ASSESSMENT OF THE EFFECTS OF MALARIA AND ANEMIA IN PREGNANT MALAWIAN WOMEN BEFORE AND AFTER TREATMENT OF MODERATE MALNUTRITION

A Thesis

presented to

the Faculty of California Polytechnic State University,

San Luis Obispo

In Partial Fulfillment

of the Requirements for the Degree of

Master of Science in Nutrition

by

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July 2020

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ABSTRACT

Assessment of the Effects of Malaria and Anemia in Pregnant Malawian Women Before and After Treatment of Moderate Malnutrition

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Background: Moderate acute malnutrition (MAM) can lead to adverse maternal and infant outcomes and possibly further complications. Supplementary foods or treatments with high quality nutrients should be administered to those with MAM in hopes to increase the chance of healthy maternal and infant outcomes. Sometimes supplementary food is not enough to overcome MAM and disease may play a role, particularly in pregnant malnourished women.

Objective: To determine if the effects of malaria and anemia moderated the effect of nutritional treatments (one of the three given nutritional interventions) used to improve malnutrition and achieve a MUAC ≥ 23 cm during study participation. Additionally, this research serves to assess whether the relationship between malaria and anemia is associated with malnutrition status.

Methods: Women were given a dose of IPTp at each antenatal visit between zero and four total IPTp doses. Infant anthropometrics – length, weight, head circumference, and MUAC were taken at birth, 6 weeks, and 3 months. Maternal hemoglobin levels were assessed at enrollment and after 10 weeks of enrollment as well as infant hemoglobin at 3 months. Anemia was defined by a hemoglobin less than 11.0 g/dL. Mild anemia was defined as hemoglobin greater than 7.0 but less than 9.9 g/dL and moderate anemia was defined by hemoglobin values 9.9 or greater but less than 11.0 g/dL. Analysis was completed using ANOVA, and if any significant differences were observed, they were compared via Tukey HSD (continuous) or Chi-squared test (categorical).

Results: Total number of IPTp doses was found to be a more statistically significant predictor of maternal weight gain during treatment than timing of the doses. It stands to reason that women receiving three or more IPTp doses was the most beneficial for women during treatment as it saw the highest increases in maternal weight gain. At baseline, women that achieved a MUAC > 23 cm during the study was 32.0% (n = 1805). The greatest proportion of women, after adjustment, that achieved a MUAC ≥ 23 cm was seen in women receiving four (47.3 %) and three (37.8 %) total IPTp doses during pregnancy. Maternal weight gain correlated closely with hemoglobin at enrollment (p-value = 0.0111). Total number of IPTp doses received during pregnancy was not found to have a statistical effect on infant hemoglobin or anemia at three months. Infant length at six weeks was higher in infants from mothers that received two or three IPTp doses compared to mothers that received one IPTp dose (p-value = 0.0218). A p-value below 0.05 by total number of IPTp doses was observed for infant weight, head circumference, and MUAC at birth, six weeks, and three months.

Conclusion: At least three IPTp was effective in improving maternal weight gain and achievement of MUAC > 23 cm as well as improved many infant outcomes. Hemoglobin at enrollment was a predictor of maternal weight gain during tx but was not associated with any other outcomes.

ACKNOWLEDGMENTS

Thank you to my graduate advisor, Dr. Peggy Papathakis, for her guidance, encouragement, and support throughout my entire experience at Cal Poly. I would also like to thank her for continuing to never give up on my work, even though slow and unproductive times. Thank you for my graduate committee chairs, Dr. Andrew Schaffner, for contributing countless hours of statistical analysis and interest in my thesis research, as well as Dr. Michael LaFrano, for always being a willing contributor. A special thank you to my wife, for supporting me over a longer period of time than desired initially and to never give up on my research. Thank you to Cal Poly and the opportunities, facilities, and supplies to create an environment conducive to learning and research.

TABLE OF CONTENTS

Page

LIST OF TABLES ix
LIST OF FIGURES x
CHAPTER

1. INTRODUCTION 1
2. LITERATURE REVIEW 3
    2.1 Maternal Outcomes in Normal Pregnancy 3
        2.1.1 Gestational Weight Gain 3
        2.1.2 Maternal MUAC 4
    2.2 Malaria and Preventative Treatments 5
        2.2.1 Malaria 5
        2.2.2 Consequences of Malaria 7
        2.2.3 Malaria Prevention During Pregnancy 7
        2.2.4 Malaria Treatment During Pregnancy 10
        2.2.5 Malarial Resistance 11
    2.3 Hemoglobin and Anemia 13
        2.3.1 Types of Anemia 13
        2.3.2 Anemia in Pregnancy 13
        2.3.3 Anemia Risk Factors 15
        2.3.4 Consequences of Anemia During Pregnancy 17
    2.4 Malawi 17
        2.4.1 Maternal and Infant Statistics in Malawi 17
    2.5 Conclusion 20
3. METHODS AND MATERIALS 21
    3.1 Methods and Objective 21
    3.2 Study Participants 22
    3.3 Randomization and Blinding 23
    3.4 Study Design 24
    3.5 Data Collection 26
    3.6 Statistical Analysis 27
4. RESULTS 28
    4.1 Baseline: Maternal Characteristics 28
    4.2 Unadjusted Maternal Health Outcomes by IPTp Dosing 29
    4.3 Adjusted Maternal Health Outcomes by IPTp Dosing 30
        4.3.1 Weight Gain During Treatment by Total IPTp Doses 32
        4.3.2 Weight Gain During Treatment by IPTp Doses in the 2nd Trimester 34
        4.3.3 Weight Gain During Treatment by IPTp Doses in the 3rd Trimester 34
        4.3.4 Weight Gain During Treatment and Covariates 35
        4.3.5 Achievement of MUAC ≥ 23 cm by Total Number of IPTp Doses 35
        4.3.6 Achievement of MUAC ≥ 23 cm by IPTp Doses in the 2nd Trimester 36
        4.3.7 Achievement of MUAC ≥ 23 cm by IPTp Doses in the 3rd Trimester 36
        4.3.8 Achievement of MUAC ≥ 23 cm and Covariates 38

4.4 Maternal Hemoglobin and Anemia 38
4.5 Unadjusted Maternal Health Outcomes by Hemoglobin and Anemia 39
4.5.1 Adjusted Maternal Weight Gain During Treatment by Hemoglobin 40
4.5.2 Adjusted Maternal Weight Gain During Treatment by Anemia 42
4.5.3 Adjusted Achievement of MUAC ≥ 23 cm by Hemoglobin and Anemia 43
4.6 Baseline: Infant Characteristics 43
4.7 Unadjusted Infant Health Outcomes by IPTp 46
4.7.1 Infant Health Outcomes by Maternal IPTp Dosing During Pregnancy 49
4.7.2 Adjusted Infant Hemoglobin and Anemia at 3 Months by IPTp 49
4.7.3 Adjusted Infant Length by IPTp 50
4.7.4 Adjusted Infant Weight by IPTp 51
4.7.5 Adjusted Infant Head Circumference by IPTp 54
4.7.6 Adjusted Infant MUAC by IPTp 55
4.7.7 Summary of Infant Outcomes by IPTp 56
4.8 Adjusted Infant Health Outcomes by Hemoglobin/Anemia 56
4.9 Adjusted Infant Health Outcomes by Maternal Hemoglobin and Anemia 56
4.10 Adjusted Infant Health Outcomes by Maternal Anemia 58
5. DISCUSSION 61
5.1 IPTp 61
5.2 Hemoglobin and Anemia 64
5.3 Strengths and Limitations 64
6. CONCLUSION 68
REFERENCES 70
APPENDIX 86

LIST OF TABLES

TABLE Page

Table 1: Institute of Medicine Weight Gain Recommendations for Pregnancy....4
Table 2: Prevalence of *Pfdhfr* and *Pfdhps* Single and Haplotype Mutations Among Febrile Pregnant Women Presenting at Outpatient Clinics....12
Table 3: Baseline and Follow-up Maternal Characteristics....28
Table 4: Distribution of IPTp Doses Among Women Enrolled....30
Table 5: Maternal Characteristics Compared Across Number of IPTp Doses Reported During Pregnancy....31
Table 6: Achievement of MUAC ≥ 23 cm by IPTp Dosing....36
Table 7: Baseline and Follow-up Maternal Hemoglobin and Anemia....39
Table 8: Univariate Analysis of Hemoglobin by Intervention Group....40
Table 9: Baseline and Follow-up Infant Characteristics....44
Table 10: Baseline and Follow-up Infant Z-scores....45
Table 11: Unadjusted Infant Health Outcomes Across Number of IPTp Doses Reported....46
Table 12: Unadjusted Infant Health Outcomes Compared Across Number of IPTp Doses Reported in Z-scores....47
Table 13: Infant Hemoglobin and Anemia by Number of Total Number IPTp Doses....50
Table 14: Infant Length by Total Number IPTp Doses....50
Table 15: Infant LFA (Z-scores) by Total IPTp Doses....51
Table 16: Infant Weight by Total Number IPTp Doses....52
Table 17: Infant WFA (Z-scores) by Total IPTp Doses....53
Table 18: Infant Head Circumference by Total Number of IPTp Doses....54
Table 19: Infant HCFA (Z-scores) by Total IPTp Doses....54
Table 20: Infant MUAC by Total Number of IPTp Doses....55
Table 21: Infant Outcomes by Maternal Hemoglobin at Enrollment and at 10 Weeks After Enrollment....57
Table 22: Adjusted Infant Health Outcomes by Maternal Anemia at Enrollment and at 10 Weeks After Enrollment....58
Table 23: Adjusted Infant Health Outcomes by Severity of Maternal Anemia at Enrollment and at 10 Weeks After Enrollment....59
Appendix Table A: Nutrient Composition and Comparison by Treatment....86
Appendix Table B: Univariate Analysis of IPTp Doses and Categorical Covariates....87
Appendix Table C: Univariate Analysis of IPTp Doses and Continuous Covariates....88

LIST OF FIGURES

FIGURE Page

Figure 1: The Relationship Between Malnutrition and Infection or Disease....2
Figure 2: Linear Correlation of Birthweight and Maternal MUAC....5
Figure 3: Folate Metabolism....9
Figure 4: Chemical Structure of Sulfadoxine and Pyrimethamine....10
Figure 5: Transfer of Iron from Mother to Fetus....15
Figure 6: Reported Data on Malawi Micronutrient Status from 2015-2016....19
Figure 7: Weight Gain During Treatment Across Total Number of IPTp Doses, Doses During the 2nd Trimester, and Doses During the 3rd Trimester....33
Figure 8: The Prevalence of Achieving MUAC ≥ 23 cm by Total Number of IPTp Doses....37
Figure 9: Hemoglobin at Enrollment as a Predictor of Maternal Weight Gain During Tx....41
Figure 10: Change in Hemoglobin as a Predictor of Maternal Weight Gain During Tx....42

1

CHAPTER 1:

INTRODUCTION

Malnutrition is a serious global health issue, especially in developing countries. Pregnant women with malnutrition are particularly susceptible to pregnancy related complications due to the increased demand of many nutrients throughout the duration of pregnancy, integral to fetal development (Darnton-Hill, 2015). Current research suggests that maternal malnutrition may lead to poor fetal development resulting in low birth weight, stillbirths, preterm birth, neonatal mortality, and halted linear growth (Papathakis et al., 2016). There is a strong relationship between malnutrition and disease, particularly in infants, because malnutrition leads to underweight and stunted children that are more susceptible to disease. Illness only worsens a person's nutritional status by ailments such as diarrhea, malabsorption, and diversion of nutrients for an immune response. Energy and micronutrient needs are increased with illness such as fever (Katona, 2008). In a 2005 study, the WHO estimated that more than half of all children deaths prior to 5 years of age are due to five main diseases – pneumonia, diarrhea, malaria, measles, and AIDS. 94% of the global deaths for children under 5 are attributed to one of the main diseases occur in the African region with undernutrition being a comorbidity in 53% of these deaths (Bryce J, 2005).

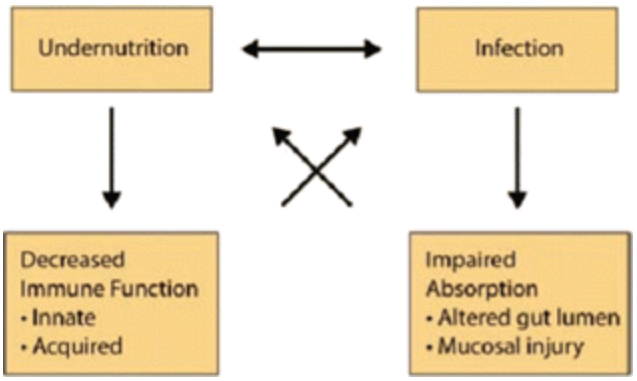

Figure 1: The relationship between malnutrition and infection or disease (Katona, 2008).

Several factors contribute to and are responsible for malnutrition, such as, personal health, socioeconomic status, and household food insecurity. Under nutrition continues to be a significant problem in Sub-Saharan Africa. In 2005, the prevalence of malnutrition was 24.3 % and continues to be a staggering 22.8 % as recent as 2018 (WFP, 2019). Malnutrition has been shown to increase the prevalence of comorbidities, including, tuberculosis, diarrhea, malaria, and anemia (Bain, 2013). During pregnancy, we see similar complications, such as anemia, increased risk of life-threatening hemorrhages, and pre-eclampsia (Wu, 2012).

CHAPTER 2:

LITERATURE REVIEW

2.1 Maternal Outcomes in Normal Pregnancy

2.1.1 Gestational Weight Gain

Pregnancy is a unique and complex biological condition, and central to that is gestational weight gain, which supports growth and development of the fetus. Women that are pregnant enter an anabolic state during which the body increases it's overall mass, fetal mass, blood volume, amniotic fluid, and placental growth (Rasmussen, 2009). Recommended weight gain depends on women BMI, as shown in Figure 1, with higher weight gain recommendations for women with lower BMI. The recommendations are based on evidence that supports fetal growth and infant outcomes. (IOM, 2009).

Developed countries have seen increases in the prevalence of excessive gestational weight gain (Kominiarek, 2017); however, in developing countries there is a high prevalence of inadequate gestational weight gain to support optimal maternal and fetal outcomes. This high prevalence of inadequate gestational weight gain has been attributed to low food availability and nutrient intake. (Rasmussen, 2009). The IOM recommends an average weekly weight gain during pregnancy in the 2nd and 3rd trimesters of 0.434 kg (IOM, 2009). Weight gain and nutritional status during pregnancy is positively related to fetal growth and infant outcomes, especially during the second and third trimesters (Stein, 2004).

