26.176485		26.2110995	-4.2110995	0.05399241
54	20.85748	21.283817		21.0706485	0.4293515	1.34662812	54	22.352505	22.422602		22.3875535	-0.3875535	0.76442481
55	28.697897	31.302204		30.0000505	-8.5000505	0.00276204	55	29.078678	28.941216		29.009947	-7.009947	0.00775882
56	27.364193	27.477657		27.420925	-5.920925	0.01650532	56	27.157133	28.194153		27.675643	-5.675643	0.01956416
57	26.144053	26.421188		26.2826205	-4.7826205	0.03633187	57	27.513794	27.761366		27.63758	-5.63758	0.0200872
58	29.687468	30.985579		30.3365235	-8.8365235	0.00218747	58	25.386196	25.447113		25.4166545	-3.4166545	0.09364498
var2csa	20.89076	21.111034		21.000897	0.499103	1.41333454	*var2csa*	22.830738	23.134779		22.9827585	-0.9827585	0.5060113
91	24.198278	24.800714		24.499496	-2.999496	0.12504368	91	26.16167	25.987904		26.074787	-4.074787	0.05934264
92	23.980652	24.092896		24.036774	-2.536774	0.17232764	92	25.246683	25.073912		25.1602975	-3.1602975	0.11185507
93	27.057728	27.641018		27.349373	-5.849373	0.01734456	93	28.878553	29.150484		29.0145185	-7.0145185	0.00773427
94	28.543747	28.579487		28.561617	-7.061617	0.00748586	94	29.967037	29.814938		29.8909875	-7.8909875	0.00421285
95	27.048021	27.422953		27.235487	-5.735487	0.01876923	95	28.165049	28.05429		28.1096695	-6.1096695	0.01448126
96	29.181314	29.460016		29.320665	-7.820665	0.00442329	96	30.384796	30.20834		30.296568	-8.296568	0.00318042
97	30.444317	32.028114		31.2362155	-9.7362155	0.00117248	97	32.047447	31.937742		31.9925945	-9.9925945	0.00098159
60	21.462776	21.558178		21.510477			60	22.055937	21.796684		21.9263105		

1xCho, Dep Ser 4Wks (Rep 1)							MCM 4 WK (Rep 1)						
Primer pair	CT			CT Average	relative to p60	realtive copy number	Primer pair	CT			CT Average	relative to p60	realtive copy number
1	25.481283	25.555683		25.518483	-4.028483	0.06127817	1	25.386269	25.171618		25.2789435	-4.2789435	0.05151215
3	28.280975	28.317415		28.299195	-6.809195	0.00891719	3	27.366545	27.182081		27.274313	-6.274313	0.01291944
4	25.99464	26.11036		26.0525	-4.5625	0.04232049	4	25.707214	26.459414		26.083314	-5.083314	0.02949647
5	28.025751	27.999273		28.012512	-6.522512	0.01087748	5	27.703506	27.949623		27.8265645	-6.8265645	0.00881047
6	26.722221	26.638336		26.6802785	-5.1902785	0.02738864	6	25.03312	24.737556		24.885338	-3.885338	0.06767008
7	28.053484	27.793446		27.923465	-6.433465	0.01157002	7	26.88515	27.814829		27.3499895	-6.3499895	0.01225922
8	27.795202	28.042223		27.9187125	-6.4287125	0.0116082	8	26.605135	26.50562		26.5553775	-5.5553775	0.02126497
9	26.397034	26.463673		26.4303535	-4.9403535	0.03256907	9	26.54068	27.196669		26.8686745	-5.8686745	0.01711406
11	28.063879	28.01194		28.0379095	-6.5479095	0.01068766	11	26.849771	26.631073		26.740422	-5.740422	0.01870513
12	26.767027	26.843803		26.805415	-5.315415	0.02511312	12	25.461561	25.706476		25.5840185	-4.5840185	0.04169394
13	25.030382	25.228168		25.129275	-3.639275	0.08025444	13	24.974714	25.850536		25.412625	-4.412625	0.04695345
15	27.383266	27.415184		27.399225	-5.909225	0.01663972	15	26.780602	26.694397		26.7374995	-5.7374995	0.01874306
17	27.097263	27.2145		27.1558815	-5.6658815	0.01969698	17	25.704113	26.014807		25.85946	-4.85946	0.03444743
18	25.393616	25.399748		25.396682	-3.906682	0.06667631	18	24.316717	23.71957		24.0181435	-3.0181435	0.12343783
19	27.320333	27.27141		27.2958715	-5.8058715	0.01787551	19	24.412592	24.475273		24.4439325	-3.4439325	0.09189101
20	29.510166	29.14595		29.328058	-7.838058	0.00437028	20	28.26944	28.60987		28.439655	-7.439655	0.00576024
21	26.985088	26.98688		26.985984	-5.495984	0.02215868	21	25.412653	26.454353		25.933503	-4.933503	0.03272409
22	25.747095	26.057728		25.9024115	-4.4124115	0.0469604	22	26.008095	25.996372		26.0022335	-5.0022335	0.03120166
23	28.159756	28.202187		28.1809715	-6.6909715	0.0096787	23	27.14094	27.523815		27.3323775	-6.3323775	0.01240979
25	29.41359	28.932568		29.173079	-7.683079	0.0048659	25	28.554	28.953222		28.753611	-7.753611	0.00463373
26	26.248213	26.4285		26.3383565	-4.8483565	0.03471357	26	27.920502	30.619402		29.269952	-8.269952	0.00323964
27	25.68179	26.141699		25.9117445	-4.4217445	0.04665759	27	24.060137	25.1712		24.6156685	-3.6156685	0.08157843
28	26.583643	28.003517		27.29358	-5.80358	0.01790393	28	25.103893	25.12175		25.1128215	-4.1128215	0.05779861
30	28.155474	28.248367		28.2019205	-6.7119205	0.00953917	30	26.612951	27.00361		26.8082805	-5.8082805	0.01784569
31	23.60832	23.580208		23.594264	-2.104264	0.23256985	31	22.309465	22.12364		22.2165525	-1.2165525	0.43030977
34	27.240486	28.459867		27.8501765	-6.3601765	0.01217296	34	25.84135	25.790762		25.816056	-4.816056	0.03549954
35	28.991583	29.024153		29.007868	-7.517868	0.00545627	35	27.476303	27.705246		27.5907745	-6.5907745	0.01037479
36	28.28952	28.661581		28.4755505	-6.9855505	0.00789114	36	27.058165	27.551277		27.304721	-6.304721	0.01264998
37	25.543901	25.406218		25.4750595	-3.9850595	0.06315061	37	28.832201	28.650074		28.7411375	-7.7411375	0.00467396
38	27.370403	28.261757		27.81608	-6.32608	0.01246408	38	26.161081	26.288935		26.225008	-5.225008	0.0267372
39	26.732607	26.69895		26.7157785	-5.2257785	0.02672292	39	27.086683	27.766346		27.4265145	-6.4265145	0.01162589
40	28.648262	28.921375		28.7848185	-7.2948185	0.00636855	40	28.102621	29.022758		28.5626895	-7.5626895	0.00528937
41	29.677824	29.54275		29.610287	-8.120287	0.00359377	41	29.503128	30.40165		29.952389	-8.952389	0.00201866
43	27.219782	27.406145		27.3129635	-5.8229635	0.01766499	43	25.941608	25.816067		25.8788375	-4.8788375	0.03398784
44	30.94446	Undetermined		30.94446	-9.45446	0.00142536	44	28.58795	28.868608		28.728279	-7.728279	0.00471581
45	27.129221	27.996355		27.562788	-6.072788	0.01485623	45	24.291391	24.719393		24.505392	-3.505392	0.08805862
46	27.054619	27.215565		27.135092	-5.645092	0.01998288	46	25.568256	26.018225		25.7932405	-4.7932405	0.03606541
47	27.799545	28.182074		27.9908095	-6.5008095	0.01104235	47	26.584906	26.038773		26.3118395	-5.3118395	0.02517543
49	26.835514	26.763779		26.7996465	-5.3096465	0.02521373	49	25.773092	25.782633		25.7778625	-4.7778625	0.03645189
50	27.630697	28.058096		27.8443965	-6.3543965	0.01222182	50	26.098553	26.502102		26.3003275	-5.3003275	0.02537713
51	27.35464	27.651329		27.5029845	-6.0129845	0.015485	51	27.079556	27.00796		27.043758	-6.043758	0.0151582
52	27.534676	27.215334		27.375005	-5.885005	0.01692143	52	26.018215	25.653086		25.8356505	-4.8356505	0.03502065
53	26.848343	28.002409		27.425376	-5.935376	0.01634082	53	24.32811	24.402172		24.365141	-3.365141	0.09704912
54	21.19713	21.536612		21.366871	0.123129	1.08909439	54	19.346067	20.057299		19.701683	1.298317	2.45941808
55	28.516039	28.689241		28.60264	-7.11264	0.00722573	55	27.27007	27.203102		27.236586	-6.236586	0.01326174
56	29.177746	30.328913		29.7533295	-8.2633295	0.00325454	56	26.938433	26.636427		26.78743	-5.78743	0.01810548
57	26.164433	26.11272		26.1385765	-4.6485765	0.03986934	57	26.388535	24.928875		25.658705	-4.658705	0.03959041
58	28.202335	28.18786		28.1950975	-6.7050975	0.00958439	58	26.624517	27.096375		26.860446	-5.860446	0.01721195
var2csa	22.003029	21.861376		21.9322025	-0.4422025	0.73601012	*var2csa*	23.374258	23.217001		23.2956295	-2.2956295	0.20367919
91	26.293718	26.51118		26.402449	-4.912449	0.03320515	92	23.464869	24.025053		23.744961	-2.744961	0.149171
92	26.11787	27.483822		26.800846	-5.310846	0.02519278	93	29.850037	30.929281		30.389659	-9.389659	0.00149084
93	30.845753	34.34782		32.5967865	-11.106787	0.00045344	94	28.975016	30.219816		29.597416	-8.597416	0.00258178
94	29.645178	30.461071		30.0531245	-8.5631245	0.00264389	95	27.933357	29.21901		28.5761835	-7.5761835	0.00524012
95	27.772577	29.112675		28.442626	-6.952626	0.0080733	96	29.398481	29.094496		29.2464885	-8.2464885	0.00329276
96	30.414204	Undetermined		30.414204	-8.924204	0.00205848	97	28.442526	26.819632		27.631079	-6.631079	0.01008896
97	31.237988	32.763123		32.0005555	-10.510556	0.0006855							
60	21.599905	21.390093		21.494999			60	20.967497	21.147501		21.057499		

No Cho, Dep Ser 2Wks (Rep 1)						
Primer pair	CT			CT Average	relative to p60	realtive copy number
1	25.316017	25.872723		25.59437	-3.44437	0.09186315
3	28.913912	29.829071		29.3714915	-7.2214915	0.00670061
4	26.352144	28.089666		27.220905	-5.070905	0.02975127
5	26.804478	28.14784		27.476159	-5.326159	0.02492679
6	25.393381	25.5333		25.4633405	-3.3133405	0.10059702
7	26.330439	27.924217		27.127328	-4.977328	0.03174497
8	27.261442	27.980938		27.62119	-5.47119	0.02254279
9	27.03753	27.827557		27.4325435	-5.2825435	0.02569188
11	26.931849	27.363306		27.1475775	-4.9975775	0.03130252
12	25.714876	25.862856		25.788866	-3.638866	0.08027719
13	23.74674	24.156092		23.951416	-1.801416	0.28689287
15	26.847305	27.291605		27.069455	-4.919455	0.03304429
17	25.270567	26.116604		25.6935855	-3.5435855	0.08575797
18	24.372187	24.32664		24.3494135	-2.1994135	0.21772614
19	24.19234	24.637718		24.415029	-2.265029	0.2080455
20	28.494488	28.92063		28.707559	-6.557559	0.01061642
21	26.099106	26.856829		26.4779675	-4.3279675	0.04979113
22	25.896505	25.89848		25.8974925	-3.7474925	0.07445474
23	27.280704	27.816616		27.54866	-5.39866	0.02370508
25	29.332039	30.79023		30.0611345	-7.9111345	0.00415443
26	25.073658	25.56491		25.319284	-3.169284	0.11116049
27	25.522682	32.51792		29.020301	-6.870301	0.00854739
28	24.572535	25.938877		25.255706	-3.105706	0.11616876
30	27.03205	27.212494		27.122272	-4.972272	0.03185642
31	22.629835	23.141336		22.8855855	-0.7355855	0.60057424
34	26.546175	29.085001		27.815588	-5.665588	0.01970099
35	28.472725	29.155184		28.8139545	-6.6639545	0.00986166
36	27.144457	28.71839		27.9314235	-5.7814235	0.01818101
37	29.549198	29.65743		29.603314	-7.453314	0.00570596
38	26.265787	26.73556		26.5006735	-4.3506735	0.04901362
39	26.536556	27.4504		26.993478	-4.843478	0.03483115
40	28.259474	29.44552		28.852497	-6.702497	0.00960168
41	30.790821	33.061275		31.926048	-9.776048	0.00114056
43	26.773981	28.64448		27.7092305	-5.5592305	0.02120825
44	28.58822	29.746408		29.167314	-7.017314	0.0077193
45	25.682308	27.906565		26.7944365	-4.6444365	0.03998391
46	27.09903	29.07588		28.087455	-5.937455	0.01631729
47	27.8227	31.056925		29.4398125	-7.2898125	0.00639069
49	23.734356	Undetermined		23.734356	-1.584356	0.33347349
50	27.622845	29.766333		28.694589	-6.544589	0.01071229
51	28.076431	32.966743		30.521587	-8.371587	0.00301926
52	26.927277	30.238684		28.5829805	-6.4329805	0.0115739
53	25.111862	26.00707		25.559466	-3.409466	0.09411275
54	20.314814	21.474743		20.8947785	1.2552215	2.38703794
55	30.177856	Undetermined		30.177856	-8.027856	0.00383155
56	27.12247	30.04867		28.58557	-6.43557	0.01155315
57	26.59697	26.987627		26.7922985	-4.6422985	0.04004321
58	30.54799	34.970196		32.759093	-10.609093	0.00064024
var2csa	27.171408	28.590828		27.881118	-5.731118	0.01882615
91	23.897573	25.048132		24.4728525	-2.3228525	0.19987189
92	32.344795	Undetermined		32.344795	-10.194795	0.00085322
93	24.115482	24.584417		24.3499495	-2.1999495	0.21764526
94	30.780722	34.171646		32.476184	-10.326184	0.00077895
95	24.903902	26.726337		25.8151195	-3.6651195	0.07882956
96	32.072964	Undetermined		32.072964	-9.922964	0.00103013
97	23.91428	25.348957		24.6316185	-2.4816185	0.17904343

No Cho, Dep Ser 4Wks (Rep 1)						
Primer pair	CT			CT Average	relative to p60	realtive copy number
1	24.810375	24.78563		24.7980025	-3.5680025	0.08431876
3	27.234951	27.739477		27.487214	-6.257214	0.01307347
4	25.88814	26.18156		26.03485	-4.80485	0.03577635
5	27.029955	27.326073		27.178014	-5.948014	0.0161983
6	25.035154	24.71208		24.873617	-3.643617	0.08001326
7	25.900011	25.847681		25.873846	-4.643846	0.04000028
8	27.485043	27.461142		27.4730925	-6.2430925	0.01320207
9	26.89106	26.943993		26.9175265	-5.6875265	0.01940367
11	27.144388	27.011608		27.077998	-5.847998	0.0173611
12	26.051884	25.708027		25.8799555	-4.6499555	0.03983125
13	23.824696	23.869762		23.847229	-2.617229	0.16298047
15	26.608294	26.777216		26.692755	-5.462755	0.02267498
17	24.428976	27.537062		25.983019	-4.753019	0.03708504
18	23.9385	24.140957		24.0397285	-2.8097285	0.1426223
19	24.900932	24.966228		24.93358	-3.70358	0.07675582
20	30.129833	29.972855		30.051344	-8.821344	0.0022106
21	25.502995	26.259298		25.8811465	-4.6511465	0.03979838
22	26.617714	26.796516		26.707115	-5.477115	0.0224504
23	27.729673	26.944405		27.337039	-6.107039	0.01450768
25	28.635542	29.438715		29.0371285	-7.8071285	0.00446499
26	24.684734	24.7657		24.725217	-3.495217	0.08868187
27	24.166483	24.530096		24.3482895	-3.1182895	0.11515991
28	23.396858	23.467785		23.4323215	-2.2023215	0.21728771
30	27.403294	28.11287		27.758082	-6.528082	0.01083556
31	22.678404	22.585632		22.632018	-1.402018	0.37839948
34	26.087856	26.117939		26.1028975	-4.8728975	0.03412807
35	28.942583	29.096087		29.019335	-7.789335	0.0045204
36	26.639858	26.54187		26.590864	-5.360864	0.02433432
37	30.163183	29.842564		30.0028735	-8.7728735	0.00228614
38	25.768139	25.861813		25.814976	-4.584976	0.04166628
39	27.416115	26.97435		27.1952325	-5.9652325	0.01600612
40	28.827135	30.750347		29.788741	-8.558741	0.00265193
41	30.400291	30.12408		30.2621855	-9.0321855	0.00191003
43	26.202969	26.512709		26.357839	-5.127839	0.02860004
44	29.210726	29.187689		29.1992075	-7.9692075	0.00399052
45	25.25943	25.09363		25.17653	-3.94653	0.06485987
46	26.708485	26.717113		26.712799	-5.482799	0.02236212
47	27.316706	27.278479		27.2975925	-6.0675925	0.01490983
49	24.515871	24.66225		24.5890605	-3.3590605	0.09745902
50	27.411898	27.74697		27.579434	-6.349434	0.01226394
51	26.435589	26.440617		26.438103	-5.208103	0.02705234
52	26.997509	27.07108		27.0342945	-5.8042945	0.01789506
53	25.39643	25.696112		25.546271	-4.316271	0.05019644
54	20.636534	20.750362		20.693448	0.536552	1.45050171
55	27.465364	27.571411		27.5183875	-6.2883875	0.01279401
56	27.22603	27.479635		27.3528325	-6.1228325	0.01434973
57	27.380823	27.298908		27.3398655	-6.1098655	0.01447929
58	28.90616	29.38618		29.14617	-7.91617	0.00413995
var2csa	24.576357	24.920115		24.748236	-3.518236	0.08727813
91	24.484097	24.716969		24.600533	-3.370533	0.09668708
92	24.203386	24.579893		24.3916395	-3.1616395	0.11175107
93	26.141714	26.42456		26.283137	-5.053137	0.03011995
94	29.776188	29.989063		29.8826255	-8.6526255	0.00248485
95	27.120262	27.069836		27.095049	-5.865049	0.01715712
96	30.286701	31.263634		30.7751675	-9.5451675	0.0013385
97	30.971241	30.605265		30.788253	-9.558253	0.00132641
60	21.217068	21.243572		21.23032		

MCM 4 Wks (Rep 2)							No Cho, Dep Ser 4Wks (Rep 2)						
Primer pair	CT			CT Average	relative to p60	realtive copy number	Primer pair	CT			CT Average	relative to p60	realtive copy number
1	24.953753	24.716894		24.8353235	-3.7323235	0.07524171	1	26.270655	26.475346		26.3730005	-2.2800005	0.20589768
3	26.94368	26.44692		26.6953	-5.5923	0.02072764	3	29.121502	29.006134		29.063818	-4.970818	0.03188854
4	25.930773	26.165602		26.0481875	-4.9451875	0.03246013	4	26.959642	26.109709		26.5346755	-2.4416755	0.18406976
5	26.45846	26.247774		26.353117	-5.250117	0.02627588	5	29.255274	28.698536		28.976905	-4.883905	0.03386867
6	24.938889	25.208843		25.073866	-3.970866	0.06377496	6	26.885458	26.959858		26.922658	-2.829658	0.14066565
7	26.282143	26.20569		26.2439165	-5.1409165	0.02834196	7	27.935225	27.78025		27.8577375	-3.7647375	0.07357006
8	25.80776	25.958824		25.883292	-4.780292	0.03639056	8	28.135424	27.912613		28.0240185	-3.9310185	0.06556099
9	25.27153	25.324993		25.2982615	-4.1952615	0.05458841	9	27.869963	27.677767		27.773865	-3.680865	0.07797389
11	26.777634	26.437067		26.6073505	-5.5043505	0.02203055	11	27.954031	27.977985		27.966008	-3.873008	0.06825091
12	25.51911	26.782305		26.1507075	-5.0477075	0.03023351	12	27.037449	27.07407		27.0557595	-2.9627595	0.12826865
13	23.98441	24.117756		24.051083	-2.948083	0.12958018	13	26.179964	26.451113		26.3155385	-2.2225385	0.21426402
15	26.068527	25.83252		25.9505235	-4.8475235	0.03473362	15	30.030125	28.851982		29.4410535	-5.3480535	0.02455136
17	24.891254	25.011118		24.951186	-3.848186	0.06943534	17	Undetermine	27.865274		27.865274	-3.772274	0.07318674
18	23.690264	23.801146		23.745705	-2.642705	0.16012772	18	26.161512	25.774723		25.9681175	-1.8751175	0.27260473
19	26.442118	24.762121		25.6021195	-4.4991195	0.04422115	19	Undetermine	29.16736		29.16736	-5.07436	0.0296801
20	27.867474	27.914028		27.890751	-6.787751	0.00905072	20	29.680246	29.850525		29.7653855	-5.6723855	0.01960838
21	24.874174	24.931095		24.9026345	-3.7996345	0.07181184	21	27.889652	27.903908		27.89678	-3.80378	0.07160579
22	24.884045	24.883629		24.883837	-3.780837	0.07275363	22	26.81577	26.733864		26.774817	-2.681817	0.15584492
23	26.494434	26.843987		26.6692105	-5.5662105	0.02110589	23	28.675558	29.109896		28.892727	-4.799727	0.03590362
25	27.934841	27.931982		27.9334115	-6.8304115	0.00878701	25	29.686302	29.949385		29.8178435	-5.7248435	0.01890821
26	24.52299	24.52269		24.52284	-3.41984	0.09343844	26	26.71852	26.905567		26.8120435	-2.7190435	0.15187502
27	23.0935	23.239555		23.1665275	-2.0635275	0.23923038	27	26.066734	25.971899		26.0193165	-1.9263165	0.26310006
28	25.14835	25.353523		25.2509365	-4.1479365	0.05640878	28	25.9944	26.233511		26.1139555	-2.0209555	0.24639493
30	26.572353	26.5746		26.5734765	-5.4704765	0.02255394	30	29.102606	28.881397		28.9920015	-4.8990015	0.03351611
31	21.827963	21.880213		21.854088	-0.751088	0.59415531	31	24.271744	24.271322		24.271533	-0.178533	0.88360103
34	24.12826	24.49185		24.310055	-3.207055	0.10828798	34	28.196587	27.773561		27.985074	-3.892074	0.06735487
35	27.08219	27.11427		27.09823	-5.99523	0.01567675	35	28.772545	29.241571		29.007058	-4.914058	0.03316814
36	26.95689	26.453676		26.705283	-5.602283	0.02058471	36	28.295174	28.337698		28.316436	-4.223436	0.05353269
37	26.867334	26.765879		26.8166065	-5.7136065	0.01905606	37	28.569748	28.484613		28.5271805	-4.4341805	0.04625713
38	25.888977	25.734499		25.811738	-4.708738	0.03824095	38	27.398676	27.577723		27.4881995	-3.3951995	0.09504803
39	25.248867	25.568102		25.4084845	-4.3054845	0.05057315	39	27.971342	27.776203		27.8737725	-3.7807725	0.07275688
40	27.189833	26.79267		26.9912515	-5.8882515	0.01688339	40	28.666632	29.28679		28.976711	-4.883711	0.03387322
41	29.021332	29.48669		29.254011	-8.151011	0.00351804	41	29.253582	29.092718		29.17315	-5.08015	0.02956123
43	25.219921	25.238337		25.229129	-4.126129	0.05726792	43	28.05094	28.119818		28.085379	-3.992379	0.06283103
44	26.458519	26.919409		26.688964	-5.585964	0.02081888	44	29.558842	30.639425		30.0991335	-6.0061335	0.01555871
45	22.814909	23.231358		23.0231335	-1.9201335	0.26423006	45	27.466646	27.137085		27.3018655	-3.2088655	0.10815217
46	22.471235	22.660736		22.5659855	-1.4629855	0.3627417	46	28.437428	28.429838		28.433633	-4.340633	0.04935592
47	25.41169	25.398846		25.405268	-4.302268	0.05068603	47	29.471535	29.17275		29.3221425	-5.2291425	0.02666068
49	23.373081	23.848454		23.6107675	-2.5077675	0.17582748	49	27.14752	27.13742		27.14247	-3.04947	0.12078641
50	25.678833	26.248259		25.963546	-4.860546	0.03442151	50	28.414454	28.20957		28.312012	-4.219012	0.0536971
51	25.710663	25.814919		25.762791	-4.659791	0.03956062	51	28.065392	27.961765		28.0135785	-3.9205785	0.06603714
52	25.72949	26.002409		25.8659495	-4.7629495	0.03683065	52	28.089882	28.265392		28.177637	-4.084637	0.05893886
53	24.5449	24.043667		24.2942835	-3.1912835	0.10947827	53	26.367302	26.621145		26.4942235	-2.4012235	0.18930396
54	20.864653	21.082333		20.973493	0.129507	1.09391982	54	22.591022	22.83513		22.713076	1.379924	2.60254661
55	25.261326	25.358028		25.309677	-4.206677	0.05415818	55	29.800934	29.219145		29.5100395	-5.4170395	0.023405
56	27.478165	27.378452		27.4283085	-6.3253085	0.01247075	56	26.74455	27.734144		27.239347	-3.146347	0.11294192
57	23.803204	24.127823		23.9655135	-2.8625135	0.13749838	57	26.765968	28.022064		27.394016	-3.301016	0.10146007
58	27.60125	28.344137		27.9726935	-6.8696935	0.00855099	58	26.799057	26.849024		26.8240405	-2.7310405	0.15061731
var2csa	21.637093	21.563585		21.600339	-0.497339	0.70841222	*var2csa*	24.28919	24.415834		24.352512	-0.259512	0.83537044
91	23.697182	23.981886		23.839534	-2.736534	0.15004488	91	26.387035	26.592022		26.4895285	-2.3965285	0.18992102
92	22.516981	22.983273		22.750127	-1.647127	0.31927533	92	25.349411	25.743654		25.5465325	-1.4535325	0.3651263
93	25.785719	26.194073		25.989896	-4.886896	0.03379852	93	29.233238	29.512762		29.373	-5.28	0.02573722
94	27.595236	27.756573		27.6759045	-6.5729045	0.01050409	94	29.841923	30.125893		29.983908	-5.890908	0.01685233
95	25.523048	25.755453		25.6392505	-4.5362505	0.04309754	95	28.508244	28.304415		28.4063295	-4.3133295	0.05029889
96	27.947662	28.29044		28.119051	-7.016051	0.00772606	96	30.460213	30.837666		30.6489395	-6.5559395	0.01062834
97	27.45979	28.054207		27.7569985	-6.6539985	0.00992995	97	31.945633	31.98733		31.9664815	-7.8734815	0.00426428
60	21.027617	21.1786		21.1031085			60	23.998785	24.187744		24.0932645	-0.0002645	0.99981668

20xCho, Dep Ser 4Wks (Rep 2)							1xCho, Dep Ser 4 Wks (Rep 2)						
Primer pair	CT			CT Average	relative to p60	realtive copy number	Primer pair	CT			CT Average	relative to p60	realtive copy number
1	24.766531	25.267637		25.017084	-4.147084	0.05644212	1	25.049553	24.79983		24.9246915	-4.2246915	0.05348613
3	26.716587	28.777245		27.746916	-6.876916	0.00850828	3	27.429405	27.4898		27.4596025	-6.7596025	0.00922905
4	25.40382	25.43309		25.418455	-4.548455	0.0427345	4	25.853682	25.771635		25.8126585	-5.1126585	0.02890257
5	26.412977	26.70577		26.5593735	-5.6893735	0.01937885	5	26.52044	27.086563		26.8035015	-6.1035015	0.0145433
6	24.639528	25.14396		24.891744	-4.021744	0.06156508	6	24.82094	24.800316		24.810628	-4.110628	0.05788655
7	26.815496	26.87218		26.843838	-5.973838	0.01591093	7	27.376019	27.887693		27.631856	-6.931856	0.00819037
8	25.801962	25.866632		25.834297	-4.964297	0.03203301	8	25.762905	25.60633		25.6846175	-4.9846175	0.03158498
9	25.289347	25.039051		25.164199	-4.294199	0.05097031	9	25.190136	25.475548		25.332842	-4.632842	0.04030655
11	26.9131	26.899931		26.9065155	-6.0365155	0.01523448	11	26.382607	26.972218		26.6774125	-5.9774125	0.01587156
12	25.435621	25.490328		25.4629745	-4.5929745	0.04143591	12	25.149378	25.769646		25.459512	-4.759512	0.03691851
13	24.2886	24.205627		24.2471135	-3.3771135	0.09624707	13	24.21252	24.252527		24.2325235	-3.5325235	0.08641805
15	26.220861	26.06301		26.1419355	-5.2719355	0.02588149	15	26.972305	26.704283		26.838294	-6.138294	0.01419676
17	25.028543	25.080761		25.054652	-4.184652	0.05499133	17	25.35288	25.336674		25.344777	-4.644777	0.03997448
18	24.086315	23.97246		24.0293875	-3.1593875	0.11192564	18	24.304056	24.149578		24.226817	-3.526817	0.08676055
19	24.661936	24.708427		24.6851815	-3.8151815	0.07104212	19	24.406912	24.771511		24.5892115	-3.8892115	0.06748864
20	28.08592	27.845331		27.9656255	-7.0956255	0.00731146	20	28.327856	28.21596		28.271908	-7.571908	0.00525568
21	24.715803	24.593218		24.6545105	-3.7845105	0.07256861	21	24.324787	25.111929		24.718358	-4.018358	0.06170974
22	25.786407	25.662003		25.724205	-4.854205	0.03457313	22	26.246864	26.321915		26.2843895	-5.5843895	0.02084161
23	26.557413	26.99934		26.7783765	-5.9083765	0.01664951	23	27.630114	27.046106		27.33811	-6.63811	0.01003991
25	28.466257	28.7971		28.6316785	-7.7616785	0.00460789	25	28.015383	28.716032		28.3657075	-7.7657075	0.00492484
26	24.452509	24.727993		24.590251	-3.720251	0.07587398	26	25.018646	25.23964		25.129143	-4.429143	0.04641893
27	23.078218	23.680124		23.379171	-2.509171	0.17565652	27	23.232197	23.666582		23.4493895	-2.7493895	0.14871381
28	25.273983	25.361092		25.3175375	-4.4475375	0.04583084	28	25.211226	25.476297		25.3437615	-4.6437615	0.04000263
30	26.595318	26.320179		26.4577485	-5.5877485	0.02079314	30	26.42849	27.085129		26.7568095	-6.0568095	0.01502169
31	22.673027	23.00691		22.8399685	-1.9699685	0.2552586	31	21.979105	22.397465		22.188285	-1.488285	0.35643601
34	24.829237	24.627878		24.7285575	-3.8585575	0.06893796	34	25.22665	25.841595		25.5341225	-4.8341225	0.03505776
35	27.761734	27.485765		27.6237495	-6.7537495	0.00926657	35	27.186438	26.573984		26.880211	-6.180211	0.01379022
36	26.36299	26.561563		26.4622765	-5.5922765	0.02072798	36	27.209978	27.763111		27.4865445	-6.7865445	0.0090583
37	27.056879	27.129898		27.0933885	-6.2233885	0.01338361	37	26.738796	26.287775		26.5132855	-5.8132855	0.01778389
38	25.639095	25.10223		25.3706625	-4.5006625	0.04417388	38	25.64587	25.823486		25.734678	-5.034678	0.0305078
39	25.644197	25.506802		25.5754995	-4.7054995	0.03832688	39	25.294182	25.298923		25.2965525	-4.5965525	0.04133328
40	27.437195	27.3534		27.3952975	-6.5252975	0.0108565	40	27.66276	27.773956		27.718358	-7.018358	0.00771372
41	28.976038	28.676899		28.8264685	-7.9564685	0.00402591	41	28.071648	28.015732		28.04369	-7.34369	0.00615643
43	25.437828	25.630787		25.5343075	-4.6643075	0.03943697	43	25.687214	26.00979		25.848502	-5.148502	0.02819334
44	26.810226	27.161346		26.985786	-6.115786	0.01441999	44	26.722315	26.526056		26.6241855	-5.9241855	0.01646806
45	22.868334	22.788254		22.828294	-1.958294	0.25733257	45	23.349747	23.503405		23.426576	-2.726576	0.15108413
46	22.816193	22.679464		22.7478285	-1.8778285	0.27209295	46	23.048433	23.524687		23.28656	-2.58656	0.16648222
47	25.36047	25.513586		25.437028	-4.567028	0.04218787	47	25.944286	25.986233		25.9652595	-5.2652595	0.02600153
49	23.920174	23.714172		23.817173	-2.947173	0.12966194	49	23.8938	23.879742		23.886771	-3.186771	0.10982124
50	25.44446	25.744846		25.594653	-4.724653	0.03782141	50	25.874071	25.571692		25.7228815	-5.0228815	0.03075828
51	25.589487	25.265965		25.427726	-4.557726	0.04246076	51	25.614294	25.966934		25.790614	-5.090614	0.02934759
52	25.8271	26.024473		25.9257865	-5.0557865	0.03006468	52	26.446121	26.368036		26.4070785	-5.7070785	0.01914248
53	24.540264	24.191748		24.366006	-3.496006	0.08863338	53	24.718233	24.781391		24.749812	-4.049812	0.06037889
54	20.734396	21.133188		20.933792	-0.063792	0.95674609	54	21.04349	21.496107		21.2697985	-0.5697985	0.67371088
55	25.832258	25.630108		25.731183	-4.861183	0.03440631	55	25.354315	26.014936		25.6846255	-4.9846255	0.03158481
56	27.154951	26.775368		26.9651595	-6.0951595	0.01462764	56	27.01584	27.918941		27.4673905	-6.7673905	0.00917936
57	24.437368	24.436842		24.437105	-3.567105	0.08437123	57	24.097416	24.256395		24.1769055	-3.4769055	0.08981464
58	27.045645	22.157572		24.6016085	-3.7316085	0.07527901	58	26.35876	27.635836		26.997298	-6.297298	0.01271524
var2csa	19.241539	19.317408		19.2794735	1.5905265	3.01159235	*var2csa*	20.132078	20.595062		20.36357	0.33643	1.26262831
91	23.917896	24.112583		24.0152395	-3.1452395	0.11302866	91	23.850943	23.915785		23.883364	-3.183364	0.11008089
92	23.18473	23.365665		23.2751975	-2.4051975	0.18878323	92	22.157995	22.874481		22.516238	-1.816238	0.28396047
93	26.339504	26.217594		26.278549	-5.408549	0.02354315	93	26.373524	26.558939		26.4662315	-5.7662315	0.01837348
94	27.712975	27.493797		27.603386	-6.733386	0.00939829	94	28.0026	27.501198		27.751899	-7.051899	0.00753645
95	26.314718	26.124937		26.2198275	-5.3498275	0.02452118	95	25.791687	26.136324		25.9640055	-5.2640055	0.02602414
96	28.41969	28.73767		28.57868	-7.70868	0.00478031	96	27.548374	27.998549		27.7734615	-7.0734615	0.00742465
97	28.854599	28.560589		28.707594	-7.837594	0.00437169	97	27.69786	28.938204		28.318032	-7.618032	0.00509031
60	20.801058	20.944466		20.872762			60	20.444769	20.925238		20.6850035		

MCM 4Wks (Rep 3)							No Cho, Dep Ser 4Wks (Rep 3)						
Primer pair	CT			CT Average	relative to p60	realtive copy number	Primer pair	CT			CT Average	relative to p60	realtive copy number
1	26.27061	26.743742		26.507176	-3.907176	0.06665348	1	22.760721	22.54842		22.6545705	-3.6545705	0.07940807
3	28.025063	29.390469		28.707766	-6.107766	0.01450037	3	24.791176	24.979286		24.885231	-5.885231	0.01691878
4	26.797583	26.937572		26.8675775	-4.2675775	0.05191958	4	22.39406	23.159046		22.776553	-3.776553	0.07296999
5	27.378376	27.756943		27.5676595	-4.9676595	0.03195843	5	23.610357	24.203468		23.9069125	-4.9069125	0.03333283
6	26.160866	26.703453		26.4321595	-3.8321595	0.07021098	6	22.943747	23.292347		23.118047	-4.118047	0.05758964
7	26.729443	26.982124		26.8557835	-4.2557835	0.05234576	7	24.032238	24.707262		24.36975	-5.36975	0.02418489
8	29.216877	29.346325		29.281601	-6.681601	0.00974177	8	24.4213	24.156162		24.288731	-5.288731	0.02558193
9	28.343369	28.51822		28.4307945	-5.8307945	0.01756936	9	23.464655	23.406515		23.435585	-4.435585	0.04621212
11	28.08661	28.313517		28.2000635	-5.6000635	0.0206164	11	24.074812	24.139017		24.1069145	-5.1069145	0.02901787
12	25.723312	26.633114		26.178213	-3.578213	0.08372411	12	21.779802	22.335949		22.0578755	-3.0578755	0.12008472
13	24.53922	24.492554		24.515887	-1.915887	0.26500895	13	21.779339	21.987846		21.8835925	-2.8835925	0.13550401
15	27.82329	28.432426		28.127858	-5.527858	0.02167449	15	23.984467	24.580128		24.2822975	-5.2822975	0.02569627
17	25.384293	25.613243		25.498768	-2.898768	0.13408614	17	22.422493	22.362408		22.3924505	-3.3924505	0.09522931
18	25.14392	25.20409		25.174005	-2.574005	0.16793735	18	21.835848	22.619558		22.227703	-3.227703	0.10674919
19	25.145658	25.31275		25.229204	-2.629204	0.16163326	19	21.48166	21.952522		21.717091	-2.717091	0.1520807
20	29.389837	29.452963		29.4214	-6.8214	0.00884207	20	31.943789	29.522396		30.7330925	-11.733093	0.00029376
21	26.486843	26.977066		26.7319545	-4.1319545	0.05703714	21	22.681816	Undetermined		22.681816	-3.681816	0.07792251
22	27.509071	27.321676		27.4153735	-4.8153735	0.03551633	22	30.943254	32.618103		31.7806785	-12.780679	0.00014211
23	27.847042	28.12761		27.987326	-5.387326	0.02389204	23	Undetermine	24.838736		24.838736	-5.838736	0.01747291
25	29.616302	29.729622		29.672962	-7.072962	0.00742722	25	32.068703	25.444443		28.756573	-9.756573	0.00115606
26	26.463142	28.01279		27.237966	-4.637966	0.04016364	26	28.965284	23.09259		26.028937	-7.028937	0.00765736
27	25.29675	27.6429		26.469825	-3.869825	0.06840165	27	Undetermine	21.703302		21.703302	-2.703302	0.15354123
28	26.129002	26.464075		26.2965385	-3.6965385	0.07713137	28	22.639162	22.584929		22.6120455	-3.6120455	0.08178355
30	28.008678	28.07497		28.041824	-5.441824	0.02300635	30	23.769983	23.836802		23.8033925	-4.8033925	0.03581251
31	24.486046	24.103422		24.294734	-1.694734	0.30891161	31	20.722921	21.014587		20.868754	-1.868754	0.2738098
34	27.364264	28.020582		27.692423	-5.092423	0.02931082	34	22.859825	23.005856		22.9328405	-3.9328405	0.06547825
35	30.012068	30.340015		30.1760415	-7.5760415	0.00524064	35	25.594389	25.87864		25.7365145	-6.7365145	0.00937793
36	28.360413	29.729467		29.04494	-6.44494	0.01147836	36	24.285765	24.327364		24.3065645	-5.3065645	0.02526765
37	32.01348	31.675074		31.844277	-9.244277	0.0016489	37	24.853533	25.107391		24.980462	-5.980462	0.01583804
38	26.450645	27.279703		26.865174	-4.265174	0.05200615	38	23.04193	23.333185		23.1875575	-4.1875575	0.05488069
39	28.317245	28.37248		28.3448625	-5.7448625	0.01864765	39	23.75615	23.93729		23.84672	-4.84672	0.03475297
40	29.374004	30.842216		30.10811	-7.50811	0.0054933	40	25.18354	25.463675		25.3236075	-6.3236075	0.01248546
41	31.815493	31.805346		31.8104195	-9.2104195	0.00168806	41	26.31738	26.094263		26.2058215	-7.2058215	0.00677379
43	27.055147	26.9821		27.0186235	-4.4186235	0.04675863	43	23.17122	22.904825		23.0380225	-4.0380225	0.06087432
44	31.879494	Undetermined		31.879494	-9.279494	0.00160914	44	24.848331	24.9341		24.8912155	-5.8912155	0.01684874
45	26.427773	27.172014		26.7998935	-4.1998935	0.05441343	45	22.323683	22.177889		22.250786	-3.250786	0.1050548
46	28.643606	29.128233		28.8859195	-6.2859195	0.01281592	46	23.111383	22.936441		23.023912	-4.023912	0.06147263
47	28.849503	30.174269		29.511886	-6.911886	0.00830453	47	24.33408	24.509958		24.422019	-5.422019	0.02332436
49	24.62235	25.12836		24.875355	-2.275355	0.20656175	49	22.646194	22.438005		22.5420995	-3.5420995	0.08584634
50	28.80413	29.576897		29.1905135	-6.5905135	0.01037666	50	23.469324	24.026106		23.747715	-4.747715	0.03722163
51	28.606634	28.898088		28.752361	-6.152361	0.01405901	51	Undetermine	23.610619		23.610619	-4.610619	0.04093223
52	28.053915	28.999567		28.526741	-5.926741	0.01643892	52	24.318336	24.272133		24.2952345	-5.2952345	0.02546687
53	26.016388	26.082575		26.0494815	-3.4494815	0.09153825	53	Undetermine	22.82897		22.82897	-3.82897	0.07036637
54	21.621227	21.823616		21.7224215	0.8775785	1.8372889	54	19.294964	19.597021		19.4459925	-0.4459925	0.73407913
55	30.04121	33.669857		31.8555335	-9.2555335	0.00163609	55	25.968344	24.08015		25.024247	-6.024247	0.01536459
56	28.882135	28.872637		28.877386	-6.277386	0.01289195	56	32.488224	24.726068		28.607146	-9.607146	0.00128222
57	27.74151	27.847013		27.7942615	-5.1942615	0.02731313	57	24.759169	22.617117		23.688143	-4.688143	0.03879076
58	31.819578	33.23413		32.526854	-9.926854	0.00102735	58	23.912762	24.13453		24.023646	-5.023646	0.03074198
var2csa	26.756691	27.334478		27.0455845	-4.4455845	0.04589292	*var2csa*	21.399336	22.214018		21.806677	-2.806677	0.14292429
91	25.129255	25.002087		25.065671	-2.465671	0.18103355	91	21.758862	22.682749		22.2208055	-3.2208055	0.10726078
92	24.799627	25.325312		25.0624695	-2.4624695	0.18143573	92	21.355995	21.899069		21.627532	-2.627532	0.16182069
93	27.449802	27.93237		27.691086	-5.091086	0.02933799	93	23.171597	23.125984		23.1487905	-4.1487905	0.0563754
94	30.000263	30.22581		30.1130365	-7.5130365	0.00547458	94	26.56922	26.920252		26.744736	-7.744736	0.00466232
95	28.257948	28.332748		28.295348	-5.695348	0.01929876	95	24.954332	24.970127		24.9622295	-5.9622295	0.01603947
96	30.546825	30.360394		30.4536095	-7.8536095	0.00432343	96	26.286798	26.558104		26.422451	-7.422451	0.00582934
97	31.605843	32.34944		31.9776415	-9.3776415	0.00150331	97	26.073029	26.136194		26.1046115	-7.1046115	0.00726606
							60	19.034452	18.968113		19.0012825		

1x Cho, Dep Ser 4Wks (Rep 3)						
Primer pair	CT			CT Average	relative to p60	realtive copy number
1	22.90937	23.08052		22.994945	-3.044945	0.12116585
3	25.264297	25.294195		25.279246	-5.329246	0.02487351
4	22.891073	23.147514		23.0192935	-3.0692935	0.11913808
5	24.14909	24.82558		24.487335	-4.537335	0.04306516
6	23.632154	23.82159		23.726872	-3.776872	0.07295385
7	24.743826	25.138054		24.94094	-4.99094	0.03144686
8	24.197515	24.376005		24.28676	-4.33676	0.0494886
9	23.278267	23.482304		23.3802855	-3.4302855	0.09276436
11	24.757502	24.808283		24.7828925	-4.8328925	0.03508766
12	23.617546	23.696012		23.656779	-3.706779	0.07658581
13	23.111671	23.21386		23.1627655	-3.2127655	0.1078602
15	24.219114	24.4095		24.314307	-4.364307	0.04855262
17	23.0599	23.415152		23.237526	-3.287526	0.10241323
18	22.78059	23.118942		22.949766	-2.999766	0.12502028
19	22.73321	22.908344		22.820777	-2.870777	0.13671306
20	26.100615	25.924244		26.0124295	-6.0624295	0.01496328
21	23.34869	23.232769		23.2907295	-3.3407295	0.09870524
22	23.631786	23.791634		23.71171	-3.76171	0.07372461
23	24.822895	25.586008		25.2044515	-5.2544515	0.02619706
25	25.13788	25.454353		25.2961165	-5.3461165	0.02458434
26	23.25753	23.369776		23.313653	-3.363653	0.09714927
27	21.901249	22.061897		21.981573	-2.031573	0.24458825
28	22.952454	23.456385		23.2044195	-3.2544195	0.10479055
30	24.425934	24.516977		24.4714555	-4.5214555	0.04354179
31	20.793997	20.817183		20.80559	-0.85559	0.55263927
34	22.992987	22.22745		22.6102185	-2.6602185	0.15819561
35	25.06266	25.661158		25.361909	-5.411909	0.02348838
36	24.343702	24.872961		24.6083315	-4.6583315	0.03960067
37	24.540445	24.718723		24.629584	-4.679584	0.03902158
38	23.513775	23.731892		23.6228335	-3.6728335	0.07840919
39	24.055685	24.120598		24.0881415	-4.1381415	0.05679306
40	25.002977	25.171997		25.087487	-5.137487	0.02840942
41	26.039753	26.112469		26.076111	-6.126111	0.01431716
43	23.018406	23.176243		23.0973245	-3.1473245	0.11286542
44	24.460629	24.59265		24.5266395	-4.5766395	0.04190774
45	22.58365	22.35458		22.469115	-2.519115	0.17444994
46	23.60557	23.786377		23.6959735	-3.7459735	0.07453317
47	24.914246	24.481571		24.6979085	-4.7479085	0.03721664
49	22.89694	22.798393		22.8476665	-2.8976665	0.13418855
50	23.909481	23.823256		23.8663685	-3.9163685	0.06623013
51	24.26407	24.139467		24.2017685	-4.2517685	0.05249164
52	24.373735	24.44962		24.4116775	-4.4616775	0.04538384
53	22.58441	22.968033		22.7762215	-2.8262215	0.14100112
54	19.875721	19.82498		19.8503505	0.0996495	1.07151311
55	24.613379	24.873363		24.743371	-4.793371	0.03606214
56	24.747059	24.736767		24.741913	-4.791913	0.03609861
57	22.51553	22.425539		22.4705345	-2.5205345	0.17427838
58	24.501421	24.806122		24.6537715	-4.7037715	0.03837282
var2csa	20.000463	20.804022		20.4022425	-0.4522425	0.73090586
91	22.225946	24.212748		23.219347	-3.269347	0.10371188
92	22.357735	22.232819		22.295277	-2.345277	0.19678921
93	23.68568	24.284733		23.9852065	-4.0352065	0.06099325
94	25.977333	26.85042		26.4138765	-6.4638765	0.01132868
95	24.877687	24.920279		24.898983	-4.948983	0.03237484
96	25.386223	25.94421		25.6652165	-5.7152165	0.0190348
97	25.60983	25.400864		25.505347	-5.555347	0.02126542
60	19.881556	20.02637		19.953963		

Chapter 2.3 and 2.4 *var* panels

MCM Day 0 (Rep 1)						
Primer pair	CT			CT Average	relative to p60	realtive copy number
1	26.931082	27.131638		27.03136	-6.51636	0.01092396
3	27.510624	27.879713		27.6951685	-7.1801685	0.00689531
4	27.560629	27.800638		27.6806335	-7.1656335	0.00696513
5	24.205105	24.413015		24.30906	-3.79406	0.07208985
6	26.082508	26.230368		26.156438	-5.641438	0.02003355
7	26.815828	26.880657		26.8482425	-6.3332425	0.01240235
8	26.434206	26.432083		26.4331445	-5.9181445	0.01653716
9	26.268742	25.922853		26.0957975	-5.5807975	0.02089357
11	25.83665	26.418888		26.127769	-5.612769	0.02043564
12	24.637186	26.362179		25.4996825	-4.9846825	0.03158356
13	24.951578	23.802095		24.3768365	-3.8618365	0.06878146
15	22.343124	25.375061		23.8590925	-3.3440925	0.09847542
17	25.62259	23.417473		24.5200315	-4.0050315	0.06228241
18	24.840776	25.254776		25.047776	-4.532776	0.04320146
19	24.264257	28.266968		26.2656125	-5.7506125	0.01857347
21	28.871113	28.48493		28.6780215	-8.1630215	0.00348888
21	27.004606	26.073793		26.5391995	-6.0241995	0.0153651
22	25.968174	25.684132		25.826153	-5.311153	0.02518742
23	28.77616	27.719587		28.2478735	-7.7328735	0.00470081
25	29.149841	28.3779		28.7638705	-8.2488705	0.00328732
26	25.851505	25.717823		25.784664	-5.269664	0.02592227
27	23.959616	23.850058		23.904837	-3.389837	0.09540198
28	27.081238	27.362242		27.22174	-6.70674	0.00957349
30	26.987844	27.575926		27.281885	-6.766885	0.00918258
31	22.62391	22.808481		22.7161955	-2.2011955	0.21745737
34	24.103598	24.13716		24.120379	-3.605379	0.08216234
35	27.194693	26.837557		27.016125	-6.501125	0.01103993
36	27.406343	27.00846		27.2074015	-6.6924015	0.00966911
37	24.014599	25.177689		24.596144	-4.081144	0.05908173
38	26.173822	25.88639		26.030106	-5.515106	0.02186692
39	26.6514	26.759901		26.7056505	-6.1906505	0.01369079
40	Undetermine	Undetermined		0	0	0
41	27.123764	27.194231		27.1589975	-6.6439975	0.00999902
43	26.724064	26.86718		26.795622	-6.280622	0.01286306
44	26.446993	28.147367		27.29718	-6.78218	0.00908574
45	25.246407	25.218082		25.2322445	-4.7172445	0.03801613
46	27.068705	27.15929		27.1139975	-6.5989975	0.01031582
47	31.080566	31.815107		31.4478365	-10.932837	0.00051155
49	25.966726	25.687374		25.82705	-5.31205	0.02517176
50	26.983055	26.909843		26.946449	-6.431449	0.0115862
51	26.648962	26.747534		26.698248	-6.183248	0.01376122
52	27.83372	27.767368		27.800544	-7.285544	0.00640963
53	26.463495	26.476458		26.4699765	-5.9549765	0.01612031
54	18.138374	18.18982		18.164097	2.350903	5.10143456
55	26.971014	27.49315		27.232082	-6.717082	0.0095051
56	25.385275	24.950813		25.168044	-4.653044	0.03974607
57	25.387789	25.965584		25.6766865	-5.1616865	0.02793686
58	27.214611	27.662815		27.438713	-6.923713	0.00823673
var2csa	24.149904	24.593025		24.3714645	-3.8564645	0.06903805
91	25.859835	26.000244		25.9300395	-5.4150395	0.02343747
92	25.170557	25.857357		25.513957	-4.998957	0.0312726
93	26.850527	27.442804		27.1466655	-6.6316655	0.01008486
94	27.854473	28.189404		28.0219385	-7.5069385	0.00549777
95	26.724829	26.950666		26.8377475	-6.3227475	0.0124929
96	28.231144	28.321583		28.2763635	-7.7613635	0.00460889
97	29.68523	29.903013		29.7941215	-9.2791215	0.00160956
60	20.304968	20.724493		20.5147305		

(Rep 1) MCM 8Wk				
Primer pair	CT		relative to p60	realtive copy number
1	Undetermined		#VALUE!	#VALUE!
3	22.823		-6.843989	0.0087047
4	21.734		-5.75451	0.01852336
5	21.724		-5.744898	0.01864719
6	29.723		-13.743952	7.2888E-05
7	20.407		-4.428381	0.04644345
8	21.153		-5.173737	0.02770448
9	21.008		-5.02854	0.03063787
11	21.952		-5.973013	0.01592003
12	21.178		-5.199047	0.02722268
13	18.528		-2.54935	0.17083198
15	20.216		-4.236788	0.05303954
17	19.885		-3.90564	0.06672448
18	Undetermined		#VALUE!	#VALUE!
19	19.972		-3.99327	0.06279224
20	24.847		-8.867527	0.00214096
21	20.890		-4.910624	0.03324718
22	20.162		-4.183415	0.0550385
23	22.425		-6.446014	0.01146982
25	18.884		-2.905306	0.13347986
26	19.588		-3.60894	0.08195978
27	17.585		-1.606388	0.32841957
28	21.386		-5.407183	0.02356545
30	22.277		-6.29776	0.01271116
31	16.316		-0.33674	0.79182855
34	18.716		-2.7374	0.14995484
35	21.978		-5.99932	0.01563237
36	20.917		-4.938473	0.03261155
37	22.396		-6.41697	0.01170306
38	22.318		-6.338713	0.01235541
39	21.385		-5.406202	0.02358148
40	Undetermined		#VALUE!	#VALUE!
41	23.881		-7.902218	0.00418018
43	21.449		-5.470036	0.02256083
44	22.430		-6.451334	0.0114276
45	19.577		-3.598297	0.08256665
46	20.677		-4.697931	0.03852848
47	25.838		-9.859358	0.00107656
49	19.157		-3.178116	0.11048206
50	26.949		-10.970465	0.00049838
51	21.262		-5.282852	0.02568639
52	21.481		-5.502297	0.02206193
53	Undetermined		#VALUE!	#VALUE!
54	14.518		1.46126	2.75348739
55	22.578		-6.599331	0.01031344
56	21.855		-5.876406	0.01702259
57	20.919		-4.939812	0.0325813
58	21.464		-5.484608	0.0223341
var2csa	16.905		-0.925665	0.5264378
91	19.170		-3.191292	0.10947763
92	18.700		-2.721226	0.15164544
93	19.782		-3.802986	0.07164521
94	23.666		-7.68744	0.00485121
95	20.652		-4.672966	0.03920099
96	23.251		-7.271511	0.00647228
97	22.818		-6.83922	0.00873353
60	15.979		0.00044	1.00030503

(Rep 1) -Met-Ser 8Wks					(Rep 1) -Met 20xSer 8Wks				
Primer pair	CT		relative to p60	realtive copy number	Primer pair	CT		relative to p60	realtive copy number
1	24.014		-5.752	0.01855165	1	20.994		-4.172	0.05545975
3	24.514		-6.252	0.01311815	3	23.176		-6.354	0.01222747
4	23.874		-5.612	0.02044426	4	22.467		-5.645	0.01998044
5	23.606		-5.344	0.02461641	5	22.921		-6.099	0.01458517
6	22.808		-4.546	0.04280954	6	21.595		-4.773	0.03657493
7	22.482		-4.220	0.05366644	7	21.369		-4.547	0.04277949
8	22.922		-4.660	0.03955325	8	21.794		-4.972	0.03185719
9	22.801		-4.539	0.0430131	9	21.377		-4.555	0.04252659
11	23.380		-5.118	0.02880568	11	21.617		-4.795	0.03603064
12	22.828		-4.566	0.04222523	12	21.373		-4.551	0.04266959
13	20.287		-2.025	0.24567279	13	20.161		-3.339	0.09885337
15	22.467		-4.205	0.05422049	15	20.470		-3.648	0.07977763
17	21.723		-3.461	0.09083266	17	19.524		-2.702	0.1536593
18	21.846		-3.584	0.08337277	18	20.440		-3.618	0.08145246
19	21.938		-3.676	0.07826109	19	21.979		-5.157	0.02802072
20	26.993		-8.731	0.00235338	20	25.900		-9.078	0.00185025
21	22.709		-4.447	0.04583889	21	21.668		-4.846	0.03477249
22	21.696		-3.434	0.0925275	22	20.249		-3.427	0.09300126
23	23.883		-5.621	0.02032236	23	22.821		-5.999	0.01563309
25	20.854		-2.592	0.16583242	25	19.526		-2.704	0.15347547
26	21.514		-3.252	0.10496476	26	20.296		-3.474	0.08999739
27	19.568		-1.306	0.40438321	27	18.345		-1.523	0.34784127
28	23.362		-5.100	0.02915154	28	21.426		-4.604	0.04111592
30	24.252		-5.990	0.0157324	30	22.935		-6.113	0.01444746
31	18.385		-0.123	0.9185117	31	17.270		-0.448	0.73309192
34	21.781		-3.519	0.08725647	34	19.335		-2.513	0.1752485
35	24.419		-6.157	0.01401145	35	22.914		-6.092	0.01466068
36	22.912		-4.650	0.03983223	36	21.882		-5.060	0.02996928
37	22.525		-4.263	0.05208797	37	22.812		-5.990	0.01573357
38	24.538		-6.276	0.01290434	38	22.963		-6.141	0.01416633
39	23.553		-5.291	0.02553336	39	22.016		-5.194	0.02732594
40	Undetermined		#VALUE!	#VALUE!	40	33.558		-16.736	9.1636E-06
41	25.498		-7.236	0.00663526	41	23.504		-6.682	0.00974196
43	23.599		-5.337	0.02474567	43	21.603		-4.781	0.03636159
44	24.279		-6.017	0.01543975	44	23.274		-6.452	0.01142264
45	21.906		-3.644	0.08000617	45	19.992		-3.170	0.11108739
46	21.960		-3.698	0.07707603	46	20.630		-3.808	0.07139065
47	27.415		-9.153	0.00175696	47	25.935		-9.113	0.00180634
49	21.294		-3.032	0.12224734	49	19.843		-3.021	0.12318854
50	28.875		-10.613	0.00063865	50	27.517		-10.695	0.00060336
51	22.830		-4.568	0.04214987	51	21.381		-4.559	0.04241681
52	24.165		-5.903	0.01671181	52	22.896		-6.074	0.01484452
53	21.903		-3.641	0.08017943	53	20.475		-3.653	0.07948696
54	16.859		1.403	2.64396486	54	15.134		1.688	3.22243889
55	24.336		-6.074	0.01484082	55	22.287		-5.465	0.02263344
56	23.162		-4.900	0.03348942	56	22.320		-5.498	0.02212506
57	23.287		-5.025	0.03071862	57	21.393		-4.571	0.04208524
58	22.832		-4.570	0.04210268	58	21.717		-4.895	0.03359932
var2csa	18.879		-0.617	0.65201181	var2csa	17.654		-0.832	0.56183251
91	20.995		-2.733	0.15041163	91	19.682		-2.860	0.13771676
92	20.332		-2.070	0.23819433	92	19.477		-2.655	0.15882127
93	21.575		-3.313	0.10062314	93	20.493		-3.671	0.07852795
94	25.273		-7.011	0.00775084	94	23.706		-6.884	0.00846545
95	21.973		-3.711	0.07635339	95	20.951		-4.129	0.0571608
96	25.277		-7.015	0.00773181	96	23.710		-6.888	0.00844106
97	24.840		-6.578	0.01046724	97	23.256		-6.434	0.0115688
60	18.262		0.000	1.00020935	60	16.822		0.000	1.00015805

(Rep 1) -Ser 20xMet 8Wks					(Rep 2) MCM Day 0				
Primer pair	CT		relative to p60	realtive copy number	Primer pair	CT		relative to p60	realtive copy number
1	19.451		-2.997	0.12529667	1	21.899		-3.555	0.08507164
3	22.419		-5.965	0.01601331	3	23.773		-5.429	0.02320681
4	21.252		-4.798	0.03594363	4	Undetermined		#VALUE!	#VALUE!
5	21.243		-4.789	0.03617225	5	23.334		-4.990	0.03145892
6	20.497		-4.043	0.06065372	6	22.177		-3.833	0.07018449
7	20.671		-4.217	0.05377584	7	21.227		-2.883	0.13552153
8	20.793		-4.339	0.04942614	8	22.398		-4.054	0.06019523
9	19.592		-3.138	0.11363325	9	21.988		-3.644	0.0800035
11	21.232		-4.778	0.03643892	11	23.879		-5.535	0.0215691
12	19.414		-2.960	0.12848181	12	22.269		-3.925	0.06585113
13	17.554		-1.100	0.4664237	13	20.289		-1.945	0.25972918
15	19.706		-3.252	0.10493952	15	22.181		-3.837	0.06999322
17	19.204		-2.750	0.14860463	17	21.486		-3.142	0.11325236
18	19.806		-3.352	0.09793131	18	21.147		-2.803	0.14326221
19	20.184		-3.730	0.07534507	19	21.455		-3.111	0.11570475
20	24.593		-8.139	0.00354669	20	23.695		-5.351	0.02450869
21	20.784		-4.330	0.04971976	21	22.241		-3.897	0.06712388
22	18.812		-2.358	0.19500202	22	21.856		-3.512	0.08763585
23	21.880		-5.426	0.02325776	23	22.980		-4.636	0.0402299
25	18.214		-1.760	0.2951567	25	21.217		-2.873	0.13648715
26	18.552		-2.098	0.23353214	26	22.218		-3.874	0.06819019
27	16.627		-0.173	0.88717155	27	20.153		-1.809	0.28548048
28	20.182		-3.728	0.07547716	28	22.960		-4.616	0.04077504
30	21.805		-5.351	0.02450573	30	23.713		-5.369	0.0241909
31	15.923		0.531	1.44454285	31	18.679		-0.335	0.79274292
34	18.596		-2.142	0.22661213	34	20.846		-2.502	0.17656276
35	22.308		-5.854	0.01728648	35	23.286		-4.942	0.03252408
36	20.921		-4.467	0.04521076	36	22.714		-4.370	0.04835366
37	21.507		-5.053	0.03012657	37	22.542		-4.198	0.05450147
38	21.628		-5.174	0.02770605	38	22.262		-3.918	0.06614647
39	20.263		-3.809	0.07132477	39	22.354		-4.010	0.06208528
40	32.438		-15.984	1.5431E-05	40	Undetermined		#VALUE!	#VALUE!
41	22.731		-6.277	0.01289097	41	24.861		-6.517	0.0109189
43	20.509		-4.055	0.06017633	43	22.649		-4.305	0.05059957
44	21.590		-5.136	0.02843984	44	23.287		-4.943	0.03251272
45	18.754		-2.300	0.20302552	45	20.532		-2.188	0.21948344
46	18.868		-2.414	0.18769017	46	21.842		-3.498	0.08850458
47	25.376		-8.922	0.00206098	47	27.995		-9.651	0.00124365
49	18.288		-1.834	0.28056073	49	19.512		-1.168	0.44512949
50	26.323		-9.869	0.00106948	50	27.821		-9.477	0.00140337
51	20.854		-4.400	0.04735892	51	22.547		-4.203	0.05430854
52	21.612		-5.158	0.02800298	52	23.450		-5.106	0.0290432
53	19.876		-3.422	0.09328475	53	21.593		-3.249	0.10516583
54	13.779		2.675	6.38797205	54	15.728		2.616	6.13178452
55	21.574		-5.120	0.02874899	55	23.300		-4.956	0.03221564
56	21.609		-5.155	0.02806314	56	23.532		-5.188	0.02742969
57	20.485		-4.031	0.06117716	57	21.786		-3.442	0.09204499
58	20.735		-4.281	0.05144433	58	23.307		-4.963	0.03206582
var2csa	14.512		1.942	3.84248599	var2csa	19.245		-0.901	0.53569436
91	18.360		-1.906	0.26682708	91	21.301		-2.957	0.1287655
92	17.979		-1.525	0.34744584	92	20.998		-2.654	0.15893392
93	19.365		-2.911	0.13292644	93	22.406		-4.062	0.05986381
94	22.460		-6.006	0.01555566	94	23.956		-5.612	0.02044548
95	20.221		-3.767	0.07343568	95	22.236		-3.892	0.06737952
96	22.547		-6.093	0.01464797	96	24.001		-5.657	0.01982373
97	22.183		-5.729	0.0188512	97	24.150		-5.806	0.01787491
60	16.454		0.000	0.99977891	60	18.344		0.000	1.00019826

(Rep 2) MCM 2Wk							(Rep 2) -Met 2Wk						
Primer pair	CT			CT Average	relative to p60	realtive copy number	Primer pair	CT			CT Average	relative to p60	realtive copy number
1	23.114	23.034		23.074	-5.514	0.02188359	1	21.674	23.617		22.646	-4.154	0.05619159
3	23.490	23.384		23.437	-5.877	0.01701787	3	24.290	24.780		24.535	-6.043	0.01516375
4	22.742	22.765		22.754	-5.194	0.0273255	4	22.400	22.439		22.419	-3.927	0.06572536
5	22.454	22.790		22.622	-5.062	0.0299293	5	23.616	23.507		23.561	-5.069	0.02978134
6	22.145	21.650		21.897	-4.337	0.04946418	6	22.688	22.817		22.752	-4.260	0.05218162
7	22.170	22.256		22.213	-4.653	0.03974252	7	22.977	22.926		22.952	-4.460	0.04545152
8	22.924	22.840		22.882	-5.322	0.02499972	8	23.610	23.729		23.670	-5.178	0.02762897
9	21.613	21.726		21.669	-4.109	0.0579397	9	22.856	22.696		22.776	-4.284	0.05133311
11	23.547	23.642		23.595	-6.035	0.01525437	11	24.960	25.327		25.143	-6.651	0.00994964
12	21.420	21.336		21.378	-3.818	0.07089462	12	22.766	22.937		22.852	-4.360	0.0487085
13	Undetermine	20.663		20.663	-3.103	0.11641905	13	Undetermine	22.423		22.423	-3.931	0.06556842
15	22.808	22.577		22.692	-5.132	0.02850919	15	23.528	23.514		23.521	-5.029	0.03062637
17	21.138	20.965		21.051	-3.491	0.08892273	17	21.347	21.337		21.342	-2.850	0.13866989
18	20.489	20.405		20.447	-2.887	0.13521785	18	21.193	21.271		21.232	-2.740	0.14964454
19	20.820	20.806		20.813	-3.253	0.10488887	19	21.552	21.783		21.668	-3.176	0.11066451
20	24.618	25.158		24.888	-7.328	0.00622178	20	25.681	25.635		25.658	-7.166	0.00696181
21	21.843	22.134		21.988	-4.428	0.04644218	21	22.262	22.551		22.407	-3.915	0.06631525
22	21.328	21.271		21.300	-3.740	0.07486081	22	22.442	22.426		22.434	-3.942	0.06507456
23	22.949	22.847		22.898	-5.338	0.0247239	23	23.289	23.397		23.343	-4.851	0.03464596
25	22.438	22.318		22.378	-4.818	0.03544804	25	22.910	23.146		23.028	-4.536	0.04310238
26	21.742	21.692		21.717	-4.157	0.05607076	26	23.586	23.466		23.526	-5.034	0.03052444
27	20.825	20.783		20.804	-3.244	0.10555812	27	21.723	Undetermined		21.723	-3.231	0.10649031
28	23.530	23.373		23.452	-5.892	0.0168422	28	24.839	24.808		24.823	-6.331	0.01241833
30	23.227	23.269		23.248	-5.688	0.01939253	30	23.645	23.664		23.654	-5.162	0.0279272
31	19.521	19.565		19.543	-1.983	0.2529768	31	20.385	20.486		20.436	-1.944	0.25996089
34	21.167	21.329		21.248	-3.688	0.07759115	34	22.309	22.392		22.350	-3.858	0.0689448
35	23.624	23.542		23.583	-6.023	0.0153795	35	24.269	23.950		24.109	-5.617	0.02037057
36	22.514	22.685		22.600	-5.040	0.03040443	36	23.125	23.159		23.142	-4.650	0.03983105
37	22.885	23.170		23.028	-5.468	0.02259979	37	22.746	22.595		22.671	-4.179	0.05522217
38	22.344	22.421		22.382	-4.822	0.0353431	38	23.157	23.167		23.162	-4.670	0.03928837
39	22.825	22.956		22.891	-5.331	0.02484965	39	23.382	23.285		23.334	-4.842	0.03487518
40	34.578	34.666		34.622	-17.062	7.3084E-06	40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
41	26.117	25.875		25.996	-8.436	0.00288749	41	26.298	26.729		26.513	-8.021	0.00384856
43	21.702	21.955		21.828	-4.268	0.05188697	43	23.108	23.112		23.110	-4.618	0.04073211
44	23.287	22.875		23.081	-5.521	0.02177406	44	24.214	24.445		24.330	-5.838	0.01748654
45	20.938	21.015		20.976	-3.416	0.09366936	45	21.701	21.672		21.687	-3.195	0.10923227
46	22.248	22.268		22.258	-4.698	0.03852701	46	22.931	23.187		23.059	-4.567	0.04219982
47	26.350	27.384		26.867	-9.307	0.00157885	47	28.373	28.776		28.574	-10.082	0.00092234
49	20.540	20.837		20.689	-3.129	0.1143264	49	21.443	21.584		21.514	-3.022	0.12314466
50	27.513	26.443		26.978	-9.418	0.00146201	50	28.494	28.679		28.586	-10.094	0.00091476
51	21.878	21.802		21.840	-4.280	0.0514705	51	22.917	22.924		22.921	-4.429	0.04643735
52	23.245	23.499		23.372	-5.812	0.01779495	52	23.697	23.546		23.621	-5.129	0.02856881
53	21.133	21.241		21.187	-3.627	0.08094353	53	21.530	21.511		21.521	-3.029	0.12254767
54	16.401	16.421		16.411	1.149	2.21823621	54	17.201	16.966		17.083	1.409	2.65484659
55	23.418	23.632		23.525	-5.965	0.01600605	55	23.834	24.188		24.011	-5.519	0.02181009
56	22.832	22.876		22.854	-5.294	0.02548629	56	23.666	23.693		23.680	-5.188	0.02743961
57	21.937	21.990		21.964	-4.404	0.04724528	57	22.630	22.684		22.657	-4.165	0.0557493
58	22.437	22.472		22.455	-4.895	0.03362066	58	23.262	Undetermined		23.262	-4.770	0.03665719
var2csa	18.560	18.905		18.733	-1.173	0.44365188	var2csa	19.995	19.212		19.603	-1.111	0.46286343
91	20.674	20.662		20.668	-3.108	0.11599813	91	21.621	21.784		21.703	-3.211	0.10802694
92	20.631	20.646		20.639	-3.079	0.11837456	92	21.523	21.496		21.510	-3.018	0.12348781
93	21.463	21.462		21.463	-3.903	0.06686537	93	22.555	Undetermined		22.555	-4.063	0.05981972
94	24.169	24.346		24.258	-6.698	0.00963483	94	24.789	25.447		25.118	-6.626	0.01012443
95	22.971	23.000		22.986	-5.426	0.02326777	95	24.483	24.798		24.641	-6.149	0.01409507
96	23.919	23.844		23.882	-6.322	0.0125013	96	24.340	24.570		24.455	-5.963	0.01603069
97	23.797	24.111		23.954	-6.394	0.01189084	97	25.364	25.435		25.399	-6.907	0.00832996
60	17.671	17.448		17.560	0.000	1.0003449	60	18.502	18.482		18.492	0.000	1.00004194

(Rep 2) -ChoSer 2Wk							(Rep 2,1) +20xMet 2Wk						
Primer pair	CT			CT Average	relative to p60	realtive copy number	Primer pair	CT			CT Average	relative to p60	realtive copy number
1	23.313	23.406		23.360	-4.462	0.04538261	1	23.264	22.349		22.807	-4.479	0.04485711
3	25.423	25.271		25.347	-6.449	0.0114423	3	24.284	24.371		24.328	-6.000	0.01562922
4	23.852	23.692		23.772	-4.874	0.03409655	4	Undetermine	22.976		22.976	-4.648	0.03989467
5	23.441	24.568		24.005	-5.107	0.0290178	5	23.432	23.502		23.467	-5.139	0.02838791
6	24.210	23.824		24.017	-5.119	0.02877395	6	22.879	22.955		22.917	-4.589	0.04155604
7	24.505	24.411		24.458	-5.560	0.02120272	7	22.995	23.232		23.113	-4.785	0.03626751
8	24.864	24.787		24.826	-5.928	0.01642704	8	24.568	24.586		24.577	-6.249	0.01314942
9	23.935	23.743		23.839	-4.941	0.0325573	9	22.915	23.204		23.059	-4.731	0.03764658
11	25.643	25.475		25.559	-6.661	0.00988178	11	25.239	25.004		25.122	-6.794	0.00901457
12	Undetermine	22.893		22.893	-3.995	0.06273203	12	23.012	22.642		22.827	-4.499	0.04422339
13	22.393	22.325		22.359	-3.461	0.09080912	13	21.765	Undetermined		21.765	-3.437	0.09232217
15	24.589	24.417		24.503	-5.605	0.02054535	15	23.453	23.292		23.373	-5.045	0.03030049
17	22.837	22.773		22.805	-3.907	0.06666487	17	21.492	21.731		21.611	-3.283	0.10270394
18	22.717	22.570		22.643	-3.745	0.07455909	18	21.199	21.219		21.209	-2.881	0.13571245
19	23.011	22.923		22.967	-4.069	0.05959706	19	21.617	21.737		21.677	-3.349	0.09815483
20	26.451	26.096		26.273	-7.375	0.00602252	20	25.460	25.745		25.603	-7.275	0.00645706
21	23.732	23.601		23.666	-4.768	0.03669531	21	22.933	22.816		22.875	-4.547	0.04279224
22	23.144	23.008		23.076	-4.178	0.05524842	22	22.013	22.181		22.097	-3.769	0.07334267
23	24.827	24.811		24.819	-5.921	0.01650145	23	23.563	23.604		23.584	-5.256	0.02617711
25	23.757	24.131		23.944	-5.046	0.03027057	25	23.187	23.448		23.317	-4.989	0.03147974
26	23.883	23.947		23.915	-5.017	0.03088517	26	22.893	23.185		23.039	-4.711	0.03817961
27	22.707	22.458		22.582	-3.684	0.07779054	27	21.675	21.774		21.724	-3.396	0.09497641
28	26.644	25.094		25.869	-6.971	0.00797231	28	24.763	24.559		24.661	-6.333	0.01240559
30	25.194	25.398		25.296	-6.398	0.01185752	30	24.116	23.888		24.002	-5.674	0.01958237
31	21.388	21.402		21.395	-2.497	0.17716009	31	20.410	20.361		20.385	-2.057	0.24027284
34	22.878	22.948		22.913	-4.015	0.06186598	34	Undetermine	22.124		22.124	-3.796	0.07200545
35	25.434	25.347		25.391	-6.493	0.01110506	35	23.999	24.335		24.167	-5.839	0.01746531
36	24.607	24.525		24.566	-5.668	0.01966871	36	23.264	23.430		23.347	-5.019	0.03084364
37	24.409	24.232		24.320	-5.422	0.02331848	37	23.397	22.988		23.193	-4.865	0.03432193
38	23.925	23.934		23.929	-5.031	0.03058283	38	23.663	23.567		23.615	-5.287	0.02561287
39	24.281	24.142		24.212	-5.314	0.02514505	39	23.408	23.432		23.420	-5.092	0.02931386
40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!	40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
41	27.577	27.309		27.443	-8.545	0.00267743	41	26.658	26.594		26.626	-8.298	0.00317716
43	24.316	24.417		24.366	-5.468	0.02258794	43	22.833	22.802		22.817	-4.489	0.04452689
44	25.200	25.358		25.279	-6.381	0.01200039	44	24.363	24.211		24.287	-5.959	0.01607823
45	22.646	22.552		22.599	-3.701	0.07689918	45	21.774	21.828		21.801	-3.473	0.09007197
46	23.889	23.842		23.866	-4.968	0.03195643	46	23.008	23.010		23.009	-4.681	0.03897415
47	28.874	28.153		28.513	-9.615	0.001275	47	27.920	28.308		28.114	-9.786	0.00113269
49	22.660	22.596		22.628	-3.730	0.07536769	49	21.846	21.601		21.724	-3.396	0.09502135
50	28.637	28.675		28.656	-9.758	0.00115521	50	29.523	29.261		29.392	-11.064	0.00046699
51	23.970	23.956		23.963	-5.065	0.02988148	51	22.934	22.836		22.885	-4.557	0.04247819
52	24.836	24.841		24.838	-5.940	0.01628444	52	24.416	24.707		24.561	-6.233	0.01329336
53	22.560	22.251		22.406	-3.508	0.0879262	53	21.837	21.918		21.878	-3.550	0.08540246
54	17.716	17.656		17.686	1.212	2.31668999	54	17.186	Undetermined		17.186	1.142	2.20673746
55	25.174	25.189		25.182	-6.284	0.01283569	55	24.158	24.214		24.186	-5.858	0.01724543
56	24.805	24.801		24.803	-5.905	0.01668535	56	23.624	23.594		23.609	-5.281	0.02571634
57	23.360	23.486		23.423	-4.525	0.04343318	57	23.137	23.265		23.201	-4.873	0.03413288
58	24.593	24.460		24.527	-5.629	0.02021204	58	23.341	23.684		23.513	-5.185	0.02749844
var2csa	20.196	20.158		20.177	-1.279	0.41217074	var2csa	16.940	17.128		17.034	1.294	2.45262472
91	22.753	22.844		22.799	-3.901	0.06696117	91	21.600	21.566		21.583	-3.255	0.10473979
92	22.419	22.437		22.428	-3.530	0.08655185	92	21.840	21.915		21.878	-3.550	0.08539122
93	23.605	23.688		23.647	-4.749	0.03720123	93	22.232	22.254		22.243	-3.915	0.06629887
94	25.870	25.877		25.873	-6.975	0.00794633	94	24.823	24.965		24.894	-6.566	0.01055697
95	25.208	25.188		25.198	-6.300	0.01269109	95	23.659	23.678		23.669	-5.341	0.02467596
96	25.453	25.731		25.592	-6.694	0.00966128	96	24.724	24.639		24.681	-6.353	0.01222943
97	25.417	25.556		25.486	-6.588	0.01039334	97	24.804	24.962		24.883	-6.555	0.01063489
60	18.934	18.862		18.898	0.000	1.00021109	60	18.260	18.395		18.328	0.000	1.00011923

(Rep 2,2) 20xMet 2Wk							(Rep 2,3) 20xMet 2Wk				
Primer pair	CT			CT Average	relative to p60	realtive copy number	Primer pair	CT		relative to p60	realtive copy number
1	20.476	20.321		20.398	-3.322	0.09998434	1	23.807		-4.797	0.03598175
3	28.582	22.472		25.527	-8.451	0.00285751	3	24.535		-5.525	0.02171526
4	21.965	22.350		22.158	-5.082	0.02953301	4	23.861		-4.851	0.03465028
5	21.605	21.523		21.564	-4.488	0.04456716	5	24.575		-5.565	0.02112812
6	20.980	21.142		21.061	-3.985	0.06313431	6	23.759		-4.749	0.03718993
7	21.271	21.236		21.254	-4.178	0.05525805	7	23.579		-4.569	0.04213798
8	21.790	21.942		21.866	-4.790	0.03614137	8	24.814		-5.804	0.01789314
9	20.749	20.665		20.707	-3.631	0.08071883	9	23.211		-4.201	0.05438765
11	22.938	22.954		22.946	-5.870	0.01710123	11	25.340		-6.330	0.01242606
12	20.302	20.513		20.407	-3.331	0.0993421	12	23.283		-4.273	0.05172222
13	19.854	20.101		19.978	-2.902	0.13382031	13	21.988		-2.978	0.12694188
15	21.499	21.597		21.548	-4.472	0.04506309	15	24.348		-5.338	0.02471619
17	19.664	19.610		19.637	-2.561	0.16943825	17	22.214		-3.204	0.10853754
18	19.375	19.462		19.419	-2.343	0.19714103	18	22.225		-3.215	0.10772171
19	20.339	20.311		20.325	-3.249	0.10518213	19	22.341		-3.331	0.09938108
20	24.756	24.755		24.755	-7.679	0.00487831	20	27.240		-8.230	0.00333172
21	21.387	20.681		21.034	-3.958	0.0643341	21	24.237		-5.227	0.02670356
22	20.488	20.579		20.533	-3.457	0.09103436	22	22.840		-3.830	0.07029579
23	21.821	21.840		21.831	-4.755	0.03704176	23	24.641		-5.631	0.02018015
25	21.617	21.642		21.629	-4.553	0.04258637	25	23.243		-4.233	0.05317913
26	21.536	21.413		21.475	-4.399	0.04740244	26	23.606		-4.596	0.04136077
27	19.737	20.694		20.215	-3.139	0.11348083	27	22.343		-3.333	0.09925063
28	22.501	22.264		22.382	-5.306	0.02526929	28	24.578		-5.568	0.02107277
30	22.547	22.439		22.493	-5.417	0.0234113	30	25.622		-6.612	0.01022376
31	18.800	18.781		18.790	-1.714	0.30473751	31	22.237		-3.227	0.10678952
34	20.285	20.379		20.332	-3.256	0.10468386	34	23.458		-4.448	0.04582803
35	22.477	22.413		22.445	-5.369	0.02419615	35	24.966		-5.956	0.01611346
36	21.430	21.687		21.559	-4.483	0.04472987	36	24.374		-5.364	0.0242834
37	23.458	23.237		23.347	-6.271	0.01294482	37	24.424		-5.414	0.02346091
38	21.018	21.484		21.251	-4.175	0.05535366	38	23.967		-4.957	0.03219474
39	21.209	21.224		21.217	-4.141	0.05669715	39	24.180		-5.170	0.02776707
40	33.146	33.752		33.449	-16.373	1.1783E-05	40	Undetermined		#VALUE!	#VALUE!
41	24.862	25.297		25.079	-8.003	0.00389709	41	26.574		-7.564	0.00528407
43	21.005	21.459		21.232	-4.156	0.0560881	43	23.690		-4.680	0.03900384
44	22.867	23.103		22.985	-5.909	0.01664138	44	25.433		-6.423	0.01165532
45	19.710	19.619		19.665	-2.589	0.16624265	45	22.748		-3.738	0.07494957
46	21.250	21.297		21.274	-4.198	0.05449649	46	23.956		-4.946	0.03245258
47	27.255	27.214		27.235	-10.159	0.00087492	47	30.166		-11.156	0.00043837
49	20.361	20.416		20.388	-3.312	0.10066719	49	22.824		-3.814	0.07107687
50	25.787	27.398		26.592	-9.516	0.00136539	50	29.868		-10.858	0.0005386
51	21.245	21.149		21.197	-4.121	0.05745652	51	24.283		-5.273	0.02585514
52	22.818	22.794		22.806	-5.730	0.01884348	52	25.593		-6.583	0.01043407
53	19.774	19.800		19.787	-2.711	0.15271387	53	22.592		-3.582	0.08347801
54	15.791	15.571		15.681	1.395	2.63015806	54	18.525		0.486	1.40007101
55	22.791	22.903		22.847	-5.771	0.01831671	55	25.188		-6.178	0.0138136
56	22.142	22.661		22.401	-5.325	0.0249384	56	25.022		-6.012	0.01549604
57	20.636	20.753		20.695	-3.619	0.08140756	57	23.502		-4.492	0.04444793
58	21.707	21.773		21.740	-4.664	0.0394445	58	24.941		-5.931	0.01638504
var2csa	15.499	15.566		15.533	1.543	2.91470424	var2csa	17.857		1.153	2.22398491
91	19.963	19.804		19.884	-2.808	0.14282808	91	22.242		-3.232	0.10640899
92	19.963	19.894		19.928	-2.852	0.13848168	92	22.243		-3.233	0.10634403
93	20.544	20.675		20.610	-3.534	0.0863582	93	23.601		-4.591	0.04148401
94	23.577	23.361		23.469	-6.393	0.01189993	94	25.861		-6.851	0.00866252
95	21.832	21.859		21.846	-4.770	0.03665894	95	25.170		-6.160	0.0139885
96	22.997	22.686		22.842	-5.766	0.0183785	96	25.533		-6.523	0.01087514
97	23.160	22.989		23.074	-5.998	0.01564184	97	25.608		-6.598	0.01032184
60	17.140	17.013		17.076	0.000	0.99983331	60	19.010		0.000	0.99973941