Many studies have shown a strong correlation between low gestational weight gain and low birthweight. A study done by Frederick et al. showed that women gaining

less than the median gestational weight gain, or 15.9 kg during pregnancy, was associated with twice the risk of low birthweight after adjusting for covariates (Frederick, 2008). A large meta-analysis done by Han et al showed that low total gestational weight gain, between 11.5 and 12.5 kg, was associated with increased risk of low birthweight in developing countries, which suggests that a weekly gain of 0.3 kg is indicative of increased risk for low birthweight; however, the meta-analysis also only showed a statistical correlation between weekly gestational weight and preterm birth, but not low birthweight (Han, 2011).

Table 1: Institute of Medicine Weight Gain Recommendations for Pregnancy

Prepregnancy Weight Category	Body Mass Index*	Recommended Range of Total Weight (lb)	Recommended Rates of Weight Gain† in the Second and Third Trimesters (lb) (Mean Range [lb/wk])
Underweight	Less than 18.5	28–40	1 (1–1.3)
Normal Weight	18.5–24.9	25–35	1 (0.8–1)
Overweight	25–29.9	15–25	0.6 (0.5–0.7)
Obese (includes all classes)	30 and greater	11–20	0.5 (0.4–0.6)

[1]Adapted from Institute of Medicine (IOM), 2016.

2.1.2 Maternal MUAC

Mid-upper arm circumference (MUAC) can be a useful indicator for maternal malnutrition as well as predict low birthweight. The arm has both subcutaneous fat and muscle, so changes in MUAC may indicate a change in sub-cutaneous fat as well as muscle (Kumar, 2018). In low socioeconomic areas and regions with limited resources available, individuals tend to have smaller amounts of subcutaneous fat, changes in MUAC are closely associated with changes in muscle mass (Tang, 2016). MUAC > 21

cm is commonly used a cut-off for severe malnutrition and MUAC > 23 cm for moderate malnutrition. An observational study done in India showed a MUAC > 23 cm to be a good predictor of low birthweight as seen in Figure 2.

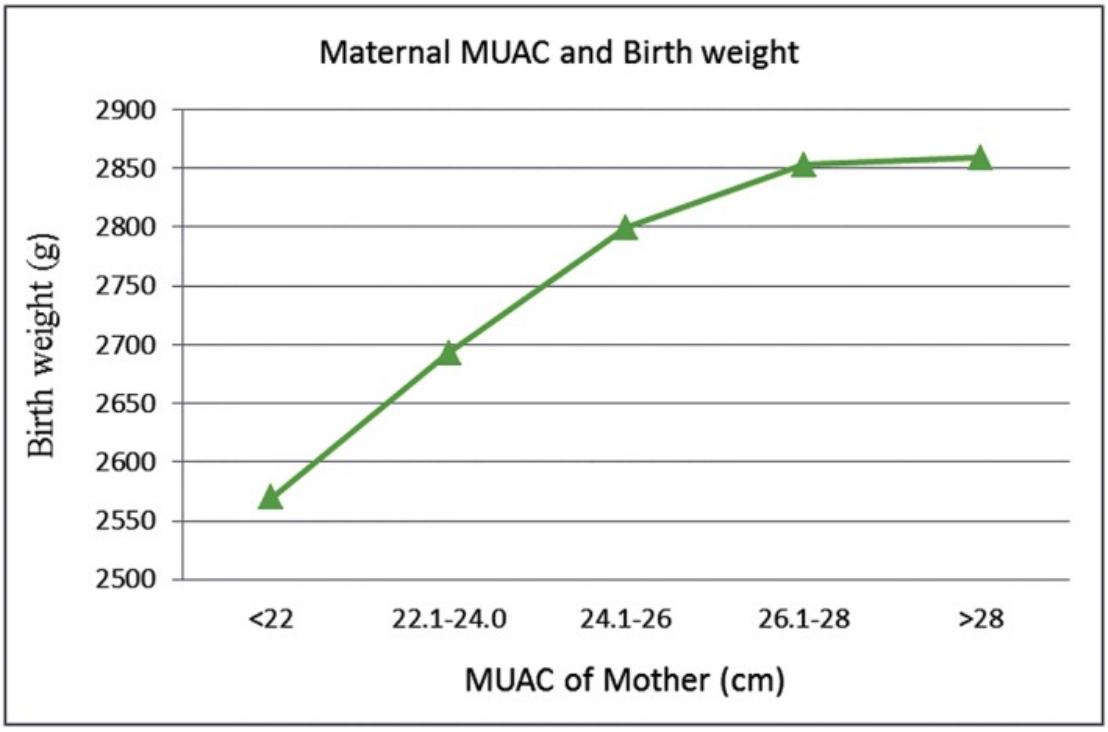

Figure 2: Linear Correlation of Birthweight and Maternal MUAC. (Adapated from Vasundhara, 2019).

2.2 Malaria and Preventative Treatments

2.2.1 Malaria

Malaria, according to the WHO, is a life-threatening disease caused by parasites that are transmitted to people through bites from carrier female Anopheles mosquitos. The disease is a preventable and curable one. Malaria is caused from a Plasmodium parasite, of which there are five different species of these parasites that may cause a malarial infection. The two most common species are P. falciparum and P. vivax. P. falciparum is the most common malarial parasite on the African continent and is

responsible for most of malarial-related deaths worldwide, while P. vivax thrives in most countries outside of Africa (WHO, 2020).

Symptoms of malaria begin as fevers and headaches; and may be difficult to diagnose as malaria. It is crucial that symptoms be treated as soon as possible, preferentially within the first 24 hours of infection because it may progress to a severe illness and can result in death. It is common for infants with malaria to develop anemia, metabolic acidosis, or respiratory stress. Anemia and malaria are prevalent comorbidities throughout most of the African continent, which increases the importance of early diagnosis and treatment. Another reason for the importance of early diagnosis of malaria is to prevent spreading infection throughout local communities; malaria should be treated as a medical emergency. The Centers for Disease Control and Prevention (CDC) identify late diagnoses of malaria as being the leading cause of death in malaria patients throughout the United States (CDC. 2019).

According to the WHO, World Health Organization, there are approximately 3.4 billion people worldwide throughout 91 different countries that are at risk for developing malarial disease, and of those 3.4 billion, 1.1 billion are considered at high risk, or more than one person out of every thousand people will develop malaria in a calendar year. The World Malaria Report reported 228 million cases of malaria throughout the globe in 2018, which led to 405,000 deaths. The WHO reported that roughly 94% of all malarial deaths occurred within the African Region (WHO, 2019). Even though malarial is a worldwide disease, over 90% of cases are from African populations where the malarial parasite affects mainly young infant and pregnant women (Snow, 2005).

2.2.2 Consequences of Malaria

Malaria has severe implications on nutritional status, especially if the person is already malnourished. Pregnant women have a higher risk than non-pregnant women of being malnourished due to the increased nutrient needs (Unger, 2016). This can be especially dangerous in high-risk countries susceptible to malaria such as Sub-Saharan African populations (Rogerson, 2017). Macronutrient deficiency has been associated with increased morbidity and mortality related to malaria in infant, non-pregnant adults, and pregnant adults. These findings suggest malaria is instrumental in interactions with the immune system (Shankar, 2000). Malaria has been shown to cause nutritional depletion and worsen nutrient status in already undernourished populations, especially infants (Rogerson, 2017). Malnourished women with malarial infections have a significantly more profound effect on fetal growth as well as birth weight than in well-nourished women with malaria (Nyakeriga, 2004). In a study conducted in the Democratic Republic of Congo, they found that undernourished women with early pregnancy malaria, with low MUAC and BMI, had a 3.6 times increased risk of having infants with stunted growth (Griffin, 2012).

2.2.3 Malaria Prevention During Pregnancy

According to a study by Feng et al, between 1999 and 2006, the use of bed nets in Malawi went from 14.4 % during pregnancy to 60.5% and showed a significant association with decreased malarial contraction, an odds ratio of 0.47 [CI 95%: 0.37, 0.60], and infant low birthweight, but was unable to show a decrease in anemia. In the study, infant birth weight was 47 g higher on average in women using bed nets compared to non-users (Feng, 2010). The use of bed nets in conjunction with at least two doses of

SP IPTp during pregnancy also showed a significant decrease in rates of infection with malaria and low birth-weight compared to no bed net use with zero or one doses of SP, odds ratio 0.38, [CI 95%: 0.27, 0.53]. Women with at least two doses of SP IPTp and without the use of bed nets did not show a significant decrease in malaria infection, odds ratio 0.87 [CI 95%: 0.67, 1.13] (Feng, 2010). Another study showed that three or more doses of IPTp decreased the incidence of low birthweight from 9.9 % to 6.9 % (Cates, 2018).

SP is a common low-cost treatment for pregnant women in Sub-Saharan regions at Antenatal Clinics for the prevention of contracting malaria. In 2020, the WHO recently issued new guidelines outlining the use of SP from two or more doses to one dose at every scheduled antenatal clinic (ANC) visit beginning in the second trimester (WHO, 2020). This new guideline stems from a meta-analysis of seven trials that showed three or more doses of SP were much more effective at overall malarial prevention, reducing risk of infection by as much as half, than the previous two-dose recommendation. Also, there is evidence that three of more doses of SP as IPTp has a greater chance to reduce the risk of severe maternal anemia and adverse birth outcomes, such as low-birth weight than the former two-dose recommendation (Kayentao, 2013). It is important that each facility using SP as IPTp continue to monitor SP efficacy and molecular resistance markers because almost 70% of malarial cases are resistant to both cholorquine and SP antimalarials.

SP is a combination drug composed of sulfadoxine and pyrimethamine. Sulfadoxine works mechanistically is by targeting Plasmodium dihydropteroate synthase, which converts dihydropteroate diphosphate and p-aminobenzoic acid (PABA) to

dihydropteroic acid. Specifically, SP acts competitive inhibitor of the enzyme, competing with PABA for incorporation into folic acid. As Figure 3 indicates, this inhibited step is crucial for the synthesis of folic acid, which is vital in the synthesis, methylation, and repair of DNA. Without folic acid, the Plasmodium falciparum bacterium have difficulty in reproduction since they are unable to get folic acid from host cells because folic acid does not diffuse through bacterial cell walls (Shultz, 1994).

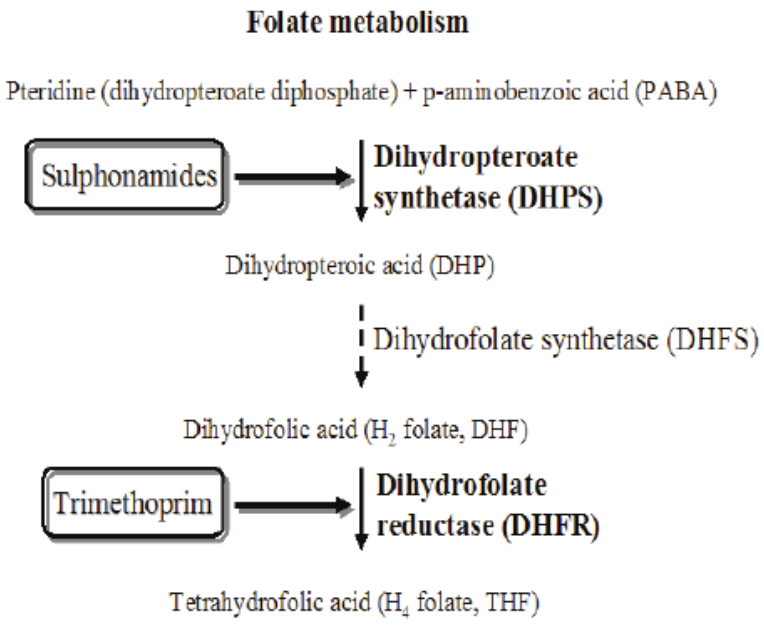

Figure 3: Folate Metabolism. (adapted, Crider, 2012).

Pyrimethamine is similar to sulfadoxine but inhibits dihydrofolate reductase (DHFR) instead of DHPS. Inhibition of dihydrofolate reductase is associated with blocking the biosynthesis of purine and pyrimidine nitrogenous bases. This leads to failure to replicate DNA and the bacterial cell is unable to reproduce (Shultz, 2013).

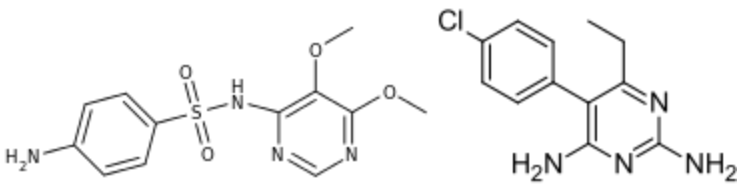

Figure 4: Chemical Structure of Sulfadoxine and Pyrimethamine. (PubChem, Accessed June 16th, 2020).

2.2.4 Malarial Treatment During Pregnancy

Lariam, mefloquine, is generally the drug of choice when treating malaria in endemic areas of the world such as Sub-Saharan Africa. In 2013, Lariam was reclassified as a pregnancy category B drug, though initially was a category C drug (FDA, 2013). Category B drugs are classified as drugs that have failed to demonstrate a risk to the fetus in animal reproduction but haven't been in many well-controlled studies with pregnant women. Category C drugs are classified as having shown an adverse effect on the fetus in animal reproduction and should be used cautiously when treating pregnant women. (Federal Register, 2017). Lariam has at least five major characteristics that make it a desirable choice for malarial treatment: a long half-life of between 14 to 28 days, a single dose administration, a well-researched pharmacokinetic profile in pregnant women, infrequent MQ resistance in Sub-Saharan Africa, and an acceptable level of toxic side effects during reproduction in animal studies (Briand, 2009; Bangchang, 1994; Nosten, 1990; Ward, 2007). Mefloquine has been found to be effective against malaria parasites that are resistant to chloroquine or sulfadoxine-pyrimethamine. It's mechanism of action involves erythrocytes within the Plasmodium species. It is known as a blood schizonticide, which destroys shizonts in the blood stream. Shizonts are mature malaria parasites that infect the red blood cells of the human host (Schlagenhauf, 1999).

2.2.5 Malarial Resistance

With a continued expansion of Plasmodium falciparum resistance to antimalarial therapies, malarial control and antimalarial strategies are important in high at-risk populations such as sub-Saharan African populations. In 1993, chloroquine was discontinued as the predominant chemotherapy drug and replaced by sulfadoxine-pyrimethamine (SP) in Malawi, and used to prevent and treat uncomplicated malaria due to a high resistance response; however, within the next decade, chloroquine-susceptible parasites reemerged in Malawi (Artimovich, 2015). Within the decade following 1993, parasite resistance to sulfadoxine-pyrimethamine (SP) was seen (Kublin, 2002).