(Rep 2,1) -Cho+20x Ser 2Wk							(Rep 2,2) 20xSer 2Wk				
Primer pair	CT			CT Average	relative to p60	realtive copy number	Primer pair	CT		relative to p60	realtive copy number
1	31.190	21.449		26.319	-8.452	0.00285492	1	22.368		-4.412	0.046982
3	23.526	23.556		23.541	-5.674	0.01959068	3	23.970		-6.014	0.01546896
4	22.232	22.330		22.281	-4.414	0.04690558	4	22.979		-5.023	0.03075959
5	22.500	22.662		22.581	-4.714	0.03810269	5	23.360		-5.404	0.02361488
6	22.021	21.950		21.986	-4.119	0.05757144	6	22.928		-4.972	0.03187175
7	21.772	21.781		21.777	-3.910	0.06654543	7	21.834		-3.878	0.0679963
8	22.959	23.222		23.091	-5.224	0.02675963	8	23.558		-5.602	0.02059492
9	21.613	21.701		21.657	-3.790	0.07230524	9	22.232		-4.276	0.05162927
11	23.798	23.741		23.769	-5.902	0.0167206	11	24.341		-6.385	0.0119682
12	21.530	21.603		21.567	-3.700	0.07695266	12	21.955		-3.999	0.06253267
13	Undetermine	20.910		20.910	-3.043	0.12130812	13	21.515		-3.559	0.08487321
15	22.653	23.187		22.920	-5.053	0.03012819	15	23.481		-5.525	0.02172404
17	20.651	20.618		20.634	-2.767	0.14686276	17	21.259		-3.303	0.10134564
18	20.389	20.371		20.380	-2.513	0.17521929	18	20.880		-2.924	0.13175156
19	20.748	20.698		20.723	-2.856	0.1381614	19	21.360		-3.404	0.09447622
20	23.717	23.792		23.754	-5.887	0.01689431	20	24.921		-6.965	0.00800466
21	21.404	21.425		21.415	-3.548	0.08549925	21	22.620		-4.664	0.03944281
22	21.240	21.302		21.271	-3.404	0.0944626	22	21.701		-3.745	0.07456787
23	22.658	22.658		22.658	-4.791	0.03612053	23	23.456		-5.500	0.02209611
25	21.799	21.830		21.814	-3.947	0.06483769	25	22.494		-4.538	0.04303684
26	22.103	22.001		22.052	-4.185	0.05496949	26	22.850		-4.894	0.03363525
27	21.229	20.764		20.997	-3.130	0.11426247	27	21.402		-3.446	0.09173112
28	23.837	23.805		23.821	-5.954	0.01613	28	23.851		-5.895	0.01680156
30	23.483	23.437		23.460	-5.593	0.02071522	30	23.813		-5.857	0.01725037
31	19.659	19.611		19.635	-1.768	0.2936102	31	21.020		-3.064	0.11955223
34	21.598	21.705		21.652	-3.785	0.07255151	34	22.291		-4.335	0.04955794
35	23.300	23.368		23.334	-5.467	0.02260797	35	23.636		-5.680	0.01949857
36	22.434	22.396		22.415	-4.548	0.04275081	36	22.971		-5.015	0.03091661
37	23.625	23.573		23.599	-5.732	0.01881022	37	24.405		-6.449	0.01144745
38	22.203	22.116		22.160	-4.293	0.05101985	38	22.779		-4.823	0.03531872
39	22.142	22.237		22.189	-4.322	0.04998907	39	22.704		-4.748	0.03722568
40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!	40	Undetermined		#VALUE!	#VALUE!
41	25.917	25.846		25.881	-8.014	0.00386825	41	25.336		-7.380	0.00600277
43	22.151	22.215		22.183	-4.316	0.05021031	43	22.825		-4.869	0.03422222
44	23.283	23.232		23.257	-5.390	0.02384082	44	23.775		-5.819	0.01771788
45	20.847	20.852		20.849	-2.982	0.12653707	45	21.903		-3.947	0.06484931
46	21.940	22.210		22.075	-4.208	0.05411082	46	22.517		-4.561	0.04237173
47	26.904	27.010		26.957	-9.090	0.00183474	47	30.282		-12.326	0.00019479
49	20.725	20.745		20.735	-2.868	0.1369948	49	22.251		-4.295	0.05092546
50	26.977	27.277		27.127	-9.260	0.0016305	50	29.575		-11.619	0.00031795
51	21.825	21.919		21.872	-4.005	0.06228562	51	22.689		-4.733	0.0375942
52	22.855	22.902		22.878	-5.011	0.03100285	52	24.131		-6.175	0.01383973
53	21.335	21.328		21.331	-3.464	0.09060334	53	21.170		-3.214	0.10779618
54	15.986	15.952		15.969	1.898	3.72792026	54	17.386		0.570	1.48484362
55	22.996	22.997		22.996	-5.129	0.02856751	55	24.152		-6.196	0.01363792
56	23.233	23.407		23.320	-5.453	0.02282647	56	23.861		-5.905	0.01668746
57	21.619	21.599		21.609	-3.742	0.07474374	57	21.868		-3.912	0.06642265
58	22.849	22.849		22.849	-4.982	0.03163931	58	22.925		-4.969	0.03192594
var2csa	17.880	17.984		17.932	-0.065	0.9557969	var2csa	19.285		-1.329	0.39801977
91	20.897	22.543		21.720	-3.853	0.06919912	91	21.375		-3.419	0.09350537
92	20.705	20.688		20.697	-2.830	0.14066916	92	21.751		-3.795	0.07205778
93	21.709	21.733		21.721	-3.854	0.06915074	93	22.612		-4.656	0.0396676
94	23.603	23.633		23.618	-5.751	0.01856758	94	24.702		-6.746	0.00931559
95	22.536	22.638		22.587	-4.720	0.03794205	95	23.884		-5.928	0.01642146
96	23.593	23.422		23.507	-5.640	0.02005188	96	23.547		-5.591	0.02074435
97	23.928	24.119		24.023	-6.156	0.01402077	97	24.224		-6.268	0.01298008
60	18.093	17.642		17.867	0.000	0.99967531	60	17.956		0.000	0.99966457

(Rep 2) -Ser+20xCho 2Wk							(Rep 2) -Ser +1xCho 2Wk						
Primer pair	CT			CT Average	relative to p60	realtive copy number	Primer pair	CT			CT Average	relative to p60	realtive copy number
1	22.520	22.432		22.476	-4.766	0.03675861	1	21.130	22.137		21.634	-4.071	0.05950303
3	23.905	23.828		23.866	-6.156	0.01402002	3	23.933	23.946		23.939	-6.376	0.01203833
4	22.427	22.562		22.495	-4.785	0.03627686	4	22.887	22.322		22.604	-5.041	0.03036962
5	22.972	23.181		23.076	-5.366	0.02424037	5	23.651	23.601		23.626	-6.063	0.01495512
6	22.711	22.701		22.706	-4.996	0.0313365	6	31.081	22.943		27.012	-9.449	0.00143079
7	23.273	23.145		23.209	-5.499	0.02210984	7	23.431	23.602		23.516	-5.953	0.01613773
8	23.802	23.673		23.737	-6.027	0.01533194	8	24.161	23.937		24.049	-6.486	0.0111561
9	22.012	21.915		21.964	-4.254	0.05242541	9	22.634	22.714		22.674	-5.111	0.0289348
11	23.913	23.930		23.922	-6.212	0.01349272	11	20.628	23.801		22.214	-4.651	0.03978867
12	21.393	21.449		21.421	-3.711	0.07637051	12	21.978	21.914		21.946	-4.383	0.04792896
13	Undetermine	21.274		21.274	-3.564	0.08455214	13	21.640	21.313		21.476	-3.913	0.06636229
15	22.687	22.630		22.658	-4.948	0.03239317	15	22.990	23.290		23.140	-5.577	0.02094821
17	21.119	21.166		21.142	-3.432	0.09263357	17	21.834	21.883		21.859	-4.296	0.05091503
18	21.239	21.251		21.245	-3.535	0.08627291	18	21.637	21.460		21.548	-3.985	0.06314269
19	21.453	21.656		21.554	-3.844	0.06961408	19	22.309	22.163		22.236	-4.673	0.03920328
20	24.818	24.749		24.784	-7.074	0.0074239	20	26.774	26.894		26.834	-9.271	0.00161876
21	21.856	21.915		21.886	-4.176	0.055332	21	22.930	23.403		23.167	-5.604	0.02056094
22	21.632	21.558		21.595	-3.885	0.06770598	22	21.872	22.273		22.072	-4.509	0.04391372
23	23.168	23.228		23.198	-5.488	0.02228754	23	23.859	23.902		23.880	-6.317	0.0125402
25	22.415	22.443		22.429	-4.719	0.03797455	25	23.353	23.597		23.475	-5.912	0.01661058
26	22.478	22.469		22.474	-4.764	0.03681213	26	22.469	22.899		22.684	-5.121	0.02873598
27	21.096	21.128		21.112	-3.402	0.0945862	27	21.701	21.964		21.832	-4.269	0.05186564
28	23.435	23.536		23.485	-5.775	0.01825615	28	23.772	24.126		23.949	-6.386	0.01195793
30	23.854	23.717		23.785	-6.075	0.01482851	30	24.496	24.629		24.563	-7.000	0.00781478
31	Undetermine	20.109		20.109	-2.399	0.18956992	31	20.430	20.349		20.390	-2.827	0.14094835
34	21.828	21.850		21.839	-4.129	0.05715385	34	22.478	22.297		22.387	-4.824	0.03529646
35	23.862	23.848		23.855	-6.145	0.0141317	35	24.327	24.445		24.386	-6.823	0.00883222
36	23.256	23.138		23.197	-5.487	0.02229574	36	23.650	23.614		23.632	-6.069	0.01489585
37	23.292	23.261		23.277	-5.567	0.02110038	37	23.000	22.401		22.701	-5.138	0.02840545
38	22.621	22.426		22.523	-4.813	0.03556727	38	23.170	23.280		23.225	-5.662	0.01974455
39	22.724	22.676		22.700	-4.990	0.03147085	39	23.288	22.935		23.112	-5.549	0.02136622
40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!	40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
41	25.294	25.589		25.441	-7.731	0.00470583	41	24.969	25.274		25.121	-7.558	0.00530608
43	22.558	22.335		22.446	-4.736	0.03751547	43	22.584	23.365		22.975	-5.412	0.02349225
44	23.657	23.452		23.554	-5.844	0.01740561	44	24.506	24.916		24.711	-7.148	0.00705053
45	20.940	20.891		20.915	-3.205	0.10840961	45	21.958	21.729		21.843	-4.280	0.05146778
46	22.380	22.279		22.330	-4.620	0.04068035	46	22.778	23.006		22.892	-5.329	0.02487731
47	27.690	27.029		27.359	-9.649	0.00124514	47	27.701	28.960		28.331	-10.768	0.00057364
49	21.913	21.800		21.857	-4.147	0.05645333	49	22.364	22.477		22.421	-4.858	0.03448266
50	27.335	27.538		27.437	-9.727	0.00118035	50	29.654	28.757		29.205	-11.642	0.00031285
51	22.441	22.355		22.398	-4.688	0.038802	51	22.628	23.247		22.938	-5.375	0.02410262
52	23.190	23.167		23.179	-5.469	0.02258446	52	24.745	24.385		24.565	-7.002	0.00780219
53	21.906	22.015		21.961	-4.251	0.05253729	53	22.117	21.916		22.017	-4.454	0.04563237
54	16.750	16.673		16.712	0.998	1.99760592	54	17.800	17.684		17.742	-0.179	0.8832174
55	23.649	23.744		23.696	-5.986	0.01577337	55	24.258	23.962		24.110	-6.547	0.01069274
56	23.541	23.492		23.516	-5.806	0.01786817	56	24.163	23.663		23.913	-6.350	0.01225874
57	22.415	22.422		22.418	-4.708	0.03824785	57	22.472	22.732		22.602	-5.039	0.03040918
58	23.502	23.431		23.467	-5.757	0.01849414	58	22.758	22.798		22.778	-5.215	0.02692385
var2csa	14.718	14.715		14.716	2.994	7.96493175	var2csa	15.762	15.649		15.705	1.858	3.62439451
91	21.546	21.617		21.581	-3.871	0.06832318	91	21.327	21.670		21.499	-3.936	0.06535718
92	21.267	21.245		21.256	-3.546	0.08559495	92	21.512	21.318		21.415	-3.852	0.06923863
93	22.133	22.267		22.200	-4.490	0.0444939	93	22.377	22.393		22.385	-4.822	0.03535516
94	24.332	24.246		24.289	-6.579	0.01046112	94	25.201	25.339		25.270	-7.707	0.0047845
95	24.179	24.102		24.140	-6.430	0.01159418	95	23.863	23.856		23.859	-6.296	0.01272238
96	24.535	24.405		24.470	-6.760	0.00922564	96	24.589	24.505		24.547	-6.984	0.00789888
97	24.549	24.651		24.600	-6.890	0.00843179	97	24.795	24.520		24.658	-7.095	0.00731669
60	17.739	17.681		17.710	0.000	0.99982084	60	17.440	17.685		17.563	0.000	1.00020034

(Rep 2) MCM 4Wk					(Rep 2) -Met 4Wk				
Primer pair	CT		relative to p60	realtive copy number	Primer pair	CT		relative to p60	realtive copy number
1	20.393		-2.664	0.15776149	1	20.373		-2.820	0.14161993
3	Undetermined		#VALUE!	#VALUE!	3	22.375		-4.822	0.035364
4	21.543		-3.814	0.07111097	4	21.846		-4.293	0.05099918
5	Undetermined		#VALUE!	#VALUE!	5	21.942		-4.389	0.04773317
6	21.714		-3.985	0.06314175	6	21.240		-3.687	0.07763194
7	Undetermined		#VALUE!	#VALUE!	7	20.680		-3.127	0.11443234
8	23.228		-5.499	0.02211216	8	21.410		-3.857	0.06901499
9	21.131		-3.402	0.09459298	9	20.939		-3.386	0.09565699
11	23.225		-5.496	0.02216254	11	22.896		-5.343	0.02464275
12	20.850		-3.121	0.11490544	12	20.932		-3.379	0.09611043
13	19.921		-2.192	0.21880051	13	19.691		-2.138	0.22726062
15	21.347		-3.618	0.08142259	15	21.790		-4.237	0.05302154
17	20.257		-2.528	0.17342202	17	20.004		-2.451	0.18292166
18	Undetermined		#VALUE!	#VALUE!	18	19.594		-2.041	0.24304154
19	Undetermined		#VALUE!	#VALUE!	19	20.224		-2.671	0.15706794
20	24.095		-6.366	0.01212594	20	23.895		-6.342	0.01233106
21	21.405		-3.676	0.07821657	21	21.589		-4.036	0.06096783
22	20.816		-3.087	0.11768774	22	20.811		-3.258	0.10453931
23	22.262		-4.533	0.04318084	23	22.008		-4.455	0.04559935
25	20.758		-3.029	0.12248509	25	20.920		-3.367	0.09694926
26	Undetermined		#VALUE!	#VALUE!	26	20.955		-3.402	0.09462907
27	19.727		-1.998	0.2503456	27	19.596		-2.043	0.24259788
28	22.987		-5.258	0.02613565	28	22.393		-4.840	0.034906
30	Undetermined		#VALUE!	#VALUE!	30	22.437		-4.884	0.03387471
31	18.435		-0.706	0.61304845	31	18.296		-0.743	0.59732994
34	19.898		-2.169	0.22243643	34	20.421		-2.868	0.13693596
35	23.340		-5.611	0.02045663	35	22.497		-4.944	0.03247802
36	21.919		-4.190	0.0548021	36	21.867		-4.314	0.05026985
37	21.684		-3.955	0.06448489	37	22.753		-5.200	0.02721103
38	21.224		-3.495	0.08868955	38	21.463		-3.910	0.06650259
39	21.767		-4.038	0.06088117	39	22.343		-4.790	0.03615319
40	Undetermined		#VALUE!	#VALUE!	40	37.064		-19.511	1.3384E-06
41	24.677		-6.948	0.00809927	41	24.130		-6.577	0.01047442
43	21.193		-3.464	0.09065221	43	21.497		-3.944	0.06495835
44	22.348		-4.619	0.04069005	44	22.111		-4.558	0.04245286
45	20.229		-2.500	0.17679606	45	19.592		-2.039	0.24339042
46	21.254		-3.525	0.08686026	46	21.473		-3.920	0.0660861
47	27.981		-10.252	0.00081995	47	26.753		-9.200	0.00170062
49	19.978		-2.249	0.21037002	49	19.535		-1.982	0.25320634
50	27.224		-9.495	0.00138564	50	26.855		-9.302	0.00158457
51	21.741		-4.012	0.06197422	51	21.359		-3.806	0.07150518
52	22.957		-5.228	0.02667481	52	22.517		-4.964	0.03203286
53	21.241		-3.512	0.08762589	53	20.341		-2.788	0.1447365
54	15.173		2.556	5.88198049	54	15.203		2.350	5.09943206
55	22.770		-5.041	0.03038367	55	21.998		-4.445	0.04592144
56	22.325		-4.596	0.04133936	56	21.917		-4.364	0.04854678
57	21.959		-4.230	0.05328384	57	25.492		-7.939	0.00407417
58	0.000		0.000	0	58	22.213		-4.660	0.03954424
var2csa	17.337		0.392	1.3119566	var2csa	17.338		0.215	1.16062005
91	19.938		-2.209	0.21621642	91	20.290		-2.737	0.15001477
92	20.478		-2.749	0.14879129	92	20.466		-2.913	0.13279416
93	20.476		-2.747	0.1489468	93	21.183		-3.630	0.0807795
94	23.938		-6.209	0.01351386	94	23.171		-5.618	0.02036642
95	22.476		-4.747	0.03724752	95	21.687		-4.134	0.05697153
96	24.120		-6.391	0.01191285	96	22.837		-5.284	0.02565921
97	23.919		-6.190	0.01370015	97	23.492		-5.939	0.01629484
60	17.729		0.000	1.00030642	60	17.553		0.000	0.99995937

(Rep 2) -ChoSer 4Wk					(Rep 2,1) 20xMet 4Wk				
Primer pair	CT		relative to p60	realtive copy number	Primer pair	CT		relative to p60	realtive copy number
1	23.569		-4.227	0.05341127	1	20.639		-2.965	0.12804235
3	26.001		-6.659	0.00989553	3	23.420		-5.746	0.01863923
4	Undetermined		#VALUE!	#VALUE!	4	22.400		-4.726	0.03778373
5	24.803		-5.461	0.02269857	5	22.654		-4.980	0.03168228
6	23.942		-4.600	0.04123548	6	21.776		-4.102	0.05821412
7	23.378		-4.036	0.06097954	7	21.880		-4.206	0.05419149
8	24.481		-5.139	0.02838633	8	22.534		-4.860	0.03444509
9	24.501		-5.159	0.02798018	9	21.482		-3.808	0.07141322
11	26.523		-7.181	0.00689245	11	24.121		-6.447	0.01145876
12	23.327		-3.985	0.06314687	12	22.202		-4.528	0.0433327
13	22.525		-3.183	0.1100844	13	Undetermined		#VALUE!	#VALUE!
15	24.158		-4.816	0.03549422	15	22.425		-4.751	0.03714248
17	Undetermined		#VALUE!	#VALUE!	17	20.317		-2.643	0.16010663
18	22.783		-3.441	0.09204895	18	20.375		-2.701	0.15374048
19	23.508		-4.166	0.05569602	19	20.844		-3.170	0.11114339
20	26.761		-7.419	0.00584474	20	24.293		-6.619	0.01017378
21	24.564		-5.222	0.02679638	21	21.657		-3.983	0.06323407
22	24.220		-4.878	0.03401512	22	21.493		-3.819	0.07085483
23	26.879		-7.537	0.00538277	23	22.885		-5.211	0.0270063
25	24.517		-5.175	0.02768509	25	21.278		-3.604	0.08223264
26	24.946		-5.604	0.02055795	26	21.948		-4.274	0.05168262
27	22.776		-3.434	0.09252224	27	20.405		-2.731	0.15063699
28	24.860		-5.518	0.02182053	28	22.730		-5.056	0.03006403
30	Undetermined		#VALUE!	#VALUE!	30	22.644		-4.970	0.03190614
31	21.806		-2.464	0.18126182	31	18.965		-1.291	0.40854814
34	23.640		-4.298	0.0508498	34	20.750		-3.076	0.1185839
35	24.494		-5.152	0.02812775	35	23.347		-5.673	0.01960628
36	24.774		-5.432	0.02316376	36	22.400		-4.726	0.03778271
37	24.285		-4.943	0.03249933	37	22.815		-5.141	0.02834541
38	23.713		-4.371	0.04834254	38	21.739		-4.065	0.05976232
39	24.430		-5.088	0.02940108	39	22.379		-4.705	0.0383325
40	Undetermined		#VALUE!	#VALUE!	40	Undetermined		#VALUE!	#VALUE!
41	26.260		-6.918	0.00827178	41	24.387		-6.713	0.00953336
43	24.158		-4.816	0.03549705	43	22.026		-4.352	0.0489662
44	25.673		-6.331	0.01242552	44	22.686		-5.012	0.03098625
45	23.217		-3.875	0.0681573	45	20.791		-3.117	0.11527839
46	24.374		-5.032	0.03057073	46	22.014		-4.340	0.04936116
47	Undetermined		#VALUE!	#VALUE!	47	26.899		-9.225	0.00167056
49	34.694		-15.352	2.3908E-05	49	20.551		-2.877	0.13613218
50	29.858		-10.516	0.00068277	50	27.407		-9.733	0.00117481
51	24.230		-4.888	0.03377915	51	21.403		-3.729	0.07539988
52	25.998		-6.656	0.00991876	52	23.153		-5.479	0.02242461
53	22.644		-3.302	0.10138773	53	20.717		-3.043	0.12130627
54	18.588		0.754	1.68623546	54	15.815		1.859	3.62845153
55	25.388		-6.046	0.01513797	55	22.705		-5.031	0.03058233
56	24.883		-5.541	0.02148282	56	22.535		-4.861	0.03440796
57	22.998		-3.656	0.07933124	57	21.884		-4.210	0.05401512
58	23.970		-4.628	0.04043272	58	22.189		-4.515	0.04373797
var2csa	20.575893		-1.234	0.42516861	var2csa	15.360		2.314	4.97295026
91	23.159		-3.817	0.07092927	91	20.829		-3.155	0.11226491
92	23.253		-3.911	0.06645585	92	21.452		-3.778	0.0729029
93	24.329		-4.987	0.03153389	93	21.434		-3.760	0.07379987
94	25.862		-6.520	0.01089409	94	24.046		-6.372	0.01207756
95	25.425		-6.083	0.01475359	95	23.426		-5.752	0.01855655
96	25.779		-6.437	0.01154421	96	23.692		-6.018	0.01542677
97	24.796		-5.454	0.02281044	97	24.139		-6.465	0.01132212
60	19.342		0.000	1.00016082	60	17.674		0.000	0.99968952

(Rep 2,2) 20xMet 4Wk					(Rep 2,3) 20xMet 4Wk				
Primer pair	CT		relative to p60	realtive copy number	Primer pair	CT		relative to p60	realtive copy number
1	21.172		-3.169	0.11120966	1	21.442		-3.191	0.10952529
3	22.833		-4.830	0.03514836	3	23.716		-5.465	0.02264317
4	22.745		-4.742	0.03736901	4	22.455		-4.204	0.0542617
5	22.726		-4.723	0.03787527	5	22.882		-4.631	0.04035049
6	22.284		-4.281	0.05145011	6	21.910		-3.659	0.07913809
7	21.832		-3.829	0.07035272	7	21.708		-3.457	0.0910372
8	22.846		-4.843	0.03484332	8	22.558		-4.307	0.05050822
9	21.869		-3.866	0.06859504	9	21.846		-3.595	0.08277764
11	23.989		-5.986	0.01577762	11	24.155		-5.904	0.01670402
12	21.642		-3.639	0.08025933	12	21.437		-3.186	0.10987088
13	20.189		-2.186	0.2197008	13	20.622		-2.371	0.19335353
15	21.812		-3.809	0.07134104	15	22.128		-3.877	0.06806656
17	20.602		-2.599	0.16502128	17	20.803		-2.552	0.17050075
18	20.555		-2.552	0.17054259	18	20.292		-2.041	0.24296864
19	21.498		-3.495	0.08871612	19	21.382		-3.131	0.11411932
20	24.631		-6.628	0.01010792	20	24.944		-6.693	0.00966498
21	22.380		-4.377	0.04813059	21	22.511		-4.260	0.05219426
22	21.626		-3.623	0.08115614	22	21.516		-3.265	0.1040464
23	22.740		-4.737	0.03750846	23	22.783		-4.532	0.04323445
25	21.989		-3.986	0.06309926	25	21.953		-3.702	0.07682875
26	22.452		-4.449	0.04577685	26	21.485		-3.234	0.10629687
27	20.509		-2.506	0.17600545	27	20.008		-1.757	0.29578894
28	22.900		-4.897	0.03356124	28	22.776		-4.525	0.0434472
30	23.308		-5.305	0.02529814	30	23.387		-5.136	0.02844715
31	18.955		-0.952	0.51699348	31	19.440		-1.189	0.43855449
34	20.897		-2.894	0.13450093	34	21.389		-3.138	0.11362176
35	Undetermined		#VALUE!	#VALUE!	35	22.997		-4.746	0.037269
36	22.441		-4.438	0.0461252	36	22.834		-4.583	0.04171827
37	22.862		-4.859	0.03445569	37	23.159		-4.908	0.03330268
38	22.144		-4.141	0.05667837	38	21.932		-3.681	0.07798346
39	22.817		-4.814	0.03553853	39	22.467		-4.216	0.053794
40	31.530		-13.527	8.4735E-05	40	Undetermined		#VALUE!	#VALUE!
41	24.863		-6.860	0.00861154	41	24.894		-6.643	0.01000831
43	22.273		-4.270	0.05181896	43	22.148		-3.897	0.06713068
44	23.380		-5.377	0.02407169	44	23.486		-5.235	0.02656024
45	20.210		-2.207	0.21663106	45	20.334		-2.083	0.23598908
46	22.443		-4.440	0.04608136	46	21.702		-3.451	0.0914398
47	28.421		-10.418	0.0007311	47	28.343		-10.092	0.00091641
49	20.767		-2.764	0.14723964	49	20.262		-2.011	0.24810281
50	27.834		-9.831	0.00109805	50	27.846		-9.595	0.00129283
51	21.949		-3.946	0.06489585	51	22.265		-4.014	0.06190329
52	23.405		-5.402	0.02365154	52	23.493		-5.242	0.02642424
53	20.668		-2.665	0.15764977	53	20.652		-2.401	0.18927687
54	16.115		1.888	3.70102274	54	15.766		2.485	5.6002491
55	22.752		-4.749	0.0371954	55	22.913		-4.662	0.03951012
56	23.296		-5.293	0.02550834	56	22.680		-4.429	0.04642179
57	22.472		-4.469	0.04516512	57	22.252		-4.001	0.06247271
58	22.627		-4.624	0.04055726	58	22.191		-3.940	0.06515117
var2csa	15.942		2.061	4.17158604	var2csa	15.215		3.036	8.20421898
91	21.260		-3.257	0.10460937	91	20.795		-2.544	0.17149364
92	21.177		-3.174	0.11079272	92	20.701		-2.450	0.18301198
93	24.642		-6.639	0.01003126	93	21.589		-3.338	0.09886584
94	24.483		-6.480	0.01120398	94	23.858		-5.607	0.02051282
95	22.283		-4.280	0.05149128	95	22.689		-4.438	0.04613483
96	23.872		-5.869	0.01711506	96	23.804		-5.553	0.02129443
97	32.080		-14.077	5.7871E-05	97	24.319		-6.068	0.01490154
60	18.003		0.000	1.00030018	60	18.251		0.000	0.99982673

(Rep 2,1) 20xSer 4Wk					(Rep 2,2) 20xSer 4Wk				
Primer pair	CT		relative to p60	realtive copy number	Primer pair	CT		relative to p60	realtive copy number
1	20.932		-3.699	0.07698963	1	19.958		-3.561	0.08470559
3	23.256		-6.023	0.01537553	3	Undetermined		#VALUE!	#VALUE!
4	21.721		-4.488	0.04457514	4	20.984		-4.587	0.0415997
5	22.515		-5.282	0.02570488	5	21.903		-5.506	0.0220032
6	21.501		-4.268	0.05190761	6	21.226		-4.829	0.03517665
7	21.316		-4.083	0.05901265	7	20.683		-4.286	0.05127083
8	21.915		-4.682	0.03894711	8	21.694		-5.297	0.02544151
9	21.463		-4.230	0.05327986	9	20.645		-4.248	0.0526285
11	23.819		-6.586	0.01041131	11	23.429		-7.032	0.00764313
12	21.346		-4.113	0.05780948	12	20.589		-4.192	0.05472106
13	19.982		-2.749	0.14877871	13	19.523		-3.126	0.11458467
15	21.593		-4.360	0.04870575	15	21.326		-4.929	0.03282746
17	20.258		-3.025	0.12281707	17	19.958		-3.561	0.08473683
18	20.264		-3.031	0.12236603	18	19.596		-3.199	0.10889307
19	20.610		-3.377	0.09624597	19	19.825		-3.428	0.09291917
20	23.526		-6.293	0.01274882	20	22.501		-6.104	0.01453674
21	21.486		-4.253	0.05246474	21	20.992		-4.595	0.04137508
22	21.223		-3.990	0.0629301	22	20.686		-4.289	0.05115869
23	22.505		-5.272	0.02588123	23	21.549		-5.152	0.02811654
25	20.722		-3.489	0.08906812	25	20.130		-3.733	0.07522656
26	Undetermined		#VALUE!	#VALUE!	26	21.026		-4.629	0.04041134
27	19.657		-2.424	0.1863233	27	19.572		-3.175	0.11071242
28	22.805		-5.572	0.02102263	28	21.965		-5.568	0.02107923
30	22.615		-5.382	0.02398772	30	22.141		-5.744	0.01865983
31	18.567		-1.334	0.39659324	31	17.986		-1.589	0.33236883
34	20.497		-3.264	0.10407194	34	19.791		-3.394	0.09510797
35	22.716		-5.483	0.02236138	35	21.726		-5.329	0.02486965
36	22.364		-5.131	0.02853958	36	21.916		-5.519	0.02180556
37	22.695		-5.462	0.02268172	37	Undetermined		#VALUE!	#VALUE!
38	21.287		-4.054	0.06022407	38	21.462		-5.065	0.02986745
39	21.664		-4.431	0.0463499	39	21.368		-4.971	0.03189288
40	Undetermined		#VALUE!	#VALUE!	40	Undetermined		#VALUE!	#VALUE!
41	23.833		-6.600	0.01030972	41	23.255		-6.858	0.00862004
43	21.752		-4.519	0.04362781	43	21.433		-5.036	0.03047939
44	22.793		-5.560	0.0211897	44	22.576		-6.179	0.01380417
45	19.936		-2.703	0.15358572	45	19.469		-3.072	0.11895081
46	21.606		-4.373	0.0482564	46	20.931		-4.534	0.0431747
47	26.471		-9.238	0.00165557	47	26.292		-9.895	0.00105042
49	19.596		-2.363	0.19432858	49	19.279		-2.882	0.13562716
50	26.989		-9.756	0.00115616	50	26.848		-10.451	0.00071421
51	21.379		-4.146	0.05648592	51	20.611		-4.214	0.05388711
52	22.605		-5.372	0.02414315	52	22.167		-5.770	0.01832123
53	20.455		-3.222	0.10718241	53	19.698		-3.301	0.10142688
54	15.248		1.985	3.959771	54	14.933		1.464	2.75898765
55	22.137		-4.904	0.03340463	55	21.354		-4.957	0.03218986
56	22.636		-5.403	0.02362629	56	21.602		-5.205	0.02711575
57	21.709		-4.476	0.04494433	57	21.358		-4.961	0.03211373
58	22.461		-5.228	0.02667284	58	21.643		-5.246	0.02635435
var2csa	16.434		0.799	1.74034098	var2csa	16.278		#REF!	#REF!
91	20.742		-3.509	0.08786467	91	19.893		-3.496	0.08864328
92	Undetermined		#VALUE!	#VALUE!	92	20.175		-3.778	0.07290709
93	21.449		-4.216	0.05382178	93	20.957		-4.560	0.04240755
94	Undetermined		#VALUE!	#VALUE!	94	23.461		-7.064	0.00747401
95	21.744		-4.511	0.04385873	95	Undetermined		#VALUE!	#VALUE!
96	30.441		-13.208	0.00010571	96	23.244		-6.847	0.00868674
97	24.117		-6.884	0.00846491	97	23.598		-7.201	0.00679541
60	17.233		0.000	1.00000416	60	16.937		-0.540	0.68778426

(Rep 2) 20xCho 4Wk					(Rep 2) 1xCho 4Wk				
Primer pair	CT		relative to p60	realtive copy number	Primer pair	CT		relative to p60	realtive copy number
1	22.107		-4.620	0.04066138	1	22.872		-4.551	0.04266453
3	23.649		-6.162	0.01396285	3	24.625		-6.304	0.01265232
4	23.364		-5.877	0.01701429	4	23.131		-4.810	0.03564027
5	23.361		-5.874	0.01705557	5	24.148		-5.827	0.01762042
6	22.414		-4.927	0.03286603	6	22.896		-4.575	0.04194856
7	22.120		-4.633	0.04030252	7	22.998		-4.677	0.03909733
8	22.781		-5.294	0.02547998	8	23.736		-5.415	0.02343281
9	22.251		-4.764	0.03679611	9	22.713		-4.392	0.04763085
11	24.308		-6.821	0.00884219	11	24.687		-6.366	0.01212292
12	21.836		-4.349	0.04907255	12	22.782		-4.461	0.04539027
13	21.364		-3.877	0.06804722	13	21.624		-3.303	0.10134635
15	22.485		-4.998	0.03128717	15	22.513		-4.192	0.05472299
17	21.380		-3.893	0.06731505	17	21.905		-3.584	0.0834086
18	21.507		-4.020	0.06163117	18	21.805		-3.484	0.08939364
19	21.599		-4.112	0.05781309	19	22.552		-4.231	0.05326043
20	24.908		-7.421	0.00583509	20	26.372		-8.051	0.00377081
21	24.737		-7.250	0.00656815	21	23.729		-5.408	0.02354719
22	22.715		-5.228	0.02668212	22	23.170		-4.849	0.03469294
23	23.474		-5.987	0.01576902	23	24.382		-6.061	0.01497439
25	22.308		-4.821	0.03536755	25	22.839		-4.518	0.04364605
26	22.965		-5.478	0.02242925	26	23.456		-5.135	0.02845039
27	20.440		-2.953	0.12918281	27	21.253		-2.932	0.1310013
28	22.913		-5.426	0.02325273	28	23.674		-5.353	0.02446839
30	23.463		-5.976	0.01588261	30	21.301		-2.980	0.1267047
31	20.188		-2.701	0.1537456	31	20.552		-2.231	0.21307349
34	21.811		-4.324	0.04994444	34	21.742		-3.421	0.09335933
35	23.655		-6.168	0.01390862	35	24.210		-5.889	0.0168715
36	23.009		-5.522	0.02176549	36	23.870		-5.549	0.02136106
37	21.806		-4.319	0.05011245	37	23.153		-4.832	0.03511602
38	22.649		-5.162	0.0279334	38	23.468		-5.147	0.02822662
39	Undetermined		#VALUE!	#VALUE!	39	23.579		-5.258	0.02613958
40	Undetermined		#VALUE!	#VALUE!	40	32.755		-14.434	4.5184E-05
41	23.924		-6.437	0.01154298	41	25.359		-7.038	0.00760817
43	22.638		-5.151	0.02814429	43	23.505		-5.184	0.02750226
44	23.566		-6.079	0.01479676	44	23.888		-5.567	0.02109503
45	20.371		-2.884	0.13544838	45	21.485		-3.164	0.11159946
46	22.814		-5.327	0.02490372	46	Undetermined		#VALUE!	#VALUE!
47	27.658		-10.171	0.00086721	47	27.962		-9.641	0.00125214
49	21.591		-4.104	0.05816165	49	21.991		-3.670	0.07855462
50	28.594		-11.107	0.00045349	50	28.602		-10.281	0.00080398
51	22.345		-4.858	0.03447712	51	23.550		-5.229	0.02667015
52	23.997		-6.510	0.0109728	52	24.726		-6.405	0.01180205
53	21.914		-4.427	0.04648777	53	22.519		-4.198	0.05447375
54	17.444		0.043	1.03016255	54	17.929		0.392	1.31239955
55	23.313		-5.826	0.0176243	55	23.423		-5.102	0.02912662
56	23.189		-5.702	0.01921238	56	24.423		-6.102	0.01455661
57	22.251		-4.764	0.03680807	57	22.991		-4.670	0.03927453
58	23.749		-6.262	0.01303299	58	24.207		-5.886	0.01691283
var2csa	12.593		4.894	29.7259498	var2csa	14.884		3.437	10.829776
91	21.746		-4.259	0.05221681	91	22.189		-3.868	0.06849559
92	21.323		-3.836	0.07001268	92	22.363		-4.042	0.06072267
93	22.744		-5.257	0.02614734	93	23.112		-4.791	0.03611265
94	23.761		-6.274	0.01292659	94	24.931		-6.610	0.01023616
95	23.929		-6.442	0.01150428	95	23.674		-5.353	0.02446654
96	24.259		-6.772	0.00915321	96	25.375		-7.054	0.00752809
97	25.151		-7.664	0.00493165	97	25.531		-7.210	0.00675188
60	17.487		0.000	1.00001594	60	18.321		0.000	1.00027175

Chapter 3.2 *var* panels (on/off ATc for 2 Weeks)

Parent Culture for OE lines						
Primer pair	CT			CT Average	relative to p60	relative copy number
1	26.931082	27.131638		27.03136	-6.51636	0.01092396
3	27.510624	27.879713		27.6951685	-7.1801685	0.00689531
4	27.560629	27.800638		27.6806335	-7.1656335	0.00696513
5	24.205105	24.413015		24.30906	-3.79406	0.07208985
6	26.082508	26.230368		26.156438	-5.641438	0.02003355
7	26.815828	26.880657		26.8482425	-6.3332425	0.01240235
8	26.434206	26.432083		26.4331445	-5.9181445	0.01653716
9	26.268742	25.922853		26.0957975	-5.5807975	0.02089357
11	25.83665	26.418888		26.127769	-5.612769	0.02043564
12	24.637186	26.362179		25.4996825	-4.9846825	0.03158356
13	24.951578	23.802095		24.3768365	-3.8618365	0.06878146
15	22.343124	25.375061		23.8590925	-3.3440925	0.09847542
17	25.62259	23.417473		24.5200315	-4.0050315	0.06228241
18	24.840776	25.254776		25.047776	-4.532776	0.04320146
19	24.264257	28.266968		26.2656125	-5.7506125	0.01857347
21	28.871113	28.48493		28.6780215	-8.1630215	0.00348888
21	27.004606	26.073793		26.5391995	-6.0241995	0.0153651
22	25.968174	25.684132		25.826153	-5.311153	0.02518742
23	28.77616	27.719587		28.2478735	-7.7328735	0.00470081
25	29.149841	28.3779		28.7638705	-8.2488705	0.00328732
26	25.851505	25.717823		25.784664	-5.269664	0.02592227
27	23.959616	23.850058		23.904837	-3.389837	0.09540198
28	27.081238	27.362242		27.22174	-6.70674	0.00957349
30	26.987844	27.575926		27.281885	-6.766885	0.00918258
31	22.62391	22.808481		22.7161955	-2.2011955	0.21745737
34	24.103598	24.13716		24.120379	-3.605379	0.08216234
35	27.194693	26.837557		27.016125	-6.501125	0.01103993
36	27.406343	27.00846		27.2074015	-6.6924015	0.00966911
37	24.014599	25.177689		24.596144	-4.081144	0.05908173
38	26.173822	25.88639		26.030106	-5.515106	0.02186692
39	26.6514	26.759901		26.7056505	-6.1906505	0.01369079
40	Undetermine	Undetermined		0	0	0
41	27.123764	27.194231		27.1589975	-6.6439975	0.00999902
43	26.724064	26.86718		26.795622	-6.280622	0.01286306
44	26.446993	28.147367		27.29718	-6.78218	0.00908574
45	25.246407	25.218082		25.2322445	-4.7172445	0.03801613
46	27.068705	27.15929		27.1139975	-6.5989975	0.01031582
47	31.080566	31.815107		31.4478365	-10.932837	0.00051155
49	25.966726	25.687374		25.82705	-5.31205	0.02517176
50	26.983055	26.909843		26.946449	-6.431449	0.0115862
51	26.648962	26.747534		26.698248	-6.183248	0.01376122
52	27.83372	27.767368		27.800544	-7.285544	0.00640963
53	26.463495	26.476458		26.4699765	-5.9549765	0.01612031
54	18.138374	18.18982		18.164097	2.350903	5.10143456
55	26.971014	27.49315		27.232082	-6.717082	0.0095051
56	25.385275	24.950813		25.168044	-4.653044	0.03974607
57	25.387789	25.965584		25.6766865	-5.1616865	0.02793686
58	27.214611	27.662815		27.438713	-6.923713	0.00823673
var2csa	24.149904	24.593025		24.3714645	-3.8564645	0.06903805
91	25.859835	26.000244		25.9300395	-5.4150395	0.02343747
92	25.170557	25.857357		25.513957	-4.998957	0.0312726
93	26.850527	27.442804		27.1466655	-6.6316655	0.01008486
94	27.854473	28.189404		28.0219385	-7.5069385	0.00549777
95	26.724829	26.950666		26.8377475	-6.3227475	0.0124929
96	28.231144	28.321583		28.2763635	-7.7613635	0.00460889
97	29.68523	29.903013		29.7941215	-9.2791215	0.00160956
60	20.304968	20.724493		20.5147305		

PfSAMS OE Day 0						
Primer pair	CT			CT Average	relative to p60	relative copy number
1	22.917	22.936		22.927	-5.263	0.02604995
3	24.571	24.691		24.631	-6.967	0.00799278
4	23.466	23.627		23.547	-5.883	0.01695008
5	23.908	23.963		23.935	-6.271	0.01294688
6	22.721	22.823		22.772	-5.108	0.0289993
7	21.787	21.755		21.771	-4.107	0.05802317
8	30.173	31.596		30.885	-13.221	0.00010476
9	23.007	22.982		22.994	-5.330	0.02485449
11	24.319	24.480		24.400	-6.736	0.00938431
12	22.934	23.170		23.052	-5.388	0.02387746
13	21.585	21.589		21.587	-3.923	0.06594088
15	23.762	23.887		23.825	-6.161	0.01397762
17	22.234	22.280		22.257	-4.593	0.04144184
18	22.148	22.249		22.198	-4.534	0.04316046
19	22.854	22.920		22.887	-5.223	0.0267752
20	23.762	23.521		23.641	-5.977	0.01587267
21	23.563	24.337		23.950	-6.286	0.01281287
22	22.883	22.527		22.705	-5.041	0.03037314
23	24.418	24.240		24.329	-6.665	0.00985556
25	23.982	23.667		23.825	-6.161	0.01397824
26	23.870	23.810		23.840	-6.176	0.01382956
27	20.859	21.148		21.004	-3.340	0.09878837
28	23.683	24.266		23.975	-6.311	0.01259812
30	24.228	24.467		24.348	-6.684	0.00972708
31	20.841	20.876		20.859	-3.195	0.10921709
34	23.395	23.533		23.464	-5.800	0.01794801
35	22.939	22.658		22.798	-5.134	0.02846949
36	23.711	23.716		23.714	-6.050	0.01509599
37	23.431	23.735		23.583	-5.919	0.01652659
38	23.510	23.513		23.512	-5.848	0.01736418
39	22.808	22.956		22.882	-5.218	0.02686677
40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
41	24.743	24.722		24.732	-7.068	0.00745133
43	22.868	22.967		22.918	-5.254	0.02621349
44	24.513	24.326		24.420	-6.756	0.00925429
45	23.236	23.192		23.214	-5.550	0.02134131
46	23.535	23.376		23.456	-5.792	0.01805379
47	28.304	28.551		28.427	-10.763	0.00057542
49	23.543	23.706		23.625	-5.961	0.01605707
50	28.683	29.504		29.093	-11.429	0.00036256
51	22.406	22.532		22.469	-4.805	0.03577626
52	25.255	26.072		25.663	-7.999	0.00390882
53	22.378	22.415		22.396	-4.732	0.03762092
54	21.619	21.947		21.783	-4.119	0.05754393
55	24.295	24.453		24.374	-6.710	0.00955266
56	23.855	23.759		23.807	-6.143	0.01415104
57	21.814	21.663		21.738	-4.074	0.05937163
58	23.601	23.379		23.490	-5.826	0.0176283
var2csa	13.917	13.950		13.933	3.731	13.2746968
91	22.618	22.900		22.759	-5.095	0.02926271
92	21.432	21.408		21.420	-3.756	0.07401664
93	22.542	22.562		22.552	-4.888	0.03377452
94	24.446	25.338		24.892	-7.228	0.00667027
95	24.235	23.982		24.108	-6.444	0.01148275
96	24.532	24.678		24.605	-6.941	0.00813957
97	23.762	23.853		23.808	-6.144	0.0141449
60	17.437	17.891		17.664	0.000	1.00021109

PfSAMS OE OFF ATc							PfSAMS OE ON ATc						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	24.534	24.283		24.409	-6.293	0.01275669	1	22.538	22.739		22.638	-5.705	0.01916413
3	25.327	25.276		25.301	-7.185	0.00687143	3	23.805	23.711		23.758	-6.825	0.00881887
4	25.185	25.311		25.248	-7.132	0.00713038	4	23.611	23.144		23.377	-6.444	0.01148241
5	25.346	25.184		25.265	-7.149	0.00704543	5	23.352	23.289		23.321	-6.388	0.0119419
6	24.358	24.229		24.294	-6.178	0.01381605	6	22.998	22.994		22.996	-6.063	0.01495714
7	22.825	22.785		22.805	-4.689	0.0387738	7	21.398	21.319		21.358	-4.425	0.04654092
8	32.029	32.077		32.053	-13.937	6.3762E-05	8	29.430	30.439		29.934	-13.001	0.00012196
9	24.510	24.380		24.445	-6.329	0.01243937	9	22.512	22.438		22.475	-5.542	0.02146631
11	25.567	25.652		25.610	-7.494	0.00554878	11	23.979	23.812		23.896	-6.963	0.00801575
12	24.574	24.470		24.522	-6.406	0.01179286	12	22.259	22.402		22.331	-5.398	0.02372356
13	22.808	22.764		22.786	-4.670	0.03928357	13	20.806	21.126		20.966	-4.033	0.06106961
15	25.490	25.344		25.417	-7.301	0.0063411	15	23.917	23.702		23.809	-6.876	0.00851172
17	23.726	23.695		23.710	-5.594	0.02070174	17	21.889	21.957		21.923	-4.990	0.03146872
18	23.552	23.433		23.493	-5.377	0.02406459	18	21.921	21.987		21.954	-5.021	0.03079905
19	24.188	23.929		24.058	-5.942	0.01626066	19	21.900	22.507		22.204	-5.271	0.02589953
20	25.286	24.802		25.044	-6.928	0.00821166	20	24.962	24.525		24.743	-7.810	0.00445513
21	24.497	24.503		24.500	-6.384	0.01197349	21	23.184	22.884		23.034	-6.101	0.01456672
22	23.691	23.859		23.775	-5.659	0.01978845	22	22.280	22.157		22.219	-5.286	0.02563696
23	25.514	25.431		25.473	-7.357	0.0061017	23	23.730	23.356		23.543	-6.610	0.01023691
25	24.822	24.788		24.805	-6.689	0.00969296	25	23.684	23.439		23.561	-6.628	0.01010798
26	24.775	24.558		24.667	-6.551	0.01066577	26	23.210	23.218		23.214	-6.281	0.01286184
27	21.856	21.779		21.817	-3.701	0.07687664	27	20.389	20.409		20.399	-3.466	0.0905021
28	24.123	25.505		24.814	-6.698	0.0096298	28	23.963	23.968		23.966	-7.033	0.00763826
30	25.607	25.783		25.695	-7.579	0.00522961	30	24.296	24.221		24.259	-7.326	0.00623254
31	21.891	21.872		21.881	-3.765	0.07354028	31	20.663	20.725		20.694	-3.761	0.07376159
34	24.654	24.482		24.568	-6.452	0.01141962	34	23.439	22.813		23.126	-6.193	0.01366604
35	24.000	23.930		23.965	-5.849	0.01735241	35	22.386	22.399		22.392	-5.459	0.02272752
36	25.111	24.853		24.982	-6.866	0.00857173	36	23.516	23.281		23.398	-6.465	0.01131712
37	24.261	24.323		24.292	-6.176	0.01383164	37	23.797	23.945		23.871	-6.938	0.0081547
38	25.190	25.578		25.384	-7.268	0.00648739	38	23.928	23.745		23.836	-6.903	0.00835425
39	23.968	24.189		24.079	-5.963	0.01603605	39	22.326	22.307		22.316	-5.383	0.02395838
40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!	40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
41	27.155	26.488		26.821	-8.705	0.00239613	41	25.803	25.483		25.643	-8.710	0.0023878
43	24.016	24.381		24.199	-6.083	0.01475473	43	22.782	22.561		22.672	-5.739	0.01872919
44	25.660	25.661		25.661	-7.545	0.00535587	44	23.842	24.379		24.110	-7.177	0.00691023
45	24.610	24.721		24.666	-6.550	0.01067251	45	23.301	23.284		23.293	-6.360	0.01217768
46	24.676	24.352		24.514	-6.398	0.01185919	46	23.009	23.132		23.071	-6.138	0.01420442
47	30.465	31.010		30.737	-12.621	0.00015869	47	28.680	28.329		28.504	-11.571	0.0003286
49	24.618	24.712		24.665	-6.549	0.01067914	49	23.563	23.270		23.416	-6.483	0.011176
50	30.419	29.309		29.864	-11.748	0.00029066	50	29.874	29.917		29.896	-12.963	0.00012528
51	23.174	22.999		23.087	-4.971	0.03189242	51	21.695	21.505		21.600	-4.667	0.03937209
52	27.091	26.836		26.963	-8.847	0.00217093	52	25.891	25.505		25.698	-8.765	0.00229854
53	23.477	23.333		23.405	-5.289	0.02557112	53	21.897	21.714		21.806	-4.873	0.03413451
54	22.818	23.184		23.001	-4.885	0.03383858	54	21.715	21.593		21.654	-4.721	0.03791258
55	26.187	26.164		26.175	-8.059	0.00374895	55	24.912	24.805		24.859	-7.926	0.0041121
56	24.778	24.873		24.826	-6.710	0.00955476	56	23.578	23.582		23.580	-6.647	0.00997666
57	22.984	22.935		22.959	-4.843	0.03483774	57	21.170	21.430		21.300	-4.367	0.04846248
58	24.348	24.118		24.233	-6.117	0.01440891	58	23.346	23.384		23.365	-6.432	0.01158518
var2csa	14.685	14.800		14.742	3.374	10.3652356	var2csa	13.109	13.119		13.114	3.819	14.1093146
91	23.333	23.442		23.387	-5.271	0.02588939	91	21.611	21.783		21.697	-4.764	0.03679983
92	22.224	22.473		22.349	-4.233	0.05319644	92	20.939	20.999		20.969	-4.036	0.06096117
93	23.749	23.730		23.739	-5.623	0.02028467	93	21.795	21.931		21.863	-4.930	0.03279686
94	25.827	25.985		25.906	-7.790	0.00451775	94	24.613	24.241		24.427	-7.494	0.00554664
95	25.544	25.416		25.480	-7.364	0.00607052	95	23.760	23.521		23.641	-6.708	0.00956839
96	25.605	25.195		25.400	-7.284	0.00641529	96	23.604	23.368		23.486	-6.553	0.01065256
97	24.757	24.914		24.836	-6.720	0.00948793	97	23.193	22.796		22.994	-6.061	0.01497417
60	18.114	18.117		18.116	0.000	1.00033554	60	16.922	16.944		16.933	0.000	0.99986554

PfSAHH Day 0							PfSAHH ON ATc						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	21.541	21.375		21.458	-3.081	0.11820861	1	20.526	20.514		20.520	-3.216	0.10764323
3	24.661	24.696		24.679	-6.302	0.01267706	3	22.484	22.112		22.298	-4.994	0.0313737
4	25.385	25.465		25.425	-7.048	0.00755504	4	22.450	22.245		22.347	-5.043	0.03032381
5	29.821	30.354		30.087	-11.710	0.0002984	5	26.382	25.167		25.774	-8.470	0.0028193
6	23.958	23.600		23.779	-5.402	0.02365013	6	22.138	21.788		21.963	-4.659	0.03957904
7	23.714	23.886		23.800	-5.423	0.02331095	7	19.808	19.879		19.844	-2.540	0.17197992
8	25.377	25.531		25.454	-7.077	0.00740602	8	21.341	21.236		21.289	-3.985	0.06317193
9	22.243	22.405		22.324	-3.947	0.06483902	9	20.931	20.940		20.935	-3.631	0.08070445
11	23.662	23.994		23.828	-5.451	0.02286304	11	22.370	22.565		22.468	-5.164	0.02790157
12	23.925	24.222		24.074	-5.697	0.01928022	12	20.651	20.661		20.656	-3.352	0.09793036
13	22.913	23.004		22.958	-4.581	0.04176932	13	19.868	20.188		20.028	-2.724	0.15134538
15	27.133	27.484		27.309	-8.932	0.00204802	15	22.112	21.980		22.046	-4.742	0.03736833
17	22.328	21.961		22.145	-3.768	0.07341301	17	19.704	19.634		19.669	-2.365	0.19414305
18	22.195	22.306		22.251	-3.874	0.0682134	18	20.220	20.200		20.210	-2.906	0.13341576
19	24.212	24.017		24.115	-5.738	0.01874113	19	20.540	20.550		20.545	-3.241	0.10579574
20	29.941	30.231		30.086	-11.709	0.00029871	20	22.999	23.702		23.351	-6.047	0.0151285
21	25.480	25.695		25.588	-7.211	0.00675063	21	21.536	21.898		21.717	-4.413	0.04695052
22	21.691	21.607		21.649	-3.272	0.103512	22	20.518	20.534		20.526	-3.222	0.10719043
23	25.190	25.484		25.337	-6.960	0.008032	23	22.446	22.357		22.401	-5.097	0.02921289
25	26.592	26.297		26.445	-8.068	0.0037271	25	22.830	23.131		22.980	-5.676	0.0195537
26	25.606	25.732		25.669	-7.292	0.00638026	26	21.203	21.398		21.300	-3.996	0.06265948
27	23.005	22.990		22.998	-4.621	0.04064735	27	19.397	19.303		19.350	-2.046	0.24212744
28	24.167	24.428		24.298	-5.921	0.01650882	28	21.994	21.848		21.921	-4.617	0.0407543
30	24.821	24.536		24.679	-6.302	0.01267489	30	22.485	22.620		22.552	-5.248	0.02630628
31	22.624	22.596		22.610	-4.233	0.05316763	31	17.693	17.849		17.771	-0.467	0.72359132
34	25.431	25.755		25.593	-7.216	0.00672662	34	20.233	20.567		20.400	-3.096	0.11693032
35	25.373	24.974		25.174	-6.797	0.00899414	35	21.617	21.740		21.678	-4.374	0.04822354
36	24.747	25.204		24.976	-6.599	0.01031724	36	21.441	21.534		21.487	-4.183	0.05504319
37	26.289	26.128		26.209	-7.832	0.00438978	37	22.801	22.633		22.717	-5.413	0.02346963
38	24.396	24.461		24.428	-6.051	0.01507765	38	21.425	21.631		21.528	-4.224	0.05351609
39	23.735	23.594		23.665	-5.288	0.02560197	39	21.516	21.553		21.535	-4.231	0.0532652
40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!	40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
41	23.315	23.241		23.278	-4.901	0.03346838	41	24.306	24.327		24.316	-7.012	0.00774617
43	23.391	23.357		23.374	-4.997	0.03131376	43	21.748	21.800		21.774	-4.470	0.04512493
44	26.293	26.017		26.155	-7.778	0.00455713	44	22.361	22.171		22.266	-4.962	0.03208885
45	25.605	25.424		25.514	-7.137	0.0071026	45	20.358	20.165		20.262	-2.958	0.12871829
46	27.214	26.603		26.909	-8.532	0.00270242	46	20.594	20.638		20.616	-3.312	0.10070673
47	32.727	32.689		32.708	-14.331	4.8521E-05	47	26.977	27.964		27.470	-10.166	0.00087013
49	25.779	25.811		25.795	-7.418	0.00584727	49	20.350	20.363		20.357	-3.053	0.12052263
50	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!	50	26.698	28.341		27.519	-10.215	0.00084115
51	26.989	27.280		27.134	-8.757	0.00231074	51	20.911	20.761		20.836	-3.532	0.08644037
52	29.097	29.422		29.260	-10.883	0.0005297	52	22.829	22.949		22.889	-5.585	0.02083132
53	22.364	22.295		22.329	-3.952	0.06459989	53	19.684	19.711		19.698	-2.394	0.19031821
54	22.475	22.636		22.556	-4.179	0.05522445	54	19.971	19.780		19.875	-2.571	0.1682545
55	23.506	23.298		23.402	-5.025	0.03070898	55	22.539	22.693		22.616	-5.312	0.02517506
56	23.464	23.560		23.512	-5.135	0.02845732	56	22.334	21.914		22.124	-4.820	0.03540809
57	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!	57	20.024	20.256		20.140	-2.836	0.14005666
58	20.809	23.292		22.051	-3.674	0.07836461	58	21.711	21.951		21.831	-4.527	0.04338331
var2csa	17.923	17.730		17.827	0.550	1.4643851	var2csa	14.928	15.004		14.966	2.338	5.05658893
91	21.678	21.699		21.689	-3.312	0.1007209	91	19.574	19.695		19.634	-2.330	0.19884814
92	21.493	21.466		21.480	-3.103	0.11642356	92	20.222	20.301		20.262	-2.958	0.1287075
93	24.302	24.276		24.289	-5.912	0.01660745	93	19.882	20.156		20.019	-2.715	0.15231891
94	27.874	27.815		27.845	-9.468	0.00141224	94	23.224	23.646		23.435	-6.131	0.01426588
95	23.615	23.659		23.637	-5.260	0.02609102	95	20.956	21.236		21.096	-3.792	0.07219246
96	25.279	25.768		25.523	-7.146	0.0070586	96	22.728	23.375		23.052	-5.748	0.01861274
97	26.229	26.481		26.355	-7.978	0.00396654	97	22.193	22.149		22.171	-4.867	0.03425918
60	18.393	18.361		18.377	0.000	1.0002929	60	17.333	17.276		17.304	0.000	0.9998295

PfSAHH OFF ATc							EV Day 0						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	19.523	20.130		19.826	-3.538	0.08608842	1	24.121	22.930		23.525	-3.137	0.11363782
3	21.220	21.418		21.319	-5.031	0.03058061	3	26.620	26.510		26.565	-6.177	0.01381707
4	21.947	21.355		21.651	-5.363	0.02429689	4	26.954	26.423		26.689	-6.301	0.01268574
5	24.156	24.513		24.335	-8.047	0.00378225	5	25.776	25.858		25.817	-5.429	0.02321333
6	21.268	20.559		20.913	-4.625	0.04051753	6	24.860	25.627		25.243	-4.855	0.03454507
7	19.170	19.120		19.145	-2.857	0.13799702	7	22.752	22.758		22.755	-2.367	0.19384985
8	20.583	20.452		20.517	-4.229	0.05331386	8	24.462	24.624		24.543	-4.155	0.05613917
9	20.189	19.962		20.075	-3.787	0.07243521	9	23.833	23.650		23.741	-3.353	0.09784768
11	21.448	21.441		21.445	-5.157	0.02803565	11	25.749	26.995		26.372	-5.984	0.0158017
12	19.568	19.690		19.629	-3.341	0.09868386	12	25.001	25.198		25.100	-4.712	0.0381656
13	18.921	19.282		19.101	-2.813	0.14226794	13	23.956	23.923		23.939	-3.551	0.08530014
15	20.985	20.562		20.774	-4.486	0.04463977	15	26.740	27.186		26.963	-6.575	0.01049042
17	18.654	18.666		18.660	-2.372	0.19314497	17	23.258	23.290		23.274	-2.886	0.13527508
18	18.996	19.408		19.202	-2.914	0.1327008	18	23.381	23.291		23.336	-2.948	0.12960569
19	19.477	19.443		19.460	-3.172	0.11091797	19	23.780	24.169		23.975	-3.587	0.08322966
20	22.683	22.857		22.770	-6.482	0.01118773	20	29.627	29.962		29.795	-9.407	0.00147328
21	20.895	20.559		20.727	-4.439	0.0461128	21	25.457	25.354		25.405	-5.017	0.03087798
22	19.588	19.574		19.581	-3.293	0.10202527	22	24.149	23.525		23.837	-3.449	0.09156589
23	21.447	21.350		21.398	-5.110	0.02894874	23	27.613	26.604		27.108	-6.720	0.00948288
25	21.954	21.836		21.895	-5.607	0.02051828	25	28.219	27.542		27.881	-7.493	0.00555236
26	20.315	20.300		20.308	-4.020	0.0616502	26	27.537	27.683		27.610	-7.222	0.00669921
27	18.514	18.441		18.478	-2.190	0.21921691	27	23.802	23.809		23.805	-3.417	0.09359326
28	20.983	20.776		20.879	-4.591	0.04148518	28	23.642	23.805		23.724	-3.336	0.09903834
30	21.743	21.565		21.654	-5.366	0.02424689	30	26.235	26.699		26.467	-6.079	0.01479477
31	16.820	17.172		16.996	-0.708	0.61230866	31	24.182	23.853		24.017	-3.629	0.08080347
34	19.634	19.540		19.587	-3.299	0.10159572	34	26.412	27.327		26.870	-6.482	0.01119048
35	20.826	20.877		20.851	-4.563	0.0422978	35	27.017	26.687		26.852	-6.464	0.01132976
36	20.686	20.661		20.673	-4.385	0.0478475	36	26.155	26.370		26.263	-5.875	0.01704263
37	22.579	22.283		22.431	-6.143	0.01414943	37	28.558	27.993		28.276	-7.888	0.00422217
38	20.812	20.657		20.734	-4.446	0.04586572	38	25.989	25.471		25.730	-5.342	0.02465443
39	20.646	20.559		20.602	-4.314	0.05026347	39	24.980	25.841		25.411	-5.023	0.03076381
40	31.920	31.406		31.663	-15.375	2.3537E-05	40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
41	23.449	23.257		23.353	-7.065	0.00746741	41	25.503	25.390		25.447	-5.059	0.03000629
43	20.993	20.693		20.843	-4.555	0.04254039	43	25.156	25.676		25.416	-5.028	0.03064649
44	22.211	21.989		22.100	-5.812	0.01779911	44	27.218	27.285		27.251	-6.863	0.00858933
45	19.443	19.505		19.474	-3.186	0.10990329	45	26.439	26.260		26.350	-5.962	0.01604353
46	19.821	19.830		19.826	-3.538	0.08610704	46	26.261	25.838		26.049	-5.661	0.0197587
47	26.653	25.413		26.033	-9.745	0.00116531	47	32.572	32.572		32.572	-12.184	0.0002149
49	19.873	19.764		19.818	-3.530	0.08654966	49	26.875	26.524		26.700	-6.312	0.01258949
50	26.798	26.443		26.620	-10.332	0.0007758	50	31.633	31.633		31.633	-11.245	0.00041208
51	19.956	19.735		19.845	-3.557	0.08493712	51	26.345	26.378		26.362	-5.974	0.01591078
52	22.556	21.896		22.226	-5.938	0.01630811	52	27.465	29.636		28.550	-8.162	0.00349022
53	18.876	18.839		18.857	-2.569	0.16850491	53	24.544	24.877		24.711	-4.323	0.04997668
54	19.129	18.962		19.045	-2.757	0.14788581	54	18.158	18.120		18.139	2.249	4.75346817
55	21.591	21.571		21.581	-5.293	0.02550431	55	27.012	27.518		27.265	-6.877	0.0085087
56	21.173	21.265		21.219	-4.931	0.03278497	56	25.453	25.277		25.365	-4.977	0.03175265
57	19.360	19.482		19.421	-3.133	0.11402274	57	24.321	24.157		24.239	-3.851	0.0692932
58	20.986	20.926		20.956	-4.668	0.03934213	58	23.674	23.520		23.597	-3.209	0.10815314
var2csa	14.232	14.402		14.317	1.971	3.91953905	var2csa	24.435	24.316		24.375	-3.987	0.06304984
91	18.854	18.868		18.861	-2.573	0.1680256	91	24.175	24.393		24.284	-3.896	0.06716891
92	19.417	19.307		19.362	-3.074	0.11876832	92	24.475	24.278		24.377	-3.989	0.06299478
93	19.230	19.263		19.246	-2.958	0.12865251	93	24.427	24.384		24.406	-4.018	0.06173979
94	22.501	22.394		22.448	-6.160	0.01398782	94	25.827	25.932		25.879	-5.491	0.02222829
95	20.426	20.463		20.444	-4.156	0.05607578	95	28.889	28.668		28.778	-8.390	0.00298007
96	21.914	22.418		22.166	-5.878	0.01700504	96	24.844	24.811		24.828	-4.440	0.04608431
97	21.275	21.392		21.334	-5.046	0.03027697	97	25.886	25.538		25.712	-5.324	0.02496494
60	16.247	16.328		16.288	0.000	1.00012477	60	20.438	20.338		20.388	0.000	1.00005649

EV Control ON ATc							EV Control OFF ATc						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	25.162	24.134		24.648	-6.337	0.01237145	1	24.464	24.483		24.473	-5.690	0.01936584
3	25.506	25.657		25.581	-7.270	0.00647845	3	26.396	26.604		26.500	-7.717	0.00475378
4	24.949	24.666		24.807	-6.496	0.01107695	4	26.354	25.233		25.794	-7.011	0.00775521
5	24.625	24.627		24.626	-6.315	0.01256085	5	24.803	24.669		24.736	-5.953	0.01614736
6	24.637	24.161		24.399	-6.088	0.01470469	6	24.736	24.811		24.773	-5.990	0.015729
7	23.163	22.652		22.907	-4.596	0.0413395	7	23.677	23.512		23.595	-4.812	0.03561042
8	24.411	24.164		24.288	-5.977	0.01587972	8	24.739	24.740		24.739	-5.956	0.01610542
9	24.423	24.300		24.362	-6.051	0.01508686	9	24.978	24.892		24.935	-6.152	0.01406305
11	23.845	23.725		23.785	-5.474	0.0224989	11	24.091	24.041		24.066	-5.283	0.02567554
12	24.532	24.142		24.337	-6.026	0.0153466	12	24.832	24.484		24.658	-5.875	0.01704096
13	22.001	21.647		21.824	-3.513	0.08758491	13	22.542	22.018		22.280	-3.497	0.08856872
15	24.649	24.313		24.481	-6.170	0.01388962	15	25.708	25.173		25.441	-6.658	0.00990488
17	21.860	21.575		21.717	-3.406	0.09432091	17	22.274	21.979		22.126	-3.343	0.09852479
18	23.195	22.815		23.005	-4.694	0.0386276	18	23.204	23.163		23.183	-4.400	0.0473531
19	23.397	23.256		23.326	-5.015	0.03092004	19	23.988	23.659		23.824	-5.041	0.03038062
21	24.431	24.434		24.433	-6.122	0.01436077	21	25.534	25.449		25.492	-6.709	0.00956136
21	23.725	23.479		23.602	-5.291	0.02554053	21	24.761	24.340		24.551	-5.768	0.01835659
22	23.886	23.921		23.903	-5.592	0.02073087	22	24.418	24.383		24.401	-5.618	0.02036468
23	25.208	25.143		25.175	-6.864	0.00858323	23	25.803	25.653		25.728	-6.945	0.00811482
25	25.698	25.396		25.547	-7.236	0.00663218	25	26.842	26.163		26.502	-7.719	0.00474595
26	23.500	23.314		23.407	-5.096	0.02924737	26	23.976	23.805		23.891	-5.108	0.02900213
27	21.667	21.626		21.647	-3.336	0.09904969	27	21.921	21.704		21.812	-3.029	0.12247678
28	24.812	24.406		24.609	-6.298	0.01270927	28	26.445	24.998		25.722	-6.939	0.00815147
30	25.870	25.510		25.690	-7.379	0.00600823	30	26.771	25.806		26.288	-7.505	0.00550369
31	18.519	18.346		18.433	-0.122	0.91915675	31	19.095	18.833		18.964	-0.181	0.88208802
34	20.767	19.779		20.273	-1.962	0.25671977	34	20.758	20.576		20.667	-1.884	0.27096179
35	24.370	24.255		24.312	-6.001	0.0156092	35	25.469	25.367		25.418	-6.635	0.0100605
36	24.247	24.124		24.185	-5.874	0.01704698	36	24.719	24.530		24.624	-5.841	0.01744392
37	22.974	22.694		22.834	-4.523	0.04349846	37	24.193	23.838		24.015	-5.232	0.02660099
38	24.394	24.471		24.432	-6.121	0.01436433	38	25.707	25.332		25.520	-6.737	0.00937687
39	23.012	23.568		23.290	-4.979	0.03170594	39	23.382	23.328		23.355	-4.572	0.04204232
40	Undetermine	34.687		34.687	-16.376	1.1762E-05	40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
41	25.978	25.964		25.971	-7.660	0.00494571	41	26.917	26.638		26.778	-7.995	0.00392065
43	24.695	24.390		24.543	-6.232	0.01330797	43	24.819	24.943		24.881	-6.098	0.01460089
44	26.668	25.846		26.257	-7.946	0.00405471	44	26.453	25.978		26.215	-7.432	0.00579011
45	24.109	23.995		24.052	-5.741	0.01870297	45	24.580	24.543		24.562	-5.779	0.01821666
46	25.295	24.988		25.141	-6.830	0.00878649	46	26.106	26.303		26.205	-7.422	0.00583288
47	27.775	27.699		27.737	-9.426	0.0014541	47	28.241	27.567		27.904	-9.121	0.00179594
49	24.014	23.812		23.913	-5.602	0.02059389	49	24.760	24.945		24.853	-6.070	0.01488584
50	23.692	23.461		23.577	-5.266	0.02599084	50	24.405	24.136		24.271	-5.488	0.02228869
51	23.851	23.749		23.800	-5.489	0.02226293	51	23.967	24.148		24.058	-5.275	0.0258296
52	22.448	22.338		22.393	-4.082	0.05904358	52	23.191	22.990		23.091	-4.308	0.05049584
53	21.754	21.528		21.641	-3.330	0.09945154	53	21.941	22.016		21.979	-3.196	0.1091503
54	14.825	15.172		14.999	3.312	9.93412459	54	15.655	15.623		15.639	3.144	8.83859147
55	23.570	23.476		23.523	-5.212	0.02697721	55	23.460	23.582		23.521	-4.738	0.03747459
56	24.572	24.432		24.502	-6.191	0.01368752	56	24.789	24.894		24.842	-6.059	0.01500246
57	23.101	22.731		22.916	-4.605	0.04109885	57	23.285	24.641		23.963	-5.180	0.02758656
58	23.482	23.446		23.464	-5.153	0.02809869	58	24.333	24.259		24.296	-5.513	0.0218996
var2csa	21.542	21.400		21.471	-3.160	0.11190493	*var2csa*	22.618	22.557		22.587	-3.804	0.07157765
91	22.932	22.808		22.870	-4.559	0.0424202	91	23.516	23.528		23.522	-4.739	0.03745908
92	22.131	22.268		22.200	-3.889	0.06750198	92	23.243	22.561		22.902	-4.119	0.05753675
93	23.230	23.008		23.119	-4.808	0.03570055	93	23.836	23.675		23.755	-4.972	0.03185735
94	25.754	25.647		25.700	-7.389	0.0059644	94	26.137	26.001		26.069	-7.286	0.00640918
95	24.368	24.217		24.293	-5.982	0.01582592	95	25.635	25.506		25.570	-6.787	0.00905328
96	25.640	25.432		25.536	-7.225	0.00668517	96	26.503	26.701		26.602	-7.819	0.00442979
97	25.579	25.299		25.439	-7.128	0.00714781	97	26.557	26.465		26.511	-7.728	0.00471739
60	18.132	18.489		18.311	0.000	1.00033066	60	18.766	18.800		18.783	0.000	1.0000476

PfSAMS Day 0 (Rep 2)					PfSAMS ON ATc (Rep 2)				
Primer pair	CT		relative to p60	relative copy number	Primer pair	CT		relative to p60	relative copy number
1	Undetermined		#VALUE!	#VALUE!	1	25.899		-8.704	0.00239793
3	26.920		-9.129244	0.00178576	3	26.977		-9.782	0.00113549
4	25.544		-7.753436	0.00463429	4	25.534		-8.339	0.003089
5	27.521		-9.730338	0.00117727	5	26.637		-9.442	0.00143754
6	23.594		-5.803063	0.01791035	6	23.367		-6.172	0.01386817
7	24.278		-6.487046	0.0111482	7	24.251		-7.056	0.00751434
8	33.053		-15.261624	2.5456E-05	8	31.915		-14.720	3.7053E-05
9	25.756		-7.965083	0.00400194	9	23.964		-6.769	0.00917201
11	27.267		-9.476029	0.00140421	11	25.167		-7.972	0.00398299
12	24.262		-6.470744	0.01127488	12	23.902		-6.707	0.00957133
13	24.678		-6.886935	0.0084494	13	22.947		-5.752	0.01855984
15	27.712		-9.92136	0.00103127	15	26.146		-8.951	0.00202009
17	24.770		-6.978749	0.00792843	17	23.962		-6.767	0.00918136
18	23.730		-5.939433	0.01629493	18	23.575		-6.380	0.01200443
19	26.465		-8.674384	0.00244766	19	25.488		-8.293	0.0031873
20	28.857		-11.066107	0.00046641	20	26.554		-9.359	0.00152245
21	27.514		-9.723029	0.00118325	21	25.963		-8.768	0.00229433
22	23.343		-5.552235	0.02131134	22	23.619		-6.424	0.01164393
23	26.908		-9.11742	0.00180046	23	26.726		-9.531	0.00135215
25	29.766		-11.975054	0.0002484	25	22.801		-5.606	0.02052503
26	28.100		-10.308758	0.00078841	26	26.180		-8.985	0.00197348
27	24.729		-6.937514	0.00815831	27	21.568		-4.373	0.04827533
28	Undetermined		#VALUE!	#VALUE!	28	26.440		-9.245	0.00164836
30	29.635		-11.843886	0.00027204	30	28.289		-11.094	0.0004574
31	25.633		-7.841946	0.00435852	31	23.715		-6.520	0.01089779
34	27.267		-9.475817	0.00140441	34	25.920		-8.725	0.00236272
35	26.574		-8.783408	0.0022695	35	25.646		-8.451	0.00285705
36	25.610		-7.818632	0.00442953	36	25.849		-8.654	0.0024819
37	25.747		-7.95559	0.00402836	37	23.823		-6.628	0.01010992
38	25.859		-8.067692	0.0037272	38	25.599		-8.404	0.00295211
39	24.944		-7.152832	0.00702721	39	22.950		-5.755	0.0185189
40	Undetermined		#VALUE!	#VALUE!	40	Undetermined		#VALUE!	#VALUE!
41	24.880		-7.08897	0.00734526	41	24.378		-7.183	0.00688371
43	24.776		-6.98545	0.00789169	43	24.273		-7.078	0.00739961
44	28.446		-10.654631	0.00062035	44	26.664		-9.469	0.00141101
45	27.723		-9.931647	0.00102394	45	25.297		-8.102	0.00363862
46	29.320		-11.529147	0.00033836	46	26.714		-9.519	0.00136256
47	32.516		-14.724556	3.6937E-05	47	31.686		-14.491	4.3439E-05
49	26.631		-8.839642	0.00218274	49	25.276		-8.081	0.00369174
50	32.533		-14.741803	3.6499E-05	50	31.260		-14.065	5.8332E-05
51	27.006		-9.215329	0.00168232	51	25.183		-7.988	0.00393867
52	27.915		-10.1236	0.00089638	52	26.800		-9.605	0.00128382
53	25.328		-7.537045	0.00538423	53	25.558		-8.363	0.00303746
54	23.928		-6.136599	0.01421345	54	26.461		-9.266	0.00162457
55	26.351		-8.560158	0.00264933	55	25.868		-8.673	0.00244916
56	24.931		-7.13971	0.00709142	56	26.149		-8.954	0.00201673
57	24.569		-6.778138	0.00911123	57	24.178		-6.983	0.00790628
58	22.924		-5.132824	0.02850139	58	27.779		-10.584	0.0006516
var2csa	16.734		1.05691	2.08047074	var2csa	15.279		1.916	3.7739675
91	24.924		-7.133301	0.00712299	91	25.237		-8.042	0.00379424
92	21.839		-4.04814	0.0604489	92	20.984		-3.789	0.07236305
93	26.584		-8.79302	0.00225443	93	25.926		-8.731	0.00235365
94	29.099		-11.307726	0.00039449	94	27.121		-9.926	0.00102814
95	27.348		-9.556654	0.00132789	95	26.503		-9.308	0.00157814
96	25.324		-7.533492	0.0053975	96	25.290		-8.095	0.00365692
97	26.923		-9.13221	0.0017821	97	27.516		-10.321	0.00078184
60	17.791		0.000278	1.00019271	60	17.195		0.000	0.99989603

PfSAMS OFF ATc (Rep 2)					EV ON ATc (Rep 2)						
Primer pair	CT		relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	24.405		-6.429	0.01160349	1	26.813	26.913		26.863	-2.801	0.1435062
3	26.595		-8.619	0.0025432	3	27.699	29.127		28.413	-4.351	0.04900501
4	25.872		-7.896	0.00419736	4	29.863	29.177		29.520	-5.458	0.02275178
5	26.809		-8.833	0.0021924	5	28.779	28.428		28.604	-4.542	0.04293851
6	23.647		-5.671	0.01963253	6	29.186	28.821		29.003	-4.941	0.03254662
7	23.894		-5.918	0.01654421	7	26.837	27.704		27.270	-3.208	0.10819528
8	32.547		-14.571	4.108E-05	8	27.357	27.811		27.584	-3.522	0.08705437
9	23.392		-5.416	0.02341614	9	26.662	27.707		27.184	-3.122	0.11483478
11	25.196		-7.220	0.00670883	11	29.268	31.425		30.346	-6.284	0.01283113
12	24.323		-6.347	0.01228047	12	26.493	25.884		26.189	-2.127	0.22896492
13	22.881		-4.905	0.03338047	13	26.316	26.569		26.442	-2.380	0.1920865
15	25.828		-7.852	0.00432912	15	26.889	27.906		27.397	-3.335	0.09906838
17	23.395		-5.419	0.02336722	17	25.702	25.216		25.459	-1.397	0.37972911
18	23.278		-5.302	0.02533959	18	25.806	26.256		26.031	-1.969	0.25539426
19	24.929		-6.953	0.00807369	19	27.180	26.654		26.917	-2.855	0.13823602
20	25.407		-7.431	0.00579352	20	Undetermine	30.218		30.218	-6.156	0.0140265
21	25.388		-7.412	0.00587124	21	Undetermine	28.767		28.767	-4.705	0.03833123
22	23.650		-5.674	0.01958347	22	27.917	26.676		27.297	-3.235	0.10623562
23	26.493		-8.517	0.00273043	23	Undetermine	29.167		29.167	-5.105	0.02904654
25	22.149		-4.173	0.05543681	25	Undetermine	28.581		28.581	-4.519	0.04361327
26	25.783		-7.807	0.00446516	26	31.732	27.618		29.675	-5.613	0.02043263
27	21.184		-3.208	0.10820859	27	Undetermine	24.474		24.474	-0.412	0.7515
28	24.904		-6.928	0.00820971	28	Undetermine	26.611		26.611	-2.549	0.17083234
30	27.553		-9.577	0.00130967	30	28.713	28.500		28.607	-4.545	0.04284219
31	22.669		-4.693	0.03865326	31	24.693	24.853		24.773	-0.711	0.61085696
34	25.416		-7.440	0.00575969	34	27.887	28.375		28.131	-4.069	0.05956516
35	24.869		-6.893	0.00841352	35	29.893	29.600		29.746	-5.684	0.01944801
36	26.109		-8.133	0.00356252	36	28.364	29.228		28.796	-4.734	0.03756903
37	23.308		-5.332	0.02482349	37	29.766	29.929		29.847	-5.785	0.01813051
38	25.198		-7.222	0.00669906	38	28.665	28.448		28.557	-4.495	0.04435935
39	23.481		-5.505	0.02201984	39	28.799	27.905		28.352	-4.290	0.05111711
40	Undetermined		#VALUE!	#VALUE!	40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
41	24.352		-6.376	0.01203854	41	29.982	28.787		29.385	-5.323	0.02498702
43	23.825		-5.849	0.01735029	43	26.735	26.792		26.764	-2.702	0.15370005
44	25.507		-7.531	0.00540549	44	28.511	29.306		28.908	-4.846	0.03475858
45	24.583		-6.607	0.01026217	45	26.435	26.930		26.683	-2.621	0.16259254
46	25.851		-7.875	0.00425883	46	26.965	27.172		27.068	-3.006	0.12444529
47	29.770		-11.794	0.00028171	47	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
49	24.916		-6.940	0.00814447	49	27.588	27.667		27.627	-3.565	0.08448594
50	29.217		-11.241	0.0004133	50	30.137	Undetermined		30.137	-6.075	0.01483579
51	24.638		-6.662	0.00987294	51	29.700	28.415		29.057	-4.995	0.03134911
52	25.865		-7.889	0.00421921	52	27.405	29.657		28.531	-4.469	0.04515688
53	24.719		-6.743	0.00933661	53	28.859	26.307		27.583	-3.521	0.08712309
54	22.706		-4.730	0.03768335	54	20.578	20.269		20.423	3.639	12.4541616
55	25.263		-7.287	0.00640362	55	27.962	28.792		28.377	-4.315	0.05024332
56	25.141		-7.165	0.0069682	56	24.903	25.690		25.296	-1.234	0.42502659
57	23.526		-5.550	0.02134127	57	26.231	24.916		25.573	-1.511	0.35075437
58	27.403		-9.427	0.00145309	58	29.248	29.440		29.344	-5.282	0.02570119
var2csa	14.690		3.286	9.75466278	var2csa	24.219	24.262		24.241	-0.179	0.88356918
91	27.623		-9.647	0.00124705	91	26.737	26.201		26.469	-2.407	0.1885745
92	21.229		-3.253	0.10489843	92	27.609	27.745		27.677	-3.615	0.08159771
93	Undetermined		#VALUE!	#VALUE!	93	26.574	27.355		26.965	-2.903	0.13372119
94	25.907		-7.931	0.00409847	94	30.513	28.840		29.677	-5.615	0.02040826
95	26.070		-8.094	0.00365865	95	26.248	26.438		26.343	-2.281	0.20575024
96	25.179		-7.203	0.00678647	96	28.374	29.142		28.758	-4.696	0.03857495
97	26.576		-8.600	0.00257747	97	27.703	27.811		27.757	-3.695	0.07720831
60	17.976		0.000	0.99969575	60	24.193	23.931		24.062	0.000	0.99993519

EV OFF ATc (Rep 2)							PfSAHH Day 0 (Rep 2)				
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT		relative to p60	relative copy number
1	21.241	20.828		21.034	-4.125	0.05729397	1	Undetermined		#VALUE!	#VALUE!
3	22.873	23.196		23.035	-6.126	0.01432125	3	24.290		-7.964873	0.00400253
4	22.642	22.392		22.517	-5.608	0.02050272	4	23.412		-7.086997	0.00735532
5	22.474	22.804		22.639	-5.730	0.01884243	5	24.248		-7.922562	0.00412165
6	22.851	22.358		22.605	-5.696	0.01929474	6	23.537		-7.212075	0.00674449
7	21.489	21.519		21.504	-4.595	0.04137286	7	24.201		-7.875697	0.00425774
8	21.875	22.468		22.171	-5.262	0.02605156	8	Undetermined		#VALUE!	#VALUE!
9	21.530	21.310		21.420	-4.511	0.04387041	9	22.500		-6.175418	0.01383611
11	23.463	23.362		23.412	-6.503	0.01102201	11	24.618		-8.293126	0.00318801
12	21.365	21.272		21.319	-4.410	0.04705381	12	23.194		-6.869315	0.00855323
13	20.890	21.399		21.145	-4.236	0.05307595	13	21.301		-4.975772	0.03177923
15	22.948	23.007		22.978	-6.069	0.01489753	15	23.502		-7.176724	0.00691179
17	20.676	20.738		20.707	-3.798	0.07189319	17	21.924		-5.59922	0.02062846
18	20.977	20.800		20.889	-3.980	0.06338016	18	Undetermined		#VALUE!	#VALUE!
19	21.807	21.921		21.864	-4.955	0.03224619	19	22.706		-6.381324	0.01199582
20	26.161	26.316		26.239	-9.330	0.00155397	20	26.417		-10.091853	0.00091632
21	23.409	23.478		23.444	-6.535	0.0107873	21	23.143		-6.817572	0.00886556
22	21.698	21.574		21.636	-4.727	0.03775792	22	22.381		-6.055938	0.01503076
23	23.462	23.230		23.346	-6.437	0.01154231	23	24.709		-8.38443	0.00299251
25	23.546	23.543		23.545	-6.636	0.01005767	25	23.342		-7.016932	0.00772135
26	21.847	22.006		21.927	-5.018	0.03087215	26	22.870		-6.545321	0.01070686
27	19.993	19.927		19.960	-3.051	0.12064299	27	21.823		-5.498494	0.02212017
28	21.711	21.417		21.564	-4.655	0.03969542	28	24.402		-8.076676	0.00370406
30	23.867	23.920		23.894	-6.985	0.00789555	30	24.365		-8.039733	0.00380014
31	18.781	19.202		18.991	-2.082	0.23613863	31	19.849		-3.524493	0.08690042
34	21.562	19.186		20.374	-3.465	0.0905813	34	20.012		-3.686894	0.07764872
35	22.748	22.232		22.490	-5.581	0.02088427	35	24.218		-7.892587	0.00420818
36	22.808	22.870		22.839	-5.930	0.01639997	36	23.406		-7.081425	0.00738378
37	23.295	23.203		23.249	-6.340	0.01234268	37	21.793		-5.468074	0.02259153
38	22.857	22.939		22.898	-5.989	0.01574332	38	23.113		-6.78835	0.00904697
39	21.768	21.473		21.621	-4.712	0.03816259	39	22.693		-6.367936	0.01210766
40	Undetermine	34.234		34.234	-17.325	6.0923E-06	40	Undetermined		#VALUE!	#VALUE!
41	24.970	25.315		25.143	-8.234	0.00332182	41	23.328		-7.002837	0.00779715
43	22.690	22.895		22.793	-5.884	0.01693897	43	22.388		-6.063105	0.01495628
44	23.759	24.309		24.034	-7.125	0.00716472	44	25.593		-9.267964	0.00162205
45	22.175	21.875		22.025	-5.116	0.02884074	45	23.644		-7.31935	0.00626118
46	22.216	22.309		22.262	-5.353	0.02445985	46	25.309		-8.98441	0.00197435
47	29.340	30.907		30.124	-13.215	0.0001052	47	28.756		-12.431145	0.00018107
49	22.454	22.812		22.633	-5.724	0.01892004	49	22.653		-6.328343	0.01244454
50	29.936	30.958		30.447	-13.538	8.4074E-05	50	28.531		-12.205682	0.0002117
51	22.172	22.229		22.201	-5.292	0.02553067	51	23.932		-7.60655	0.00513098
52	24.841	25.808		25.325	-8.416	0.00292844	52	25.739		-9.413794	0.00146611
53	20.682	20.734		20.708	-3.799	0.07182817	53	21.684		-5.359273	0.02436117
54	14.343	14.869		14.606	2.303	4.93565139	54	14.141		2.18389	4.5437706
55	24.422	24.266		24.344	-7.435	0.0057788	55	24.257		-7.931865	0.00409516
56	22.503	22.273		22.388	-5.479	0.02241901	56	23.612		-7.287123	0.00640262
57	20.248	20.414		20.331	-3.422	0.09330615	57	22.675		-6.349828	0.01226059
58	22.451	22.308		22.379	-5.470	0.02255515	58	22.977		-6.652274	0.00994182
var2csa	17.525	17.207		17.366	-0.457	0.72872935	var2csa	15.146		1.178842	2.26394985
91	20.633	20.487		20.560	-3.651	0.07963119	91	23.901		-7.57613	0.00524032
92	20.559	19.573		20.066	-3.157	0.11209715	92	21.374		-5.048627	0.03021425
93	21.356	21.179		21.267	-4.358	0.04875238	93	28.576		-12.251012	0.00020515
94	25.262	24.825		25.043	-8.134	0.00355858	94	25.118		-8.792863	0.00225468
95	22.251	22.369		22.310	-5.401	0.02366581	95	24.462		-8.136922	0.00355257
96	24.216	23.998		24.107	-7.198	0.00680962	96	23.875		-7.550162	0.0053355
97	21.805	21.864		21.834	-4.925	0.03291429	97	25.306		-8.981473	0.00197837
60	16.986	16.832		16.909	0.000	0.99997817	60	16.325		-0.000361	0.99974981

PfSAHH ON ATc (Rep 2)					PfSAHH OFF ATc (Rep 2)				
Primer pair	CT		relative to p60	relative copy number	Primer pair	CT		relative to p60	relative copy number
1	25.656		-5.148	0.028199	1	22.874		-4.379	0.04805772
3	28.225		-7.717	0.00475299	3	23.432		-4.937	0.03264432
4	26.703		-6.195	0.01364577	4	22.665		-4.170	0.05556969
5	27.594		-7.086	0.00736184	5	23.517		-5.022	0.03077964
6	24.320		-3.812	0.07117562	6	22.903		-4.408	0.04709942
7	25.266		-4.758	0.036946	7	22.603		-4.108	0.05797549
8	25.888		-5.380	0.0240163	8	24.767		-6.272	0.01293572
9	23.963		-3.455	0.09121026	9	22.447		-3.952	0.06461442
11	26.454		-5.946	0.01622443	11	23.749		-5.254	0.0262074
12	25.479		-4.971	0.03189544	12	Undetermined		#VALUE!	#VALUE!
13	23.586		-3.078	0.11839171	13	22.325		-3.830	0.07029408
15	24.965		-4.457	0.04553507	15	Undetermined		#VALUE!	#VALUE!
17	23.518		-3.010	0.12410903	17	21.374		-2.879	0.1359794
18	24.230		-3.722	0.07577065	18	Undetermined		#VALUE!	#VALUE!
19	Undetermined		#VALUE!	#VALUE!	19	21.779		-3.284	0.10269117
20	27.335		-6.827	0.0088078	20	24.374		-5.879	0.0169956
21	25.522		-5.014	0.03094699	21	22.760		-4.265	0.05202205
22	25.327		-4.819	0.03542712	22	21.286		-2.791	0.14445439
23	26.770		-6.262	0.01303324	23	23.129		-4.634	0.0402667
25	24.633		-4.125	0.05732006	25	22.245		-3.750	0.07433415
26	25.215		-4.707	0.03828502	26	24.173		-5.678	0.01953476
27	23.352		-2.844	0.13927295	27	20.648		-2.153	0.22476745
28	25.254		-4.746	0.03726838	28	23.959		-5.464	0.02265482
30	27.891		-7.383	0.00598909	30	24.119		-5.624	0.02027299
31	20.952		-0.444	0.73486238	31	19.937		-1.442	0.36804013
34	23.654		-3.146	0.11296001	34	22.428		-3.933	0.06546965
35	26.737		-6.229	0.01332932	35	23.802		-5.307	0.02526053
36	26.500		-5.992	0.01571546	36	22.660		-4.165	0.05573189
37	23.746		-3.238	0.1059792	37	20.464		-1.969	0.25541125
38	25.173		-4.665	0.0394296	38	22.665		-4.170	0.05555321
39	25.849		-5.341	0.02466867	39	22.735		-4.240	0.05291667
40	Undetermined		#VALUE!	#VALUE!	40	Undetermined		#VALUE!	#VALUE!
41	26.354		-5.846	0.01738934	41	24.596		-6.101	0.01456496
43	25.739		-5.231	0.02662014	43	22.850		-4.355	0.04887613
44	27.465		-6.957	0.00805041	44	24.295		-5.800	0.01794555
45	25.558		-5.050	0.03019208	45	22.815		-4.320	0.05005719
46	25.148		-4.640	0.04010236	46	23.222		-4.727	0.03774724
47	30.478		-9.970	0.00099684	47	27.697		-9.202	0.00169783
49	24.405		-3.897	0.06712602	49	23.418		-4.923	0.03295655
50	31.837		-11.329	0.00038865	50	27.866		-9.371	0.00151072
51	26.430		-5.922	0.01649351	51	22.734		-4.239	0.052956
52	27.901		-7.393	0.00594756	52	24.350		-5.855	0.01727411
53	24.128		-3.620	0.08131791	53	21.126		-2.631	0.16145612
54	16.650		3.858	14.5032669	54	18.998		-0.503	0.70546578
55	26.452		-5.944	0.01624212	55	22.256		-3.761	0.07377327
56	26.450		-5.942	0.01626623	56	23.318		-4.823	0.03531836
57	25.609		-5.101	0.02914556	57	22.360		-3.865	0.06865155
58	28.301		-7.793	0.00450932	58	Undetermined		#VALUE!	#VALUE!
var2csa	15.520		4.988	31.7398411	var2csa	18.268		0.227	1.17070534
91	24.689		-4.181	0.05511776	91	20.898		-2.403	0.1890959
92	23.652		-3.144	0.11311154	92	20.722		-2.227	0.21360362
93	24.885		-4.377	0.04811652	93	21.520		-3.025	0.12285087
94	27.016		-6.508	0.01098905	94	25.127		-6.632	0.01008364
95	23.545		-3.037	0.12179645	95	24.137		-5.642	0.02002793
96	27.000		-6.492	0.01111071	96	24.532		-6.037	0.01523451
97	27.338		-6.830	0.00878967	97	25.175		-6.680	0.00975045
60	17.508		3.000	8.00116457	60	18.495		0.000	1.00016637

Chapter 3.3 *var* panels

PfSAMS KD1 Day 0							PfSAMS KD2 Day 0						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	30.527	30.008		30.268	-11.728	0.00029488	1	Undetermine	32.971		32.971	-14.287	5.00222E-05
3	24.699	24.969		24.834	-6.294	0.01274162	3	23.877	23.636		23.757	-5.073	0.029718292
4	16.864	17.296		17.080	1.460	2.75166817	4	15.314	15.371		15.342	3.342	10.13787236
5	25.480	25.325		25.403	-6.863	0.00859188	5	24.486	24.537		24.511	-5.827	0.01761037
6	23.624	23.714		23.669	-5.129	0.02857767	6	23.223	22.922		23.073	-4.389	0.047741129
7	21.125	21.482		21.304	-2.764	0.14724566	7	21.575	21.599		21.587	-2.903	0.133704646
8	20.994	20.883		20.938	-2.398	0.18967895	8	20.784	20.691		20.738	-2.054	0.240895108
9	25.451	25.583		25.517	-6.977	0.00793596	9	23.497	23.396		23.446	-4.762	0.036843834
11	25.574	25.632		25.603	-7.063	0.00748066	11	25.125	20.830		22.978	-4.294	0.050991373
12	23.286	23.130		23.208	-4.668	0.03934438	12	22.889	22.706		22.798	-4.114	0.057763621
13	19.818	20.156		19.987	-1.447	0.36682351	13	20.101	20.250		20.176	-1.492	0.355622004
15	16.690	16.556		16.623	1.917	3.77564729	15	23.629	23.382		23.505	-4.821	0.035370379
17	24.263	24.504		24.383	-5.843	0.01741591	17	23.012	22.881		22.947	-4.263	0.052097646
18	25.129	25.332		25.231	-6.691	0.00967999	18	24.008	24.147		24.077	-5.393	0.023792662
19	24.178	24.187		24.182	-5.642	0.02002247	19	23.488	23.338		23.413	-4.729	0.037696215
20	22.672	22.517		22.595	-4.055	0.06017423	20	22.190	22.161		22.176	-3.492	0.088897059
21	24.973	25.147		25.060	-6.520	0.01089851	21	23.662	24.192		23.927	-5.243	0.026403107
22	24.283	24.507		24.395	-5.855	0.01727594	22	23.606	23.445		23.526	-4.842	0.034875616
23	24.454	24.428		24.441	-5.901	0.01673623	23	22.913	22.969		22.941	-4.257	0.052300498
25	24.219	24.401		24.310	-5.770	0.01833048	25	23.268	23.276		23.272	-4.588	0.041581324
26	16.267	16.162		16.215	2.325	5.01226086	26	15.727	15.330		15.528	3.156	8.910543921
27	16.103	16.245		16.174	2.366	5.1555041	27	14.855	14.911		14.883	3.801	13.94030286
28	23.767	23.690		23.728	-5.188	0.02742552	28	24.287	24.379		24.333	-5.649	0.01992589
30	25.658	25.597		25.627	-7.087	0.00735407	30	24.524	24.540		24.532	-5.848	0.017361603
31	23.397	23.333		23.365	-4.825	0.03527756	31	22.599	22.627		22.613	-3.929	0.065660566
34	23.612	23.599		23.606	-5.066	0.02985729	34	22.284	22.336		22.310	-3.626	0.080979975
35	22.435	22.271		22.353	-3.813	0.07113397	35	21.727	21.616		21.672	-2.988	0.126062318
36	26.557	26.559		26.558	-8.018	0.00385773	36	24.832	25.144		24.988	-6.304	0.012652954
37	26.907	26.195		26.551	-8.011	0.00387553	37	27.394	27.480		27.437	-8.753	0.002317417
38	26.652	26.517		26.584	-8.044	0.00378796	38	25.736	25.605		25.670	-6.986	0.00788825
39	23.697	23.786		23.742	-5.202	0.02717592	39	23.717	23.450		23.583	-4.899	0.033508316
40	36.477	33.029		34.753	-16.213	1.3164E-05	40	33.281	32.784		33.032	-14.348	4.79425E-05
41	22.938	22.942		22.940	-4.400	0.0473709	41	21.955	21.904		21.930	-3.246	0.105433117
43	24.968	24.941		24.955	-6.415	0.01172163	43	24.499	24.229		24.364	-5.680	0.019504712
44	18.157	18.443		18.300	0.240	1.1811085	44	15.666	15.715		15.690	2.994	7.965185716
45	22.645	22.349		22.497	-3.957	0.06439705	45	22.773	22.383		22.578	-3.894	0.067251045
46	23.579	23.260		23.420	-4.880	0.03397005	46	22.529	22.701		22.615	-3.931	0.065559197
47	21.871	22.080		21.976	-3.436	0.09242747	47	20.943	20.911		20.927	-2.243	0.211282327
49	23.107	22.934		23.020	-4.480	0.04479925	49	23.123	22.874		22.998	-4.314	0.050258572
50	23.819	23.620		23.720	-5.180	0.0275888	50	23.013	23.250		23.132	-4.448	0.045827821
51	23.346	23.387		23.366	-4.826	0.0352453	51	22.190	22.438		22.314	-3.630	0.08078059
52	23.725	23.533		23.629	-5.089	0.02938627	52	22.691	22.741		22.716	-4.032	0.061130937
53	23.437	23.592		23.514	-4.974	0.03181477	53	21.860	21.756		21.808	-3.124	0.114720411
54	21.957	22.223		22.090	-3.550	0.08539151	54	21.719	21.690		21.704	-3.020	0.123239531
55	23.965	23.801		23.883	-5.343	0.02464194	55	23.700	23.679		23.690	-5.006	0.031129042
56	26.237	26.480		26.358	-7.818	0.00443059	56	24.729	24.502		24.616	-5.932	0.016384398
57	20.691	20.927		20.809	-2.269	0.20751809	57	18.948	19.225		19.087	-0.403	0.756545628
58	24.425	24.521		24.473	-5.933	0.01636943	58	23.853	23.490		23.672	-4.988	0.031519523
var2csa	22.328	22.206		22.267	-3.727	0.07552602	*var2csa*	19.965	19.889		19.927	-1.243	0.422417497
91	26.209	26.117		26.163	-7.623	0.00507288	91	24.442	24.514		24.478	-5.794	0.018027829
92	15.663	16.147		15.905	2.635	6.21299331	92	14.547	14.354		14.450	4.234	18.81500151
93	21.890	21.828		21.859	-3.319	0.10022168	93	20.276	19.899		20.088	-1.404	0.377968131
94	18.216	18.408		18.312	0.228	1.17114646	94	15.929	15.998		15.963	2.721	6.592798555
95	24.678	24.150		24.414	-5.874	0.01705332	95	23.353	23.556		23.454	-4.770	0.03663923
96	22.480	22.390		22.435	-3.895	0.06722066	96	20.439	20.183		20.311	-1.627	0.323735504
97	32.190	34.130		33.160	-14.620	3.9714E-05	97	Undetermine	33.747		33.747	-15.063	2.92175E-05
60	18.490	18.589		18.540	0.000	1.00026586	60	18.809	18.558		18.684	0.000	1.000331033

PfSAMS KD3 Day 0							PfSAMS KD1 OFF GlcN 2Wk						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	Undetermine	33.551		33.551	-14.135	5.55895E-05	1	32.099	34.524		33.312	-14.655	3.87689E-05
3	25.017	24.987		25.002	-5.586	0.020813336	3	23.327	23.259		23.293	-4.636	0.040226541
4	17.162	17.288		17.225	2.191	4.566904116	4	15.998	15.571		15.785	2.872	7.322424942
5	25.637	25.421		25.529	-6.113	0.014447403	5	23.702	23.774		23.738	-5.081	0.029547738
6	23.178	23.155		23.166	-3.750	0.07431303	6	22.166	21.931		22.048	-3.391	0.095303169
7	21.359	21.364		21.361	-1.945	0.259653126	7	20.176	19.737		19.957	-1.300	0.406164344
8	21.474	21.394		21.434	-2.018	0.24683964	8	19.887	19.629		19.758	-1.101	0.466157699
9	23.802	23.988		23.895	-4.479	0.044845934	9	23.287	23.232		23.260	-4.603	0.041157838
11	25.517	25.392		25.454	-6.038	0.01521677	11	23.895	23.831		23.863	-5.206	0.027094221
12	24.587	24.238		24.412	-4.996	0.031333386	12	22.108	21.892		22.000	-3.343	0.098548792
13	20.278	20.495		20.386	-0.970	0.51038665	13	18.865	18.828		18.847	-0.190	0.876681677
15	24.266	23.896		24.081	-4.665	0.0394109	15	16.299	16.145		16.222	2.435	5.407602101
17	23.371	23.382		23.376	-3.960	0.064242863	17	22.392	22.479		22.436	-3.779	0.072850693
18	24.204	24.594		24.399	-4.983	0.031625158	18	23.703	21.942		22.823	-4.166	0.055720596
19	23.672	23.121		23.397	-3.981	0.063347873	19	22.976	22.728		22.852	-4.195	0.054603699
20	23.769	23.832		23.801	-4.385	0.047866263	20	20.515	20.424		20.470	-1.813	0.28468374
21	24.447	24.643		24.545	-5.129	0.028583433	21	23.346	22.860		23.103	-4.446	0.045874222
22	24.694	24.664		24.679	-5.263	0.026038813	22	22.820	22.721		22.771	-4.114	0.057768126
23	23.764	23.812		23.788	-4.372	0.048306244	23	22.745	22.722		22.734	-4.077	0.059265815
25	24.641	24.322		24.482	-5.066	0.029858272	25	21.767	21.679		21.723	-3.066	0.1194429
26	16.820	16.956		16.888	2.528	5.768135278	26	15.321	15.182		15.251	3.406	10.59649876
27	16.432	16.561		16.497	2.919	7.565615723	27	14.877	14.874		14.876	3.781	13.75063654
28	23.907	24.379		24.143	-4.727	0.037759985	28	22.637	22.770		22.703	-4.046	0.060523847
30	24.525	24.706		24.616	-5.200	0.02721272	30	23.928	23.623		23.776	-5.119	0.02878482
31	24.009	24.124		24.067	-4.651	0.039813776	31	21.529	21.513		21.521	-2.864	0.137366965
34	23.504	23.579		23.542	-4.126	0.057279472	34	21.690	21.622		21.656	-2.999	0.125080084
35	24.478	24.345		24.411	-4.995	0.031348582	35	20.327	20.268		20.297	-1.640	0.320806438
36	25.723	25.535		25.629	-6.213	0.013479936	36	24.694	24.623		24.659	-6.002	0.015604895
37	27.492	27.304		27.398	-7.982	0.0039551	37	24.908	24.651		24.779	-6.122	0.014353163
38	25.658	25.532		25.595	-6.179	0.013805591	38	24.996	24.743		24.870	-6.213	0.013481899
39	24.859	24.517		24.688	-5.272	0.025877903	39	22.396	22.187		22.291	-3.634	0.080537277
40	38.686	37.781		38.234	-18.818	2.16438E-06	40	35.536	31.820		33.678	-15.021	3.00779E-05
41	23.941	23.946		23.944	-4.528	0.043351914	41	20.877	20.543		20.710	-2.053	0.241015611
43	24.411	24.324		24.367	-4.951	0.032324918	43	23.777	23.545		23.661	-5.004	0.031166739
44	18.334	18.278		18.306	1.110	2.159004874	44	16.260	15.968		16.114	2.543	5.828382256
45	24.269	24.011		24.140	-4.724	0.03784921	45	21.127	20.987		21.057	-2.400	0.189438636
46	24.014	23.649		23.831	-4.415	0.046863467	46	21.642	21.538		21.590	-2.933	0.130955858
47	22.133	22.507		22.320	-2.904	0.133572693	47	21.497	21.214		21.356	-2.699	0.154006698
49	23.522	23.536		23.529	-4.113	0.05778813	49	20.991	20.714		20.853	-2.196	0.218287133
50	24.578	24.453		24.515	-5.099	0.029168368	50	21.801	21.617		21.709	-3.052	0.120555594
51	21.437	21.519		21.478	-2.062	0.23951958	51	21.619	21.378		21.498	-2.841	0.13954874
52	24.627	24.574		24.600	-5.184	0.027499477	52	22.155	21.942		22.048	-3.391	0.095309214
53	23.392	23.464		23.428	-4.012	0.061986785	53	20.902	20.707		20.805	-2.148	0.225664674
54	21.586	21.842		21.714	-2.298	0.20329174	54	20.997	20.730		20.864	-2.207	0.216652532
55	23.534	23.976		23.755	-4.339	0.049405398	55	21.833	21.732		21.783	-3.126	0.114567202
56	25.349	24.363		24.856	-5.440	0.023036039	56	24.772	24.521		24.646	-5.989	0.015740362
57	22.180	22.124		22.152	-2.736	0.150123527	57	19.683	19.349		19.516	-0.859	0.551432995
58	25.701	25.568		25.634	-6.218	0.013429938	58	23.989	23.997		23.993	-5.336	0.02476198
var2csa	21.933	23.285		22.609	-3.193	0.109356395	var2csa	20.117	19.792		19.955	-1.298	0.40678616
91	25.522	25.364		25.443	-6.027	0.015336914	91	24.367	23.889		24.128	-5.471	0.022542971
92	16.527	16.753		16.640	2.776	6.848739563	92	14.648	14.652		14.650	4.007	16.08284242
93	21.645	21.462		21.553	-2.137	0.227326141	93	20.125	19.815		19.970	-1.313	0.402473165
94	18.716	18.615		18.665	0.751	1.682437603	94	16.336	16.131		16.233	2.424	5.365189023
95	25.098	24.928		25.013	-5.597	0.020660064	95	22.579	22.437		22.508	-3.851	0.069295477
96	23.397	23.352		23.374	-3.958	0.064333077	96	21.722	21.504		21.613	-2.956	0.128894222
97	Undetermine	Undetermined		#DIV/0!	#DIV/0!	0	97	Undetermine	Undetermined		#DIV/0!	#DIV/0!	0
60	19.395	19.436		19.416	0.000	1.000203806	60	18.755	18.558		18.657	0.000	1.000217672

PfSAMS KD1 ON GlcN 2Wk							PfSAMS KD2 OFF GlcN 2Wk						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	35.166	35.599		35.382	-16.121	1.40276E-05	1	33.012	Undetermined		33.012	-13.599	8.05755E-05
3	22.905	22.489		22.697	-3.436	0.092390159	3	23.360	23.230		23.295	-3.882	0.067825096
4	16.127	15.793		15.960	3.301	9.856512346	4	15.646	15.573		15.609	3.804	13.9645773
5	23.985	23.780		23.882	-4.621	0.040630798	5	24.286	24.269		24.277	-4.864	0.034334303
6	21.759	21.697		21.728	-2.467	0.18089739	6	22.911	23.116		23.014	-3.601	0.082430297
7	19.920	19.831		19.876	-0.615	0.653080845	7	21.266	20.936		21.101	-1.688	0.310402052
8	19.764	19.703		19.734	-0.473	0.720562511	8	20.651	20.232		20.441	-1.028	0.490232658
9	23.316	23.243		23.279	-4.018	0.061715385	9	23.265	23.702		23.483	-4.070	0.059526752
11	22.230	22.932		22.581	-3.320	0.100132589	11	24.516	24.263		24.389	-4.976	0.031771312
12	19.922	20.181		20.052	-0.791	0.577979408	12	23.135	23.298		23.216	-3.803	0.071627579
13	17.743	17.601		17.672	1.589	3.008316795	13	19.627	19.539		19.583	-0.170	0.888592888
15	15.823	17.342		16.582	2.679	6.403388312	15	23.518	23.223		23.370	-3.957	0.064369385
17	22.586	21.858		22.222	-2.961	0.128404574	17	22.750	22.605		22.677	-3.264	0.104072081
18	22.837	22.523		22.680	-3.419	0.093483561	18	23.875	23.819		23.847	-4.434	0.046258891
19	22.811	22.557		22.684	-3.423	0.093250224	19	23.449	23.200		23.325	-3.912	0.066453684
20	20.821	20.772		20.796	-1.535	0.345023133	20	21.944	21.745		21.845	-2.432	0.185329764
21	23.332	23.251		23.291	-4.030	0.061208974	21	23.699	23.508		23.604	-4.191	0.054766709
22	22.656	22.416		22.536	-3.275	0.10329648	22	23.946	23.826		23.886	-4.473	0.045025793
23	22.016	22.245		22.131	-2.870	0.136815776	23	23.235	22.972		23.103	-3.690	0.077456842
25	21.574	21.464		21.519	-2.258	0.209098628	25	22.518	22.685		22.602	-3.189	0.109688486
26	15.494	15.506		15.500	3.761	13.55855475	26	15.645	15.362		15.504	3.909	15.02615516
27	14.832	14.805		14.819	4.442	21.73761869	27	15.366	15.326		15.346	4.067	16.7617834
28	21.993	21.699		21.846	-2.585	0.166644083	28	23.870	23.742		23.806	-4.393	0.047594049
30	23.230	22.671		22.951	-3.690	0.07750687	30	24.699	24.433		24.566	-5.153	0.028101257
31	20.631	20.416		20.523	-1.262	0.416829272	31	22.557	22.562		22.559	-3.146	0.112933703
34	20.622	20.378		20.500	-1.239	0.423764019	34	21.839	21.794		21.817	-2.404	0.188959637
35	20.375	20.002		20.188	-0.927	0.525852649	35	21.437	21.282		21.360	-1.947	0.259426813
36	23.800	23.859		23.830	-4.569	0.042143502	36	25.292	24.896		25.094	-5.681	0.019490015
37	25.757	26.114		25.936	-6.675	0.009789779	37	26.752	26.538		26.645	-7.232	0.006651927
38	24.199	24.150		24.175	-4.914	0.033173556	38	25.321	25.294		25.308	-5.895	0.016809534
39	21.541	21.424		21.482	-2.221	0.214421652	39	23.498	23.470		23.484	-4.071	0.059496289
40	32.656	31.439		32.047	-12.786	0.000141559	40	32.418	37.232		34.825	-15.412	2.29395E-05
41	20.787	20.245		20.516	-1.255	0.419000102	41	22.367	22.138		22.253	-2.840	0.139702331
43	23.448	22.892		23.170	-3.909	0.066554868	43	24.240	24.323		24.281	-4.868	0.03423765
44	16.651	16.300		16.476	2.785	6.894032023	44	16.513	15.977		16.245	3.168	8.987625432
45	20.934	20.769		20.852	-1.591	0.331991682	45	21.994	21.684		21.839	-2.426	0.186078077
46	21.237	21.134		21.186	-1.925	0.263384162	46	22.376	22.343		22.360	-2.947	0.129711473
47	21.079	20.864		20.971	-1.710	0.305600646	47	21.293	21.290		21.291	-1.878	0.272051653
49	20.548	20.578		20.563	-1.302	0.405585646	49	22.466	21.946		22.206	-2.793	0.144285477
50	21.435	21.368		21.401	-2.140	0.226802744	50	22.687	22.515		22.601	-3.188	0.109698028
51	20.447	20.007		20.227	-0.966	0.51193995	51	22.433	22.315		22.374	-2.961	0.128443697
52	21.423	21.345		21.384	-2.123	0.229519081	52	22.566	22.372		22.469	-3.056	0.120243724
53	20.814	20.775		20.795	-1.534	0.345413171	53	22.155	21.912		22.033	-2.620	0.162612154
54	19.807	19.563		19.685	-0.424	0.745164061	54	21.674	21.447		21.561	-2.148	0.225692127
55	21.003	20.967		20.985	-1.724	0.3026811	55	23.470	22.958		23.214	-3.801	0.071751933
56	24.172	23.994		24.083	-4.822	0.035349571	56	24.734	24.716		24.725	-5.312	0.025169328
57	19.366	18.938		19.152	0.109	1.078695746	57	19.709	19.530		19.620	-0.207	0.866428738
58	24.011	23.817		23.914	-4.653	0.039749045	58	24.391	23.791		24.091	-4.678	0.03905623
var2csa	20.355	20.175		20.265	-1.004	0.498735395	var2csa	19.680	19.486		19.583	-0.170	0.888871022
91	23.606	23.397		23.501	-4.240	0.052908708	91	24.717	24.588		24.652	-5.239	0.026471321
92	14.932	14.714		14.823	4.438	21.6710252	92	15.633	15.674		15.654	3.759	13.53876746
93	19.853	19.608		19.730	-0.469	0.72226767	93	20.194	19.904		20.049	-0.636	0.643583837
94	16.681	16.323		16.502	2.759	6.768363258	94	16.776	16.653		16.715	2.698	6.490780639
95	21.267	21.192		21.229	-1.968	0.255531314	95	23.401	23.400		23.400	-3.987	0.063050735
96	21.107	20.905		21.006	-1.745	0.298301575	96	21.478	21.354		21.416	-2.003	0.249458719
97	38.693	36.419		37.556	-18.295	3.10896E-06	97	Undetermine	38.062		38.062	-18.649	2.43243E-06
60	19.333	19.189		19.261	0.000	1.000038817	60	19.552	19.273		19.413	0.000	1.000312658

PfSAMS KD2 ON GlcN 2Wk							PfSAMS KD3 OFF GlcN 2Wk					
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT		CT Average	relative to p60	relative copy number
1	Undetermine	39.563		39.563	-19.767	1.1208E-06	1	32.216	Undetermined	32.216	-13.580	8.16839E-05
3	22.612	22.606		22.609	-2.813	0.14232944	3	22.577	22.399	22.488	-3.852	0.069264671
4	15.758	15.638		15.698	4.098	17.1280677	4	15.155	14.774	14.965	3.671	12.73985101
5	23.559	23.560		23.559	-3.763	0.07363398	5	23.565	23.477	23.521	-4.885	0.033849997
6	22.186	21.995		22.090	-2.294	0.20383992	6	21.775	21.799	21.787	-3.151	0.112582032
7	20.001	19.767		19.884	-0.088	0.94108308	7	19.550	19.473	19.511	-0.875	0.545231946
8	19.903	19.940		19.921	-0.125	0.91671298	8	19.695	19.763	19.729	-1.093	0.468783436
9	22.626	22.511		22.569	-2.773	0.14633888	9	21.961	21.617	21.789	-3.153	0.112403978
11	23.400	23.184		23.292	-3.496	0.08863265	11	23.820	23.808	23.814	-5.178	0.027615633
12	21.425	21.140		21.282	-1.486	0.35687902	12	22.634	22.333	22.484	-3.848	0.069455993
13	19.377	19.313		19.345	0.451	1.36733524	13	18.911	19.248	19.080	-0.444	0.735288079
15	23.428	23.367		23.397	-3.601	0.08238726	15	22.894	23.046	22.970	-4.334	0.049581784
17	22.127	22.108		22.118	-2.322	0.20003128	17	21.500	21.550	21.525	-2.889	0.135030573
18	23.214	22.828		23.021	-3.225	0.10695839	18	23.529	23.363	23.446	-4.810	0.035650349
19	23.003	22.847		22.925	-3.129	0.1143098	19	22.424	22.420	22.422	-3.786	0.072504357
20	20.759	20.637		20.698	-0.902	0.5352319	20	21.509	21.285	21.397	-2.761	0.147519748
21	23.413	23.354		23.384	-3.588	0.08316069	21	22.407	22.500	22.454	-3.818	0.070925833
22	23.220	22.997		23.108	-3.312	0.10065994	22	23.116	22.934	23.025	-4.389	0.04772629
23	22.317	22.264		22.291	-2.495	0.17741741	23	22.012	22.140	22.076	-3.440	0.092143103
25	21.485	21.304		21.395	-1.599	0.33021636	25	21.642	21.528	21.585	-2.949	0.129473792
26	15.853	15.709		15.781	4.015	16.1664779	26	15.599	15.317	15.458	3.178	9.050597257
27	14.792	14.658		14.725	5.071	33.6099851	27	14.589	14.389	14.489	4.147	17.71465545
28	23.158	22.996		23.077	-3.281	0.10289277	28	23.169	23.287	23.228	-4.592	0.041454715
30	23.623	23.604		23.614	-3.818	0.07091347	30	23.155	23.094	23.125	-4.489	0.044542875
31	21.119	20.895		21.007	-1.211	0.43209981	31	21.508	21.384	21.446	-2.810	0.142586717
34	20.777	20.589		20.683	-0.887	0.54072464	34	21.578	21.298	21.438	-2.802	0.143371135
35	20.146	20.130		20.138	-0.342	0.78893563	35	21.333	20.854	21.094	-2.458	0.182059509
36	23.955	23.900		23.928	-4.132	0.05703685	36	24.103	23.993	24.048	-5.412	0.02348374
37	25.712	25.352		25.532	-5.736	0.01876326	37	24.505	24.300	24.403	-5.767	0.018366913
38	24.691	24.405		24.548	-4.752	0.0371044	38	24.104	23.967	24.036	-5.400	0.023688251
39	21.737	21.812		21.775	-1.979	0.25370423	39	22.716	22.592	22.654	-4.018	0.061716219
40	33.401	32.242		32.821	-13.025	0.00011993	40	35.676	31.649	33.663	-15.027	2.99584E-05
41	20.800	20.724		20.762	-0.966	0.51207428	41	21.828	21.432	21.630	-2.994	0.125550136
43	23.727	23.512		23.620	-3.824	0.07061603	43	22.942	22.810	22.876	-4.240	0.052914741
44	15.890	15.340		15.615	4.181	18.1400056	44	15.845	15.698	15.771	2.865	7.284116517
45	21.602	21.499		21.550	-1.754	0.29637671	45	21.812	21.749	21.781	-3.145	0.113066778
46	21.189	20.985		21.087	-1.291	0.40867418	46	21.922	21.738	21.830	-3.194	0.10926642
47	21.022	20.977		21.000	-1.204	0.4341324	47	20.897	20.587	20.742	-2.106	0.232343228
49	21.371	21.354		21.362	-1.566	0.33763515	49	20.857	20.789	20.823	-2.187	0.219580295
50	21.623	21.396		21.510	-1.714	0.30488752	50	21.708	22.183	21.945	-3.309	0.100869855
51	20.714	20.267		20.491	-0.695	0.61783393	51	19.842	19.479	19.660	-1.024	0.491621188
52	21.761	21.700		21.731	-1.935	0.26157064	52	22.520	22.151	22.335	-3.699	0.076981442
53	21.353	21.363		21.358	-1.562	0.33867655	53	20.918	20.700	20.809	-2.173	0.221763756
54	20.209	20.014		20.112	-0.316	0.80356827	54	19.865	19.758	19.812	-1.176	0.442710194
55	21.533	22.113		21.823	-2.027	0.24537837	55	21.293	21.277	21.285	-2.649	0.159406405
56	23.709	21.830		22.770	-2.974	0.12731233	56	23.483	23.882	23.682	-5.046	0.030267147
57	18.785	18.655		18.720	1.076	2.10769337	57	20.304	20.290	20.297	-1.661	0.31624871
58	23.548	23.378		23.463	-3.667	0.07872041	58	23.395	23.271	23.333	-4.697	0.038553456
var2csa	18.373	18.197		18.285	1.511	2.84970978	var2csa	20.501	20.241	20.371	-1.735	0.300410158
91	23.947	23.811		23.879	-4.083	0.05900295	91	23.652	23.441	23.547	-4.911	0.033249616
92	15.428	15.228		15.328	4.468	22.1287798	92	14.785	14.751	14.768	3.868	14.60131096
93	19.992	20.003		19.998	-0.202	0.86957478	93	19.577	19.244	19.411	-0.775	0.584442298
94	15.810	15.894		15.852	3.944	15.3920235	94	16.373	16.239	16.306	2.330	5.026992372
95	21.538	21.638		21.588	-1.792	0.28873692	95	22.381	22.475	22.428	-3.792	0.072181703
96	20.286	19.805		20.046	-0.250	0.84113746	96	21.732	21.820	21.776	-3.140	0.113423029
97	Undetermine	37.597		37.597	-17.801	4.3776E-06	97	Undetermine	37.261	37.261	-18.625	2.47418E-06
60	19.878	19.714		19.796	0.000	1.00032722	60	18.740	18.531	18.636	0.000	1.000231538

PfSAMS KD3 ON GlcN 2Wk							PfSAMS KD1 OFF GlcN 4Wk						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	33.935	30.746		32.340	-13.109	0.00011315	1	33.106	Undetermined		33.106	-10.012	0.00096847
3	22.465	22.114		22.290	-3.059	0.12000284	3	28.601	28.215		28.408	-5.314	0.02514078
4	15.754	15.551		15.653	3.578	11.9453482	4	19.597	19.345		19.471	3.623	12.3214403
5	23.125	23.005		23.065	-3.834	0.07011593	5	28.801	29.161		28.981	-5.887	0.01689712
6	21.551	21.300		21.425	-2.194	0.21847975	6	26.663	26.184		26.424	-3.330	0.09946608
7	19.516	19.342		19.429	-0.198	0.87179117	7	22.626	22.469		22.548	0.546	1.46049103
8	19.319	19.347		19.333	-0.102	0.93191159	8	22.784	22.557		22.670	0.424	1.34134095
9	21.571	21.561		21.566	-2.335	0.19821286	9	26.743	26.542		26.642	-3.548	0.0854722
11	21.877	21.656		21.766	-2.535	0.17249064	11	28.468	28.607		28.537	-5.443	0.02298279
12	20.112	20.135		20.123	-0.892	0.538793	12	25.174	25.262		25.218	-2.124	0.22943494
13	18.291	18.218		18.254	0.977	1.96804881	13	22.234	21.942		22.088	1.006	2.00850841
15	22.288	22.099		22.194	-2.963	0.12828897	15	20.154	19.892		20.023	3.071	8.40331171
17	21.180	21.119		21.150	-1.919	0.26451556	17	25.763	25.764		25.764	-2.670	0.15716458
18	22.509	22.281		22.395	-3.164	0.11154912	18	26.805	27.255		27.030	-3.936	0.06534699
19	22.158	21.835		21.997	-2.766	0.14705538	19	26.193	25.715		25.954	-2.860	0.137757
20	20.546	20.361		20.453	-1.222	0.42859294	20	25.298	24.944		25.121	-2.027	0.24537939
21	22.641	22.398		22.520	-3.289	0.1023301	21	26.799	26.450		26.624	-3.530	0.08655029
22	22.748	22.402		22.575	-3.344	0.09849406	22	26.730	26.763		26.747	-3.653	0.07950831
23	21.529	21.418		21.473	-2.242	0.21132949	23	26.679	26.484		26.582	-3.488	0.08915651
25	20.933	20.750		20.841	-1.610	0.3274997	25	28.766	27.777		28.271	-5.177	0.02763601
26	15.657	15.514		15.585	3.646	12.5148188	26	20.134	19.804		19.969	3.125	8.72456378
27	14.800	14.859		14.829	4.402	21.1356635	27	17.797	17.813		17.805	5.289	39.0988969
28	21.222	21.369		21.296	-2.065	0.23904349	28	25.661	25.928		25.795	-2.701	0.15378994
30	22.357	22.339		22.348	-3.117	0.11527815	30	29.104	28.582		28.843	-5.749	0.01859981
31	19.568	19.589		19.579	-0.348	0.78588357	31	27.287	27.406		27.347	-4.253	0.05245267
34	19.635	19.670		19.652	-0.421	0.74667302	34	26.970	26.592		26.781	-3.687	0.07763174
35	20.280	20.144		20.212	-0.981	0.50663424	35	25.365	25.340		25.352	-2.258	0.20903371
36	22.952	22.776		22.864	-3.633	0.08057932	36	29.564	29.543		29.553	-6.459	0.01136413
37	25.434	25.356		25.395	-6.164	0.01394387	37	30.672	29.699		30.186	-7.092	0.0073323
38	23.392	23.275		23.334	-4.103	0.05821208	38	29.638	28.682		29.160	-6.066	0.01492245
39	21.464	21.163		21.313	-2.082	0.23613471	39	26.721	26.238		26.479	-3.385	0.0956963
40	32.811	32.113		32.462	-13.231	0.000104	40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	0
41	20.679	20.394		20.536	-1.305	0.40458816	41	23.923	23.805		23.864	-0.770	0.58643861
43	22.796	22.515		22.655	-3.424	0.09315665	43	28.151	28.305		28.228	-5.134	0.02847494
44	16.262	16.261		16.261	2.970	7.83298122	44	20.258	19.880		20.069	3.025	8.13747102
45	20.834	20.870		20.852	-1.621	0.32507105	45	25.832	25.862		25.847	-2.753	0.14835711
46	20.450	20.919		20.684	-1.453	0.36515667	46	24.724	24.748		24.736	-1.642	0.3203427
47	20.914	21.050		20.982	-1.751	0.29711967	47	25.367	25.077		25.222	-2.128	0.22878336
49	19.824	19.810		19.817	-0.586	0.66627677	49	26.319	25.881		26.100	-3.006	0.12448247
50	21.147	20.842		20.994	-1.763	0.2945641	50	25.850	25.767		25.809	-2.715	0.15235112
51	18.968	18.813		18.890	0.341	1.26620632	51	25.946	25.718		25.832	-2.738	0.14990023
52	20.977	20.805		20.891	-1.660	0.3164341	52	26.518	25.884		26.201	-3.107	0.11607929
53	20.462	20.466		20.464	-1.233	0.42532955	53	26.817	26.643		26.730	-3.636	0.08043343
54	18.612	18.600		18.606	0.625	1.54261816	54	24.557	24.308		24.433	-1.339	0.39536128
55	19.598	19.410		19.504	-0.273	0.82763955	55	28.646	28.287		28.466	-5.372	0.02413972
56	20.010	22.607		21.308	-2.077	0.23695106	56	27.964	27.769		27.866	-4.772	0.03659715
57	19.437	19.367		19.402	-0.171	0.8882388	57	24.110	23.794		23.952	-0.858	0.55167175
58	22.981	22.785		22.883	-3.652	0.07955244	58	26.413	25.911		26.162	-3.068	0.11924883
var2csa	20.109	20.161		20.135	-0.904	0.53438725	var2csa	21.428	21.171		21.299	1.795	3.46994309
91	22.841	22.724		22.782	-3.551	0.08529955	91	29.859	28.826		29.342	-6.248	0.01315313
92	15.529	15.489		15.509	3.722	13.1974793	92	20.147	19.911		20.029	3.065	8.36713957
93	19.753	19.363		19.558	-0.327	0.79720187	93	25.830	25.710		25.770	-2.676	0.15649283
94	16.474	16.250		16.362	2.869	7.30624939	94	21.554	21.399		21.477	1.617	3.06812125
95	20.373	20.134		20.253	-1.022	0.49230882	95	24.742	24.785		24.763	-1.669	0.3144161
96	20.958	20.769		20.863	-1.632	0.32252994	96	23.730	23.596		23.663	-0.569	0.67404018
97	39.365	Undetermined		39.365	-20.134	8.6896E-07	97	Undetermine	Undetermined		#DIV/0!	#DIV/0!	0
60	19.327	19.136		19.231	0.000	0.99978619	60	23.268	22.920		23.094	0.000	0.99976609

PfSAMS KD1 ON GlcN 4Wk							PfSAMS KD2 OFF GlcN 4Wk						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	33.575	30.475		32.025	-12.624	0.00015841	1	33.973	33.962		33.968	-9.487	0.00139402
3	25.700	25.405		25.552	-6.151	0.0140688	3	29.188	29.545		29.367	-4.886	0.03383124
4	18.400	17.714		18.057	1.344	2.53892451	4	20.393	20.318		20.356	4.125	17.4523874
5	25.617	25.154		25.386	-5.985	0.01579	5	30.117	31.689		30.903	-6.422	0.01166523
6	23.595	23.447		23.521	-4.120	0.05751177	6	27.785	27.790		27.788	-3.307	0.10107485
7	20.418	20.420		20.419	-1.018	0.4938184	7	24.379	24.601		24.490	-0.009	0.9937191
8	21.013	20.924		20.968	-1.567	0.33745043	8	24.773	24.495		24.634	-0.153	0.89928574
9	23.810	23.845		23.828	-4.427	0.04649789	9	29.552	29.443		29.498	-5.017	0.03088523
11	24.166	24.291		24.228	-4.827	0.0352215	11	29.960	29.334		29.647	-5.166	0.02785218
12	21.618	21.233		21.426	-2.025	0.24576655	12	27.486	27.749		27.617	-3.136	0.11373412
13	19.667	19.555		19.611	-0.210	0.86448779	13	23.952	23.712		23.832	0.649	1.56781029
15	18.140	18.153		18.146	1.255	2.38603053	15	28.646	28.299		28.473	-3.992	0.06286706
17	23.583	23.296		23.439	-4.038	0.06085843	17	26.680	27.325		27.003	-2.522	0.17415237
18	24.198	23.839		24.019	-4.618	0.04072818	18	28.278	28.780		28.529	-4.048	0.06045785
19	23.711	23.286		23.499	-4.098	0.05841026	19	26.751	26.592		26.672	-2.191	0.21907412
20	22.630	22.378		22.504	-3.103	0.11640404	20	27.297	27.144		27.220	-2.739	0.14974363
21	24.550	24.449		24.499	-5.098	0.02918979	21	28.133	28.599		28.366	-3.885	0.06768481
22	23.544	23.182		23.363	-3.962	0.06417906	22	27.932	27.857		27.895	-3.414	0.09383988
23	23.303	23.398		23.351	-3.950	0.06472115	23	27.846	27.002		27.424	-2.943	0.13003213
25	23.694	23.505		23.600	-4.199	0.05446239	25	29.520	29.223		29.372	-4.891	0.03371256
26	17.557	17.468		17.513	1.888	3.70216706	26	21.338	21.273		21.305	3.176	9.03674367
27	16.198	16.236		16.217	3.184	9.08771128	27	19.480	19.466		19.473	5.008	32.1815855
28	23.335	23.173		23.254	-3.853	0.06919802	28	27.826	27.669		27.748	-3.267	0.1039045
30	25.291	24.873		25.082	-5.681	0.01949095	30	29.709	30.391		30.050	-5.569	0.02105959
31	22.406	21.943		22.175	-2.774	0.14621427	31	28.124	27.723		27.923	-3.442	0.09200522
34	21.942	21.677		21.809	-2.408	0.18835262	34	27.312	26.994		27.153	-2.672	0.15691645
35	21.943	21.818		21.881	-2.480	0.17926168	35	27.461	27.701		27.581	-3.100	0.11664647
36	25.610	25.163		25.387	-5.986	0.01578231	36	29.687	30.965		30.326	-5.845	0.01740151
37	24.758	24.676		24.717	-5.316	0.02510222	37	33.647	33.262		33.454	-8.973	0.00198977
38	25.286	24.855		25.070	-5.669	0.01965033	38	30.281	29.655		29.968	-5.487	0.0222959
39	21.797	21.546		21.672	-2.271	0.20724274	39	28.160	28.280		28.220	-3.739	0.07490633
40	35.956	33.812		34.884	-15.483	2.183E-05	40	34.258	37.164		35.711	-11.230	0.00041621
41	21.420	21.310		21.365	-1.964	0.25627906	41	26.325	26.387		26.356	-1.875	0.27255806
43	23.913	23.927		23.920	-4.519	0.04362695	43	30.172	28.947		29.560	-5.079	0.02958807
44	17.841	17.718		17.779	1.622	3.07779576	44	22.139	21.897		22.018	2.463	5.51256209
45	23.333	23.013		23.173	-3.772	0.07320061	45	27.541	28.166		27.853	-3.372	0.09657389
46	21.695	21.543		21.619	-2.218	0.21488251	46	25.851	25.586		25.719	-1.238	0.42407123
47	22.628	22.448		22.538	-3.137	0.11366953	47	26.579	26.803		26.691	-2.210	0.21612435
49	22.899	22.921		22.910	-3.509	0.0878352	49	28.153	28.012		28.082	-3.601	0.08240345
50	23.185	23.172		23.179	-3.778	0.0729125	50	28.378	27.451		27.914	-3.433	0.09256977
51	21.357	21.125		21.241	-1.840	0.27929178	51	26.890	26.749		26.819	-2.338	0.19772758
52	23.345	22.913		23.129	-3.728	0.07546947	52	27.503	27.147		27.325	-2.844	0.139255
53	21.744	21.784		21.764	-2.363	0.19440335	53	28.254	28.504		28.379	-3.898	0.0670853
54	21.484	21.180		21.332	-1.931	0.26229469	54	26.118	25.635		25.876	-1.395	0.38013269
55	22.432	22.409		22.421	-3.020	0.12329558	55	28.198	28.635		28.416	-3.935	0.06536449
56	24.713	25.656		25.185	-5.784	0.01815264	56	26.008	28.301		27.154	-2.673	0.15674996
57	19.262	19.013		19.137	0.264	1.20038816	57	24.169	23.730		23.950	0.531	1.44503356
58	21.621	21.727		21.674	-2.273	0.20683991	58	26.708	26.182		26.445	-1.964	0.25632161
var2csa	20.401	20.264		20.332	-0.931	0.5243598	var2csa	22.390	22.294		22.342	2.139	4.40376658
91	23.737	23.513		23.625	-4.224	0.05350552	91	30.740	30.569		30.655	-6.174	0.0138527
92	16.485	16.338		16.411	2.990	7.943936	92	21.860	21.796		21.828	2.653	6.28868773
93	22.580	22.380		22.480	-3.079	0.11832899	93	27.643	27.663		27.653	-3.172	0.11095238
94	18.244	17.846		18.045	1.356	2.55958692	94	23.250	23.411		23.330	1.151	2.22028442
95	23.287	23.116		23.202	-3.801	0.07176449	95	26.984	27.201		27.093	-2.612	0.16362414
96	20.691	20.556		20.623	-1.222	0.42857215	96	24.588	24.379		24.484	-0.003	0.99814028
97	36.476	Undetermined		36.476	-17.075	7.2437E-06	97	Undetermine	Undetermined		#DIV/0!	#DIV/0!	0
60	19.479	19.324		19.401	0.000	0.99969541	60	24.588	24.373		24.481	0.000	1.00015493

PfSAMS KD2 ON GlcN 4Wk							PfSAMS KD3 OFF GlcN 4Wk						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	32.975	34.275		33.625	-13.716	7.43148E-05	1	Undetermine	Undetermined		0	0	0
3	25.934	25.476		25.705	-5.796	0.018000216	3	27.665	27.558		27.612	-7.160851	0.00698826
4	18.221	17.886		18.054	1.855	3.618562023	4	20.236	19.978		20.107	0.3437525	1.26905316
5	26.006	25.915		25.960	-6.051	0.015078639	5	27.891	27.704		27.797	-7.346378	0.00614497
6	24.240	23.894		24.067	-4.158	0.056009098	6	25.859	25.585		25.722	-5.2705615	0.02590615
7	20.769	20.586		20.678	-0.769	0.587010616	7	21.425	21.373		21.399	-0.9482855	0.51824798
8	22.202	22.001		22.101	-2.192	0.218811808	8	22.338	22.262		22.300	-1.849038	0.2775774
9	24.398	24.156		24.277	-4.368	0.048431198	9	26.141	26.347		26.244	-5.7932015	0.01803319
11	24.794	24.584		24.689	-4.780	0.036399237	11	27.108	27.317		27.213	-6.7615245	0.00921676
12	22.246	21.970		22.108	-2.199	0.217833916	12	25.979	25.895		25.937	-5.4862305	0.022309
13	20.922	20.734		20.828	-0.919	0.528857703	13	22.936	22.915		22.925	-2.474309	0.17995287
15	25.099	24.920		25.010	-5.101	0.02913937	15	25.072	25.139		25.106	-4.6545155	0.03970555
17	23.390	23.286		23.338	-3.429	0.092826659	17	25.304	25.334		25.319	-4.8677725	0.03424952
18	24.831	24.604		24.718	-4.809	0.035679631	18	27.310	26.973		27.141	-6.690205	0.00968384
19	23.985	23.841		23.913	-4.004	0.062324361	19	26.287	25.976		26.132	-5.6806625	0.01949621
20	23.573	23.524		23.548	-3.639	0.080242925	20	25.105	24.953		25.029	-4.577941	0.04186995
21	24.810	24.760		24.785	-4.876	0.034053715	21	26.836	26.444		26.640	-6.189216	0.01370441
22	24.308	23.862		24.085	-4.176	0.055325811	22	23.457	22.912		23.185	-2.733577	0.15035273
23	24.350	23.967		24.159	-4.250	0.05256681	23	26.358	26.175		26.267	-5.815906	0.01775161
25	24.447	24.144		24.295	-4.386	0.047823516	25	25.996	25.786		25.891	-5.440262	0.02303127
26	18.428	18.260		18.344	1.565	2.958836545	26	19.217	18.839		19.028	1.4234005	2.68216966
27	17.301	17.198		17.250	2.659	6.317216429	27	17.958	17.677		17.818	2.6331935	6.2039777
28	23.266	23.013		23.139	-3.230	0.106572639	28	26.188	25.846		26.017	-5.5661045	0.02110744
30	25.821	25.653		25.737	-5.828	0.017604432	30	28.258	28.113		28.185	-7.7343835	0.0046959
31	22.684	22.629		22.657	-2.748	0.148893224	31	24.411	24.287		24.349	-3.897986	0.06707942
34	22.262	22.118		22.190	-2.281	0.205754872	34	24.660	24.619		24.640	-4.1887225	0.05483639
35	22.787	22.619		22.703	-2.794	0.144185801	35	24.894	24.662		24.778	-4.327144	0.04981956
36	25.846	25.912		25.879	-5.970	0.015954907	36	29.005	29.134		29.069	-8.618446	0.00254442
37	27.599	27.478		27.538	-7.629	0.005050056	37	24.852	24.841		24.847	-4.3955975	0.04751091
38	26.111	25.919		26.015	-6.106	0.014519424	38	27.984	28.155		28.069	-7.6182165	0.00508965
39	22.753	22.559		22.656	-2.747	0.148960425	39	26.232	26.318		26.275	-5.824349	0.01764803
40	Undetermine	34.109		34.109	-14.200	5.31302E-05	40	32.950	Undetermined		32.950	-12.49866	0.00017279
41	22.933	22.553		22.743	-2.834	0.14024731	41	24.319	23.962		24.141	-3.68962	0.07750214
43	25.505	25.269		25.387	-5.478	0.022437045	43	27.584	26.694		27.139	-6.687815	0.0096999
44	18.915	18.547		18.731	1.178	2.262548158	44	20.263	20.002		20.132	0.3186735	1.24718329
45	24.248	23.913		24.080	-4.171	0.05550042	45	25.177	25.141		25.159	-4.7081655	0.03825612
46	21.840	21.831		21.835	-1.926	0.263120398	46	24.763	24.699		24.731	-4.2800345	0.05147321
47	23.493	23.383		23.438	-3.529	0.086641018	47	26.620	26.521		26.571	-6.1199055	0.01437887
49	23.721	23.653		23.687	-3.778	0.072885341	49	24.218	24.018		24.118	-3.6669885	0.0787275
50	23.915	23.920		23.918	-4.009	0.06213231	50	25.500	25.721		25.611	-5.1595275	0.0279787
51	21.617	21.452		21.535	-1.626	0.324089009	51	24.783	24.372		24.577	-4.1260715	0.0572702
52	23.736	23.631		23.683	-3.774	0.073077419	52	25.725	25.697		25.711	-5.2596325	0.02610315
53	23.836	24.087		23.961	-4.052	0.060268167	53	24.342	24.239		24.290	-3.8394025	0.06985937
54	21.494	21.584		21.539	-1.630	0.323106908	54	23.116	22.867		22.991	-2.540155	0.17192426
55	22.379	22.113		22.246	-2.337	0.197954669	55	23.983	23.920		23.951	-3.500417	0.0883628
56	23.915	26.334		25.124	-5.215	0.026918382	56	26.615	26.784		26.700	-6.248572	0.01315202
57	18.898	18.982		18.940	0.969	1.95720178	57	23.331	23.171		23.251	-2.7999325	0.14359401
58	22.389	22.188		22.289	-2.380	0.192136032	58	22.895	22.725		22.810	-2.3593015	0.19488548
var2csa	22.224	21.873		22.049	-2.140	0.226948365	var2csa	19.901	20.159		20.030	0.421228	1.33906686
91	24.584	24.445		24.515	-4.606	0.041069395	91	25.261	25.212		25.237	-4.785665	0.03625528
92	17.365	17.325		17.345	2.564	5.913097231	92	20.340	19.952		20.146	0.3049325	1.23536084
93	22.706	22.613		22.660	-2.751	0.148559163	93	25.909	25.981		25.945	-5.493916	0.02219047
94	18.827	18.964		18.895	1.014	2.018833596	94	21.345	21.183		21.264	-0.813346	0.56906052
95	23.404	23.317		23.360	-3.451	0.091416	95	23.795	23.600		23.698	-3.246623	0.10535838
96	19.792	19.581		19.687	0.222	1.166729404	96	23.784	23.855		23.819	-3.3684255	0.09682843
97	Undetermine	33.177		33.177	-13.268	0.00010137	97	39.050	Undetermined		39.050	-18.599186	2.5182E-06
60	19.937	19.882		19.909	0.000	0.99975431	60	20.522	20.380		20.451	4.5E-05	1.00003119

PfSAMS KD3 ON GlcN 4Wk						
Primer pair	CT			CT Average	relative to p60	relative copy number
1	34.485	36.246		35.365	-12.819	0.00013836
3	27.804	27.942		27.873	-5.327	0.02491039
4	19.301	19.130		19.216	3.330	10.059272
5	27.635	26.862		27.248	-4.702	0.03840673
6	24.426	24.354		24.390	-1.844	0.27855227
7	20.806	20.746		20.776	1.770	3.41117436
8	21.753	21.760		21.756	0.790	1.72848969
9	25.208	24.984		25.096	-2.550	0.17075823
11	25.183	25.104		25.144	-2.598	0.16518705
12	23.107	22.872		22.989	-0.443	0.73554525
13	20.631	20.631		20.631	1.915	3.77021161
15	25.418	25.016		25.217	-2.671	0.15699511
17	24.143	23.926		24.035	-1.489	0.35632238
18	25.264	25.392		25.328	-2.782	0.14541853
19	24.159	23.926		24.043	-1.497	0.35435234
20	24.566	24.404		24.485	-1.939	0.2608192
21	25.197	24.906		25.052	-2.506	0.17608293
22	25.751	25.314		25.533	-2.987	0.12615231
23	24.756	24.481		24.618	-2.072	0.23775227
25	26.128	26.417		26.273	-3.727	0.075534
26	19.731	19.654		19.693	2.853	7.22711825
27	17.783	17.792		17.788	4.758	27.0639498
28	23.479	23.607		23.543	-0.997	0.5009776
30	26.870	26.591		26.731	-4.185	0.05499388
31	24.268	23.966		24.117	-1.571	0.3365882
34	23.681	23.455		23.568	-1.022	0.49251532
35	24.318	24.469		24.393	-1.847	0.27793684
36	26.330	26.229		26.279	-3.733	0.07519203
37	28.651	28.372		28.511	-5.965	0.01600498
38	26.687	26.529		26.608	-4.062	0.05986848
39	24.651	24.523		24.587	-2.041	0.24291702
40	33.557	38.966		36.261	-13.715	7.4351E-05
41	22.947	22.824		22.886	-0.340	0.79023519
43	26.688	26.315		26.501	-3.955	0.06447012
44	20.198	19.625		19.912	2.634	6.20952968
45	24.304	23.993		24.149	-1.603	0.32925585
46	22.492	22.297		22.395	0.151	1.11055204
47	24.913	24.795		24.854	-2.308	0.2019283
49	23.695	23.625		23.660	-1.114	0.46201321
50	24.682	24.422		24.552	-2.006	0.24895338
51	21.939	21.759		21.849	0.697	1.62077014
52	23.969	23.818		23.894	-1.348	0.39294042
53	24.203	24.166		24.184	-1.638	0.32123344
54	20.997	21.142		21.069	1.477	2.78279185
55	22.655	22.421		22.538	0.008	1.00543513
56	26.836	26.574		26.705	-4.159	0.05598862
57	21.480	21.193		21.336	1.210	2.31268616
58	24.295	23.883		24.089	-1.543	0.34318942
var2csa	23.136	23.000		23.068	-0.522	0.69642336
91	26.118	26.131		26.125	-3.579	0.08370668
92	20.493	20.210		20.351	2.195	4.57764949
93	25.200	25.007		25.103	-2.557	0.16989343
94	21.556	21.480		21.518	1.028	2.03914378
95	22.706	22.454		22.580	-0.034	0.97664252
96	21.776	21.781		21.779	0.767	1.70228843
97	39.136	38.015		38.575	-16.029	1.4951E-05
60	22.619	22.473		22.546	0.000	1.00003535

WT Day 0						
Primer pair	CT			CT Average	relative to p60	relative copy number
1	25.476511	25.561333		25.518922	-5.430922	0.02318086
3	30.60869	31.306717		30.9577035	-10.869704	0.00053443
4	26.496288	26.545574		26.520931	-6.432931	0.0115743
5	32.4614	31.004639		31.7330195	-11.64502	0.00031225
6	26.231243	26.127779		26.179511	-6.091511	0.01466468
7	27.898521	28.118597		28.008559	-7.920559	0.00412738
8	27.370888	27.041368		27.206128	-7.118128	0.0071983
9	31.968561	31.701813		31.835187	-11.747187	0.0002909
11	29.370226	28.808144		29.089185	-9.001185	0.00195152
12	27.248745	27.38681		27.3177775	-7.2297775	0.00666224
13	25.348827	26.667913		26.00837	-5.92037	0.01651167
15	20.264917	21.072247		20.668582	-0.580582	0.66869396
17	28.413597	31.312887		29.863242	-9.775242	0.00114119
18	26.712713	27.098206		26.9054595	-6.8174595	0.00886625
19	27.827793	32.538708		30.1832505	-10.095251	0.00091417
20	29.566423	29.930674		29.7485485	-9.6605485	0.00123562
21	28.968014	31.49547		30.231742	-10.143742	0.00088395
22	Undetermine	Undetermined		#DIV/0!	#DIV/0!	0
23	30.77069	32.17296		31.471825	-11.383825	0.00037422
25	28.019365	28.681862		28.3506135	-8.2626135	0.00325616
26	23.997192	24.46033		24.228761	-4.140761	0.05669004
27	21.009655	21.960419		21.485037	-1.397037	0.37970818
28	26.256527	26.552858		26.4046925	-6.3166925	0.01254545
30	28.869373	29.394323		29.131848	-9.043848	0.00189466
31	22.267063	23.013851		22.640457	-2.552457	0.17046447
34	26.315783	26.553408		26.4345955	-6.3465955	0.01228809
35	30.71697	30.958294		30.837632	-10.749632	0.00058082
36	29.675795	30.307222		29.9915085	-9.9035085	0.00104411
37	32.26741	Undetermined		32.26741	-12.17941	0.00021559
38	30.40016	32.46224		31.4312	-11.3432	0.00038491
39	34.947166	29.55263		32.249898	-12.161898	0.00021822
40	32.328667	30.47032		31.3994935	-11.311494	0.00039346
41	Undetermine	Undetermined		#DIV/0!	#DIV/0!	0
43	Undetermine	30.143808		30.143808	-10.055808	0.00093951
44	Undetermine	34.01111		34.01111	-13.92311	6.4376E-05
45	23.246964	22.377533		22.8122485	-2.7242485	0.15132807
46	Undetermine	Undetermined		#DIV/0!	#DIV/0!	0
47	31.511246	29.287685		30.3994655	-10.311466	0.00078694
49	24.18532	23.164715		23.6750175	-3.5870175	0.08321472
50	31.912865	33.936756		32.9248105	-12.836811	0.00013669
51	25.918337	29.381012		27.6496745	-7.5616745	0.00529309
52	27.823355	30.096561		28.959958	-8.871958	0.00213439
53	27.24735	27.112534		27.179942	-7.091942	0.00733015
54	27.163858	25.244177		26.2040175	-6.1160175	0.01441768
55	26.103935	26.49328		26.2986075	-6.2106075	0.01350271
56	28.747679	27.80991		28.2787945	-8.1907945	0.00342236
57	25.082743	25.59421		25.3384765	-5.2504765	0.02626934
58	25.988726	26.367975		26.1783505	-6.0903505	0.01467648
var2csa	26.208424	27.30495		26.756687	-6.668687	0.00982936
91	30.139912	31.52676		30.833336	-10.745336	0.00058255
92	23.453566	23.381353		23.4174595	-3.3294595	0.09947932
93	34.467724	34.631897		34.5498105	-14.461811	4.4316E-05
94	Undetermine	32.942787		32.942787	-12.854787	0.000135
95	Undetermine	Undetermined		#DIV/0!	#DIV/0!	0
96	29.97863	29.757599		29.8681145	-9.7801145	0.00113734
97	34.621964	33.484505		34.0532345	-13.965235	6.2524E-05
60	20.107254	20.069069		20.0881615	-0.0001615	0.99988806

(Rep 1) WT OFF GlcN 2Wks					(Rep 1) WT ON GlcN 2Wks				
Primer pair	CT		relative to p60	relative copy number	Primer pair	CT		relative to p60	relative copy number
1	21.582		-4.991	0.03144453	1	22.421		-5.005	0.0311402
3	23.788		-7.197	0.00681634	3	24.860		-7.444	0.00574335
4	22.780		-6.189	0.01370513	4	23.380		-5.964	0.01602516
5	23.145		-6.554	0.01064345	5	25.226		-7.810	0.00445581
6	23.210		-6.619	0.01017655	6	24.461		-7.045	0.00757471
7	21.427		-4.836	0.03501143	7	23.281		-5.865	0.01716255
8	22.466		-5.875	0.01704184	8	24.846		-7.430	0.00579779
9	23.753		-7.162	0.00698339	9	25.026		-7.610	0.00511864
11	21.745		-5.154	0.02809087	11	23.395		-5.979	0.01585099
12	Undetermined		#VALUE!	#VALUE!	12	24.004		-6.588	0.01039191
13	19.394		-2.803	0.14333114	13	21.374		-3.958	0.06434804
15	16.451		0.140	1.1020449	15	18.272		-0.856	0.55256573
17	21.660		-5.069	0.02978604	17	22.375		-4.959	0.03215679
18	23.319		-6.728	0.00943357	18	23.952		-6.536	0.0107793
19	21.882		-5.291	0.02554952	19	23.416		-6.000	0.01562729
20	23.391		-6.800	0.00897541	20	26.638		-9.222	0.00167511
21	22.992		-6.401	0.01183641	21	24.350		-6.934	0.00817749
22	32.139		-15.548	2.0869E-05	22	Undetermined		#VALUE!	#VALUE!
23	24.789		-8.198	0.00340527	23	25.778		-8.362	0.00304025
25	19.657		-3.066	0.11942005	25	21.423		-4.007	0.06220537
26	Undetermined		#VALUE!	#VALUE!	26	20.697		-3.281	0.10289587
27	17.420		-0.829	0.56278626	27	18.746		-1.330	0.3977812
28	21.048		-4.457	0.04552892	28	23.423		-6.007	0.0155445
30	23.978		-7.387	0.00597314	30	25.702		-8.286	0.00320288
31	18.376		-1.785	0.29020615	31	20.815		-3.399	0.09476998
34	20.133		-3.542	0.08583757	34	22.405		-4.989	0.03148346
35	22.689		-6.098	0.01459475	35	25.827		-8.411	0.00293814
36	22.956		-6.365	0.01213163	36	23.330		-5.914	0.01658288
37	20.224		-3.633	0.0805841	37	21.161		-3.745	0.07459041
38	23.183		-6.592	0.01036521	38	24.345		-6.929	0.00820904
39	Undetermined		#VALUE!	#VALUE!	39	23.014		-5.598	0.02063936
40	33.358		-16.767	8.9698E-06	40	Undetermined		#VALUE!	#VALUE!
41	24.784		-8.193	0.00341748	41	26.921		-9.505	0.00137661
43	23.504		-6.913	0.00829837	43	25.202		-7.786	0.004531
44	20.978		-4.387	0.04778766	44	22.185		-4.769	0.03668601
45	18.990		-2.399	0.18963905	45	20.020		-2.604	0.1644784
46	21.299		-4.708	0.03826661	46	22.435		-5.019	0.03084895
47	27.836		-11.245	0.00041212	47	30.826		-13.410	9.189E-05
49	18.862		-2.271	0.20715018	49	20.112		-2.696	0.15427434
50	30.271		-13.680	7.617E-05	50	30.531		-13.115	0.00011268
51	22.350		-5.759	0.01846942	51	23.738		-6.322	0.01250201
52	24.220		-7.629	0.00505071	52	25.604		-8.188	0.00342922
53	21.874		-5.283	0.02569152	53	22.636		-5.220	0.02682459
54	21.334		-4.743	0.03733193	54	22.692		-5.276	0.02580209
55	23.539		-6.948	0.00809807	55	24.013		-6.597	0.01032684
56	23.640		-7.049	0.00754988	56	24.632		-7.216	0.00672486
57	20.718		-4.127	0.05721618	57	22.366		-4.950	0.03235032
58	23.145		-6.554	0.01064494	58	23.302		-5.886	0.01691097
var2csa	20.667		-4.076	0.05928266	var2csa	21.505		-4.089	0.0587683
91	24.485		-7.894	0.00420299	91	25.487		-8.071	0.00371928
92	17.416		-0.825	0.5644509	92	18.854		-1.438	0.36902284
93	21.756		-5.165	0.02786579	93	22.653		-5.237	0.02651293
94	24.704		-8.113	0.00361084	94	26.477		-9.061	0.0018723
95	22.495		-5.904	0.01670033	95	24.166		-6.750	0.00928937
96	23.798		-7.207	0.00676609	96	25.765		-8.349	0.00306695
97	21.866		-5.275	0.02583055	97	24.322		-6.906	0.00833955
60	16.591		0.000	0.99992098	60	17.416		0.000	0.99999723

(Rep 1) WT OFF GlcN 4Wks					(Rep 1) WT ON GlcN 4Wks				
Primer pair	CT		relative to p60	relative copy number	Primer pair	CT		relative to p60	relative copy number
1	21.644		-3.276	0.10323399	1	21.509		-4.045	0.06058385
3	23.885		-5.517	0.02183445	3	23.191		-5.727	0.01887499
4	22.202		-3.834	0.07012001	4	21.914		-4.450	0.04573778
5	23.726		-5.358	0.02439048	5	22.797		-5.333	0.02481056
6	22.841		-4.473	0.04502387	6	22.622		-5.158	0.02799878
7	22.386		-4.018	0.06174097	7	21.416		-3.952	0.06461276
8	22.966		-4.598	0.04129517	8	21.842		-4.378	0.04808441
9	23.828		-5.460	0.02271705	9	23.306		-5.842	0.01742949
11	21.881		-3.513	0.08760469	11	21.694		-4.230	0.05329581
12	22.936		-4.568	0.04214622	12	21.712		-4.248	0.05262288
13	19.709		-1.341	0.39487447	13	18.952		-1.488	0.3566263
15	16.127		2.241	4.72822602	15	15.726		1.738	3.33541899
17	20.885		-2.517	0.17473906	17	20.507		-3.043	0.12132645
18	22.704		-4.336	0.04952999	18	21.898		-4.434	0.04627212
19	22.182		-3.814	0.0710916	19	21.640		-4.176	0.05530482
20	25.639		-7.271	0.00647542	20	24.905		-7.441	0.00575372
21	23.302		-4.934	0.03271373	21	21.712		-4.248	0.05262394
22	Undetermined		#VALUE!	#VALUE!	22	Undetermined		#VALUE!	#VALUE!
23	24.643		-6.275	0.01291477	23	23.548		-6.084	0.01473921
25	19.280		-0.912	0.53144784	25	18.738		-1.274	0.41352114
26	19.590		-1.222	0.42861433	26	18.808		-1.344	0.39387234
27	17.389		0.979	1.97046756	27	16.740		0.724	1.65196944
28	22.156		-3.788	0.07238909	28	21.263		-3.799	0.07183427
30	24.576		-6.208	0.01352958	30	23.670		-6.206	0.01354174
31	19.201		-0.833	0.56134943	31	18.167		-0.703	0.61446596
34	20.809		-2.441	0.18421877	34	19.874		-2.410	0.18809755
35	24.353		-5.985	0.01578452	35	22.700		-5.236	0.02652561
36	23.451		-5.083	0.02950152	36	Undetermined		#VALUE!	#VALUE!
37	22.900		-4.532	0.04323079	37	21.867		-4.403	0.04726991
38	22.397		-4.029	0.06125736	38	22.326		-4.862	0.03437742
39	21.358		-2.990	0.12584327	39	20.118		-2.654	0.15888061
40	Undetermined		#VALUE!	#VALUE!	40	Undetermined		#VALUE!	#VALUE!
41	25.350		-6.982	0.00791093	41	23.590		-6.126	0.01431895
43	23.108		-4.740	0.03741439	43	22.951		-5.487	0.0222912
44	21.354		-2.986	0.12619039	44	20.535		-3.071	0.11897118
45	18.919		-0.551	0.68249056	45	18.565		-1.101	0.46617838
46	20.509		-2.141	0.22679929	46	20.238		-2.774	0.14616006
47	28.412		-10.044	0.00094696	47	28.544		-11.080	0.00046208
49	19.708		-1.340	0.39490212	49	19.190		-1.726	0.30236792
50	28.439		-10.071	0.00092975	50	27.645		-10.181	0.00086122
51	22.575		-4.207	0.05416092	51	21.970		-4.506	0.04400444
52	24.522		-6.154	0.01404091	52	23.365		-5.901	0.01673205
53	22.236		-3.868	0.06847898	53	21.575		-4.111	0.05787764
54	20.692		-2.324	0.19968557	54	20.539		-3.075	0.118688
55	23.801		-5.433	0.02314188	55	22.854		-5.390	0.02384383
56	22.860		-4.492	0.0444507	56	22.578		-5.114	0.02886606
57	21.290		-2.922	0.13192474	57	19.891		-2.427	0.18595726
58	23.457		-5.089	0.02938315	58	22.795		-5.331	0.02484503
var2csa	19.525		-1.157	0.44857652	var2csa	19.678		-2.214	0.21552095
91	24.472		-6.104	0.01454097	91	23.363		-5.899	0.01675357
92	17.714		0.654	1.57380086	92	16.775		0.689	1.61196453
93	20.981		-2.613	0.16348215	93	20.471		-3.007	0.12440704
94	25.421		-7.053	0.00752873	94	24.302		-6.838	0.00874164
95	21.742		-3.374	0.09646658	95	21.323		-3.859	0.06892719
96	24.201		-5.833	0.01754588	96	22.882		-5.418	0.02339566
97	22.867		-4.499	0.04423457	97	21.274		-3.810	0.07131839
60	18.368		0.000	1.00004852	60	17.464		0.000	1.00001525

(Rep 2) WT OFF GlcN 2Wks				(Rep 2) WT ON GlcN 2Wks			
Primer pair	CT	relative to p60	relative copy number	Primer pair	CT	relative to p60	relative copy number
1	20.556	-3.898	0.06708286	1	21.356	-4.262	0.05211881
3	23.508	-6.850	0.00866587	3	23.632	-6.538	0.0107609
4	21.657	-4.999	0.03126584	4	22.491	-5.397	0.0237353
5	22.632	-5.974	0.01591327	5	23.414	-6.320	0.01251288
6	22.457	-5.799	0.01795681	6	22.738	-5.644	0.02000135
7	20.656	-3.998	0.06258423	7	21.612	-4.518	0.04364872
8	21.711	-5.053	0.03012295	8	22.872	-5.778	0.01822358
9	22.958	-6.300	0.01269039	9	Undetermined	#VALUE!	#VALUE!
11	20.811	-4.153	0.05619424	11	21.321	-4.227	0.05339265
12	Undetermined	#VALUE!	#VALUE!	12	22.680	-5.586	0.02082341
13	18.559	-1.901	0.26774007	13	19.455	-2.361	0.19467911
15	16.451	0.207	1.15443184	15	15.824	1.270	2.41243321
17	21.660	-5.002	0.03120195	17	20.527	-3.433	0.09258704
18	23.319	-6.661	0.00988201	18	22.717	-5.623	0.02029677
19	21.291	-4.633	0.04031364	19	21.719	-4.625	0.0405309
20	23.191	-6.533	0.01079969	20	23.925	-6.831	0.00878585
21	Undetermined	#VALUE!	#VALUE!	21	22.398	-5.304	0.02532126
22	34.361	-17.703	4.6876E-06	22	Undetermined	#VALUE!	#VALUE!
23	Undetermined	#VALUE!	#VALUE!	23	23.818	-6.724	0.00945654
25	18.202	-1.544	0.34292025	25	18.763	-1.669	0.3144647
26	18.646	-1.988	0.25201509	26	Undetermined	#VALUE!	#VALUE!
27	16.378	0.280	1.21414018	27	16.860	0.234	1.17614669
28	20.690	-4.032	0.06111473	28	21.172	-4.078	0.0592268
30	23.606	-6.948	0.00810111	30	24.374	-7.280	0.00643243
31	17.903	-1.245	0.42178918	31	18.809	-1.715	0.3045253
34	19.656	-2.998	0.12520603	34	20.505	-3.411	0.09400882
35	22.224	-5.566	0.02110404	35	22.958	-5.864	0.01716942
36	23.239	-6.581	0.01044392	36	21.940	-4.846	0.03477849
37	19.515	-2.857	0.1380483	37	20.166	-3.072	0.11894809
38	22.365	-5.707	0.01914006	38	22.521	-5.427	0.02324359
39	19.517	-2.859	0.13787082	39	20.638	-3.544	0.08573173
40	Undetermined	#VALUE!	#VALUE!	40	Undetermined	#VALUE!	#VALUE!
41	23.128	-6.470	0.01128116	41	24.391	-7.297	0.00635742
43	22.327	-5.669	0.01965817	43	22.995	-5.901	0.01673996
44	19.900	-3.242	0.10567881	44	20.431	-3.337	0.09898179
45	18.217	-1.559	0.33946506	45	17.844	-0.750	0.5944927
46	20.166	-3.508	0.08788179	46	20.268	-3.174	0.11080593
47	27.348	-10.690	0.00060537	47	27.426	-10.332	0.00077597
49	18.478	-1.820	0.28329872	49	18.254	-1.160	0.44742445
50	30.042	-13.384	9.357E-05	50	27.398	-10.304	0.00079111
51	21.545	-4.887	0.03379126	51	21.803	-4.709	0.03824002
52	22.794	-6.136	0.01422195	52	23.370	-6.276	0.0129081
53	21.225	-4.567	0.04217781	53	21.838	-4.744	0.03730824
54	20.180	-3.522	0.08705591	54	20.579	-3.485	0.08928341
55	22.608	-5.950	0.016174	55	22.569	-5.475	0.02247991
56	22.748	-6.090	0.01467603	56	23.379	-6.285	0.01282702
57	19.888	-3.230	0.10654745	57	20.678	-3.584	0.08337277
58	22.748	-6.090	0.01467661	58	21.994	-4.900	0.03348596
var2csa	19.698	-3.040	0.12154488	var2csa	20.308	-3.214	0.10780164
91	23.424	-6.766	0.00918783	91	24.338	-7.244	0.00659664
92	16.448	0.210	1.15643967	92	17.192	-0.098	0.93411883
93	20.673	-4.015	0.06186126	93	21.913	-4.819	0.03543761
94	23.717	-7.059	0.00749778	94	24.447	-7.353	0.00611774
95	21.503	-4.845	0.03479254	95	25.212	-8.118	0.00359858
96	22.975	-6.317	0.01254597	96	23.701	-6.607	0.01025792
97	21.299	-4.641	0.04007388	97	22.383	-5.289	0.02558248
60	16.658	0.000	0.99987386	60	17.094	0.000	1.00028423