Plasmodium falciparum develop or adapt sulfadoxine-pyrimethamine resistance polymorphisms within the dihydrofolate reductase-thymidylate synthase (encoded by pfdhfr-ts) and dropteroate synthase genes (encoded by pfdhps) (Kublin, 2002). More specifically, mutations occur at three different codons of the Pfdhfr gene: N51I, C59R, and S108N. Mutations at these codons is commonly referred to as the triple mutation; whereas, mutations at the A437G and G540E codons within the Pfdhps gene are referred to as the double mutation (McCollum, 2010). Studies in Eastern Africa have shown the emergence of a third mutation within the Pfdhps at codon 581G, which in conjunction with the other five mutations makes a sextuple mutation (Spalding, 2010). This is especially important when considering the use of IPTp and SP treatment for prevention of malaria because there is evidence that sextuple mutations are associated with reduced birth weight and increased risk of parasitemia and placental infection by Plasmodium falciparum, which suggests that SP contributes to malaria in women with sextuple mutant strains (Harrington, 2013; Minja, 2013). Table 1 shows the prevalence of each of the six

mutations observed in P. falciparum-positive women during pregnancy in a recent study. Overall, the proportion of P. falciparum-positive was 128/998 or 12.8%. 8/101 or 8% of women were observed carrying the sextuple mutant, which indicates that SP-IPTp treatment is losing efficacy and may become ineffective overtime (Anthony, 2015). There has been no evidence of parasite-susceptibility to SP after discontinued use for several years, unlike that seen with chloroquine (Artimovich, 2015). These findings show a difficulty factor associated with anti-malarial and first-line treatments when dealing with high-risk populations.

Table 2: Prevalence of *Pfdhfr* and *Pfdhps* Single and Haplotype Mutations Among Febrile Pregnant Women Presenting at Outpatient Clinics[a] (Mbonye, 2015).

Pfdhfr and *Pfdhps* haplotype(s)	Prevalence of *Pfdhfr* and *Pfdhps* mutations[b]
dhfr 51I	111/111 (100)
dhfr 59R	112/116 (97)
dhfr 108N	120/120 (100)
dhps 437G	120/120 (100)
dhps 540E	119/120 (99)
dhps 581G	8/103 (8)
dhps 613S	0/111 (0)
dhfr double mutant 51I+108N	111/111 (100)
dhfr double mutant 59R+108N	112/116 (97)
dhps double mutant 437G+540E	119/120 (99)
dhfr triple mutant 51I+59R+108N	107/110 (97)
Quadruple mutant (51I+59R+108N)+437G	107/110 (97)
Quintuple mutant (51I+59R+108N)+ (437G+540E)	106/110 (96)
Sextuple mutant (51I+59R+108N)+ (437G+540E+581G)	8/101 (8)

[a] The proportion of *P. falciparum*-positive mutations was 128/998 (12.8%).
[b] Prevalence data are expressed as follows: number of mutations/total number of samples tested (%).

2.3 Hemoglobin and Anemia

2.3.1 Types of Anemia

Anemia is defined by the World Health Organization (WHO) as a hemoglobin concentration below 11.0 g/dL and as of 2016, has high prevalence in both developed and developing countries, being the highest in South-East Asian at 48.1 % followed by Africa with a prevalence rate of 46.2 % (WHO, 2017). The most common types of anemia are iron-deficiency anemia, pernicious anemia, aplastic anemia, and hemolytic anemia. Iron-deficiency anemia is the most common type of anemia, and iron is essential in human energy production, oxygen transport, oxygen utilization, cell proliferation, and pathogen destruction. Iron-deficiency primarily occurs in persons with increased iron requirements, such as children, adolescents, women during childbearing years, and pregnancy. With diminished oxygen carrying capacity due to iron-deficiency, symptoms of weakness, fatigue, shortness of breath, and decreased physical capacity are common (Lynch, 2018).

Pernicious anemia is a direct result of vitamin B-12 deficiency and results in megaloblastic, or larger than normal, red blood cells. Vitamin B-12 deficiency is not always a from lack of dietary intake, but is a consequence of intrinsic factor deficiency due to the parietal cells in the corpus of the stomach not producing enough intrinsic factor to assist in Vitamin B-12 absorption in the small intestine (Toh, 2004).

2.3.2 Anemia in Pregnancy

Iron is needed in the greatest quantities during the third trimester, where the most iron is transferred to the fetus (WHO, 2015). The most iron transferred to the fetus occurs after 30 weeks of gestation (Allen, 2000). Iron absorption is increased during iron transfer from the mother to fetus and is regulated by the placenta. Typically, serum

ferritin significantly decreases between weeks 12 to 25 of gestation to increase iron utilization as red blood cells are synthesized (Harris, 1992). Figure 1 depicts serum transferrin transporting iron from maternal blood to transferrin receptors on the epithelial surface of embryonic placental villi, then holotransferrin is endocytosed, iron is released in the fetal blood, and apotransferrin returns to the maternal blood. Then, a similar cascade occurs where iron binds to ferritin in placental cells, which is then transferred to apotransferrin, enters the fetal side of the placenta, and enters the fetal blood stream as holotransferrin (Starreveld, 1995). With low maternal iron status, there is an increased number of placental transferrin receptors to promote iron uptake by the placenta, thus leaving the mother at greater risk for iron-deficiency anemia (Allen, 2000).

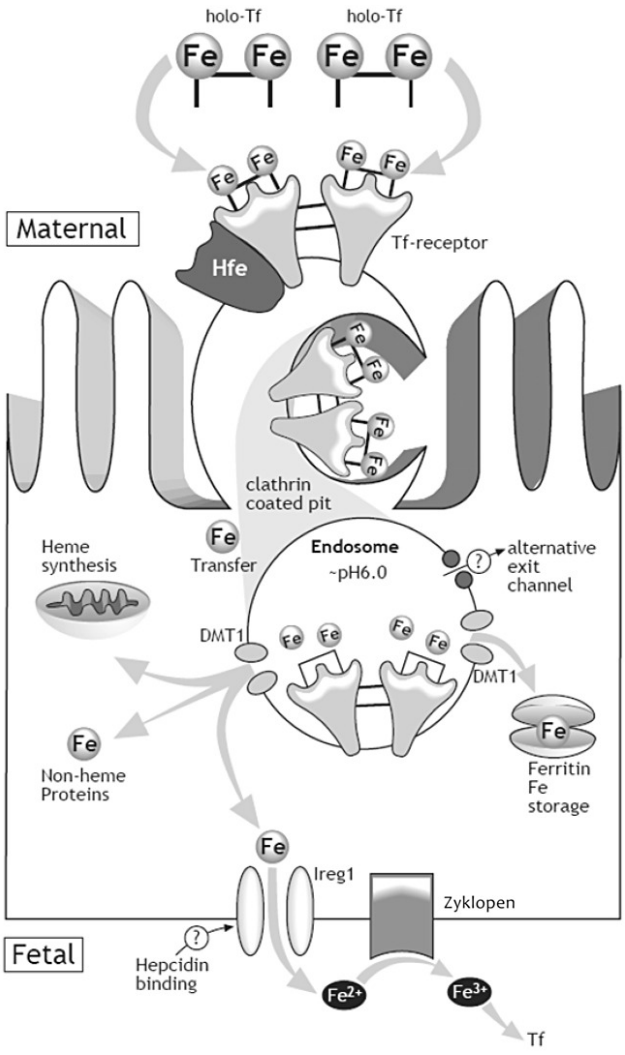

Figure 5: Transfer of iron from mother to fetus (Mcardle, 2011).

2.3.3 Anemia Risk Factors

There are several risk factors for anemia in general, such as: malaria, schistosomiasis, tuberculosis, HIV/AIDS, geophagia, parasites, and socioeconomic status (WHO, 2015). Parasites, such as Plasmodium falciparum, significantly increase the risk of iron deficiency anemia during pregnancy. Malaria prevention is a key focus to lowering the risk of iron deficiency anemia in malaria endemic areas such as Africa and South America (Anlaakuu, 2017). Low socioeconomic status shows the highest risk for

iron deficiency anemia in infant and pregnant women – there is the least access to interventions, treatments, and services (Mawani, 2016).

Nutritional considerations with iron-deficiency anemia are based on the two main forms of dietary iron: heme and nonheme. Generally, heme iron is found in meats and nonheme iron is found in meats, as well as plant-based foods. Some dietary considerations with iron to consider are absorption enhancers and inhibitors. Inhibitors of iron absorption include dairy products, eggs, polyphenols such as in coffee, phytates, divalent cations, and oxalates. Enhancers of iron absorption include fruits and vegetables. Some challenges specific to Sub-Saharan African regions include limitations to availability of iron rich foods as well as high costs (Mwangi, 2017). From a bioavailability standpoint, there is evidence that African diets are low in ascorbic acid, or Vitamin C, which enhances the absorption of iron, as well as low bioavailability of iron due to diets high in legume and cereals, containing inhibitors of iron absorption (Zimmerman, 2005).

The WHO has developed guidelines specifically for the prevention, control, and treatment of anemia in reproductive age women. Daily iron and folic acid supplementation is recommended as part of antenatal care (ANC), especially in areas where the prevalence of anemia is above 20% (WHO, 2017). Iron recommendations for pregnant women is 27 mg per day, but for iron-deficient anemia, 60-180 mg per day should be consumed (Woteki, 1993). The management of malaria control, such as IPTp or use of bed nets is important in prevention and control of anemia. The WHO also promotes education on anemia and basic hygiene as preventative measures (WHO, 2017).

2.3.4 Consequences of Anemia During Pregnancy

Common iron-deficient anemia complications are an increased maternal cardiac output, which leads to fatigue; and for infants, risk is increased for low-birth weight, pre-mature delivery, abortion or still birth, and low iron stores at birth (Jung, 2019). If iron-deficient anemia occurs during early pregnancy, risk for preterm and low-birth weight increased by two-to-three times normal risk. Iron-deficient anemia during late pregnancy has shown a risk for lower scores on intelligence, language, gross motor, and attention tests in infant. Infants are at high risk for impaired physical and cognitive development and increased mortality (Rahman, 2016). Anemia is a major factor in maternal deaths, and women with severe anemia are as high as 2.36 times more likely to die than women without severe anemia (Daru, 2018).

Anemia during pregnancy has been associated with increased risk of still birth, neonatal deaths, and low birthweight babies (Patel, 2018). Another study conducted in China showed similar results, with anemia in pregnant women leading to an increase in the prevalence of preterm birth, low birthweight, and neonatal complications. They also found increased prevalence of anemia in lower socioeconomic areas (Lin, 2018) which is consistent with another study in Pakistan (Ayub, 2009).

2.4 Malawi

2.4.1 Maternal and Infant Statistics in Malawi

Malawi is a small country in the southeast region of Sub-Saharan Africa and has a reported population near 18,098,000 as of 2016 (WHO, 2018). The maternal mortality rate was 634 deaths per 100,000 live births in 2015 and has been decreasing by an annual

rate of 2 % since 1990 (UNICEF, 2020). As of 2018, the maternal mortality rate decreased to 439 deaths per 100,000 women but remains one of the worst in the world with the 13th highest rate. The infant mortality rate has steadily decreased from 76 deaths per 1,000 births in 2004 to 35 deaths per 1,000 births in 2018 (UNICEF, 2018).

Within Malawi, there were 5,830,741 reported confirmed cases of Malaria in 2018, with as many as 11,513,684 suspected cases, which is over half of the entire population of 18,600,000 – Roughly one in every four persons in Malawi have a confirmed case of Malaria – of those reported cases, all are due to the Plasmodium species. Of all the confirmed cases of malaria, approximately 4,000 people died. (WHO, 2018).

Malnutrition remains a problem in Malawi for women and children. In addition to high stunting levels, ranging from 30 to 43 percent, 63 percent of children under five are anemia and 33 percent of women are anemic (USAID, 2018). In 2000, UNICEF reported 15.9% of all infants had low birth-weight (LBW), which by definition is weighing less than 2.5 kg. By 2006, 12.5 % of infants were born with LBW, which by comparison is a significant decrease from the year 2000; however, the trend did not stick, in 2010, the rate of LBW was 13.5 % (UNICEF, 2018).

Micronutrient deficiencies are common in Malawi; and in 2001, the Ministry of Health's Micronutrient Survey showed that 60 % of infant under 5 had low vitamin A status. Vitamin A is a vital component of proper immune function and healthy growth and development of infant, and low Vitamin A status lowers the chance of infant surviving serious illnesses. Low vitamin A status is a result of diets low in caloric and nutrient intakes, disease, and chronically malnourished pregnant and breastfeeding women

(UNICEF, 2010). Vitamin A status has improved significantly since 2001; as recently as 2016, vitamin A deficiency prevalence was very low with 4% of infant under 5 and less than 1% of the remaining population. This positive result with vitamin A deficiency is thought to be attributed to vitamin A supplementation in infant. Zinc deficiency was a major micronutrient that had a range between 60 and 66 % of the total population being zinc deficient. Zinc is an important micronutrient for normal infant growth, proper immune function, and healthy pregnancies. Zinc deficiency may be caused by excess losses, inadequate dietary intake, and malabsorption. Iron deficiency was reported to be 22 % in infant under 5, 18.2 % in pregnant women, and 15 % in reproductive age women (UNICEF, 2018). A major challenge for Malawians to receive proper nutrition is household food security status, or the ability of the household to obtain adequate food for the entire family (UNICEF, 2018).

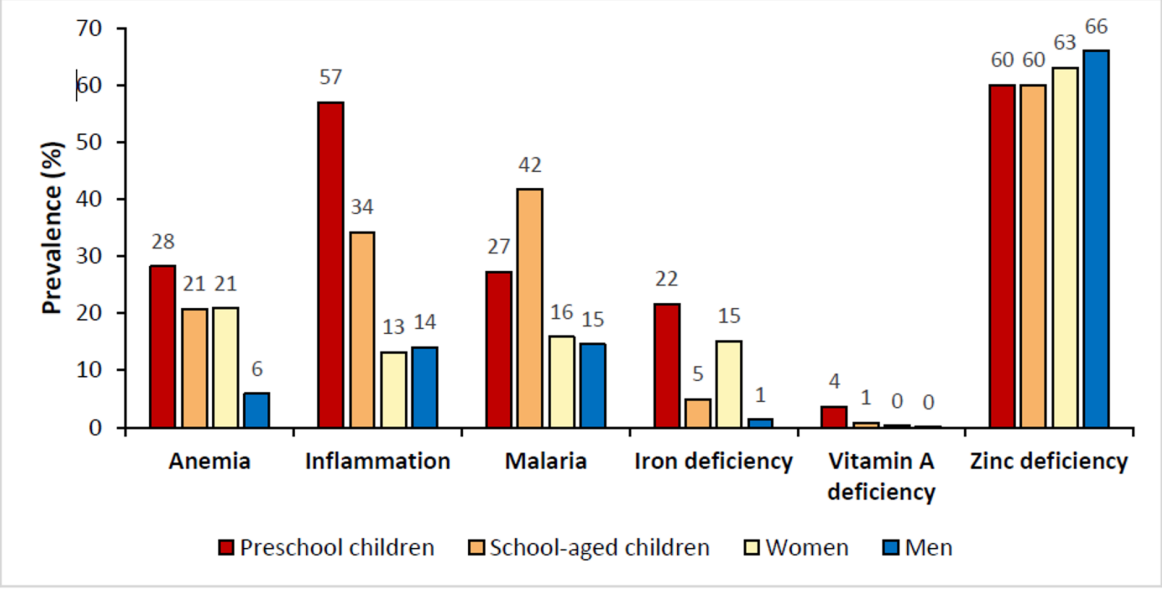

Figure 6: Reported data on Malawi micronutrient status from 2015-2016 (UNICEF, 2018).

2.5 Conclusion

Overall, maternal and infant outcomes continue to be a public health issue in Malawi. Rates of maternal weight gain, on average, are below recommended weekly weight gain during pregnancy and prevalence of anemia is high. Infant outcomes remain low on growth charts and while improving, maternal and infant mortality rates continue to be some of the highest in the world. Increasing energy and micronutrient intakes, through supplementation, relative to increased needs during pregnancy is an ideal approach to improving maternal and infant outcomes; however, more research is necessary to further understand and develop a successful treatment plan for malnourished pregnancies (Papathakis, 2016).

Disease has been shown to play a role in maternal and infant outcomes, particularly malaria in high-transmission risk regions, such as Sub-Saharan Africa. The WHO released guidelines for intermittent preventative treatment in these high-risk areas, recommending women receive a minimum of three IPTp doses for improved infant outcomes (WHO 2020). To our knowledge, there aren't studies that examine the relationship between IPTp dosing and maternal weight gain or MUAC (cm). The current study seeks to find results consistent with the WHO recommendations for improved infant outcomes as well providing additional information on the effects of IPTp dosing on maternal outcomes not previously studied. Anemia and hemoglobin levels are also examined in this study to assess if a relationship exists between IPTp dosing and anemia status and the effects on maternal and infant outcomes.

CHAPTER 3:

METHODS AND MATERIALS

3.1 Methods and Objective

The objective of this research was to determine if the effects of intermittent preventative treatment in pregnancy specific to malaria as well as anemia influenced maternal weight gain and achievement a MUAC ≥ 23 cm during study participation. Additionally, this research serves to assess whether the relationship between IPTp and anemia influences infant health outcomes.

- Research questions to answer through data analysis:
    - Does hx of malaria, malaria during study, anemia and hemoglobin level at enrollment and measured after 10 weeks of enrollment the participants ability to achieve MUAC ≥ 23.0 cm through nutritional intervention?
    - Does anemia severity, mild and moderate, predict weight gain during treatment and achievement of MUAC ≥ 23 cm?
    - The objective of this research was to determine the effects of maternal hemoglobin, anemia, total maternal IPTp doses, maternal IPTp doses in the second and third trimesters, and bed net use on maternal weight gain, recovery from moderate malnutrition (MUAC ≥ 23 cm), and infant outcomes – weight, length, head circumference, and MUAC at birth, six weeks and/or 3 months as well as infant hemoglobin (measured at 3 months), length-for-age (LFA or stunting), and weight-for-age (WFA or underweight)?
- Hypothesis for each research question:

- Women with malaria or anemia are less likely to achieve a MUAC ≥ 23.0 cm during study.
- Maternal hemoglobin, anemia, number of IPTp dose therapy by trimester as well as total number of IPTp doses, illness, and bed net use predict infant outcomes at birth, six weeks, and three months. I expect women that report hx of malaria, malaria during study, lack of bed net use, and have low hemoglobin status to have infants with z-scores at least 1 standard deviation below average for length and weight; as well as head circumference, MUAC at birth, six weeks, and three months and hemoglobin at 3 months.
- IPTp dose therapy in pregnant women is for the prevention of malaria and is common in high-risk regions such as Malawi. Women receiving three or more IPTp doses will have a significantly higher mean MUAC at delivery, and give birth to infants with significantly higher z-scores, head circumference, MUAC, and hemoglobin levels than the results of women receiving less than three IPTp treatment doses, which is consistent with WHO recommendations.

3.2 Study Participants

This research was a secondary analysis of a large assessor-blinded randomized controlled clinical trial. which assessed providing the Malawian standard of care when treating moderate malnutrition in pregnancy against two other nutrition interventions – between March 2014 and December 2015, 15 antenatal clinics in southern Malawi recruiting pregnant women with moderate malnutrition into the Mamachiponde study.

The 15 antenatal clinics were within the Blantyre, Chikhwawa, Mulanje, and Zomba health districts.

Between March 2014 and December 2015, 15 antenatal clinics in southern Malawi recruited pregnant women with moderate malnutrition into the Mamachiponde study. The 15 antenatal clinics were within the Blantyre, Chikhwawa, Mulanje, and Zomba health districts.

Inclusion criteria for the Mamachiponde study:

- ≥ 16 years old
- Hemoglobin ≥ 7.0 g/L
- Moderate malnutrition as defined by a MUAC ≥ 20.6 cm to ≤ 23.0 cm
- Willing to attend antenatal clinical visits every two weeks throughout pregnancy and remain in the area for delivery and until 3 months postpartum
- Consented to participate in the study via written and verbal consent
- Consent to HIV testing if not done previously

Exclusion criteria for the Mamachiponde study:

- Severe malnutrition as defined by a MUAC < 20.6 cm
- Pregnancy complications, such as preeclampsion, hypertension, and gestational diabetes
- < 16 years old
- Severe anemia as defined by a hemoglobin level < 7.0 g/L
- Participation in another nutrition study or nutritional intervention program

In this sub-study, women receiving more 5 doses of IPTp were excluded due to the limited sample size. After the exclusion criteria were taken into account, the total sample size used was 1805.

3.3 Randomization and Blinding

Study participants were randomized into blocks of 60 and received one of the three nutritional intervention treatments (RUSF, CSB-UNIMMAP, or CSB-IFA) via a random number generator, which assigned each subject to a lettered group A, B, or C. Each participant received a pregnancy ID number by picking an envelope with the enclosed ID number contained inside. The research team was also blinded to treatment groups for each study participant. Each participant was given a large opaque bucket, so the researchers were unable to differentiate between different nutritional intervention foods. Nurses involved in the study were aware of which treatment group each participant was in, and this was necessary to provide proper preparation or consumption instructions of intervention foods to each study participant. Upon completion of enrollment measurements and information at the clinics, the study subject would collect her food and be transported by a study driver, who was responsible for matching pregnancy ID number to corresponding treatment group and confirm appropriate intervention food was received. Study participants were unable to be blinded to which treatment group they were a part of because the RUSF, CSB-UNIMMAP, and CSB-IFA are visually different from each other. All data entry and analysis members were blinded to group assignments.

3.4 Study Design

Mamachiponde is a researcher-blinded, randomized controlled trial designed to determine whether a micronutrient-fortified ready-to-use supplementary food (RUSF) or

a fortified corn-soy blended flour (CSB+) plus a multiple micronutrient supplement provided to pregnant women in Malawi, Africa with moderate malnutrition (defined as a MUAC ≥ 20.6 cm to ≥ 23.0 cm) improved recovery from malnutrition and improved both maternal and infant birth outcomes, compared to women receiving the standard of care supplements. Standard of care supplemental food is a fortified CSB flour plus both daily iron and folic acid supplements (CSB-IFA). CSB-IFA is the control intervention. Each treatment food was given to participants in a 14-day supply.

The three intervention groups are described in detail below:

1) RUSF: This treatment provided 900 kcal/day, 38 g protein/day, and 200% of the Recommended Dietary Allowance (RDA) for most micronutrients during pregnancy (except for vitamin A, vitamin B3, folic acid, iodine, magnesium, and calcium, which will remain near 100%). The energy content of RUSF was designed to provide 360-450 calories required during the second and third trimesters of pregnancy, and an additional 450-550 calories to support recovery from moderate malnutrition. Women randomized to this treatment arm did not receive any additional supplements since micronutrients were already in the formulated food.

2) CSB + UNIMMAP: This treatment included 5 kg every two weeks of a corn-soy

blended (CSB+) flour plus a standard antenatal micronutrient tablet (UNIMMAP) that contains 15 micronutrients, which together provided a similar amount of energy, protein, and micronutrients as the RUSF treatment.

3) CSB-IFA: This is the control group, which is the Malawian standard of care treatment for moderate and severe malnutrition during pregnancy. This treatment

included 5 kg CSB every two weeks plus daily iron (60 mg) and folic acid (400 mcg) supplements. Enrolled women visited the clinic every 2 weeks for measurements, health checks and received their two-week supply of treatment food. Women who achieved a MUAC > 23.1 cm were considered 'graduated'. Women whose MUAC remained > 23.1 cm for two consecutive visits no longer received food treatment, however, she still received iron and folic acid supplements. After graduation, women came to clinic every month to ensure no relapse into malnutrition had occurred. If relapse (MUAC <23.1) occurred, mothers went back onto their assigned treatment food/supplements. Women who graduated and/or relapsed were still included in the analysis.

All participants visited the clinic every two weeks for anthropometric measurements, check-up on health status, and receive the next 14-day supply of their nutritional intervention food. Women measured with a MUAC > 23.1 cm at any point during their interventional time were considered as graduated or no longer moderately malnourished. If these women maintained graduated status for two or more consecutive visits, then they no longer received the food supplement, but only received an iron and folic acid supplement. After graduating, women were required to visit the clinic monthly to check for relapse back to malnourished status. If women relapsed, they were given their previously assigned nutritional intervention food again.

3.5 Data Collection

Eligible women that were willing to consent to the study requirements had anthropometric measurements recorded by a study team member at each antenatal clinic visit every two weeks following enrollment. The measurements recorded at each visit included weight, height, MUAC, tricep skinfold (TSF), and blood pressure. Weight was

measured in kilograms with a Detecto Slimpro scale and height was recorded in centimeters with a Seca Stadiometer. MUAC was measured twice using TALC measuring tape - an additional third measurement was taken if the first two measures differed by more than 1 mm and the closest two measurements were used.

3.6 Statistical Analysis

Statistical analysis was done using JMP Pro 13.1 software. Analyses included ANOVA, logistic regression, two sample t-test, Chi-square analysis, and Tukey HSD test. Continuous variables were analyzed by univariately by ANOVA and logistic regression. Chi-square analysis was used to analyze categorical data univariately. For multivariate analyses, continuous variables were looked at using Tukey HSD to determine the effect of an individual variable after taking all the other model variables into account. Categorical multivariate analysis was done by using odds ratios.

Variables created included number of IPTp doses (count), which was created by adding total number of IPTp dose dates for each study participant. IPTp doses up to 10 weeks of enrollment was created to assess whether IPTp dosing during the first 10 weeks of study enrollment had an effect on hemoglobin. IPTp doses by trimester was a variable created by using the estimated age of gestation at enrollment and date of IPTp dose. Mild and moderate hemoglobin (anemia) was created by using a data filter to only include data within the ranges defined – mild defined greater than 7.0 g/dL and less than 9.9 g/dL and moderate greater than or equal to 9.9 g/dL and less than 11.0 g/dL.

CHAPTER 4:

RESULTS

4.1 Baseline: Maternal Characteristics

The age range for the 1805 study participants was 16 to 45 years old. BMI at enrollment ranged from 15.3 kg/m² to 24.1 kg/m². On average, women enrolled in the study at 24.5 weeks gestation. Over 20 % of women reported illness within 2 months of enrollment into the study, of which, 13.5 % are attributed to malaria. Weight gain during treatment in this context refers to weight gain from enrollment to delivery, which on average was low, 0.269 kg, compared to the recommended or expected weight gain during pregnancy of 0.454 kg per week (Rasmussen, 2009). The group enrollment was as followed: 604 (33.5%) were in the RUSF group, 599 (33.2%) in the CSB-UNIMMAP group, and 602 (33.4%) in the CSB-IFA group, p. Additional baseline maternal characteristics are listed in Table 3.

Table 3: Baseline and Follow-up Maternal Characteristics

Characteristic	N = 1805[2]	Mean[1]
Age, years		21.6±5.3
BMI at Enroll (kg/m²)		19.7±1.4
Weeks Gestation at Enroll	1760	24.5±6.4
MUAC at Enroll (cm)		22.3±0.6
MUAC at Delivery (cm)	1505	22.2±0.9
Weeks on Treatment	1799	9.08±5.7
Weight gain during tx (kg)		2.69±2.6
Characteristic		%
Clinic Region		
Blantyre	314	17.4
Chikhwawa	739	41
Mulange	87	4.8
Zomba	665	36.8
Season		

Rainy	568	31.5
Dry	1237	68.5
Achieve MUAC ≥ 23 cm	578	32.7
Hx of Illness	363	20.1
Hx of Malaria	243	13.5
Hx of Iron supplement	996	55.2
Hx of Folate supplement	50	2.8
Malaria during tx	111	6.5
Bed Net Use during tx	1410	78.1

[1]Values expressed as Means and Standard Deviation (SD) or Percentages (%)
[2]Sample size N is 1805 unless otherwise specified in column.

4.2 Unadjusted Maternal Health Outcomes by IPTp Dosing

Women received between zero and four IPTp doses during pregnancy with most receiving two total IPTp doses. Not all doses were received during the study; maternal records were reviewed and, prior IPTp dosing was recorded. Most women received no IPTp (95.6 %) doses during the 1st trimester, consistent with WHO guidelines on IPTp dosing during the 1st trimester (WHO, 2020). IPTp dosing during the 2nd trimester ranged between zero and four doses; however, four doses during the 2nd trimester was only recorded for one woman, with the majority of women receiving 1 dose in the 2nd trimester. The 3rd trimester had a range of zero to three IPTp doses with the majority of women receiving one IPTp dose.

Table 4: Distribution of IPTp Doses Among Women Enrolled

Characteristic	N = 1648	%
Total Number IPTp Doses during pregnancy		
0	43	2.6
1	363	22
2	745	45.2
3	442	26.8
4	55	3.3
IPTp Doses in 1st Trimester		
0	1576	95.6
1	65	3.9
2	7	0.4
IPTp Doses in 2nd Trimester		
0	292	17.7
1	955	58
2	383	23.2
3	17	1
4	1	0.1
IPTp Doses in 3rd Trimester		
0	541	32.8
1	717	43.5
2	343	20.8
3	47	2.9

[1]Values expressed as n and percenaget (%).