(Rep 2) WT OFF GlcN 4Wks					(Rep 2) WT ON GlcN 4Wks				
Primer pair	CT		relative to p60	relative copy number	Primer pair	CT		relative to p60	relative copy number
1	21.280		-3.324	0.09988813	1	22.408		-4.116	0.05768656
3	23.765		-5.809	0.01783708	3	24.429		-6.137	0.01421303
4	21.635		-3.679	0.07809035	4	21.806		-3.514	0.08754229
5	23.458		-5.502	0.02206973	5	23.908		-5.616	0.02039088
6	22.876		-4.920	0.0330281	6	23.125		-4.833	0.035076
7	22.318		-4.362	0.04861391	7	22.134		-3.842	0.06974502
8	23.198		-5.242	0.02642109	8	22.964		-4.672	0.03922616
9	23.609		-5.653	0.01987917	9	Undetermined		#VALUE!	#VALUE!
11	21.742		-3.786	0.07249946	11	22.321		-4.029	0.061276
12	22.515		-4.559	0.04242643	12	22.405		-4.113	0.05778136
13	Undetermined		#VALUE!	#VALUE!	13	19.857		-1.565	0.33792453
15	16.464		1.492	2.81325826	15	17.130		1.162	2.23734851
17	21.253		-3.297	0.1017161	17	20.958		-2.666	0.15753584
18	22.168		-4.212	0.05397159	18	22.949		-4.657	0.03963731
19	21.744		-3.788	0.07239285	19	22.435		-4.143	0.05661703
20	25.457		-7.501	0.00551905	20	23.835		-5.543	0.02144824
21	22.354		-4.398	0.04743698	21	22.860		-4.568	0.04215469
22	34.846		-16.890	8.232E-06	22	34.185		-15.893	1.6435E-05
23	24.492		-6.536	0.01077449	23	24.572		-6.280	0.0128684
25	19.200		-1.244	0.42222269	25	19.812		-1.520	0.3487316
26	19.312		-1.356	0.39077498	26	18.917		-0.625	0.64824317
27	18.152		-0.196	0.87284053	27	17.883		0.409	1.32732531
28	21.414		-3.458	0.09100912	28	21.465		-3.173	0.11088929
30	24.369		-6.413	0.01173316	30	24.523		-6.231	0.01331336
31	18.930		-0.974	0.5091593	31	19.720		-1.428	0.37173386
34	20.885		-2.929	0.13133888	34	21.176		-2.884	0.13546856
35	24.016		-6.060	0.01498404	35	23.514		-5.222	0.02678461
36	22.879		-4.923	0.03295758	36	22.691		-4.399	0.047407
37	23.259		-5.303	0.02532781	37	23.721		-5.429	0.02321357
38	29.753		-11.797	0.00028109	38	23.116		-4.824	0.03530176
39	21.582		-3.626	0.08098228	39	21.949		-3.657	0.07925847
40	27.556		-9.600	0.00128829	40	Undetermined		#VALUE!	#VALUE!
41	26.285		-8.329	0.00311078	41	25.155		-6.863	0.00858901
43	Undetermined		#VALUE!	#VALUE!	43	23.253		-4.961	0.0321093
44	21.193		-3.237	0.10603835	44	21.194		-2.902	0.1338255
45	19.117		-1.161	0.447337	45	18.880		-0.588	0.66514464
46	20.468		-2.512	0.17525555	46	20.998		-2.706	0.15321899
47	28.390		-10.434	0.00072271	47	27.837		-9.545	0.00133885
49	19.519		-1.563	0.33839614	49	19.285		-0.993	0.50241868
50	28.668		-10.712	0.000596	50	28.358		-10.066	0.00093279
51	22.114		-4.158	0.0560096	51	22.544		-4.252	0.0524694
52	23.783		-5.827	0.01762125	52	24.239		-5.947	0.01620735
53	22.296		-4.340	0.04939138	53	22.700		-4.408	0.04710781
54	21.011		-3.055	0.12033323	54	21.207		-2.915	0.13258156
55	23.456		-5.500	0.02209765	55	23.466		-5.174	0.02770834
56	22.781		-4.825	0.03528312	56	23.820		-5.528	0.02167146
57	21.561		-3.605	0.08220876	57	20.895		-2.603	0.16458275
58	22.791		-4.835	0.03502828	58	23.798		-5.506	0.02201113
var2csa	20.019		-2.063	0.23934557	var2csa	20.138		-1.846	0.27813824
91	24.008		-6.052	0.01507705	91	24.421		-6.129	0.0142916
92	17.683		0.273	1.20825084	92	18.213		0.079	1.05654997
93	20.833		-2.877	0.13609472	93	21.415		-3.123	0.11476503
94	24.859		-6.903	0.00835728	94	24.837		-6.545	0.01070906
95	22.269		-4.313	0.05032346	95	22.820		-4.528	0.04334679
96	24.215		-6.259	0.01305779	96	23.981		-5.689	0.01937924
97	22.931		-4.975	0.03180131	97	22.478		-4.186	0.05493936
60	17.956		0.000	0.99970546	60	18.292		0.000	0.9996618

Chapter 3.5 *var* panels (grown off blasticidin for 2 months)

(Rep 1) PfSAMS OE OFF drug					(Rep 2) PfSAMS OE OFF drug				
Primer pair	CT		relative to p60	relative copy number	Primer pair	CT		relative to p60	relative copy number
1	Undetermined		#VALUE!	#VALUE!	1	23.265		-5.421	0.02333359
3	22.560		-5.019204	0.03083678	3	23.468		-5.624	0.02027744
4	22.281		-4.740317	0.03741299	4	23.270		-5.426	0.02325267
5	22.333		-4.792015	0.03609606	5	23.347		-5.503	0.02204762
6	21.970		-4.428767	0.04643103	6	22.778		-4.934	0.03271044
7	22.183		-4.642332	0.04004228	7	22.788		-4.944	0.032496
8	33.476		-15.93535	1.5958E-05	8	32.376		-14.532	4.22E-05
9	20.913		-3.371645	0.09661259	9	22.018		-4.174	0.05540247
11	22.304		-4.763022	0.03682879	11	23.192		-5.348	0.02454985
12	21.992		-4.450758	0.04572865	12	22.890		-5.046	0.03026905
13	20.421		-2.879721	0.13586813	13	21.431		-3.587	0.08321792
15	23.185		-5.64445	0.01999177	15	24.096		-6.252	0.01311729
17	20.813		-3.272272	0.10350182	17	21.822		-3.978	0.06345761
18	21.294		-3.752663	0.07418838	18	22.107		-4.263	0.05208407
19	22.415		-4.874289	0.03409517	19	23.699		-5.855	0.01727819
20	22.878		-5.336907	0.02474178	20	23.881		-6.037	0.01523022
21	22.671		-5.130398	0.02854936	21	23.698		-5.854	0.01728642
22	21.632		-4.091288	0.05866777	22	22.643		-4.799	0.03591922
23	22.153		-4.612418	0.04088122	23	23.292		-5.448	0.02291358
25	21.242		-3.70098	0.07689428	25	22.290		-4.446	0.04587532
26	22.645		-5.103884	0.02907889	26	23.362		-5.518	0.02181791
27	20.808		-3.266909	0.10388729	27	21.570		-3.726	0.07558685
28	21.014		-3.473341	0.09003683	28	22.254		-4.410	0.04703593
30	23.517		-5.976214	0.01588475	30	24.879		-7.035	0.00762451
31	20.205		-2.664196	0.15776007	31	21.268		-3.424	0.09317076
34	22.198		-4.657093	0.03963468	34	23.257		-5.413	0.02347004
35	22.377		-4.835963	0.03501306	35	23.160		-5.316	0.02510421
36	22.160		-4.618565	0.0407074	36	23.106		-5.262	0.02606853
37	21.942		-4.401293	0.04732371	37	22.815		-4.971	0.03189071
38	22.460		-4.918711	0.03306134	38	23.403		-5.559	0.02121761
39	21.826		-4.284945	0.05129831	39	22.790		-4.946	0.03243072
40	Undetermined		#VALUE!	#VALUE!	40	Undetermined		#VALUE!	#VALUE!
41	22.380		-4.83883	0.03494355	41	Undetermined		#VALUE!	#VALUE!
43	21.662		-4.120915	0.05747526	43	22.610		-4.766	0.03674618
44	22.212		-4.671233	0.03924811	44	23.176		-5.332	0.02483201
45	21.939		-4.397677	0.04744247	45	22.912		-5.068	0.02980374
46	22.203		-4.661816	0.03950513	46	22.980		-5.136	0.02843917
47	25.803		-8.261895	0.00325778	47	27.378		-9.534	0.00134849
49	22.153		-4.612418	0.04088122	49	22.777		-4.933	0.03274572
50	25.990		-8.44898	0.00286156	50	27.534		-9.690	0.001211
51	22.249		-4.707503	0.0382737	51	23.196		-5.352	0.02448935
52	22.630		-5.089236	0.02937564	52	23.395		-5.551	0.02132419
53	21.732		-4.190993	0.05475016	53	22.915		-5.071	0.02974065
54	19.957		-2.416052	0.1873682	54	20.877		-3.033	0.12213131
55	22.211		-4.669693	0.03929003	55	23.263		-5.419	0.02337717
56	22.188		-4.646553	0.0399253	56	23.172		-5.328	0.02490127
57	20.583		-3.041705	0.12143827	57	21.803		-3.959	0.06431776
58	21.683		-4.141718	0.05665244	58	22.816		-4.972	0.03185695
var2csa	17.552		-0.011263	0.99222348	var2csa	17.968		-0.124	0.91775629
91	22.330		-4.788805	0.03617646	91	23.139		-5.295	0.02546324
92	20.904		-3.362532	0.09722479	92	21.434		-3.590	0.08302502
93	22.017		-4.475823	0.04494103	93	22.929		-5.085	0.02946053
94	22.227		-4.685711	0.03885621	94	23.370		-5.526	0.02170706
95	22.223		-4.68162	0.03896655	95	23.361		-5.517	0.02184211
96	22.158		-4.617422	0.04073967	96	23.280		-5.436	0.02309749
97	22.899		-5.358261	0.02437826	97	24.123		-6.279	0.0128795
60	17.541		-3E-05	0.99997921	60	17.844		0.000	0.99975951

(Rep 1) PfSAHH OE OFF drug					(Rep 2) PfSAHH OE OFF drug				
Primer pair	CT		relative to p60	relative copy number	Primer pair	CT		relative to p60	relative copy number
1	22.330		-4.812	0.03558801	1	22.940		-5.423	0.02330467
3	22.792		-5.274	0.02583964	3	24.970		-7.453	0.00570629
4	22.757		-5.239	0.02647119	4	24.212		-6.695	0.0096526
5	22.767		-5.249	0.02630429	5	24.731		-7.214	0.00673652
6	22.161		-4.643	0.04002255	6	22.884		-5.367	0.02423567
7	22.307		-4.789	0.03617734	7	25.298		-7.781	0.00454754
8	23.197		-5.679	0.01951423	8	24.587		-7.070	0.00744405
9	21.751		-4.233	0.05316092	9	23.609		-6.092	0.01465549
11	23.301		-5.783	0.01816339	11	25.267		-7.750	0.00464466
12	22.326		-4.808	0.03569043	12	23.289		-5.772	0.01829911
13	20.648		-3.130	0.11425094	13	21.317		-3.800	0.0718047
15	22.519		-5.001	0.03123034	15	22.484		-4.967	0.03198136
17	21.131		-3.613	0.08173258	17	22.095		-4.578	0.04187706
18	20.791		-3.273	0.10343153	18	21.801		-4.284	0.05134622
19	21.654		-4.136	0.05688531	19	22.552		-5.035	0.03049169
20	23.657		-6.139	0.0141899	20	26.470		-8.953	0.00201788
21	22.193		-4.675	0.03914362	21	22.975		-5.458	0.02275137
22	21.773		-4.255	0.05235906	22	22.745		-5.228	0.02668843
23	22.723		-5.205	0.02710328	23	24.682		-7.165	0.00697034
25	21.554		-4.036	0.06093952	25	22.659		-5.142	0.02832396
26	21.787		-4.269	0.05186755	26	22.623		-5.106	0.02903452
27	20.349		-2.831	0.14054656	27	21.159		-3.642	0.08010433
28	21.799		-4.281	0.05143638	28	23.313		-5.796	0.0180027
30	22.897		-5.379	0.02403141	30	24.405		-6.888	0.00844475
31	19.236		-1.718	0.3040539	31	19.577		-2.060	0.23976667
34	20.662		-3.144	0.11314651	34	21.138		-3.621	0.0812805
35	22.705		-5.187	0.02745864	35	24.835		-7.318	0.00626883
36	22.324		-4.806	0.03575168	36	23.545		-6.028	0.01532532
37	22.153		-4.635	0.04025023	37	23.877		-6.360	0.01217148
38	22.457		-4.939	0.03260787	38	23.325		-5.808	0.01785262
39	22.451		-4.933	0.03273664	39	22.864		-5.347	0.02457108
40	Undetermined		#VALUE!	#VALUE!	40	Undetermined		#VALUE!	#VALUE!
41	Undetermined		#VALUE!	#VALUE!	41	Undetermined		#VALUE!	#VALUE!
43	21.900		-4.382	0.04796274	43	23.351		-5.834	0.0175308
44	22.612		-5.094	0.02927563	44	25.186		-7.669	0.00491277
45	21.499		-3.981	0.06332109	45	22.440		-4.923	0.03295365
46	22.442		-4.924	0.03293289	46	25.320		-7.803	0.00447782
47	26.844		-9.326	0.00155816	47	28.305		-10.788	0.0005654
49	20.676		-3.158	0.11205213	49	21.591		-4.074	0.05937243
50	27.558		-10.040	0.00094978	50	28.485		-10.968	0.00049921
51	22.360		-4.842	0.034875	51	24.491		-6.974	0.00795717
52	22.754		-5.236	0.02653974	52	25.564		-8.047	0.00378192
53	21.775		-4.257	0.05230606	53	22.336		-4.819	0.03542619
54	15.199		2.319	4.98986921	54	14.829		2.688	6.44354221
55	22.151		-4.633	0.04030481	55	24.602		-7.085	0.00736394
56	22.383		-4.865	0.03431673	56	24.148		-6.631	0.01009225
57	21.299		-3.781	0.07274309	57	22.696		-5.179	0.02761113
58	23.140		-5.622	0.02030399	58	23.844		-6.327	0.01245809
var2csa	19.146		-1.628	0.32364811	var2csa	19.251		-1.734	0.30059314
91	21.384		-3.866	0.0685755	91	21.973		-4.456	0.04555613
92	20.591		-3.073	0.11883914	92	21.323		-3.806	0.07149881
93	21.704		-4.186	0.0549217	93	22.570		-5.053	0.03012531
94	22.743		-5.225	0.02674235	94	25.501		-7.984	0.00394932
95	22.892		-5.374	0.02411857	95	24.977		-7.460	0.00568106
96	22.943		-5.425	0.02326944	96	23.645		-6.128	0.01429558
97	23.353		-5.835	0.01751223	97	25.305		-7.788	0.00452351
60	17.518		0.000	0.99969991	60	17.157		0.360	1.28347216

Chapter 3.6 *var* panels

KD1 (at time of Rescue 1 Day 0)					PfSAMS Rescue 1 (Day 0)				
Primer pair	CT		relative to p60	relative copy number	Primer pair	CT		relative to p60	relative copy number
1	31.720		-15.360	2.37828E-05	1	32.020		-16.153	1.37221E-05
3	20.432		-4.072	0.059464893	3	19.963		-4.096	0.058470632
4	15.700		0.660	1.580106719	4	16.876		-1.009	0.496887102
5	20.015		-3.655	0.079391205	5	19.628		-3.761	0.073740806
6	20.005		-3.645	0.079915602	6	21.180		-5.313	0.025157023
7	16.606		-0.246	0.843086755	7	17.913		-2.046	0.242220017
8	16.743		-0.383	0.76690299	8	16.753		-0.886	0.541082317
9	19.416		-3.056	0.120211681	9	19.979		-4.112	0.057837941
11	21.921		-5.561	0.021176383	11	21.357		-5.490	0.022255921
12	16.758		-0.398	0.759149537	12	16.398		-0.531	0.69210652
13	16.757		-0.397	0.759503227	13	16.557		-0.690	0.619746447
15	14.524		1.836	3.570977392	15	13.946		1.921	3.787525233
17	19.702		-3.342	0.098597372	17	19.725		-3.858	0.068950174
18	20.809		-4.449	0.045785259	18	20.789		-4.922	0.032981253
19	20.614		-4.254	0.052411674	19	21.232		-5.365	0.024265493
20	20.467		-4.107	0.058013682	20	21.040		-5.173	0.027727743
21	21.416		-5.056	0.030062692	21	22.592		-6.725	0.009451008
22	21.436		-5.076	0.029638536	22	21.821		-5.954	0.016132468
23	20.801		-4.441	0.046050288	23	20.906		-5.039	0.03042523
25	15.972		0.388	1.308218027	25	16.984		-1.117	0.461049322
26	13.667		2.693	6.46692106	26	13.411		2.456	5.48771668
27	12.852		3.508	11.37349699	27	13.550		2.317	4.983875398
28	20.412		-4.052	0.06027669	28	19.426		-3.559	0.084847326
30	21.840		-5.480	0.022411453	30	21.419		-5.552	0.021309817
31	17.997		-1.637	0.321485818	31	19.204		-3.337	0.098988998
34	19.265		-2.905	0.133512433	34	19.515		-3.648	0.07976911
35	17.467		-1.107	0.464301549	35	17.442		-1.575	0.335530542
36	21.956		-5.596	0.020680215	36	22.275		-6.408	0.011778111
37	22.540		-6.180	0.013794558	37	22.767		-6.900	0.008373973
38	21.579		-5.219	0.026846243	38	22.141		-6.274	0.012920422
39	18.892		-2.532	0.172867312	39	20.284		-4.417	0.046808846
40	29.634		-13.274	0.00010098	40	27.768		-11.901	0.000261547
41	18.756		-2.396	0.189943072	41	18.897		-3.030	0.122396397
43	21.214		-4.854	0.034578042	43	19.795		-3.928	0.065699946
44	14.572		1.788	3.452357819	44	13.405		2.462	5.509922398
45	18.877		-2.517	0.174689161	45	20.677		-4.810	0.035657343
46	18.371		-2.011	0.248047617	46	17.979		-2.112	0.231330757
47	20.235		-3.875	0.0681351	47	19.499		-3.632	0.080634665
49	19.365		-3.005	0.124596893	49	20.610		-4.743	0.037355125
50	21.012		-4.652	0.039765359	50	20.042		-4.175	0.055374516
51	18.866		-2.506	0.176103929	51	19.887		-4.020	0.061628992
52	20.795		-4.435	0.046234833	52	22.518		-6.651	0.009952744
53	18.704		-2.344	0.196955553	53	19.714		-3.847	0.069494711
54	19.440		-3.080	0.118256386	54	19.823		-3.956	0.064435252
55	21.350		-4.990	0.031478269	55	21.202		-5.335	0.024781821
56	23.998		-7.638	0.005021903	56	22.610		-6.743	0.009333138
57	16.497		-0.137	0.909393754	57	15.785		0.082	1.058209299
58	21.631		-5.271	0.025904562	58	21.449		-5.582	0.02087241
var2csa	15.790		0.570	1.484208732	var2csa	11.796		4.071	16.80901173
91	21.784		-5.424	0.023299923	91	20.944		-5.077	0.029624241
92	13.317		3.043	8.243425704	92	14.214		1.653	3.145684541
93	18.895		-2.535	0.172551031	93	18.390		-2.523	0.173953909
94	14.982		1.378	2.598269358	94	14.665		1.202	2.300684251
95	18.961		-2.601	0.164850481	95	20.242		-4.375	0.048195926
96	18.380		-2.020	0.246577147	96	19.526		-3.659	0.079172433
97	33.200		-16.840	8.52537E-06	97	33.697		-17.830	4.2925E-06
60	16.360		0.000	1.000315432	60	15.867		0.000	1.000063078

KD2 (at time of Rescue 2 Day 0)					PfSAMS Rescue 2 (Day 0)				
Primer pair	CT		relative to p60	relative copy number	Primer pair	CT		relative to p60	relative copy number
1	34.670		-16.267	1.26806E-05	1	Undetermined		#VALUE!	#VALUE!
3	21.749		-3.346	0.098358271	3	23.480		-4.2515761	0.05249864
4	14.254		4.149	17.74037996	4	15.968		3.25982411	9.57866177
5	21.862		-3.459	0.090931996	5	21.826		-2.5976321	0.16520943
6	20.877		-2.474	0.179976085	6	23.509		-4.2805182	0.05145595
7	19.413		-1.010	0.496676209	7	20.604		-1.3764598	0.38516279
8	18.612		-0.209	0.865233066	8	16.426		2.80198415	6.97398928
9	20.578		-2.175	0.221497062	9	21.775		-2.5465285	0.17116641
11	23.148		-4.745	0.037289801	11	24.017		-4.7888972	0.03617415
12	20.780		-2.377	0.192496986	12	21.246		-2.0184294	0.24682673
13	18.123		0.280	1.21398048	13	18.370		0.85790952	1.8124102
15	21.314		-2.911	0.132918307	15	24.471		-5.2432563	0.02640113
17	20.679		-2.276	0.206521437	17	22.227		-2.9986769	0.12511469
18	21.853		-3.450	0.091522133	18	24.353		-5.1251609	0.02865318
19	21.334		-2.931	0.131125433	19	22.903		-3.6752249	0.07827932
20	21.273		-2.870	0.13678357	20	22.619		-3.3912913	0.09530586
21	21.471		-3.068	0.119233853	21	21.191		-1.9626338	0.25655966
22	21.774		-3.371	0.096640065	22	22.985		-3.7574469	0.07394278
23	20.891		-2.488	0.178210472	23	20.304		-1.0764014	0.4742102
25	16.517		1.886	3.695395963	25	17.419		1.80856641	3.50294031
26	14.147		4.256	19.10976465	26	15.114		4.11353047	17.3099598
27	14.008		4.395	21.03293245	27	14.713		4.51546033	22.8712027
28	19.224		-0.821	0.56620252	28	20.338		-1.109635	0.46341125
30	22.505		-4.102	0.058229884	30	23.202		-3.9741141	0.06363154
31	20.542		-2.139	0.227027828	31	21.268		-2.0400817	0.24314997
34	20.536		-2.133	0.228006612	34	20.717		-1.4889685	0.35626717
35	19.414		-1.011	0.496281068	35	20.691		-1.4629962	0.36273901
36	23.614		-5.211	0.027003881	36	25.728		-6.5004184	0.01104534
37	23.898		-5.495	0.022174026	37	24.359		-5.131457	0.02852841
38	22.973		-4.570	0.042092713	38	25.278		-6.0495345	0.01509763
39	20.652		-2.249	0.210404299	39	22.908		-3.6802146	0.07800906
40	Undetermined		#VALUE!	#VALUE!	40	Undetermined		#VALUE!	#VALUE!
41	20.285		-1.882	0.271316698	41	20.413		-1.1852729	0.43974136
43	17.751		0.652	1.571667667	43	18.788		0.4403127	1.3568984
44	14.659		3.744	13.39677313	44	15.660		3.56820329	11.8614073
45	19.928		-1.525	0.347535727	45	21.754		-2.5262534	0.1735889
46	20.336		-1.933	0.261870795	46	21.308		-2.0802218	0.23647805
47	20.321		-1.918	0.264706632	47	21.520		-2.2918021	0.20422026
49	20.368		-1.965	0.256219469	49	23.393		-4.1648566	0.05575107
50	20.600		-2.197	0.218030078	50	21.939		-2.7114875	0.15267254
51	19.850		-1.447	0.366663	51	22.696		-3.4678294	0.09038146
52	21.545		-3.142	0.11331584	52	24.690		-5.4618994	0.02268843
53	20.627		-2.224	0.214016108	53	23.864		-4.6359946	0.04021856
54	20.212		-1.809	0.285356629	54	21.544		-2.3155276	0.20088927
55	22.466		-4.063	0.059835494	55	25.499		-6.2708461	0.01295052
56	23.676		-5.273	0.025853467	56	25.819		-6.591458	0.01036987
57	18.006		0.397	1.316707261	57	18.715		0.51315244	1.42716529
58	22.350		-3.947	0.064816874	58	22.391		-3.1625258	0.11168243
var2csa	15.203		3.200	9.187284397	var2csa	16.359		2.86855634	7.3033397
91	22.381		-3.978	0.063439471	91	25.288		-6.0600077	0.01498842
92	13.700		4.703	26.04661581	92	14.895		4.3328336	20.1517553
93	19.200		-0.797	0.57537098	93	20.313		-1.0849101	0.47142165
94	14.482		3.921	15.14283917	94	15.695		3.53324445	11.5774406
95	21.290		-2.887	0.135163708	95	21.572		-2.3444869	0.19689701
96	19.194		-0.791	0.5781045	96	19.629		-0.4008166	0.75742944
97	Undetermined		#VALUE!	#VALUE!	97	Undetermined		#VALUE!	#VALUE!
60	18.403		0.000	1.000263212	60	19.228		-0.0004184	0.99971006

Chapter 3.7 *var* panels

EV Parent						
Primer pair	CT			CT Average	relative to p60	relative copy number
1	24.464	24.483		24.473	-5.690	0.01936584
3	26.396	26.604		26.500	-7.717	0.00475378
4	26.354	25.233		25.794	-7.011	0.00775521
5	24.803	24.669		24.736	-5.953	0.01614736
6	24.736	24.811		24.773	-5.990	0.015729
7	23.677	23.512		23.595	-4.812	0.03561042
8	24.739	24.740		24.739	-5.956	0.01610542
9	24.978	24.892		24.935	-6.152	0.01406305
11	24.091	24.041		24.066	-5.283	0.02567554
12	24.832	24.484		24.658	-5.875	0.01704096
13	22.542	22.018		22.280	-3.497	0.08856872
15	25.708	25.173		25.441	-6.658	0.00990488
17	22.274	21.979		22.126	-3.343	0.09852479
18	23.204	23.163		23.183	-4.400	0.0473531
19	23.988	23.659		23.824	-5.041	0.03038062
21	25.534	25.449		25.492	-6.709	0.00956136
21	24.761	24.340		24.551	-5.768	0.01835659
22	24.418	24.383		24.401	-5.618	0.02036468
23	25.803	25.653		25.728	-6.945	0.00811482
25	26.842	26.163		26.502	-7.719	0.00474595
26	23.976	23.805		23.891	-5.108	0.02900213
27	21.921	21.704		21.812	-3.029	0.12247678
28	26.445	24.998		25.722	-6.939	0.00815147
30	26.771	25.806		26.288	-7.505	0.00550369
31	19.095	18.833		18.964	-0.181	0.88208802
34	20.758	20.576		20.667	-1.884	0.27096179
35	25.469	25.367		25.418	-6.635	0.0100605
36	24.719	24.530		24.624	-5.841	0.01744392
37	24.193	23.838		24.015	-5.232	0.02660099
38	25.707	25.332		25.520	-6.737	0.00937687
39	23.382	23.328		23.355	-4.572	0.04204232
40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
41	26.917	26.638		26.778	-7.995	0.00392065
43	24.819	24.943		24.881	-6.098	0.01460089
44	26.453	25.978		26.215	-7.432	0.00579011
45	24.580	24.543		24.562	-5.779	0.01821666
46	26.106	26.303		26.205	-7.422	0.00583288
47	28.241	27.567		27.904	-9.121	0.00179594
49	24.760	24.945		24.853	-6.070	0.01488584
50	24.405	24.136		24.271	-5.488	0.02228869
51	23.967	24.148		24.058	-5.275	0.0258296
52	23.191	22.990		23.091	-4.308	0.05049584
53	21.941	22.016		21.979	-3.196	0.1091503
54	15.655	15.623		15.639	3.144	8.83859147
55	23.460	23.582		23.521	-4.738	0.03747459
56	24.789	24.894		24.842	-6.059	0.01500246
57	23.285	24.641		23.963	-5.180	0.02758656
58	24.333	24.259		24.296	-5.513	0.0218996
var2csa	22.618	22.557		22.587	-3.804	0.07157765
91	23.516	23.528		23.522	-4.739	0.03745908
92	23.243	22.561		22.902	-4.119	0.05753675
93	23.836	23.675		23.755	-4.972	0.03185735
94	26.137	26.001		26.069	-7.286	0.00640918
95	25.635	25.506		25.570	-6.787	0.00905328
96	26.503	26.701		26.602	-7.819	0.00442979
97	26.557	26.465		26.511	-7.728	0.00471739
60	18.766	18.800		18.783	0.000	1.0000476

EV Clone B6				
Primer pair	CT		relative to p60	relative copy number
1	23.804		-5.610	0.02046933
3	25.496		-7.302	0.00633504
4	23.958		-5.764	0.01840535
5	25.492		-7.298	0.0063551
6	24.138		-5.944	0.01624779
7	25.354		-7.160	0.00699422
8	25.205		-7.011	0.00775307
9	24.423		-6.229	0.01333369
11	25.475		-7.281	0.00642849
12	25.421		-7.227	0.00667714
13	Undetermined		#VALUE!	#VALUE!
15	23.733		-5.539	0.0215006
17	22.565		-4.371	0.04831292
18	23.225		-5.031	0.03057974
19	24.119		-5.925	0.01645318
20	26.885		-8.691	0.00241971
21	25.390		-7.196	0.00681954
22	24.591		-6.397	0.01186497
23	25.390		-7.196	0.00682134
25	23.112		-4.918	0.03308342
26	24.822		-6.628	0.01011086
27	21.956		-3.762	0.07370279
28	24.483		-6.289	0.01278659
30	25.566		-7.372	0.00603487
31	21.603		-3.409	0.09411588
34	24.016		-5.822	0.01767409
35	25.832		-7.638	0.00502123
36	24.902		-6.708	0.00956639
37	22.293		-4.099	0.05833761
38	24.290		-6.096	0.0146223
39	24.600		-6.406	0.01179602
40	Undetermined		#VALUE!	#VALUE!
41	24.608		-6.414	0.01172326
43	24.017		-5.823	0.01766692
44	25.658		-7.464	0.00566435
45	23.871		-5.677	0.01954788
46	25.961		-7.767	0.00459161
47	30.659		-12.465	0.00017686
49	25.258		-7.064	0.00747459
50	31.183		-12.989	0.00012302
51	25.696		-7.502	0.00551546
52	27.085		-8.891	0.00210632
53	23.792		-5.598	0.02065025
54	14.796		3.398	10.5405775
55	25.479		-7.285	0.00641391
56	24.878		-6.684	0.00972297
57	25.914		-7.720	0.00474268
58	22.471		-4.277	0.05159403
var2csa	22.636		-4.442	0.04601902
91	23.738		-5.544	0.02143969
92	23.530		-5.336	0.02475507
93	24.513		-6.319	0.01252969
94	25.878		-7.684	0.00486356
95	25.601		-7.407	0.00589329
96	27.103		-8.909	0.00208
97	28.561		-10.367	0.00075718
60	18.194		0.000	0.99972971

EV Clone D6					EV Clone E1				
Primer pair	CT		relative to p60	relative copy number	Primer pair	CT		relative to p60	relative copy number
1	24.227		-6.155	0.01403779	1	24.405		-6.717	0.00950656
3	26.440		-8.368	0.00302713	3	26.203		-8.515	0.00273401
4	24.820		-6.748	0.00930476	4	23.317		-5.629	0.02020633
5	24.529		-6.457	0.01138265	5	23.997		-6.309	0.01261176
6	23.821		-5.749	0.01859966	6	23.719		-6.031	0.0152932
7	25.131		-7.059	0.00750073	7	25.221		-7.533	0.00539752
8	25.779		-7.707	0.00478472	8	25.653		-7.965	0.0040015
9	24.749		-6.677	0.009772	9	24.372		-6.684	0.00972693
11	26.776		-8.704	0.00239867	11	27.403		-9.715	0.00118953
12	25.901		-7.829	0.00439834	12	24.985		-7.297	0.00635842
13	22.888		-4.816	0.03550222	13	22.475		-4.787	0.03622323
15	24.406		-6.334	0.01239574	15	23.764		-6.076	0.01482617
17	23.466		-5.394	0.02378457	17	22.747		-5.059	0.02999128
18	22.590		-4.518	0.04363586	18	19.423		-1.735	0.30040735
19	Undetermined		#VALUE!	#VALUE!	19	22.153		-4.465	0.04527348
20	27.408		-9.336	0.0015473	20	26.839		-9.151	0.00175958
21	25.783		-7.711	0.00477152	21	24.719		-7.031	0.00764676
22	25.801		-7.729	0.00471408	22	24.880		-7.192	0.00683672
23	25.020		-6.948	0.00810159	23	24.965		-7.277	0.00644967
25	24.349		-6.277	0.01289401	25	23.003		-5.315	0.02512214
26	25.080		-7.008	0.00777173	26	23.882		-6.194	0.01365603
27	22.738		-4.666	0.03939742	27	20.918		-3.230	0.10658704
28	25.455		-7.383	0.00598909	28	24.784		-7.096	0.00730764
30	25.227		-7.155	0.00701806	30	24.627		-6.939	0.00815098
31	20.206		-2.134	0.22786322	31	21.153		-3.465	0.09056215
34	23.957		-5.885	0.01692266	34	24.642		-6.954	0.00806364
35	26.273		-8.201	0.00339938	35	25.563		-7.875	0.0042585
36	24.013		-5.941	0.0162736	36	24.431		-6.743	0.00933885
37	22.618		-4.546	0.04279369	37	22.138		-4.450	0.04576844
38	23.688		-5.616	0.02039128	38	22.848		-5.160	0.02796798
39	26.363		-8.291	0.00319344	39	25.757		-8.069	0.00372386
40	Undetermined		#VALUE!	#VALUE!	40	Undetermined		#VALUE!	#VALUE!
41	26.865		-8.793	0.00225448	41	25.997		-8.309	0.00315324
43	24.614		-6.542	0.01073085	43	24.356		-6.668	0.00983617
44	26.475		-8.403	0.00295511	44	25.657		-7.969	0.00399084
45	26.604		-8.532	0.0027011	45	23.698		-6.010	0.01551366
46	26.413		-8.341	0.00308462	46	25.654		-7.966	0.00399885
47	30.784		-12.712	0.00014907	47	29.673		-11.985	0.00024666
49	23.709		-5.637	0.02009632	49	23.978		-6.290	0.01277658
50	30.674		-12.602	0.00016087	50	30.092		-12.404	0.00018452
51	24.877		-6.805	0.00894044	51	24.737		-7.049	0.00755368
52	26.153		-8.081	0.00369282	52	23.771		-6.083	0.01474721
53	Undetermined		#VALUE!	#VALUE!	53	22.142		-4.454	0.04563524
54	14.493	14.348	3.579	11.9464743	54	14.281		3.407	10.6048624
55	25.243		-7.171	0.0069373	55	24.974		-7.286	0.00640923
56	24.661		-6.589	0.01038788	56	24.192		-6.504	0.01102092
57	26.179		-8.107	0.00362601	57	23.467		-5.779	0.01821422
58	25.002		-6.930	0.0082017	58	22.947		-5.259	0.02611387
var2csa	22.893		-4.821	0.0353776	var2csa	21.125		-3.437	0.09235347
91	22.702		-4.630	0.04037754	91	23.658		-5.970	0.01595414
92	23.795		-5.723	0.01893363	92	21.005		-3.317	0.10034148
93	23.444		-5.372	0.02413919	93	23.942		-6.254	0.01310068
94	26.159		-8.087	0.00367732	94	25.594		-7.906	0.00417009
95	25.599		-7.527	0.00542156	95	25.704		-8.016	0.00386396
96	27.617		-9.545	0.00133845	96	23.822		-6.134	0.01424274
97	28.838		-10.766	0.00057442	97	27.699		-10.011	0.00096902
60	18.072		0.000	1.00031543	60	17.688		0.000	1.0000506

EV Clone E11					EV Clone F1				
Primer pair	CT		relative to p60	relative copy number	Primer pair	CT		relative to p60	relative copy number
1	21.500		-4.680	0.03900187	1	22.593		-5.655	0.01984369
3	23.691		-6.871	0.0085431	3	24.998		-8.060	0.00374822
4	20.423		-3.603	0.08228071	4	23.334		-6.396	0.01187737
5	22.673		-5.853	0.01730391	5	24.138		-7.200	0.00680191
6	21.847		-5.027	0.03067028	6	21.547		-4.609	0.04098634
7	22.686		-5.866	0.01714555	7	23.766		-6.828	0.00879942
8	22.925		-6.105	0.01452775	8	24.454		-7.516	0.00546472
9	22.474		-5.654	0.01986348	9	23.846		-6.908	0.00832791
11	23.982		-7.162	0.00698373	11	25.303		-8.365	0.00303206
12	21.354		-4.534	0.04317665	12	23.608		-6.670	0.00982302
13	20.326		-3.506	0.08804965	13	20.607		-3.669	0.07861116
15	23.639		-6.819	0.00885665	15	23.658		-6.720	0.00948835
17	20.827		-4.007	0.06221007	17	21.560		-4.622	0.04060561
18	20.014		-3.194	0.10925794	18	21.478		-4.540	0.04299879
19	21.352		-4.532	0.0432233	19	21.597		-4.659	0.03957244
20	25.544		-8.724	0.00236514	20	25.786		-8.848	0.00216998
21	22.164		-5.344	0.02462006	21	23.996		-7.058	0.00750223
22	22.100		-5.280	0.02572946	22	23.927		-6.989	0.00787395
23	22.292		-5.472	0.02252872	23	24.349		-7.411	0.00587468
25	22.380		-5.560	0.02119038	25	22.905		-5.967	0.01599072
26	22.620		-5.800	0.01794983	26	23.954		-7.016	0.00772727
27	20.971		-4.151	0.05629236	27	21.141		-4.203	0.05431423
28	22.702		-5.882	0.01696219	28	23.688		-6.750	0.00929013
30	23.261		-6.441	0.01150726	30	23.788		-6.850	0.00866765
31	19.362		-2.542	0.17165062	31	20.868		-3.930	0.06560652
34	23.296		-6.476	0.01123349	34	23.783		-6.845	0.00869885
35	23.934		-7.114	0.00722104	35	25.148		-8.210	0.00337746
36	22.745		-5.925	0.01645974	36	24.283		-7.345	0.00615205
37	22.306		-5.486	0.02231201	37	21.679		-4.741	0.03740293
38	21.785		-4.965	0.0320273	38	23.681		-6.743	0.00933571
39	23.957		-7.137	0.00710448	39	25.183		-8.245	0.00329567
40	Undetermined		#VALUE!	#VALUE!	40	Undetermined		#VALUE!	#VALUE!
41	25.317		-8.497	0.00276834	41	25.666		-8.728	0.00235804
43	21.847		-5.027	0.03068118	43	23.432		-6.494	0.01109277
44	23.854		-7.034	0.00762947	44	25.647		-8.709	0.00238942
45	22.540		-5.720	0.01897703	45	22.683		-5.745	0.01865063
46	23.544		-6.724	0.00945934	46	25.136		-8.198	0.00340527
47	28.062		-11.242	0.0004129	47	30.778		-13.840	6.8203E-05
49	22.991		-6.171	0.01387528	49	24.250		-7.312	0.00629496
50	28.013		-11.193	0.00042709	50	30.468		-13.530	8.4566E-05
51	22.151		-5.331	0.02483805	51	23.911		-6.973	0.00796024
52	24.168		-7.348	0.00613728	52	25.305		-8.367	0.00302788
53	Undetermined		#VALUE!	#VALUE!	53	22.978		-6.040	0.01520206
54	13.798		3.022	8.12557552	54	13.664		3.274	9.67571986
55	24.400		-7.580	0.00522506	55	25.100		-8.162	0.00349251
56	23.032		-6.212	0.01349365	56	24.198		-7.260	0.00652485
57	23.253		-6.433	0.01157628	57	21.781		-4.843	0.0348489
58	22.397		-5.577	0.0209477	58	22.181		-5.243	0.02640692
var2csa	22.444		-5.624	0.02027256	var2csa	21.917		-4.979	0.03171887
91	18.621		-1.801	0.28690162	91	22.489		-5.551	0.02132356
92	21.513		-4.693	0.03865814	92	21.229		-4.291	0.05106717
93	18.945		-2.125	0.2292669	93	22.989		-6.051	0.01508656
94	24.375		-7.555	0.00531725	94	23.844		-6.906	0.00833985
95	24.487		-7.667	0.00492158	95	23.870		-6.932	0.00819076
96	26.091		-9.271	0.0016188	96	26.331		-9.393	0.00148709
97	24.230		-7.410	0.00588036	97	22.600		-5.662	0.01975028
60	16.820		0.000	0.99967497	60	16.938		0.000	1.00032029

EV Clone F6							EV Clone F10						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	24.614	23.749289		24.182	-7.437	0.00577238	1	20.999	20.994	20.997		-4.012	0.06199186
3	25.276	25.12157		25.199	-8.454	0.0028516	3	22.855	22.662	22.758		-5.773	0.01828435
4	23.729	24.205385		23.967	-7.222	0.00669734	4	21.282	21.219	21.250		-4.265	0.05200211
5	Undetermine	23.627312		23.627	-6.882	0.00847652	5	21.500	21.137	21.319		-4.334	0.04958811
6	23.188	23.209911		23.199	-6.454	0.01140495	6	20.589	21.142	20.866		-3.881	0.06789248
7	24.956	24.681965		24.819	-8.074	0.00371154	7	25.856	21.685	23.771		-6.786	0.00906258
8	25.145	24.985313		25.065	-8.320	0.00312843	8	22.233	21.913	22.073		-5.088	0.02940165
9	23.276	22.949759		23.113	-6.368	0.01210878	9	Undetermine	22.004	22.004		-5.019	0.03084144
11	Undetermine	25.481611		25.482	-8.737	0.00234433	11	23.839	23.409	23.624		-6.639	0.01003499
12	23.687	23.344025		23.515	-6.770	0.00915968	12	22.231	21.916	22.074		-5.089	0.02938484
13	21.893	21.638872		21.766	-5.021	0.03080472	13	17.701	17.578	17.639		-0.654	0.63529915
15	23.410	23.471807		23.441	-6.696	0.00964598	15	22.783	22.995	22.889		-5.904	0.01670086
17	22.222	21.971928		22.097	-5.352	0.02448629	17	20.508	20.653	20.581		-3.596	0.08271475
18	21.341	20.923952		21.132	-4.387	0.0477825	18	19.734	19.633	19.684		-2.699	0.15404657
19	Undetermine	22.210356		22.210	-5.465	0.02263414	19	20.306	20.141	20.224		-3.239	0.10593645
20	26.539	27.341148		26.940	-10.195	0.00085293	20	23.803	24.930	24.366		-7.381	0.00599837
21	24.606	24.940632		24.774	-8.029	0.00382972	21	19.638	20.355	19.996		-3.011	0.12403817
22	23.595	23.54452		23.570	-6.825	0.00882014	22	20.781	20.830	20.806		-3.821	0.07078066
23	24.277	23.869705		24.073	-7.328	0.00622289	23	20.225	20.445	20.335		-3.350	0.09807658
25	23.546	23.696339		23.621	-6.876	0.00851313	25	21.418	20.925	21.171		-4.186	0.05492956
26	23.754	23.880796		23.817	-7.072	0.00742961	26	22.404	22.534	22.469		-5.484	0.02234304
27	22.469	22.380379		22.425	-5.680	0.01950916	27	19.689	19.540	19.614		-2.629	0.16160845
28	24.072	22.990221		23.531	-6.786	0.00906222	28	22.755	21.873	22.314		-5.329	0.02486974
30	24.812	24.64459		24.728	-7.983	0.00395188	30	21.988	21.954	21.971		-4.986	0.03154566
31	20.837	20.872782		20.855	-4.110	0.05792245	31	19.457	19.624	19.541		-2.556	0.17009852
34	23.972	24.115902		24.044	-7.299	0.0063508	34	20.710	20.446	20.578		-3.593	0.08285956
35	25.743	25.555378		25.649	-8.904	0.00208718	35	22.962	22.958	22.960		-5.975	0.01589841
36	24.465	24.643776		24.555	-7.810	0.00445754	36	Undetermine	21.327	21.327		-4.342	0.04931687
37	21.554	20.946016		21.250	-4.505	0.04404454	37	22.279	22.520	22.400		-5.415	0.02344616
38	22.440	22.45799		22.449	-5.704	0.01918642	38	20.161	20.319	20.240		-3.255	0.104757
39	25.755	25.610096		25.682	-8.937	0.00203971	39	21.658	21.622	21.640		-4.655	0.03969996
40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!	40	33.058	Undetermine	33.058		-16.073	1.451E-05
41	25.691	26.256927		25.974	-9.229	0.00166639	41	23.852	24.767	24.309		-7.324	0.00623899
43	23.024	22.934504		22.979	-6.234	0.01328532	43	21.395	21.397	21.396		-4.411	0.04700503
44	25.312	25.38044		25.346	-8.601	0.00257504	44	23.826	23.608	23.717		-6.732	0.00940718
45	22.605	22.636295		22.621	-5.876	0.01703055	45	21.639	21.490	21.564		-4.579	0.04182667
46	24.740	24.851955		24.796	-8.051	0.00377101	46	23.235	23.000	23.118		-6.133	0.01425244
47	27.901	28.494581		28.198	-11.453	0.00035676	47	26.893	27.759	27.326		-10.341	0.00077103
49	23.652	23.652477		23.652	-6.907	0.00833024	49	22.106	21.805	21.956		-4.971	0.03189178
50	29.152	29.09193		29.122	-12.377	0.000188	50	27.520	27.198	27.359		-10.374	0.00075356
51	24.604	24.727615		24.666	-7.921	0.00412731	51	21.264	21.833	21.548		-4.563	0.04229807
52	24.644	Undetermined		24.644	-7.899	0.00418812	52	22.884	23.018	22.951		-5.966	0.01599587
53	23.535	23.330114		23.433	-6.688	0.00970125	53	19.899	19.820	19.860		-2.875	0.13633161
54	17.936	12.915426		15.426	1.319	2.49578646	54	19.576	19.740	19.658		-2.673	0.15679528
55	24.614	24.713486		24.664	-7.919	0.00413241	55	20.967	21.291	21.129		-4.144	0.05655787
56	23.810	23.574526		23.692	-6.947	0.0081047	56	21.954	21.820	21.887		-4.902	0.03344316
57	25.340	24.837812		25.089	-8.344	0.0030775	57	21.892	22.122	22.007		-5.022	0.03078149
58	23.188	22.793806		22.991	-6.246	0.0131781	58	21.792	21.425	21.609		-4.624	0.04056179
var2csa	21.907	21.814022		21.860	-5.115	0.02884758	var2csa	21.810	21.741	21.775		-4.790	0.03613456
91	22.574	22.595882		22.585	-5.840	0.01746069	91	20.110	20.359	20.235		-3.250	0.10514353
92	22.922	23.20908		23.066	-6.321	0.01251193	92	20.567	20.959	20.763		-3.778	0.07288956
93	22.925	22.869501		22.897	-6.152	0.01405927	93	20.299	20.450	20.374		-3.389	0.09544352
94	25.762	25.532385		25.647	-8.902	0.00209001	94	23.261	23.450	23.355		-6.370	0.0120902
95	25.091	24.879635		24.985	-8.240	0.00330704	95	23.567	23.331	23.449		-6.464	0.01132872
96	26.350	26.742125		26.546	-9.801	0.00112094	96	24.904	24.779	24.841		-7.856	0.00431537
97	28.441	27.380432		27.911	-11.166	0.00043526	97	23.195	22.935	23.065		-6.080	0.01478156
60	16.836	16.654625		16.745	0.000	0.99977856	60	17.134	16.835	16.985		0.000	1.00032965

EV Clone G12					PfSAMS OE Parent						
Primer pair	CT		relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	26.547		-9.717	0.00118815	1	22.917	22.936		22.927	-5.263	0.02604995
3	26.194		-9.364	0.0015179	3	24.571	24.691		24.631	-6.967	0.00799278
4	23.193		-6.363	0.01215056	4	23.466	23.627		23.547	-5.883	0.01695008
5	22.920		-6.090	0.01467853	5	23.908	23.963		23.935	-6.271	0.01294688
6	22.801		-5.971	0.01593787	6	22.721	22.823		22.772	-5.108	0.0289993
7	24.575		-7.745	0.00466238	7	21.787	21.755		21.771	-4.107	0.05802317
8	24.616		-7.786	0.00453145	8	30.173	31.596		30.885	-13.221	0.00010476
9	22.577		-5.747	0.01862076	9	23.007	22.982		22.994	-5.330	0.02485449
11	24.908		-8.078	0.00370011	11	24.319	24.480		24.400	-6.736	0.00938431
12	23.994		-7.164	0.00697113	12	22.934	23.170		23.052	-5.388	0.02387746
13	20.806		-3.976	0.06356031	13	21.585	21.589		21.587	-3.923	0.06594088
15	22.595		-5.765	0.01838978	15	23.762	23.887		23.825	-6.161	0.01397762
17	21.479		-4.649	0.03985642	17	22.234	22.280		22.257	-4.593	0.04144184
18	20.945		-4.115	0.05771752	18	22.148	22.249		22.198	-4.534	0.04316046
19	22.408		-5.578	0.02093139	19	22.854	22.920		22.887	-5.223	0.0267752
20	25.710		-8.880	0.00212263	20	23.762	23.521		23.641	-5.977	0.01587267
21	23.699		-6.869	0.00855596	21	23.563	24.337		23.950	-6.286	0.01281287
22	23.487		-6.657	0.00990931	22	22.883	22.527		22.705	-5.041	0.03037314
23	23.980		-7.150	0.0070418	23	24.418	24.240		24.329	-6.665	0.00985556
25	23.899		-7.069	0.00744854	25	23.982	23.667		23.825	-6.161	0.01397824
26	23.823		-6.993	0.00785248	26	23.870	23.810		23.840	-6.176	0.01382956
27	22.292		-5.462	0.02268109	27	20.859	21.148		21.004	-3.340	0.09878837
28	24.452		-7.622	0.00507477	28	23.683	24.266		23.975	-6.311	0.01259812
30	24.679		-7.849	0.00433814	30	24.228	24.467		24.348	-6.684	0.00972708
31	20.198		-3.368	0.09684048	31	20.841	20.876		20.859	-3.195	0.10921709
34	23.560		-6.730	0.00942079	34	23.395	23.533		23.464	-5.800	0.01794801
35	24.817		-7.987	0.00394089	35	22.939	22.658		22.798	-5.134	0.02846949
36	24.422		-7.592	0.00518234	36	23.711	23.716		23.714	-6.050	0.01509599
37	20.712		-3.882	0.06781715	37	23.431	23.735		23.583	-5.919	0.01652659
38	23.160		-6.330	0.01242807	38	23.510	23.513		23.512	-5.848	0.01736418
39	24.647		-7.817	0.0044346	39	22.808	22.956		22.882	-5.218	0.02686677
40	Undetermined		#VALUE!	#VALUE!	40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
41	25.743		-8.913	0.00207404	41	24.743	24.722		24.732	-7.068	0.00745133
43	23.251		-6.421	0.01167139	43	22.868	22.967		22.918	-5.254	0.02621349
44	23.791		-6.961	0.00802413	44	24.513	24.326		24.420	-6.756	0.00925429
45	20.308		-3.478	0.08976097	45	23.236	23.192		23.214	-5.550	0.02134131
46	24.850		-8.020	0.00385239	46	23.535	23.376		23.456	-5.792	0.01805379
47	28.617		-11.787	0.00028291	47	28.304	28.551		28.427	-10.763	0.00057542
49	20.290		-3.460	0.09084822	49	23.543	23.706		23.625	-5.961	0.01605707
50	28.684		-11.854	0.00027012	50	28.683	29.504		29.093	-11.429	0.00036256
51	24.194		-7.364	0.00607022	51	22.406	22.532		22.469	-4.805	0.03577626
52	25.340		-8.510	0.00274298	52	25.255	26.072		25.663	-7.999	0.00390882
53	Undetermined		#VALUE!	#VALUE!	53	22.378	22.415		22.396	-4.732	0.03762092
54	12.881		3.949	15.4437157	54	21.619	21.947		21.783	-4.119	0.05754393
55	23.809		-6.979	0.007925	55	24.295	24.453		24.374	-6.710	0.00955266
56	23.731		-6.901	0.00836736	56	23.855	23.759		23.807	-6.143	0.01415104
57	24.733		-7.903	0.00417826	57	21.814	21.663		21.738	-4.074	0.05937163
58	23.402		-6.572	0.01051081	58	23.601	23.379		23.490	-5.826	0.0176283
var2csa	20.743		-3.913	0.06640179	var2csa	13.917	13.950		13.933	3.731	13.2746968
91	22.499		-5.669	0.01965455	91	22.618	22.900		22.759	-5.095	0.02926271
92	23.249		-6.419	0.01168861	92	21.432	21.408		21.420	-3.756	0.07401664
93	22.741		-5.911	0.01662334	93	22.542	22.562		22.552	-4.888	0.03377452
94	24.951		-8.121	0.00359184	94	24.446	25.338		24.892	-7.228	0.00667027
95	24.245		-7.415	0.00585759	95	24.235	23.982		24.108	-6.444	0.01148275
96	26.489		-9.659	0.00123682	96	24.532	24.678		24.605	-6.941	0.00813957
97	27.617		-10.787	0.00056611	97	23.762	23.853		23.808	-6.144	0.0141449
60	16.830		0.000	1.00017746	60	17.437	17.891		17.664	0.000	1.00021109

PfSAMS OE Clone A3					PfSAMS OE Clone A4				
Primer pair	CT		relative to p60	relative copy number	Primer pair	CT		relative to p60	relative copy number
1	24.350		-5.897	0.01678657	1	21.426		-5.823	0.01766656
3	25.142		-6.689	0.00969444	3	20.237		-4.634	0.04026087
4	24.282		-5.829	0.01758914	4	21.685		-6.082	0.01476543
5	26.012		-7.559	0.00530345	5	22.860		-7.257	0.00653611
6	24.770		-6.317	0.01253854	6	22.173		-6.570	0.01052358
7	23.893		-5.440	0.02302767	7	20.585		-4.982	0.03163245
8	Undetermined		#VALUE!	#VALUE!	8	30.452		-14.849	3.3889E-05
9	24.456		-6.003	0.01558917	9	21.533		-5.930	0.01639804
11	25.410		-6.957	0.00805142	11	23.723		-8.120	0.00359463
12	Undetermined		#VALUE!	#VALUE!	12	22.621		-7.018	0.0077147
13	24.194		-5.741	0.01870374	13	20.748		-5.145	0.02827126
15	28.879		-10.426	0.00072702	15	23.424		-7.821	0.00442144
17	24.672		-6.219	0.01342677	17	21.225		-5.622	0.0203014
18	24.460		-6.007	0.0155442	18	21.144		-5.541	0.02147384
19	25.521		-7.068	0.00745094	19	21.802		-6.199	0.01361499
20	27.227		-8.774	0.00228482	20	25.324		-9.721	0.00118476
21	26.596		-8.143	0.00353757	21	22.937		-7.334	0.00620007
22	24.496		-6.043	0.01516143	22	21.563		-5.960	0.01606016
23	26.694		-8.241	0.00330429	23	23.854		-8.251	0.00328181
25	25.605		-7.152	0.00703076	25	22.837		-7.234	0.00664427
26	26.821		-8.368	0.00302754	26	21.742		-6.139	0.0141867
27	24.479		-6.026	0.01535104	27	21.427		-5.824	0.01765711
28	25.813		-7.360	0.00608834	28	24.162		-8.559	0.00265195
30	26.569		-8.116	0.00360389	30	23.376		-7.773	0.00457156
31	23.994		-5.541	0.02147296	31	19.939		-4.336	0.04952779
34	25.738		-7.285	0.00641246	34	22.706		-7.103	0.00727397
35	25.427		-6.974	0.00795209	35	21.496		-5.893	0.01682301
36	25.990		-7.537	0.00538554	36	22.565		-6.962	0.00801984
37	22.889		-4.436	0.04619003	37	23.523		-7.920	0.00413014
38	25.937		-7.484	0.00558562	38	21.639		-6.036	0.01523582
39	26.141		-7.688	0.0048507	39	22.280		-6.677	0.00977155
40	Undetermined		#VALUE!	#VALUE!	40	Undetermined		#VALUE!	#VALUE!
41	26.277		-7.824	0.00441267	41	23.761		-8.158	0.00350073
43	24.802		-6.349	0.01226403	43	22.630		-7.027	0.00766871
44	26.925		-8.472	0.00281533	44	24.655		-9.052	0.00188408
45	26.671		-8.218	0.00335869	45	21.912		-6.309	0.01261188
46	27.267		-8.814	0.00222161	46	22.938		-7.335	0.00619244
47	31.905		-13.452	8.9257E-05	47	29.835		-14.232	5.1968E-05
49	26.326		-7.873	0.00426527	49	22.278		-6.675	0.00978507
50	33.327		-14.874	3.3313E-05	50	30.002		-14.399	4.6278E-05
51	25.622		-7.169	0.00694674	51	22.155		-6.552	0.01065943
52	28.003		-9.550	0.00133387	52	25.606		-10.003	0.00097482
53	24.942		-6.489	0.01113059	53	21.409		-5.806	0.01787609
54	24.297		-5.844	0.0174036	54	21.484		-5.881	0.01697157
55	26.146		-7.693	0.0048341	55	24.607		-9.004	0.00194772
56	26.014		-7.561	0.0052949	56	23.164		-7.561	0.00529522
57	24.362		-5.909	0.01664459	57	21.720		-6.117	0.01440825
58	27.400		-8.947	0.00202567	58	23.175		-7.572	0.00525513
var2csa	14.414		4.039	16.4337127	var2csa	12.419		3.184	9.08777427
91	24.974		-6.521	0.01088909	91	19.996		-4.393	0.04759375
92	Undetermined		#VALUE!	#VALUE!	92	21.732		-6.129	0.01429167
93	26.871		-8.418	0.00292457	93	20.951		-5.348	0.02454883
94	Undetermined		#VALUE!	#VALUE!	94	24.845		-9.242	0.00165157
95	26.618		-8.165	0.00348386	95	23.720		-8.117	0.00360077
96	27.190		-8.737	0.00234367	96	23.496		-7.893	0.00420694
97	26.572		-8.119	0.00359656	97	23.854		-8.251	0.00328291
60	18.453		0.000	1.00029945	60	15.603		0.000	0.99978515

PfSAMS OE Clone B2					PfSAMS OE Clone B7				
Primer pair	CT		relative to p60	relative copy number	Primer pair	CT		relative to p60	relative copy number
1	23.407		-7.0548916	0.00752083	1	23.701		-6.996	0.00783185
3	24.971		-8.6181297	0.00254498	3	25.792		-9.087	0.00183882
4	23.733		-7.380722	0.00600041	4	24.500		-7.795	0.00450421
5	23.900		-7.5472927	0.00534612	5	25.361		-8.656	0.00247936
6	23.381		-7.0281563	0.00766151	6	23.788		-7.083	0.00737795
7	21.700		-5.3474369	0.02456185	7	21.772		-5.067	0.02983945
8	32.558		-16.205652	1.3232E-05	8	31.945		-15.240	2.5844E-05
9	23.569		-7.2163029	0.00672475	9	23.602		-6.897	0.00839211
11	24.871		-8.5186863	0.00272659	11	24.894		-8.189	0.00342638
12	23.930		-7.5776348	0.00523485	12	24.495		-7.790	0.00451777
13	21.273		-4.9206276	0.03301745	13	22.981		-6.276	0.01290868
15	25.854		-9.5012531	0.00137987	15	26.113		-9.408	0.00147241
17	22.454		-6.1018047	0.01456041	17	23.156		-6.451	0.01143148
18	22.539		-6.1867332	0.01372801	18	22.998		-6.293	0.01275269
19	22.601		-6.248806	0.01314989	19	22.985		-6.280	0.01287298
20	24.336		-7.984087	0.00394957	20	24.480		-7.775	0.00456576
21	23.905		-7.5529728	0.00532511	21	24.689		-7.984	0.00394996
22	22.950		-6.5976753	0.01032528	22	23.738		-7.033	0.00763625
23	24.942		-8.5894718	0.00259604	23	25.402		-8.697	0.00240954
25	23.767		-7.4141521	0.00586297	25	23.416		-6.711	0.00954758
26	25.228		-8.8754234	0.00212927	26	25.910		-9.205	0.00169461
27	22.918		-6.5657349	0.01055642	27	22.443		-5.738	0.01874235
28	24.445		-8.0924683	0.00366374	28	25.210		-8.505	0.00275342
30	23.680		-7.3279743	0.00622386	30	25.769		-9.064	0.00186862
31	21.720		-5.3677311	0.02421876	31	22.507		-5.802	0.01792423
34	23.554		-7.2014618	0.00679429	34	23.697		-6.992	0.0078572
35	23.227		-6.8747272	0.0085212	35	23.349		-6.644	0.00999968
36	23.784		-7.4316216	0.00579241	36	24.733		-8.028	0.00383242
37	23.280		-6.9274712	0.0082153	37	21.840		-5.135	0.02846084
38	24.190		-7.8373852	0.00437232	38	24.663		-7.958	0.00402235
39	25.384		-9.0314541	0.001911	39	24.540		-7.835	0.00438087
40	39.603		-23.250219	1.0023E-07	40	Undetermined		#VALUE!	#VALUE!
41	25.347		-8.9942226	0.00196096	41	24.883		-8.178	0.00345283
43	23.642		-7.2893791	0.00639261	43	24.526		-7.821	0.00442288
44	25.732		-9.3801098	0.00150074	44	25.397		-8.692	0.00241852
45	24.521		-8.1685123	0.00347562	45	24.278		-7.573	0.00525044
46	25.296		-8.9440708	0.00203033	46	25.408		-8.703	0.00239983
47	30.944		-14.591455	4.0507E-05	47	30.234		-13.529	8.4572E-05
49	24.201		-7.8489876	0.0043373	49	24.906		-8.201	0.00339896
50	30.329		-13.976742	6.2027E-05	50	31.048		-14.343	4.8124E-05
51	22.875		-6.522213	0.01087973	51	23.471		-6.766	0.00918845
52	25.626		-9.2738876	0.00161541	52	26.815		-10.110	0.00090514
53	22.871		-6.5190983	0.01090325	53	23.391		-6.686	0.00970974
54	22.476		-6.1234379	0.01434371	54	23.252		-6.547	0.01069246
55	25.966		-9.6134701	0.00127661	55	25.548		-8.843	0.00217808
56	25.277		-8.924387	0.00205822	56	25.183		-8.478	0.00280541
57	21.998		-5.6459427	0.0199711	57	22.519		-5.814	0.01777563
58	23.691		-7.3385887	0.00617824	58	24.712		-8.007	0.00388738
var2csa	13.623		2.72963238	6.63286599	var2csa	13.775		2.930	7.62075336
91	23.862		-7.5094204	0.00548832	91	23.780		-7.075	0.0074187
92	22.677		-6.3243904	0.01247868	92	22.315		-5.610	0.02047654
93	25.737		-9.3849621	0.0014957	93	25.189		-8.484	0.00279206
94	25.367		-9.0142612	0.00193391	94	25.779		-9.074	0.00185548
95	25.772		-9.4194832	0.00146034	95	26.680		-9.975	0.00099364
96	24.444		-8.0919132	0.00366515	96	24.710		-8.005	0.0038939
97	24.131		-7.7782078	0.0045554	97	24.546		-7.841	0.00436008
60	16.352		0	1	60	16.705		0.000	0.99981498

PfSAMS OE Clone F4					PfSAMS OE Clone H7				
Primer pair	CT		relative to p60	relative copy number	Primer pair	CT		relative to p60	relative copy number
1	23.752		-5.363	0.02430325	1	Undetermined		#VALUE!	#VALUE!
3	26.356		-7.967	0.00399695	3	26.361		-7.5499161	0.0053364
4	24.969		-6.580	0.01045526	4	24.313		-5.5021847	0.02206365
5	26.241		-7.852	0.00432879	5	25.651		-6.8398484	0.00872972
6	24.497		-6.108	0.01449571	6	24.526		-5.7146653	0.01904208
7	24.361		-5.972	0.01593564	7	23.977		-5.1660031	0.02785339
8	34.326		-15.937	1.5944E-05	8	32.724		-13.912598	6.4847E-05
9	24.356		-5.967	0.01598336	9	24.303		-5.4921712	0.02221732
11	26.722		-8.333	0.00310211	11	26.312		-7.5014142	0.00551886
12	25.418		-7.029	0.00765642	12	24.590		-5.778983	0.0182118
13	24.250		-5.861	0.01720083	13	23.534		-4.7234448	0.0378531
15	27.127		-8.738	0.00234167	15	26.584		-7.7725667	0.00457324
17	24.266		-5.877	0.01701109	17	23.969		-5.1583432	0.02800167
18	23.977		-5.588	0.02079357	18	23.895		-5.0838631	0.02948524
19	24.732		-6.343	0.01232093	19	25.411		-6.5999783	0.01030881
20	26.311		-7.922	0.00412288	20	26.425		-7.6137036	0.0051056
21	26.287		-7.898	0.00419155	21	26.218		-7.40742	0.00589039
22	24.481		-6.092	0.01466003	22	24.219		-5.408202	0.02354881
23	26.020		-7.631	0.00504338	23	26.275		-7.4638661	0.00566438
25	25.528		-7.139	0.00709528	25	25.419		-6.6083096	0.01024945
26	25.370		-6.981	0.00791344	26	25.795		-6.983548	0.0079021
27	23.741		-5.352	0.02448569	27	24.237		-5.4255379	0.02326753
28	25.889		-7.500	0.0055225	28	26.100		-7.2888974	0.00639475
30	26.436		-8.047	0.00378052	30	25.610		-6.7987641	0.0089819
31	23.755		-5.366	0.02424093	31	24.319		-5.5084275	0.02196838
34	25.222		-6.833	0.00876908	34	26.224		-7.413329	0.00586632
35	24.941		-6.552	0.01065593	35	24.944		-6.132573	0.01425317
36	24.877		-6.488	0.01113949	36	26.243		-7.4324177	0.00578921
37	20.665		-2.276	0.20651324	37	21.636		-2.8245743	0.14116219
38	25.401		-7.012	0.00774613	38	22.874		-4.0628155	0.05983712
39	24.952		-6.563	0.01057945	39	24.740		-5.9286069	0.01641767
40	Undetermined		#VALUE!	#VALUE!	40	Undetermined		#VALUE!	#VALUE!
41	25.763		-7.374	0.0060266	41	25.640		-6.8293904	0.00879323
43	25.302		-6.913	0.00830084	43	25.394		-6.5832528	0.01042902
44	27.232		-8.843	0.00217713	44	26.658		-7.8466824	0.00434423
45	25.900		-7.511	0.00548345	45	26.143		-7.3321217	0.006206
46	26.405		-8.016	0.00386405	46	26.491		-7.6799515	0.00487646
47	31.429		-13.040	0.00011874	47	26.144		-7.3325013	0.00620436
49	25.807		-7.418	0.0058487	49	26.293		-7.4821881	0.0055929
50	31.336		-12.947	0.00012664	50	25.723		-6.9115685	0.00830636
51	24.812		-6.423	0.01165168	51	25.262		-6.450858	0.01143137
52	28.693		-10.304	0.00079099	52	27.505		-8.6937112	0.00241508
53	23.186		-4.797	0.03596682	53	24.364		-5.5534619	0.02129322
54	24.477		-6.088	0.01470376	54	23.333		-4.5224389	0.04351212
55	25.779		-7.390	0.00596045	55	25.604		-6.7925118	0.00902091
56	25.549		-7.160	0.00699461	56	26.126		-7.3154133	0.00627829
57	24.463		-6.074	0.0148409	57	24.546		-5.7353066	0.01877157
58	24.810		-6.421	0.01167291	58	25.655		-6.8440446	0.00870437
var2csa	14.267		4.122	17.4060421	var2csa	14.288		4.52263845	22.9852818
91	24.940		-6.551	0.01066452	91	24.733		-5.9219903	0.01649314
92	23.374		-4.985	0.03158328	92	23.177		-4.3655919	0.0485094
93	27.222		-8.833	0.00219303	93	25.363		-6.5519799	0.01065755
94	26.621		-8.232	0.00332518	94	26.997		-8.186242	0.00343317
95	27.305		-8.916	0.00207022	95	27.200		-8.3894337	0.00298215
96	26.805		-8.416	0.00292684	96	26.584		-7.7731637	0.00457135
97	27.118		-8.729	0.00235644	97	26.932		-8.1210869	0.00359178
60	18.389		0.000	1.00013226	60	18.811		0.00043405	1.00030091

PfSAMS OE Clone F11					PfSAMS OE Clone H9				
Primer pair	CT		relative to p60	relative copy number	Primer pair	CT		relative to p60	relative copy number
1	24.761		-7.524	0.00543432	1	22.747		-6.017	0.0154437
3	27.251		-10.014	0.00096706	3	23.179		-6.449	0.01144332
4	26.165		-8.928	0.00205369	4	22.629		-5.899	0.01676337
5	26.504		-9.267	0.00162357	5	23.652		-6.922	0.00824658
6	24.777		-7.540	0.00537456	6	22.513		-5.783	0.01815604
7	23.658		-6.421	0.01166763	7	20.165		-3.435	0.09245286
8	35.716		-18.479	2.7364E-06	8	32.803		-16.073	1.4506E-05
9	25.493		-8.256	0.00327067	9	23.529		-6.799	0.00897967
11	26.607		-9.370	0.00151141	11	24.150		-7.420	0.00583838
12	25.764		-8.527	0.00271056	12	23.135		-6.405	0.01179855
13	23.738		-6.501	0.01103798	13	20.700		-3.970	0.06380184
15	25.933		-8.696	0.0024109	15	24.371		-7.641	0.00500958
17	24.877		-7.640	0.00501297	17	21.765		-5.035	0.03050605
18	25.008		-7.771	0.00457821	18	18.857		-2.127	0.22890946
19	25.826		-8.589	0.00259744	19	21.869		-5.139	0.02838014
20	26.487		-9.250	0.00164249	20	23.523		-6.793	0.00901891
21	27.218		-9.981	0.00098941	21	18.422		-1.692	0.3094581
22	24.448		-7.211	0.0067511	22	22.243		-5.513	0.02189143
23	26.964		-9.727	0.00118013	23	24.123		-7.393	0.00595104
25	25.848		-8.611	0.00255816	25	22.300		-5.570	0.02105513
26	26.643		-9.406	0.00147358	26	22.566		-5.836	0.01750344
27	24.625		-7.388	0.00597059	27	21.025		-4.295	0.05095658
28	26.993		-9.756	0.00115641	28	23.436		-6.706	0.00957729
30	27.356		-10.119	0.00089899	30	23.495		-6.765	0.00919176
31	24.568		-7.331	0.00620974	31	20.784		-4.054	0.0601916
34	26.330		-9.093	0.00183118	34	21.997		-5.267	0.02596758
35	25.008		-7.771	0.00457717	35	21.737		-5.007	0.03110476
36	26.409		-9.172	0.00173351	36	22.916		-6.186	0.01373043
37	21.682		-4.445	0.04590517	37	23.910		-7.180	0.00689586
38	26.954		-9.717	0.00118835	38	22.797		-6.067	0.0149135
39	25.945		-8.708	0.00239148	39	22.524		-5.794	0.01802924
40	37.801		-20.564	6.4505E-07	40	Undetermined		#VALUE!	#VALUE!
41	25.401		-8.164	0.00348563	41	24.627		-7.897	0.00419403
43	25.669		-8.432	0.00289548	43	23.321		-6.591	0.01037539
44	28.203		-10.966	0.00049995	44	25.150		-8.420	0.00291971
45	26.380		-9.143	0.00176917	45	20.578		-3.848	0.06943112
46	25.412		-8.175	0.00345916	46	22.804		-6.074	0.01484198
47	32.605		-15.368	2.3648E-05	47	28.794		-12.064	0.0002335
49	26.264		-9.027	0.00191723	49	22.550		-5.820	0.01770661
50	32.732		-15.495	2.1655E-05	50	28.759		-12.029	0.00023931
51	25.448		-8.211	0.00337588	51	22.157		-5.427	0.02325196
52	28.557		-11.320	0.00039118	52	25.570		-8.840	0.00218208
53	23.564		-6.327	0.01245632	53	21.636		-4.906	0.03334996
54	24.573		-7.336	0.00618798	54	21.263		-4.533	0.04319548
55	26.209		-8.972	0.00199195	55	24.415		-7.685	0.00486034
56	26.283		-9.046	0.0018918	56	24.265		-7.535	0.00539045
57	24.687		-7.450	0.00572017	57	20.226		-3.496	0.08863833
58	23.449		-6.212	0.01348801	58	23.668		-6.938	0.00815458
var2csa	15.116		2.121	4.34970287	var2csa	21.022		-4.292	0.05105031
91	24.907		-7.670	0.00491003	91	21.988		-5.258	0.02613821
92	23.578		-6.341	0.01233218	92	21.235		-4.505	0.04402885
93	26.938		-9.701	0.00120122	93	22.919		-6.189	0.01370361
94	27.659		-10.422	0.000729	94	24.632		-7.902	0.00418077
95	28.524		-11.287	0.00040014	95	24.177		-7.447	0.00572994
96	26.156		-8.919	0.00206583	96	22.919		-6.189	0.01370656
97	26.818		-9.581	0.00130601	97	21.530		-4.800	0.03590255
60	17.237		0.000	0.99983509	60	16.730		0.000	1.00015105

PfSAMS OE Clone B11					PfSAMS OE Clone C7				
Primer pair	CT		relative to p60	relative copy number	Primer pair	CT		relative to p60	relative copy number
1	22.630		-5.6348629	0.02012506	1	22.424		-5.247	0.0263355
3	26.192		-9.1970062	0.00170383	3	23.961		-6.784	0.00907271
4	23.341		-6.3450432	0.01230132	4	22.662		-5.485	0.02233555
5	25.447		-8.4519577	0.00285566	5	17.619		-0.442	0.73604812
6	22.909		-5.9130878	0.01659523	6	22.330		-5.153	0.02811293
7	22.929		-5.9334335	0.01636283	7	20.164		-2.987	0.12613133
8	33.097		-16.10154	1.4222E-05	8	30.843		-13.666	7.6918E-05
9	22.001		-5.0052032	0.0311375	9	22.733		-5.556	0.02125872
11	25.268		-8.2728577	0.00323312	11	23.849		-6.672	0.00980486
12	23.000		-6.0046158	0.01557509	12	22.669		-5.492	0.02221331
13	22.252		-5.2565956	0.02615815	13	22.676		-5.499	0.02211465
15	25.560		-8.564764	0.00264088	15	24.338		-7.161	0.0069868
17	21.786		-4.7908707	0.0361247	17	21.337		-4.160	0.05592406
18	21.983		-4.9876404	0.03151887	18	21.400		-4.223	0.05354351
19	23.957		-6.9618416	0.00802189	19	21.496		-4.319	0.0501127
20	24.661		-7.665062	0.00492704	20	22.701		-5.524	0.02172879
21	24.614		-7.618309	0.00508933	21	22.785		-5.608	0.02050235
22	22.731		-5.7353401	0.01877114	22	22.161		-4.984	0.03159952
23	25.535		-8.53936	0.0026878	23	23.580		-6.403	0.01181364
25	24.717		-7.7210884	0.00473937	25	22.284		-5.107	0.02900817
26	26.108		-9.1122952	0.00180687	26	22.630		-5.453	0.02282117
27	23.498		-6.5024681	0.01102966	27	20.747		-3.570	0.08421347
28	25.072		-8.0764866	0.00370455	28	23.544		-6.367	0.01211484
30	23.916		-6.9208183	0.00825327	30	23.801		-6.624	0.01013967
31	22.344		-5.348465	0.02454435	31	21.443		-4.266	0.05197663
34	24.482		-7.4862251	0.00557727	34	20.769		-3.592	0.08295353
35	23.678		-6.6829948	0.00973236	35	21.865		-4.688	0.03880452
36	24.468		-7.4723759	0.00563107	36	22.429		-5.252	0.02624836
37	21.175		-4.179985	0.05516951	37	25.849		-8.672	0.00245127
38	24.368		-7.3726559	0.00603406	38	22.529		-5.352	0.02447909
39	24.834		-7.8384762	0.00436902	39	22.760		-5.583	0.02085546
40	Undetermined		#VALUE!	#VALUE!	40	35.499		-18.322	3.0524E-06
41	24.956		-7.9605885	0.00401443	41	24.169		-6.992	0.00785668
43	24.319		-7.3232422	0.00624431	43	22.693		-5.516	0.02185189
44	23.118		-6.1224518	0.01435352	44	24.582		-7.405	0.00590012
45	25.284		-8.2882252	0.00319886	45	22.591		-5.414	0.02344987
46	24.739		-7.7431145	0.00466756	46	23.158		-5.981	0.01583041
47	30.257		-13.261717	0.00010182	47	28.854		-11.677	0.0003053
49	25.203		-8.2074585	0.00338305	49	23.310		-6.133	0.01425005
50	31.051		-14.055094	5.8748E-05	50	27.934		-10.757	0.00057784
51	23.558		-6.5620193	0.01058365	51	21.138		-3.961	0.06422327
52	26.587		-9.5918388	0.00129589	52	24.736		-7.559	0.00530448
53	23.311		-6.3157978	0.01255323	53	21.792		-4.615	0.04082053
54	22.871		-5.8750401	0.01703871	54	20.841		-3.664	0.07890663
55	25.760		-8.7641544	0.00229999	55	24.796		-7.619	0.00508779
56	24.866		-7.8706207	0.00427275	56	23.895		-6.718	0.00949825
57	23.107		-6.1115608	0.01446228	57	20.491		-3.314	0.10054499
58	24.674		-7.6782818	0.0048821	58	24.358		-7.181	0.00688975
var2csa	13.540		3.45533657	10.9688211	var2csa	21.460		-4.283	0.05136632
91	23.340		-6.3441982	0.01230853	91	22.158		-4.981	0.03165714
92	22.326		-5.3309917	0.02484343	92	21.345		-4.168	0.05564587
93	25.110		-8.1145058	0.0036082	93	23.195		-6.018	0.01543198
94	38.210		-21.21442	4.1098E-07	94	23.749		-6.572	0.01051169
95	25.785		-8.7898674	0.00225936	95	25.163		-7.986	0.00394538
96	24.410		-7.4146023	0.00586114	96	22.518		-5.341	0.0246774
97	25.284		-8.2889576	0.00319724	97	23.537		-6.360	0.01217774
60	16.995		0	1	60	17.177		0	1

PfSAMS OE Clone E11					PfSAMS OE Clone G7				
Primer pair	CT		relative to p60	relative copy number	Primer pair	CT		relative to p60	relative copy number
1	24.460		-5.724287	0.0189155	1	22.934		-6.5072002	0.01099354
3	27.349		-8.6138134	0.00255261	3	25.850		-9.4236832	0.00145609
4	25.702		-6.9663391	0.00799692	4	24.265		-7.8386955	0.00436835
5	25.828		-7.0926647	0.00732648	5	24.817		-8.3899517	0.00298107
6	25.297		-6.5612469	0.01058932	6	23.976		-7.5491772	0.00533914
7	24.161		-5.4250336	0.02327567	7	22.617		-6.1907482	0.01368986
8	33.499		-14.763428	3.5956E-05	8	30.922		-14.494999	4.3308E-05
9	24.449		-5.713274	0.01906045	9	24.216		-7.7896614	0.00451937
11	26.845		-8.1093483	0.00362112	11	25.220		-8.7935734	0.00225357
12	25.892		-7.1560097	0.00701175	12	24.222		-7.795639	0.00450069
13	25.467		-6.7311592	0.00941281	13	22.742		-6.3156509	0.01255451
15	28.431		-9.6953964	0.00120613	15	26.000		-9.5729923	0.00131293
17	24.854		-6.1184826	0.01439306	17	23.690		-7.2635689	0.00650801
18	24.772		-6.0367661	0.01523184	18	22.506		-6.0791759	0.0147906
19	26.946		-8.2105007	0.00337593	19	24.714		-8.2869492	0.00320169
20	26.950		-8.2143555	0.00336692	20	24.685		-8.2581215	0.00326631
21	27.451		-8.7149258	0.00237983	21	23.946		-7.5189667	0.00545212
22	24.681		-5.9458885	0.01622218	22	24.090		-7.6637154	0.00493164
23	27.749		-9.0130711	0.00193551	23	26.374		-9.9470959	0.00101304
25	26.580		-7.8448734	0.00434968	25	23.705		-7.2780972	0.0064428
26	27.463		-8.726984	0.00236002	26	25.531		-9.10466	0.00181645
27	24.841		-6.1054459	0.01452371	27	22.431		-6.0044403	0.01557698
28	26.751		-8.0157604	0.00386381	28	24.595		-8.1688023	0.00347493
30	27.533		-8.7973728	0.00224764	30	25.318		-8.8917389	0.00210533
31	24.976		-6.2406826	0.01322414	31	22.666		-6.2390766	0.01323887
34	27.500		-8.7648582	0.00229887	34	24.004		-7.5776024	0.00523497
35	25.498		-6.7620277	0.00921355	35	23.601		-7.1745663	0.00692214
36	25.189		-6.4534302	0.01141101	36	24.991		-8.5646019	0.00264118
37	24.777		-6.0415268	0.01518166	37	25.507		-9.0806408	0.00184695
38	25.937		-7.2017498	0.00679293	38	25.168		-8.7415562	0.0023363
39	26.697		-7.9612179	0.00401268	39	25.167		-8.7404232	0.00233814
40	Undetermined		#VALUE!	#VALUE!	40	37.759		-21.332273	3.7874E-07
41	26.661		-7.9254436	0.00411343	41	24.927		-8.5000343	0.00276207
43	26.139		-7.4034004	0.00590683	43	24.450		-8.0232334	0.00384385
44	27.898		-9.1623077	0.0017453	44	26.090		-9.6629963	0.00123353
45	27.305		-8.5696182	0.00263201	45	23.560		-7.132967	0.00712464
46	27.800		-9.0646667	0.00186751	46	25.412		-8.9850883	0.00197342
47	35.599		-16.86377	8.3849E-06	47	30.622		-14.195137	5.3314E-05
49	26.567		-7.8310127	0.00439168	49	23.359		-6.9323254	0.0081877
50	32.330		-13.594673	8.0834E-05	50	30.693		-14.266253	5.0749E-05
51	25.730		-6.994276	0.00784356	51	23.520		-7.0928535	0.00732552
52	28.732		-9.9965	0.00097893	52	27.132		-10.705038	0.00059905
53	25.382		-6.6461029	0.00998444	53	23.808		-7.3815708	0.00599689
54	25.141		-6.4053993	0.0117973	54	22.809		-6.3826656	0.01198467
55	27.533		-8.7969475	0.0022483	55	25.169		-8.7424774	0.00233481
56	27.368		-8.6321812	0.00252031	56	25.595		-9.168251	0.00173813
57	24.344		-5.6079159	0.0205045	57	22.997		-6.5702229	0.01052364
58	25.944		-7.2080917	0.00676314	58	22.708		-6.2817116	0.01285335
var2csa	15.583		3.1529541	8.89475027	var2csa	13.413		3.01405239	8.07830374
91	25.568		-6.8319225	0.00877781	91	23.912		-7.4856396	0.00557953
92	24.335		-5.5996571	0.02062221	92	22.020		-5.5930595	0.02071674
93	27.742		-9.0063267	0.00194458	93	24.970		-8.5434189	0.00268025
94	27.315		-8.5794697	0.0026141	94	25.328		-8.9009304	0.00209196
95	27.938		-9.202116	0.0016978	95	25.938		-9.5110016	0.00137058
96	26.308		-7.5719585	0.00525549	96	25.541		-9.1140442	0.00180468
97	26.421		-7.685709	0.00485703	97	25.905		-9.4779549	0.00140233
60	18.736		0	1	60	16.427		0	1

PfSAMS OE Clone H4					PfSAMS OE Clone H8				
Primer pair	CT		relative to p60	relative copy number	Primer pair	CT		relative to p60	relative copy number
1	22.484		-6.876	0.00851528	1	22.847		-7.329	0.00621876
3	23.661		-8.053	0.00376547	3	23.945		-8.427	0.00290545
4	24.183		-8.575	0.00262236	4	24.541		-9.023	0.00192238
5	25.219		-9.611	0.00127879	5	23.946		-8.428	0.00290433
6	24.220		-8.612	0.00255613	6	23.721		-8.203	0.00339383
7	22.542		-6.934	0.00817842	7	22.406		-6.888	0.00844573
8	32.164		-16.556	1.038E-05	8	31.729		-16.211	1.3186E-05
9	23.696		-8.088	0.00367627	9	23.723		-8.205	0.00338834
11	24.939		-9.331	0.00155223	11	24.499		-8.981	0.00197878
12	23.823		-8.215	0.00336581	12	23.111		-7.593	0.00518053
13	22.935		-7.327	0.00622744	13	22.400		-6.882	0.00847671
15	26.179		-10.571	0.00065758	15	25.493		-9.975	0.0009934
17	22.974		-7.366	0.0060601	17	22.979		-7.461	0.00567679
18	23.388		-7.780	0.00455106	18	22.776		-7.258	0.00653204
19	25.435		-9.827	0.00110111	19	24.895		-9.377	0.0015035
20	27.172		-11.564	0.0003302	20	27.569		-12.051	0.00023566
21	26.491		-10.883	0.00052951	21	26.487		-10.969	0.00049885
22	22.745		-7.137	0.00710474	22	22.737		-7.219	0.00671001
23	26.467		-10.859	0.00053849	23	26.563		-11.045	0.00047324
25	24.755		-9.147	0.00176445	25	24.522		-9.004	0.0019473
26	24.685		-9.077	0.00185132	26	23.554		-8.036	0.00381003
27	23.566		-7.958	0.00402142	27	22.496		-6.978	0.00793135
28	25.655		-10.047	0.00094548	28	24.471		-8.953	0.00201818
30	25.629		-10.021	0.00096253	30	25.790		-10.272	0.00080859
31	23.248		-7.640	0.00501263	31	23.217		-7.699	0.00481246
34	24.787		-9.179	0.00172559	34	25.207		-9.689	0.00121165
35	23.268		-7.660	0.00494356	35	23.737		-8.219	0.0033569
36	25.172		-9.564	0.00132109	36	24.947		-9.429	0.00145043
37	23.819		-8.211	0.00337413	37	25.008		-9.490	0.00139086
38	25.222		-9.614	0.00127649	38	25.230		-9.712	0.0011924
39	24.860		-9.252	0.00163979	39	25.842		-10.324	0.00078038
40	34.221		-18.613	2.4938E-06	40	Undetermined		#VALUE!	#VALUE!
41	24.545		-8.937	0.00204006	41	24.157		-8.639	0.00250917
43	24.320		-8.712	0.00238404	43	23.846		-8.328	0.00311275
44	25.568		-9.960	0.00100431	44	24.776		-9.258	0.00163343
45	24.201		-8.593	0.00258948	45	24.313		-8.795	0.00225062
46	24.827		-9.219	0.00167752	46	26.237		-10.719	0.00059338
47	31.410		-15.802	1.7505E-05	47	30.818		-15.300	2.4788E-05
49	24.628		-9.020	0.00192564	49	24.891		-9.373	0.00150852
50	31.519		-15.911	1.6229E-05	50	31.261		-15.743	1.8236E-05
51	23.747		-8.139	0.00354691	51	23.562		-8.044	0.00378903
52	27.708		-12.100	0.00022785	52	27.427		-11.909	0.00026012
53	24.348		-8.740	0.00233939	53	23.871		-8.353	0.00305869
54	23.239		-7.631	0.00504524	54	22.716		-7.198	0.00680869
55	25.621		-10.013	0.00096804	55	25.928		-10.410	0.00073488
56	24.855		-9.247	0.00164589	56	24.724		-9.206	0.00169312
57	23.409		-7.801	0.00448399	57	22.987		-7.469	0.00564337
58	23.972		-8.364	0.00303467	58	21.257		-5.739	0.01872739
var2csa	13.342		2.266	4.80934026	var2csa	12.885		2.633	6.20505071
91	23.604		-7.996	0.00391606	91	24.662		-9.144	0.00176806
92	22.177		-6.569	0.01053504	92	22.201		-6.683	0.00973246
93	25.205		-9.597	0.00129096	93	24.977		-9.459	0.0014206
94	26.192		-10.584	0.00065153	94	25.600		-10.082	0.00092286
95	25.943		-10.335	0.00077407	95	26.481		-10.963	0.00050108
96	25.737		-10.129	0.00089297	96	25.194		-9.676	0.00122206
97	25.835		-10.227	0.00083461	97	24.831		-9.313	0.00157261
60	15.608		0.000	1.00022045	60	15.518		0.000	1.00031821

PfSAMS KD Parent							PfSAMS KD Clone A5				
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT		relative to p60	relative copy number
1	32.099	34.524		33.312	-14.655	3.87689E-05	1	Undetermined		#VALUE!	#VALUE!
3	23.327	23.259		23.293	-4.636	0.040226541	3	22.180		-6.265407	0.01299944
4	15.998	15.571		15.785	2.872	7.322424942	4	14.170		1.744878	3.3516651
5	23.702	23.774		23.738	-5.081	0.029547738	5	23.542		-7.626506	0.00506049
6	22.166	21.931		22.048	-3.391	0.095303169	6	21.954		-6.03945	0.01520353
7	20.176	19.737		19.957	-1.300	0.406164344	7	20.451		-4.536242	0.0430978
8	19.887	19.629		19.758	-1.101	0.466157699	8	20.827		-4.911736	0.03322157
9	23.287	23.232		23.260	-4.603	0.041157838	9	23.258		-7.3433	0.0061581
11	23.895	23.831		23.863	-5.206	0.027094221	11	23.353		-7.437957	0.00576703
12	22.108	21.892		22.000	-3.343	0.098548792	12	22.685		-6.770144	0.00916186
13	18.865	18.828		18.847	-0.190	0.876681677	13	19.343		-3.428414	0.09288478
15	16.299	16.145		16.222	2.435	5.407602101	15	22.436		-6.520518	0.01089252
17	22.392	22.479		22.436	-3.779	0.072850693	17	21.936		-6.020677	0.01540266
18	23.703	21.942		22.823	-4.166	0.055720596	18	22.906		-6.990655	0.00786327
19	22.976	22.728		22.852	-4.195	0.054603699	19	21.595		-5.680465	0.01949888
20	20.515	20.424		20.470	-1.813	0.28468374	20	22.887		-6.972138	0.00796484
21	23.346	22.860		23.103	-4.446	0.045874222	21	22.798		-6.88271	0.00847418
22	22.820	22.721		22.771	-4.114	0.057768126	22	23.185		-7.270118	0.00647853
23	22.745	22.722		22.734	-4.077	0.059265815	23	22.787		-6.871987	0.0085374
25	21.767	21.679		21.723	-3.066	0.1194429	25	16.382		-0.466605	0.72366555
26	15.321	15.182		15.251	3.406	10.59649876	26	14.363		1.552039	2.93231278
27	14.877	14.874		14.876	3.781	13.75063654	27	14.000		1.915176	3.77159823
28	22.637	22.770		22.703	-4.046	0.060523847	28	23.982		-8.066855	0.00372936
30	23.928	23.623		23.776	-5.119	0.02878482	30	24.384		-8.468705	0.00282271
31	21.529	21.513		21.521	-2.864	0.137366965	31	22.540		-6.624963	0.01013182
34	21.690	21.622		21.656	-2.999	0.125080084	34	23.149		-7.234372	0.00664105
35	20.327	20.268		20.297	-1.640	0.320806438	35	21.450		-5.535052	0.02156668
36	24.694	24.623		24.659	-6.002	0.015604895	36	23.891		-7.97608	0.00397156
37	24.908	24.651		24.779	-6.122	0.014353163	37	26.195		-10.279962	0.00080431
38	24.996	24.743		24.870	-6.213	0.013481899	38	24.149		-8.234426	0.0033204
39	22.396	22.187		22.291	-3.634	0.080537277	39	22.780		-6.864673	0.00858079
40	35.536	31.820		33.678	-15.021	3.00779E-05	40	31.203		-15.287799	2.4998E-05
41	20.877	20.543		20.710	-2.053	0.241015611	41	22.550		-6.635287	0.01005957
43	23.777	23.545		23.661	-5.004	0.031166739	43	23.161		-7.245957	0.00658794
44	16.260	15.968		16.114	2.543	5.828382256	44	15.572		0.343245	1.26860682
45	21.127	20.987		21.057	-2.400	0.189438636	45	20.860		-4.945415	0.03245501
46	21.642	21.538		21.590	-2.933	0.130955858	46	20.957		-5.041644	0.03036085
47	21.497	21.214		21.356	-2.699	0.154006698	47	20.434		-4.518516	0.0436306
49	20.991	20.714		20.853	-2.196	0.218287133	49	21.964		-6.048964	0.0151036
50	21.801	21.617		21.709	-3.052	0.120555594	50	20.347		-4.432092	0.04632414
51	21.619	21.378		21.498	-2.841	0.13954874	51	21.695		-5.780278	0.01819546
52	22.155	21.942		22.048	-3.391	0.095309214	52	24.410		-8.49511	0.00277151
53	20.902	20.707		20.805	-2.148	0.225664674	53	24.710		-8.794835	0.0022516
54	20.997	20.730		20.864	-2.207	0.216652532	54	22.123		-6.207534	0.0135315
55	21.833	21.732		21.783	-3.126	0.114567202	55	24.434		-8.519145	0.00272572
56	24.772	24.521		24.646	-5.989	0.015740362	56	24.244		-8.32945	0.00310875
57	19.683	19.349		19.516	-0.859	0.551432995	57	20.238		-4.323033	0.04996172
58	23.989	23.997		23.993	-5.336	0.02476198	58	21.763		-5.847983	0.01736128
var2csa	20.117	19.792		19.955	-1.298	0.40678616	var2csa	19.713		-3.797503	0.07191801
91	24.367	23.889		24.128	-5.471	0.022542971	91	23.437		-7.521733	0.00544168
92	14.648	14.652		14.650	4.007	16.08284242	92	13.777		2.138225	4.40220094
93	20.125	19.815		19.970	-1.313	0.402473165	93	19.138		-3.223092	0.10709091
94	16.336	16.131		16.233	2.424	5.365189023	94	15.543		0.37178	1.29394832
95	22.579	22.437		22.508	-3.851	0.069295477	95	23.707		-7.792298	0.00451112
96	21.722	21.504		21.613	-2.956	0.128894222	96	21.744		-5.828559	0.01759661
97	Undetermine	Undetermined		#DIV/0!	#DIV/0!	0	97	Undetermined		#VALUE!	#VALUE!
60	18.755	18.558		18.657	0.000	1.000217672	60	15.915		0.00023	1.00015944

PfSAMS KD Clone C8					PfSAMS KD Clone D6				
Primer pair	CT		relative to p60	relative copy number	Primer pair	CT		relative to p60	relative copy number
1	31.521		-15.213	2.6334E-05	1	32.602		-15.457	2.22294E-05
3	22.748		-6.440	0.0115145	3	23.664		-6.519	0.010907082
4	14.320		1.988	3.96721614	4	15.029		2.116	4.334975981
5	16.965		-0.657	0.63412537	5	23.842		-6.697	0.009638244
6	21.842		-5.534	0.02158102	6	21.130		-3.985	0.063167619
7	20.178		-3.870	0.06837193	7	20.274		-3.129	0.114325569
8	20.202		-3.894	0.06726678	8	20.448		-3.303	0.101347542
9	21.863		-5.555	0.02127561	9	19.559		-2.414	0.18757455
11	23.137		-6.829	0.00879396	11	24.477		-7.332	0.006204562
12	17.518		-1.210	0.43233454	12	18.183		-1.038	0.487050746
13	19.615		-3.307	0.10105496	13	20.020		-2.875	0.136359961
15	14.374		1.934	3.82096356	15	15.108		2.037	4.104979451
17	21.437		-5.129	0.02857804	17	22.923		-5.778	0.018222996
18	20.774		-4.466	0.04523957	18	23.146		-6.001	0.015610559
19	22.501		-6.193	0.01367307	19	23.491		-6.346	0.012297101
20	22.660		-6.352	0.01224551	20	23.246		-6.101	0.014569983
21	21.572		-5.264	0.02602731	21	23.543		-6.398	0.01186181
22	22.498		-6.190	0.01369317	22	23.636		-6.491	0.011121329
23	21.579		-5.271	0.0258941	23	22.914		-5.769	0.018334287
25	15.225		1.083	2.11908727	25	16.113		1.032	2.044285934
26	13.944		2.364	5.14895621	26	14.593		2.552	5.865088997
27	14.002		2.306	4.94357939	27	14.442		2.703	6.51120698
28	18.841		-2.533	0.17283389	28	19.170		-2.025	0.245627501
30	23.221		-6.913	0.00829928	30	24.837		-7.692	0.004834525
31	22.464		-6.156	0.01402612	31	23.559		-6.414	0.011730278
34	21.003		-4.695	0.03859412	34	23.623		-6.478	0.011216482
35	20.219		-3.911	0.06645419	35	21.624		-4.479	0.04484724
36	23.484		-7.176	0.00691331	36	24.750		-7.605	0.005137147
37	24.914		-8.606	0.002567	37	23.400		-6.255	0.013096907
38	23.349		-7.041	0.00759475	38	24.375		-7.230	0.006660748
39	22.468		-6.160	0.01398663	39	23.437		-6.292	0.012759618
40	28.515		-12.207	0.00021152	40	29.208		-12.063	0.000233767
41	21.413		-5.105	0.02906039	41	22.504		-5.359	0.024370134
43	16.561		-0.253	0.8393864	43	17.506		-0.361	0.778373231
44	13.990		2.318	4.98503454	44	15.440		1.705	3.260185378
45	21.912		-5.604	0.02055775	45	22.747		-5.602	0.020589106
46	21.835		-5.527	0.02168029	46	22.566		-5.421	0.023333993
47	19.755		-3.447	0.09170658	47	20.982		-3.837	0.069990213
49	20.509		-4.201	0.05436082	49	22.853		-5.708	0.019135561
50	19.720		-3.412	0.09392851	50	21.049		-3.904	0.066798245
51	22.373		-6.065	0.01494014	51	18.459		-1.314	0.402243218
52	23.232		-6.924	0.00823635	52	24.508		-7.363	0.006074612
53	19.412		-3.104	0.11629911	53	21.390		-4.245	0.05274876
54	21.848		-5.540	0.02149865	54	19.716		-2.571	0.168344092
55	23.776		-7.468	0.00564755	55	24.566		-7.421	0.005834319
56	24.420		-8.112	0.00361323	56	25.233		-8.088	0.003675981
57	Undetermined		#VALUE!	#VALUE!	57	Undetermined		#VALUE!	#VALUE!
58	21.851		-5.543	0.0214431	58	23.272		-6.127	0.014309331
var2csa	19.768		-3.460	0.0908632	var2csa	21.332		-4.187	0.054909135
91	23.347		-7.039	0.00760335	91	23.780		-6.635	0.01005995
92	13.728		2.580	5.97997934	92	14.443		2.702	6.505062868
93	19.971		-3.663	0.07892822	93	20.667		-3.522	0.087080772
94	14.952		1.356	2.55955499	94	16.249		0.896	1.860544633
95	23.244		-6.936	0.00816907	95	24.329		-7.184	0.00687854
96	18.898		-2.590	0.16609229	96	22.666		-5.521	0.021780472
97	Undetermined		#VALUE!	#VALUE!	97	Undetermined		#VALUE!	#VALUE!
60	16.308		0.000	1.00021767	60	17.145		0.000	1.000311965

PfSAMS KD Clone E1					PfSAMS KD Clone F12				
Primer pair	CT		relative to p60	relative copy number	Primer pair	CT		relative to p60	relative copy number
1	31.812		-14.394	4.64415E-05	1	34.458		-17.614	4.98394E-06
3	23.109		-5.691	0.019356514	3	Undetermined		#VALUE!	#VALUE!
4	15.201		2.217	4.64837996	4	15.251		1.593	3.017167937
5	23.842		-6.424	0.011647491	5	Undetermined		#VALUE!	#VALUE!
6	21.915		-4.497	0.044300721	6	22.894		-6.050	0.015088467
7	20.716		-3.298	0.101659152	7	20.946		-4.102	0.058235674
8	19.735		-2.317	0.200661118	8	21.168		-4.324	0.049930183
9	19.250		-1.832	0.280887631	9	23.163		-6.319	0.012524502
11	23.921		-6.503	0.011029032	11	24.317		-7.473	0.005626982
12	18.023		-0.605	0.657380242	12	21.840		-4.996	0.031340739
13	14.937		2.481	5.583102335	13	20.490		-3.646	0.079870137
15	14.790		2.628	6.180771799	15	14.856		1.988	3.965503345
17	21.314		-3.896	0.067178528	17	22.273		-5.429	0.023217927
18	23.241		-5.823	0.01766432	18	23.693		-6.849	0.008677337
19	23.112		-5.694	0.019310429	19	23.658		-6.814	0.008885927
20	22.420		-5.002	0.031214799	20	21.749		-4.905	0.033373227
21	23.719		-6.301	0.012680777	21	22.694		-5.850	0.01733486
22	22.380		-4.962	0.032088608	22	23.349		-6.505	0.011012288
23	22.638		-5.220	0.026827083	23	23.506		-6.662	0.009873477
25	16.197		1.221	2.331790217	25	16.584		0.260	1.197176612
26	14.585		2.833	7.126368074	26	14.469		2.375	5.185578701
27	14.417		3.001	8.008305439	27	14.386		2.458	5.494171711
28	18.916		-1.498	0.353944975	28	19.349		-2.505	0.176172665
30	24.691		-7.273	0.006466129	30	24.259		-7.415	0.005859345
31	22.127		-4.709	0.03823469	31	23.327		-6.483	0.011180302
34	22.103		-4.685	0.038880889	34	22.924		-6.080	0.014786045
35	20.997		-3.579	0.083651603	35	21.594		-4.750	0.037155304
36	24.601		-7.183	0.006883653	36	24.201		-7.357	0.006100385
37	26.145		-8.727	0.002359603	37	24.791		-7.947	0.004053809
38	24.581		-7.163	0.006976939	38	24.384		-7.540	0.005371959
39	21.381		-3.963	0.06412319	39	22.768		-5.924	0.016472428
40	25.728		-8.310	0.003152017	40	29.506		-12.662	0.000154314
41	16.711		0.707	1.632427591	41	22.102		-5.258	0.026133062
43	16.850		0.568	1.482286169	43	17.991		-1.147	0.451470304
44	14.624		2.794	6.936087026	44	15.129		1.715	3.283703806
45	22.889		-5.471	0.022543651	45	21.870		-5.026	0.03070182
46	22.601		-5.183	0.027521196	46	22.925		-6.081	0.014776783
47	21.157		-3.739	0.074898207	47	20.809		-3.965	0.064025304
49	23.203		-5.785	0.018132944	49	21.761		-4.917	0.033091351
50	20.993		-3.575	0.083921775	50	20.202		-3.358	0.097510068
51	15.092		2.326	5.01254749	51	22.291		-5.447	0.02292701
52	24.211		-6.793	0.009017686	52	24.111		-7.267	0.006491268
53	21.655		-4.237	0.053042295	53	22.337		-5.493	0.022212229
54	16.517		0.901	1.866930213	54	22.186		-5.342	0.024660434
55	24.435		-7.017	0.007722844	55	24.280		-7.436	0.005775522
56	25.410		-7.992	0.003927209	56	25.829		-8.985	0.001973388
57	Undetermined		#VALUE!	#VALUE!	57	20.602		-3.758	0.073926376
58	23.675		-6.257	0.01307725	58	23.435		-6.591	0.01037236
var2csa	20.356		-2.938	0.130531689	var2csa	21.354		-4.510	0.043902077
91	23.806		-6.388	0.011940665	91	22.397		-5.553	0.021292793
92	13.997		3.421	10.71254233	92	14.393		2.451	5.466187698
93	20.211		-2.793	0.144289078	93	20.994		-4.150	0.05634514
94	15.526		1.892	3.710282404	94	15.585		1.259	2.393092231
95	22.780		-5.362	0.024307882	95	24.318		-7.474	0.005623344
96	21.236		-3.818	0.070886613	96	22.488		-5.644	0.019998201
97	Undetermined		#VALUE!	#VALUE!	97	Undetermined		#VALUE!	#VALUE!
60	17.418		0.000	0.999875934	60	16.844		0.000	0.999961878

PfSAMS KD Clone G4					PfSAMS KD Clone G11						
Primer pair	CT		relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	Undetermined		#VALUE!	#VALUE!	1	29.146	29.215	29.181		-11.590	0.00032441
3	24.203		-6.261207	0.01303734	3	18.680	23.736	21.208		-3.617	0.08149653
4	16.133		1.808962	3.50390097	4	20.583	20.866	20.725		-3.134	0.11394689
5	25.772		-7.829883	0.00439512	5	25.208	25.711	25.460		-7.869	0.00427863
6	21.996		-4.054262	0.06019294	6	23.274	23.616	23.445		-5.854	0.01728987
7	20.840		-2.898286	0.13413094	7	20.334	20.676	20.505		-2.914	0.13267859
8	22.868		-4.92631	0.03288766	8	21.860	Undetermine	21.860		-4.269	0.05185339
9	23.936		-5.993915	0.01569104	9	25.644	17.701	21.673		-4.082	0.05906413
11	25.504		-7.562135	0.0052914	11	25.406	24.395	24.900		-7.309	0.00630556
12	23.068		-5.125652	0.02864343	12	18.427	21.785	20.106		-2.515	0.17493283
13	Undetermined		#VALUE!	#VALUE!	13	21.426	22.331	21.879		-4.288	0.05120731
15	16.281		1.661112	3.16260198	15	16.184	16.310	16.247		1.344	2.53848723
17	23.722		-5.780414	0.01819374	17	25.569	25.807	25.688		-8.097	0.00365258
18	24.812		-6.87024	0.00854775	18	24.758	25.496	25.127		-7.536	0.00538959
19	25.338		-7.3955	0.00593926	19	24.626	25.596	25.111		-7.520	0.00544747
20	22.512		-4.570115	0.04209769	20	17.382	16.402	16.892		0.699	1.62326327
21	22.906		-4.963556	0.03204946	21	24.821	25.755	25.288		-7.697	0.00481974
22	24.388		-6.445905	0.01147068	22	24.020	Undetermine	24.020		-6.429	0.01160435
23	24.756		-6.813793	0.00888882	23	24.476	25.474	24.975		-7.384	0.00598529
25	17.778		0.16416	1.12051348	25	15.720	16.838	16.279		1.312	2.48274269
26	15.963		1.979477	3.94350098	26	15.320	15.945	15.632		1.959	3.88725163
27	15.616		2.325951	5.01396178	27	14.325	14.808	14.567		3.024	8.13577341
28	20.923		-2.981235	0.12663648	28	18.559	18.298	18.429		-0.838	0.55959152
30	25.755		-7.8128	0.00444747	30	25.775	25.990	25.883		-8.292	0.00319149
31	24.724		-6.781883	0.00908761	31	23.665	24.498	24.081		-6.490	0.01112471
34	24.798		-6.855583	0.00863503	34	24.391	24.801	24.596		-7.005	0.00778534
35	22.117		-4.175437	0.0553437	35	22.194	22.729	22.462		-4.871	0.03418377
36	25.461		-7.518527	0.00545378	36	25.183	25.467	25.325		-7.734	0.0046966
37	26.355		-8.41304	0.00293375	37	26.562	27.529	27.046		-9.455	0.00142494
38	25.737		-7.795293	0.00450177	38	25.504	26.405	25.955		-8.364	0.0030361
39	25.156		-7.214445	0.00673342	39	23.895	24.891	24.393		-6.802	0.00896108
40	33.620		-15.67778	1.9077E-05	40	30.353	27.434	28.893		-11.302	0.000396
41	23.965		-6.023078	0.01537704	41	22.972	Undetermine	22.972		-5.381	0.02399682
43	19.653		-1.711273	0.30539048	43	18.562	18.787	18.674		-1.083	0.47190243
44	16.467		1.475222	2.78026423	44	16.223	Undetermine	16.223		1.368	2.58125917
45	24.310		-6.367937	0.01210765	45	14.800	15.630	15.215		2.376	5.19090649
46	23.431		-5.488723	0.02227049	46	23.262	18.998	21.130		-3.539	0.08603142
47	21.922		-3.979803	0.06338112	47	21.044	21.531	21.287		-3.696	0.07713805
49	23.909		-5.96659	0.01599107	49	23.824	24.798	24.311		-6.720	0.00948497
50	22.192		-4.249574	0.05257155	50	21.400	21.719	21.560		-3.969	0.0638794
51	23.814		-5.871711	0.01707807	51	23.138	Undetermine	23.138		-5.547	0.02138822
52	24.221		-6.27865	0.01288066	52	23.438	23.960	23.699		-6.108	0.01449656
53	23.947		-6.004932	0.01557168	53	22.913	26.402	24.657		-7.066	0.0074607
54	20.126		-2.18394	0.22007391	54	23.475	24.211	23.843		-6.252	0.01312074
55	25.755		-7.813182	0.00444629	55	24.893	16.360	20.627		-3.036	0.12195679
56	20.514		-2.572126	0.16815621	56	26.512	26.975	26.744		-9.153	0.00175693
57	19.698		-1.755506	0.2961693	57	Undetermine	Undetermine	#DIV/0!		#DIV/0!	#DIV/0!
58	24.218		-6.27591	0.01290514	58	21.857	22.567	22.212		-4.621	0.04064621
var2csa	22.830		-4.88794	0.03377407	var2csa	15.257	16.161	15.709		1.882	3.68611607
91	25.588		-7.646463	0.00499097	91	24.410	24.827	24.618		-7.027	0.00766609
92	15.512		2.429565	5.38730969	92	14.250	14.999	14.625		2.966	7.81587286
93	22.113		-4.171098	0.0555104	93	23.200	24.298	23.749		-6.158	0.01400681
94	16.976		0.96594	1.95333582	94	15.877	16.308	16.092		1.499	2.8261842
95	25.716		-7.773542	0.00457015	95	24.522	25.302	24.912		-7.321	0.00625271
96	20.186		-2.244445	0.21103512	96	22.663	23.390	23.026		-5.435	0.02311407
97	Undetermined		#VALUE!	#VALUE!	97	Undetermine	Undetermine	#DIV/0!		#DIV/0!	#DIV/0!
60	17.942		0.000458	1.00031751	60	17.030	18.152	17.591		0.000	0.99992341

PfSAMS KD Clone H2						
Primer pair	CT			CT Average	relative to p60	relative copy number
1	35.230	33.193	34.211		-14.377	4.69841E-05
3	26.652	26.592	26.622		-6.788	0.009047676
4	26.278	26.967	26.623		-6.789	0.00904582
5	26.300	Undetermine	26.300		-6.466	0.011310316
6	26.948	26.934	26.941		-7.107	0.007253683
7	22.193	22.762	22.477		-2.643	0.160041061
8	24.793	25.562	25.177		-5.343	0.02463024
9	27.183	27.244	27.214		-7.380	0.006005008
11	28.259	28.482	28.370		-8.536	0.002693764
12	20.444	22.313	21.378		-1.544	0.34283065
13	24.175	25.001	24.588		-4.754	0.037065724
15	19.121	18.915	19.018		0.816	1.760039735
17	Undetermine	25.996	25.996		-6.162	0.01396822
18	27.211	26.976	27.094		-7.260	0.006525151
19	25.716	25.976	25.846		-6.012	0.015495112
20	21.419	21.439	21.429		-1.595	0.331008232
21	27.861	27.940	27.900		-8.066	0.003730852
22	25.342	26.093	25.717		-5.883	0.01694082
23	25.430	26.319	25.874		-6.040	0.015196585
25	18.705	19.915	19.310		0.524	1.43786826
26	18.135	18.864	18.500		1.334	2.521662154
27	17.291	17.634	17.463		2.371	5.1735856
28	22.193	21.003	21.598		-1.764	0.294494889
30	27.856	28.827	28.341		-8.507	0.002748265
31	25.558	26.330	25.944		-6.110	0.014478596
34	17.487	18.489	17.988		1.846	3.595507702
35	26.674	27.116	26.895		-7.061	0.007488814
36	23.452	23.718	23.585		-3.751	0.074276441
37	25.582	26.125	25.854		-6.020	0.015410976
38	28.217	29.238	28.727		-8.893	0.002103027
39	26.503	27.648	27.075		-7.241	0.006610016
40	29.870	29.622	29.746		-9.912	0.001037822
41	19.583	20.255	19.919		-0.085	0.942809696
43	22.097	21.857	21.977		-2.143	0.226344554
44	19.303	20.125	19.714		0.120	1.086763864
45	26.606	26.450	26.528		-6.694	0.009659271
46	24.450	24.716	24.583		-4.749	0.037186789
47	23.839	23.709	23.774		-3.940	0.065145011
49	26.775	27.015	26.895		-7.061	0.007487965
50	23.720	24.180	23.950		-4.116	0.057673884
51	26.475	27.429	26.952		-7.118	0.007199406
52	25.861	26.212	26.037		-6.203	0.013578228
53	23.910	24.799	24.354		-4.520	0.043571135
54	18.704	19.525	19.115		0.719	1.646446918
55	Undetermine	28.504	28.504		-8.670	0.002454368
56	27.820	27.568	27.694		-7.860	0.004304286
57	Undetermine	23.989	23.989		-4.155	0.056138721
58	22.985	24.210	23.597		-3.763	0.073646231
var2csa	25.401	25.960	25.680		-5.846	0.017383007
91	25.376	26.244	25.810		-5.976	0.015884539
92	17.964	18.646	18.305		1.529	2.885799365
93	26.655	27.191	26.923		-7.089	0.007342856
94	19.956	20.972	20.464		-0.630	0.646170145
95	26.469	27.313	26.891		-7.057	0.007508146
96	25.318	25.830	25.574		-5.740	0.018712764
97	Undetermine	Undetermine	#DIV/0!		#DIV/0!	#DIV/0!
60	19.310	20.359	19.834		0.000	0.99984648

Chapter 4.1 *var* panels (TI on for 36-48 hours)

(Rep 1) MCM Day 0							(Rep 1) MCM						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	22.368	21.997		22.183	-4.852	0.03462982	1	29.490	29.966		29.728	-4.415	0.04687258
3	23.616	23.628		23.622	-6.291	0.01276926	3	33.464	31.717		32.590	-7.277	0.00644606
4	23.122	22.828		22.975	-5.644	0.01999293	4	30.620	30.585		30.602	-5.289	0.02557248
5	23.100	22.972		23.036	-5.705	0.01917102	5	32.779	30.883		31.831	-6.518	0.01091114
6	22.142	22.007		22.075	-4.744	0.03732665	6	30.363	29.865		30.114	-4.801	0.03588288
7	22.630	22.388		22.509	-5.178	0.02762189	7	29.237	29.731		29.484	-4.171	0.055509
8	22.973	22.860		22.917	-5.586	0.02082235	8	31.798	30.137		30.967	-5.654	0.01985482
9	22.508	22.356		22.432	-5.101	0.02914154	9	30.428	30.940		30.684	-5.371	0.02416513
11	24.404	24.649		24.526	-7.195	0.00682435	11	30.358	30.454		30.406	-5.093	0.0292976
12	23.315	23.140		23.228	-5.897	0.01678265	12	30.533	31.335		30.934	-5.621	0.02032134
13	21.372	21.116		21.244	-3.913	0.06637563	13	29.423	29.100		29.262	-3.949	0.06475873
15	21.743	21.643		21.693	-4.362	0.04862646	15	31.108	30.589		30.848	-5.535	0.02156467
17	21.319	21.009		21.164	-3.833	0.07016899	17	30.123	29.581		29.852	-4.539	0.04302093
18	20.746	20.592		20.669	-3.338	0.09888068	18	28.099	28.639		28.369	-3.056	0.12024597
19	21.363	21.363		21.363	-4.032	0.06112659	19	29.767	30.248		30.008	-4.695	0.03861652
20	23.931	23.952		23.942	-6.611	0.01023276	20	33.825	32.346		33.085	-7.772	0.00457355
21	22.004	21.782		21.893	-4.562	0.04233832	21	31.643	31.534		31.588	-6.275	0.01290959
22	21.950	21.719		21.834	-4.503	0.04408765	22	29.258	28.731		28.994	-3.681	0.07794682
23	23.188	22.980		23.084	-5.753	0.01853807	23	33.521	33.285		33.403	-8.090	0.00367094
25	24.646	24.471		24.559	-7.228	0.00667089	25	38.080	31.468		34.774	-9.461	0.0014192
26	21.756	21.632		21.694	-4.363	0.0485936	26	30.811	29.839		30.325	-5.012	0.03099348
27	19.931	19.808		19.870	-2.539	0.17212147	27	28.815	28.970		28.892	-3.579	0.08365903
28	22.993	22.856		22.925	-5.594	0.02070597	28	30.723	30.099		30.411	-5.098	0.02920508
30	23.395	23.116		23.255	-5.924	0.01646658	30	31.159	34.257		32.708	-7.395	0.00594074
31	18.959	18.971		18.965	-1.634	0.32220047	31	27.747	27.545		27.646	-2.333	0.19852389
34	20.598	20.452		20.525	-3.194	0.10924643	34	27.858	28.219		28.038	-2.725	0.15123327
35	23.440	23.410		23.425	-6.094	0.01463674	35	35.847	32.674		34.260	-8.947	0.00202591
36	22.785	22.662		22.723	-5.392	0.0238067	36	31.180	31.961		31.570	-6.257	0.013071
37	21.209	21.118		21.164	-3.833	0.07018351	37	30.413	31.193		30.803	-5.490	0.02225014
38	22.449	22.297		22.373	-5.042	0.03035968	38	30.849	30.126		30.487	-5.174	0.02769781
39	22.727	22.622		22.674	-5.343	0.02463006	39	28.995	28.921		28.958	-3.645	0.07992765
40	35.561	33.562		34.561	-17.230	6.5029E-06	40	36.377	28.393		32.385	-7.072	0.00743061
41	25.779	25.625		25.702	-8.371	0.00302115	41	31.659	30.333		30.996	-5.683	0.01946523
43	22.701	22.436		22.568	-5.237	0.02650737	43	31.960	29.777		30.869	-5.556	0.02125793
44	23.547	23.544		23.546	-6.215	0.01346583	44	31.933	30.813		31.373	-6.060	0.0149912
45	20.989	20.782		20.886	-3.555	0.08508667	45	30.159	29.511		29.835	-4.522	0.0435304
46	22.373	22.320		22.346	-5.015	0.0309179	46	33.193	Undetermined		33.193	-7.880	0.00424544
47	27.409	27.805		27.607	-10.276	0.00080671	47	Undetermine	38.603		38.603	-13.290	9.9831E-05
49	21.571	21.448		21.509	-4.178	0.05523814	49	29.399	30.590		29.994	-4.681	0.03897737
50	22.712	22.403		22.557	-5.226	0.02671111	50	30.332	30.181		30.257	-4.944	0.03249073
51	21.977	21.984		21.981	-4.650	0.03983079	51	29.761	29.210		29.485	-4.172	0.0554666
52	23.013	23.165		23.089	-5.758	0.01847633	52	31.767	32.220		31.994	-6.681	0.00974791
53	21.367	21.185		21.276	-3.945	0.06492982	53	31.323	30.119		30.721	-5.408	0.02355448
54	14.599	14.452		14.526	2.805	6.98999052	54	22.690	22.635		22.662	2.651	6.28038282
55	22.461	22.241		22.351	-5.020	0.03081519	55	30.295	29.143		29.719	-4.406	0.04716147
56	22.908	14.160		18.534	-1.203	0.4343032	56	26.539	27.252		26.895	-1.582	0.33400035
57	22.459	22.013		22.236	-4.905	0.03337817	57	29.883	31.189		30.536	-5.223	0.0267789
58	23.580	23.342		23.461	-6.130	0.014276	58	30.860	31.096		30.978	-5.665	0.01970367
var2csa	20.516	20.229		20.373	-3.042	0.12141773	var2csa	27.952	27.984		27.968	-2.655	0.15875374
91	21.000	20.821		20.911	-3.580	0.08362224	91	29.758	28.525		29.141	-3.828	0.07039867
92	20.798	20.724		20.761	-3.430	0.09281064	92	29.108	29.404		29.256	-3.943	0.06502262
93	21.640	21.628		21.634	-4.303	0.05067149	93	29.741	30.825		30.283	-4.970	0.03191196
94	24.320	24.263		24.291	-6.960	0.00802977	94	34.182	31.184		32.683	-7.370	0.00604675
95	22.235	22.921		22.578	-5.247	0.02633019	95	30.947	29.688		30.318	-5.005	0.0311488
96	24.499	24.137		24.318	-6.987	0.007883	96	30.624	30.512		30.568	-5.255	0.02618617
97	24.727	24.238		24.482	-7.151	0.00703528	97	32.519	31.665		32.092	-6.779	0.00910476
60	17.531	17.130		17.331	0.000	1.000122	60	25.296	25.329		25.313	0.000	1.00018994

(Rep 1, 1) 1mM TI							(Rep 1, 2) 1mM TI						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	25.135	24.910		25.022	-6.609	0.01024154	1	24.654	24.682		24.668	-6.306	0.01263547
3	26.484	26.517		26.501	-8.088	0.00367615	3	26.556	26.371		26.464	-8.102	0.00364016
4	25.796	25.839		25.818	-7.405	0.00590221	4	25.234	25.297		25.266	-6.904	0.00835135
5	26.017	25.598		25.808	-7.395	0.00594237	5	25.473	25.390		25.431	-7.069	0.00744683
6	24.712	24.707		24.709	-6.296	0.01272368	6	24.227	23.982		24.105	-5.743	0.01867631
7	24.189	24.248		24.219	-5.806	0.01787755	7	23.832	23.715		23.774	-5.412	0.02349331
8	25.741	25.530		25.636	-7.223	0.00669575	8	25.275	25.100		25.187	-6.825	0.00881761
9	25.187	25.174		25.181	-6.768	0.00917703	9	24.742	24.917		24.829	-6.467	0.01130075
11	27.631	28.242		27.936	-9.523	0.00135895	11	27.122	26.469		26.796	-8.434	0.00289214
12	25.306	25.233		25.270	-6.857	0.00862826	12	25.204	24.772		24.988	-6.626	0.01012579
13	23.457	23.308		23.383	-4.970	0.03191172	13	23.178	22.939		23.059	-4.697	0.03855964
15	24.670	24.884		24.777	-6.364	0.01214338	15	24.506	24.315		24.411	-6.049	0.01510749
17	23.644	23.933		23.789	-5.376	0.02408734	17	23.329	23.214		23.271	-4.909	0.03327388
18	23.149	22.658		22.904	-4.491	0.04448508	18	22.378	22.332		22.355	-3.993	0.06281683
19	23.834	24.120		23.977	-5.564	0.02113756	19	23.443	23.306		23.374	-5.012	0.0309859
20	27.413	27.308		27.361	-8.948	0.00202536	20	26.465	26.849		26.657	-8.295	0.00318406
21	24.925	24.978		24.952	-6.539	0.01075513	21	24.582	24.345		24.463	-6.101	0.01456438
22	23.934	23.728		23.831	-5.418	0.02338699	22	23.483	23.253		23.368	-5.006	0.03111578
23	26.324	25.926		26.125	-7.712	0.00476866	23	25.608	25.350		25.479	-7.117	0.00720307
25	28.228	27.374		27.801	-9.388	0.00149272	25	27.215	27.377		27.296	-8.934	0.00204461
26	25.355	25.242		25.299	-6.886	0.00845737	26	25.005	24.761		24.883	-6.521	0.01088723
27	22.977	22.757		22.867	-4.454	0.04562358	27	22.527	22.499		22.513	-4.151	0.05629109
28	25.851	25.866		25.859	-7.446	0.00573648	28	25.530	25.422		25.476	-7.114	0.00721914
30	25.890	26.091		25.991	-7.578	0.00523526	30	25.851	25.894		25.872	-7.510	0.00548424
31	22.338	21.967		22.152	-3.739	0.07486992	31	21.896	21.812		21.854	-3.492	0.08888036
34	22.760	22.428		22.594	-4.181	0.05513843	34	21.980	21.859		21.920	-3.558	0.08492655
35	26.653	26.891		26.772	-8.359	0.00304614	35	26.504	26.273		26.389	-8.027	0.00383469
36	26.405	25.738		26.072	-7.659	0.00494823	36	25.525	25.206		25.366	-7.004	0.00779343
37	25.784	25.527		25.656	-7.243	0.00660366	37	25.632	25.356		25.494	-7.132	0.00712806
38	24.320	24.191		24.256	-5.843	0.01742512	38	24.216	24.119		24.167	-5.805	0.01788132
39	25.515	25.276		25.396	-6.983	0.00790637	39	25.160	24.850		25.005	-6.643	0.01000416
40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!	40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
41	26.892	26.998		26.945	-8.532	0.00270158	41	26.880	26.823		26.852	-8.490	0.00278158
43	25.223	25.146		25.184	-6.771	0.00915436	43	24.602	24.295		24.449	-6.087	0.01471564
44	27.451	27.232		27.341	-8.928	0.00205277	44	26.923	26.544		26.733	-8.371	0.00301998
45	24.155	24.162		24.158	-5.745	0.01864098	45	24.325	23.787		24.056	-5.694	0.01931663
46	25.375	25.493		25.434	-7.021	0.0076992	46	24.584	24.326		24.455	-6.093	0.0146481
47	30.860	30.914		30.887	-12.474	0.00017578	47	29.704	29.832		29.768	-11.406	0.00036848
49	24.964	24.873		24.918	-6.505	0.0110079	49	24.716	24.480		24.598	-6.236	0.01326449
50	26.444	25.783		26.114	-7.701	0.00480727	50	25.841	24.562		25.202	-6.840	0.00873018
51	25.565	25.308		25.436	-7.023	0.00768733	51	25.085	24.488		24.787	-6.425	0.01164138
52	26.310	26.216		26.263	-7.850	0.00433444	52	25.473	25.488		25.481	-7.119	0.00719636
53	24.352	24.115		24.233	-5.820	0.01769759	53	23.903	23.715		23.809	-5.447	0.02292263
54	17.537	17.289		17.413	1.000	1.99991336	54	17.129	16.766		16.948	1.414	2.6655126
55	25.292	25.132		25.212	-6.799	0.00897772	55	25.209	25.160		25.185	-6.823	0.00883474
56	24.775	25.008		24.892	-6.479	0.01121327	56	24.690	24.571		24.630	-6.268	0.01297241
57	23.860	23.460		23.660	-5.247	0.02633421	57	22.969	22.709		22.839	-4.477	0.04490116
58	25.314	25.335		25.325	-6.912	0.00830508	58	25.503	25.295		25.399	-7.037	0.00761446
var2csa	23.390	23.141		23.265	-4.852	0.03461407	var2csa	23.003	22.889		22.946	-4.584	0.04169812
91	23.840	23.522		23.681	-5.268	0.02594761	91	23.308	23.198		23.253	-4.891	0.03369985
92	22.946	22.926		22.936	-4.523	0.04350165	92	22.755	22.739		22.747	-4.385	0.04785746
93	24.585	24.596		24.591	-6.178	0.01381242	93	24.204	24.013		24.108	-5.746	0.01862875
94	27.634	27.882		27.758	-9.345	0.00153808	94	26.496	27.240		26.868	-8.506	0.00275036
95	25.202	25.749		25.475	-7.062	0.0074821	95	25.413	25.359		25.386	-7.024	0.00768159
96	24.777	24.903		24.840	-6.427	0.01162215	96	24.795	24.766		24.780	-6.418	0.01169133
97	26.463	26.485		26.474	-8.061	0.00374383	97	25.706	25.369		25.538	-7.176	0.00691747
60	18.610	18.216		18.413	0.000	1.00011576	60	18.540	18.185		18.362	0.000	0.99968917

(Rep 2) MCM Day 0							(Rep 2) MCM						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	25.969	25.761		25.865	-5.538	0.02152246	1	26.002	25.824		25.913	-6.092	0.01465795
3	25.809	25.771		25.790	-5.463	0.02266742	3	26.404	25.965		26.184	-6.363	0.01214762
4	26.321	25.899		26.110	-5.783	0.01816401	4	25.726	25.868		25.797	-5.976	0.01588827
5	27.554	27.485		27.520	-7.193	0.00683539	5	27.746	27.484		27.615	-7.794	0.00450532
6	25.762	25.651		25.707	-5.380	0.02401985	6	26.245	25.665		25.955	-6.134	0.01424209
7	26.032	25.706		25.869	-5.542	0.02146314	7	26.237	26.299		26.268	-6.447	0.01146229
8	25.381	25.171		25.276	-4.949	0.03237737	8	25.151	25.263		25.207	-5.386	0.02391599
9	25.220	24.821		25.020	-4.693	0.03865306	9	25.650	25.142		25.396	-5.575	0.0209777
11	27.359	27.665		27.512	-7.185	0.00687208	11	27.690	29.154		28.422	-8.601	0.00257511
12	25.451	25.280		25.365	-5.038	0.03042843	12	25.467	25.131		25.299	-5.478	0.02243284
13	21.543	21.297		21.420	-1.093	0.46865916	13	21.871	21.681		21.776	-1.955	0.25792383
15	24.766	24.765		24.765	-4.438	0.04612487	15	25.695	25.403		25.549	-5.728	0.0188683
17	24.458	24.634		24.546	-4.219	0.05370088	17	24.599	24.645		24.622	-4.801	0.0358725
18	25.147	25.122		25.134	-4.807	0.03571363	18	25.018	25.188		25.103	-5.282	0.02570474
19	25.618	25.623		25.620	-5.293	0.02549977	19	25.526	25.786		25.656	-5.835	0.01751918
20	27.263	27.214		27.238	-6.911	0.00830884	20	27.121	27.076		27.099	-7.278	0.00644506
21	26.913	26.951		26.932	-6.605	0.01027329	21	26.670	26.765		26.718	-6.897	0.00839226
22	23.644	23.634		23.639	-3.312	0.10072101	22	24.196	23.869		24.033	-4.212	0.0539726
23	27.201	27.132		27.166	-6.839	0.00873222	23	27.414	27.291		27.353	-7.532	0.00540464
25	27.228	26.976		27.102	-6.775	0.00913075	25	27.861	27.702		27.781	-7.960	0.00401479
26	24.867	24.891		24.879	-4.552	0.04263809	26	25.108	25.501		25.305	-5.484	0.02235041
27	22.512	22.302		22.407	-2.080	0.23644885	27	22.845	22.858		22.852	-3.031	0.12236946
28	25.968	25.788		25.878	-5.551	0.02132861	28	26.629	26.255		26.442	-6.621	0.01015822
30	26.906	26.783		26.845	-6.518	0.01091488	30	26.716	27.114		26.915	-7.094	0.00731915
31	21.591	21.462		21.527	-1.200	0.4354245	31	21.922	21.811		21.866	-2.045	0.24225024
34	24.300	24.382		24.341	-4.014	0.0618859	34	24.677	24.379		24.528	-4.707	0.03828629
35	26.813	27.306		27.059	-6.732	0.00940508	35	27.570	27.577		27.573	-7.752	0.00463756
36	26.447	26.289		26.368	-6.041	0.0151848	36	26.735	26.543		26.639	-6.818	0.00886123
37	23.146	22.752		22.949	-2.622	0.16244002	37	23.300	23.311		23.305	-3.484	0.0893473
38	26.496	26.091		26.293	-5.966	0.01599368	38	25.936	25.568		25.752	-5.931	0.01638919
39	26.759	26.517		26.638	-6.311	0.012596	39	26.880	26.259		26.569	-6.748	0.0093004
40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!	40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
41	25.498	25.499		25.498	-5.171	0.02774975	41	25.912	25.833		25.873	-6.052	0.01507702
43	25.330	25.257		25.293	-4.966	0.0319872	43	25.314	25.266		25.290	-5.469	0.02258147
44	26.105	25.802		25.954	-5.627	0.02024166	44	25.991	32.805		29.398	-9.577	0.00130914
45	25.521	25.210		25.365	-5.038	0.03042875	45	25.347	25.953		25.650	-5.829	0.01759399
46	25.887	26.157		26.022	-5.695	0.01930283	46	26.014	26.508		26.261	-6.440	0.01151992
47	30.266	30.231		30.249	-9.922	0.0010311	47	30.246	30.771		30.509	-10.688	0.00060632
49	24.528	24.486		24.507	-4.180	0.05517073	49	24.852	24.794		24.823	-5.002	0.03121484
50	25.488	25.477		25.483	-5.156	0.02805375	50	25.914	25.954		25.934	-6.113	0.0144458
51	26.140	26.545		26.343	-6.016	0.01545672	51	26.768	26.293		26.530	-6.709	0.00955644
52	24.448	24.507		24.477	-4.150	0.05631478	52	24.793	24.591		24.692	-4.871	0.0341775
53	24.606	24.352		24.479	-4.152	0.0562424	53	24.778	24.645		24.711	-4.890	0.03371845
54	17.910	17.759		17.834	2.493	5.6284454	54	18.566	18.282		18.424	1.397	2.63325001
55	24.561	23.940		24.250	-3.923	0.06590953	55	24.546	24.413		24.479	-4.658	0.03960358
56	25.895	25.572		25.734	-5.407	0.02357488	56	25.873	25.738		25.806	-5.985	0.01578972
57	25.281	24.922		25.101	-4.774	0.03653704	57	25.516	25.243		25.379	-5.558	0.02121982
58	25.273	25.347		25.310	-4.983	0.03162634	58	25.229	24.737		24.983	-5.162	0.02793439
var2csa	20.437	20.311		20.374	-0.047	0.96790174	var2csa	20.174	20.188		20.181	-0.360	0.77911111
91	23.796	23.846		23.821	-3.494	0.08877148	91	24.304	23.925		24.114	-4.293	0.05099551
92	23.524	23.354		23.439	-3.112	0.11565448	92	23.564	23.411		23.488	-3.667	0.0787391
93	23.800	23.793		23.796	-3.469	0.09027733	93	24.254	24.216		24.235	-4.414	0.046909
94	27.168	27.491		27.330	-7.003	0.00779784	94	27.394	27.627		27.510	-7.689	0.00484521
95	24.773	24.564		24.669	-4.342	0.04931564	95	24.740	24.544		24.642	-4.821	0.03537634
96	27.167	26.675		26.921	-6.594	0.01035408	96	27.892	27.156		27.524	-7.703	0.0047982
97	27.934	27.584		27.759	-7.432	0.00579099	97	28.198	28.118		28.158	-8.337	0.00309275
60	20.431	20.223		20.327	0.000	0.99984613	60	19.903	19.738		19.821	0.000	1.00027799

(Rep 2, 1) 1mM TI							(Rep 2, 2) 1mM TI						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	23.951	23.924		23.938	-4.482	0.04476395	1	25.152	24.888		25.020	-6.396	0.01187576
3	25.173	24.925		25.049	-5.593	0.02071113	3	25.830	25.610		25.720	-7.096	0.00731064
4	25.225	24.789		25.007	-5.551	0.02132707	4	25.725	25.665		25.695	-7.071	0.00743797
5	25.812	25.651		25.732	-6.276	0.01290696	5	26.483	26.690		26.587	-7.963	0.00400885
6	23.392	23.541		23.466	-4.010	0.06204929	6	24.484	24.748		24.616	-5.992	0.01571284
7	23.579	23.402		23.490	-4.034	0.06102571	7	25.139	24.799		24.969	-6.345	0.01230399
8	24.407	24.296		24.352	-4.896	0.03358938	8	25.677	25.497		25.587	-6.963	0.00801658
9	23.566	23.682		23.624	-4.168	0.05562914	9	24.633	24.837		24.735	-6.111	0.01446631
11	25.208	25.003		25.106	-5.650	0.01992155	11	25.988	25.765		25.877	-7.253	0.00655762
12	23.381	23.476		23.429	-3.973	0.06369359	12	24.660	24.298		24.479	-5.855	0.01727729
13	21.136	21.193		21.164	-1.708	0.30597825	13	21.374	21.327		21.350	-2.726	0.15109449
15	23.770	23.641		23.705	-4.249	0.05257588	15	24.110	24.464		24.287	-5.663	0.01973519
17	22.706	22.531		22.619	-3.163	0.11165432	17	24.006	23.926		23.966	-5.342	0.02465328
18	23.253	23.250		23.252	-3.796	0.07199517	18	24.629	24.844		24.736	-6.112	0.01445652
19	23.562	23.287		23.424	-3.968	0.06388174	19	24.877	24.998		24.938	-6.314	0.01257228
20	25.177	25.013		25.095	-5.639	0.02006992	20	26.097	25.928		26.013	-7.389	0.00596753
21	24.386	24.270		24.328	-4.872	0.03415669	21	25.443	25.237		25.340	-6.716	0.00951377
22	22.230	22.131		22.181	-2.725	0.15125251	22	23.282	23.026		23.154	-4.530	0.04327992
23	25.322	25.240		25.281	-5.825	0.01763905	23	25.963	26.018		25.990	-7.366	0.00606024
25	25.590	25.663		25.626	-6.170	0.01388702	25	26.463	26.407		26.435	-7.811	0.00445185
26	23.250	22.934		23.092	-3.636	0.08043173	26	23.589	23.643		23.616	-4.992	0.03142066
27	21.635	21.605		21.620	-2.164	0.22315608	27	22.760	22.637		22.698	-4.074	0.05935461
28	24.944	24.789		24.867	-5.411	0.02351065	28	24.913	24.843		24.878	-6.254	0.0131036
30	25.500	24.974		25.237	-5.781	0.01818856	30	26.138	26.226		26.182	-7.558	0.00530753
31	20.583	20.508		20.546	-1.090	0.46989245	31	20.974	20.810		20.892	-2.268	0.20764305
34	22.457	22.132		22.294	-2.838	0.13982905	34	22.982	22.731		22.857	-4.233	0.05318864
35	24.751	24.592		24.671	-5.215	0.02691754	35	25.634	25.511		25.573	-6.949	0.00809495
36	24.789	24.862		24.826	-5.370	0.02418781	36	25.907	25.657		25.782	-7.158	0.00700308
37	24.848	24.508		24.678	-5.222	0.02678852	37	22.761	22.576		22.669	-4.045	0.06058826
38	24.618	24.549		24.583	-5.127	0.02861212	38	25.458	25.563		25.511	-6.887	0.00844974
39	24.414	24.532		24.473	-5.017	0.03088337	39	24.896	24.736		24.816	-6.192	0.01367579
40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!	40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
41	24.397	24.259		24.328	-4.872	0.03415268	41	25.155	24.886		25.021	-6.397	0.01187007
43	23.398	23.190		23.294	-3.838	0.06992834	43	24.123	24.249		24.186	-5.562	0.02116615
44	24.942	24.504		24.723	-5.267	0.02597196	44	26.176	25.753		25.964	-7.340	0.0061706
45	22.857	22.833		22.845	-3.389	0.09545083	45	23.464	23.626		23.545	-4.921	0.03301334
46	24.271	23.900		24.086	-4.630	0.04039881	46	25.682	25.347		25.515	-6.891	0.00842698
47	29.335	29.510		29.422	-9.966	0.00099953	47	30.704	31.167		30.936	-12.312	0.0001967
49	23.288	23.347		23.317	-3.861	0.06880501	49	23.418	23.680		23.549	-4.925	0.03291632
50	23.828	23.510		23.669	-4.213	0.05392272	50	24.792	24.697		24.745	-6.121	0.0143723
51	24.217	23.903		24.060	-4.604	0.04111966	51	25.684	25.163		25.423	-6.799	0.00897826
52	24.017	23.789		23.903	-4.447	0.04584294	52	24.892	24.657		24.775	-6.151	0.0140729
53	23.609	23.291		23.450	-3.994	0.06275782	53	25.021	24.716		24.869	-6.245	0.0131886
54	16.879	16.599		16.739	2.717	6.57389767	54	17.825	17.663		17.744	0.880	1.84053285
55	23.666	23.663		23.665	-4.209	0.05408791	55	24.655	24.512		24.583	-5.959	0.01607186
56	24.400	24.325		24.362	-4.906	0.03334932	56	25.606	25.366		25.486	-6.862	0.0085971
57	23.588	23.293		23.440	-3.984	0.06318281	57	24.785	24.371		24.578	-5.954	0.01613406
58	23.710	23.792		23.751	-4.295	0.05094998	58	23.496	23.534		23.515	-4.891	0.0337096
var2csa	18.337	18.216		18.277	1.179	2.26492299	var2csa	18.960	18.891		18.925	-0.301	0.81162123
91	21.958	21.845		21.902	-2.446	0.1835771	91	23.567	23.397		23.482	-4.858	0.03448548
92	22.023	22.453		22.238	-2.782	0.14537771	92	23.171	22.893		23.032	-4.408	0.04709947
93	22.548	22.570		22.559	-3.103	0.11636854	93	24.189	23.812		24.001	-5.377	0.02406809
94	26.388	25.893		26.140	-6.684	0.00972249	94	26.870	27.098		26.984	-8.360	0.00304394
95	23.661	23.818		23.740	-4.284	0.05134444	95	24.273	24.362		24.317	-5.693	0.01932407
96	25.606	25.474		25.540	-6.084	0.01474119	96	26.514	26.363		26.438	-7.814	0.00444268
97	25.805	25.880		25.843	-6.387	0.01195099	97	26.971	26.772		26.872	-8.248	0.00328997
60	19.552	19.360		19.456	0.000	0.99988529	60	18.590	18.657		18.624	0.000	1.00009462

(Rep 3) MCM Day 0							(Rep 3) MCM						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	23.104	22.617		22.860	-4.441	0.04603471	1	24.355	24.257		24.306	-6.061	0.01497658
3	24.891	24.720		24.805	-6.386	0.01195365	3	26.575	26.521		26.548	-8.303	0.00316645
4	23.517	23.587		23.552	-5.133	0.02849905	4	25.931	25.750		25.841	-7.596	0.00516932
5	24.422	24.389		24.406	-5.987	0.01576896	5	26.551	26.616		26.584	-8.339	0.00308846
6	22.536	22.494		22.515	-4.096	0.05847203	6	24.324	24.326		24.325	-6.080	0.01478453
7	22.896	22.779		22.837	-4.418	0.04676712	7	24.870	24.611		24.740	-6.495	0.01108445
8	23.922	23.403		23.662	-5.243	0.02640019	8	25.378	25.190		25.284	-7.039	0.00760517
9	23.426	23.451		23.439	-5.020	0.03082689	9	24.685	24.503		24.594	-6.349	0.01226514
11	24.232	23.964		24.098	-5.679	0.01951857	11	24.356	24.492		24.424	-6.179	0.01380339
12	23.299	23.208		23.253	-4.834	0.03504956	12	24.111	23.755		23.933	-5.688	0.01939918
13	21.088	20.978		21.033	-2.614	0.16336564	13	22.264	22.438		22.351	-4.106	0.05807258
15	22.562	21.981		22.272	-3.853	0.06922632	15	24.635	24.825		24.730	-6.485	0.01116407
17	22.588	22.425		22.506	-4.087	0.05883302	17	22.993	22.931		22.962	-4.717	0.03802893
18	21.596	21.582		21.589	-3.170	0.11109301	18	23.261	23.207		23.234	-4.989	0.03148799
19	22.501	22.480		22.491	-4.072	0.05947153	19	25.218	24.892		25.055	-6.810	0.00891223
20	24.285	24.303		24.294	-5.875	0.01703598	20	26.591	26.626		26.609	-8.364	0.00303622
21	22.737	22.738		22.738	-4.319	0.05011096	21	24.766	24.717		24.741	-6.496	0.011077
22	22.524	22.420		22.472	-4.053	0.0602481	22	23.712	23.501		23.606	-5.361	0.02432543
23	23.489	23.528		23.509	-5.090	0.02936751	23	25.514	25.610		25.562	-7.317	0.00627279
25	24.447	24.661		24.554	-6.135	0.01422633	25	26.551	26.553		26.552	-8.307	0.00315769
26	22.216	21.746		21.981	-3.562	0.0846758	26	24.514	24.234		24.374	-6.129	0.01428809
27	20.902	20.868		20.885	-2.466	0.1809828	27	22.194	21.987		22.090	-3.845	0.06958359
28	23.542	23.495		23.519	-5.100	0.0291637	28	25.150	24.727		24.938	-6.693	0.0096618
30	23.959	23.934		23.946	-5.527	0.02168037	30	25.765	25.771		25.768	-7.523	0.00543612
31	19.816	19.649		19.733	-1.314	0.40233119	31	20.599	20.483		20.541	-2.296	0.20361849
34	22.102	21.843		21.973	-3.554	0.08515585	34	23.100	22.586		22.843	-4.598	0.04128979
35	25.089	24.663		24.876	-6.457	0.01138264	35	26.314	25.552		25.933	-7.688	0.00484879
36	24.275	24.188		24.231	-5.812	0.01779536	36	25.809	25.305		25.557	-7.312	0.0062918
37	23.517	23.499		23.508	-5.089	0.02938497	37	22.609	22.596		22.602	-4.357	0.04879452
38	22.593	22.385		22.489	-4.070	0.05954301	38	25.265	25.103		25.184	-6.939	0.00814958
39	23.643	23.464		23.553	-5.134	0.02847087	39	24.992	24.779		24.885	-6.640	0.01002355
40	36.445	Undetermined		36.445	-18.026	3.7468E-06	40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	0
41	24.968	24.789		24.878	-6.459	0.01136311	41	26.499	25.841		26.170	-7.925	0.0041151
43	23.196	23.198		23.197	-4.778	0.03644679	43	24.775	24.835		24.805	-6.560	0.01060051
44	25.195	24.995		25.095	-6.676	0.00977888	44	25.630	25.423		25.526	-7.281	0.00642937
45	21.649	21.450		21.550	-3.131	0.11418274	45	23.976	23.721		23.849	-5.604	0.0205673
46	23.734	23.557		23.645	-5.226	0.02671244	46	25.740	25.839		25.790	-7.545	0.00535638
47	27.986	28.198		28.092	-9.673	0.00122518	47	30.223	30.266		30.245	-12.000	0.00024419
49	22.186	21.971		22.078	-3.659	0.07913861	49	23.791	23.738		23.764	-5.519	0.02180205
50	23.690	23.528		23.609	-5.190	0.02739532	50	25.606	25.418		25.512	-7.267	0.00649234
51	22.305	22.318		22.312	-3.893	0.06732922	51	24.809	24.728		24.768	-6.523	0.01087031
52	23.897	23.695		23.796	-5.377	0.0240599	52	25.557	25.467		25.512	-7.267	0.00649439
53	22.433	22.435		22.434	-4.015	0.06185909	53	24.008	23.894		23.951	-5.706	0.01915814
54	14.816	14.829		14.823	3.596	12.0956006	54	17.428	17.212		17.320	0.925	1.89858751
55	23.439	23.265		23.352	-4.933	0.03272979	55	24.871	24.866		24.869	-6.624	0.01014085
56	21.111	20.975		21.043	-2.624	0.16223944	56	24.461	24.663		24.562	-6.317	0.01254205
57	23.459	23.421		23.440	-5.021	0.03078921	57	24.846	24.251		24.549	-6.304	0.01265992
58	24.401	24.387		24.394	-5.975	0.01589492	58	24.879	24.988		24.933	-6.688	0.00969713
var2csa	15.799	15.427		15.613	2.806	6.99500942	var2csa	17.416	17.224		17.320	0.925	1.89897841
91	21.979	21.774		21.876	-3.457	0.09103954	91	23.546	23.357		23.451	-5.206	0.02708677
92	21.943	21.687		21.815	-3.396	0.09501203	92	22.190	21.938		22.064	-3.819	0.07085399
93	22.577	22.575		22.576	-4.157	0.0560628	93	24.273	24.005		24.139	-5.894	0.01681575
94	24.920	24.926		24.923	-6.504	0.01101551	94	26.873	26.673		26.773	-8.528	0.00270841
95	23.537	23.496		23.517	-5.098	0.02920246	95	25.444	25.486		25.465	-7.220	0.00670699
96	25.949	25.612		25.780	-7.361	0.00608225	96	26.769	26.646		26.708	-8.463	0.00283431
97	26.405	26.175		26.290	-7.871	0.0042717	97	26.899	26.638		26.769	-8.524	0.00271734
60	18.501	18.337		18.419	0.000	1.00026066	60	18.338	18.152		18.245	0.000	1.00012651

(Rep 3, 1) 1mM TI							(Rep 3, 2) 1mM TI						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	25.664	25.336		25.500	-5.016	0.03089737	1	26.230	25.648		25.939	-4.239	0.05295081
3	28.182	27.999		28.091	-7.607	0.00513055	3	28.751	28.153		28.452	-6.752	0.00927964
4	26.120	25.604		25.862	-5.378	0.02405248	4	26.226	26.436		26.331	-4.631	0.04035473
5	27.318	27.330		27.324	-6.840	0.00872803	5	27.455	27.824		27.639	-5.939	0.01629589
6	24.712	24.666		24.689	-4.205	0.05421634	6	25.716	25.209		25.463	-3.763	0.07368381
7	23.491	23.406		23.448	-2.964	0.12811435	7	24.393	24.169		24.281	-2.581	0.16710954
8	25.220	24.909		25.064	-4.580	0.04179779	8	25.533	25.286		25.410	-3.710	0.07643743
9	24.440	24.541		24.491	-4.007	0.06220746	9	25.505	25.316		25.411	-3.711	0.07638054
11	25.861	25.457		25.659	-5.175	0.02768522	11	26.244	26.166		26.205	-4.505	0.04403843
12	25.241	24.491		24.866	-4.382	0.04795957	12	26.149	25.943		26.046	-4.346	0.04916952
13	22.876	22.914		22.895	-2.411	0.18802143	13	23.788	23.672		23.730	-2.030	0.24482987
15	25.552	25.366		25.459	-4.975	0.03179875	15	26.342	25.778		26.060	-4.360	0.04870607
17	22.917	22.726		22.821	-2.337	0.19790535	17	24.251	23.948		24.099	-2.399	0.18955488
18	24.143	23.864		24.003	-3.519	0.08721956	18	24.574	24.664		24.619	-2.919	0.13223431
19	24.432	24.231		24.331	-3.847	0.06948132	19	25.266	24.969		25.117	-3.417	0.09359232
20	26.954	26.928		26.941	-6.457	0.01138278	20	27.809	27.703		27.756	-6.056	0.01502998
21	24.507	24.381		24.444	-3.960	0.06426404	21	24.921	24.928		24.924	-3.224	0.10699872
22	24.400	23.919		24.159	-3.675	0.07827365	22	25.122	24.722		24.922	-3.222	0.10718122
23	26.357	25.669		26.013	-5.529	0.02165461	23	26.292	26.385		26.339	-4.639	0.04013918
25	27.588	28.122		27.855	-7.371	0.00604237	25	28.251	29.238		28.744	-7.044	0.00757556
26	25.673	25.544		25.608	-5.124	0.02867128	26	26.346	26.161		26.253	-4.553	0.04258591
27	22.422	22.601		22.511	-2.027	0.24531018	27	23.269	23.121		23.195	-1.495	0.35483175
28	25.749	25.626		25.687	-5.203	0.02713934	28	26.986	26.941		26.964	-5.264	0.02603063
30	26.844	26.740		26.792	-6.308	0.01262039	30	27.885	27.143		27.514	-5.814	0.01777322
31	22.791	22.692		22.741	-2.257	0.20913965	31	23.387	23.313		23.350	-1.650	0.31867837
34	24.678	24.338		24.508	-4.024	0.06145555	34	25.517	25.734		25.626	-3.926	0.06579877
35	26.657	26.662		26.659	-6.175	0.01383625	35	26.837	27.022		26.930	-5.230	0.02665323
36	26.784	26.533		26.658	-6.174	0.01384767	36	27.137	26.920		27.029	-5.329	0.02488571
37	26.667	27.245		26.956	-6.472	0.01126623	37	27.767	27.875		27.821	-6.121	0.01436461
38	26.225	25.777		26.001	-5.517	0.02183803	38	26.868	26.674		26.771	-5.071	0.02975163
39	25.968	25.654		25.811	-5.327	0.0249161	39	26.629	26.768		26.699	-4.999	0.03128177
40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!	40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
41	25.761	25.616		25.688	-5.204	0.02712351	41	26.355	26.529		26.442	-4.742	0.03736332
43	25.371	24.992		25.181	-4.697	0.03854003	43	25.734	25.441		25.588	-3.888	0.06756159
44	26.276	26.008		26.142	-5.658	0.01980473	44	26.771	26.594		26.683	-4.983	0.03162222
45	25.268	25.135		25.202	-4.718	0.03800337	45	25.754	25.793		25.774	-4.074	0.05938531
46	26.147	25.456		25.801	-5.317	0.02508036	46	26.682	26.506		26.594	-4.894	0.03363604
47	30.277	29.945		30.111	-9.627	0.00126469	47	33.098	32.313		32.705	-11.005	0.00048644
49	25.236	25.319		25.278	-4.794	0.03605837	49	26.456	26.384		26.420	-4.720	0.03794517
50	25.320	26.777		26.048	-5.564	0.02113192	50	28.434	26.233		27.334	-5.634	0.02014304
51	25.195	24.973		25.084	-4.600	0.04122873	51	26.683	26.262		26.472	-4.772	0.03659632
52	26.651	26.494		26.572	-6.088	0.01469581	52	27.283	27.121		27.202	-5.502	0.02206832
53	25.390	25.221		25.305	-4.821	0.03537105	53	26.740	26.416		26.578	-4.878	0.03400996
54	19.142	18.976		19.059	1.425	2.68551262	54	19.782	19.676		19.729	1.971	3.92032701
55	26.270	26.126		26.198	-5.714	0.01905487	55	27.367	27.167		27.267	-5.567	0.02109487
56	24.903	24.749		24.826	-4.342	0.04931265	56	18.956	24.588		21.772	-0.072	0.95135191
57	24.978	24.544		24.761	-4.277	0.0515655	57	25.764	25.604		25.684	-3.984	0.06318752
58	25.185	25.101		25.143	-4.659	0.0395835	58	24.792	24.605		24.698	-2.998	0.12514838
var2csa	18.943	18.743		18.843	1.641	3.11792019	var2csa	18.988	19.173		19.080	2.620	6.14624595
91	24.778	24.691		24.734	-4.250	0.05253785	91	25.761	25.866		25.814	-4.114	0.05775862
92	24.214	23.968		24.091	-3.607	0.08209601	92	24.483	24.352		24.417	-2.717	0.15204255
93	25.721	25.424		25.573	-5.089	0.02938574	93	26.127	26.724		26.425	-4.725	0.03780271
94	27.706	27.714		27.710	-7.226	0.00667979	94	29.117	28.303		28.710	-7.010	0.00775752
95	25.956	25.654		25.805	-5.321	0.02502096	95	25.931	25.854		25.892	-4.192	0.05469933
96	26.342	26.142		26.242	-5.758	0.01848125	96	26.764	26.822		26.793	-5.093	0.02929968
97	26.002	26.006		26.004	-5.520	0.02179415	97	27.353	27.572		27.462	-5.762	0.01842312
60	20.564	20.404		20.484	0.000	1.00012131	60	21.662	21.739		21.700	0.000	0.99965591

Chapter 4.2 *var* panels

(Rep 1) MCM Day 0							(Rep 1) MCM 2Wks						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	23.090	22.998		23.044	-5.299	0.02539745	1	24.963	28.705		26.834	-8.941	0.00203457
3	24.913	24.873		24.893	-7.148	0.00705117	3	26.698	26.527		26.613	-8.720	0.00237211
4	24.330	24.671		24.501	-6.756	0.00925496	4	25.587	25.341		25.464	-7.571	0.00525822
5	24.183	23.976		24.080	-6.335	0.01239137	5	26.515	26.243		26.379	-8.486	0.00278832
6	22.903	23.114		23.009	-5.264	0.02602907	6	25.270	25.117		25.194	-7.301	0.00634309
7	22.615	22.899		22.757	-5.012	0.03099515	7	25.139	25.188		25.164	-7.271	0.00647614
8	23.187	22.924		23.056	-5.311	0.02519651	8	25.890	25.755		25.823	-7.930	0.00410136
9	23.125	23.183		23.154	-5.409	0.02353597	9	25.451	25.458		25.454	-7.561	0.00529446
11	24.884	24.934		24.909	-7.164	0.00697138	11	26.491	26.008		26.249	-8.356	0.00305151
12	23.154	23.310		23.232	-5.487	0.02229902	12	25.383	24.961		25.172	-7.279	0.0064374
13	20.833	21.469		21.151	-3.406	0.09433785	13	23.452	22.866		23.159	-5.266	0.02599397
15	22.570	22.150		22.360	-4.615	0.04081668	15	23.983	24.155		24.069	-6.176	0.01383199
17	22.612	22.346		22.479	-4.734	0.03758831	17	24.357	24.210		24.284	-6.391	0.01191971
18	22.372	22.656		22.514	-4.769	0.03668307	18	24.497	24.344		24.421	-6.528	0.01083899
19	22.871	22.869		22.870	-5.125	0.02865103	19	24.799	24.701		24.750	-6.857	0.00862507
20	24.387	24.312		24.350	-6.605	0.01027546	20	26.929	26.465		26.697	-8.804	0.00223734
21	23.257	23.373		23.315	-5.570	0.02105652	21	24.936	24.742		24.839	-6.946	0.00811118
22	22.333	21.982		22.158	-4.413	0.04694933	22	23.807	23.554		23.681	-5.788	0.01810033
23	24.217	24.206		24.211	-6.466	0.01131024	23	26.345	26.179		26.262	-8.369	0.00302424
25	24.959	25.597		25.278	-7.533	0.00539876	25	27.330	26.926		27.128	-9.235	0.00165994
26	22.305	22.363		22.334	-4.589	0.0415379	26	24.779	24.353		24.566	-6.673	0.00980184
27	20.687	20.766		20.727	-2.982	0.12660392	27	23.140	22.943		23.041	-5.148	0.02819683
28	24.095	24.392		24.244	-6.499	0.01105978	28	26.391	26.228		26.309	-8.416	0.00292724
30	24.434	24.429		24.431	-6.686	0.00971058	30	27.164	26.685		26.925	-9.032	0.00191056
31	19.331	19.181		19.256	-1.511	0.35092789	31	20.837	20.807		20.822	-2.929	0.13127394
34	20.908	20.899		20.903	-3.158	0.112001	34	22.570	22.391		22.481	-4.588	0.04159288
35	23.633	23.338		23.485	-5.740	0.01870501	35	26.834	26.585		26.710	-8.817	0.00221783
36	24.281	24.141		24.211	-6.466	0.01131344	36	26.196	26.518		26.357	-8.464	0.00283225
37	23.776	21.824		22.800	-5.055	0.03007792	37	21.914	21.716		21.815	-3.922	0.06597789
38	23.150	22.776		22.963	-5.218	0.02686434	38	24.837	24.659		24.748	-6.855	0.00863622
39	22.943	22.848		22.896	-5.151	0.02815398	39	25.000	24.923		24.962	-7.069	0.00744961
40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!	40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
41	24.203	23.953		24.078	-6.333	0.01240604	41	27.774	27.265		27.519	-9.626	0.00126515
43	22.775	23.105		22.940	-5.195	0.02730123	43	25.839	25.253		25.546	-7.653	0.0049687
44	25.113	25.477		25.295	-7.550	0.00533668	44	26.897	26.774		26.836	-8.943	0.00203253
45	21.471	21.341		21.406	-3.661	0.07904593	45	23.232	23.112		23.172	-5.279	0.02575743
46	22.958	23.291		23.124	-5.379	0.02402838	46	24.828	24.816		24.822	-6.929	0.00820461
47	28.979	29.292		29.135	-11.390	0.00037253	47	30.733	30.776		30.754	-12.861	0.00013439
49	22.184	22.107		22.146	-4.401	0.04734197	49	24.116	23.986		24.051	-6.158	0.01400542
50	23.969	23.860		23.914	-6.169	0.01389412	50	25.222	25.199		25.210	-7.317	0.00626924
51	23.481	23.752		23.616	-5.871	0.0170845	51	25.015	24.880		24.947	-7.054	0.00752312
52	24.576	24.420		24.498	-6.753	0.00927104	52	25.782	25.921		25.851	-7.958	0.00402072
53	22.982	22.674		22.828	-5.083	0.02950312	53	23.992	24.253		24.123	-6.230	0.01332442
54	15.529	15.805		15.667	2.078	4.22135892	54	17.481	17.259		17.370	0.523	1.43699894
55	24.015	23.857		23.936	-6.191	0.01368682	55	24.907	24.933		24.920	-7.027	0.00766705
56	23.121	22.849		22.985	-5.240	0.026458	56	21.862	23.363		22.612	-4.719	0.03795887
57	22.555	22.775		22.665	-4.920	0.03303067	57	24.877	24.400		24.639	-6.746	0.00931942
58	24.374	24.546		24.460	-6.715	0.00951922	58	24.867	24.780		24.823	-6.930	0.00819995
var2csa	21.454	21.214		21.334	-3.589	0.08309155	var2csa	21.187	21.290		21.238	-3.345	0.09838338
91	22.683	22.004		22.344	-4.599	0.04127413	91	23.844	23.625		23.735	-5.842	0.01743766
92	21.745	21.493		21.619	-3.874	0.06819678	92	22.697	22.252		22.474	-4.581	0.04177447
93	22.774	22.706		22.740	-4.995	0.03135862	93	24.745	24.654		24.699	-6.806	0.00893526
94	24.846	24.816		24.831	-7.086	0.00736086	94	27.499	27.514		27.507	-9.614	0.00127644
95	23.303	23.436		23.369	-5.624	0.02027131	95	24.814	24.782		24.798	-6.905	0.0083442
96	24.628	24.742		24.685	-6.940	0.00814434	96	26.863	26.674		26.769	-8.876	0.00212884
97	24.577	24.624		24.600	-6.855	0.00863693	97	27.215	26.750		26.982	-9.089	0.00183568
60	17.644	17.847		17.745	0.000	0.99978064	60	18.077	17.710		17.893	0.000	0.99991163

(Rep 1) MCM 4Wks							(Rep 1) (-)TrpArg 2Wks						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	22.143	21.918		22.031	-4.783	0.03633035	1	23.821	23.662		23.741	-5.705	0.01916642
3	25.159	25.177		25.168	-7.920	0.00412897	3	25.918	25.995		25.956	-7.920	0.00412762
4	23.575	23.421		23.498	-6.250	0.01313954	4	24.710	24.437		24.573	-6.537	0.01076627
5	23.206	23.126		23.166	-5.918	0.01653466	5	24.725	24.610		24.668	-6.632	0.0100855
6	21.899	21.840		21.870	-4.622	0.0406158	6	23.792	23.620		23.706	-5.670	0.01964309
7	22.931	22.756		22.843	-5.595	0.02068771	7	23.463	23.342		23.402	-5.366	0.02424482
8	23.278	23.174		23.226	-5.978	0.01586688	8	23.608	23.554		23.581	-5.545	0.0214144
9	23.494	23.489		23.492	-6.244	0.01319675	9	24.169	24.301		24.235	-6.199	0.01361592
11	24.455	24.359		24.407	-7.159	0.00699654	11	24.594	24.617		24.605	-6.569	0.01052984
12	23.839	23.633		23.736	-6.488	0.01113963	12	23.528	23.570		23.549	-5.513	0.02190244
13	20.642	20.490		20.566	-3.318	0.1002518	13	21.904	21.735		21.820	-3.784	0.07261289
15	21.734	21.630		21.682	-4.434	0.04625823	15	23.703	23.419		23.561	-5.525	0.02171439
17	21.945	21.675		21.810	-4.562	0.04234097	17	22.647	22.708		22.677	-4.641	0.04007396
18	21.404	21.249		21.326	-4.078	0.05919028	18	22.922	22.744		22.833	-4.797	0.03598054
19	21.280	20.970		21.125	-3.877	0.06806304	19	23.800	23.895		23.848	-5.812	0.01780461
20	25.471	25.159		25.315	-8.067	0.00372942	20	25.767	25.719		25.743	-7.707	0.00478732
21	22.654	22.458		22.556	-5.308	0.02524239	21	23.940	23.843		23.892	-5.856	0.01726771
22	21.564	21.183		21.373	-4.125	0.05730145	22	22.951	23.149		23.050	-5.014	0.03094508
23	23.528	23.474		23.501	-6.253	0.01310992	23	24.696	25.462		25.079	-7.043	0.00758365
25	26.422	26.228		26.325	-9.077	0.00185167	25	26.256	25.700		25.978	-7.942	0.00406658
26	21.515	21.518		21.516	-4.268	0.05189529	26	22.610	22.369		22.490	-4.454	0.04563048
27	20.423	20.473		20.448	-3.200	0.10884983	27	20.963	20.933		20.948	-2.912	0.13284239
28	23.823	23.848		23.836	-6.588	0.01039752	28	24.482	24.162		24.322	-6.286	0.0128165
30	24.237	24.112		24.175	-6.927	0.00821997	30	25.243	25.207		25.225	-7.189	0.00685412
31	19.136	18.918		19.027	-1.779	0.29132512	31	19.395	19.418		19.407	-1.371	0.38674284
34	20.616	20.555		20.586	-3.338	0.09891887	34	21.367	21.234		21.300	-3.264	0.10407334
35	24.472	24.337		24.404	-7.156	0.00701004	35	24.249	24.615		24.432	-6.396	0.01187319
36	22.745	22.863		22.804	-5.556	0.02126098	36	24.288	24.570		24.429	-6.393	0.01190236
37	23.442	23.298		23.370	-6.122	0.01435694	37	22.970	22.970		22.970	-4.934	0.03271137
38	22.931	23.122		23.026	-5.778	0.01822232	38	23.208	23.149		23.178	-5.142	0.02831088
39	21.856	21.796		21.826	-4.578	0.04186761	39	24.151	23.965		24.058	-6.022	0.01538821
40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!	40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
41	25.931	26.011		25.971	-8.723	0.00236662	41	26.215	26.226		26.221	-8.185	0.00343723
43	22.576	22.422		22.499	-5.251	0.0262537	43	24.594	24.485		24.540	-6.504	0.01102125
44	24.909	24.722		24.816	-7.568	0.00527127	44	24.964	25.015		24.989	-6.953	0.00806975
45	21.301	21.534		21.418	-4.170	0.05556647	45	21.493	21.559		21.526	-3.490	0.08900107
46	22.162	22.289		22.226	-4.978	0.03173524	46	23.024	23.522		23.273	-5.237	0.02651456
47	27.407	27.240		27.323	-10.075	0.00092679	47	29.715	28.679		29.197	-11.161	0.0004368
49	21.705	21.508		21.606	-4.358	0.04874953	49	22.174	22.152		22.163	-4.127	0.0572281
50	23.841	23.628		23.734	-6.486	0.01115495	50	22.927	23.902		23.414	-5.378	0.0240413
51	22.761	22.550		22.655	-5.407	0.02356044	51	23.947	23.661		23.804	-5.768	0.01835059
52	23.484	23.546		23.515	-6.267	0.0129865	52	24.453	24.639		24.546	-6.510	0.01097114
53	21.977	21.886		21.932	-4.684	0.03890634	53	22.887	22.901		22.894	-4.858	0.03448099
54	15.299	15.300		15.300	1.948	3.85928477	54	16.226	16.191		16.208	1.828	3.54922245
55	23.980	23.995		23.988	-6.740	0.00935734	55	23.768	24.011		23.889	-5.853	0.01729522
56	22.617	22.332		22.474	-5.226	0.02671054	56	21.731	23.339		22.535	-4.499	0.04422597
57	21.940	21.751		21.845	-4.597	0.04130834	57	23.403	22.782		23.093	-5.057	0.03004795
58	22.672	22.523		22.597	-5.349	0.02453022	58	23.823	23.620		23.721	-5.685	0.01943217
var2csa	19.472	19.210		19.341	-2.093	0.23440431	var2csa	18.669	18.516		18.593	-0.557	0.67980506
91	21.222	20.886		21.054	-3.806	0.07147302	91	22.220	22.298		22.259	-4.223	0.05354598
92	20.952	21.012		20.982	-3.734	0.07514467	92	21.867	21.985		21.926	-3.890	0.06746341
93	21.539	21.636		21.587	-4.339	0.04940649	93	22.697	22.691		22.694	-4.658	0.03960893
94	24.770	24.561		24.666	-7.418	0.00584935	94	25.690	25.572		25.631	-7.595	0.00517201
95	23.652	23.567		23.610	-6.362	0.01216154	95	23.343	23.212		23.278	-5.242	0.02642692
96	24.517	24.269		24.393	-7.145	0.00706697	96	25.576	25.448		25.512	-7.476	0.00561639
97	23.932	23.631		23.781	-6.533	0.01079603	97	25.702	24.832		25.267	-7.231	0.0066576
60	17.527	16.970		17.248	0.000	0.99983401	60	18.164	17.909		18.036	0.000	0.99976713