Table 5: Maternal Characteristics Compared Across Number of IPTp Doses Reported During Pregnancy

Total Number of IPTp Doses (n = 1648)							
Characteristics	0 n = 43	1 n = 363	2 n = 745	3 n = 442	4 n = 55	Overall	P-value
Subject Age	22.6±0.8[ab]	21.7±0.3[a]	20.7±0.2[b]	21.1±0.2[ab]	20.7±0.6[ab]	21.1±0.1	0.0036
MUAC at Delivery (cm)[4]	22.3±0.2	22.1±0.1	22.2±0.03	22.2±0.05	22.3±0.1	22.2±0.02	0.2274
Wt. Gain during treatment (kg)	2.42±0.4[abc]	1.97±0.1[b]	2.61±0.1[a]	3.34±0.1[c]	3.8±0.3[c]	2.70±0.1	<0.0001
Δ in Hemoglobin (g/dL)[5]	1.30±0.3	1.40±0.1	1.20±0.1	1.13±0.1	0.73±0.2	1.20±0.1	0.0907
Hemoglobin 10 weeks after Enroll (g/dL)[5]	11.43±0.3	11.38±0.1	11.47±0.1	11.32±0.1	10.94±0.2	11.39±0.04	0.1693
	% (n)	% (n)	% (n)	% (n)	% (n)	Overall % (n)	P-value
Malaria during Tx	4.7 (2)	7.4 (27)	7.5 (56)	5.2 (23)	5.5 (3)	6.5 (111)	0.5418
Achieve MUAC ≥ 23 cm	27.9 (12)[a]	26.7 (97)[a]	31.8 (237)[a]	37.8 (167)[b]	47.3 (26)[b]	32.7 (539)	0.0020

[1]Values expressed as Means±SE or percent (n)
[2] P-values calculated using ANOVA or Chi-squared analysis.
[3]Groups that do not share a common letter differ significantly (Tukey's HSD p-value < 0.05).
[4]MUAC at Delivery has sample size n = 1381, with IPTp 0,1,2,3,4 being n = 30, 262, 639, 397, and 53, respectively.
[5]Δ in Hemoglobin and Hemoglobin at 10 Weeks has sample size n = 1442, with IPTp 0, 1,2 ,3 ,4 being n = 36, 279, 659, 413, and 55, respectively.

There was no improvement observed in baseline data for MUAC at enrollment (22.3 cm) and MUAC at delivery (22.2 cm) nor was there any statistical difference by IPTp doses as shown in Table 3. Weight gain during treatment was higher among women who received three or four IPTp doses and lowest in women receiving only one IPTp dose (p-value < 0.0001) as. Shown in Table 3. Hemoglobin increased in all treatment groups but was not different across IPTp doses. Malaria reported during treatment was also not different across IPTp doses. Those receiving three or four IPTp doses during

pregnancy had the highest proportion, 37.8 % and 47.3 % respectively, of women achieving a MUAC ≥ 23 cm (p-value = 0.0020).

4.3 Adjusted Analysis of Maternal Health Outcomes by IPTp Dosing

Maternal health outcomes of weight gain during tx and achievement of MUAC ≥ 23 cm were analyzed by IPTp doses after adjusting for maternal age, clinic region, intervention, hemoglobin at enrollment (g/dL), education, weeks on treatment, and seasonality. Models were fit with and without malaria during tx and malaria was not found to statistically contribute to the model and therefore not included in models and results reported below. As shown in Appendix B, univariate analysis of covariates and IPTp dosing, there was a difference in proportion of women receiving IPTp doses based on clinic region, education level, hemoglobin at enrollment, and weeks on treatment. The highest proportion of women receiving IPTp doses occurred in Zomba (37.0 %) and Chikhwawa (40.4 %) (p-value < 0.0001). Women with 4-6 and 7-8 years of education had the highest proportion of women that received IPTp doses (37.2 %) and (26.1 %) respectively (p-value = 0.0007). Also, women that received treatment the longest received three or four IPTp doses throughout pregnancy (p-value < 0.0001).

4.3.1 Weight Gain During Treatment by Total IPTp Doses

The average weight gain during treatment for all women enrolled in the study was 2.69±2.6 kg with the highest weight gain during treatment seen with women in the RUSF intervention group, 2.89±2.7 kg. After adjustment for age, clinic region, intervention, hemoglobin at enrollment (g/dL), education, weeks on treatment, and seasonality, differences in weight gain during tx across the IPTp doses was observed (p-value = 0.0017).

Women receiving three total IPTp doses had statistically higher weight gains during treatment than those receiving one IPTp dose: (Tukey HSD p-value = 0.0014). The average weight gain was 3.08 [95% CI: 2.88, 3.55] kg for women receiving three IPTp doses during treatment and 2.45 [95% CI: 2.24, 2.93] kg for those receiving one IPTp dose.

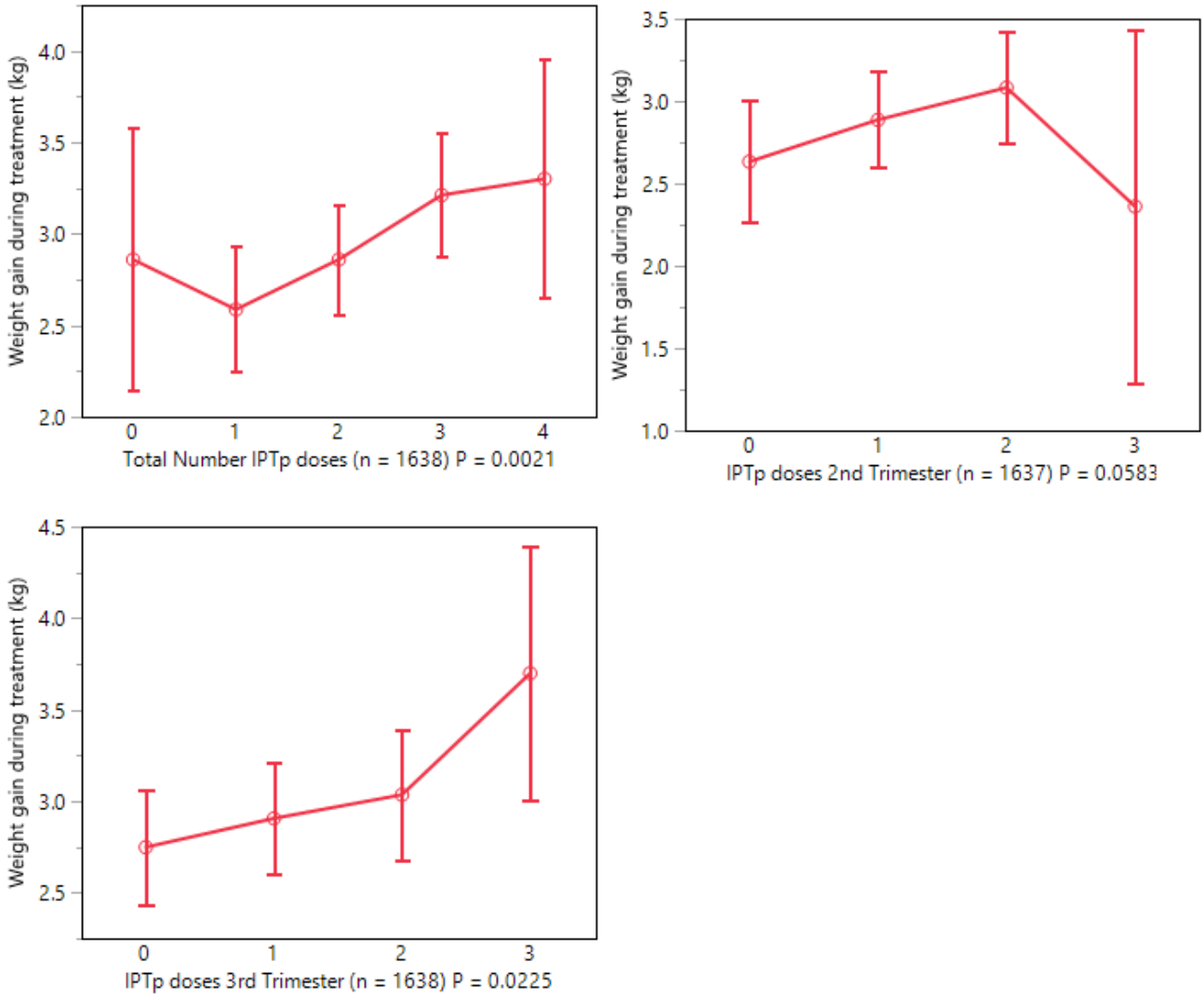

Figure 7: Weight gain during treatment across total number of IPTp doses, doses during the 2nd trimester, and doses during the 3rd trimester.

4.3.2 Weight Gain During Treatment by IPTp Doses in the 2nd Trimester

Adjusted mean weight gain during treatment for zero, one, two, and three IPTp doses given during the 2nd trimester was 2.50 kg [95% CI: 2.26, 3.00], 2.76 kg [95% CI: 2.59, 3.18], 2.95 kg [95% CI: 2.74, 3.42], and 2.27 kg [95% CI: 1.29, 3.43], respectively. Statistically, there is no difference in weight gain during tx by any number of IPTp doses received in the 2nd trimester (p-value = 0.0594). Although, the highest average weight gain before adjustment, 3.14 kg, was observed from women receiving three IPTp doses during the 2nd trimester, there is no statistical difference from those receiving any other number of IPTp doses, and the least squares mean average was the lowest at 2.27 kg for three IPTp doses, which may be attributed to having a low sample size of women receiving three IPTp doses (n = 17). Four IPTp doses during the 2nd trimester has been excluded from Figure 1 due to having n = 1.

4.3.3 Weight Gain During Treatment by IPTp Doses in the 3rd Trimester

IPTp doses given during the 3rd trimester, depicted in Figure 7, show a higher weight gain during tx with increased IPTp doses (p-value = 0.0249) and women receiving three IPTp doses had a statistically higher weight gain during tx than those receiving no IPTp doses (Tukey HSD p-value = 0.0246). There was no difference in weight gain during tx between women receiving zero, one, or two IPTp doses during the 3rd trimester (p-value = 0.1605).

Adjusted mean weight gain by women receiving no IPTp doses was 2.62 kg [95% CI: 2.43, 3.06] while those receiving one, two, or three doses had greater weight gains of 2.78 kg [95% CI: 2.60, 3.21], 2.89 kg [95% CI: 2.68, 3.39], and 3.58 kg [95% CI: 3.01, 4.39], respectively (p-value = 0.0249).

4.3.4 Weight Gain During Treatment and Covariates

There was a difference observed, univariately, for weight gain during tx for each of the following covariates: seasonality, subject age, education, weeks on treatment, and bed net use. Women gained 3.21 kg during tx, on average, during the dry season, compared to 2.74 kg during the rainy season (p-value < 0.0001). Education, maternal age, and weeks on treatment has a positive trendline with weight gain during tx. The older maternal age, the higher weight gain during tx observed. More weeks on treatment, intuitively, led to higher maternal weight gain during tx. Women that reported using a bed net had average weight gain during tx of 3.07 kg compared to 2.65 kg for women that reported not using a bed net (p-value = 0.0020).

4.3.5 Achievement of MUAC ≥ 23 cm by Total Number of IPTp Doses

The highest proportion of women achieving a MUAC ≥ 23 cm was 52.0 % seen in women receiving four total IPTp doses (p-value = 0.0002). Women receiving three total IPTp doses had a higher proportion achieve a MUAC ≥ 23 cm, 39.1 %, compared to those receiving one IPTp doses, 26.8 % (Tukey HSD p-value 0.0020). There was no statistical difference between those receiving three or four IPTp doses (Tukey HSD p-value = 0.4321) and between three or two IPTp doses (Tukey HSD p-value = 0.0998); however, those receiving four IPTp doses had a statistically higher proportion of women achieve a MUAC ≥ 23 cm than those receiving two total IPTp doses (Tukey HSD p-value = 0.0416). The data suggests that total IPTp doses are a strong predictor in women achieving a MUAC ≥ 23 cm (p-value = 0.0002).

Table 6: Achievement of MUAC ≥ 23 cm by IPTp Dosing

	Achievement of MUAC ≥ 23 cm					
IPTp doses	0	1	2	3	4[3]	
	%	%	%	%	%	P-value
Total Number IPTp doses	26.5[ab c]	26.8[c]	32.3[bc]	39.1[ab]	52.0[a]	0.0002
IPTp doses 2nd Trimester	25.0[a]	34.1[b]	35.1[b]	28.3[ab]	-	0.0230
IPTp doses 3rd Trimester	30.0	33.3	36.6	44.1	-	0.0808

[1]The prevalence of achieving MUAC ≥ 23 cm across total number of IPTp doses after adjusting for age, clinic region, intervention, hemoglobin at enrollment (g/dL), weeks on treatment, education, bed net use, and seasonality.
[2]Groups that do not share a common letter differ significantly (p-value < 0.05).
[3]Missing data in column for four IPTp doses is because there are no women within the sample that received four IPTp doses in the 3rd trimester and only one in the 2nd trimester, which was excluded.

4.3.6 Achievement of MUAC ≥ 23 cm by IPTp Doses in the 2nd Trimester

The number of IPTp doses during the 2nd trimester was associated with the proportion of women achieving a MUAC ≥ 23 cm (p-value = 0.0230). Women receiving one (Tukey HSD p-value = 0.0349) or two (Tukey HSD p-value = 0.0493) IPTp doses during the 2nd trimester had a statistically higher proportion of women achieved a MUAC ≥ 23 cm than those receiving no IPTp doses during the 2nd trimester. There was no difference observed for women receiving three IPTp doses during the 2nd trimester compared to women receiving zero, one, or two IPTp doses (Tukey HSD p-value = 0.8350). A small sample size of 18 women received three IPTp doses during the 2nd trimester led to a reduced power.