(Rep 1) (-)TrpArg 4Wks							(Rep 1) (-)TrpArg +1xKyn 2Wks						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	21.533	21.100		21.317	-3.846	0.06955934	1	23.967	23.731		23.849	-4.324	0.04994316
3	24.654	24.557		24.605	-7.134	0.00711813	3	25.215	24.959		25.087	-5.562	0.02117111
4	23.134	22.925		23.029	-5.558	0.02122354	4	25.014	24.585		24.799	-5.274	0.02583614
5	23.919	23.769		23.844	-6.373	0.01206741	5	25.860	25.547		25.704	-6.179	0.01380521
6	22.371	22.296		22.333	-4.862	0.03437876	6	23.892	24.130		24.011	-4.486	0.04462853
7	24.255	23.879		24.067	-6.596	0.01033657	7	22.691	22.451		22.571	-3.046	0.12105638
8	24.096	23.745		23.920	-6.449	0.01144298	8	23.702	23.368		23.535	-4.010	0.06206867
9	22.380	22.206		22.293	-4.822	0.03534549	9	24.643	24.435		24.539	-5.014	0.03094618
11	24.779	24.503		24.641	-7.170	0.0069438	11	25.117	24.792		24.955	-5.430	0.02320242
12	22.993	22.683		22.838	-5.367	0.02423047	12	24.202	23.924		24.063	-4.538	0.04304718
13	21.523	21.262		21.392	-3.921	0.06601085	13	21.482	21.192		21.337	-1.812	0.28478153
15	23.304	22.891		23.098	-5.627	0.02024019	15	23.501	23.348		23.425	-3.900	0.06699371
17	21.268	21.113		21.190	-3.719	0.07591514	17	23.127	23.264		23.195	-3.670	0.07854417
18	21.768	21.449		21.609	-4.138	0.0568157	18	22.735	22.533		22.634	-3.109	0.11590237
19	21.927	21.825		21.876	-4.405	0.04720166	19	24.004	23.636		23.820	-4.295	0.05094246
20	26.611	26.534		26.572	-9.101	0.00182069	20	24.433	23.992		24.212	-4.687	0.0388181
21	22.482	22.388		22.435	-4.964	0.03203399	21	24.091	23.619		23.855	-4.330	0.04972685
22	22.383	22.168		22.276	-4.805	0.0357776	22	23.318	23.427		23.372	-3.847	0.0694728
23	23.694	23.599		23.647	-6.176	0.01383273	23	25.317	25.114		25.216	-5.691	0.01936302
25	26.667	26.488		26.578	-9.107	0.00181385	25	25.602	25.137		25.369	-5.844	0.01740595
26	22.683	22.311		22.497	-5.026	0.03069163	26	22.442	22.208		22.325	-2.800	0.14358501
27	20.526	20.522		20.524	-3.053	0.12047168	27	21.147	21.377		21.262	-1.737	0.2999865
28	23.762	23.519		23.640	-6.169	0.01389374	28	24.758	24.513		24.636	-5.111	0.02894154
30	24.986	24.802		24.894	-7.423	0.00582535	30	25.927	25.458		25.693	-6.168	0.01390906
31	19.559	19.407		19.483	-2.012	0.24793331	31	19.880	19.872		19.876	-0.351	0.78384266
34	21.505	21.236		21.370	-3.899	0.06701612	34	21.683	21.480		21.582	-2.057	0.24035755
35	25.330	24.429		24.880	-7.409	0.00588545	35	24.568	24.188		24.378	-4.853	0.03460427
36	24.337	23.253		23.795	-6.324	0.01248289	36	24.648	24.715		24.681	-5.156	0.02803952
37	24.438	23.912		24.175	-6.704	0.00959127	37	24.480	23.893		24.187	-4.662	0.03950646
38	22.736	22.451		22.593	-5.122	0.02870786	38	23.714	23.541		23.627	-4.102	0.0582232
39	22.993	22.740		22.866	-5.395	0.0237608	39	23.909	23.652		23.780	-4.255	0.05236345
40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!	40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
41	25.697	25.629		25.663	-8.192	0.0034191	41	26.400	26.177		26.288	-6.763	0.00920428
43	23.166	22.872		23.019	-5.548	0.02137044	43	24.542	24.708		24.625	-5.100	0.02915871
44	25.503	25.432		25.467	-7.996	0.00391599	44	25.811	25.307		25.559	-6.034	0.01526232
45	21.279	20.974		21.126	-3.655	0.07936964	45	21.978	21.773		21.876	-2.351	0.19602411
46	23.746	23.659		23.703	-6.232	0.01330757	46	24.171	24.008		24.089	-4.564	0.04226271
47	28.246	28.332		28.289	-10.818	0.00055388	47	29.472	29.634		29.553	-10.028	0.00095802
49	22.340	22.167		22.253	-4.782	0.03633644	49	23.173	22.852		23.012	-3.487	0.08916127
50	24.157	24.132		24.144	-6.673	0.00979747	50	23.629	23.658		23.644	-4.119	0.05756497
51	23.461	23.464		23.462	-5.991	0.01571858	51	23.448	22.947		23.198	-3.673	0.07841133
52	24.462	24.462		24.462	-6.991	0.00786029	52	25.460	25.280		25.370	-5.845	0.01739635
53	22.597	22.471		22.534	-5.063	0.02990783	53	22.576	22.583		22.580	-3.055	0.12035183
54	16.431	16.236		16.333	1.138	2.20045596	54	18.274	16.440		17.357	2.168	4.49375821
55	23.876	23.775		23.825	-6.354	0.01222209	55	24.940	24.974		24.957	-5.432	0.02316584
56	22.547	22.469		22.508	-5.037	0.03044966	56	22.896	22.875		22.885	-3.360	0.09737218
57	21.972	21.621		21.796	-4.325	0.04988602	57	23.156	22.848		23.002	-3.477	0.08980605
58	22.000	22.006		22.003	-4.532	0.04322624	58	25.112	24.605		24.858	-5.333	0.02480347
var2csa	17.228	16.704		16.966	0.505	1.41898762	var2csa	19.443	19.372		19.408	0.117	1.08472136
91	21.780	21.738		21.759	-4.288	0.05118121	91	22.682	22.296		22.489	-2.964	0.1281521
92	20.885	20.756		20.820	-3.349	0.09812819	92	22.644	22.671		22.657	-3.132	0.11404764
93	22.350	22.099		22.224	-4.753	0.03707589	93	22.871	22.793		22.832	-3.307	0.10101252
94	25.476	25.731		25.603	-8.132	0.00356348	94	25.836	25.557		25.696	-6.171	0.01387548
95	23.969	23.790		23.879	-6.408	0.01177392	95	23.313	23.230		23.271	-3.746	0.07451889
96	23.301	22.532		22.917	-5.446	0.02294764	96	25.779	25.609		25.694	-6.169	0.0138979
97	24.861	24.421		24.641	-7.170	0.00694518	97	25.460	25.411		25.436	-5.911	0.01662423
60	17.643	17.300		17.471	0.000	0.9998146	60	19.454	19.595		19.525	0.000	1.00034213

(Rep 1) (-)TrpArg +1xKyn 4Wks							(Rep 2) MCM Day 0						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	23.860	23.923		23.891	-5.632	0.02016034	1	24.956	24.900		24.928	-6.517	0.01091895
3	25.294	25.095		25.194	-6.935	0.00817066	3	25.206	24.785		24.995	-6.584	0.01042267
4	24.462	24.308		24.385	-6.126	0.01431773	4	24.619	24.643		24.631	-6.220	0.01341685
5	24.872	24.489		24.680	-6.421	0.01166711	5	24.437	24.426		24.431	-6.020	0.01540462
6	23.518	23.405		23.462	-5.203	0.02715701	6	23.769	23.582		23.675	-5.264	0.02601791
7	24.863	24.704		24.783	-6.524	0.01086472	7	22.963	22.830		22.896	-4.485	0.04464432
8	24.985	25.189		25.087	-6.828	0.00880194	8	24.006	23.873		23.940	-5.529	0.02166401
9	24.469	24.481		24.475	-6.216	0.0134522	9	23.971	23.637		23.804	-5.393	0.02379745
11	25.235	25.163		25.199	-6.940	0.00814406	11	23.930	24.119		24.025	-5.614	0.02042497
12	23.852	23.469		23.660	-5.401	0.02366007	12	23.832	23.561		23.697	-5.286	0.02563604
13	21.217	21.203		21.210	-2.951	0.12934566	13	21.742	20.967		21.354	-2.943	0.1299986
15	23.602	23.221		23.411	-5.152	0.02811912	15	22.981	23.125		23.053	-4.642	0.0400563
17	22.420	22.299		22.359	-4.100	0.05829648	17	22.015	21.913		21.964	-3.553	0.08519647
18	22.187	21.962		22.074	-3.815	0.07103447	18	22.257	22.235		22.246	-3.835	0.07005933
19	22.455	22.745		22.600	-4.341	0.04935315	19	22.765	22.495		22.630	-4.219	0.0536941
20	27.312	26.964		27.138	-8.879	0.00212424	20	24.430	24.576		24.503	-6.092	0.0146568
21	22.947	22.587		22.767	-4.508	0.04394403	21	23.524	23.549		23.537	-5.126	0.02863922
22	21.940	21.974		21.957	-3.698	0.07704774	22	25.081	25.128		25.105	-6.694	0.00966096
23	25.010	25.140		25.075	-6.816	0.00887567	23	24.255	24.162		24.209	-5.798	0.01797702
25	26.770	27.206		26.988	-8.729	0.00235682	25	25.336	24.976		25.156	-6.745	0.00932159
26	22.786	22.447		22.617	-4.358	0.04877408	26	23.175	23.266		23.220	-4.809	0.03566829
27	21.649	21.254		21.451	-3.192	0.10939442	27	21.431	20.989		21.210	-2.799	0.14365847
28	25.450	25.346		25.398	-7.139	0.00709427	28	22.121	22.502		22.312	-3.901	0.06694822
30	25.445	25.228		25.336	-7.077	0.0074041	30	24.439	25.174		24.806	-6.395	0.01187906
31	20.810	20.514		20.662	-2.403	0.18902684	31	19.567	19.490		19.528	-1.117	0.4609517
34	23.217	22.432		22.825	-4.566	0.04223104	34	20.852	20.779		20.815	-2.404	0.18888087
35	25.670	25.590		25.630	-7.371	0.006041	35	24.324	24.498		24.411	-6.000	0.01562602
36	24.971	24.471		24.721	-6.462	0.01134431	36	24.657	24.910		24.784	-6.373	0.01206834
37	23.535	23.513		23.524	-5.265	0.02600871	37	24.299	24.144		24.222	-5.811	0.01781595
38	23.223	23.280		23.251	-4.992	0.03142118	38	23.228	23.134		23.181	-4.770	0.03665526
39	22.698	22.719		22.709	-4.450	0.04576379	39	23.588	23.642		23.615	-5.204	0.02712731
40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!	40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
41	24.659	24.580		24.620	-6.361	0.01216836	41	25.965	25.946		25.956	-7.545	0.0053553
43	23.796	23.760		23.778	-5.519	0.02180561	43	24.112	24.246		24.179	-5.768	0.01835166
44	25.626	25.420		25.523	-7.264	0.00650683	44	24.884	25.495		25.189	-6.778	0.0091115
45	22.187	22.161		22.174	-3.915	0.0662778	45	21.189	20.985		21.087	-2.676	0.15648242
46	24.120	23.558		23.839	-5.580	0.02089915	46	22.875	22.798		22.837	-4.426	0.04653345
47	29.357	29.588		29.473	-11.214	0.00042102	47	29.472	29.218		29.345	-10.934	0.00051099
49	23.572	23.514		23.543	-5.284	0.02566545	49	19.802	19.641		19.722	-1.311	0.40307313
50	24.752	24.520		24.636	-6.377	0.01202865	50	24.547	24.125		24.336	-5.925	0.01646092
51	23.715	23.459		23.587	-5.328	0.02490152	51	23.097	23.133		23.115	-4.704	0.03836358
52	26.246	25.814		26.030	-7.771	0.00457787	52	22.618	22.381		22.500	-4.089	0.05876901
53	23.268	22.859		23.064	-4.805	0.03577489	53	22.855	22.799		22.827	-4.416	0.04683525
54	17.076	16.708		16.892	1.367	2.57966191	54	16.552	16.431		16.491	1.920	3.78369815
55	25.187	25.344		25.266	-7.007	0.00777681	55	22.915	22.758		22.836	-4.425	0.04654431
56	22.695	22.421		22.558	-4.299	0.05079511	56	22.492	22.756		22.624	-4.213	0.05392094
57	23.170	23.287		23.228	-4.969	0.03192318	57	22.442	22.320		22.381	-3.970	0.06382166
58	23.006	22.672		22.839	-4.580	0.04180599	58	23.799	23.758		23.778	-5.367	0.02422549
var2csa	20.229	19.968		20.098	-1.839	0.27947342	var2csa	17.852	17.597		17.725	0.686	1.60935656
91	21.543	21.366		21.455	-3.196	0.10915268	91	21.943	21.943		21.943	-3.532	0.08645664
92	21.788	21.700		21.744	-3.485	0.08931111	92	21.241	20.896		21.069	-2.658	0.15846575
93	22.443	22.134		22.288	-4.029	0.06125207	93	22.620	22.545		22.583	-4.172	0.05549238
94	25.940	25.617		25.779	-7.520	0.00545003	94	25.307	25.374		25.341	-6.930	0.00820309
95	24.402	24.169		24.285	-6.026	0.01534102	95	24.478	24.161		24.319	-5.908	0.01665321
96	25.623	25.571		25.597	-7.338	0.00618179	96	24.324	24.531		24.427	-6.016	0.01545199
97	26.229	26.000		26.114	-7.855	0.00431798	97	24.198	24.168		24.183	-5.772	0.01829958
60	18.268	18.251		18.259	0.000	0.9996618	60	18.443	18.380		18.411	0.000	0.99986103

(Rep 2) MCM 3Wks							(Rep 2) (-)TrpArg 3Wks						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	19.924	19.815		19.870	-3.800	0.07181375	1	20.856	20.823		20.840	-4.603	0.04115648
3	21.509	21.936		21.723	-5.653	0.01987731	3	22.755	22.644		22.699	-6.462	0.01134199
4	21.921	22.378		22.150	-6.080	0.01478415	4	22.565	22.601		22.583	-6.346	0.01229092
5	20.519	20.673		20.596	-4.526	0.04339796	5	21.812	21.839		21.825	-5.588	0.02078877
6	21.360	21.969		21.664	-5.594	0.0207028	6	21.592	21.998		21.795	-5.558	0.02122851
7	20.704	20.630		20.667	-4.597	0.04131964	7	21.387	21.530		21.459	-5.222	0.02680184
8	21.893	22.392		22.143	-6.073	0.01485687	8	Undetermine	22.375		22.375	-6.138	0.01419994
9	21.668	21.824		21.746	-5.676	0.01956505	9	22.001	22.269		22.135	-5.898	0.01676968
11	21.277	21.390		21.333	-5.263	0.02603615	11	23.036	22.916		22.976	-6.739	0.00936096
12	21.741	21.893		21.817	-5.747	0.01862084	12	22.207	21.978		22.093	-5.856	0.01726851
13	19.454	19.742		19.598	-3.528	0.08668271	13	Undetermine	20.543		20.543	-4.306	0.0505676
15	25.626	24.871		25.249	-9.179	0.00172555	15	21.600	21.609		21.604	-5.367	0.02422511
17	21.753	20.980		21.367	-5.297	0.0254414	17	20.443	20.358		20.401	-4.164	0.05579948
18	20.843	21.179		21.011	-4.941	0.03255132	18	20.824	20.622		20.723	-4.486	0.0446319
19	19.627	19.482		19.554	-3.484	0.0893548	19	19.894	19.609		19.752	-3.515	0.08749452
20	24.584	25.046		24.815	-8.745	0.00233065	20	26.975	27.332		27.153	-10.916	0.00051742
21	21.523	21.704		21.614	-5.544	0.02143614	21	22.537	22.460		22.499	-6.262	0.01303407
22	25.166	24.869		25.018	-8.948	0.00202539	22	26.267	26.152		26.209	-9.972	0.00099538
23	21.509	22.215		21.862	-5.792	0.01804463	23	22.400	22.449		22.425	-6.188	0.01371872
25	26.190	26.126		26.158	-10.088	0.00091875	25	24.132	24.305		24.218	-7.981	0.00395691
26	20.911	20.620		20.765	-4.695	0.03859963	26	21.491	21.374		21.432	-5.195	0.02729156
27	19.003	19.173		19.088	-3.018	0.12344716	27	Undetermine	18.655		18.655	-2.418	0.18705391
28	20.516	20.387		20.451	-4.381	0.04799134	28	20.653	20.483		20.568	-4.331	0.04968697
30	21.929	21.482		21.706	-5.636	0.02011266	30	22.534	22.688		22.611	-6.374	0.01205688
31	16.328	16.594		16.461	-0.391	0.76252841	31	17.524	17.626		17.575	-1.338	0.39559401
34	18.421	18.624		18.522	-2.452	0.18270854	34	19.456	19.638		19.547	-3.310	0.10082009
35	21.797	21.696		21.746	-5.676	0.01955377	35	23.314	23.238		23.276	-7.039	0.00760334
36	22.417	22.532		22.474	-6.404	0.0118068	36	22.273	22.594		22.434	-6.197	0.01363445
37	24.275	23.853		24.064	-7.994	0.00392235	37	25.439	25.721		25.580	-9.343	0.00153984
38	21.530	21.323		21.427	-5.357	0.02440782	38	21.607	21.817		21.712	-5.475	0.0224828
39	21.941	21.775		21.858	-5.788	0.01809806	39	22.422	22.399		22.410	-6.173	0.01385558
40		Undetermined		#DIV/0!	#DIV/0!	#DIV/0!	40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
41	24.132	24.324		24.228	-8.158	0.00350122	41	25.516	25.801		25.659	-9.422	0.00145818
43	21.822	21.990		21.906	-5.836	0.01750614	43	22.123	22.338		22.231	-5.994	0.01569329
44	23.990	23.913		23.952	-7.882	0.00424047	44	24.479	24.526		24.502	-8.265	0.00324989
45	19.881	19.815		19.848	-3.778	0.07292089	45	18.955	18.917		18.936	-2.699	0.15398193
46	21.693	21.446		21.569	-5.499	0.02210737	46	21.201	21.016		21.109	-4.872	0.03415978
47	28.467	27.846		28.157	-12.087	0.00022989	47	28.422	28.344		28.383	-12.146	0.00022066
49	17.987	17.903		17.945	-1.875	0.2726738	49	18.675	18.731		18.703	-2.466	0.18100852
50	27.805	29.024		28.415	-12.345	0.00019228	50	28.411	28.796		28.604	-12.367	0.00018935
51	20.970	21.229		21.100	-5.030	0.03061528	51	22.176	22.204		22.190	-5.953	0.01614682
52	22.448	22.627		22.537	-6.467	0.01130148	52		22.216		22.216	-5.979	0.01585884
53	20.540	19.584		20.062	-3.992	0.0628471	53	20.834	20.955		20.895	-4.658	0.03961802
54	15.753	15.753		15.753	0.317	1.24580304	54		15.641		15.641	0.596	1.51193488
55	21.570	21.602		21.586	-5.516	0.02184957	55		22.374		22.374	-6.137	0.01420911
56	22.152	22.399		22.276	-6.206	0.0135475	56		21.012		21.012	-4.775	0.03652429
57	21.383	21.352		21.368	-5.298	0.02542481	57		21.988		21.988	-5.751	0.01857112
58	21.680	21.647		21.663	-5.593	0.02071054	58	21.930	21.924		21.927	-5.690	0.01936522
var2csa	15.519	15.459		15.489	0.581	1.49602937	var2csa	16.401	16.371		16.386	-0.149	0.90194665
91	19.908	19.920		19.914	-3.844	0.06964297	91	20.383	20.168		20.276	-4.039	0.06084451
92	19.245	19.370		19.307	-3.237	0.10603438	92	19.921	19.803		19.862	-3.625	0.0810279
93	20.806	20.731		20.769	-4.699	0.03850505	93	20.739	20.526		20.633	-4.396	0.04750407
94	23.896	23.927		23.911	-7.841	0.00436046	94	23.951	24.138		24.045	-7.808	0.00446384
95	22.739	22.604		22.672	-6.602	0.01029756	95	23.272	23.563		23.417	-7.180	0.00689429
96	24.215	24.272		24.244	-8.174	0.00346333	96	24.715	24.837		24.776	-8.539	0.00268779
97	23.639	24.179		23.909	-7.839	0.00436657	97	24.455	24.132		24.294	-8.057	0.00375621
60	16.031	16.110		16.070	0.000	0.99980871	60	16.193	16.281		16.237	0.000	1.00000381

(Rep 2) (-)Arg 3Wks							(Rep 2) (-)Trp 3Wks						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	24.275	25.149		24.712	-4.487	0.04458079	1	24.180	24.153		24.166	-3.063	0.11961838
3	25.691	25.611		25.651	-5.426	0.02326483	3	25.955	26.490		26.223	-5.120	0.02876086
4	23.937	24.896		24.417	-4.192	0.05472893	4	25.568	25.139		25.354	-4.251	0.05253268
5	25.206	25.271		25.238	-5.013	0.03095947	5	26.198	26.369		26.284	-5.181	0.0275689
6	23.827	24.313		24.070	-3.845	0.06958275	6	23.878	24.289		24.083	-2.980	0.12670629
7	22.620	22.671		22.645	-2.420	0.18680313	7	23.562	23.487		23.524	-2.421	0.18667705
8	23.790	23.412		23.601	-3.376	0.09630834	8	23.882	23.884		23.883	-2.780	0.14560002
9	23.583	24.306		23.944	-3.719	0.07591404	9	24.463	24.501		24.482	-3.379	0.09611477
11	25.165	25.238		25.201	-4.976	0.03176628	11	25.583	25.327		25.455	-4.352	0.04896
12	24.257	24.390		24.324	-4.099	0.05836693	12	25.273	24.794		25.033	-3.930	0.06558895
13	21.538	22.305		21.922	-1.697	0.30845779	13	21.927	22.767		22.347	-1.244	0.42216094
15	24.443	25.127		24.785	-4.560	0.04239629	15	23.611	24.906		24.258	-3.155	0.11222842
17	23.199	23.122		23.160	-2.935	0.13071952	17	23.938	23.445		23.691	-2.588	0.16626443
18	21.897	21.884		21.890	-1.665	0.31528731	18	22.684	22.559		22.621	-1.518	0.34904925
19	22.012	21.150		21.581	-1.356	0.39068046	19	23.235	21.935		22.585	-1.482	0.35790138
20	29.188	29.851		29.519	-9.294	0.00159249	20	27.475	26.731		27.103	-6.000	0.01562555
21	29.608	29.344		29.476	-9.251	0.00164122	21	24.137	25.289		24.713	-3.610	0.08189269
22	24.381	24.419		24.400	-4.175	0.05535425	22	27.022	26.519		26.770	-5.667	0.01967492
23	27.845	26.987		27.416	-7.191	0.00684289	23	28.183	Undetermined		28.183	-7.080	0.00739254
25	24.719	24.603		24.661	-4.436	0.04619402	25	30.020	33.899		31.960	-10.857	0.00053922
26	29.443	27.990		28.716	-8.491	0.00277873	26	24.280	24.583		24.432	-3.329	0.09953516
27	24.194	23.984		24.089	-3.864	0.06866447	27	26.019	Undetermined		26.019	-4.916	0.03313432
28	20.702	20.618		20.660	-0.435	0.73973093	28	24.266	24.723		24.495	-3.392	0.09528897
30	23.384	23.283		23.333	-3.108	0.11594424	30	Undetermine	Undetermined		23.333497	-2.230	0.2130853
31	24.628	22.961		23.794	-3.569	0.08424676	31	Undetermine	Undetermined		23.794235	-2.691	0.15483086
34	23.320	23.508		23.414	-3.189	0.10964896	34	Undetermine	Undetermined		23.414036	-2.311	0.20151568
35	26.394	25.769		26.081	-5.856	0.01726068	35	Undetermine	Undetermined		26.0813665	-4.978	0.03172213
36	25.281	25.526		25.403	-5.178	0.02761472	36	Undetermine	Undetermined		25.4034185	-4.300	0.05075105
37	26.940	26.905		26.923	-6.698	0.00963439	37	Undetermine	Undetermined		26.9225915	-5.820	0.01770632
38	23.869	24.414		24.141	-3.916	0.06623362	38	Undetermine	Undetermined		24.1412925	-3.038	0.12172585
39	25.793	25.883		25.838	-5.613	0.02043572	39	Undetermine	Undetermined		25.837763	-4.735	0.0375573
40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!	40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
41	25.215	24.583		24.899	-4.674	0.03917884	41	25.497	Undetermined		24.8987815	-3.796	0.07200388
43	24.719	25.173		24.946	-4.721	0.03791882	43	26.335	25.179		25.757	-4.654	0.03972932
44	26.124	25.680		25.902	-5.677	0.01954708	44	26.388	27.620		27.004	-5.901	0.01673969
45	21.603	21.699		21.651	-1.426	0.37207142	45	22.242	22.342		22.292	-1.189	0.43863611
46	23.275	23.713		23.494	-3.269	0.10375775	46	24.559	24.438		24.498	-3.395	0.09502932
47	29.647	31.332		30.490	-10.265	0.00081282	47	Undetermine	31.657		31.657	-10.554	0.00066494
49	20.901	20.889		20.895	-0.670	0.62845637	49	22.010	22.215		22.113	-1.010	0.49667757
50	31.717	11.387		21.552	-1.327	0.39862525	50	33.450	31.929		32.689	-11.586	0.00032526
51	24.990	25.537		25.263	-5.038	0.03043501	51	26.371	26.608		26.490	-5.387	0.02390389
52	24.902	24.960		24.931	-4.706	0.03830508	52	25.399	25.304		25.352	-4.249	0.05260685
53	25.460	25.632		25.546	-5.321	0.02501499	53	26.351	25.886		26.118	-5.015	0.03091736
54	18.275	18.230		18.253	1.972	3.92443374	54	18.993	18.775		18.884	2.219	4.65566096
55	26.682	26.165		26.424	-6.199	0.01361545	55	27.988	27.022		27.505	-6.402	0.01182543
56	24.927	25.249		25.088	-4.863	0.03436198	56	25.382	26.240		25.811	-4.708	0.03824773
57	24.496	24.558		24.527	-4.302	0.05069162	57	24.651	24.771		24.711	-3.608	0.08200186
58	22.924	22.668		22.796	-2.571	0.16825777	58	22.803	22.709		22.756	-1.653	0.31805992
var2csa	16.637	16.848		16.743	3.482	11.1767203	var2csa	17.825	17.833		17.829	3.274	9.67116373
91	25.006	25.219		25.112	-4.887	0.03379185	91	26.299	26.475		26.387	-5.284	0.02566336
92	22.977	23.604		23.290	-3.065	0.11946791	92	23.903	23.971		23.937	-2.834	0.14027137
93	24.543	24.290		24.416	-4.191	0.05473318	93	24.810	24.427		24.619	-3.516	0.08743608
94	27.594	28.323		27.958	-7.733	0.00469885	94	28.421	28.229		28.325	-7.222	0.00669776
95	24.275	24.490		24.382	-4.157	0.05604442	95	25.776	24.838		25.307	-4.204	0.05426521
96	26.040	25.908		25.974	-5.749	0.01859546	96	26.933	26.920		26.926	-5.823	0.01766078
97	25.589	25.855		25.722	-5.497	0.02214878	97	26.983	27.155		27.069	-5.966	0.01600031
60	20.151	20.299		20.225	0.000	1.00025095	60	21.020	21.186		21.103	0.000	1.00008145

(Rep 2) MCM 6Wks							(Rep 2) (-)TrpArg 6Wks						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	22.576	22.596		22.586	-1.947	0.25933925	1	21.854	22.202		22.028	-5.228	0.02668297
3	25.364	26.126		25.745	-5.106	0.02903281	3	24.350	23.753		24.051	-7.251	0.00656385
4	24.897	25.013		24.955	-4.316	0.05020944	4	23.606	23.741		23.674	-6.874	0.00852825
5	24.228	24.020		24.124	-3.485	0.08931523	5	22.956	22.835		22.896	-6.096	0.01462314
6	22.030	22.580		22.305	-1.666	0.31506469	6	22.577	22.562		22.570	-5.770	0.01832832
7	22.651	22.521		22.586	-1.947	0.25933997	7	23.360	23.444		23.402	-6.602	0.01029589
8	23.684	23.506		23.595	-2.956	0.12887828	8	24.203	24.453		24.328	-7.528	0.00541925
9	21.744	21.605		21.675	-1.036	0.48771508	9	22.794	22.919		22.856	-6.056	0.01502943
11	24.660	24.542		24.601	-3.962	0.06416534	11	24.803	24.903		24.853	-8.053	0.00376567
12	24.268	24.526		24.397	-3.758	0.073917	12	22.350	22.235		22.293	-5.493	0.02220911
13	21.783	21.719		21.751	-1.112	0.46264019	13	21.718	21.631		21.674	-4.874	0.03409329
15	24.266	24.572		24.419	-3.780	0.07278939	15	22.958	22.946		22.952	-6.152	0.01406087
17	21.994	21.932		21.963	-1.324	0.39932132	17	21.269	20.926		21.097	-4.297	0.05086531
18	21.866	21.930		21.898	-1.259	0.4177731	18	20.959	20.903		20.931	-4.131	0.0570743
19	23.292	23.414		23.353	-2.714	0.15244038	19	22.358	21.690		22.024	-5.224	0.02675288
20	29.392	29.587		29.489	-8.850	0.00216638	20	29.402	29.459		29.430	-12.630	0.00015771
21	25.267	25.202		25.235	-4.596	0.04136311	21	24.390	23.724		24.057	-7.257	0.00653856
22	23.949	23.975		23.962	-3.323	0.09993369	22	28.452	27.914		28.183	-11.383	0.00037442
23	23.829	24.228		24.029	-3.390	0.09539358	23	23.580	23.367		23.473	-6.673	0.00979729
25	26.628	26.744		26.686	-6.047	0.01512157	25	25.726	25.895		25.810	-9.010	0.001939
26	25.378	25.439		25.409	-4.770	0.03665663	26	21.849	21.714		21.782	-4.982	0.03165213
27	21.256	21.354		21.305	-0.666	0.63021325	27	19.775	19.637		19.706	-2.906	0.13345178
28	24.235	24.334		24.285	-3.646	0.07990555	28	21.942	21.694		21.818	-5.018	0.03086786
30	24.010	24.980		24.495	-3.856	0.0690493	30	23.569	23.771		23.670	-6.870	0.00855095
31	22.925	22.722		22.824	-2.185	0.21997981	31	18.757	18.662		18.709	-1.909	0.26619613
34	24.563	24.218		24.391	-3.752	0.07424355	34	20.639	20.036		20.337	-3.537	0.08612465
35	25.451	25.157		25.304	-4.665	0.03941123	35	25.397	25.388		25.392	-8.592	0.00259082
36	25.501	25.778		25.640	-5.001	0.03123867	36	24.258	24.225		24.242	-7.442	0.00575175
37	27.788	27.560		27.674	-7.035	0.00762589	37	26.525	26.251		26.388	-9.588	0.00129942
38	23.697	23.783		23.740	-3.101	0.11654702	38	23.291	22.716		23.003	-6.203	0.01357093
39	24.647	24.597		24.622	-3.983	0.06324348	39	24.008	24.192		24.100	-7.300	0.00634614
40	Undetermine	32.155		32.155	-11.516	0.00034143	40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
41	23.326	23.272		23.299	-2.660	0.1581852	41	25.267	24.870		25.069	-8.269	0.00324287
43	23.252	23.384		23.318	-2.679	0.15617365	43	23.253	23.117		23.185	-6.385	0.01196518
44	25.473	25.598		25.536	-4.897	0.0335738	44	26.368	26.703		26.535	-9.735	0.0011732
45	22.473	22.618		22.546	-1.907	0.26670772	45	20.683	20.336		20.510	-3.710	0.07642784
46	24.884	24.778		24.831	-4.192	0.05472521	46	22.588	22.631		22.610	-5.810	0.01782972
47	9.462	32.620		21.041	-0.402	0.75670663	47	30.807	32.022		31.414	-14.614	3.9872E-05
49	22.692	22.827		22.760	-2.121	0.22993779	49	19.917	19.385		19.651	-2.851	0.13857631
50	32.521	32.590		32.556	-11.917	0.00025867	50	30.447	30.832		30.639	-13.839	6.8236E-05
51	25.911	25.638		25.775	-5.136	0.0284466	51	22.948	23.523		23.235	-6.435	0.01155417
52	26.151	26.246		26.198	-5.559	0.02120649	52	25.088	25.079		25.084	-8.284	0.00320932
53	23.513	23.247		23.380	-2.741	0.14957443	53	21.840	21.744		21.792	-4.992	0.03142118
54	19.214	18.853		19.034	1.605	3.04264391	54	16.897	14.494		15.695	1.105	2.15029784
55	25.575	25.519		25.547	-4.908	0.03330306	55	23.659	23.769		23.714	-6.914	0.00829243
56	23.928	23.849		23.888	-3.249	0.10518286	56	21.651	21.850		21.751	-4.951	0.0323365
57	23.915	24.377		24.146	-3.507	0.08796159	57	22.800	22.810		22.805	-6.005	0.01556945
58	21.803	21.375		21.589	-0.950	0.51771607	58	22.350	22.226		22.288	-5.488	0.02228093
var2csa	18.191	17.783		17.987	2.652	6.28588117	var2csa	14.660	14.574		14.617	2.183	4.53979373
91	22.694	22.948		22.821	-2.182	0.22034659	91	21.213	21.171		21.192	-4.392	0.04761985
92	22.530	22.499		22.514	-1.875	0.27258083	92	20.535	20.004		20.270	-3.470	0.090265
93	24.331	Undetermined		24.331	-3.692	0.07736174	93	21.976	21.824		21.900	-5.100	0.02915868
94	27.283	27.791		27.537	-6.898	0.00838651	94	26.504	26.577		26.540	-9.740	0.00116912
95	23.637	23.657		23.647	-3.008	0.12430649	95	23.684	23.402		23.543	-6.743	0.00933368
96	26.066	26.741		26.404	-5.765	0.01839369	96	26.614	26.289		26.451	-9.651	0.00124344
97	25.759	26.268		26.014	-5.375	0.02410493	97	25.734	25.783		25.759	-8.959	0.00201008
60	20.637	20.641		20.639	0.000	1.00012165	60	16.974	16.629		16.801	-0.001	0.9991516

(Rep 2) (-)Arg 6Wks							(Rep 2) (-)Trp 6Wks						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	23.379	23.383		23.381	-2.891	0.13481571	1	20.868	20.733		20.800	-4.909	0.03327912
3	26.783	27.259		27.021	-6.531	0.0108122	3	22.746	22.553		22.649	-6.758	0.00923887
4	25.983	25.767		25.875	-5.385	0.02392949	4	22.738	22.754		22.746	-6.855	0.00863596
5	26.008	25.520		25.764	-5.274	0.02584679	5	21.377	21.438		21.407	-5.516	0.02185078
6	24.257	23.972		24.115	-3.625	0.08107023	6	22.382	22.203		22.293	-6.402	0.01182783
7	24.514	23.866		24.190	-3.700	0.0769585	7	20.827	20.783		20.805	-4.914	0.03316316
8	24.937	24.859		24.898	-4.408	0.04710082	8	21.928	21.915		21.922	-6.031	0.01529567
9	23.192	23.296		23.244	-2.754	0.14826002	9	20.751	20.789		20.770	-4.879	0.03397611
11	26.245	25.835		26.040	-5.550	0.02134777	11	22.168	22.270		22.219	-6.328	0.01244997
12	24.842	25.146		24.994	-4.504	0.04407287	12	20.854	21.008		20.931	-5.040	0.03039495
13	23.796	23.632		23.714	-3.224	0.10703529	13	19.720	19.955		19.837	-3.946	0.06486794
15	25.886	25.670		25.778	-5.288	0.02559603	15	20.663	20.753		20.708	-4.817	0.0354678
17	23.403	23.366		23.384	-2.894	0.13452036	17	19.530	19.235		19.383	-3.492	0.0888961
18	23.180	22.940		23.060	-2.570	0.168399	18	19.820	19.761		19.790	-3.899	0.06701038
19	23.808	23.822		23.815	-3.325	0.09978768	19	20.646	20.531		20.588	-4.697	0.03854238
20	31.733	31.131		31.432	-10.942	0.00050829	20	25.293	25.021		25.157	-9.266	0.00162403
21	25.736	25.644		25.690	-5.200	0.02719764	21	21.502	21.749		21.626	-5.735	0.01877968
22	26.805	26.812		26.808	-6.318	0.01253303	22	28.022	26.623		27.323	-11.432	0.00036199
23	25.499	25.792		25.645	-5.155	0.02806015	23	22.003	22.154		22.079	-6.188	0.01371775
25	28.277	29.351		28.814	-8.324	0.00312005	25	23.352	23.214		23.283	-7.392	0.00595461
26	26.820	26.432		26.626	-6.136	0.01421588	26	20.657	20.957		20.807	-4.916	0.03313287
27	22.695	22.477		22.586	-2.096	0.23389915	27	18.666	18.570		18.618	-2.727	0.15105214
28	24.768	24.709		24.739	-4.249	0.05260594	28	20.867	20.951		20.909	-5.018	0.03086589
30	26.352	27.002		26.677	-6.187	0.01372124	30	22.266	22.485		22.376	-6.485	0.01116728
31	23.206	23.228		23.217	-2.727	0.15107606	31	16.990	17.200		17.095	-1.204	0.43407553
34	25.611	25.356		25.483	-4.993	0.03140011	34	19.447	17.821		18.634	-2.743	0.14939725
35	26.501	26.680		26.590	-6.100	0.01457611	35	22.486	21.971		22.229	-6.338	0.01236396
36	26.354	26.730		26.542	-6.052	0.01507299	36	22.912	22.648		22.780	-6.889	0.00843606
37	28.395	28.593		28.494	-8.004	0.00389496	37	22.400	22.558		22.479	-6.588	0.01039522
38	25.768	25.829		25.799	-5.309	0.02522717	38	21.243	21.209		21.226	-5.335	0.02477608
39	26.425	25.998		26.211	-5.721	0.01895352	39	21.606	21.618		21.612	-5.721	0.01895525
40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!	40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
41	25.492	24.770		25.131	-4.641	0.04008306	41	24.439	24.644		24.541	-8.650	0.00248861
43	24.751	24.557		24.654	-4.164	0.05578444	43	21.913	22.286		22.100	-6.209	0.01352242
44	27.486	27.453		27.469	-6.979	0.00792443	44	22.894	22.883		22.889	-6.998	0.00782534
45	24.011	23.965		23.988	-3.498	0.08852057	45	18.889	18.802		18.846	-2.955	0.1289768
46	25.687	25.982		25.835	-5.345	0.02461105	46	20.826	20.856		20.841	-4.950	0.03235324
47	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!	47	28.052	27.897		27.974	-12.083	0.00023043
49	23.273	23.460		23.366	-2.876	0.13618021	49	18.264	18.181		18.222	-2.331	0.19869431
50	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!	50	28.852	28.741		28.797	-12.906	0.00013032
51	27.421	26.897		27.159	-6.669	0.00982832	51	21.272	21.000		21.136	-5.245	0.02637593
52	26.973	27.466		27.219	-6.729	0.00942545	52	23.637	23.663		23.650	-7.759	0.00461663
53	23.965	24.384		24.174	-3.684	0.07778887	53	20.509	20.493		20.501	-4.610	0.04096079
54	19.962	19.475		19.719	0.771	1.70693072	54	13.631	13.731		13.681	2.210	4.6274872
55	25.619	25.725		25.672	-5.182	0.02754372	55	21.769	22.335		22.052	-6.161	0.01397302
56	25.462	25.329		25.395	-4.905	0.03336925	56	21.726	21.831		21.779	-5.888	0.01689144
57	24.992	25.327		25.160	-4.670	0.03929512	57	20.838	20.912		20.875	-4.984	0.03159086
58	22.293	21.939		22.116	-1.626	0.32398962	58	21.798	21.269		21.534	-5.643	0.02001794
var2csa	17.286	16.460		16.873	3.617	12.2712303	var2csa	12.232	12.231		12.231	3.660	12.6374414
91	24.282	24.131		24.207	-3.717	0.07606399	91	19.728	19.542		19.635	-3.744	0.07462834
92	23.333	23.262		23.298	-2.808	0.14280268	92	19.817	19.288		19.552	-3.661	0.07903725
93	25.991	25.392		25.692	-5.202	0.02717124	93	20.639	20.555		20.597	-4.706	0.03831798
94	28.633	29.063		28.848	-8.358	0.00304818	94	24.506	24.382		24.444	-8.553	0.00266262
95	25.033	24.936		24.984	-4.494	0.04437614	95	22.184	22.279		22.232	-6.341	0.01233972
96	27.389	26.924		27.156	-6.666	0.00984523	96	23.908	24.187		24.048	-8.157	0.00350413
97	26.257	26.493		26.375	-5.885	0.0169213	97	23.339	23.265		23.302	-7.411	0.00587501
60	20.481	20.500		20.490	0.000	0.99967635	60	15.865	15.917		15.891	0.000	0.99986381

(Rep 3) MCM Day 0							(Rep 3) MCM 3Wks						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	23.601	24.460		24.031	-7.110	0.00724034	1	23.497	23.593		23.545	-5.613	0.02043499
3	25.401	24.999		25.200	-8.279	0.00321992	3	26.313	25.953		26.133	-8.201	0.00339795
4	25.584	22.796		24.190	-7.269	0.00648407	4	25.636	26.994		26.315	-8.383	0.00299546
5	25.020	24.682		24.851	-7.930	0.00409947	5	24.793	25.200		24.997	-7.065	0.00746992
6	24.598	25.523		25.060	-8.139	0.00354683	6	24.782	24.754		24.768	-6.836	0.0087536
7	23.021	22.515		22.768	-5.847	0.01737424	7	23.874	24.242		24.058	-6.126	0.01431861
8	24.390	24.461		24.425	-7.504	0.0055082	8	24.945	24.871		24.908	-6.976	0.00794218
9	23.827	23.574		23.701	-6.780	0.00910094	9	23.753	24.374		24.064	-6.132	0.01426181
11	26.189	25.783		25.986	-9.065	0.00186681	11	26.754	26.523		26.638	-8.706	0.0023939
12	24.149	24.341		24.245	-7.324	0.00624066	12	24.203	24.019		24.111	-6.179	0.01380349
13	22.224	22.200		22.212	-5.291	0.02553798	13	23.125	22.935		23.030	-5.098	0.02919465
15	22.966	23.432		23.199	-6.278	0.01288757	15	23.571	23.602		23.586	-5.654	0.01985441
17	22.293	22.766		22.530	-5.609	0.02049441	17	22.240	22.450		22.345	-4.413	0.04694284
18	22.229	21.981		22.105	-5.184	0.02750187	18	22.212	22.206		22.209	-4.277	0.05158759
19	23.568	23.174		23.371	-6.450	0.0114402	19	24.380	23.960		24.170	-6.238	0.01324504
20	25.539	25.600		25.570	-8.649	0.00249191	20	31.190	31.814		31.502	-13.570	8.2239E-05
21	23.902	23.466		23.684	-6.763	0.00920871	21	25.495	25.019		25.257	-7.325	0.00623764
22	22.805	22.857		22.831	-5.910	0.01663223	22	23.645	23.598		23.621	-5.689	0.01937792
23	25.117	24.788		24.953	-8.032	0.00382178	23	25.981	26.140		26.061	-8.129	0.00357291
25	26.847	26.391		26.619	-9.698	0.00120403	25	29.742	27.721		28.731	-10.799	0.0005611
26	23.983	23.781		23.882	-6.961	0.00802812	26	24.661	24.758		24.709	-6.777	0.00911659
27	21.447	21.190		21.319	-4.398	0.04744375	27	21.587	21.730		21.658	-3.726	0.07554683
28	24.504	23.996		24.250	-7.329	0.00621987	28	23.335	22.992		23.163	-5.231	0.0266191
30	25.947	24.521		25.234	-8.313	0.00314473	30	25.800	26.400		26.100	-8.168	0.00347721
31	20.916	21.179		21.047	-4.126	0.05725635	31	22.183	22.250		22.216	-4.284	0.05132233
34	23.646	21.819		22.732	-5.811	0.01780755	34	23.675	23.585		23.630	-5.698	0.01926234
35	25.479	25.709		25.594	-8.673	0.00245038	35	27.455	28.142		27.798	-9.866	0.00107135
36	25.937	25.420		25.679	-8.758	0.0023105	36	25.572	26.379		25.975	-8.043	0.00379065
37	26.249	24.947		25.598	-8.677	0.00244294	37	28.712	28.944		28.828	-10.896	0.00052476
38	23.703	23.783		23.743	-6.822	0.00883731	38	24.279	25.425		24.852	-6.920	0.008258
39	23.651	21.647		22.649	-5.728	0.01886761	39	26.614	26.203		26.408	-8.476	0.00280797
40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!	40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	0
41	24.991	25.708		25.349	-8.428	0.00290275	41	26.907	27.817		27.362	-9.430	0.00144984
43	24.423	24.546		24.484	-7.563	0.0052869	43	25.345	25.945		25.645	-7.713	0.00476491
44	25.708	25.679		25.694	-8.773	0.00228621	44	30.197	29.845		30.021	-12.089	0.00022952
45	24.218	23.944		24.081	-7.160	0.00699192	45	22.556	22.804		22.680	-4.748	0.03721484
46	23.951	23.587		23.769	-6.848	0.00868112	46	24.531	24.188		24.360	-6.428	0.01161715
47	28.377	28.184		28.281	-11.360	0.00038058	47	31.075	Undetermined		31.075	-13.143	0.00011056
49	23.796	23.538		23.667	-6.746	0.00931965	49	25.015	24.658		24.837	-6.905	0.00834618
50	23.604	23.856		23.730	-6.809	0.0089193	50	32.584	31.087		31.836	-13.904	6.5255E-05
51	24.349	24.138		24.244	-7.323	0.00624589	51	25.250	25.439		25.344	-7.412	0.00587023
52	25.342	24.710		25.026	-8.105	0.00363233	52	28.065	27.428		27.746	-9.814	0.00111063
53	22.889	23.513		23.201	-6.280	0.0128722	53	25.134	25.411		25.273	-7.341	0.00616992
54	16.624	16.540		16.582	0.339	1.26509524	54	17.292	18.588		17.940	-0.008	0.99426753
55	25.292	24.714		25.003	-8.082	0.0036907	55	26.675	31.210		28.943	-11.011	0.00048474
56	21.276	21.325		21.300	-4.379	0.04804759	56	24.498	24.614		24.556	-6.624	0.0101381
57	22.663	21.959		22.311	-5.390	0.02384274	57	25.247	25.304		25.276	-7.344	0.00615676
58	24.549	23.742		24.145	-7.224	0.00668741	58	24.184	24.487		24.335	-6.403	0.01181339
var2csa	20.342	19.355		19.848	-2.927	0.13147656	var2csa	20.018	20.331		20.174	-2.242	0.21135146
91	22.914	22.475		22.695	-5.774	0.01828088	91	23.359	22.924		23.142	-5.210	0.02702585
92	22.543	21.122		21.832	-4.911	0.03323349	92	23.147	23.296		23.221	-5.289	0.02557247
93	23.756	23.652		23.704	-6.783	0.00908313	93	23.809	23.797		23.803	-5.871	0.01708726
94	26.789	26.768		26.778	-9.857	0.00107805	94	28.491	29.293		28.892	-10.960	0.00050206
95	24.447	24.199		24.323	-7.402	0.00591164	95	25.756	25.999		25.877	-7.945	0.00405724
96	24.000	23.909		23.955	-7.034	0.00763313	96	27.907	27.980		27.943	-10.011	0.00096886
97	24.631	24.557		24.594	-7.673	0.00489838	97	26.965	27.199		27.082	-9.150	0.00175989
60	16.900	16.943		16.921	0.000	0.99966527	60	18.109	17.754		17.932	0.000	1.00024714

(Rep 3) (-)TrpArg 3Wks							(Rep 3) (-)Arg 3Wks						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	20.601	20.816		20.709	-4.532	0.0432362	1	21.649	21.544		21.597	-5.242	0.0264282
3	21.859	21.871		21.865	-5.688	0.0193935	3	23.009	22.150		22.580	-6.225	0.01337322
4	23.651	24.055		23.853	-7.676	0.00489093	4	23.999	23.875		23.937	-7.582	0.00521923
5	21.757	21.897		21.827	-5.650	0.01991816	5	22.927	23.267		23.097	-6.742	0.00934228
6	22.957	23.315		23.136	-6.959	0.00803813	6	22.545	21.961		22.253	-5.898	0.01676959
7	22.146	21.596		21.871	-5.694	0.01931687	7	22.432	22.380		22.406	-6.051	0.0150799
8	22.376	22.378		22.377	-6.200	0.01360123	8	22.903	22.985		22.944	-6.589	0.01038677
9	21.806	21.554		21.680	-5.503	0.02205313	9	21.971	21.905		21.938	-5.583	0.02086426
11	23.780	24.282		24.031	-7.854	0.0043221	11	25.828	25.900		25.864	-9.509	0.0013721
12	21.345	21.197		21.271	-5.094	0.02928159	12	22.330	22.327		22.329	-5.974	0.01591298
13	19.906	20.229		20.067	-3.890	0.067438	13	20.893	20.888		20.890	-4.535	0.04313208
15	21.714	21.975		21.845	-5.668	0.01967422	15	21.760	21.493		21.627	-5.272	0.02588523
17	19.934	19.921		19.927	-3.750	0.07429984	17	21.152	21.297		21.224	-4.869	0.03421472
18	18.949	18.675		18.812	-2.635	0.16101824	18	20.314	20.319		20.316	-3.961	0.06419766
19	19.966	20.859		20.412	-4.235	0.05308893	19	21.335	21.317		21.326	-4.971	0.0318838
20	26.977	27.159		27.068	-10.891	0.00052676	20	26.127	26.303		26.215	-9.860	0.00107617
21	23.437	24.461		23.949	-7.772	0.00457582	21	22.241	26.994		24.617	-8.262	0.00325664
22	21.571	21.696		21.633	-5.456	0.02277514	22	20.725	20.604		20.664	-4.309	0.05043982
23	23.799	24.356		24.078	-7.901	0.00418437	23	23.510	22.905		23.207	-6.852	0.00865538
25	24.513	24.566		24.540	-8.363	0.00303773	25	24.832	24.797		24.814	-8.459	0.00284099
26	22.473	22.629		22.551	-6.374	0.01205752	26	21.684	21.709		21.696	-5.341	0.02466454
27	17.903	18.339		18.121	-1.944	0.25991567	27	19.458	19.931		19.695	-3.340	0.09877981
28	21.956	22.246		22.101	-5.924	0.0164704	28	23.746	20.744		22.245	-5.890	0.0168589
30	22.572	22.438		22.505	-6.328	0.01245069	30	22.493	21.434		21.963	-5.608	0.02049785
31	17.659	18.194		17.927	-1.750	0.29740205	31	Undetermine	Undetermined		#DIV/0!	#DIV/0!	0
34	20.159	19.879		20.019	-3.842	0.06973376	34	20.567	20.745		20.656	-4.301	0.05074134
35	23.395	23.170		23.283	-7.106	0.00725996	35	24.416	24.279		24.348	-7.993	0.00392631
36	23.904	23.615		23.760	-7.583	0.00521683	36	23.569	23.166		23.368	-7.013	0.00774479
37	25.131	25.380		25.255	-9.078	0.00184999	37	24.704	24.879		24.791	-8.436	0.00288661
38	22.386	22.671		22.529	-6.352	0.01224416	38	21.436	21.852		21.644	-5.289	0.02557951
39	22.685	22.429		22.557	-6.380	0.01200359	39	22.542	22.609		22.576	-6.221	0.01340815
40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	0	40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	0
41	26.519	26.199		26.359	-10.182	0.00086109	41	26.438	26.609		26.524	-10.169	0.00086872
43	23.496	23.727		23.612	-7.435	0.00578035	43	22.773	22.706		22.739	-6.384	0.0119702
44	25.907	25.989		25.948	-9.771	0.00114447	44	24.926	25.231		25.079	-8.724	0.0023655
45	20.654	20.332		20.493	-4.316	0.05019093	45	21.131	21.165		21.148	-4.793	0.03607042
46	21.447	21.440		21.444	-5.267	0.02597615	46	21.706	21.317		21.512	-5.157	0.02803738
47	28.185	30.090		29.137	-12.960	0.00012549	47	26.454	26.292		26.373	-10.018	0.00096449
49	19.868	19.937		19.902	-3.725	0.07561682	49	21.403	21.215		21.309	-4.954	0.03225881
50	Undetermine	Undetermined		#DIV/0!	#DIV/0!	0	50	Undetermine	34.037		34.037	-17.682	4.7558E-06
51	22.459	22.821		22.640	-6.463	0.01133632	51	22.211	22.211		22.211	-5.856	0.01726445
52	25.418	25.543		25.481	-9.304	0.00158246	52	22.499	22.879		22.689	-6.334	0.01239395
53	21.908	21.044		21.476	-5.299	0.02539354	53	21.423	20.987		21.205	-4.850	0.03466453
54	15.928	14.091		15.010	1.167	2.24600128	54	14.817	14.740		14.779	1.576	2.9816419
55	24.860	24.596		24.728	-8.551	0.00266629	55	23.991	22.863		23.427	-7.072	0.00743092
56	21.788	21.749		21.768	-5.591	0.02073955	56	22.611	22.580		22.595	-6.240	0.01322681
57	20.639	20.829		20.734	-4.557	0.04248929	57	23.820	21.526		22.673	-6.318	0.01253433
58	22.496	22.831		22.663	-6.486	0.0111539	58	22.772	22.982		22.877	-6.522	0.01088307
var2csa	16.411	15.323		15.867	0.310	1.23985186	var2csa	16.914	16.860		16.887	-0.532	0.69161904
91	20.581	20.428		20.504	-4.327	0.04981781	91	20.581	20.480		20.531	-4.176	0.05532974
92	20.648	20.659		20.653	-4.476	0.0449251	92	20.250	20.284		20.267	-3.912	0.06644779
93	20.904	20.980		20.942	-4.765	0.03678435	93	20.924	20.879		20.901	-4.546	0.04279851
94	25.079	24.924		25.002	-8.825	0.00220558	94	24.560	24.677		24.619	-8.264	0.00325379
95	22.414	22.225		22.319	-6.142	0.01415682	95	23.511	23.501		23.506	-7.151	0.00703681
96	24.556	24.599		24.577	-8.400	0.00295981	96	25.543	25.373		25.458	-9.103	0.00181812
97	24.463	23.609		24.036	-7.859	0.00430741	97	24.546	24.460		24.503	-8.148	0.00352537
60	16.106	16.248		16.177	0.000	1.00009497	60	16.362	16.348		16.355	0.000	1.00003674

(Rep 3) (-)Trp 3Wks							(Rep 3) MCM 6Wks						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	21.293	21.190		21.242	-4.986	0.03156242	1	24.739	24.425		24.582	-7.096	0.00730889
3	22.748	22.846		22.797	-6.541	0.01073956	3	27.126	26.383		26.754	-9.268	0.00162165
4	23.015	23.332		23.174	-6.918	0.00826984	4	27.205	26.472		26.838	-9.352	0.00152983
5	22.679	22.638		22.659	-6.403	0.01181984	5	25.763	25.568		25.665	-8.179	0.00344943
6	22.999	22.711		22.855	-6.599	0.0103153	6	25.560	25.538		25.549	-8.063	0.0037386
7	22.708	22.467		22.587	-6.331	0.01241904	7	23.954	23.810		23.882	-6.396	0.01187167
8	26.222	26.107		26.165	-9.909	0.00104028	8	25.522	25.523		25.523	-8.037	0.00380811
9	25.685	25.317		25.501	-9.245	0.00164786	9	24.623	24.846		24.734	-7.248	0.00657687
11	24.761	24.507		24.634	-8.378	0.00300604	11	27.223	27.361		27.292	-9.806	0.00111734
12	21.703	21.717		21.710	-5.454	0.02281229	12	24.487	24.475		24.481	-6.995	0.00784033
13	19.993	19.938		19.965	-3.709	0.07646228	13	23.919	23.688		23.803	-6.317	0.01254066
15	26.408	25.801		26.104	-9.848	0.001085	15	25.465	25.327		25.396	-7.910	0.00415782
17	24.417	24.179		24.298	-8.042	0.00379417	17	23.883	23.390		23.636	-6.150	0.0140792
18	20.141	20.384		20.262	-4.006	0.06222411	18	23.797	23.731		23.764	-6.278	0.01288536
19	21.227	21.853		21.540	-5.284	0.02566337	19	24.177	23.864		24.021	-6.535	0.01078522
20	25.193	25.361		25.277	-9.021	0.00192484	20	30.257	30.903		30.580	-13.094	0.00011436
21	20.967	21.164		21.065	-4.809	0.03566219	21	24.910	24.946		24.928	-7.442	0.00575064
22	20.770	20.513		20.642	-4.386	0.04783708	22	23.584	23.867		23.725	-6.239	0.0132357
23	23.590	23.357		23.474	-7.218	0.00671711	23	26.736	25.958		26.347	-8.861	0.00215063
25	24.571	24.243		24.407	-8.151	0.00351852	25	28.843	29.251		29.047	-11.561	0.00033101
26	21.785	21.342		21.564	-5.308	0.02525106	26	26.126	25.855		25.991	-8.505	0.00275329
27	21.283	21.156		21.219	-4.963	0.03205142	27	22.904	22.908		22.906	-5.420	0.02335432
28	24.352	24.220		24.286	-8.030	0.00382588	28	24.931	25.221		25.076	-7.590	0.00519166
30	23.231	22.992		23.112	-6.856	0.00863531	30	27.223	27.323		27.273	-9.787	0.00113182
31	18.175	18.175		18.175	-1.919	0.26437488	31	23.137	22.987		23.062	-5.576	0.02096583
34	19.842	19.879		19.861	-3.605	0.08219868	34	23.946	24.245		24.095	-6.609	0.01024139
35	23.260	23.635		23.447	-7.191	0.00684144	35	28.133	26.666		27.400	-9.914	0.00103678
36	23.233	23.163		23.198	-6.942	0.00813238	36	26.139	25.841		25.990	-8.504	0.00275438
37	24.017	23.909		23.963	-7.707	0.00478668	37	29.673	29.238		29.455	-11.969	0.00024943
38	24.991	24.939		24.965	-8.709	0.00238922	38	25.608	25.342		25.475	-7.989	0.00393528
39	24.709	24.811		24.760	-8.504	0.00275477	39	25.844	25.217		25.531	-8.045	0.00378713
40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	0	40	Undetermine	32.576		32.576	-15.090	2.8664E-05
41	25.842	25.672		25.757	-9.501	0.00137995	41	25.913	25.745		25.829	-8.343	0.00307962
43	22.373	22.465		22.419	-6.163	0.01395421	43	25.233	25.190		25.212	-7.726	0.00472353
44	24.221	24.007		24.114	-7.858	0.00431006	44	27.518	28.253		27.886	-10.400	0.00074034
45	23.669	23.629		23.649	-7.393	0.00595042	45	24.967	24.664		24.815	-7.329	0.00621891
46	24.009	24.020		24.014	-7.758	0.00461862	46	24.816	25.179		24.997	-7.511	0.00548058
47	26.067	26.135		26.101	-9.845	0.00108734	47	32.779	32.184		32.481	-14.995	3.0617E-05
49	20.616	20.702		20.659	-4.403	0.04726636	49	25.656	25.736		25.696	-8.210	0.00337723
50	Undetermine	Undetermined		#DIV/0!	#DIV/0!	0	50	31.985	9.491		20.738	-3.252	0.10495621
51	22.170	22.218		22.194	-5.938	0.01630903	51	26.429	26.523		26.476	-8.990	0.00196684
52	23.379	22.982		23.180	-6.924	0.008233	52	27.610	27.897		27.753	-10.267	0.0008113
53	21.728	20.806		21.267	-5.011	0.03101348	53	24.816	24.595		24.706	-7.220	0.00670972
54	15.187	16.929		16.058	0.198	1.14699373	54	18.379	19.263		18.821	-1.335	0.3964249
55	25.640	25.029		25.335	-9.079	0.00184944	55	26.890	26.684		26.787	-9.301	0.00158514
56	22.553	22.674		22.613	-6.357	0.01219735	56	25.531	25.624		25.578	-8.092	0.00366586
57	20.793	20.755		20.774	-4.518	0.04364115	57	23.639	23.887		23.763	-6.277	0.01289211
58	23.204	23.393		23.299	-7.043	0.00758456	58	24.541	24.763		24.652	-7.166	0.00696245
var2csa	17.828	17.837		17.832	-1.576	0.335312	var2csa	19.576	19.486		19.531	-2.045	0.24235303
91	21.236	21.356		21.296	-5.040	0.030391	91	23.958	24.007		23.982	-6.496	0.01107884
92	21.174	21.161		21.168	-4.912	0.03322152	92	23.017	23.481		23.249	-5.763	0.01841461
93	21.764	21.785		21.775	-5.519	0.02180893	93	24.926	25.280		25.103	-7.617	0.00509327
94	26.335	24.563		25.449	-9.193	0.00170803	94	28.943	28.925		28.934	-11.448	0.00035799
95	22.644	22.561		22.603	-6.347	0.01228714	95	26.536	25.810		26.173	-8.687	0.0024263
96	24.878	24.776		24.827	-8.571	0.00262963	96	25.520	25.412		25.466	-7.980	0.00396159
97	24.155	23.998		24.076	-7.820	0.00442482	97	25.581	26.332		25.957	-8.471	0.00281882
60	16.244	16.268		16.256	0.000	1.00012859	60	17.456	17.516		17.486	0.000	1.00020381

(Rep 3) (-)TrpArg 6Wks							(Rep 3) (-)Arg 6Wks						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	26.784	26.272		26.528	-3.010	0.12412912	1	26.402	26.170		26.286	-4.799	0.03591869
3	27.995	28.460		28.227	-4.709	0.03822417	3	28.243	28.300		28.272	-6.785	0.00906952
4	28.773	29.250		29.011	-5.493	0.02220055	4	27.793	28.669		28.231	-6.744	0.00932821
5	29.611	28.291		28.951	-5.433	0.02314977	5	27.596	27.972		27.784	-6.297	0.01271993
6	27.322	28.219		27.770	-4.252	0.05247296	6	27.409	27.910		27.660	-6.173	0.01386149
7	26.649	26.985		26.817	-3.299	0.1015947	7	26.980	27.338		27.159	-5.672	0.01961652
8	26.921	27.391		27.156	-3.638	0.08034244	8	28.016	28.434		28.225	-6.738	0.0093665
9	26.956	26.697		26.826	-3.308	0.10094707	9	26.501	25.809		26.155	-4.668	0.03933766
11	27.890	27.586		27.738	-4.220	0.05366062	11	28.465	29.208		28.837	-7.350	0.00613136
12	26.162	26.152		26.157	-2.639	0.16058327	12	26.526	25.698		26.112	-4.625	0.04052239
13	25.371	25.905		25.638	-2.120	0.22998633	13	25.758	25.593		25.675	-4.188	0.05485021
15	28.258	27.906		28.082	-4.564	0.04227766	15	27.751	27.965		27.858	-6.371	0.01208176
17	25.988	26.246		26.117	-2.599	0.16508352	17	26.318	25.994		26.156	-4.669	0.03930865
18	25.768	25.722		25.745	-2.227	0.21361102	18	25.788	25.617		25.702	-4.215	0.05383954
19	25.617	25.426		25.522	-2.004	0.24936813	19	26.162	27.174		26.668	-5.181	0.02756404
20	Undetermine	32.494		32.494	-8.976	0.00198588	20	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
21	27.201	27.008		27.104	-3.586	0.08326774	21	29.124	28.316		28.720	-7.233	0.00664713
22	26.300	25.982		26.141	-2.623	0.16234097	22	25.754	25.910		25.832	-4.345	0.04921021
23	27.915	29.198		28.557	-5.039	0.03042635	23	28.493	28.313		28.403	-6.916	0.00827934
25	29.324	30.923		30.123	-6.605	0.0102706	25	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
26	28.623	28.733		28.678	-5.160	0.02796651	26	27.738	28.701		28.219	-6.732	0.00940541
27	25.197	24.931		25.064	-1.546	0.34252816	27	24.900	24.768		24.834	-3.347	0.0982851
28	27.985	28.493		28.239	-4.721	0.03792546	28	25.777	26.016		25.896	-4.409	0.0470629
30	30.115	29.427		29.771	-6.253	0.01311062	30	29.640	28.796		29.218	-7.731	0.00470762
31	26.381	26.273		26.327	-2.809	0.14270636	31	25.305	25.227		25.266	-3.779	0.07283446
34	28.854	28.523		28.688	-5.170	0.02777304	34	27.435	26.799		27.117	-5.630	0.02018773
35	29.705	29.183		29.444	-5.926	0.01644538	35	28.590	28.289		28.439	-6.952	0.0080749
36	28.771	29.454		29.112	-5.594	0.02069615	36	27.925	28.508		28.216	-6.729	0.0094244
37	31.144	30.925		31.035	-7.517	0.00546064	37	Undetermine	33.025		33.025	-11.538	0.0003363
38	27.237	27.255		27.246	-3.728	0.07548135	38	27.812	27.157		27.485	-5.998	0.01565082
39	27.719	27.769		27.744	-4.226	0.05342286	39	28.495	29.223		28.859	-7.372	0.00603618
40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!	40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
41	27.858	27.759		27.809	-4.291	0.05109948	41	27.837	27.305		27.571	-6.084	0.01474137
43	28.098	28.596		28.347	-4.829	0.03518777	43	27.468	28.418		27.943	-6.456	0.01139073
44	30.296	29.807		30.052	-6.534	0.0107947	44	29.755	31.738		30.746	-9.259	0.00163162
45	26.207	27.291		26.749	-3.231	0.10651876	45	26.524	27.336		26.930	-5.443	0.02298845
46	27.733	27.218		27.476	-3.958	0.06434842	46	27.839	28.869		28.354	-6.867	0.00856568
47	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!	47	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
49	28.656	28.716		28.686	-5.168	0.027817	49	28.393	28.609		28.501	-7.014	0.00773778
50	32.842	Undetermined		32.842	-9.324	0.00156057	50	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
51	29.386	28.764		29.075	-5.557	0.0212421	51	28.707	29.499		29.103	-7.616	0.00509776
52	30.233	30.106		30.169	-6.651	0.00994874	52	31.111	30.162		30.637	-9.150	0.00176058
53	27.913	27.893		27.903	-4.385	0.04786344	53	27.820	27.787		27.804	-6.317	0.01254676
54	22.913	22.948		22.930	0.588	1.50269269	54	22.152	21.534		21.843	-0.356	0.78133682
55	32.281	31.485		31.883	-8.365	0.00303396	55	30.186	30.911		30.548	-9.061	0.00187175
56	27.214	27.569		27.391	-3.873	0.06823279	56	28.803	27.532		28.168	-6.681	0.00974826
57	27.646	28.374		28.010	-4.492	0.04443364	57	28.158	27.646		27.902	-6.415	0.0117185
58	24.450	24.215		24.333	-0.815	0.56843567	58	26.779	26.225		26.502	-5.015	0.03092996
var2csa	21.616	21.893		21.755	1.763	3.3945645	var2csa	20.448	20.562		20.505	0.982	1.97540915
91	27.286	27.228		27.257	-3.739	0.07487654	91	25.868	26.498		26.183	-4.696	0.03857874
92	26.788	27.159		26.973	-3.455	0.09116517	92	25.586	25.552		25.569	-4.082	0.0590567
93	27.640	28.286		27.963	-4.445	0.04591424	93	27.475	27.471		27.473	-5.986	0.01577796
94	30.362	30.000		30.181	-6.663	0.00986684	94	31.928	32.202		32.065	-10.578	0.00065423
95	27.296	26.854		27.075	-3.557	0.08496085	95	27.632	26.845		27.238	-5.751	0.01856358
96	29.574	28.514		29.044	-5.526	0.02170305	96	29.151	30.624		29.888	-8.401	0.00295907
97	28.520	29.272		28.896	-5.378	0.024046	97	29.169	27.960		28.564	-7.077	0.00740569
60	23.426	23.611		23.518	0.000	0.99977822	60	21.518	21.456		21.487	0.000	1.00011923

(Rep 3) (-)Trp 6Wks						
Primer pair	CT			CT Average	relative to p60	relative copy number
1	28.253	26.950		27.602	-4.345	0.04921886
3	28.269	28.359		28.314	-5.057	0.03004706
4	28.016	29.683		28.850	-5.593	0.02072163
5	28.167	28.221		28.194	-4.937	0.03265005
6	27.492	28.668		28.080	-4.823	0.03533116
7	27.522	27.849		27.686	-4.429	0.04643141
8	28.202	28.824		28.513	-5.256	0.02617103
9	27.286	26.893		27.090	-3.833	0.07019188
11	29.227	27.940		28.583	-5.326	0.02492184
12	26.438	26.954		26.696	-3.439	0.09220242
13	25.590	25.761		25.676	-2.419	0.18702681
15	28.650	28.939		28.794	-5.537	0.02153181
17	26.654	28.317		27.486	-4.229	0.0533397
18	26.717	26.418		26.567	-3.310	0.10079647
19	27.231	27.247		27.239	-3.982	0.06327533
20	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
21	27.781	29.257		28.519	-5.262	0.02605394
22	26.377	26.135		26.256	-2.999	0.12509821
23	29.485	29.716		29.601	-6.344	0.01231408
25	31.642	Undetermined		31.642	-8.385	0.00299084
26	28.762	29.670		29.216	-5.959	0.01607347
27	25.581	25.792		25.686	-2.429	0.18565403
28	27.186	26.748		26.967	-3.710	0.07643525
30	31.895	29.874		30.884	-7.627	0.00505766
31	26.441	25.688		26.065	-2.808	0.14283446
34	29.362	28.351		28.856	-5.599	0.02062477
35	28.171	28.476		28.323	-5.066	0.02984432
36	28.918	28.668		28.793	-5.536	0.02155288
37	32.640	31.702		32.171	-8.914	0.00207323
38	27.910	27.941		27.925	-4.668	0.03932958
39	28.376	28.309		28.343	-5.086	0.02945064
40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
41	27.593	26.664		27.129	-3.872	0.06831741
43	27.746	28.528		28.137	-4.880	0.03395362
44	29.892	29.969		29.930	-6.673	0.00979739
45	27.775	28.207		27.991	-4.734	0.03757834
46	28.457	29.198		28.828	-5.571	0.02104039
47	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
49	29.827	28.642		29.234	-5.977	0.01587487
50	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
51	30.356	32.109		31.232	-7.975	0.00397413
52	31.663	30.822		31.242	-7.985	0.00394581
53	27.910	27.777		27.843	-4.586	0.0416253
54	23.250	23.291		23.270	-0.013	0.99085235
55	29.893	31.668		30.781	-7.524	0.00543393
56	28.266	28.463		28.364	-5.107	0.029013
57	28.417	28.253		28.335	-5.078	0.02960901
58	25.463	25.814		25.639	-2.382	0.19189513
var2csa	21.895	21.823		21.859	1.398	2.63494162
91	26.632	27.452		27.042	-3.785	0.07253854
92	25.994	26.577		26.286	-3.029	0.12253548
93	28.353	28.232		28.292	-5.035	0.03049478
94	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
95	28.576	26.993		27.785	-4.528	0.04335483
96	29.750	30.753		30.251	-6.994	0.00784416
97	28.781	30.630		29.705	-6.448	0.01145066
60	23.315	23.199		23.257	0.000	0.99990851

Chapter 4.4, 4.5, 4.7 *var* panels

(Rep 1) MCM Day 0							(Rep 1) MCM 3Wks						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	24.956	24.900		24.928	-6.517	0.01091895	1	19.924	19.815		19.870	-3.800	0.07181375
3	25.206	24.785		24.995	-6.584	0.01042267	3	21.509	21.936		21.723	-5.653	0.01987731
4	24.619	24.643		24.631	-6.220	0.01341685	4	21.921	22.378		22.150	-6.080	0.01478415
5	24.437	24.426		24.431	-6.020	0.01540462	5	20.519	20.673		20.596	-4.526	0.04339796
6	23.769	23.582		23.675	-5.264	0.02601791	6	21.360	21.969		21.664	-5.594	0.0207028
7	22.963	22.830		22.896	-4.485	0.04464432	7	20.704	20.630		20.667	-4.597	0.04131964
8	24.006	23.873		23.940	-5.529	0.02166401	8	21.893	22.392		22.143	-6.073	0.01485687
9	23.971	23.637		23.804	-5.393	0.02379745	9	21.668	21.824		21.746	-5.676	0.01956505
11	23.930	24.119		24.025	-5.614	0.02042497	11	21.277	21.390		21.333	-5.263	0.02603615
12	23.832	23.561		23.697	-5.286	0.02563604	12	21.741	21.893		21.817	-5.747	0.01862084
13	21.742	20.967		21.354	-2.943	0.1299986	13	19.454	19.742		19.598	-3.528	0.08668271
15	22.981	23.125		23.053	-4.642	0.0400563	15	25.626	24.871		25.249	-9.179	0.00172555
17	22.015	21.913		21.964	-3.553	0.08519647	17	21.753	20.980		21.367	-5.297	0.0254414
18	22.257	22.235		22.246	-3.835	0.07005933	18	20.843	21.179		21.011	-4.941	0.03255132
19	22.765	22.495		22.630	-4.219	0.0536941	19	19.627	19.482		19.554	-3.484	0.0893548
20	24.430	24.576		24.503	-6.092	0.0146568	20	24.584	25.046		24.815	-8.745	0.00233065
21	23.524	23.549		23.537	-5.126	0.02863922	21	21.523	21.704		21.614	-5.544	0.02143614
22	25.081	25.128		25.105	-6.694	0.00966096	22	25.166	24.869		25.018	-8.948	0.00202539
23	24.255	24.162		24.209	-5.798	0.01797702	23	21.509	22.215		21.862	-5.792	0.01804463
25	25.336	24.976		25.156	-6.745	0.00932159	25	26.190	26.126		26.158	-10.088	0.00091875
26	23.175	23.266		23.220	-4.809	0.03566829	26	20.911	20.620		20.765	-4.695	0.03859963
27	21.431	20.989		21.210	-2.799	0.14365847	27	19.003	19.173		19.088	-3.018	0.12344716
28	22.121	22.502		22.312	-3.901	0.06694822	28	20.516	20.387		20.451	-4.381	0.04799134
30	24.439	25.174		24.806	-6.395	0.01187906	30	21.929	21.482		21.706	-5.636	0.02011266
31	19.567	19.490		19.528	-1.117	0.4609517	31	16.328	16.594		16.461	-0.391	0.76252841
34	20.852	20.779		20.815	-2.404	0.18888087	34	18.421	18.624		18.522	-2.452	0.18270854
35	24.324	24.498		24.411	-6.000	0.01562602	35	21.797	21.696		21.746	-5.676	0.01955377
36	24.657	24.910		24.784	-6.373	0.01206834	36	22.417	22.532		22.474	-6.404	0.0118068
37	24.299	24.144		24.222	-5.811	0.01781595	37	24.275	23.853		24.064	-7.994	0.00392235
38	23.228	23.134		23.181	-4.770	0.03665526	38	21.530	21.323		21.427	-5.357	0.02440782
39	23.588	23.642		23.615	-5.204	0.02712731	39	21.941	21.775		21.858	-5.788	0.01809806
40	Undetermined	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!	40		Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
41	25.965	25.946		25.956	-7.545	0.0053553	41	24.132	24.324		24.228	-8.158	0.00350122
43	24.112	24.246		24.179	-5.768	0.01835166	43	21.822	21.990		21.906	-5.836	0.01750614
44	24.884	25.495		25.189	-6.778	0.0091115	44	23.990	23.913		23.952	-7.882	0.00424047
45	21.189	20.985		21.087	-2.676	0.15648242	45	19.881	19.815		19.848	-3.778	0.07292089
46	22.875	22.798		22.837	-4.426	0.04653345	46	21.693	21.446		21.569	-5.499	0.02210737
47	29.472	29.218		29.345	-10.934	0.00051099	47	28.467	27.846		28.157	-12.087	0.00022989
49	19.802	19.641		19.722	-1.311	0.40307313	49	17.987	17.903		17.945	-1.875	0.2726738
50	24.547	24.125		24.336	-5.925	0.01646092	50	27.805	29.024		28.415	-12.345	0.00019228
51	23.097	23.133		23.115	-4.704	0.03836358	51	20.970	21.229		21.100	-5.030	0.03061528
52	22.618	22.381		22.500	-4.089	0.05876901	52	22.448	22.627		22.537	-6.467	0.01130148
53	22.855	22.799		22.827	-4.416	0.04683525	53	20.540	19.584		20.062	-3.992	0.0628471
54	16.552	16.431		16.491	1.920	3.78369815	54	15.753	15.753		15.753	0.317	1.24580304
55	22.915	22.758		22.836	-4.425	0.04654431	55	21.570	21.602		21.586	-5.516	0.02184957
56	22.492	22.756		22.624	-4.213	0.05392094	56	22.152	22.399		22.276	-6.206	0.0135475
57	22.442	22.320		22.381	-3.970	0.06382166	57	21.383	21.352		21.368	-5.298	0.02542481
58	23.799	23.758		23.778	-5.367	0.02422549	58	21.680	21.647		21.663	-5.593	0.02071054
var2csa	17.852	17.597		17.725	0.686	1.60935656	var2csa	15.519	15.459		15.489	0.581	1.49602937
91	21.943	21.943		21.943	-3.532	0.08645664	91	19.908	19.920		19.914	-3.844	0.06964297
92	21.241	20.896		21.069	-2.658	0.15846575	92	19.245	19.370		19.307	-3.237	0.10603438
93	22.620	22.545		22.583	-4.172	0.05549238	93	20.806	20.731		20.769	-4.699	0.03850505
94	25.307	25.374		25.341	-6.930	0.00820309	94	23.896	23.927		23.911	-7.841	0.00436046
95	24.478	24.161		24.319	-5.908	0.01665321	95	22.739	22.604		22.672	-6.602	0.01029756
96	24.324	24.531		24.427	-6.016	0.01545199	96	24.215	24.272		24.244	-8.174	0.00346333
97	24.198	24.168		24.183	-5.772	0.01829958	97	23.639	24.179		23.909	-7.839	0.00436657
60	18.443	18.380		18.411	0.000	0.99986103	60	16.031	16.110		16.070	0.000	0.99980871