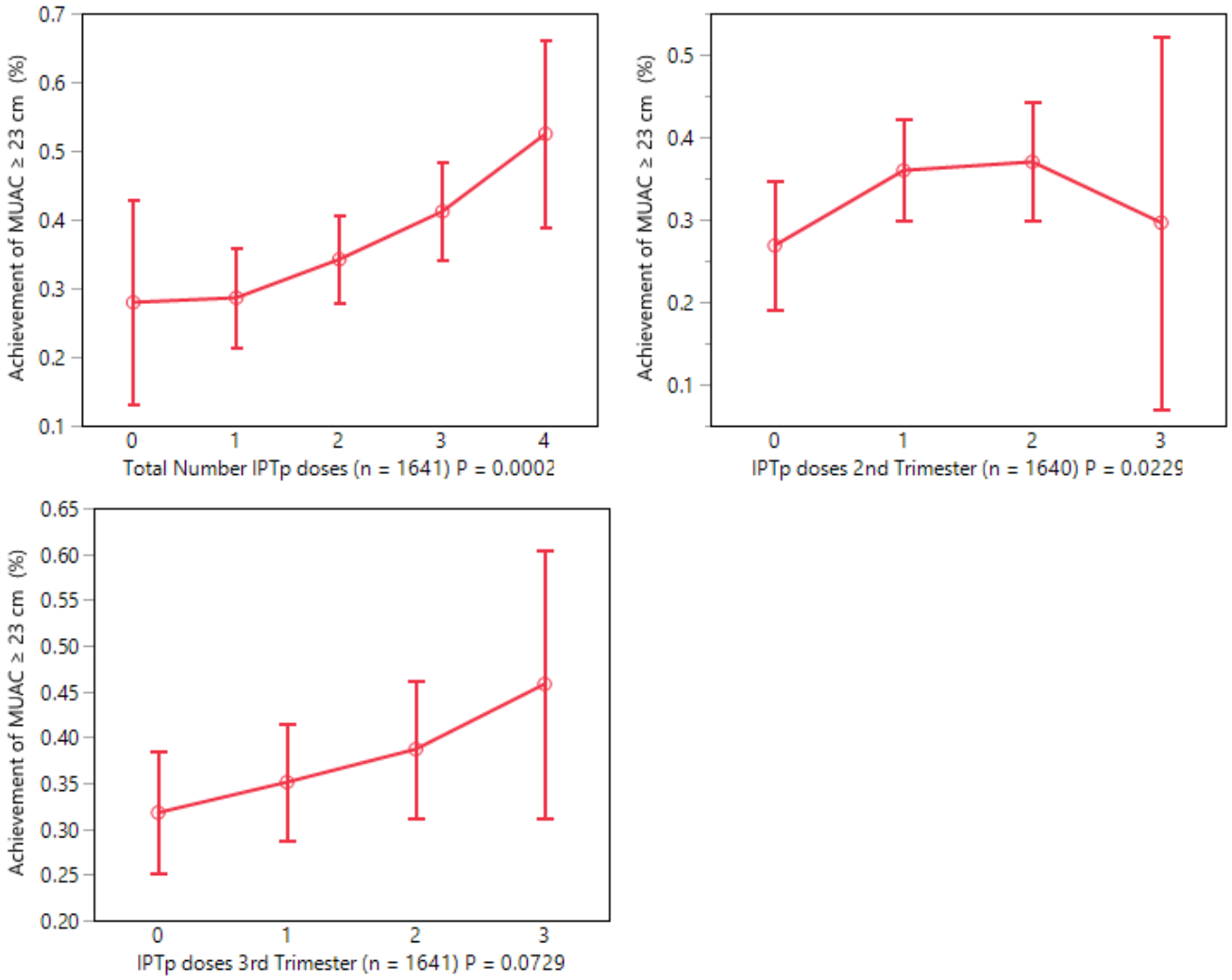

Figure 8: The Prevalence of Achieving MUAC ≥ 23 cm by Total Number of IPTp Doses

4.3.7 Achievement of MUAC ≥ 23 cm by IPTp Doses in the 3rd Trimester

Although there were no statistical differences, after adjustment, in proportions of women achieving a MUAC ≥ 23 cm among women receiving IPTp doses during the 3rd trimester (p-value = 0.0729), women receiving three IPTp doses during the 3rd trimester had the highest proportion achieve a MUAC ≥ 23 cm with 45.8 %. As depicted in Figure 8, the data suggests that four IPTp doses received in the 3rd trimester, helped achieve a

MUAC ≥ 23 cm, but again, there is not statistically significant. The sample size for women receiving 3 IPTp doses during the 3rd trimester was small with n = 47 leading to a low power for comparisons with this group.

4.3.8 Achievement of MUAC ≥ 23 cm and Covariates

Seasonality, weeks on treatment, and bed net use all had statistical differences observed when looking at unadjusted proportions of women achieving a MUAC ≥ 23 cm. A higher proportion of women achieved a MUAC ≥ 23 cm during the dry season than the rainy season (p-value < 0.0001). More women that reported using bed nets (38.47 %) achieved a MUAC ≥ 23 cm than those that reported not using bed nets (32.20 %).

4.4 Maternal Hemoglobin and Anemia

Hemoglobin values at enrollment ranged between 7.0 g/dL and 15.5 g/dL with the median at 10.1 g/dL. On average, improvements in hemoglobin values were observed after receiving treatment for ten weeks. A similar pattern was seen in anemia status with improvements between enrollment and after 10 weeks of treatment. At enrollment, 1280 (70.9 %) women were anemic and 603 (38.5 %) women were anemic at the second measurement as shown in Table 7. The majority of anemic women at time of enrollment were moderately anemic (Hemoglobin between 7.0 and 9.8 g/dL), while the majority of anemic women at the second measurement 10 weeks later were mildly anemic (hemoglobin between 9.9 and 11 g/dL). Hemoglobin values and anemia status at 10 weeks had a sample size of 1566 compared to 1805 at enrollment due to 239 women delivering before they had been enrolled in the study for 10 weeks.

Table 7: Baseline and Follow-up Maternal Hemoglobin and Anemia

	N	Mean	SD
Hemoglobin at Enroll (g/dL)	1805	10.1	1.5
Hemoglobin at 10 weeks (g/dL)	1566	11.3	1.7
Δ in Hemoglobin (g/dL)	1566	1.2	1.8
		N	%
Anemia at Enroll (Hgb < 11 g/dL)		1280	70.9
Mild (Hgb 9.9 – 11 g/dL)		399	22.1
Moderate (Hgb 7.0 – 9.8 g/dL)		677	42.4
Anemia at 10 weeks (Hgb < 11 g/dL)		603	38.5
Mild (Hgb 9.9 – 11 g/dL)		305	51.1
Moderate (Hgb 7.0 – 9.8 g/dL)		295	18.8

[1]Values expressed as Means and Standard Deviation (SD) or percent (n).

4.5 Unadjusted Maternal Health Outcomes by Hemoglobin and Anemia

To investigate whether hemoglobin or anemia had an impact on maternal weight gain during treatment and recovery or achievement of MUAC ≥ 23 cm analysis was done by adjusting for age, clinic region, intervention, education, weeks on treatment, total number IPTp doses, bed net use, and seasonality. Model was fit with and without malaria during tx and malaria was not found to statistically contribute to the model and therefore not included in models. The effect of the changes in anemia status on weight gain and MUAC were modeled by creating a new variable that tracked anemia status across both observed times. The variable had four levels: no anemia, acquired anemia, recovered anemia, and anemic at both measured times.

Women with one to three years of education or secondary and tertiary education had higher hemoglobin values at enrollment than women of other education levels (p-value = 0.0137). Women from the ANCs in Mulange and Zomba had higher hemoglobin

at enrollment values (10.47 and 10.57 g/dL) than women attending ANCs in Chikhwawa and Blantyre (9.80 and 9.84 g/dL) (p-value < 0.0001). When bed net use was reported, hemoglobin at enrollment was higher, 10.17 g/dL, compared to no bed net use, 9.94 g/dL (p-value = 0.0061).

Unadjusted hemoglobin at enrollment was not different across intervention groups (p-value = 0.3108); however, hemoglobin after ten weeks and the change in hemoglobin was different depending on the treatment group each woman was placed in. As shown in Table 8 Hemoglobin after ten weeks was the highest in the CSB-UNIMMAP group (p-value = 0.0214) and the biggest change in hemoglobin was observed in the CSB-IFA group (p-value = 0.0166).

Table 8: Univariate Analysis of Hemoglobin by Intervention Group

	Intervention			
	RUSF	CSB-UNIMMAP	CSB-IFA	P-Value
Hemoglobin at enroll (g/dL)	10.17±0.1	10.21±0.1	10.07±0.1	0.3108
Hemoglobin after 10 weeks (g/dL)	11.18±0.1[b]	11.46±0.1[a]	11.35±0.1[ab]	0.0214
Δ Hgb (g/dL)	1.00±0.1[a]	1.25±0.1[ab]	1.28±0.1[b]	0.0166

[1]Values expressed as Means±Standard Error
[2]Groups that do not share a common letter differ significantly (p-value < 0.05).

4.5.1 Adjusted Maternal Weight Gain During Treatment by Hemoglobin

After adjustment, hemoglobin at enrollment was a significant predictor of maternal weight gain during treatment (p-value = 0.0111). Women entering the study with hemoglobin values above the mean of 10.1 g/dL had a higher weight gain during treatment than the average participant weight gain of 2.69±2.6 kg. A linear relationship was observed; as hemoglobin increased, weight gain during tx increased.

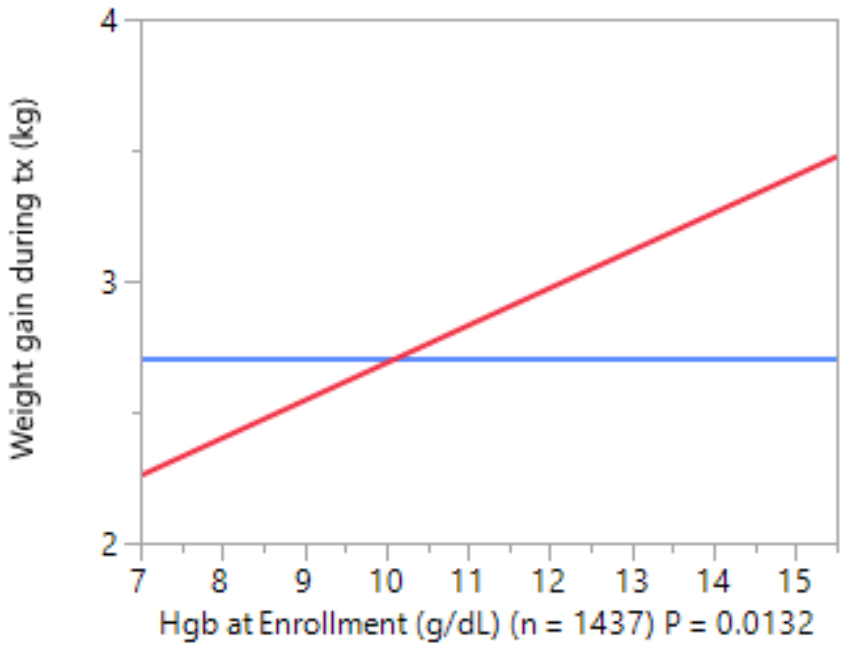

Figure 9: Hemoglobin at Enrollment as a Predictor of Maternal Weight Gain During Tx

After adjusting for covariates, the second hemoglobin measurement, taken after 10 weeks of treatment, was not a significant predictor of maternal weight gain during tx (p-value = 0.7350). The change in hemoglobin however was a significant predictor of weight gain during treatment (p-value = 0.0288). Change in hemoglobin was defined as second measurement minus measurement at enrollment. Figure 10 shows that the greatest weight gain during treatment occured when the hemoglobin at enrollment was lower than hemoglobin at second measurement (negative Δ Hgb).

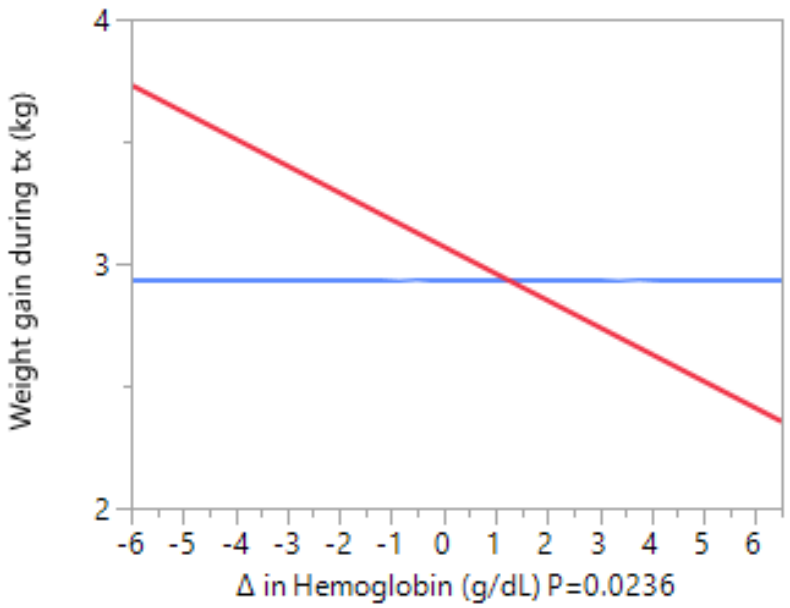

Figure 10: Change in Hemoglobin as a Predictor of Maternal Weight Gain During Tx

4.5.2 Adjusted Maternal Weight Gain During Treatment by Anemia

Anemia at enrollment, defined by hemoglobin < 11 g/dL, was not found to be a predictor of maternal weight gain during tx. Both anemic and non-anemic women at enrollment had average weight gains during treatment of 2.97 kg [anemic, 95% CI: 2.64, 3.30; non-anemic, 95% CI: 2.60, 3.33] (p-value = 0.9754). Anemia at the second measurement was also not found to be a predictor of maternal weight gain during tx (p-value = 0.0829) after adjusting for model covariates: age, clinic region, intervention, education, weeks on treatment, total number IPTp doses, and seasonality.

Moderate anemia, defined as hemoglobin between 7.0 and less than 9.9 g/dL, at enrollment was not a significant predictor of weight gain during treatment (p-value = 0.3484). Nor was mild anemia, defined as hemoglobin greater than or equal to 9.9, but less than 11.0 g/dL, at enrollment (p-value = 0.4599). Neither mild nor moderate anemia

at the second measure was found to be a predictor of weight gain during tx (p-value = 0.5010 and 0.2287, respectively).

4.5.3 Adjusted Achievement of MUAC ≥ 23 cm by Hemoglobin and Anemia

There was no difference in achievement of MUAC ≥ 23 cm observed when using hemoglobin at enrollment, hemoglobin at the second measurement, or the change in hemoglobin to predict the proportion of women that achieve a MUAC ≥ 23 cm (p-value = 0.1683, 0.7025, and 0.1808, respectively).

Similarly, to hemoglobin, anemia at enrollment, mild anemia at enrollment, and moderate anemia at enrollment was not found to have a statistically significant association with the proportion of women that achieve a MUAC ≤ 23 cm (p-values = 0.6445, 0.3165, and 0.9092, respectively). Anemia at 10 weeks, mild anemia at 10 weeks, and moderate anemia at 10 weeks were also not found to predict achievement of MUAC ≤ 23 cm (p-values = 0.1896, 0.5569, and 0.2527, respectively).