(Rep 1) MCM +10uM Spd 3Wks							(Rep 1) MCM +30nM Spd 3Wks						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	22.732	Undetermined		22.732	-5.532	0.02160823	1	21.572	21.661		21.617	-5.227	0.02670894
3	24.702	24.367		24.534	-7.334	0.00619608	3	23.169	23.312		23.240	-6.850	0.00866624
4	23.421	24.241		23.831	-6.631	0.0100922	4	23.270	23.279		23.275	-6.885	0.00846311
5	23.356	23.583		23.470	-6.270	0.01296249	5	22.254	22.865		22.560	-6.170	0.01389036
6	23.536	23.491		23.513	-6.313	0.01257565	6	22.326	22.163		22.244	-5.854	0.01728807
7	22.612	22.603		22.608	-5.408	0.0235596	7	22.147	21.778		21.963	-5.573	0.02101378
8	23.514	23.485		23.500	-6.300	0.01269442	8	22.346	22.535		22.440	-6.050	0.01508986
9	23.644	23.643		23.643	-6.443	0.01149047	9	22.150	22.465		22.307	-5.917	0.01654457
11	24.712	25.000		24.856	-7.656	0.00495838	11	23.357	23.441		23.399	-7.009	0.00776396
12	23.587	23.312		23.449	-6.249	0.0131472	12	22.386	22.498		22.442	-6.052	0.01506962
13	22.140	22.011		22.075	-4.875	0.03406833	13	20.875	20.782		20.828	-4.438	0.04611967
15	22.939	22.602		22.770	-5.570	0.0210483	15	21.426	21.733		21.580	-5.190	0.02739843
17	21.293	21.547		21.420	-4.220	0.05367105	17	20.347	20.491		20.419	-4.029	0.06126173
18	21.433	21.441		21.437	-4.237	0.05302145	18	20.608	20.803		20.705	-4.315	0.05022988
19	20.459	21.260		20.859	-3.659	0.07915254	19	19.855	20.239		20.047	-3.657	0.07929829
20	25.960	28.436		27.198	-9.998	0.00097782	20	26.876	26.543		26.710	-10.320	0.00078247
21	24.163	24.250		24.207	-7.007	0.00777713	21	22.802	22.917		22.859	-6.469	0.01128548
22	27.125	26.398		26.761	-9.561	0.00132376	22	25.608	25.773		25.691	-9.301	0.00158567
23	23.854	24.309		24.082	-6.882	0.00848097	23	23.178	22.337		22.757	-6.367	0.01211494
25	26.405	26.306		26.356	-9.156	0.00175318	25	24.974	24.599		24.787	-8.397	0.00296671
26	22.873	23.244		23.058	-5.858	0.01723733	26	21.876	21.662		21.769	-5.379	0.02403188
27	20.152	20.293		20.222	-3.022	0.12306624	27	18.987	18.986		18.986	-2.596	0.1653578
28	21.758	21.581		21.669	-4.469	0.04514996	28	20.682	20.828		20.755	-4.365	0.04853338
30	24.458	24.543		24.501	-7.301	0.006342	30	23.741	23.009		23.375	-6.985	0.00789261
31	19.350	19.398		19.374	-2.174	0.22160149	31	Undetermine	26.892		26.892	-10.502	0.00068934
34	20.460	20.750		20.605	-3.405	0.09438959	34	20.197	20.011		20.104	-3.714	0.07621155
35	24.569	24.998		24.784	-7.584	0.0052125	35	24.480	11.410		17.945	-1.555	0.34035554
36	24.843	24.816		24.830	-7.630	0.00504932	36	23.287	23.418		23.353	-6.963	0.00801732
37	26.619	26.403		26.511	-9.311	0.00157409	37	25.626	29.451		27.539	-11.149	0.00044044
38	23.734	23.348		23.541	-6.341	0.01233706	38	22.233	22.402		22.317	-5.927	0.01643036
39	24.254	24.209		24.232	-7.032	0.00764221	39	22.961	23.091		23.026	-6.636	0.01005432
40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!	40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
41	25.813	26.538		26.176	-8.976	0.00198614	41	26.801	Undetermined		26.801	-10.411	0.00073455
43	23.418	23.748		23.583	-6.383	0.01198174	43	22.791	22.264		22.528	-6.138	0.01420399
44	26.404	26.648		26.526	-9.326	0.00155797	44	24.642	Undetermined		24.642	-8.252	0.00327968
45	20.623	19.878		20.251	-3.051	0.12069941	45	18.976	19.014		18.995	-2.605	0.16440596
46	22.658	22.870		22.764	-5.564	0.02113724	46	Undetermine	26.136		26.136	-9.746	0.0011642
47	28.442	30.338		29.390	-12.190	0.000214	47	28.059	28.485		28.272	-11.882	0.00026493
49	19.901	20.622		20.262	-3.062	0.1197732	49	18.715	18.910		18.812	-2.422	0.18653419
50	30.592	29.287		29.939	-12.739	0.00014627	50	27.647	30.390		29.018	-12.628	0.00015793
51	23.732	23.817		23.775	-6.575	0.01048985	51	22.489	22.428		22.459	-6.069	0.01489828
52	23.584	24.278		23.931	-6.731	0.00941233	52	23.009	23.054		23.032	-6.642	0.01001595
53	22.889	23.265		23.077	-5.877	0.01701066	53	21.423	21.536		21.479	-5.089	0.02937535
54	17.486	16.917		17.201	-0.001	0.99911801	54	15.676	15.930		15.803	0.587	1.50216288
55	24.728	24.297		24.512	-7.312	0.00629111	55	22.884	22.984		22.934	-6.544	0.01071799
56	23.769	23.547		23.658	-6.458	0.0113723	56	22.749	22.723		22.736	-6.346	0.0122963
57	23.149	23.338		23.244	-6.044	0.01515904	57	22.475	22.506		22.490	-6.100	0.01457365
58	22.455	22.753		22.604	-5.404	0.02361589	58	21.965	21.758		21.861	-5.471	0.02253916
var2csa	17.974	17.955		17.965	-0.765	0.58860757	var2csa	16.888	17.139		17.013	-0.623	0.64913277
91	21.466	21.669		21.568	-4.368	0.04844302	91	20.838	20.816		20.827	-4.437	0.04616398
92	20.994	21.462		21.228	-4.028	0.06129648	92	19.976	19.946		19.961	-3.571	0.08413521
93	22.809	22.537		22.673	-5.473	0.02251581	93	21.646	21.337		21.492	-5.102	0.0291249
94	26.458	26.524		26.491	-9.291	0.00159669	94	24.491	24.630		24.560	-8.170	0.00347118
95	24.593	24.664		24.629	-7.429	0.00580433	95	23.650	23.426		23.538	-7.148	0.00705025
96	24.943	25.841		25.392	-8.192	0.00341952	96	25.327	25.562		25.445	-9.055	0.00188066
97	25.354	24.460		24.907	-7.707	0.0047847	97	24.193	24.442		24.318	-7.928	0.00410752
60	17.171	17.247		17.209	-0.009	0.99404047	60	16.412	16.368		16.390	0.000	0.9999799

(Rep 1) (-)TrpArg +10uM Spd 3Wks							(Rep 1) (-)TrpArg +30nM Spd 3Wks						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	23.692	23.601		23.646	-4.839	0.03492936	1	24.960			24.960	-4.298	0.0508263
3	24.427	24.390		24.409	-5.602	0.02059579	3	26.633			26.633	-5.971	0.01594646
4	24.645	24.337		24.491	-5.684	0.01945453	4	24.996			24.996	-4.334	0.04957117
5	24.609	24.286		24.448	-5.641	0.02004409	5	25.750			25.750	-5.088	0.02941054
6	24.200	23.283		23.741	-4.934	0.03270818	6	24.987			24.987	-4.325	0.04989918
7	22.010	22.004		22.007	-3.200	0.10883613	7	23.536			23.536	-2.874	0.13639144
8	23.497	23.381		23.439	-4.632	0.04033303	8	23.800			23.800	-3.138	0.1135949
9	22.983	22.980		22.981	-4.174	0.05538663	9	24.575			24.575	-3.913	0.06636535
11	24.531	24.602		24.567	-5.760	0.01845616	11	26.347			26.347	-5.685	0.01943766
12	24.655	23.497		24.076	-5.269	0.0259317	12	25.158			25.158	-4.496	0.04432634
13	21.738	24.531		23.134	-4.327	0.04980801	13	22.913			22.913	-2.251	0.21009402
15	24.406	24.200		24.303	-5.496	0.0221639	15	24.316			24.316	-3.654	0.07941817
17	21.989	22.983		22.486	-3.679	0.07808845	17	23.395			23.395	-2.733	0.15044186
18	22.248	24.645		23.446	-4.639	0.04012726	18	22.776			22.776	-2.114	0.23092912
19	21.660	22.010		21.835	-3.028	0.12258603	19	22.841			22.841	-2.179	0.22080978
20	28.511	28.527		28.519	-9.712	0.00119222	20	29.424			29.424	-8.762	0.00230344
21	23.661	23.636		23.648	-4.841	0.03488895	21	25.125			25.125	-4.463	0.04532812
22	27.012	26.807		26.909	-8.102	0.00363858	22	28.663			28.663	-8.001	0.0039026
23	23.661	23.865		23.763	-4.956	0.03222677	23	25.366			25.366	-4.704	0.03836693
25	27.299	27.846		27.573	-8.766	0.00229739	25	28.296			28.296	-7.634	0.005034
26	23.022	22.873		22.948	-4.141	0.05669746	26	24.941			24.941	-4.279	0.05150456
27	19.678	19.700		19.689	-0.882	0.54250863	27	20.902			20.902	-0.240	0.84696426
28	21.752	21.785		21.769	-2.962	0.12836284	28	23.901			23.901	-3.239	0.10588646
30	24.985	24.679		24.832	-6.025	0.01535788	30	27.021	26.897		26.959	-6.297	0.01271915
31	20.423	20.254		20.339	-1.532	0.34585147	31	23.024			23.024	-2.362	0.19449891
34	21.948	21.888		21.918	-3.111	0.11575541	34	24.525	24.319		24.422	-3.760	0.07380941
35	25.339	25.678		25.509	-6.702	0.00960698	35	26.737			26.737	-6.075	0.01482861
36	24.414	24.820		24.617	-5.810	0.01782512	36	25.635	25.659		25.647	-4.985	0.03158338
37	25.725	25.823		25.774	-6.967	0.00799274	37	26.995			26.995	-6.333	0.01240372
38	23.623	23.576		23.600	-4.793	0.03608152	38	25.573	25.183		25.378	-4.716	0.03805632
39	24.634	24.475		24.555	-5.748	0.01861144	39	26.458			26.458	-5.796	0.01800134
40	33.595	Undetermined		33.595	-14.788	3.535E-05	40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
41	25.640	25.772		25.706	-6.899	0.0083808	41	27.326			27.326	-6.664	0.00986445
43	24.816	25.217		25.017	-6.210	0.01351244	43	25.726	26.226		25.976	-5.314	0.02513803
44	25.632	25.594		25.613	-6.806	0.00893517	44	26.745			26.745	-6.083	0.01474857
45	21.166	21.013		21.089	-2.282	0.20556948	45	21.810	21.661		21.735	-1.073	0.47527536
46	22.632	22.673		22.653	-3.846	0.06956293	46	23.911			23.911	-3.249	0.10521483
47	30.300	29.767		30.033	-11.226	0.0004174	47	31.449	32.007		31.728	-11.066	0.00046657
49	20.641	20.438		20.540	-1.733	0.30089718	49	22.217			22.217	-1.555	0.34024456
50	29.924	30.474		30.199	-11.392	0.00037202	50	31.723	31.950		31.837	-11.175	0.00043265
51	23.850	24.121		23.986	-5.179	0.02761313	51	26.356	26.428		26.392	-5.730	0.01883887
52	24.201	23.781		23.991	-5.184	0.02750651	52	24.901	25.655		25.278	-4.616	0.04078764
53	23.502	23.532		23.517	-4.710	0.03820682	53	27.554	27.449		27.502	-6.840	0.00873089
54	17.354	17.262		17.308	1.499	2.8266828	54	18.647	18.707		18.677	1.985	3.9594046
55	24.409	24.698		24.553	-5.746	0.01862856	55	27.149	26.859		27.004	-6.342	0.01232598
56	23.772	23.464		23.618	-4.811	0.03562693	56	25.703			25.703	-5.041	0.03037371
57	22.766	22.746		22.756	-3.949	0.06473387	57	24.857			24.857	-4.195	0.05459308
58	22.963	23.240		23.102	-4.295	0.05095828	58	Undetermine	Undetermined		23.102	-2.440	0.18434248
var2csa	15.419	14.951		15.185	3.622	12.3125954	var2csa	17.217	17.497		17.357	3.305	9.88229073
91	22.999	23.114		23.057	-4.250	0.05256892	91	Undetermine	Undetermined		23.057	-2.395	0.190169
92	22.012	21.971		21.991	-3.184	0.11001282	92	23.599	23.701		23.650	-2.988	0.12601829
93	22.817	22.855		22.836	-4.029	0.06125396	93	Undetermine	Undetermined		22.836	-2.174	0.22158728
94	25.787	25.833		25.810	-7.003	0.0077954	94	27.562	27.534		27.548	-6.886	0.00845593
95	23.458	23.375		23.417	-4.610	0.04095557	95	Undetermine	Undetermined		23.417	-2.755	0.14815749
96	24.644	24.868		24.756	-5.949	0.01619133	96	26.111	29.169		27.640	-6.978	0.00793327
97	23.895	23.960		23.928	-5.121	0.02874297	97	Undetermine	Undetermined		23.928	-3.266	0.1039782
60	18.745	18.869		18.807	0.000	1.00007868	60	20.613	20.712		20.662	0.000	0.99972382

(Rep 1) MCM 6Wks							(Rep 1) MCM +10uM Spd 6Wks						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	22.576	22.596		22.586	-1.947	0.25933925	1	22.428	22.741		22.585	-0.974	0.50911818
3	25.364	26.126		25.745	-5.106	0.02903281	3	26.012	26.416		26.214	-4.603	0.04114865
4	24.897	25.013		24.955	-4.316	0.05020944	4	25.389	25.827		25.608	-3.997	0.06263529
5	24.228	24.020		24.124	-3.485	0.08931523	5	24.403	24.803		24.603	-2.992	0.12570248
6	22.030	22.580		22.305	-1.666	0.31506469	6	22.467	22.421		22.444	-0.833	0.56136557
7	22.651	22.521		22.586	-1.947	0.25933997	7	23.446	23.006		23.226	-1.615	0.32649663
8	23.684	23.506		23.595	-2.956	0.12887828	8	24.223	24.254		24.238	-2.627	0.16183948
9	21.744	21.605		21.675	-1.036	0.48771508	9	22.815	22.267		22.541	-0.930	0.52488053
11	24.660	24.542		24.601	-3.962	0.06416534	11	25.299	25.394		25.347	-3.736	0.07507043
12	24.268	24.526		24.397	-3.758	0.073917	12	24.790	24.502		24.646	-3.035	0.12201357
13	21.783	21.719		21.751	-1.112	0.46264019	13	22.775	22.680		22.727	-1.116	0.46124526
15	24.266	24.572		24.419	-3.780	0.07278939	15	25.310	25.378		25.344	-3.733	0.07519218
17	21.994	21.932		21.963	-1.324	0.39932132	17	23.277	22.923		23.100	-1.489	0.35630954
18	21.866	21.930		21.898	-1.259	0.4177731	18	21.926	21.705		21.816	-0.205	0.86780873
19	23.292	23.414		23.353	-2.714	0.15244038	19	23.899	23.712		23.805	-2.194	0.21847665
20	29.392	29.587		29.489	-8.850	0.00216638	20	30.271	30.094		30.183	-8.572	0.00262804
21	25.267	25.202		25.235	-4.596	0.04136311	21	25.253	25.930		25.591	-3.980	0.06335556
22	23.949	23.975		23.962	-3.323	0.09993369	22	24.647	24.762		24.705	-3.094	0.11715086
23	23.829	24.228		24.029	-3.390	0.09539358	23	24.605	24.436		24.520	-2.909	0.13310586
25	26.628	26.744		26.686	-6.047	0.01512157	25	26.468	26.714		26.591	-4.980	0.03168573
26	25.378	25.439		25.409	-4.770	0.03665663	26	25.998	25.706		25.852	-4.241	0.05288368
27	21.256	21.354		21.305	-0.666	0.63021325	27	21.941	21.915		21.928	-0.317	0.80256937
28	24.235	24.334		24.285	-3.646	0.07990555	28	24.608	24.370		24.489	-2.878	0.13600919
30	24.010	24.980		24.495	-3.856	0.0690493	30	25.334	25.492		25.413	-3.802	0.07169004
31	22.925	22.722		22.824	-2.185	0.21997981	31	23.878	23.391		23.634	-2.023	0.24599186
34	24.563	24.218		24.391	-3.752	0.07424355	34	25.458	25.705		25.582	-3.971	0.06378515
35	25.451	25.157		25.304	-4.665	0.03941123	35	25.362	25.137		25.249	-3.638	0.08029934
36	25.501	25.778		25.640	-5.001	0.03123867	36	25.522	25.670		25.596	-3.985	0.06316611
37	27.788	27.560		27.674	-7.035	0.00762589	37	28.502	28.367		28.435	-6.824	0.00882832
38	23.697	23.783		23.740	-3.101	0.11654702	38	24.469	24.804		24.637	-3.026	0.12280575
39	24.647	24.597		24.622	-3.983	0.06324348	39	24.860	25.535		25.197	-3.586	0.08324881
40	Undetermined	32.155		32.155	-11.516	0.00034143	40	30.937	Undetermined		30.937	-9.326	0.00155861
41	23.326	23.272		23.299	-2.660	0.1581852	41	23.597	23.506		23.551	-1.940	0.26056433
43	23.252	23.384		23.318	-2.679	0.15617365	43	23.862	23.593		23.727	-2.116	0.2306359
44	25.473	25.598		25.536	-4.897	0.0335738	44	25.331	25.736		25.533	-3.922	0.06595764
45	22.473	22.618		22.546	-1.907	0.26670772	45	23.622	23.342		23.482	-1.871	0.27340281
46	24.884	24.778		24.831	-4.192	0.05472521	46	25.163	25.326		25.244	-3.633	0.08059762
47	9.462	32.620		21.041	-0.402	0.75670663	47	32.731	11.548		22.140	-0.529	0.69310364
49	22.692	22.827		22.760	-2.121	0.22993779	49	23.631	23.533		23.582	-1.971	0.25508951
50	32.521	32.590		32.556	-11.917	0.00025867	50	34.226	33.561		33.893	-12.282	0.00020075
51	25.911	25.638		25.775	-5.136	0.0284466	51	26.668	27.295		26.982	-5.371	0.02416845
52	26.151	26.246		26.198	-5.559	0.02120649	52	26.552	26.709		26.631	-5.020	0.03082245
53	23.513	23.247		23.380	-2.741	0.14957443	53	23.615	23.574		23.595	-1.984	0.25287267
54	19.214	18.853		19.034	1.605	3.04264391	54	19.856	19.535		19.695	1.916	3.77286375
55	25.575	25.519		25.547	-4.908	0.03330306	55	25.816	25.896		25.856	-4.245	0.05273907
56	23.928	23.849		23.888	-3.249	0.10518286	56	24.526	24.544		24.535	-2.924	0.13179618
57	23.915	24.377		24.146	-3.507	0.08796159	57	24.476	24.712		24.594	-2.983	0.12646653
58	21.803	21.375		21.589	-0.950	0.51771607	58	22.238	21.816		22.027	-0.416	0.74960969
var2csa	18.191	17.783		17.987	2.652	6.28588117	var2csa	19.293	18.908		19.101	2.510	5.69657391
91	22.694	22.948		22.821	-2.182	0.22034659	91	23.319	23.410		23.365	-1.754	0.2965596
92	22.530	22.499		22.514	-1.875	0.27258083	92	22.981	22.978		22.980	-1.369	0.38724271
93	24.331	Undetermined		24.331	-3.692	0.07736174	93	24.482	24.825		24.654	-3.043	0.12135298
94	27.283	27.791		27.537	-6.898	0.00838651	94	26.959	27.591		27.275	-5.664	0.01972563
95	23.637	23.657		23.647	-3.008	0.12430649	95	23.998	24.211		24.105	-2.494	0.17753988
96	26.066	26.741		26.404	-5.765	0.01839369	96	27.652	26.212		26.932	-5.321	0.02501997
97	25.759	26.268		26.014	-5.375	0.02410493	97	25.939	26.876		26.408	-4.797	0.03597948
60	20.637	20.641		20.639	0.000	1.00012165	60	21.590	21.631		21.611	0.000	1.00024055

(Rep 1) MCM +30nM Spd 6Wks							(Rep 1) (-)TrpArg +10uM Spd 6Wks						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	22.610	22.835		22.722	-3.037	0.12182406	1	20.489	20.506		20.497	-4.997	0.0313048
3	25.791	25.977		25.884	-6.199	0.01361174	3	22.020	22.251		22.135	-6.635	0.01005866
4	25.594	25.990		25.792	-6.107	0.01451122	4	23.271	23.349		23.310	-7.810	0.00445558
5	23.749	23.900		23.825	-4.140	0.05673516	5	21.311	21.310		21.311	-5.811	0.01781345
6	22.459	21.967		22.213	-2.528	0.1733841	6	21.204	21.682		21.443	-5.943	0.0162552
7	22.529	22.579		22.554	-2.869	0.13685851	7	21.515	21.572		21.543	-6.043	0.01516446
8	24.332	24.256		24.294	-4.609	0.04099046	8	22.710	22.483		22.596	-7.096	0.00730793
9	21.581	21.695		21.638	-1.953	0.25823429	9	21.174	20.757		20.965	-5.465	0.02263726
11	25.404	24.976		25.190	-5.505	0.02201651	11	22.928	23.227		23.078	-7.578	0.00523406
12	24.021	24.281		24.151	-4.466	0.04524101	12	21.239	21.271		21.255	-5.755	0.01851752
13	22.361	22.361		22.361	-2.676	0.15648014	13	20.238	20.535		20.386	-4.886	0.03381338
15	25.174	25.202		25.188	-5.503	0.02205312	15	21.700	21.439		21.570	-6.070	0.01488892
17	22.001	22.197		22.099	-2.414	0.1876916	17	19.657	19.551		19.604	-4.104	0.05814887
18	21.549	21.736		21.642	-1.957	0.25753501	18	19.899	19.852		19.876	-4.376	0.04817395
19	23.265	23.010		23.137	-3.452	0.09135285	19	20.658	20.789		20.723	-5.223	0.02676803
20	30.013	29.750		29.881	-10.196	0.00085241	20	27.290	26.955		27.122	-11.622	0.00031716
21	24.829	25.223		25.026	-5.341	0.02467226	21	22.321	22.533		22.427	-6.927	0.00821876
22	24.611	24.428		24.519	-4.834	0.03505431	22	26.464	26.639		26.551	-11.051	0.00047118
23	24.586	24.578		24.582	-4.897	0.03356699	23	22.226	22.029		22.128	-6.628	0.01011284
25	26.728	26.458		26.593	-6.908	0.00832788	25	23.752	23.775		23.763	-8.263	0.00325452
26	25.989	26.513		26.251	-6.566	0.0105534	26	20.462	20.968		20.715	-5.215	0.02692637
27	21.833	21.820		21.827	-2.142	0.22659517	27	18.503	18.618		18.561	-3.061	0.11983751
28	23.676	23.840		23.758	-4.073	0.05940617	28	21.225	20.771		20.998	-5.498	0.02212802
30	24.859	24.524		24.692	-5.007	0.0311082	30	22.739	22.738		22.739	-7.239	0.00662166
31	22.273	22.211		22.242	-2.557	0.16993195	31	17.155	17.268		17.211	-1.711	0.30542065
34	23.775	23.665		23.720	-4.035	0.06100372	34	18.766	18.568		18.667	-3.167	0.11132936
35	24.794	24.875		24.834	-5.149	0.02817971	35	22.821	22.786		22.804	-7.304	0.00633014
36	25.683	25.482		25.582	-5.897	0.01677743	36	22.392	22.462		22.427	-6.927	0.0082204
37	27.478	27.548		27.513	-7.828	0.0044016	37	23.499	23.331		23.415	-7.915	0.00414292
38	24.370	24.395		24.382	-4.697	0.03854241	38	21.257	21.492		21.375	-5.875	0.01704406
39	24.313	24.310		24.311	-4.626	0.040491	39	22.173	22.216		22.195	-6.695	0.00965355
40	35.360	38.691		37.025	-17.340	6.0258E-06	40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
41	23.580	23.400		23.490	-3.805	0.07153988	41	23.324	22.720		23.022	-7.522	0.00544073
43	22.758	23.140		22.949	-3.264	0.10409744	43	21.511	21.401		21.456	-5.956	0.01610857
44	25.391	25.634		25.512	-5.827	0.01761086	44	24.255	24.344		24.300	-8.800	0.0022439
45	23.331	23.191		23.261	-3.576	0.08383907	45	19.142	18.883		19.012	-3.512	0.08762632
46	24.700	24.901		24.800	-5.115	0.02884774	46	20.767	20.813		20.790	-5.290	0.02555809
47	35.854	33.037		34.446	-14.761	3.6028E-05	47	28.996	28.670		28.833	-13.333	9.6893E-05
49	22.677	22.721		22.699	-3.014	0.12383118	49	18.505	18.450		18.478	-2.978	0.12696436
50	33.843	33.037		33.440	-13.755	7.2314E-05	50	29.660	28.812		29.236	-13.736	7.3301E-05
51	25.627	25.744		25.685	-6.000	0.01562183	51	21.474	21.915		21.695	-6.195	0.01365316
52	27.170	26.757		26.963	-7.278	0.00644116	52	23.801	23.468		23.635	-8.135	0.00355766
53	23.021	23.206		23.114	-3.429	0.09287084	53	20.406	20.370		20.388	-4.888	0.03378233
54	17.677	17.027		17.352	2.333	5.03775688	54	14.049	13.580		13.815	1.685	3.21625044
55	25.394	25.676		25.535	-5.850	0.01733264	55	21.876	22.302		22.089	-6.589	0.0103899
56	23.915	23.441		23.678	-3.993	0.06281622	56	21.802	21.891		21.846	-6.346	0.01229013
57	23.974	24.285		24.130	-4.445	0.045918	57	21.199	21.293		21.246	-5.746	0.01863223
58	23.207	Undetermined		23.207	-3.522	0.08707691	58	19.980	20.146		20.063	-4.563	0.04230113
var2csa	17.949	17.728		17.839	1.846	3.59602737	var2csa	14.656	14.188		14.422	1.078	2.11121572
91	23.363	23.289		23.326	-3.641	0.08014343	91	19.421	19.551		19.486	-3.986	0.06309793
92	22.436	22.505		22.471	-2.786	0.14501374	92	18.376	18.239		18.307	-2.807	0.14285926
93	24.854	24.576		24.715	-5.030	0.03060509	93	20.433	20.374		20.404	-4.904	0.0334017
94	26.784	26.841		26.812	-7.127	0.00715331	94	24.435	24.200		24.317	-8.817	0.00221687
95	24.226	24.468		24.347	-4.662	0.03950775	95	22.738	22.752		22.745	-7.245	0.00659394
96	27.255	26.010		26.632	-6.947	0.00810212	96	24.521	24.883		24.702	-9.202	0.00169781
97	25.495	25.791		25.643	-5.958	0.01608952	97	23.506	23.657		23.581	-8.081	0.00369194
60	19.718	19.651		19.685	0.000	1.00027383	60	15.436	15.576		15.506	-0.006	0.99602061

(Rep 1) (-)TrpArg +30nM Spd 6Wks							(Rep 2) MCM Day 0						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	22.724	22.701		22.713	-3.940	0.06517525	1	23.601	24.460		24.031	-7.110	0.00724034
3	25.725	25.938		25.831	-7.058	0.00750342	3	25.401	24.999		25.200	-8.279	0.00321992
4	25.795	25.787		25.791	-7.018	0.00771588	4	25.584	22.796		24.190	-7.269	0.00648407
5	24.554	24.668		24.611	-5.838	0.01748619	5	25.020	24.682		24.851	-7.930	0.00409947
6	24.165	24.426		24.295	-5.522	0.02175682	6	24.598	25.523		25.060	-8.139	0.00354683
7	22.698	22.720		22.709	-3.936	0.06533917	7	23.021	22.515		22.768	-5.847	0.01737424
8	23.878	23.853		23.865	-5.092	0.02930992	8	24.390	24.461		24.425	-7.504	0.0055082
9	22.645	22.387		22.516	-3.743	0.07470264	9	23.827	23.574		23.701	-6.780	0.00910094
11	24.797	24.693		24.745	-5.972	0.01593447	11	26.189	25.783		25.986	-9.065	0.00186681
12	24.518	24.600		24.559	-5.786	0.01812032	12	24.149	24.341		24.245	-7.324	0.00624066
13	22.453	22.349		22.401	-3.628	0.08089532	13	22.224	22.200		22.212	-5.291	0.02553798
15	24.575	24.847		24.711	-5.938	0.01630879	15	22.966	23.432		23.199	-6.278	0.01288757
17	22.237	22.181		22.209	-3.436	0.09237258	17	22.293	22.766		22.530	-5.609	0.02049441
18	22.326	22.382		22.354	-3.581	0.08357131	18	22.229	21.981		22.105	-5.184	0.02750187
19	22.251	22.355		22.303	-3.530	0.08654771	19	23.568	23.174		23.371	-6.450	0.0114402
20	26.791	29.727		28.259	-9.486	0.00139482	20	25.539	25.600		25.570	-8.649	0.00249191
21	24.487	24.237		24.362	-5.589	0.02077433	21	23.902	23.466		23.684	-6.763	0.00920871
22	25.477	25.847		25.662	-6.889	0.00843684	22	22.805	22.857		22.831	-5.910	0.01663223
23	24.754	24.495		24.625	-5.852	0.01731697	23	25.117	24.788		24.953	-8.032	0.00382178
25	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!	25	26.847	26.391		26.619	-9.698	0.00120403
26	24.725	24.368		24.546	-5.773	0.01828377	26	23.983	23.781		23.882	-6.961	0.00802812
27	20.759	20.970		20.865	-2.092	0.23462091	27	21.447	21.190		21.319	-4.398	0.04744375
28	22.957	22.922		22.939	-4.166	0.05569432	28	24.504	23.996		24.250	-7.329	0.00621987
30	25.801	25.650		25.726	-6.953	0.0080739	30	25.947	24.521		25.234	-8.313	0.00314473
31	21.608	21.305		21.456	-2.683	0.15569473	31	20.916	21.179		21.047	-4.126	0.05725635
34	22.583	22.559		22.571	-3.798	0.07190017	34	23.646	21.819		22.732	-5.811	0.01780755
35	26.147	25.464		25.806	-7.033	0.00763847	35	25.479	25.709		25.594	-8.673	0.00245038
36	25.975	25.646		25.811	-7.038	0.00761055	36	25.937	25.420		25.679	-8.758	0.0023105
37	25.713	26.361		26.037	-7.264	0.00650538	37	26.249	24.947		25.598	-8.677	0.00244294
38	24.438	24.567		24.502	-5.729	0.01884896	38	23.703	23.783		23.743	-6.822	0.00883731
39	24.277	24.731		24.504	-5.731	0.01882559	39	23.651	21.647		22.649	-5.728	0.01886761
40	38.123	39.379		38.751	-19.978	9.6848E-07	40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
41	24.511	24.948		24.730	-5.957	0.01610262	41	24.991	25.708		25.349	-8.428	0.00290275
43	24.136	24.430		24.283	-5.510	0.02194457	43	24.423	24.546		24.484	-7.563	0.0052869
44	26.002	26.502		26.252	-7.479	0.00560515	44	25.708	25.679		25.694	-8.773	0.00228621
45	22.499	22.546		22.523	-3.750	0.07434685	45	24.218	23.944		24.081	-7.160	0.00699192
46	24.195	23.921		24.058	-5.285	0.02564474	46	23.951	23.587		23.769	-6.848	0.00868112
47	32.500	Undetermined		32.500	-13.727	7.375E-05	47	28.377	28.184		28.281	-11.360	0.00038058
49	22.253	22.285		22.269	-3.496	0.08864091	49	23.796	23.538		23.667	-6.746	0.00931965
50	32.674	33.701		33.188	-14.415	4.5788E-05	50	23.604	23.856		23.730	-6.809	0.0089193
51	25.519	24.888		25.204	-6.431	0.01159292	51	24.349	24.138		24.244	-7.323	0.00624589
52	25.887	26.189		26.038	-7.265	0.00650186	52	25.342	24.710		25.026	-8.105	0.00363233
53	23.253	23.332		23.292	-4.519	0.04360317	53	22.889	23.513		23.201	-6.280	0.0128722
54	18.267	16.276		17.271	1.502	2.83181496	54	16.624	16.540		16.582	0.339	1.26509524
55	23.694	Undetermined		23.694	-4.921	0.03300286	55	25.292	24.714		25.003	-8.082	0.0036907
56	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!	56	21.276	21.325		21.300	-4.379	0.04804759
57	23.499	23.562		23.530	-4.757	0.03698036	57	22.663	21.959		22.311	-5.390	0.02384274
58	22.607	22.606		22.606	-3.833	0.07015297	58	24.549	23.742		24.145	-7.224	0.00668741
var2csa	15.808	16.491		16.150	2.623	6.16203125	var2csa	20.342	19.355		19.848	-2.927	0.13147656
91	23.611	23.320		23.466	-4.693	0.03867041	91	22.914	22.475		22.695	-5.774	0.01828088
92	22.008	22.257		22.132	-3.359	0.09744706	92	22.543	21.122		21.832	-4.911	0.03323349
93	24.621	24.344		24.482	-5.709	0.01911049	93	23.756	23.652		23.704	-6.783	0.00908313
94	27.056	27.620		27.338	-8.565	0.00264022	94	26.789	26.768		26.778	-9.857	0.00107805
95	23.816	23.863		23.839	-5.066	0.02984358	95	24.447	24.199		24.323	-7.402	0.00591164
96	25.932	25.686		25.809	-7.036	0.00761974	96	24.000	23.909		23.955	-7.034	0.00763313
97	25.133	25.517		25.325	-6.552	0.01065808	97	24.631	24.557		24.594	-7.673	0.00489838
60	18.946	18.600		18.773	0.000	0.99984509	60	16.900	16.943		16.921	0.000	0.99966527

(Rep 2) MCM 3Wks							(Rep 2) MCM +100uM Spd 3Wks						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	23.497	23.593		23.545	-5.613	0.02043499	1	21.444	21.273		21.358	-4.131	0.05705978
3	26.313	25.953		26.133	-8.201	0.00339795	3	23.185	22.517		22.851	-5.624	0.02027869
4	25.636	26.994		26.315	-8.383	0.00299546	4	23.645	23.258		23.451	-6.224	0.01337345
5	24.793	25.200		24.997	-7.065	0.00746992	5	21.816	21.860		21.838	-4.611	0.04091207
6	24.782	24.754		24.768	-6.836	0.0087536	6	22.283	22.644		22.463	-5.236	0.02652659
7	23.874	24.242		24.058	-6.126	0.01431861	7	21.557	21.435		21.496	-4.269	0.05186823
8	24.945	24.871		24.908	-6.976	0.00794218	8	22.573	21.970		22.272	-5.045	0.03030008
9	23.753	24.374		24.064	-6.132	0.01426181	9	22.801	22.546		22.673	-5.446	0.0229381
11	26.754	26.523		26.638	-8.706	0.0023939	11	25.555	25.517		25.536	-8.309	0.00315358
12	24.203	24.019		24.111	-6.179	0.01380349	12	22.419	22.340		22.379	-5.152	0.02811819
13	23.125	22.935		23.030	-5.098	0.02919465	13	20.064	20.318		20.191	-2.964	0.12815419
15	23.571	23.602		23.586	-5.654	0.01985441	15	24.580	24.212		24.396	-7.169	0.00695009
17	22.240	22.450		22.345	-4.413	0.04694284	17	23.511	23.510		23.510	-6.283	0.01283831
18	22.212	22.206		22.209	-4.277	0.05158759	18	19.250	19.017		19.134	-1.907	0.26669977
19	24.380	23.960		24.170	-6.238	0.01324504	19	21.096	21.290		21.193	-3.966	0.06398267
20	31.190	31.814		31.502	-13.570	8.2239E-05	20	25.927	24.939		25.433	-8.206	0.00338602
21	25.495	25.019		25.257	-7.325	0.00623764	21	23.099	23.099		23.099	-5.872	0.01706875
22	23.645	23.598		23.621	-5.689	0.01937792	22	21.062	20.899		20.981	-3.754	0.07412831
23	25.981	26.140		26.061	-8.129	0.00357291	23	23.358	23.545		23.451	-6.224	0.01337413
25	29.742	27.721		28.731	-10.799	0.0005611	25	24.452	24.020		24.236	-7.009	0.00776312
26	24.661	24.758		24.709	-6.777	0.00911659	26	21.640	21.773		21.706	-4.479	0.04483336
27	21.587	21.730		21.658	-3.726	0.07554683	27	18.746	18.923		18.834	-1.607	0.32822954
28	23.335	22.992		23.163	-5.231	0.0266191	28	21.717	21.967		21.842	-4.615	0.04081983
30	25.800	26.400		26.100	-8.168	0.00347721	30	23.201	23.261		23.231	-6.004	0.01557862
31	22.183	22.250		22.216	-4.284	0.05132233	31	19.288	19.250		19.269	-2.042	0.24276848
34	23.675	23.585		23.630	-5.698	0.01926234	34	21.029	20.824		20.926	-3.699	0.07699302
35	27.455	28.142		27.798	-9.866	0.00107135	35	23.144	23.931		23.538	-6.311	0.01259756
36	25.572	26.379		25.975	-8.043	0.00379065	36	23.821	23.988		23.905	-6.678	0.00976935
37	28.712	28.944		28.828	-10.896	0.00052476	37	26.966	26.676		26.821	-9.594	0.00129412
38	24.279	25.425		24.852	-6.920	0.008258	38	22.474	23.114		22.794	-5.567	0.02109512
39	26.614	26.203		26.408	-8.476	0.00280797	39	22.884	22.802		22.843	-5.616	0.02039092
40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	0	40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	0
41	26.907	27.817		27.362	-9.430	0.00144984	41	26.689	26.519		26.604	-9.377	0.00150424
43	25.345	25.945		25.645	-7.713	0.00476491	43	24.166	23.351		23.758	-6.531	0.01081025
44	30.197	29.845		30.021	-12.089	0.00022952	44	25.687	25.862		25.775	-8.548	0.0026722
45	22.556	22.804		22.680	-4.748	0.03721484	45	22.588	22.613		22.601	-5.374	0.02412071
46	24.531	24.188		24.360	-6.428	0.01161715	46	23.500	23.159		23.330	-6.103	0.01455294
47	31.075	Undetermined		31.075	-13.143	0.00011056	47	26.380	26.580		26.480	-9.253	0.00163858
49	25.015	24.658		24.837	-6.905	0.00834618	49	19.888	19.501		19.694	-2.467	0.18083452
50	32.584	31.087		31.836	-13.904	6.5255E-05	50	31.830	30.836		31.333	-14.106	5.6703E-05
51	25.250	25.439		25.344	-7.412	0.00587023	51	22.899	22.551		22.725	-5.498	0.02212822
52	28.065	27.428		27.746	-9.814	0.00111063	52	25.743	25.855		25.799	-8.572	0.00262767
53	25.134	25.411		25.273	-7.341	0.00616992	53	21.965	21.948		21.957	-4.730	0.03768399
54	17.292	18.588		17.940	-0.008	0.99426753	54	15.871	15.385		15.628	1.599	3.02952897
55	26.675	31.210		28.943	-11.011	0.00048474	55	25.444	25.367		25.405	-8.178	0.00345171
56	24.498	24.614		24.556	-6.624	0.0101381	56	22.016	22.194		22.105	-4.878	0.03401467
57	25.247	25.304		25.276	-7.344	0.00615676	57	20.740	20.718		20.729	-3.502	0.08825914
58	24.184	24.487		24.335	-6.403	0.01181339	58	23.449	23.722		23.585	-6.358	0.01218773
var2csa	20.018	20.331		20.174	-2.242	0.21135146	var2csa	17.913	18.229		18.071	-0.844	0.55707153
91	23.359	22.924		23.142	-5.210	0.02702585	91	20.888	21.344		21.116	-3.889	0.06751529
92	23.147	23.296		23.221	-5.289	0.02557247	92	20.868	20.740		20.804	-3.577	0.08377836
93	23.809	23.797		23.803	-5.871	0.01708726	93	21.579	21.778		21.679	-4.452	0.04569997
94	28.491	29.293		28.892	-10.960	0.00050206	94	25.725	25.841		25.783	-8.556	0.00265763
95	25.756	25.999		25.877	-7.945	0.00405724	95	22.661	22.641		22.651	-5.424	0.02329369
96	27.907	27.980		27.943	-10.011	0.00096886	96	24.574	24.318		24.446	-7.219	0.00671067
97	26.965	27.199		27.082	-9.150	0.00175989	97	24.680	24.247		24.464	-7.237	0.00663093
60	18.109	17.754		17.932	0.000	1.00024714	60	17.340	17.214		17.277	-0.050	0.96586134

(Rep 2) MCM +10uM Spd 3Wks							(Rep 2) MCM +30nM Spd 3Wks						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	21.418	21.327		21.373	-3.641	0.08016943	1	Undetermine	Undetermined		0	0	0
3	22.588	22.620		22.604	-4.872	0.03414935	3	22.773	22.471		22.622	-5.073	0.02971098
4	22.996	23.174		23.085	-5.353	0.02446874	4	23.630	23.517		23.574	-6.025	0.01535934
5	21.917	21.675		21.796	-4.064	0.05978395	5	22.207	22.172		22.189	-4.640	0.04009513
6	21.555	21.656		21.605	-3.873	0.06824277	6	22.263	22.287		22.275	-4.726	0.03778202
7	21.454	21.405		21.429	-3.697	0.07708212	7	21.506	21.447		21.477	-3.928	0.06571971
8	22.162	21.923		22.043	-4.723	0.03787716	8	22.395	22.328		22.361	-4.812	0.03558965
9	21.426	21.387		21.406	-4.086	0.05886537	9	21.754	21.975		21.865	-4.316	0.05021407
10	16.376	16.295		16.335	0.985	1.97888531	10	16.312	16.278		16.295	1.254	2.38471854
11	23.525	23.650		23.587	-6.267	0.01298152	11	23.478	23.593		23.536	-5.987	0.01577079
12	22.195	22.445		22.320	-5.000	0.0312562	12	21.149	20.842		20.996	-3.447	0.09171474
13	20.081	20.132		20.107	-2.787	0.14491913	13	20.463	20.531		20.497	-2.948	0.12956464
15	20.623	20.543		20.583	-3.263	0.10418829	15	22.236	21.913		22.075	-4.526	0.04341876
17	20.014	19.971		19.993	-2.673	0.15682941	17	20.837	20.784		20.810	-3.261	0.10428088
18	20.186	20.188		20.187	-2.867	0.13705587	18	21.763	21.422		21.593	-4.044	0.06063714
19	20.985	21.255		21.120	-3.800	0.07177511	19	21.977	21.618		21.798	-4.249	0.0526096
20	22.595	23.274		22.934	-5.614	0.02041177	20	23.425	23.386		23.405	-5.856	0.01726201
21	21.793	21.626		21.710	-4.390	0.04770701	21	21.313	21.370		21.341	-3.792	0.07216917
22	21.458	21.437		21.448	-4.128	0.05720216	22	20.520	20.476		20.498	-2.949	0.12952985
23	23.123	23.154		23.138	-5.818	0.0177208	23	22.562	22.478		22.520	-4.971	0.03188134
25	23.597	23.407		23.502	-6.182	0.01377025	25	24.301	23.846		24.074	-6.525	0.01086052
26	21.693	21.595		21.644	-4.324	0.04992887	26	22.237	22.195		22.216	-4.667	0.039362
27	19.547	19.787		19.667	-2.347	0.19654322	27	19.465	19.451		19.458	-1.909	0.26634618
28	22.299	22.159		22.229	-4.909	0.03328357	28	23.929	23.751		23.840	-6.291	0.0127688
30	22.660	22.700		22.680	-5.360	0.02435421	30	23.509	23.563		23.536	-5.987	0.01576636
31	18.260	18.387		18.324	-1.004	0.49871811	31	18.385	18.407		18.396	-0.847	0.55595981
34	20.538	20.502		20.520	-3.200	0.10883425	34	20.372	20.437		20.405	-2.856	0.13815604
35	22.728	22.629		22.679	-5.359	0.02437234	35	23.133	22.994		23.063	-5.514	0.02187736
36	22.494	22.570		22.532	-5.212	0.02698033	36	22.881	23.239		23.060	-5.511	0.02193188
37	21.340	21.263		21.302	-3.982	0.06329744	37	22.556	22.577		22.566	-5.017	0.03087947
38	22.491	22.494		22.493	-5.173	0.02772366	38	22.822	22.739		22.780	-5.231	0.0266228
39	21.213	21.189		21.201	-3.881	0.06785771	39	21.822	21.880		21.851	-4.302	0.05069631
40	Undetermine	32.773		32.773	-15.453	2.2301E-05	40	Undetermine	34.448		34.448	-16.899	8.1805E-06
41	24.849	24.708		24.779	-7.459	0.00568478	41	25.242	25.604		25.423	-7.874	0.0042633
43	21.816	21.624		21.720	-4.400	0.04736526	43	22.794	22.789		22.791	-5.242	0.02641696
44	23.313	23.400		23.357	-6.037	0.01523258	44	24.524	24.565		24.544	-6.995	0.00783776
45	20.374	20.317		20.346	-3.026	0.12280511	45	21.379	21.341		21.360	-3.811	0.07123808
46	21.633	21.725		21.679	-4.359	0.04874344	46	21.239	21.245		21.242	-3.693	0.07731639
47	26.177	25.906		26.041	-8.721	0.00236926	47	27.192	27.561		27.377	-9.828	0.0011003
49	20.981	20.926		20.954	-3.634	0.0805724	49	21.741	21.760		21.751	-4.202	0.05434454
50	25.604	26.124		25.864	-8.544	0.00267888	50	27.958	27.478		27.718	-10.169	0.00086866
51	22.461	22.318		22.390	-5.070	0.02977881	51	21.889	21.970		21.930	-4.381	0.04800871
52	23.928	23.772		23.850	-6.530	0.01082384	52	24.227	24.107		24.167	-6.618	0.01018178
53	20.804	20.780		20.792	-3.472	0.09011337	53	20.699	20.661		20.680	-3.131	0.11417728
54	15.347	15.358		15.352	1.968	3.91102227	54	14.945	14.862		14.903	2.646	6.25865651
55	23.833	23.902		23.867	-6.547	0.01069243	55	23.231	22.722		22.977	-5.428	0.02323527
56	22.239	22.420		22.330	-5.010	0.03104056	56	22.475	22.477		22.476	-4.927	0.03287125
57	21.387	21.354		21.371	-4.051	0.06034682	57	21.430	21.557		21.493	-3.944	0.0649571
58	22.786	23.020		22.903	-5.583	0.02085849	58	24.128	24.432		24.280	-6.731	0.00941364
75	16.850	16.900		16.875	0.445	1.36143301	75	16.747	16.867		16.807	0.742	1.67284294
79	17.756	17.709		17.732	-0.412	0.75145911	79	16.750	16.792		16.771	0.778	1.71519547
var2csa	16.260	16.026		16.143	1.177	2.26101333	var2csa	16.199	16.172		16.185	1.364	2.57319181
91	20.018	19.971		19.995	-2.675	0.15659836	91	20.749	20.719		20.734	-3.185	0.10997565
92	20.527	20.762		20.644	-3.324	0.09982552	92	20.010	19.992		20.001	-2.452	0.18278866
93	20.569	20.524		20.546	-3.226	0.10686786	93	21.628	21.613		21.621	-4.072	0.05947363
94	23.781	23.640		23.711	-6.391	0.01191876	94	24.544	24.308		24.426	-6.877	0.00850784
95	22.332	22.297		22.314	-4.994	0.03137425	95	23.701	23.680		23.690	-6.141	0.01416587
96	23.486	23.310		23.398	-6.078	0.01480441	96	24.388	24.893		24.640	-7.091	0.00733393
97	23.280	23.277		23.279	-5.959	0.01608018	97	23.337	23.288		23.312	-5.763	0.01841201
60	17.372	17.269		17.320	0.000	0.99976852	60	17.592	17.505		17.549	0.000	1.00023188

(Rep 2) (-)TrpArg +100uM Spd 3Wks							(Rep 2) MCM 6Wks						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	25.318	24.147		24.733	-7.828	0.00440234	1	24.739	24.425		24.582	-7.096	0.00730889
3	27.010	26.241		26.625	-9.720	0.00118541	3	27.126	26.383		26.754	-9.268	0.00162165
4	24.674	24.545		24.610	-7.705	0.00479397	4	27.205	26.472		26.838	-9.352	0.00152983
5	25.354	25.934		25.644	-8.739	0.00234016	5	25.763	25.568		25.665	-8.179	0.00344943
6	24.808	25.146		24.977	-8.072	0.00371614	6	25.560	25.538		25.549	-8.063	0.0037386
7	24.728	24.496		24.612	-7.707	0.00478615	7	23.954	23.810		23.882	-6.396	0.01187167
8	25.953	25.478		25.715	-8.810	0.00222783	8	25.522	25.523		25.523	-8.037	0.00380811
9	26.810	26.241		26.525	-9.620	0.00127042	9	24.623	24.846		24.734	-7.248	0.00657687
11	27.303	27.221		27.262	-10.357	0.00076262	11	27.223	27.361		27.292	-9.806	0.00111734
12	26.950	26.517		26.733	-9.828	0.0011	12	24.487	24.475		24.481	-6.995	0.00784033
13	23.862	23.429		23.645	-6.740	0.00935302	13	23.919	23.688		23.803	-6.317	0.01254066
15	27.570	27.536		27.553	-10.648	0.00062328	15	25.465	25.327		25.396	-7.910	0.00415782
17	27.333	26.910		27.121	-10.216	0.00084054	17	23.883	23.390		23.636	-6.150	0.0140792
18	25.973	25.895		25.934	-9.029	0.00191453	18	23.797	23.731		23.764	-6.278	0.01288536
19	26.246	25.940		26.093	-9.188	0.00171494	19	24.177	23.864		24.021	-6.535	0.01078522
20	28.798	28.576		28.687	-11.782	0.00028397	20	30.257	30.903		30.580	-13.094	0.00011436
21	24.794	26.398		25.596	-8.691	0.00241938	21	24.910	24.946		24.928	-7.442	0.00575064
22	24.129	24.801		24.465	-7.560	0.00529911	22	23.584	23.867		23.725	-6.239	0.0132357
23	26.194	25.818		26.006	-9.101	0.00182082	23	26.736	25.958		26.347	-8.861	0.00215063
25	29.291	28.811		29.051	-12.146	0.00022059	25	28.843	29.251		29.047	-11.561	0.00033101
26	23.022	23.005		23.014	-6.109	0.01449178	26	26.126	25.855		25.991	-8.505	0.00275329
27	23.792	23.856		23.824	-6.919	0.00826435	27	22.904	22.908		22.906	-5.420	0.02335432
28	24.986	24.122		24.554	-7.649	0.00498262	28	24.931	25.221		25.076	-7.590	0.00519166
30	27.413	27.204		27.309	-10.404	0.00073818	30	27.223	27.323		27.273	-9.787	0.00113182
31	25.875	21.824		23.849	-6.944	0.0081199	31	23.137	22.987		23.062	-5.576	0.02096583
34	22.836	22.732		22.784	-5.879	0.0169888	34	23.946	24.245		24.095	-6.609	0.01024139
35	26.070	27.332		26.701	-9.796	0.001125	35	28.133	26.666		27.400	-9.914	0.00103678
36	25.938	25.941		25.940	-9.035	0.00190662	36	26.139	25.841		25.990	-8.504	0.00275438
37	24.984	24.764		24.874	-7.969	0.00399075	37	29.673	29.238		29.455	-11.969	0.00024943
38	26.158	25.575		25.867	-8.962	0.00200588	38	25.608	25.342		25.475	-7.989	0.00393528
39	26.304	26.512		26.408	-9.503	0.00137809	39	25.844	25.217		25.531	-8.045	0.00378713
40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	0	40	Undetermine	32.576		32.576	-15.090	2.8664E-05
41	27.453	26.918		27.185	-10.280	0.00080416	41	25.913	25.745		25.829	-8.343	0.00307962
43	26.196	25.962		26.079	-9.174	0.00173097	43	25.233	25.190		25.212	-7.726	0.00472353
44	29.541	29.278		29.409	-12.504	0.00017211	44	27.518	28.253		27.886	-10.400	0.00074034
45	24.745	24.773		24.759	-7.854	0.00432197	45	24.967	24.664		24.815	-7.329	0.00621891
46	26.409	26.535		26.472	-9.567	0.00131876	46	24.816	25.179		24.997	-7.511	0.00548058
47	29.913	29.412		29.663	-12.758	0.00014439	47	32.779	32.184		32.481	-14.995	3.0617E-05
49	25.380	25.439		25.409	-8.504	0.00275362	49	25.656	25.736		25.696	-8.210	0.00337723
50	Undetermine	Undetermined		#DIV/0!	#DIV/0!	0	50	31.985	9.491		20.738	-3.252	0.10495621
51	Undetermine	Undetermined		#DIV/0!	#DIV/0!	0	51	26.429	26.523		26.476	-8.990	0.00196684
52	25.163	25.077		25.120	-8.215	0.00336546	52	27.610	27.897		27.753	-10.267	0.0008113
53	Undetermine	Undetermined		#DIV/0!	#DIV/0!	0	53	24.816	24.595		24.706	-7.220	0.00670972
54	18.190	18.208		18.199	-1.294	0.40774907	54	18.379	19.263		18.821	-1.335	0.3964249
55	25.099	27.428		26.264	-9.359	0.00152333	55	26.890	26.684		26.787	-9.301	0.00158514
56	26.469	25.920		26.194	-9.289	0.00159837	56	25.531	25.624		25.578	-8.092	0.00366586
57	24.431	24.591		24.511	-7.606	0.00513152	57	23.639	23.887		23.763	-6.277	0.01289211
58	24.808	25.289		25.049	-8.144	0.00353637	58	24.541	24.763		24.652	-7.166	0.00696245
var2csa	20.862	20.865		20.863	-3.958	0.06432663	var2csa	19.576	19.486		19.531	-2.045	0.24235303
91	24.380	25.140		24.760	-7.855	0.0043197	91	23.958	24.007		23.982	-6.496	0.01107884
92	23.317	23.040		23.179	-6.274	0.01292663	92	23.017	23.481		23.249	-5.763	0.01841461
93	25.463	25.668		25.565	-8.660	0.00247168	93	24.926	25.280		25.103	-7.617	0.00509327
94	28.797	28.795		28.796	-11.891	0.00026331	94	28.943	28.925		28.934	-11.448	0.00035799
95	25.777	26.520		26.148	-9.243	0.00164995	95	26.536	25.810		26.173	-8.687	0.0024263
96	26.765	27.156		26.961	-10.056	0.00093957	96	25.520	25.412		25.466	-7.980	0.00396159
97	27.288	27.398		27.343	-10.438	0.00072086	97	25.581	26.332		25.957	-8.471	0.00281882
60	16.818	16.992		16.905	0.000	0.99992895	60	17.456	17.516		17.486	0.000	1.00020381

(Rep 2) MCM +100uMSpd 6Wks							(Rep 2) MCM +10uMSpd 6Wks						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	24.921	25.169		25.045	-6.177	0.01382152	1	19.464	19.591		19.528	-4.382	0.04796916
3	27.376	28.150		27.763	-8.895	0.0021008	3	20.872	20.971		20.922	-5.776	0.01825248
4	27.971	28.420		28.196	-9.328	0.00155644	4	21.638	21.720		21.679	-6.533	0.01079854
5	26.655	26.435		26.545	-7.677	0.00488731	5	20.118	20.305		20.212	-5.066	0.02986254
6	27.265	27.467		27.366	-8.498	0.00276653	6	20.722	20.917		20.819	-5.673	0.01959588
7	24.012	23.939		23.976	-5.108	0.0290002	7	19.677	19.668		19.672	-4.526	0.04339725
8	24.717	25.188		24.953	-6.085	0.01473533	8	20.500	20.599		20.550	-5.404	0.02362043
9	25.886	25.702		25.794	-6.926	0.00822283	9	19.962	20.023		19.992	-4.846	0.03475954
11	27.226	27.227		27.227	-8.359	0.00304588	11	21.904	21.820		21.862	-6.716	0.00951336
12	25.620	25.701		25.661	-6.793	0.00902071	12	20.480	20.613		20.546	-5.400	0.02367618
13	23.687	23.674		23.681	-4.813	0.03558592	13	18.967	19.175		19.071	-3.925	0.06585168
15	24.479	24.315		24.397	-5.529	0.02166041	15	19.241	19.478		19.360	-4.214	0.05389628
17	24.147	23.913		24.030	-5.162	0.02793464	17	18.555	18.618		18.586	-3.440	0.09211858
18	24.966	24.902		24.934	-6.066	0.01492823	18	18.758	18.808		18.783	-3.637	0.08037257
19	24.565	24.720		24.643	-5.775	0.01826772	19	19.724	19.784		19.754	-4.608	0.041018
20	28.730	29.577		29.153	-10.285	0.0008013	20	24.602	24.972		24.787	-9.641	0.00125257
21	24.905	25.108		25.007	-6.139	0.01419323	21	20.728	20.541		20.634	-5.488	0.02227751
22	23.786	23.516		23.651	-4.783	0.0363148	22	19.671	19.761		19.716	-4.570	0.04210198
23	28.324	27.844		28.084	-9.216	0.00168163	23	21.569	21.607		21.588	-6.442	0.01150228
25	29.001	28.282		28.641	-9.773	0.00114261	25	22.672	22.510		22.591	-7.445	0.00573877
26	26.397	26.406		26.401	-7.533	0.00539846	26	19.394	19.483		19.439	-4.293	0.05102348
27	23.321	23.238		23.279	-4.411	0.04699378	27	17.433	17.384		17.408	-2.262	0.20842735
28	24.459	25.675		25.067	-6.199	0.01361241	28	20.902	20.819		20.860	-5.714	0.01904568
30	28.956	28.213		28.585	-9.717	0.00118858	30	21.559	21.608		21.583	-6.437	0.011539
31	23.485	23.691		23.588	-4.720	0.03794651	31	16.209	16.260		16.235	-1.089	0.47021517
34	24.602	24.358		24.480	-5.612	0.02044655	34	19.015	18.381		18.698	-3.552	0.08526893
35	26.942	27.434		27.188	-8.320	0.00312935	35	21.203	20.977		21.090	-5.944	0.01624574
36	27.743	27.875		27.809	-8.941	0.00203476	36	20.826	20.695		20.761	-5.615	0.02040768
37	26.128	25.633		25.881	-7.013	0.00774448	37	21.496	21.590		21.543	-6.397	0.01186541
38	26.479	27.227		26.853	-7.985	0.00394731	38	21.399	21.419		21.409	-6.263	0.01302359
39	25.474	25.259		25.366	-6.498	0.01106181	39	19.955	20.396		20.175	-5.029	0.03062079
40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!	40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
41	27.178	26.677		26.928	-8.060	0.00374793	41	24.380	24.302		24.341	-9.195	0.00170653
43	25.848	26.380		26.114	-7.246	0.006588	43	20.862	20.999		20.930	-5.784	0.01814452
44	28.373	28.313		28.343	-9.475	0.0014049	44	22.465	22.419		22.442	-7.296	0.00636341
45	25.032	24.931		24.981	-6.113	0.01444402	45	19.010	19.162		19.086	-3.940	0.06514953
46	25.895	25.296		25.595	-6.727	0.00943858	46	19.965	19.933		19.949	-4.803	0.03582177
47	Undetermine	32.709		32.709	-13.841	6.815E-05	47	26.907	26.938		26.923	-11.777	0.00028498
49	25.761	25.671		25.716	-6.848	0.00868175	49	19.600	19.736		19.668	-4.522	0.04352028
50	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!	50	27.235	27.104		27.170	-12.024	0.00024013
51	27.224	27.188		27.206	-8.338	0.00308986	51	20.556	20.577		20.566	-5.420	0.02335593
52	28.292	27.762		28.027	-9.159	0.00174949	52	23.256	23.202		23.229	-8.083	0.00368823
53	24.934	24.719		24.827	-5.959	0.01607961	53	19.302	19.302		19.302	-4.156	0.05608314
54	20.324	19.912		20.118	-1.250	0.42046045	54	12.443	12.642		12.543	2.603	6.07738842
55	26.958	27.451		27.204	-8.336	0.00309374	55	22.293	22.358		22.325	-7.179	0.00689903
56	27.318	27.519		27.419	-8.551	0.00266645	56	20.622	20.583		20.603	-5.457	0.02276839
57	24.376	24.479		24.427	-5.559	0.02120627	57	19.339	19.544		19.441	-4.295	0.05092774
58	24.794	24.766		24.780	-5.912	0.01660291	58	20.996	20.871		20.934	-5.788	0.01810095
var2csa	18.965	19.175		19.070	-0.202	0.86945273	var2csa	13.917	14.030		13.973	1.173	2.25418825
91	24.179	24.687		24.433	-5.565	0.02112162	91	18.330	18.326		18.328	-3.182	0.11017639
92	23.300	23.351		23.326	-4.458	0.04551355	92	18.630	18.699		18.665	-3.519	0.0872579
93	25.730	26.704		26.217	-7.349	0.00613403	93	19.174	19.000		19.087	-3.941	0.06509863
94	27.949	29.047		28.498	-9.630	0.00126206	94	23.390	23.204		23.297	-8.151	0.00351773
95	24.596	24.568		24.582	-5.714	0.0190469	95	20.708	20.842		20.775	-5.629	0.02020617
96	26.265	26.558		26.411	-7.543	0.00536108	96	22.408	22.465		22.437	-7.291	0.00638761
97	25.877	25.862		25.870	-7.002	0.00780423	97	21.342	21.466		21.404	-6.258	0.01306549
60	18.858	18.878		18.868	0.000	0.99999549	60	15.349	14.944		15.146	0.000	0.99968138

(Rep 2) MCM +30nMSpd 6Wks							(Rep 2) (-)TrpArg +100uM Spd 6Wks						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	21.635	21.482		21.559	-4.468	0.04519544	1	29.401	29.407		29.404	-4.040	0.06079346
3	22.831	22.798		22.815	-5.724	0.01892333	3	30.891	30.201		30.546	-5.182	0.02754604
4	23.538	23.676		23.607	-6.516	0.01092454	4	29.938	29.496		29.717	-4.353	0.04893018
5	22.623	22.739		22.681	-5.590	0.02076076	5	29.814	29.175		29.494	-4.130	0.05709758
6	22.399	22.550		22.475	-5.384	0.02395423	6	29.302	29.452		29.377	-4.013	0.06195221
7	21.585	21.500		21.543	-4.452	0.04569911	7	27.877	28.850		28.364	-3.000	0.12503878
8	22.380	22.396		22.388	-5.297	0.02543269	8	27.576	27.224		27.400	-2.036	0.24382889
9	21.794	21.736		21.765	-4.674	0.03916402	9	29.888	29.212		29.550	-4.186	0.05493066
11	22.885	22.941		22.913	-5.822	0.0176774	11	29.398	29.891		29.645	-4.281	0.05144726
12	20.954	20.994		20.974	-3.883	0.06777312	12	26.803	27.645		27.224	-1.860	0.27541051
13	20.258	20.223		20.241	-3.150	0.11268809	13	26.172	26.320		26.246	-0.882	0.54262559
15	21.786	21.812		21.799	-4.708	0.03825236	15	28.635	28.558		28.597	-3.233	0.10638993
17	20.522	20.637		20.579	-3.488	0.0891158	17	32.092	30.361		31.226	-5.862	0.0171891
18	20.868	20.980		20.924	-3.833	0.07017792	18	26.291	27.326		26.808	-1.444	0.36743195
19	21.552	21.475		21.514	-4.423	0.04663161	19	30.162	28.916		29.539	-4.175	0.05534927
20	25.976	26.025		26.000	-8.909	0.00207962	20	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
21	21.713	21.771		21.742	-4.651	0.03979192	21	28.539	30.795		29.667	-4.303	0.05065732
22	20.369	20.316		20.342	-3.251	0.10501585	22	28.491	28.366		28.428	-3.064	0.11954784
23	22.642	22.790		22.716	-5.625	0.02026093	23	29.923	Undetermined		29.923	-4.559	0.04242972
25	24.462	24.505		24.484	-7.393	0.00595151	25	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
26	21.480	21.496		21.488	-4.397	0.04746989	26	28.237	27.561		27.899	-2.535	0.17255916
27	18.960	19.244		19.102	-2.011	0.24808828	27	25.323	25.763		25.543	-0.179	0.8834614
28	22.865	23.212		23.039	-5.948	0.01620361	28	28.805	28.786		28.796	-3.432	0.09267818
30	23.722	23.573		23.647	-6.556	0.0106247	30	30.826	32.159		31.492	-6.128	0.01429605
31	18.139	17.974		18.056	-0.965	0.51210551	31	27.127	27.652		27.389	-2.025	0.24562205
34	20.784	20.741		20.762	-3.671	0.07849663	34	30.267	30.229		30.248	-4.884	0.03387246
35	23.242	22.977		23.110	-6.019	0.01542236	35	Undetermine	30.537		30.537	-5.173	0.0277218
36	22.828	22.840		22.834	-5.743	0.01867179	36	30.528	30.335		30.431	-5.067	0.02982592
37	22.767	22.869		22.818	-5.727	0.01887963	37	Undetermine	32.842		32.842	-7.478	0.00560833
38	23.026	22.901		22.964	-5.873	0.01706872	38	30.208	30.154		30.181	-4.817	0.03546891
39	22.150	21.998		22.074	-4.983	0.03161451	39	30.427	29.878		30.152	-4.788	0.03619106
40	33.260	Undetermined		33.260	-16.169	1.3575E-05	40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
41	26.310	25.880		26.095	-9.004	0.00194787	41	30.625	30.768		30.696	-5.332	0.02481965
43	22.517	22.594		22.555	-5.464	0.02265168	43	32.144	31.327		31.736	-6.372	0.01207394
44	24.485	24.531		24.508	-7.417	0.00585256	44	31.296	31.180		31.238	-5.874	0.01704826
45	21.356	21.334		21.345	-4.254	0.05242021	45	26.673	26.550		26.612	-1.248	0.42108649
46	21.172	21.008		21.090	-3.999	0.06254247	46	31.333	Undetermined		31.333	-5.969	0.01596103
47	28.996	28.975		28.986	-11.895	0.00026265	47	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
49	21.480	21.453		21.466	-4.375	0.04818407	49	28.519	30.150		29.335	-3.971	0.06378697
50	29.052	28.918		28.985	-11.894	0.00026277	50	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
51	22.707	22.715		22.711	-5.620	0.02033536	51	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
52	25.013	24.969		24.991	-7.900	0.00418728	52	31.140	31.045		31.092	-5.728	0.01886211
53	20.842	20.969		20.906	-3.815	0.07105778	53	29.436	Undetermined		29.436	-4.072	0.05944021
54	15.115	14.951		15.033	2.058	4.16428122	54	23.641	23.696		23.669	1.695	3.23884516
55	22.709	23.384		23.047	-5.956	0.01611235	55	32.204	32.026		32.115	-6.751	0.00928339
56	22.513	22.603		22.558	-5.467	0.02260191	56	30.623	Undetermined		30.623	-5.259	0.02611325
57	21.437	21.464		21.451	-4.360	0.04870093	57	29.798	28.745		29.271	-3.907	0.06663863
58	22.993	23.180		23.086	-5.995	0.01567502	58	28.478	28.197		28.338	-2.974	0.12731528
var2csa	15.875	15.627		15.751	1.340	2.53116578	var2csa	23.693	23.562		23.627	1.737	3.33227739
91	20.587	20.762		20.674	-3.583	0.08343034	91	29.230	28.326		28.778	-3.414	0.09382931
92	19.946	19.900		19.923	-2.832	0.14042844	92	29.297	30.993		30.145	-4.781	0.03638382
93	21.443	21.503		21.473	-4.382	0.04795182	93	29.149	31.348		30.249	-4.885	0.03384927
94	24.904	24.837		24.871	-7.780	0.00454985	94	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
95	22.677	22.815		22.746	-5.655	0.01985018	95	27.809	27.763		27.786	-2.422	0.18660008
96	24.251	24.354		24.303	-7.212	0.00674647	96	30.966	31.872		31.419	-6.055	0.01504195
97	23.383	23.541		23.462	-6.371	0.01208144	97	30.679	31.685		31.182	-5.818	0.01772299
60	17.030	17.153		17.091	0.000	0.99974149	60	25.467	25.260		25.364	0.000	1.00021733

(Rep 3) MCM Day 0							(Rep 3) MCM 3Wks						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	21.614	21.505		21.560	-4.206	0.05419936	1	Undetermine	19.945		0		0
3	22.732	22.683		22.708	-5.354	0.02445557	3	21.602	21.555		21.579	-5.680	0.01950703
4	23.339	23.219		23.279	-5.925	0.01646049	4	22.733	22.934		22.834	-6.935	0.00817514
5	22.616	22.658		22.637	-5.283	0.02568652	5	21.275	21.227		21.251	-5.352	0.02448935
6	22.218	21.908		22.063	-4.709	0.03823649	6	22.433	22.553		22.493	-6.594	0.01035236
7	21.455	21.514		21.484	-4.130	0.0571031	7	20.277	20.507		20.392	-4.493	0.04440746
8	22.475	22.591		22.533	-5.179	0.02760681	8	21.486	21.450		21.468	-5.569	0.02106592
9	22.021	21.813		21.917	-4.563	0.0423184	9	20.674	20.481		20.577	-4.678	0.03905544
11	23.498	23.454		23.476	-6.122	0.01435794	11	22.208	22.269		22.238	-6.339	0.01234915
12	22.255	22.010		22.133	-4.779	0.03642507	12	19.935	19.990		19.962	-4.063	0.05981224
13	20.401	20.320		20.361	-3.007	0.12443619	13	18.848	19.165		19.006	-3.107	0.1160309
15	21.697	21.463		21.580	-4.226	0.05345042	15	20.584	20.402		20.493	-4.594	0.04139571
17	20.876	20.733		20.804	-3.450	0.09147561	17	19.575	19.638		19.607	-3.708	0.07653498
18	20.298	20.243		20.271	-2.917	0.13243478	18	19.325	19.258		19.292	-3.393	0.09521525
19	21.367	21.224		21.295	-3.941	0.06510339	19	19.952	20.272		20.112	-4.213	0.05392232
20	25.805	25.783		25.794	-8.440	0.00287917	20	24.894	25.373		25.133	-9.234	0.00166042
21	22.316	22.177		22.246	-4.892	0.03367274	21	21.650	20.941		21.295	-5.396	0.02374428
22	21.781	21.632		21.707	-4.353	0.04894114	22	20.679	20.637		20.658	-4.759	0.0369386
23	23.433	22.922		23.178	-5.824	0.01765447	23	22.226	21.810		22.018	-6.119	0.01438423
25	25.121	25.352		25.236	-7.882	0.00423786	25	23.422	23.517		23.470	-7.571	0.00526063
26	21.757	21.330		21.544	-4.190	0.05480453	26	20.396	18.917		19.656	-3.757	0.07394403
27	19.691	19.679		19.685	-2.331	0.19873426	27	18.192	18.235		18.214	-2.315	0.20103108
28	23.673	23.272		23.473	-6.119	0.01439236	28	21.347	21.369		21.358	-5.459	0.02273301
30	23.455	23.699		23.577	-6.223	0.01338811	30	22.463	22.732		22.598	-6.699	0.00962616
31	18.367	18.306		18.336	-0.982	0.50613109	31	16.876	16.846		16.861	-0.962	0.51322808
34	19.686	19.869		19.777	-2.423	0.18640358	34	18.967	19.301		19.134	-3.235	0.10622833
35	23.008	22.896		22.952	-5.598	0.02064616	35	22.150	21.524		21.837	-5.938	0.01631286
36	22.610	22.602		22.606	-5.252	0.02624451	36	21.693	21.788		21.741	-5.842	0.01743726
37	22.189	22.142		22.165	-4.811	0.03561865	37	21.688	21.770		21.729	-5.830	0.01757936
38	22.139	22.196		22.168	-4.814	0.03556171	38	21.305	21.383		21.344	-5.445	0.02295299
39	22.677	22.741		22.709	-5.355	0.02442957	39	20.715	20.888		20.802	-4.903	0.03343201
40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!	40	34.198	Undetermined		34.198	-18.299	3.1E-06
41	25.729	25.621		25.675	-8.321	0.00312735	41	24.574	24.281		24.427	-8.528	0.00270877
43	22.477	22.490		22.484	-5.130	0.0285654	43	21.688	21.653		21.671	-5.772	0.01830542
44	23.799	23.896		23.848	-6.494	0.01109695	44	23.137	23.000		23.068	-7.169	0.00694662
45	20.866	20.748		20.807	-3.453	0.09131996	45	19.373	20.255		19.814	-3.915	0.06629763
46	21.907	21.749		21.828	-4.474	0.04499768	46	20.414	20.239		20.327	-4.428	0.04646842
47	28.938	28.911		28.925	-11.571	0.00032873	47	26.753	27.214		26.984	-11.085	0.00046043
49	21.516	21.507		21.512	-4.158	0.056033	49	19.345	19.330		19.337	-3.438	0.09223813
50	28.781	29.075		28.928	-11.574	0.00032808	50	28.370	27.663		28.016	-12.117	0.0002251
51	22.392	22.446		22.419	-5.065	0.02987438	51	21.598	21.421		21.509	-5.610	0.02046843
52	24.600	24.498		24.549	-7.195	0.00682554	52	23.573	23.459		23.516	-7.617	0.0050936
53	20.972	20.893		20.932	-3.578	0.08371825	53	19.609	19.681		19.645	-3.746	0.07453568
54	13.949	14.022		13.985	3.369	10.328111	54	12.648	12.461		12.555	3.344	10.1572363
55	23.234	22.893		23.064	-5.710	0.01910673	55	21.799	22.347		22.073	-6.174	0.01385331
56	22.798	22.921		22.859	-5.505	0.02201416	56	21.610	21.715		21.663	-5.764	0.0184046
57	21.767	21.692		21.729	-4.375	0.04818781	57	20.284	20.432		20.358	-4.459	0.04546329
58	23.171	23.021		23.096	-5.742	0.01868526	58	21.576	21.695		21.636	-5.737	0.01875392
var2csa	18.947	18.951		18.949	-1.595	0.33098116	var2csa	16.836	16.931		16.883	-0.984	0.50548091
91	20.770	20.647		20.708	-3.354	0.09776989	91	19.469	19.417		19.443	-3.544	0.08576129
92	20.888	20.867		20.877	-3.523	0.08696574	92	19.249	19.651		19.450	-3.551	0.08531567
93	21.607	21.626		21.617	-4.263	0.05209544	93	20.211	20.410		20.311	-4.412	0.04698187
94	24.832	24.855		24.844	-7.490	0.00556418	94	24.700	24.807		24.754	-8.855	0.00216022
95	22.240	21.957		22.098	-4.744	0.03731076	95	20.567	20.831		20.699	-4.800	0.0358935
96	24.313	24.241		24.277	-6.923	0.00824193	96	23.851	23.576		23.714	-7.815	0.00444128
97	24.264	23.975		24.120	-6.766	0.00919027	97	22.568	22.808		22.688	-6.789	0.00904264
60	17.407	17.300		17.354	0.000	1.00031647	60	15.558	16.240		15.899	0.000	1.00008041

(Rep 3) MCM +10uM Spd 3Wks							(Rep 3) MCM 6Wks						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	19.611	19.779		19.695	-4.243	0.05282252	1	20.963	21.161		21.062	-3.894	0.06724561
3	21.223	21.342		21.282	-5.830	0.01757355	3	22.267	22.310		22.288	-5.120	0.02875084
4	22.622	21.891		22.257	-6.805	0.00894428	4	23.150	23.145		23.147	-5.979	0.01585115
5	20.495	20.615		20.555	-5.103	0.02910149	5	21.843	21.964		21.903	-4.735	0.03753966
6	21.818	21.870		21.844	-6.392	0.01190714	6	22.743	22.346		22.545	-5.377	0.02406888
7	19.806	19.762		19.784	-4.332	0.04965277	7	21.236	21.408		21.322	-4.154	0.05616876
8	20.784	20.786		20.785	-5.333	0.02480941	8	22.371	22.010		22.190	-5.022	0.03076653
9	19.805	19.825		19.815	-4.363	0.04860141	9	21.031	21.376		21.203	-4.035	0.06099106
11	21.805	21.773		21.789	-6.337	0.01237083	11	23.223	23.247		23.235	-6.067	0.01491867
12	19.598	19.593		19.595	-4.143	0.05658798	12	21.369	21.164		21.266	-4.098	0.0583769
13	18.588	18.648		18.618	-3.166	0.11141242	13	19.939	19.903		19.921	-2.753	0.14833901
15	19.909	20.191		20.050	-4.598	0.0412997	15	21.782	21.749		21.765	-4.597	0.04130696
17	19.186	18.799		18.993	-3.541	0.08592984	17	20.432	20.542		20.487	-3.319	0.10019333
18	18.685	18.505		18.595	-3.143	0.11319719	18	19.883	19.965		19.924	-2.756	0.14802918
19	19.640	19.619		19.630	-4.178	0.05526428	19	20.881	20.904		20.893	-3.725	0.0756451
20	24.946	24.812		24.879	-9.427	0.00145281	20	26.737	26.550		26.643	-9.475	0.00140514
21	21.182	20.693		20.937	-5.485	0.02232034	21	22.305	22.401		22.353	-5.185	0.027492
22	19.793	19.745		19.769	-4.317	0.05017277	22	21.011	20.929		20.970	-3.802	0.07169986
23	21.447	21.222		21.335	-5.883	0.01695009	23	22.610	22.690		22.650	-5.482	0.02237903
25	22.595	22.420		22.508	-7.056	0.00751612	25	24.576	24.726		24.651	-7.483	0.00558931
26	20.188	19.874		20.031	-4.579	0.04183366	26	21.345	21.428		21.386	-4.218	0.05373039
27	17.826	17.829		17.827	-2.375	0.19272759	27	19.225	19.342		19.284	-2.116	0.23070325
28	20.983	21.234		21.108	-5.656	0.01982622	28	21.569	22.215		21.892	-4.724	0.03784223
30	21.845	21.879		21.862	-6.410	0.01175787	30	22.938	23.169		23.053	-5.885	0.01691636
31	16.913	16.891		16.902	-1.450	0.36607585	31	18.328	18.359		18.344	-1.176	0.4427282
34	18.993	18.698		18.845	-3.393	0.09516323	34	20.680	20.759		20.719	-3.551	0.0852944
35	21.770	21.681		21.726	-6.274	0.01292567	35	22.709	23.229		22.969	-5.801	0.0179361
36	21.404	21.265		21.335	-5.883	0.01694986	36	22.378	21.957		22.168	-5.000	0.03125159
37	21.884	22.295		22.090	-6.638	0.01004152	37	22.177	21.890		22.033	-4.865	0.0343068
38	20.642	20.403		20.522	-5.070	0.02976391	38	22.396	22.403		22.399	-5.231	0.0266202
39	20.684	20.648		20.666	-5.214	0.02694251	39	21.934	21.990		21.962	-4.794	0.036052
40	Undetermined	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!	40	34.026	Undetermined		34.026	-16.858	8.4213E-06
41	24.223	23.407		23.815	-8.363	0.00303752	41	25.011	25.637		25.324	-8.156	0.00350592
43	20.235	20.278		20.256	-4.804	0.03578549	43	21.716	21.293		21.504	-4.336	0.04950192
44	22.414	21.815		22.115	-6.663	0.00986923	44	22.797	23.870		23.334	-6.166	0.01393058
45	19.216	19.180		19.198	-3.746	0.07454656	45	20.568	20.621		20.595	-3.427	0.09299742
46	19.727	20.174		19.950	-4.498	0.04424036	46	21.403	21.359		21.381	-4.213	0.05391533
47	27.883	25.469		26.676	-11.224	0.00041808	47	28.915	27.417		28.166	-10.998	0.00048902
49	18.961	19.299		19.130	-3.678	0.07813566	49	20.593	21.278		20.935	-3.767	0.07343747
50	26.948	27.106		27.027	-11.575	0.00032774	50	28.804	28.719		28.762	-11.594	0.00032357
51	20.463	20.556		20.510	-5.058	0.03002304	51	21.973	21.998		21.986	-4.818	0.03545661
52	22.929	22.902		22.916	-7.464	0.00566455	52	24.483	24.750		24.616	-7.448	0.00572575
53	19.358	19.467		19.412	-3.960	0.06425199	53	20.680	20.510		20.595	-3.427	0.09295347
54	12.513	11.961		12.237	3.215	9.28711178	54	15.257	14.753		15.005	2.163	4.47912623
55	21.569	22.202		21.885	-6.433	0.01157174	55	23.460	23.543		23.501	-6.333	0.01240404
56	20.971	20.973		20.972	-5.520	0.02179095	56	21.322	21.344		21.333	-4.165	0.05574424
57	20.257	20.163		20.210	-4.758	0.03695438	57	21.416	21.728		21.572	-4.404	0.04723308
58	21.377	21.449		21.413	-5.961	0.01605217	58	22.393	22.405		22.399	-5.231	0.02662766
var2csa	16.474	16.465		16.469	-1.017	0.49403015	var2csa	17.803	17.800		17.801	-0.633	0.64467233
91	18.700	18.805		18.752	-3.300	0.10152553	91	19.895	19.920		19.908	-2.740	0.14973432
92	19.333	19.992		19.663	-4.211	0.05400512	92	20.443	20.620		20.531	-3.363	0.09716158
93	19.471	19.470		19.471	-4.019	0.06169122	93	20.774	20.589		20.682	-3.514	0.08756457
94	23.683	23.650		23.666	-8.214	0.00336692	94	23.747	24.648		24.198	-7.030	0.0076529
95	20.822	20.805		20.814	-5.362	0.02432347	95	22.322	22.174		22.248	-5.080	0.0295695
96	22.886	23.233		23.059	-7.607	0.00512768	96	24.437	24.327		24.382	-7.214	0.00673623
97	22.322	22.592		22.457	-7.005	0.00778492	97	23.374	23.653		23.514	-6.346	0.01229643
60	16.304	14.599		15.452	0.000	1.00028562	60	17.011	17.324		17.168	0.000	1.0002676

(Rep 3) MCM +10uM Spd 6Wks							(Rep 4) MCM Day 0						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	21.362	21.507		21.434	-4.005	0.06226327	1	20.233	19.958		20.096	-3.593	0.08289684
3	22.468	22.561		22.515	-5.086	0.02944381	3	21.699	21.481		21.590	-5.087	0.02942357
4	22.432	23.205		22.819	-5.390	0.02385568	4	21.721	21.730		21.726	-5.223	0.02678179
5	21.951	22.179		22.065	-4.636	0.04022688	5	21.272	21.144		21.208	-4.705	0.03834628
6	22.371	22.940		22.656	-5.227	0.02670726	6	21.316	21.635		21.476	-4.973	0.03185072
7	22.217	22.221		22.219	-4.790	0.03614804	7	19.822	19.841		19.832	-3.329	0.09952843
8	22.834	22.706		22.770	-5.341	0.02467114	8	21.247	21.243		21.245	-4.742	0.03736819
9	21.482	21.378		21.430	-4.001	0.06246614	9	20.554	20.413		20.483	-3.980	0.0633677
11	22.854	23.245		23.050	-5.621	0.02032224	11	22.698	22.507		22.603	-6.100	0.01458345
12	21.431	21.320		21.376	-3.947	0.06485455	12	20.312	20.577		20.444	-3.941	0.06509757
13	20.403	20.371		20.387	-2.958	0.12868374	13	18.727	18.910		18.819	-2.316	0.20086937
15	20.968	21.115		21.042	-3.613	0.08175465	15	20.989	20.872		20.931	-4.428	0.04646936
17	20.270	20.782		20.526	-3.097	0.11684015	17	19.426	19.475		19.450	-2.947	0.12963346
18	20.273	20.203		20.238	-2.809	0.14270501	18	19.397	19.288		19.343	-2.840	0.13968819
19	21.186	21.318		21.252	-3.823	0.07065971	19	19.762	19.783		19.773	-3.270	0.1036731
20	26.409	26.062		26.235	-8.806	0.00223364	20	22.081	22.407		22.244	-5.741	0.01869581
21	22.540	22.346		22.443	-5.014	0.03094198	21	20.740	20.332		20.536	-4.033	0.06107812
22	21.605	21.484		21.544	-4.115	0.05769218	22	20.312	19.932		20.122	-3.619	0.08137012
23	22.900	22.857		22.879	-5.450	0.02288407	23	21.584	21.355		21.469	-4.966	0.0319923
25	24.949	24.337		24.643	-7.214	0.00673615	25	22.873	22.864		22.869	-6.366	0.012127
26	21.653	21.521		21.587	-4.158	0.05601986	26	20.477	20.166		20.321	-3.818	0.0708791
27	19.685	19.686		19.685	-2.256	0.20929289	27	18.378	18.653		18.516	-2.013	0.24780531
28	21.465	20.993		21.229	-3.800	0.07180888	28	21.728	21.592		21.660	-5.157	0.02802241
30	23.403	23.191		23.297	-5.868	0.01712604	30	21.712	21.914		21.813	-5.310	0.02520676
31	18.565	18.621		18.593	-1.164	0.44618693	31	16.967	16.761		16.864	-0.361	0.77854833
34	20.912	20.722		20.817	-3.388	0.09552565	34	18.560	18.792		18.676	-2.173	0.22175207
35	23.505	23.198		23.352	-5.923	0.01648514	35	21.366	20.933		21.150	-4.647	0.03992062
36	22.812	22.900		22.856	-5.427	0.02323939	36	21.661	21.604		21.632	-5.129	0.028572
37	21.390	21.274		21.332	-3.903	0.06683981	37	21.002	20.849		20.926	-4.423	0.0466308
38	22.403	22.379		22.391	-4.962	0.03208991	38	21.180	21.274		21.227	-4.724	0.03783849
39	21.793	21.921		21.857	-4.428	0.04645049	39	20.510	20.461		20.485	-3.982	0.0632765
40	33.638	33.954		33.796	-16.367	1.1832E-05	40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!
41	25.857	25.373		25.615	-8.186	0.00343376	41	23.706	23.679		23.693	-7.190	0.00685017
43	21.543	Undetermined		21.543	-4.114	0.05777045	43	20.848	21.104		20.976	-4.473	0.04502971
44	23.789	23.868		23.828	-6.399	0.01184741	44	21.992	21.895		21.944	-5.441	0.02302683
45	20.497	20.598		20.548	-3.119	0.11514093	45	19.770	19.712		19.741	-3.238	0.10598254
46	21.288	21.402		21.345	-3.916	0.06624244	46	20.297	20.235		20.266	-3.763	0.07365404
47	26.817	25.655		26.236	-8.807	0.00223316	47	25.974	26.175		26.074	-9.571	0.00131443
49	21.372	21.414		21.393	-3.964	0.06407672	49	19.325	19.265		19.295	-2.792	0.14435315
50	27.126	27.659		27.393	-9.964	0.00100148	50	27.103	27.266		27.184	-10.681	0.00060895
51	22.381	22.527		22.454	-5.025	0.03071244	51	20.689	20.698		20.694	-4.191	0.05475413
52	24.823	24.285		24.554	-7.125	0.00716443	52	21.890	21.972		21.931	-5.428	0.02322548
53	20.814	20.890		20.852	-3.423	0.09323306	53	19.598	19.491		19.544	-3.041	0.12147055
54	15.661	15.635		15.648	1.781	3.43667425	54	14.220	13.430		13.825	2.678	6.4004263
55	23.166	23.655		23.410	-5.981	0.01582708	55	21.974	21.912		21.943	-5.440	0.02303218
56	21.775	21.454		21.614	-4.185	0.0549604	56	21.611	21.591		21.601	-5.098	0.02920051
57	21.916	21.874		21.895	-4.466	0.0452389	57	19.848	19.901		19.875	-3.372	0.09660526
58	22.008	22.230		22.119	-4.690	0.03874459	58	21.421	21.614		21.518	-5.015	0.0309368
var2csa	16.527	16.778		16.653	0.776	1.71265859	var2csa	18.190	18.216		18.203	-1.700	0.30781437
91	20.304	20.440		20.372	-2.943	0.13001057	91	19.556	19.810		19.683	-3.180	0.11031849
92	20.931	20.990		20.961	-3.532	0.08647088	92	19.142	19.405		19.273	-2.770	0.14658471
93	20.951	21.129		21.040	-3.611	0.08182537	93	20.478	20.320		20.399	-3.896	0.06717383
94	24.971	24.711		24.841	-7.412	0.00587126	94	23.149	22.985		23.067	-6.564	0.01056871
95	21.985	21.860		21.923	-4.494	0.04439187	95	21.139	20.980		21.060	-4.557	0.04248985
96	23.973	25.179		24.576	-7.147	0.00705556	96	22.535	22.531		22.533	-6.030	0.01530548
97	23.781	24.142		23.962	-6.533	0.01080205	97	22.178	22.217		22.197	-5.694	0.01931138
60	17.461	17.396		17.429	0.000	1.00022485	60	16.411	16.594		16.503	0.000	1.0003085