4.6 Baseline: Infant Characteristics

Infant hemoglobin was measured at 3 months after birth and ranged between 2.5 g/dL and 15.5 g/dL and 76.59 % (n = 1099) were anemic at 3 months. Infant length at birth ranged from 37.5 cm to 56 cm, at 6 weeks from 40.1 cm to 61.5 cm, and from 46.3 cm to 66 cm at 3 months. Infant weight at birth ranged from 0.6 kg to 4.72 kg, 6 weeks from 1.55 kg to 6.76 kg, and from 2 kg to 8.09 kg at 3 months. Head circumference at birth ranged from 27.9 cm to 39.5 cm, 6 weeks from 31.7 cm to 45.8 cm, and head circumference from 34.1 cm to 49.4 cm at 3 months. Infant MUAC at birth ranged between 6.2 cm and 13 cm, MUAC at 6 weeks ranged from 6.1 cm to 17.3 cm, and MUAC at 3 months ranged from 7 cm to 18.5 cm

Table 9: Baseline and Follow-up Infant Characteristics

Characteristic	N	Mean	SD
Hemoglobin at 3 months (g/dL)	1435	9.94	1.52
Anemia at 3 months	1435	1099[2]	76.59[2]
Length (cm)			
Birth	1431	47.1	2.21
6 weeks	1467	53.7	2.45
3 months	1449	58.2	2.53
Weight (kg)			
Birth	1451	2.74	0.41
6 weeks	1467	4.37	0.64
3 months	1450	5.55	0.75
Head Circumference (cm)			
Birth	1395	34.2	1.49
6 weeks	1460	38	1.46
months	1445	40.3	1.52
MUAC (cm)			
Birth	1426	9.56	0.85
6 weeks	1467	11.8	1.08
3 months	1449	13.1	1.15

[1]SD represents standard deviation.
[2]Anemia at 3 months represented as n and percent.

Table 10: Baseline and Follow-up Infant Z-scores

	N	Z-score	SD
LFA (Stunting)			
Birth	1400	-1.29	1.19
6 weeks	1267	-1.19	1.17
3 months	1255	-1.19	1.15
WFA (Underweight)			
Birth	1453	-1.28	0.98
6 weeks	1267	-0.90	1.06
3 months	1256	-0.87	1.06
WFL (Wasting)			
Birth	1213	-0.48	1.03
6 weeks	1258	0.29	1.03
months	1255	0.19	1.08
HCFA (Small Head)			
Birth	1372	0.05	1.26
6 weeks	1262	0.12	1.15
3 months	1252	0.23	1.17

	N	%
Stunted (LFA < -2)		
Birth	287	19.75
6 Weeks	146	11.52
3 Months	152	12.10
Underweight (WFA < -2)		
Birth	325	23.21
6 Weeks	260	20.52
3 Months	266	21.20
Wasting (WFL < -2)		
Birth	68	5.61
6 Weeks	23	1.83
3 Months	31	2.47
Small Head (HCFA < -2)		
Birth	62	4.52
6 Weeks	40	3.17
3 Months	34	2.72

[1]Values expressed as Means and Standard Deviation (SD) or percent (n).

4.7 Unadjusted Infant Health Outcomes by IPTp

Table 11: Unadjusted Infant Health Outcomes Across Number of IPTp Doses Reported

Characteristics	Number of IPTp Doses Reported (n = 1347)					Overall	P-value
	0 n = 32	1 n = 255	2 n = 621	3 n = 389	4 n = 50		
Hemoglobin at 3 Months (g/dL)	10.05±0.3	9.87±0.1	9.89±0.1	10.09±0.1	10.07±0.2	9.96±0.1	0.2868
Anemia at 3 Months	18 (56.3)	196 (79.4)	464 (76.3)	286 (74.5)	41 (80.4)	1099 (76.6)	0.8029
Length (cm)							
Birth	47.69±0.4^{ab}	46.75±0.2^{b}	47.12±0.1^{ab}	47.30±0.1^{a}	47.26±0.3^{ab}	47.12±0.2	0.0198
6 Weeks	53.21±0.5^{ab}	53.21±0.2^{b}	53.84±0.1^{a}	54.0±0.1^{a}	54.20±0.3^{ab}	53.78±0.2	0.0008
3 Months	58.39±0.4^{ab}	57.86±0.2^{b}	58.29±0.1^{ab}	58.51±0.1^{a}	58.57±0.3^{ab}	58.29±0.2	0.0234
Weight (kg)							
Birth	2.87±0.1^{a}	2.66±0.03^{b}	2.73±0.02^{ab}	2.79±0.02^{a}	2.81±0.05^{ab}	2.74±0.02	0.0007
6 Weeks	4.42±0.1^{ab}	4.24±0.04^{b}	4.39±0.02^{a}	4.46±0.03^{a}	4.63±0.1^{a}	4.39±0.1	<0.0001
3 Months	5.72±0.1^{ab}	5.45±0.1^{b}	5.56±0.03^{ab}	5.65±0.04^{a}	5.82±0.1^{a}	5.58±0.1	0.0012
Head Circumference (cm)							
Birth	34.77±0.3^{a}	33.95±0.1^{b}	34.18±0.1^{ab}	34.42±0.1^{a}	34.43±0.2^{ab}	34.24±0.1	0.0004
6 Weeks	37.85±0.3^{ab}	37.81±0.1^{b}	38.07±0.1^{ab}	38.20±0.1^{a}	38.61±0.2^{a}	38.07±0.1	0.0010
3 Months	40.50±0.3^{ab}	40.14±0.1^{b}	40.30±0.1^{ab}	40.55±0.1^{a}	40.77±0.2^{a}	40.37±0.1	0.0022
MUAC (cm)							

Birth	9.76±0.1[ab]	9.41±0.1[b]	9.54±0.03[ab]	9.66±0.04[a]	9.71±0.1[ab]	9.56±0.1	0.0026
6 Weeks	11.73±0.2[ab]	11.62±0.1[b]	11.89±0.04[a]	11.99±0.1[a]	12.26±0.1[a]	11.88±0.1	<0.0001
3 Months	13.25±0.2	12.92±0.1	13.04±0.04	13.20±0.1	13.36±0.1	13.08±0.1	0.0068

[1]Values expressed as Means±SE
[2] P-values calculated using ANOVA (continuous values).
[3]Groups that do not share a common letter differ significantly (Tukey's HSD p-value < 0.05).

Table 12: Unadjusted Infant Health Outcomes Compared Across Number of IPTp Doses Reported in Z-scores.

	Number of IPTp Doses Reported						
	0	1	2	3	4	Overall	P-value
LFA							
Birth	-1.07±0.2[ab]	-1.50±0.1[b]	-1.29±0.05[ab]	-1.17±0.1[a]	-1.18±0.2[ab]	-1.28±0.1	0.0157
6 Weeks	-1.28±0.2[ab]	-1.44±0.1[b]	-1.17±0.1[a]	-1.03±0.1[a]	-1.02±0.2[ab]	-1.18±0.1	0.0019
3 Months	-1.63±0.2[ab]	-1.40±0.1[b]	-1.19±0.05[ab]	1.00±0.1[a]	-0.92±0.2[ab]	-1.16±0.1	0.0002
WFA							
Birth	-1.04±0.2[ab]	-1.47±0.1[b]	-1.28±0.04[ab]	-1.15±0.04[a]	-1.04±0.1[ab]	-1.27±0.01	0.0003
6 Weeks	-0.82±0.2[ab]	-1.17±0.1[b]	-0.88±0.04[a]	-0.74±0.1[a]	-0.48±0.2[a]	-0.88±0.1	< 0.0001
3 Months	-0.87±0.2[ab]	-1.08±0.1[b]	-0.87±0.04[ab]	-0.68±0.1[a]	-0.45±0.2[a]	-0.83±0.1	< 0.0001
WFL							
Birth	-0.29±0.2	-0.46±0.1	-0.53±0.04	-0.44±0.1	-0.29±0.2	-0.48±0.2	0.3723
6 Weeks	0.52±0.2[ab]	0.20±0.1[b]	0.28±0.04[ab]	0.32±0.1[ab]	0.69±0.2[a]	0.30±0.1	0.0377
3 Months	0.69±0.2	0.14±0.1	0.19±0.05	0.22±0.1	0.45±0.2	0.21±0.1	0.0937

HCFA							
Birth	0.37±0.2[ab]	-0.19±0.1[b]	0.03±0.1[ab]	0.25±0.1[a]	0.24±0.2[ab]	0.07±0.1	0.0004
6 Weeks	-0.03±0.2[ab]	-0.04±0.1[b]	0.13±0.04[ab]	0.28±0.1[a]	0.52±0.2[a]	0.15±0.1	0.0027
3 Months	-0.003±0.2[ab]	0.05±0.1[b]	0.18±0.1[b]	0.49±0.1[a]	0.69±0.2[a]	0.27±0.1	< 0.0001

[1]Values expressed as Means±SE
[2] P-values calculated using ANOVA (continuous values).
[3]Groups that do not share a common letter differ significantly (Tukey's HSD p-value < 0.05).

4.7.1 Infant Health Outcomes by Maternal IPTp Dosing During Pregnancy

The following sub-sections of this study examine the relationship between total number of maternal IPTp doses received during pregnancy on infant outcomes: hemoglobin at three months, anemia at three months, length, weight, head circumference, and MUAC. The results in the below sections refer to adjusted mean values with covariates being maternal MUAC at delivery, maternal malaria during treatment, maternal age, clinic region, maternal weight gain during treatment, maternal hemoglobin at enrollment, intervention, bed net use, and seasonality.

Unadjusted infant hemoglobin and anemia at three months was different when comparing values by the following covariates: maternal hemoglobin at enrollment, maternal weight gain during treatment, maternal age, clinic region, maternal MUAC at delivery, and seasonality. Maternal hemoglobin at enrollment had a statistical effect on infant length and weight, but not head circumference or MUAC. Infant length, weight, head circumference, and MUAC were all statistically different by maternal weight gain during treatment and MUAC at delivery. Unadjusted analysis of maternal age showed it was a significant predictor of infant length, weight, and MUAC.

4.7.2 Adjusted Infant Hemoglobin and Anemia at 3 Months by IPTp

Total number of maternal IPTp doses during pregnancy had no statistically significant effect on infant hemoglobin at 3 months (p-value = 0.7810). The average hemoglobin at 3 months for zero, one, two, three, and four total number of IPTp doses given to the mother during pregnancy are shown in Table 13. There was no statistical difference observed in infant anemia at 3 months, defined by hemoglobin values below 11.0 g/dL, across total number of IPTp doses (p-value 0.8209).

Table 13: Infant Hemoglobin and Anemia by Number of Total Number IPTp Doses

Infant Characteristics	Total Number of IPTp doses					P-value
	0 n = 32	1 n = 255	2 n = 621	3 n = 389	4 n = 50	
Hemoglobin at 3 Months (g/dL)	10.00±0.2	10.00±0.1	9.92±0.1	10.05±0.1	9.90±0.2	0.7810
Anemia at 3 Months (Hgb > 11 g/dL)	18 (56.3)	196 (79.4)	464 (76.3)	286 (74.5)	41 (80.4)	0.8209

[1] Values are adjusted means±SE or n (%)

4.7.3 Adjusted Infant Length by IPTp

After adjusting for maternal MUAC at delivery, malaria during treatment, maternal age, clinic region, maternal weight gain during treatment, maternal hemoglobin at enrollment, intervention, and seasonality, there was no statistically significant effect on infant length at birth or 3 months by total number of maternal IPTp doses (p-value = 0.1752, 0.1498). There is some evidence that women receiving two or three IPTp doses had infants with a statistically significant higher length at six weeks than those with mothers receiving one IPTp doses (Tukey HSD p-value = 0.0247 and 0.0090, respectively). The mean average infant length at six weeks for two IPTp is shown in Table 14. No difference from total number of IPTp doses was observed with LFA (p-value = 0.1040). As seen in Table 15, infants born to women that received three IPTp doses during pregnancy had higher LFA z-scores at six weeks and three months than infants born to women who received one IPTp dose (Tukey HSD p-value = 0.0056 and 0.0276, respectively).

Table 14: Infant length by total number IPTp doses

	Number of IPTp doses					
	0	1	2	3	4	P-value
	Mean	Mean	Mean	Mean	Mean	
Length (cm)						
Birth	47.68±0.4	46.80±0.2	47.09±0.1	47.17±0.2	47.11±0.3	0.1752
6 Weeks	53.78±0.5^{ab}	53.23±0.2^{b}	53.79±0.2^{a}	53.90±0.2^{a}	53.90±0.4^{ab}	0.0163
3 Months	58.72±0.5	58.00±0.2	58.37±0.2	58.48±0.2	58.44±0.4	0.1498

[1]Values are adjusted means±SE.
[2]Groups that do not share a common letter differ significantly (Tukey's HSD p-value < 0.05)

Table 15: Infant LFA (Z-scores) by Total IPTp Doses

	Number of IPTp doses					
	0	1	2	3	4	P-value
	Mean	Mean	Mean	Mean	Mean	
LFA (Z-score)						
Birth	-1.22±0.3	-1.53±0.1	-1.31±0.1	-1.24±0.1	-	0.0545
6 Weeks	-1.30±0.3^{ab}	-1.51±0.1^{b}	-1.22±0.1^{a}	-1.13±0.1^{a}	-1.25±0.2^{ab}	0.0147
3 Months	-1.43±0.3^{ab}	-1.40±0.1^{b}	-1.20±0.1^{ab}	-1.09±0.1^{a}	-1.09±0.2^{ab}	0.0487

[1]Values are adjusted means±SE.
[2]Groups that do not share a common letter differ significantly (Tukey's HSD p-value < 0.05).

4.7.4 Adjusted Infant Weight by IPTp

Total number of IPTp doses was a statistically significant predictor of infant weight at birth, six weeks, and three months before and after adjustment. Infant weight at birth was statistically higher in infants from women receiving three total IPTp doses than one IPTp dose (Tukey HSD p-value = 0.0286).

Women receiving two, three, or four IPTp doses gave birth to infants with statistically higher weights at six weeks than women receiving one IPTp dose (Tukey HSD p-value = 0.0469, 0.0063, and 0.0271, respectively). Infants from women receiving one IPTp dose during pregnancy had an average weight at six weeks of 4.26 [95% CI: 4.10, 4.32] kg compared to average weight at six weeks of 4.39 [95% CI: 4.26, 4.42] kg,

4.48 [95% CI: 4.29, 4.49] kg, and 4.61 [95% CI: 4.31, 4.69] kg for two, three, and four IPTp doses, respectively. Women receiving no IPTp doses during pregnancy gave birth to infants with statistically higher weight at 3 months than infants from women receiving one IPTp dose (Tukey HSD p-value = 0.0471).