(Rep 4) MCM 3Wks							(Rep 4,1) MCM + 2mM Put 3Wks						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	21.358	21.366		21.362	-4.561	0.04236091	1	20.813	20.664		20.738	-2.369	0.19352207
3	23.150	23.003		23.077	-6.276	0.01290742	3	22.519	22.411		22.465	-4.096	0.05848316
4	22.949	22.684		22.816	-6.015	0.01545853	4	22.720	22.616		22.668	-4.299	0.05079469
5	22.280	22.414		22.347	-5.546	0.0213977	5	21.568	21.884		21.726	-3.357	0.09759794
6	22.595	22.648		22.621	-5.820	0.01769884	6	22.253	21.945		22.099	-3.730	0.07535766
7	22.340	22.194		22.267	-5.466	0.02262038	7	22.278	22.203		22.240	-3.871	0.06832588
8	22.880	23.733		23.307	-6.506	0.01100389	8	22.327	22.385		22.356	-3.987	0.06307305
9	21.777	21.965		21.871	-5.070	0.02977005	9	20.790	20.585		20.687	-2.318	0.20048997
11	23.522	23.652		23.587	-6.786	0.00906253	11	22.805	22.750		22.778	-4.409	0.04708195
12	21.608	21.639		21.623	-4.822	0.03534321	12	20.484	20.398		20.441	-2.072	0.23782545
13	20.555	20.668		20.611	-3.810	0.0712775	13	19.694	19.531		19.612	-1.243	0.42237138
15	22.114	22.286		22.200	-5.399	0.02369778	15	21.673	21.762		21.717	-3.348	0.09818317
17	20.723	20.793		20.758	-3.957	0.06440797	17	19.842	19.888		19.865	-1.496	0.35459485
18	20.755	20.810		20.782	-3.981	0.06330769	18	19.859	19.871		19.865	-1.496	0.35461353
19	20.898	20.843		20.870	-4.069	0.05957416	19	20.696	20.580		20.638	-2.269	0.20746042
20	25.112	25.313		25.212	-8.411	0.0029375	20	25.000	25.433		25.217	-6.848	0.00868353
21	21.963	21.954		21.959	-5.158	0.02801361	21	21.899	21.758		21.828	-3.459	0.09091669
22	21.431	21.363		21.397	-4.596	0.04135131	22	20.864	20.743		20.803	-2.434	0.18501344
23	22.642	22.780		22.711	-5.910	0.01662927	23	22.240	22.164		22.202	-3.833	0.0701876
25	24.585	24.660		24.622	-7.821	0.00442076	25	23.333	23.460		23.396	-5.027	0.03066471
26	21.406	21.436		21.421	-4.620	0.04066892	26	21.285	20.530		20.907	-2.538	0.17214522
27	20.416	20.301		20.358	-3.557	0.08494363	27	19.594	19.568		19.581	-1.212	0.43173726
28	23.333	23.479		23.406	-6.605	0.01027179	28	21.636	21.549		21.593	-3.224	0.10705592
30	22.926	22.959		22.943	-6.142	0.01416203	30	22.314	22.460		22.387	-4.018	0.06172828
31	18.674	18.734		18.704	-1.903	0.2673435	31	18.281	17.840		18.060	0.309	1.23848638
34	21.210	21.335		21.273	-4.472	0.04506499	34	20.186	20.394		20.290	-1.921	0.2640208
35	23.151	23.157		23.154	-6.353	0.01223229	35	22.512	22.027		22.270	-3.901	0.0669498
36	22.924	22.763		22.844	-6.043	0.01517066	36	22.928	22.816		22.872	-4.503	0.04409215
37	20.798	20.904		20.851	-4.050	0.06035464	37	22.965	23.005		22.985	-4.616	0.04077641
38	22.607	22.707		22.657	-5.856	0.01726597	38	21.332	21.587		21.460	-3.091	0.11738948
39	22.389	22.473		22.431	-5.630	0.02019481	39	21.451	21.409		21.430	-3.061	0.11982418
40	Undetermined	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!	40	32.532	32.340		32.436	-14.067	5.827E-05
41	25.127	25.020		25.073	-8.272	0.00323466	41	23.558	23.249		23.404	-5.035	0.0305084
43	22.323	22.498		22.410	-5.609	0.02048233	43	21.743	21.776		21.759	-3.390	0.09536836
44	23.941	24.005		23.973	-7.172	0.00693464	44	22.701	22.795		22.748	-4.379	0.0480557
45	20.266	20.407		20.337	-3.536	0.08623298	45	20.022	20.187		20.105	-1.736	0.30026901
46	21.675	21.811		21.743	-4.942	0.03253239	46	21.208	21.379		21.294	-2.925	0.13168163
47	29.210	28.438		28.824	-12.023	0.00024023	47	26.451	26.975		26.713	-8.344	0.00307696
49	19.990	19.992		19.991	-3.190	0.10959436	49	20.358	20.472		20.415	-2.046	0.24215665
50	28.828	28.549		28.689	-11.888	0.00026387	50	27.912	27.606		27.759	-9.390	0.00149042
51	22.606	22.725		22.665	-5.864	0.01716562	51	28.122	27.782		27.952	-9.583	0.0013038
52	23.997	24.244		24.120	-7.319	0.0062609	52	23.863	24.112		23.988	-5.619	0.02035308
53	21.219	21.372		21.295	-4.494	0.04436468	53	23.908	23.764		23.836	-5.467	0.02260465
54	15.421	15.140		15.281	1.520	2.86831208	54	14.458	15.027		14.742	3.627	12.3509662
55	23.747	23.828		23.787	-6.986	0.00788692	55	24.446	24.262		24.354	-5.985	0.01579022
56	22.696	23.213		22.955	-6.154	0.01404742	56	21.924	21.773		21.849	-3.480	0.0896386
57	21.973	21.917		21.945	-5.144	0.02828672	57	22.004	21.794		21.899	-3.530	0.08656421
58	21.700	22.001		21.850	-5.049	0.03019613	58	22.210	21.951		22.081	-3.712	0.0763263
var2csa	17.841	17.678		17.760	-0.959	0.5145585	var2csa	16.770	16.491		16.631	1.738	3.33679718
91	20.609	20.446		20.527	-3.726	0.07555285	91	19.862	19.926		19.894	-1.525	0.34740959
92	20.447	20.330		20.389	-3.588	0.08317873	92	20.161	21.341		20.751	-2.382	0.19185663
93	21.116	21.145		21.131	-4.330	0.04973804	93	20.576	20.576		20.576	-2.207	0.21659983
94	24.928	24.525		24.726	-7.925	0.00411366	94	23.576	23.093		23.335	-4.966	0.03200512
95	22.502	22.436		22.469	-5.668	0.01966685	95	21.767	21.781		21.774	-3.405	0.09439233
96	24.377	24.216		24.296	-7.495	0.00554304	96	23.475	22.745		23.110	-4.741	0.03739046
97	23.843	23.666		23.755	-6.954	0.00806811	97	22.996	22.890		22.943	-4.574	0.04198477
60	16.830	16.772		16.801	0.000	1.00009531	60	18.655	18.623		18.639	-0.270	0.82937818

(Rep 4,2) MCM + 2mM Put 3Wks							(Rep 4,1) MCM + 100uM Spd 3Wks						
Primer pair	CT			CT Average	relative to p60	relative copy number	Primer pair	CT			CT Average	relative to p60	relative copy number
1	24.014	24.305		24.160	-2.793	0.14432064	1	21.909	21.795		21.852	-5.278	0.02577553
3	27.347	26.316		26.831	-5.464	0.02265189	3	23.472	23.327		23.399	-6.825	0.0088184
4	25.699	25.643		25.671	-4.304	0.05062064	4	23.370	23.403		23.386	-6.812	0.00889875
5	25.981	26.003		25.992	-4.625	0.0405357	5	23.170	23.126		23.148	-6.574	0.01049577
6	25.492	24.975		25.233	-3.866	0.06855996	6	22.550	22.826		22.688	-6.114	0.01443507
7	25.474	25.500		25.487	-4.120	0.0575187	7	21.781	21.769		21.775	-5.201	0.02719314
8	26.165	25.546		25.856	-4.489	0.04453738	8	22.785	22.748		22.767	-6.193	0.01366996
9	25.331	25.710		25.521	-4.154	0.05619052	9	21.975	22.186		22.081	-5.507	0.02199557
11	27.233	27.491		27.362	-5.995	0.01568316	11	23.504	23.891		23.697	-7.123	0.00717161
12	24.610	24.655		24.633	-3.266	0.10398757	12	21.773	21.822		21.798	-5.224	0.0267632
13	23.211	23.242		23.226	-1.859	0.2755881	13	19.784	19.805		19.795	-3.221	0.10726509
15	25.329	26.213		25.771	-4.404	0.0472449	15	20.796	20.999		20.897	-4.323	0.0499505
17	23.977	24.537		24.257	-2.890	0.13492153	17	20.764	20.762		20.763	-4.189	0.05482877
18	23.372	23.363		23.368	-2.001	0.24985734	18	20.822	20.955		20.889	-4.315	0.05025072
19	24.013	24.446		24.230	-2.863	0.13747198	19	21.005	21.165		21.085	-4.511	0.04385604
20	30.250	29.593		29.922	-8.555	0.00265929	20	24.933	24.989		24.961	-8.387	0.00298728
21	26.928	26.420		26.674	-5.307	0.02526076	21	22.301	22.158		22.230	-5.656	0.01983315
22	24.782	24.695		24.739	-3.372	0.09659709	22	21.553	21.620		21.586	-5.012	0.03098042
23	25.526	25.292		25.409	-4.042	0.06070735	23	23.233	23.223		23.228	-6.654	0.00993078
25	28.645	28.340		28.493	-7.126	0.00716132	25	20.818	20.580		20.699	-4.125	0.05730167
26	24.653	24.729		24.691	-3.324	0.0998666	26	21.295	21.252		21.274	-4.700	0.03848564
27	22.987	21.827		22.407	-1.040	0.48621574	27	19.502	19.453		19.478	-2.904	0.13362066
28	25.447	25.521		25.484	-4.117	0.05763312	28	22.854	22.902		22.878	-6.304	0.01265617
30	27.157	27.922		27.539	-6.172	0.01386632	30	23.801	23.718		23.759	-7.185	0.00687033
31	22.451	22.791		22.621	-1.254	0.41934716	31	18.360	18.399		18.379	-1.805	0.28611241
34	25.353	25.360		25.357	-3.990	0.06294133	34	20.356	20.438		20.397	-3.823	0.07066105
35	28.963	27.472		28.217	-6.850	0.0086656	35	22.887	22.985		22.936	-6.362	0.01215941
36	26.251	26.339		26.295	-4.928	0.03284978	36	23.016	22.797		22.907	-6.333	0.01240872
37	26.686	25.988		26.337	-4.970	0.03190482	37	21.694	21.587		21.640	-5.066	0.02984503
38	24.968	25.882		25.425	-4.058	0.06004201	38	22.421	22.781		22.601	-6.027	0.01533585
39	26.173	26.686		26.429	-5.062	0.02992786	39	21.786	21.745		21.765	-5.191	0.02736903
40	Undetermine	Undetermined		#DIV/0!	#DIV/0!	#DIV/0!	40	38.311	34.803		36.557	-19.983	9.6527E-07
41	27.717	27.514		27.615	-6.248	0.01315479	41	25.190	24.975		25.082	-8.508	0.00274639
43	25.490	25.654		25.572	-4.205	0.0542195	43	23.153	22.643		22.898	-6.324	0.01247943
44	28.181	27.705		27.943	-6.576	0.01047957	44	24.118	23.975		24.047	-7.473	0.00563044
45	23.823	23.726		23.774	-2.407	0.18850707	45	20.631	20.434		20.533	-3.959	0.06431772
46	26.466	25.734		26.100	-4.733	0.03760134	46	21.607	21.659		21.633	-5.059	0.03000359
47	29.367	28.990		29.179	-7.812	0.00445141	47	27.422	26.907		27.165	-10.591	0.00064853
49	24.743	24.525		24.634	-3.267	0.10387652	49	19.770	19.896		19.833	-3.259	0.10446188
50	30.839	31.364		31.102	-9.735	0.00117374	50	27.897	27.169		27.533	-10.959	0.0005024
51	30.729	30.489		30.609	-9.242	0.00165159	51	22.293	22.377		22.335	-5.761	0.01844407
52	28.380	28.757		28.569	-7.202	0.00679291	52	23.270	22.844		23.057	-6.483	0.0111788
53	28.360	27.999		28.179	-6.812	0.00889941	53	21.400	21.588		21.494	-4.920	0.03303058
54	19.798	18.645		19.222	2.145	4.42432234	54	15.289	15.240		15.265	1.309	2.47852704
55	28.340	28.774		28.557	-7.190	0.00684793	55	23.008	23.380		23.194	-6.620	0.01016812
56	25.000	24.950		24.975	-3.608	0.08201082	56	23.520	23.687		23.604	-7.030	0.00765428
57	24.901	24.992		24.947	-3.580	0.08363366	57	21.937	21.717		21.827	-5.253	0.02622574
58	25.032	25.216		25.124	-3.757	0.07396105	58	22.800	22.757		22.779	-6.205	0.01355991
var2csa	21.898	22.267		22.082	-0.715	0.60911182	var2csa	14.993	14.961		14.977	1.597	3.02517164
91	23.489	23.322		23.406	-2.039	0.2433805	91	21.007	21.133		21.070	-4.496	0.0443199
92	24.491	23.905		24.198	-2.831	0.14052152	92	20.257	20.222		20.239	-3.665	0.07881218
93	24.255	24.394		24.325	-2.958	0.12872637	93	21.874	22.104		21.989	-5.415	0.02343839
94	29.287	27.694		28.491	-7.124	0.00717083	94	25.000	24.926		24.963	-8.389	0.00298331
95	25.763	25.437		25.600	-4.233	0.05317073	95	21.899	21.914		21.907	-5.333	0.02481452
96	27.355	27.623		27.489	-6.122	0.01435913	96	24.127	24.006		24.067	-7.493	0.00555277
97	28.724	28.207		28.466	-7.099	0.00729573	97	23.574	23.615		23.595	-7.021	0.00770123
60	21.561	21.172		21.367	0.000	1.00027556	60	16.578	16.570		16.574	0.000	1.00007764

(Rep 4,2) MCM + 100uM Spd 3Wks						
Primer pair	CT			CT Average	relative to p60	relative copy number
1	21.653	21.450		21.551	-5.240	0.02645431
3	22.861	22.854		22.858	-6.547	0.01069686
4	23.253	23.262		23.257	-6.946	0.00810893
5	22.663	22.689		22.676	-6.365	0.01213192
6	22.725	22.843		22.784	-6.473	0.01125489
7	21.765	21.807		21.786	-5.475	0.02248907
8	22.380	22.557		22.469	-6.158	0.01400882
9	21.497	21.476		21.487	-5.176	0.02766756
11	23.373	23.271		23.322	-7.011	0.00775443
12	21.266	21.230		21.248	-4.937	0.03264257
13	19.642	19.588		19.615	-3.304	0.10124703
15	21.117	20.803		20.960	-4.649	0.0398683
17	20.562	20.596		20.579	-4.268	0.05191253
18	20.709	20.629		20.669	-4.358	0.04876497
19	21.002	20.935		20.969	-4.658	0.03961958
20	24.591	24.469		24.530	-8.219	0.00335642
21	21.562	21.735		21.648	-5.337	0.02473204
22	21.388	21.365		21.377	-5.066	0.02985791
23	22.840	22.845		22.842	-6.531	0.01081005
25	20.223	20.432		20.327	-4.016	0.06179301
26	21.180	20.879		21.029	-4.718	0.03798376
27	19.304	19.190		19.247	-2.936	0.1306807
28	22.356	22.373		22.365	-6.054	0.01505217
30	22.849	23.190		23.020	-6.709	0.00956158
31	17.846	17.891		17.869	-1.558	0.3397393
34	19.895	20.145		20.020	-3.709	0.07647052
35	22.806	22.854		22.830	-6.519	0.01090273
36	22.764	22.981		22.872	-6.561	0.01058754
37	21.832	21.476		21.654	-5.343	0.02463504
38	22.225	22.007		22.116	-5.805	0.01789017
39	21.372	21.354		21.363	-5.052	0.03013477
40	33.920	36.303		35.111	-18.800	2.1902E-06
41	24.964	25.180		25.072	-8.761	0.00230502
43	22.222	22.426		22.324	-6.013	0.01548343
44	23.464	23.399		23.431	-7.120	0.00718808
45	19.920	19.960		19.940	-3.629	0.08084865
46	21.231	21.239		21.235	-4.924	0.0329458
47	26.596	26.220		26.408	-10.097	0.00091304
49	19.759	19.666		19.712	-3.401	0.09464854
50	27.228	26.886		27.057	-10.746	0.00058213
51	21.872	22.215		22.043	-5.732	0.01880838
52	22.691	22.571		22.631	-6.320	0.01251409
53	21.178	21.203		21.191	-4.880	0.03397218
54	15.007	14.744		14.876	1.435	2.70469985
55	22.816	23.121		22.969	-6.658	0.00990385
56	23.221	23.373		23.297	-6.986	0.00788899
57	21.627	21.663		21.645	-5.334	0.02479242
58	22.557	22.462		22.509	-6.198	0.01361677
var2csa	14.420	14.475		14.448	1.863	3.63863541
91	20.856	20.808		20.832	-4.521	0.04355181
92	20.005	19.992		19.999	-3.688	0.07760191
93	21.669	21.429		21.549	-5.238	0.02649097
94	24.648	24.985		24.817	-8.506	0.00275123
95	21.656	21.648		21.652	-5.341	0.02467483
96	23.921	23.960		23.941	-7.630	0.00504973
97	23.451	23.586		23.518	-7.207	0.00676601
60	16.306	16.316		16.311	0.000	1.00001525

Appendix B

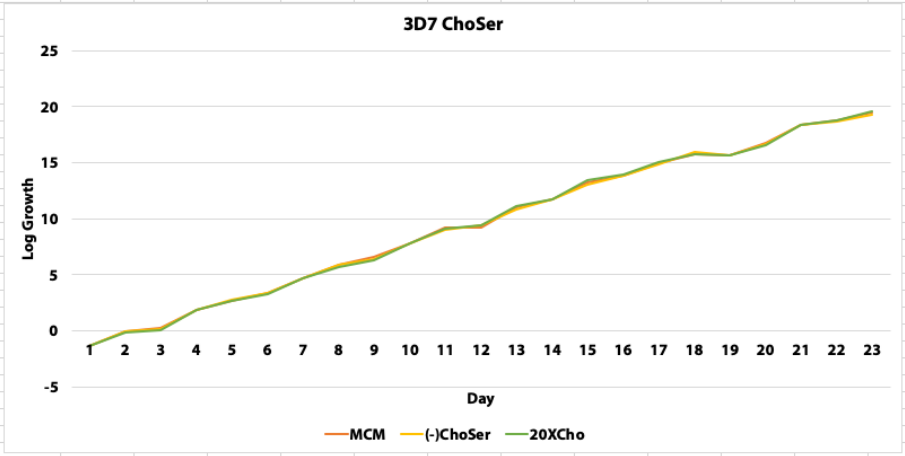
Growth assay and SAMS/SAHH qPCR data

Growth assays are shown first, organized by corresponding chapter and section. Raw numbers are log growth by day. PfSAMS/PfSAHH qPCR data presented last by corresponding chapter and section.

Chapter 2.5 Choline, Methionine, Serine growth assays

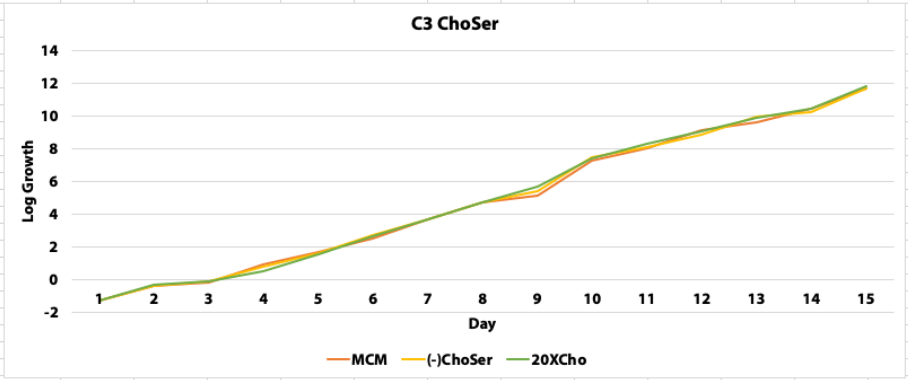
Rep 1

	MCM	(-)ChoSer	20XCho
1	-1.30103	-1.30103	-1.30103
2	-0.0555173	-0.0809219	-0.1135093
3	0.23044892	0.14612804	0.07918125
4	1.87506126	1.90308999	1.91381385
5	2.65321251	2.78532984	2.67209786
6	3.43136376	3.38021124	3.30103
7	4.69897	4.69897	4.69897
8	5.8573325	5.86923172	5.70757018
9	6.5563025	6.38021124	6.34242268
10	7.83884909	7.77815125	7.84509804
11	9.26951294	8.97312785	9.08990511
12	9.25527251	9.39794001	9.462398
13	11.045323	10.8260748	11.146128
14	11.69897	11.69897	11.69897
15	13.2227165	13.045323	13.4065402
16	13.7993405	13.8864907	13.9912261
17	14.9344985	14.8512583	15.075547
18	15.7558749	15.9190781	15.770852
19	15.7075702	15.6812412	15.6627578
20	16.7993405	16.6901961	16.5682017
21	18.3710679	18.4099331	18.4031205
22	18.7558749	18.6812412	18.7781513
23	19.5185139	19.30103	19.5682017



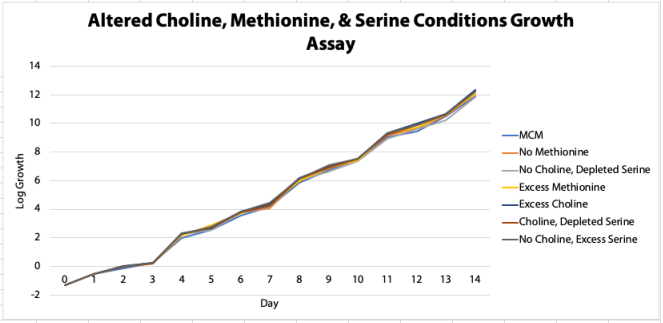
Rep 2

	MCM	(-)ChoSer	20XCho
1	-1.30103	-1.30103	-1.30103
2	-0.4202164	-0.4202164	-0.3098039
3	-0.1674911	-0.1249387	-0.1023729
4	0.93951925	0.80617997	0.47712125
5	1.64345268	1.62324929	1.53147892
6	2.53147892	2.69019608	2.61278386
7	3.69897	3.69897	3.69897
8	4.69897	4.69897	4.69897
9	5.11394335	5.38021124	5.65321251
10	7.23299611	7.43775056	7.37474835
11	7.98227123	8.06445799	8.30103
12	9.09342169	8.84509804	9.03342376
13	9.62324929	9.97312785	9.90848502
14	10.39794	10.2304489	10.4149733
15	11.6434527	11.6434527	11.8388491



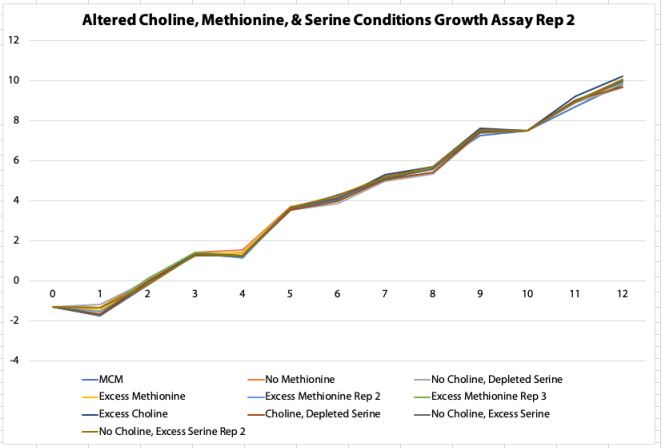
Rep 1

Day	MCM	No Methionine	No Choline, Depleted Serine	Excess Methionine	Excess Methionine Rep 2	Excess Methionine Rep 3	Excess Choline	Choline, Depleted Serine	No Choline, Excess Serine	No Choline, Excess Serine Rep 2
0	-1.30103	-1.30103	-1.30103	-1.30103	-1.30103	-1.30103	-1.30103	-1.30103	-1.30103	-1.30103
1	-0.522879	-0.522879	-0.522879	-0.522879	-0.522879	-0.522879	-0.522879	-0.522879	-0.522879	-0.522879
2	-0.113509	-0.036212	-0.040959	0.021189	-0.004365	-0.008774	0	-0.022276	-0.026872	-0.031517
3	0.278754	0.230449	0.255273	0.176091	0.176091	0.255273	0.255273	0.176091	0.255273	0.342423
4	1.963788	2.198657	2.187521	2.167317	2.243038	2.255273	2.298853	2.262451	2.245513	2.240549
5	2.568202	2.724276	2.568202	2.892095	2.755875	2.732394	2.653213	2.740363	2.778151	2.78533
6	3.556303	3.792392	3.69897	3.70757	3.69897	3.724276	3.845098	3.78533	3.819544	3.716003
7	4.146128	4.041393	4.146128	4.380211	4.361728	4.30103	4.342423	4.230449	4.431364	4.462398
8	5.863323	6.093422	6.017033	5.986772	6.079181	6.079181	6.206826	6.136721	6.120574	6.123852
9	6.748188	6.863323	6.653213	6.924279	6.963788	6.954243	6.954243	6.908485	7.071882	7.089905
10	7.462398	7.447158	7.361728	7.39794	7.380211	7.414973	7.556303	7.531479	7.556303	7.414973
11	9.029384	9.149219	8.944483	9.240549	9.257679	9.227887	9.30103	9.187521	9.320146	9.383815
12	9.447158	9.681241	9.623249	9.690196	9.748188	9.623249	10.0086	9.863323	9.880814	9.778151
13	10.47712	10.5563	10.20412	10.61278	10.62325	10.63347	10.6902	10.63347	10.6902	10.6902
14	11.87506	12.04922	11.85733	12.04532	12.1271	11.99564	12.36736	12.23553	12.20683	12.19866



Rep 2

Day	MCM	No Methionine	No Choline, Depleted Serine	Excess Methionine	Excess Methionine Rep 2	Excess Methionine Rep 3	Excess Choline	Choline, Depleted Serine	No Choline, Excess Serine	No Choline, Excess Serine Rep 2
0	-1.30103	-1.30103	-1.30103	-1.30103	-1.30103	-1.30103	-1.30103	-1.30103	-1.30103	-1.30103
1	-1.337242	-1.346787	-1.187087	-1.468521	-1.522879	-1.677781	-1.744727	-1.638272	-1.744727	-1.346787
2	-0.004365	0.056905	-0.013228	-0.107905	-0.19382	0.08636	-0.060481	-0.055517	-0.236572	-0.207608
3	1.298853	1.432969	1.298853	1.354108	1.404834	1.40654	1.350248	1.25042	1.311754	1.354108
4	1.20412	1.531479	1.431364	1.361728	1.146128	1.278754	1.278754	1.278754	1.230449	1.278754
5	3.542825	3.689309	3.552668	3.609594	3.630428	3.669317	3.666518	3.549003	3.597695	3.649335
6	4.313867	4.173186	3.838849	4.012837	4.017033	4.120574	4.079181	3.977724	4.146128	4.267172
7	5.025306	5.235528	4.995635	5.158362	5.187521	5.167317	5.283301	5.060698	5.093422	5.164353
8	5.690196	5.60206	5.342423	5.60206	5.623249	5.591065	5.690196	5.431364	5.591065	5.690196
9	7.257679	7.485721	7.359835	7.522444	7.523746	7.487138	7.600973	7.432969	7.459392	7.542825
10	7.477121	7.477121	7.477121	7.477121	7.477121	7.477121	7.477121	7.477121	7.477121	7.477121
11	8.681241	8.991226	8.892095	8.995635	9.017033	9.012837	9.209515	8.995635	8.934498	8.913814
12	9.80618	10.06819	9.653213	9.845098	9.908485	9.977724	10.19033	9.690196	9.968483	10.06446

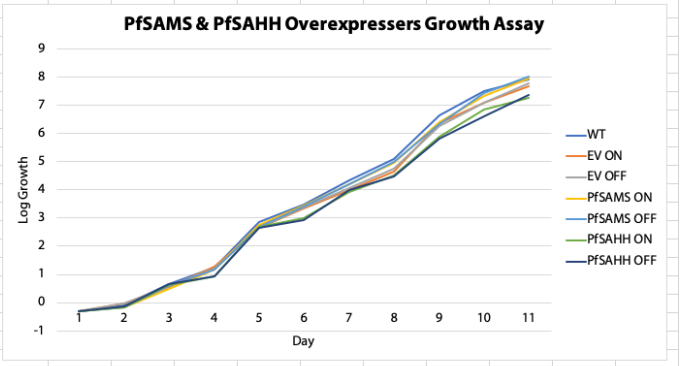


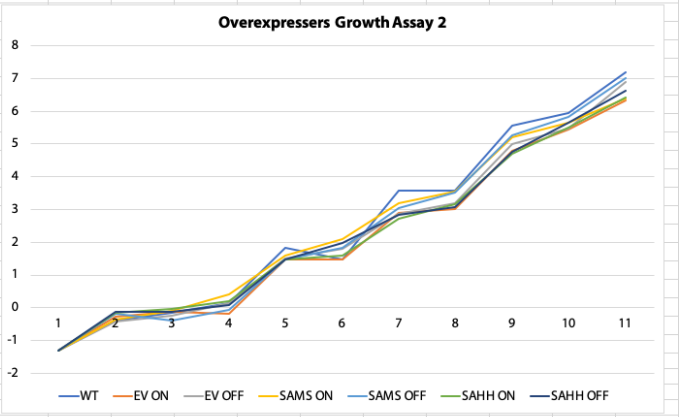
Chapter 3.4 OE growth assays

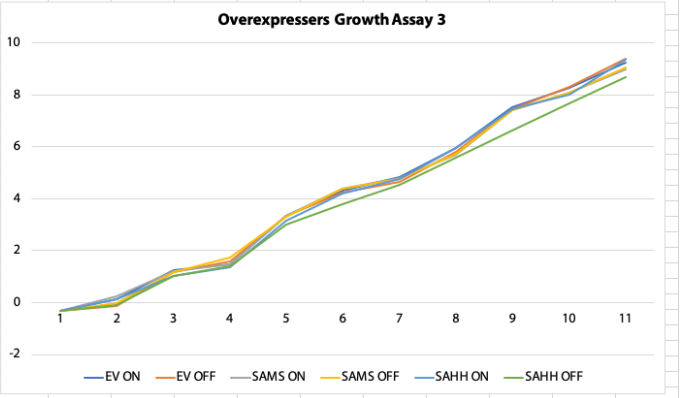
Rep 1							
	WT	EV ON	EV OFF	PfSAMS ON	PfSAMS OFF	PfSAHH ON	PfSAHH OFF
1	-0.30103	-0.30103	-0.30103	-0.30103	-0.30103	-0.30103	-0.30103
2	-0.091515	-0.036212	-0.022276	-0.1804561	-0.16749109	-0.1739252	-0.1366771
3	0.660865	0.556303	0.583199	0.5010593	0.600972896	0.61489722	0.6599162
4	1.230449	1.274158	1.220108	1.1903317	1.158362492	0.94448267	0.94448267
5	2.859739	2.657056	2.657056	2.7686381	2.681241237	2.69548168	2.66181269
6	3.463893	3.33646	3.390935	3.4265113	3.414973348	2.99122608	2.94448267
7	4.326336	3.982271	4.075547	4.2148438	4.214843848	3.91381385	3.98677173
8	5.079181	4.662758	4.755875	4.9395193	4.986771734	4.50514998	4.462398
9	6.632457	6.371068	6.260071	6.3838154	6.322219295	5.88649073	5.83250891
10	7.488551	7.071882	7.093422	7.3404441	7.445604203	6.83250891	6.60205999
11	7.90309	7.681241	7.792392	7.9395193	8.004321374	7.25527251	7.36172784

Rep 2							
	WT	EV ON	EV OFF	SAMS ON	SAMS OFF	SAHH ON	SAHH OFF
1	-1.30103	-1.30103	-1.30103	-1.30103	-1.30103	-1.30103	-1.30103
2	-0.376751	-0.283997	-0.408935	-0.3665315	-0.16749109	-0.14874165	-0.1366771
3	-0.161151	-0.124939	-0.236572	-0.1023729	-0.38721614	-0.03621217	-0.1249387
4	0.146128	-0.167491	0.152288	0.4232459	-0.07058107	0.1903317	0.07188201
5	1.83187	1.475671	1.50515	1.580925	1.460897843	1.48000694	1.48855072
6	1.477121	1.462398	1.799341	2.1003705	1.832508913	1.60205999	1.96848295
7	3.584331	2.897627	2.880814	3.1789769	3.049218023	2.71600334	2.84509804
8	3.579784	3	3.20412	3.544068	3.51851394	3.17609126	3.07918125
9	5.546543	4.78533	5.004321	5.2121876	5.257678575	4.70757018	4.75587486
10	5.94939	5.447158	5.477121	5.6532125	5.838849091	5.50514998	5.64345268
11	7.176091	6.322219	6.880814	6.39794	7.012837225	6.43136376	6.63346846

Rep 3						
	EV ON	EV OFF	SAMS ON	SAMS OFF	SAHH ON	SAHH OFF
1	-0.30103	-0.30103	-0.30103	-0.30103	-0.30103	-0.30103
2	0.113943	-0.130768	0.25042	-0.0222764	0.123851641	-0.10237291
3	1.240549	1.173186	1.225309	1.1903317	1.041392685	1.03342376
4	1.477121	1.568202	1.477121	1.7323938	1.361727836	1.41497335
5	3.356026	3.334454	3.334454	3.3117539	3.139879086	3.01703334
6	4.311754	4.220108	4.403121	4.4014005	4.190331698	3.77815125
7	4.826075	4.653213	4.792392	4.748188	4.748188027	4.54406804
8	5.968483	5.812913	5.70757	5.7160033	5.959041392	5.56820172
9	7.510545	7.434569	7.419956	7.4149733	7.432969291	6.63346846
10	8.255273	8.30963	8.071882	8.064458	8.017033339	7.66275783
11	9.245513	9.369216	8.963788	9.0374265	9.361727836	8.69019608



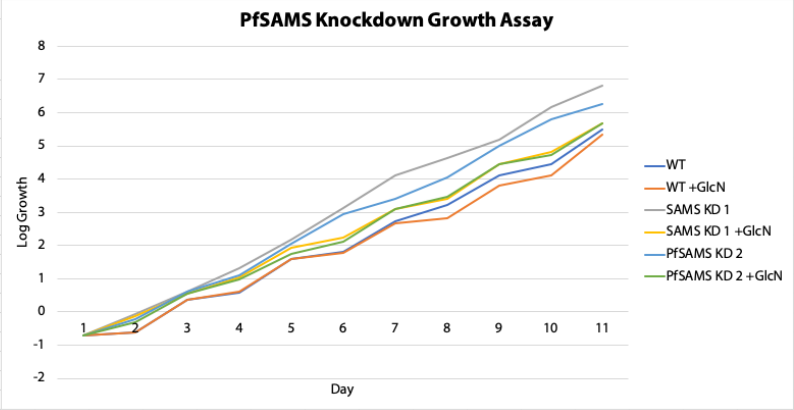




Chapter 3.4 KD growth assays

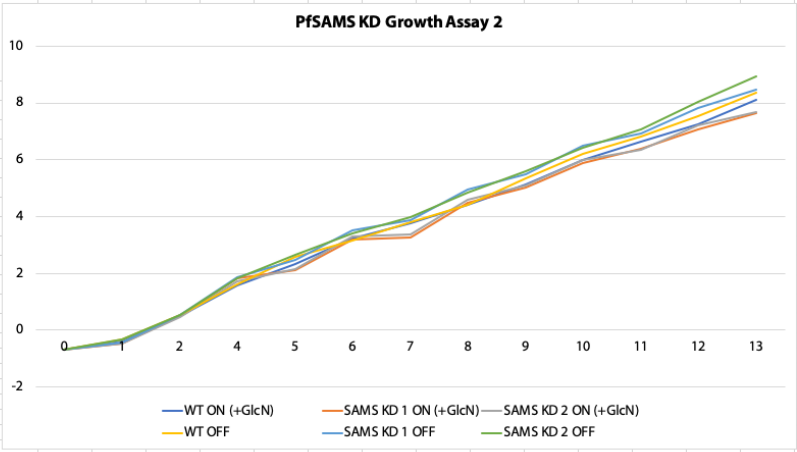
Rep 1

	WT	WT +GlcN	SAMS KD 1	SAMS KD 1 +GlcN	PfSAMS KD 2	PfSAMS KD 2 +GlcN
1	-0.699	-0.69897	-0.699	-0.699	-0.699	-0.699
2	-0.6198	-0.61979	-0.0757	-0.1135	-0.2147	-0.3098
3	0.3711	0.38021	0.618	0.5587	0.6021	0.5563
4	0.5798	0.61278	1.3096	1.0294	1.1139	0.9685
5	1.5999	1.59879	2.1644	1.9191	2.0607	1.7597
6	1.8129	1.77085	3.1239	2.2304	2.9395	2.1239
7	2.7372	2.66464	4.0969	3.0899	3.3892	3.0864
8	3.2304	2.82607	4.6335	3.3979	4.0374	3.4624
9	4.1239	3.79449	5.1903	4.4456	5.0043	4.4409
10	4.4472	4.12385	6.1553	4.8062	5.8062	4.716
11	5.5038	5.35025	6.8062	5.6702	6.248	5.6609
12	5.5682	5.30103	7.4829	6.0755	7.2856	6.0212
13	6.6981	6.45179	8.1761	6.2095	7.8751	6.1106
14	7.2405	6.75587	9.0086	6.5185	8.7952	6.6812
15	8.2068	7.75587	9.7634	7.4116	9.3522	7.316



Rep 2

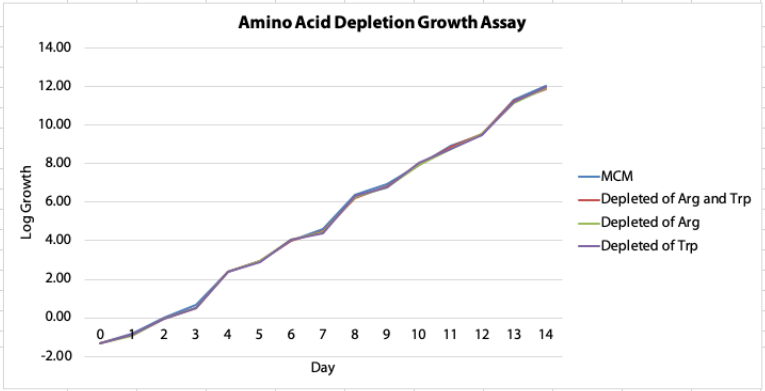
	WT ON (+GlcN)	SAMS KD 1 ON (+GlcN)	SAMS KD 2 ON (+GlcN)	WT OFF	SAMS KD 1 OFF	SAMS KD 2 OFF
0	-0.699	-0.69897	-0.699	-0.699	-0.699	-0.699
1	-0.3565	-0.48149	-0.4559	-0.3098	-0.4318	-0.3279
2	0.5051	0.50106	0.4713	0.5428	0.549	0.5478
4	1.5798	1.82478	1.7275	1.6232	1.8561	1.8189
5	2.3424	2.1038	2.1614	2.5855	2.4624	2.6561
6	3.2405	3.19866	3.2967	3.1492	3.5092	3.4065
7	3.7559	3.25527	3.3802	3.8129	3.8633	3.9777
8	4.4183	4.46687	4.5832	4.4133	4.9355	4.8525
9	5.1399	5.0086	5.0899	5.3541	5.5065	5.6053
10	6	5.88649	6.0086	6.2175	6.4955	6.4249
11	6.6532	6.39794	6.3617	6.8195	6.9395	7.0792
12	7.233	7.06819	7.2122	7.5185	7.8299	8.0453
13	8.1206	7.65321	7.6721	8.3636	8.4669	8.9494



Chapter 4.3 Amino acid starvation growth assays

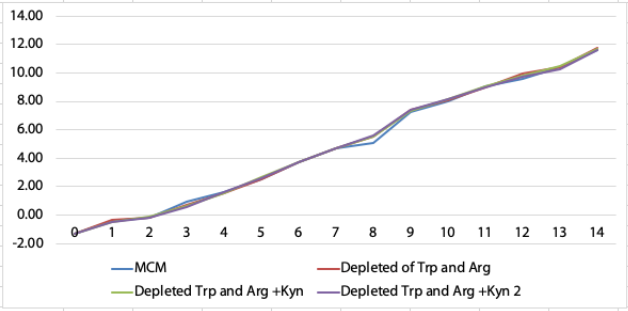
Rep 1

	MCM	Depleted of Arg and Trp	Depleted of Arg	Depleted of Trp
0	-1.30	-1.30	-1.30	-1.30
1	-0.82	-0.85	-0.92	-0.85
2	-0.02	-0.03	-0.04	-0.04
3	0.67	0.49	0.52	0.52
4	2.36	2.38	2.38	2.37
5	2.93	2.95	2.92	2.86
6	3.97	3.99	4.05	4.03
7	4.58	4.51	4.46	4.40
8	6.36	6.23	6.28	6.31
9	6.92	6.83	6.79	6.76
10	7.91	7.95	7.95	8.03
11	8.92	8.88	8.78	8.79
12	9.48	9.53	9.52	9.49
13	11.30	11.23	11.16	11.17
14	12.01	11.87	11.92	11.99



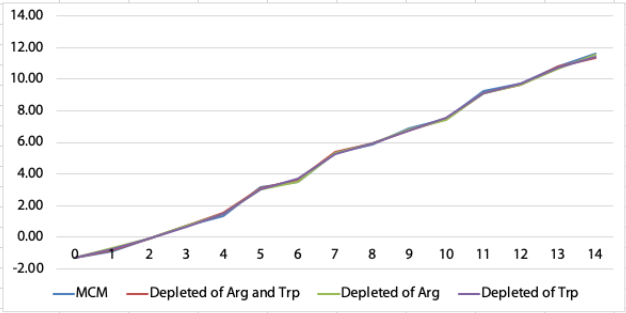
Rep 2

	MCM	Depleted of Trp and Arg	Depleted Trp and Arg +Kyn	Depleted Trp and Arg +Kyn 2
0	-1.30	-1.30	-1.30	-1.30
1	-0.42	-0.36	-0.48	-0.49
2	-0.17	-0.21	-0.13	-0.15
3	0.94	0.75	0.63	0.57
4	1.64	1.54	1.57	1.59
5	2.53	2.56	2.65	2.60
6	3.70	3.70	3.70	3.70
7	4.70	4.70	4.70	4.70
8	5.11	5.52	5.53	5.57
9	7.23	7.37	7.36	7.40
10	7.98	8.07	8.18	8.17
11	9.09	8.98	9.04	8.98
12	9.62	9.99	9.81	9.76
13	10.40	10.43	10.51	10.23
14	11.64	11.74	11.71	11.61



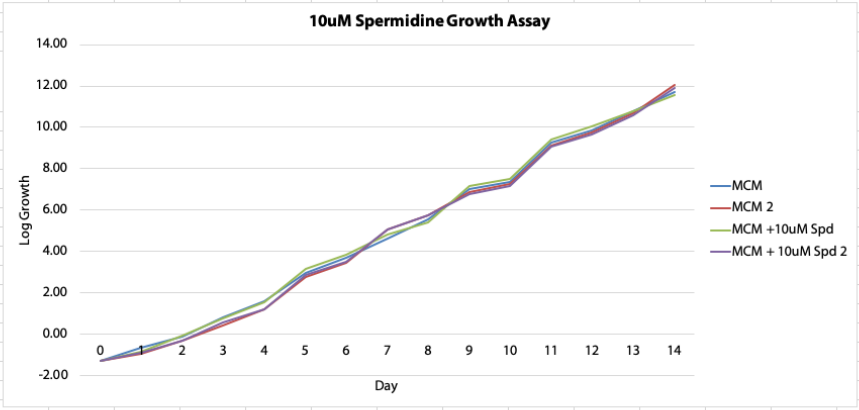
Rep 3

	MCM	Depleted of Arg and Trp	Depleted of Arg	Depleted of Trp
0	-1.30	-1.30	-1.30	-1.30
1	-0.89	-0.85	-0.72	-0.74
2	-0.12	-0.10	-0.06	-0.11
3	0.70	0.69	0.71	0.62
4	1.34	1.51	1.45	1.49
5	3.16	3.06	3.03	3.05
6	3.53	3.61	3.51	3.67
7	5.35	5.37	5.30	5.27
8	5.85	5.92	5.91	5.94
9	6.91	6.74	6.79	6.72
10	7.51	7.49	7.41	7.53
11	9.24	9.12	9.13	9.09
12	9.74	9.67	9.63	9.74
13	10.81	10.83	10.71	10.72
14	11.60	11.36	11.54	11.45

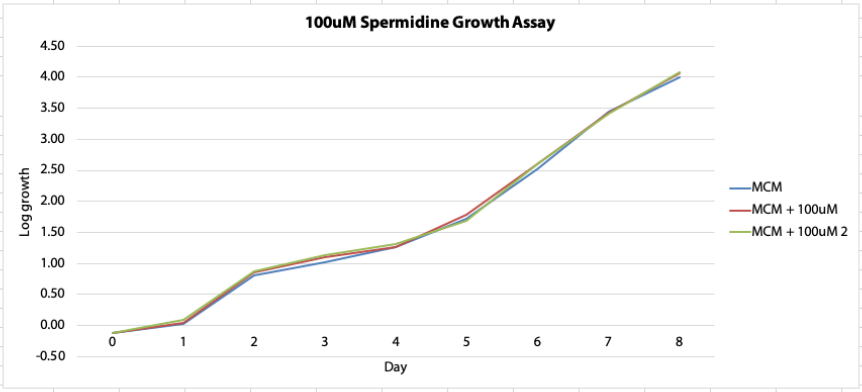


Chapter 4.6 Polyamine growth assays

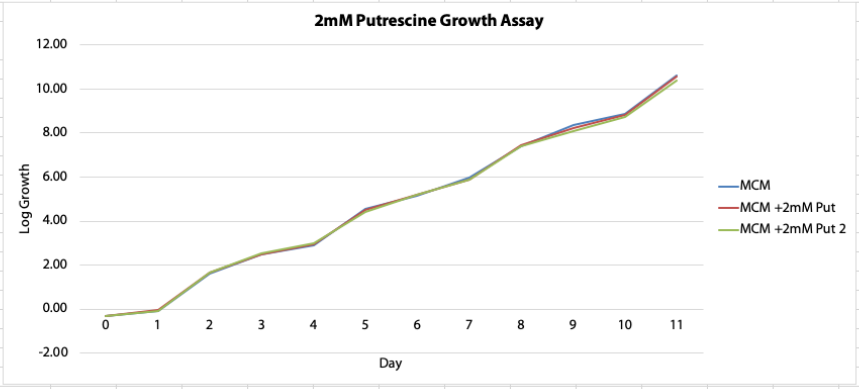
	MCM	MCM 2	MCM +10uM Spd	MCM + 10uM Spd 2
0	-1.30103	-1.30103	-1.30103	-1.30103
1	-0.638272	-0.958607	-0.853872	-0.886057
2	-0.086186	-0.283997	-0.05061	-0.309804
3	0.80618	0.447158	0.778151	0.579784
4	1.60206	1.20412	1.556303	1.20412
5	2.959041	2.778151	3.173186	2.863323
6	3.69897	3.462398	3.832509	3.518514
7	4.612784	5.08636	4.819544	5.075547
8	5.556303	5.740363	5.414973	5.763428
9	7.0086	6.851258	7.176091	6.799341
10	7.342423	7.255273	7.518514	7.176091
11	9.245513	9.11059	9.40654	9.075547
12	9.845098	9.732394	10.02531	9.672098
13	10.77085	10.6721	10.77085	10.57978
14	11.70757	12.03743	11.57978	11.92428



	MCM	MCM + 100uM	MCM + 100uM 2
0	-0.124939	-0.124939	-0.124939
1	0.033424	0.037426	0.08636
2	0.800717	0.848805	0.865696
3	1.025306	1.09482	1.138618
4	1.260071	1.258877	1.318063
5	1.718086	1.792392	1.683497
6	2.51021	2.604766	2.591065
7	3.441302	3.42529	3.416016
8	3.997277	4.057143	4.087027



	MCM	MCM +2mM Put	MCM +2mM Put
0	-0.30103	-0.30103	-0.30103
1	-0.102373	-0.05061	-0.080922
2	1.62941	1.658965	1.664642
3	2.481443	2.49276	2.511883
4	2.908485	2.963788	2.968483
5	4.537819	4.49276	4.403121
6	5.149219	5.201397	5.209515
7	5.986772	5.886491	5.908485
8	7.40824	7.444045	7.401401
9	8.38739	8.232996	8.103804
10	8.892095	8.80618	8.755875
11	10.60959	10.55388	10.39794



Chapter 3.1 PfSAHH qPCR data

Line	Sample# 1	2	3	4	5	6	Average	Standard Dev
Untransfected WT	0.4404559	0.48251	0.40773	0.44356			0.44356295	0.030608522
EV Control	0.10618067	0.43255	0.42982	0.32285			0.3228478	0.153210859
PfSAHH OFF aTc	1.5130081	1.02991	1.095	2.44098	1.5068	1.4551	1.50680088	0.504462271
PfSAHH ON aTc	2.63243024	2.36993	1.93244	1.49462	1.96336	1.38736	1.96335593	0.482784218

*Average SAHH = average relative copy number (to p60)

Chapter 3.7 PfSAMS qPCR data

Line	Average SAMS	Standard Dev
WT	0.94780841	0.006033747
PfSAMS ON aTc	2.413025087	0.342238399
PfSAMS OFF aTc	1.319860421	0.075058353
Clone B7 (var2csa)	1.041648261	0.026780853
Clone B11 (var2csa)	1.635632914	0.441708137
Clone H9 (het.)	0.465645773	0.199587517
Clone C7 (het.)	0.380951436	0.276420974

*Average SAMS = average relative copy number (to p60), n=4 well readings per sample

References

1. Organization, W. H. World malaria report 2019. (2019).

2. Deitsch, K. W. & Dzikowski, R. Variant Gene Expression and Antigenic Variation by Malaria Parasites. *Annual Review of Microbiology* **71**, 625–641 (2017).

3. Cowman, A. F., Healer, J., Marapana, D. & Marsh, K. Malaria: Biology and Disease. *Cell* **167**, 610–624 (2016).

4. Alonso, P. L., Noor, A., Santelli, A. C., Ghani, A. & Quadros, C. de. Global technical strategy for malaria 2016–2030. (2015).

5. Gazzinelli, R. T., Kalantari, P., Fitzgerald, K. A. & Golenbock, D. T. Innate sensing of malaria parasites. *Nature Reviews Immunology* **14**, 744–757 (2014).

6. Miller, L. H., Baruch, D. I., Marsh, K. & Doumbo, O. K. The pathogenic basis of malaria. *Nature* **415**, 673–679 (2002).

7. Phillips, M. A. *et al.* Malaria. *Nature Publishing Group* **3**, 1–25 (2017).

8. Boddey, J. A. & Cowman, A. F. Plasmodium Nesting: Remaking the Erythrocyte from the Inside Out. *Annual Review of Microbiology* **67**, 243–269 (2013).

9. Turner, L. *et al.* Severe malaria is associated with parasite binding to endothelial protein C receptor. *Nature* **498**, 1–6 (2013).

10. Miller, L., Good, M. & Milon, G. Malaria pathogenesis. *Science* **264**, 1878–1883 (1994).

11. Gardner, M. J. *et al.* Genome sequence of the human malaria parasite Plasmodium falciparum. *Nature* **419**, 498–511 (2002).

12. Baruch, D. I. *et al.* Cloning the P. falciparum gene encoding PfEMP1, a malarial variant antigen and adherence receptor on the surface of parasitized human erythrocytes. *cell.com* (n.d.).

13. Smith, J. D., Chitnis, C. E., Craig, A. G., Cell, D. R. & 1995. Switches in expression of Plasmodium falciparum var genes correlate with changes in antigenic and cytoadherent phenotypes of infected erythrocytes. *ncbi.nlm.nih.gov* (n.d.).

14. Su, X. Z. *et al.* The large diverse gene family var encodes proteins involved in cytoadherence and antigenic variation of Plasmodium falciparum-infected erythrocytes. *Cell* **82**, 89–100 (1995).

15. Otto, T. D. *et al.* Evolutionary analysis of the most polymorphic gene family in falciparum malaria. *Wellcome Open Research* **4**, 193–29 (2019).

16. Scherf, A. *et al.* Antigenic variation in malaria: in situ switching, relaxed and mutually exclusive transcription of var genes during intra-erythrocytic development in Plasmodium falciparum. *The EMBO journal* **17**, 5418–5426 (1998).

17. Deitsch, K. W., Pinal, A. del & Wellems, T. E. Intra-cluster recombination and var transcription switches in the antigenic variation of Plasmodium falciparum. *Molecular and biochemical parasitology* **101**, 107–116 (1999).

18. Epp, C., Li, F., Howitt, C. A., Chookajorn, T. & Deitsch, K. W. Chromatin associated sense and antisense noncoding RNAs are transcribed from the var gene family of virulence genes of the malaria parasite Plasmodium falciparum. *RNA* **15**, 116–127 (2009).

19. Scherf, A., Lopez-Rubio, J. J. & Riviere, L. Antigenic Variation in Plasmodium falciparum. *Annual Review of Microbiology* **62**, 445–470 (2008).

20. Deitsch, K. W., Lukehart, S. A. & Stringer, J. R. Common strategies for antigenic variation by bacterial, fungal and protozoan pathogens. *Nature reviews. Microbiology* **7**, 493–503 (2009).

21. Deitsch, K. W. & Kirkman, L. A. Recombination and Diversification of the Variant Antigen Encoding Genes in the Malaria Parasite Plasmodium falciparum. *Microbiology Spectrum* **2**, 1–12 (2014).

22. Dzikowski, R., Frank, M. & Deitsch, K. Mutually Exclusive Expression of Virulence Genes by Malaria Parasites Is Regulated Independently of Antigen Production. *PLoS pathogens* **2**, e22-11 (2006).

23. Voss, T. S. *et al.* A var gene promoter controls allelic exclusion of virulence genes in Plasmodium falciparum malaria. *Nature* **439**, 1004–1008 (2005).

24. Horrocks, P., Pinches, R., Christodoulou, Z., Kyes, S. A. & Newbold, C. I. Variable var transition rates underlie antigenic variation in malaria. *Proceedings of the National Academy of Sciences* **101**, 11129–11134 (2004).

25. Bachmann, A. *et al.* Highly co-ordinated var gene expression and switching in clinical Plasmodium falciparum isolates from non-immune malaria patients. *Cellular microbiology* **13**, 1397–1409 (2011).

26. Wassmer, S. C. *et al.* Investigating the Pathogenesis of Severe Malaria: A Multidisciplinary and Cross-Geographical Approach. *The American Journal of Tropical Medicine and Hygiene* **93**, 42–56 (2015).

27. Cortés, A., Crowley, V. M., Vaquero, A. & Voss, T. S. A View on the Role of Epigenetics in the Biology of Malaria Parasites. *PLoS pathogens* **8**, e1002943-7 (2012).

28. Gao, X., Reid, M. A., Kong, M. & Locasale, J. W. Metabolic interactions with cancer epigenetics. *Molecular Aspects of Medicine* **54**, 1–9 (2016).

29. Flueck, C. *et al.* Plasmodium falciparum Heterochromatin Protein 1 Marks Genomic Loci Linked to Phenotypic Variation of Exported Virulence Factors. *PLoS pathogens* **5**, e1000569-16 (2009).

30. Perez-Toledo, K. *et al.* Plasmodium falciparum heterochromatin protein 1 binds to tri-methylated histone 3 lysine 9 and is linked to mutually exclusive expression of var genes. *Nucleic Acids Research* **37**, 2596–2606 (2009).

31. Cortés, A. & Deitsch, K. W. Malaria Epigenetics. *Cold Spring Harbor Perspectives in Medicine* a025528-24 (2017) doi:10.1101/cshperspect.a025528.

32. Gupta, A. P. *et al.* Dynamic Epigenetic Regulation of Gene Expression during the Life Cycle of Malaria Parasite Plasmodium falciparum. *PLoS pathogens* **9**, e1003170-15 (2013).

33. Lopez-Rubio, J. J., Mancio-Silva, L. & Scherf, A. Genome-wide analysis of heterochromatin associates clonally variant gene regulation with perinuclear repressive centers in malaria parasites. *Cell Host and Microbe* **5**, 179–190 (2009).

34. Salcedo-Amaya, A. M. *et al.* Dynamic histone H3 epigenome marking during the intraerythrocytic cycle of Plasmodium falciparum. *Proceedings of the National Academy of Sciences* **106**, 9655–9660 (2009).

35. Lopez-Rubio, J. J. *et al.* 5' flanking region of var genes nucleate histone modification patterns linked to phenotypic inheritance of virulence traits in malaria parasites. *Molecular Microbiology* **0**, 071119190133003-??? (2007).

36. Chookajorn, T. *et al.* Epigenetic memory at malaria virulence genes. *Proceedings of the National Academy of Sciences* **104**, 899–902 (2007).

37. Jiang, L. *et al.* PfSETvs methylation of histone H3K36 represses virulence genes in Plasmodium falciparum. *Nature* **499**, 223–227 (2013).

38. Volz, J. C. *et al.* PfSET10, a Plasmodium falciparum methyltransferase, maintains the active var gene in a poised state during parasite division. *Cell Host and Microbe* **11**, 7–18 (2012).

39. Cui, L., Fan, Q., Cui, L. & Miao, J. Histone lysine methyltransferases and demethylases in Plasmodium falciparum. *International journal for parasitology* **38**, 1083–1097 (2008).

40. Ukaegbu, U. E. *et al.* Recruitment of PfSET2 by RNA Polymerase II to Variant Antigen Encoding Loci Contributes to Antigenic Variation in P. falciparum. *PLoS pathogens* **10**, e1003854-14 (2014).

41. Brancucci, N. M. B. *et al.* Heterochromatin Protein 1 Secures Survival and Transmission of Malaria Parasites. *Cell Host and Microbe* **16**, 165–176 (2014).

42. Coleman, B. I. *et al.* A Plasmodium falciparum histone deacetylase regulates antigenic variation and gametocyte conversion. *Cell Host and Microbe* **16**, 177–186 (2014).

43. Ponts, N. *et al.* Genome-wide mapping of DNA methylation in the human malaria parasite Plasmodium falciparum. *Cell Host and Microbe* **14**, 696–706 (2013).

44. Kraemer, S. M. *et al.* Patterns of gene recombination shape var gene repertoires in Plasmodium falciparum: comparisons of geographically diverse isolates. *BMC Genomics* **8**, 214–18 (2007).

45. Trimnell, A. R. *et al.* Global genetic diversity and evolution of var genes associated with placental and severe childhood malaria☆. *Molecular and biochemical parasitology* **148**, 169–180 (2006).

46. Salanti, A. *et al.* Selective upregulation of a single distinctly structured var gene in chondroitin sulphate A-adhering Plasmodium falciparum involved in pregnancy-associated malaria. *Molecular microbiology* **49**, 179–191 (2003).

47. Salanti, A. *et al.* Evidence for the involvement of VAR2CSA in pregnancy-associated malaria. *The Journal of Experimental Medicine* **200**, 1197–1203 (2004).

48. Amulic, B., Salanti, A., Lavstsen, T., Nielsen, M. A. & Deitsch, K. W. An Upstream Open Reading Frame Controls Translation of var2csa, a Gene Implicated in Placental Malaria. *PLoS pathogens* **5**, e1000256-12 (2009).

49. Bancells, C. & Deitsch, K. W. A molecular switch in the efficiency of translation reinitiation controls expression of var2csa, a gene implicated in pregnancy-associated malaria. *Molecular microbiology* **90**, 472–488 (2013).

50. Mok, B. W. *et al.* Default Pathway of var2csa Switching and Translational Repression in Plasmodium falciparum. *PLoS ONE* **3**, e1982-11 (2008).

51. Ukaegbu, U. E. *et al.* A Unique Virulence Gene Occupies a Principal Position in Immune Evasion by the Malaria Parasite Plasmodium falciparum. *PLoS genetics* **11**, e1005234 (2015).

52. Bachmann, A. *et al.* Controlled human malaria infection with Plasmodium falciparum demonstrates impact of naturally acquired immunity on virulence gene expression. *PLoS pathogens* **15**, e1007906-24 (2019).

53. Mugnier, M. R., Cross, G. A. M. & Papavasiliou, F. N. The in vivo dynamics of antigenic variation in Trypanosoma brucei. *Science (New York, N.Y.)* **347**, 1470–1473 (2015).

54. Cross, G. A. M., Kim, H.-S. & Wickstead, B. Capturing the variant surface glycoprotein repertoire (the VSGnome) of Trypanosoma brucei Lister 427. *Molecular and biochemical parasitology* **195**, 59–73 (2014).

55. Recker, M. *et al.* Antigenic variation in Plasmodium falciparum malaria involves a highly structured switching pattern. *PLoS pathogens* **7**, e1001306 (2011).

56. Horrocks, P. *et al.* Variable var Transition Rates Underlie Antigenic Variation in Malaria. *Proceedings of the National Academy of Sciences of the United States of America* **101**, 11129–11134 (2004).

57. Mentch, S. J. & Locasale, J. W. One-carbon metabolism and epigenetics: understanding the specificity. *Annals of the New York Academy of Sciences* **1363**, 91–98 (2015).

58. Mentch, S. J. *et al.* Histone Methylation Dynamics and Gene Regulation Occur through the Sensing of One-Carbon Metabolism. *Cell metabolism* **22**, 861–873 (2015).

59. Reid, M. A., Dai, Z., biology, J. L. N. cell & 2017. The impact of cellular metabolism on chromatin dynamics and epigenetics. *nature.com* **19**, 1298–1306 (2017).

60. Deguchi, T. & Barchas, J. Inhibition of transmethylations of biogenic amines by S-adenosylhomocysteine. Enhancement of transmethylation by adenosylhomocysteinase. *Journal of Biological Chemistry* **246**, 3175–3181 (1971).

61. Hoffman, D. R., Cornatzer, W. E. & Duerre, J. A. Relationship between tissue levels of S-adenosylmethionine, S-adenylhomocysteine, and transmethylation reactions. *Canadian journal of biochemistry* **57**, 56–65 (1979).

62. Locasale, J. W. & Cantley, L. C. Metabolic Flux and the Regulation of Mammalian Cell Growth. *Cell metabolism* **14**, 443–451 (2011).

63. Shiraki, N. *et al.* Methionine Metabolism Regulates Maintenance and Differentiation of Human Pluripotent Stem Cells. *Cell metabolism* **19**, 780–794 (2014).

64. Sadhu, M. J. *et al.* Nutritional control of epigenetic processes in yeast and human cells. *Genetics* **195**, 831–844 (2013).

65. Shyh-Chang, N. *et al.* Influence of threonine metabolism on S-adenosylmethionine and histone methylation. *Science (New York, N.Y.)* **339**, 222–226 (2013).

66. Ulanovskaya, O. A., Zuhl, A. M. & Cravatt, B. F. NNMT promotes epigenetic remodeling in cancer by creating a metabolic methylation sink. *Nature chemical biology* **9**, 300–306 (2013).

67. Ye, C., Sutter, B. M., Wang, Y., Kuang, Z. & Tu, B. P. A Metabolic Function for Phospholipid and Histone Methylation. *Molecular Cell* **66**, 1–23 (2017).

68. Bozdech, Z. *et al.* The Transcriptome of the Intraerythrocytic Developmental Cycle of Plasmodium falciparum. *PLOS Biology* **1**, e5-16 (2003).

69. Ruiz, J. L. & Gómez-Díaz, E. The second life of Plasmodium in the mosquito host: gene regulation on the move. *Briefings in functional genomics* (2019) doi:10.1093/bfgp/elz007.

70. Rovira-Graells, N. *et al.* Transcriptional variation in the malaria parasite Plasmodium falciparum. *Genome Research* **22**, 925–938 (2012).

71. Llorà-Batlle, O., Tintó-Font, E. & Cortés, A. Transcriptional variation in malaria parasites: why and how. *Briefings in functional genomics* **18**, 329–341 (2019).

72. Mancio-Silva, L. *et al.* Nutrient sensing modulates malaria parasite virulence. *Nature Publishing Group* **547**, 213–216 (2017).

73. Brancucci, N. M. B. *et al.* Lysophosphatidylcholine Regulates Sexual Stage Differentiation in the Human Malaria Parasite Plasmodium falciparum. *Cell* 1–29 (2017) doi:10.1016/j.cell.2017.10.020.

74. Kafsack, B. F. C. *et al.* A transcriptional switch underlies commitment to sexual development in malaria parasites. *Nature* **507**, 248–252 (2014).

75. Beri, D. *et al.* A disrupted transsulphuration pathway results in accumulation of redox metabolites and induction of gametocytogenesis in malaria. *Nature Publishing Group* **7**, 1–13 (2017).

76. Kilian, N., Choi, J.-Y., Voelker, D. R. & Mamoun, C. B. Role of phospholipid synthesis in the development and differentiation of malaria parasites in the blood. *Journal of Biological Chemistry* **293**, 17308–17316 (2018).

77. Pessi, G., Kociubinski, G., Mamoun, C. B. & Borst, P. A Pathway for Phosphatidylcholine Biosynthesis in Plasmodium falciparum Involving Phosphoethanolamine Methylation. *Proceedings of the National Academy of Sciences of the United States of America* **101**, 6206–6211 (2004).

78. Mamoun, C. B., Prigge, S. T. & Vial, H. Targeting the Lipid Metabolic Pathways for the Treatment of Malaria. *Drug development research* **71**, 44–55 (2010).

79. Witola, W. H. *et al.* Disruption of the Plasmodium falciparum PfPMT gene results in a complete loss of phosphatidylcholine biosynthesis via the serine-decarboxylase-phosphoethanolamine-methyltransferase pathway and severe growth and survival defects. *Journal of Biological Chemistry* **283**, 27636–27643 (2008).

80. Bobenchik, A. M. *et al.* Plasmodium falciparum phosphoethanolamine methyltransferase is essential for malaria transmission. *Proceedings of the National Academy of Sciences of the United States of America* **110**, 18262–18267 (2013).

81. Gulati, S. *et al.* Profiling the Essential Nature of Lipid Metabolism in Asexual Blood and Gametocyte Stages of Plasmodium falciparum. *Cell Host and Microbe* **18**, 371–381 (2015).

82. Witola, W. H. & Mamoun, C. B. Choline Induces Transcriptional Repression and Proteasomal Degradation of the Malarial Phosphoethanolamine Methyltransferase. *Eukaryotic Cell* **6**, 1618–1624 (2007).

83. Yang, M. & Vousden, K. H. Serine and one-carbon metabolism in cancer. *Nature Publishing Group* **16**, 1–13 (2016).

84. Lavstsen, T., Magistrado, P., Hermsen, C. C., journal, A. S. M. & 2005. Expression of Plasmodium falciparum erythrocyte membrane protein 1 in experimentally infected humans. *Springer* (n.d.) doi:10.1186/1475-2875-4-21.

85. Duffy, M. F. *et al.* Transcribed var genes associated with placental malaria in Malawian women. *Infection and immunity* **74**, 4875–4883 (2006).

86. Wang, Y.-P. & Lei, Q.-Y. Metabolite sensing and signaling in cell metabolism. *Signal Transduction and Targeted Therapy* **3**, 1–9 (2018).

87. Ganesan, S. M., Falla, A., Goldfless, S. J., Nasamu, A. S. & Niles, J. C. Synthetic RNA-protein modules integrated with native translation mechanisms to control gene expression in malaria parasites. *Nature communications* **7**, 10727 (2016).

88. Prommana, P. *et al.* Inducible Knockdown of Plasmodium Gene Expression Using the glmS Ribozyme. *PLoS ONE* **8**, e73783-10 (2013).

89. Dzikowski, R. & Deitsch, K. W. Active Transcription is Required for Maintenance of Epigenetic Memory in the Malaria Parasite Plasmodium falciparum. *Journal of Molecular Biology* **382**, 288–297 (2008).

90. Tsalikis, J., Croitoru, D. O., Philpott, D. J. & Girardin, S. E. Nutrient sensing and metabolic stress pathways in innate immunity. *Cellular microbiology* **40**, n/a-n/a (2013).
91. Ganeshan, K. & Chawla, A. Metabolic Regulation of Immune Responses. *Annual Review of Immunology* **32**, 609–634 (2014).

92. Gerriets, V. A. & Rathmell, J. C. Metabolic pathways in T cell fate and function. *Trends in immunology* **33**, 168–173 (2012).

93. Laplante, M. & Sabatini, D. M. mTOR Signaling in Growth Control and Disease. *Cell* **149**, 274–293 (2012).

94. Wek, R. C., Jiang, H. Y. & Anthony, T. G. Coping with stress: eIF2 kinases and translational control. *Biochemical Society Transactions* **34**, 7–11 (2006).

95. Kim, J. & Guan, K.-L. Amino Acid Signaling in TOR Activation. *Annual Review of Biochemistry* **80**, 1001–1032 (2011).

96. Dever, T. E. *et al.* Phosphorylation of initiation factor 2α by protein kinase GCN2 mediates gene-specific translational control of GCN4 in yeast. *Cell* **68**, 585–596 (1992).

97. Babbitt, S. E. *et al.* Plasmodium falciparum responds to amino acid starvation by entering into a hibernatory state. *Proceedings of the National Academy of Sciences of the United States of America* **109**, E3278-87 (2012).

98. Fennell, C. *et al.* PfeIK1, a eukaryotic initiation factor 2α kinase of the human malaria parasite Plasmodium falciparum, regulates stress-response to amino-acid starvation. *Malaria journal* **8**, 99–15 (2009).

99. Payne, S. H. & Loomis, W. F. Retention and loss of amino acid biosynthetic pathways based on analysis of whole-genome sequences. *Eukaryotic Cell* **5**, 272–276 (2006).

100. Divo, A. A., Geary, T. G., Davis, N. L. & Jensen, J. B. Nutritional Requirements of Plasmodium falciparum in Culture. I. Exogenously Supplied Dialyzable Components Necessary for Continuous Growth. *J Protozoology* **32**, 59–64 (1985).

101. McLean, K. J. & Jacobs-Lorena, M. The response of Plasmodium falciparum to isoleucine withdrawal is dependent on the stage of progression through the intraerythrocytic cell cycle. *Malaria journal* **19**, 1–9 (2020).

102. Munn, D. H. & Mellor, A. L. Indoleamine 2,3 dioxygenase and metabolic control of immune responses. *Trends in immunology* **34**, 137–143 (2013).

103. Munn, D. H. *et al.* Prevention of Allogeneic Fetal Rejection by Tryptophan Catabolism. *Science (New York, N.Y.)* **281**, 1191–1193 (1998).

104. Kudo, Y. The role of placental indoleamine 2,3-dioxygenase in human pregnancy. *Obstetrics & Gynecology Science* **56**, 209–8 (2013).

105. Rodriguez, P. C., Quiceno, D. G. & Ochoa, A. C. l-arginine availability regulates T-lymphocyte cell-cycle progression. *Blood* **109**, 1568–1573 (2006).

106. Viola, A., Munari, F., Sánchez-Rodríguez, R., Scolaro, T. & Castegna, A. The Metabolic Signature of Macrophage Responses. *Frontiers in Immunology* **10**, 140–16 (2019).

107. Lu, P. D., Harding, H. P. & Ron, D. Translation reinitiation at alternative open reading frames regulates gene expression in an integrated stress response. *Journal of Cell Biology* **167**, 27–33 (2004).

108. Liu, J., Istvan, E. S., Gluzman, I. Y., Gross, J. & Goldberg, D. E. Plasmodium falciparum ensures its amino acid supply with multiple acquisition pathways and redundant proteolytic enzyme systems. *Proceedings of the National Academy of Sciences of the United States of America* **103**, 8840–8845 (2006).

109. Niemand, J., Louw, A. I., Birkholtz, L. & Kirk, K. Polyamine uptake by the intraerythrocytic malaria parasite, Plasmodium falciparum. *International journal for parasitology* **42**, 921–929 (2012).

110. Biljon, R. van *et al.* Inducing controlled cell cycle arrest and re-entry during asexual proliferation of Plasmodium falciparum malaria parasites. *Scientific Reports* **8**, 1–14 (2018).

111. Ramya, T. N. C., Surolia, N. & Surolia, A. Polyamine synthesis and salvage pathways in the malaria parasite Plasmodium falciparum. *Biochemical and biophysical research communications* **348**, 579–584 (2006).

112. Bouharoun-Tayoun, H. & Druilhe, P. Antibody-Dependent Cell-Mediated Inhibition (ADCI) of Plasmodium falciparum: One- and Two-Step ADCI Assays. *Methods in molecular biology (Clifton, N.J.)* **1325**, 131–144 (2015).

113. Jafarshad, A. *et al.* A novel antibody-dependent cellular cytotoxicity mechanism involved in defense against malaria requires costimulation of monocytes FcgammaRII and FcgammaRIII. *The Journal of Immunology* **178**, 3099–3106 (2007).

114. Deitsch, K. W., Driskill, C. L., research, T. W. N. acids & 2001. Transformation of malaria parasites by the spontaneous uptake and expression of DNA from human erythrocytes. *academic.oup.com* (n.d.) doi:10.1093/nar/29.3.850.

115. McMahon, K. E. & Farrell, P. M. Measurement of free choline concentrations in maternal and neonatal blood by micropyrolysis gas chromatography. *Clin Chim Acta* **149**, 1–12 (1985).

116. Sweiry, J. H., Page, K. R., Dacke, C. G., Abramovich, D. R. & Yudilevich, D. L. Evidence of saturable uptake mechanisms at maternal and fetal sides of the perfused human placenta by rapid paired-tracer dilution: studies with calcium and choline. *J Dev Physiol* **8**, 435–45 (1986).

117. Poran, A. *et al.* Single-cell RNA sequencing reveals a signature of sexual commitment in malaria parasites. *Nature* **551**, 95–99 (2017).

118. Jiang, X. & Wang, H. Macrophage subsets at the maternal-fetal interface. *Cellular & Molecular Immunology* 1–3 (2020) doi:10.1038/s41423-020-0435-6.

119. Kumar, R., Loughland, J. R., Ng, S. S., Boyle, M. J. & Engwerda, C. R. The regulation of CD4+ T cells during malaria. *Immunol Rev* **293**, 70–87 (2020).

120. Guha, R. *et al.* Plasmodium falciparum malaria drives epigenetic reprogramming of human monocytes toward a regulatory phenotype. 1–41 (2020) doi:10.1101/2020.10.21.346197.

121. Netea, M. G. *et al.* Defining trained immunity and its role in health and disease. *Nat Rev Immunol* **20**, 375–388 (2020).

122. Boutlis, C. S., Yeo, T. W. & Anstey, N. M. Malaria tolerance – for whom the cell tolls? *Trends Parasitol* **22**, 371–377 (2006).

123. Dever, T. E. & Ivanov, I. P. Roles of polyamines in translation. *Journal of Biological Chemistry* **293**, 18719–18729 (2018).

124. Phillips, M. A. Polyamines in protozoan pathogens. *Journal of Biological Chemistry* **293**, 18746–18756 (2018).

125. Bissati, K. E. *et al.* Novel Synthetic Polyamines Have Potent Antimalarial Activities in vitro and in vivo by Decreasing Intracellular Spermidine and Spermine Concentrations. *Frontiers in Cellular and Infection Microbiology* **9**, 851–9 (2019).

126. Brummelen, A. C. van *et al.* Co-inhibition of Plasmodium falciparum S-adenosylmethionine decarboxylase/ornithine decarboxylase reveals perturbation-specific compensatory mechanisms by transcriptome, proteome, and metabolome analyses. *Journal of Biological Chemistry* **284**, 4635–4646 (2009).

127. Müller, S. *et al.* In the Human Malaria Parasite Plasmodium falciparum, Polyamines Are Synthesized by a Bifunctional Ornithine Decarboxylase, S-Adenosylmethionine Decarboxylase*. *J Biol Chem* **275**, 8097–8102 (2000).

128. Wrenger, C., Lüersen, K., Krause, T., Müller, S. & Walter, R. D. The Plasmodium falciparum Bifunctional Ornithine Decarboxylase, S-Adenosyl-l-methionine Decarboxylase, Enables a Well Balanced Polyamine Synthesis without Domain-Domain Interaction. *Journal of Biological Chemistry* **276**, 29651–29656 (2001).

129. Ivanov, I. P. *et al.* Polyamine Control of Translation Elongation Regulates Start Site Selection on Antizyme Inhibitor mRNA via Ribosome Queuing. *Molecular Cell* **70**, 254-264.e6 (2018).

130. Kwon, H., Wu, G., Bazer, F. W. & Spencer, T. E. Developmental Changes in Polyamine Levels and Synthesis in the Ovine Conceptus. *Biol Reprod* **69**, 1626–1634 (2003).

131. Lenis, Y. Y., Elmetwally, M. A., Maldonado-Estrada, J. G. & Bazer, F. W. Physiological importance of polyamines. *Zygote* **25**, 244–255 (2017).

132. Kaiser, A. Translational control of eIF5A in various diseases. *Amino Acids* **42**, 679–684 (2012).

133. Colvin, H. N. & Cordy, R. J. Insights into malaria pathogenesis gained from host metabolomics. *Plos Pathog* **16**, e1008930 (2020).

134. TOME, E. M., FISER, M. S., PAYNE, M. C. & GERNER, W. E. Excess putrescine accumulation inhibits the formation of modified eukaryotic initiation factor 5A (eIF-5A) and induces apoptosis. *Biochem J* **328**, 847–854 (1997).

135. Lomvardas, S. *et al.* Interchromosomal interactions and olfactory receptor choice. *Cell* **126**, 403–413 (2006).

136. Hnisz, D., Shrinivas, K., Young, R. A., Chakraborty, A. K. & Sharp, P. A. A Phase Separation Model for Transcriptional Control. *Cell* **169**, 13–23 (2017).

137. Gurumurthy, A., Shen, Y., Gunn, E. M. & Bungert, J. Phase Separation and Transcription Regulation: Are Super-Enhancers and Locus Control Regions Primary Sites of Transcription Complex Assembly? *BioEssays : news and reviews in molecular, cellular and developmental biology* **41**, e1800164 (2019).

138. Duraisingh, M. T. *et al.* Heterochromatin Silencing and Locus Repositioning Linked to Regulation of Virulence Genes in Plasmodium falciparum. *Cell* **121**, 13–24 (2005).

139. Ralph, S. A., Scheidig-Benatar, C. & Scherf, A. Antigenic variation in Plasmodium falciparum is associated with movement of var loci between subnuclear locations. *Proceedings of the National Academy of Sciences* **102**, 5414–5419 (2005).

140. Voss, T. S. *et al.* A var gene promoter controls allelic exclusion of virulence genes in Plasmodium falciparum malaria. *Nature* **439**, 1004–1008 (2005).

141. Dzikowski, R. *et al.* Mechanisms underlying mutually exclusive expression of virulence genes by malaria parasites. *Embo Rep* **8**, 959–965 (2007).

142. Frank, M., Dzikowski, R. & Deitsch, K. Variable switching rates of malaria virulence genes are associated with chromosomal position. *Molecular microbiology* **64**, 1486–1498 (2007).

143. Ploeg, L. H. V. der. Control of antigenic variation in African trypanosomes. *New Biologist* **3**, 324–30 (1991).

144. Navarro, M. & Gull, K. A pol I transcriptional body associated with VSG mono-allelic expression in Trypanosoma brucei. *Nature* **414**, 759–763 (2001).

145. Hozumi, N. & Tonegawa, S. Evidence for somatic rearrangement of immunoglobulin genes coding for variable and constant regions. *Proc National Acad Sci* **73**, 3628–3632 (1976).

146. Toumey, C. Compare and contrast as microscopes get up close and personal. *Nat Nanotechnol* **6**, 191–193 (2011).

147. Warrenfeltz, S. *et al.* Eukaryotic Genomic Databases, Methods and Protocols. *Methods Mol Biology* **1757**, 69–113 (2018).

148. Kanehisa, M. & Goto, S. KEGG: Kyoto Encyclopedia of Genes and Genomes. *Nucleic Acids Res* **28**, 27–30 (2000).

149. Wishart, D. S. *et al.* HMDB 4.0: the human metabolome database for 2018. *Nucleic Acids Res* **46**, gkx1089- (2017).

150. GR, C., WE, C., M, W. & PG, C. *The Primate Malarias*. (U.S. Department of Health, Education and Welfare, Bethesda, 1971).

151. Chen, D. S. *et al.* A Molecular Epidemiological Study of var Gene Diversity to Characterize the Reservoir of Plasmodium falciparum in Humans in Africa. *Plos One* **6**, e16629 (2011).