IPTp dosing was a significant predictor of infant WFA (p-value = 0.0024). Infants from mothers that received two or three IPTp doses had statistically higher WFA z-scores, -1.32±0.1 and -1.22±0.1, than infants whose mothers received one IPTp dose, -1.50±0.1 (Tukey HSD p-value = 0.0343 and 0.0016, respectively). Seen in Table 17, infant WFA at six weeks was significantly improved in infants whose mother's received two, three, or four IPTp doses during pregnancy than infants whose mother's received one IPTp dose (Tukey HSD P-values = 0.0049, 0.0005, and 0.0110, respectively). At three months, three IPTp doses had a significantly improved z-score than one IPT dose (Tukey HSD p-value = 0.0083). Although not statistically significant, a positive trendline is observed from one IPTp dose to four IPTp doses, values improving with each additional dose.

Proportionately, WFA z-scores at birth below two standard deviations of the mean (> -2) were statistically different by IPTp dosing when considering the overall p-value of 0.0458; however, Tukey HSD was unable to identify any difference between individual groups. 11.1 % of infants from women receiving no IPTp doses during pregnancy had WFA z-scores below -2 compared to 26.4 %, 22.3 %, and 18.0 % of infants whose mothers received one, two, or three IPTp doses. While no individual pairing is statistically significant, there is almost 8 % fewer infants stunting from women receiving three IPTp doses compared to one IPTp dose.

Table 16: Infant Weight by Total Number IPTp Doses

	Number of IPTp doses					
	0	1	2	3	4	P-value
	n =					
Weight (kg)						
Birth	2.84±0.1[ab]	2.64±0.03[b]	2.70±0.03[ab]	2.74±0.03[a]	2.75±0.1[ab]	0.0117
6 Weeks	4.48±0.1[ab]	4.26±0.1[b]	4.39±0.04[a]	4.48±0.05[a]	4.61±0.1[a]	0.0041
3 Months	5.84±0.2[a]	5.43±0.1[b]	5.50±0.1[ab]	5.56±0.1[ab]	5.67±0.1[ab]	0.0133

[1] Values are adjusted means±SE.
[2]Groups that do not share a common letter differ significantly (Tukey's HSD p-value < 0.05).

Table 17: Infant WFA (Z-scores) by Total IPTp Doses

	Number of IPTp doses					
	0	1	2	3	4	P-value
WFA (Z-score)						
Birth	-1.13±0.2[ab]	-1.52±0.1[b]	-1.32±0.1[a]	-1.22±0.1[a]	-	0.0024
6 Weeks	-0.83±0.3[ab]	-1.25±0.1[b]	-0.93±0.1[a]	-0.85±0.1[a]	-0.68±0.2[a]	0.0004
3 Months	-0.58±0.3[ab]	-1.08±0.1[b]	-0.90±0.1[ab]	-0.76±0.1[a]	-0.63±0.2[ab]	0.0050

[1]Values are adjusted means±SE.
[2]Groups that do not share a common letter differ significantly (Tukey's HSD p-value < 0.05).

4.7.5 Adjusted Infant Head Circumference by IPTp

Total number of maternal IPTp doses during pregnancy was not found to be a statistically significant predictor of infant head circumference at birth (p-value = 0.8661). Three total maternal IPTp doses during pregnancy had a statistically higher head circumference at birth than one IPTp dose (Tukey HSD p-value = 0.0227). Although the effect of total number of IPTp doses on head circumference at six weeks was found to have a statistical p-value below 0.05 (p-value = 0.0402); there was no difference between any observations for head circumference at six weeks as shown in Table 18. There was a difference observed for total number of IPTp doses and head circumference measured at three months (p-value 0.0205). Women receiving three doses of IPTp gave birth to

infants with statistically higher head circumference values at three months than infants from mothers who were administered one total IPTp dose during pregnancy (Tukey HSD p-value = 0.0353). There was no observable statistical difference between infant head circumference at three months between zero, two, or four IPTp doses.

Infant HCFA z-scores at birth were different by IPTp dosing after adjustment for covariates (p-value = 0.0064) as shown in Table 19. Infants whose mothers received three IPTp doses during pregnancy had improved z-scores compared to infants whose mothers received one IPTp dose during pregnancy (Tukey HSD p-value = 0.0035). At six weeks, no statistical difference was observed for HCFA z-scored by IPTp doses; however, a positive trend was observed, or an improved z-score with each additional IPTp dose was seen. At three months, a similar pattern was observed for HCFA z-scores as at birth; three IPTp doses had improved z-scores than one IPTp dose.

Table 18: Infant Head Circumference by Total Number of IPTp Doses

Infant Characteristics	Number of IPTp doses 0	1	2	3	4	P-value
	%	%	%	%	%	
Head Circumference						
Birth	34.73±0.3[ab]	33.94±0.1[b]	34.13±0.1[ab]	34.32±0.1[a]	34.26±0.2[ab]	0.0086
6 Weeks	37.73±0.3	37.70±0.1	37.97±0.1	38.00±0.1	38.27±0.2	0.0402
3 Months	40.66±0.3[ab]	40.07±0.1[b]	40.27±0.1[ab]	40.43±0.1[a]	40.57±0.2[ab]	0.0205

[1] Values are adjusted means±SE

[2]Groups that do not share a common letter differ significantly (Tukey's HSD p-value < 0.05).

Table 19: Infant HCFA (Z-scores) by Total IPTp Doses

	Number of IPTp doses					
	0	1	2	3	4	P-value
HCFA (Z-score)						
Birth	0.19±0.3[ab]	-0.20±0.1[b]	0.01±0.1[ab]	0.18±0.1[a]	-	0.0064
6 Weeks	-0.10±0.3[ab]	-0.13±0.1[b]	0.09±0.1[a]	0.14±0.1[a]	0.29±0.2[a]	0.0586
3 Months	0.02±0.3[ab]	-0.01±0.1[b]	0.20±0.1[ab]	0.41±0.1[a]	0.50±0.2[ab]	0.0014

[1]Values are adjusted means±SE.
[2]Groups that do not share a common letter differ significantly (Tukey's HSD p-value < 0.05).

4.7.6 Adjusted Infant MUAC by IPTp

Significant differences in infant MUAC by total number of maternal IPTp doses during pregnancy were seen at birth and six weeks, but not three months. At birth, infants from mothers who received three total IPTp doses had a statistically higher MUAC at birth of 9.55 [95% CI: 9.43, 9.68] cm compared to those from women receiving one total IPTp dose, 9.33 [95% CI: 9.18, 9.47] cm (Tukey HSD p-value = 0.0096). Infant MUAC at six weeks was statistically higher in infants with mothers that received two, three, or four total IPTp does compared to women that received one total IPTp dose during pregnancy (Tukey HSD p-value = 0.0343, 0.0073, and 0.0259, respectively). Infants from women receiving one IPTp dose during pregnancy had average MUAC values at six weeks of 11.47 [95% CI: 11.29, 11.65] cm. Although total number of IPTp doses found to be a statistically significant predictor of infant MUAC at 3 months (p-value = 0.0327), there was no evidence of a difference in predicting infant MUAC at 3 months whether the mother had between zero and four IPTp doses.

Table 20: Infant MUAC by Total Number of IPTp Doses

Infant Characteristics	Number of IPTp doses					P-value
	0	1	2	3	4	
	%	%	%	%	%	
MUAC						
Birth	9.58±0.2[ab]	9.33±0.1[b]	9.47±0.1[ab]	9.55±0.1[a]	9.58±0.1[ab]	0.0289
6 Weeks	11.53±0.2[ab]	11.47±0.1[b]	11.71±0.1[a]	11.77±0.1[a]	11.97±0.2[a]	0.0037
3 Months	13.32±0.2	12.77±0.1	12.82±0.1	12.96±0.1	13.07±0.2	0.0327

[1]Values are adjusted means±SE
[2]Groups that do not share a common letter differ significantly (Tukey's HSD p-value < 0.05).

4.7.7 Summary of Infant Outcomes by IPTp

Total number of IPTp doses received during pregnancy did not have a statistical effect on infant hemoglobin or anemia at three months. Infant length at six weeks was higher in infants from mothers that received two or three IPTp doses compared to mothers that received one IPTp dose (p-value = 0.0218). A p-value below 0.05 by total number of IPTp doses was observed for infant weight, head circumference, and MUAC at birth, six weeks, and three months. With a few exceptions described in the sections above, the general trend observed was women receiving three IPTp doses gave birth to infants with higher weight, head circumference, and MUAC values than infants from mothers that received one IPTp dose during pregnancy.

4.8 Adjusted Infant Health Outcomes by Hemoglobin/Anemia

Only infant characteristics at three months were considered when looking at the effect of infant hemoglobin and anemia at three months. Observable differences were seen in infant length at three months by infant hemoglobin at three months (p-value = 0.0266). A positive trendline was observed, the higher infant hemoglobin values at three months, the longer infant lengths at three months were on average. No differences were

observed with infant weight, head circumference, or MUAC at three months (p-value = 0.0797, 0.8034, and 0.1659, respectively). In terms of infant anemia, defined by hemoglobin below 11.0 g/dL, there was no effect of infant anemia observed for infant length, weight, head circumference, or MUAC at three months.

4.9 Adjusted Infant Health Outcomes by Maternal Hemoglobin and Anemia

Maternal hemoglobin at study enrollment were found to have a statistically significant effect on infant hemoglobin at three months (p-value = 0.0071). A positive trendline was observed, meaning that higher maternal hemoglobin values at enrollment yielded higher infant hemoglobin values, on average, at three months. Maternal hemoglobin measured at 10 weeks after enrollment was not found to be a predictor of infant hemoglobin at three months (p-value = 0.3501); therefore, maternal hemoglobin at enrollment was a stronger predictor of infant hemoglobin at three months. No observable differences were seen with change in maternal hemoglobin (second measurement minus first measurement) on infant hemoglobin at three months.

Maternal hemoglobin at enrollment had no statistical effect on any infant outcomes analyzed, see Table 21. Significant differences were observed in length (cm) at birth, six weeks, and three months, as well as head circumference (cm) at birth and six weeks, when using the second maternal hemoglobin measurement, taken 10 weeks after enrollment. A positive trendline was observed; a higher maternal hemoglobin at the second measurement yielded, on average, higher infant length at birth, six weeks, and three months. Similarly, a higher maternal hemoglobin at the second measurement, yielded, on average, higher infant head circumference at birth and six weeks. Although

length was statistically impacted by the second maternal hemoglobin measurement, there was no difference observed in LFA (p-value = 0.1066)

Table 21: Infant Outcomes by Maternal Hemoglobin at Enrollment and at 10 Weeks After Enrollment

	Maternal Hgb at Enroll (n = 1225) P-value	Maternal Hgb at 10 weeks (n = 1158) P-value
Length (cm)		
Birth	0.1118	0.0294
6 Weeks	0.9524	0.0452
3 Months	0.9277	0.0139
Weight (kg)		
Birth	0.7345	0.431
6 Weeks	0.428	0.108
3 Months	0.5073	0.4502
Head Circumference (cm)		
Birth	0.5378	0.0306
6 Weeks	0.7756	0.0349
3 Months	0.9649	0.0633
MUAC (cm)		
Birth	0.7534	0.4781
6 Weeks	0.4449	0.3358
3 Months	0.6422	0.6241

[1]Hgb represents hemoglobin (g/dL).
[2]P-values < 0.05 represent a statistical difference observed based on student's t-test.

4.10 Adjusted Infant Health Outcomes by Maternal Anemia

No statistical differences were observed for infant hemoglobin at three months by maternal anemia, regardless of anemia at enrollment or anemia at second measurement during treatment (p-value = 0.0995 and 0.6767, respectively). Similarly, there is no evidence that either maternal anemia at enrollment or at 10 weeks were statistically significant predictors of any infant outcomes measured. See Table 12 below for more details. When considering severity of anemia, shown in Table 23, infants whose mothers

had moderate anemia at enrollment had statistically lower length at six weeks and three months as well as lower weight and head circumference at six weeks than their respective counterparts. As far as severity of anemia after ten weeks of study participation, only infant MUAC at three months was statistically different, lower in this case, in infants whose mothers were moderately anemic after ten weeks (p-value = 0.0220).

Table 22: Adjusted Infant Health Outcomes by Maternal Anemia at Enrollment and at 10 Weeks After Enrollment

	Maternal Anemia at Enroll P-value	Maternal Anemia at 10 weeks P-value
Length (cm)		
Birth	0.8841	0.0816
6 Weeks	0.4411	0.2658
3 Months	0.9179	0.2183
Weight (kg)		
Birth	0.3486	0.7269
6 Weeks	0.4585	0.8778
3 Months	0.9115	0.3489
Head Circumference (cm)		
Birth	0.981	0.2294
6 Weeks	0.4668	0.3595
3 Months	0.9027	0.6137
MUAC (cm)		
Birth	0.5723	0.1987
6 Weeks	0.4297	0.7939
3 Months	0.9502	0.0641

[1]Anemia is defined as hemoglobin values below 11.0 g/dL.
[2]P-values < 0.05 represent a statistical difference observed based on student's t-tes

Table 23: Adjusted Infant Health Outcomes by Severity of Maternal Anemia at Enrollment and at 10 Weeks After Enrollment

	Maternal Anemia at Enroll		Maternal Anemia at 10 weeks	
	Mild P-value	Moderate P-value	Mild P-value	Moderate P-value
Length (cm)				
Birth	0.1172	0.1491	0.2275	0.5124
6 Weeks	0.0509	0.0062	0.9120	0.6363
3 Months	0.3123	0.0241	0.2851	0.1351
Weight (kg)				
Birth	0.0651	0.3219	0.3732	0.6939
6 Weeks	0.3448	0.0385	0.2284	0.2670
3 Months	0.4016	0.2635	0.3218	0.0664

Head Circumference (cm)				
Birth	0.0540	0.1143	0.2594	0.4176
6 Weeks	0.2384	0.0415	0.2212	0.2481
3 Months	0.5680	0.3236	0.2748	0.0849
MUAC (cm)				
Birth	0.4001	0.8292	0.9830	0.6026
6 Weeks	0.8027	0.7071	0.1651	0.1125
3 Months	0.2123	0.9332	0.1788	0.0220

[1]Anemia is defined as hemoglobin values below 11.0 g/dL.
[2]P-values < 0.05 represent a statistical difference observed based on student's t-test.

CHAPTER 5:

DISCUSSION

In the current secondary analysis of data from a randomized controlled clinical trial, we found that intermittent preventative treatment in pregnancy (IPTp) with sulfadoxine-pyrimethamine (SP) was a significant predictor of maternal weight gain during treatment and achievement of MUAC ≥ 23 cm, as well as several infant health outcomes. We also found that hemoglobin, at enrollment and measured after 10 weeks of enrollment, was a significant predictor of maternal and infant outcomes observed in the study. There was no evidence that IPTp usage reduced the prevalence of malaria during treatment as shown in Table 5.

5.1 IPTp

Four total number of IPTp doses had the highest overall maternal weight gain during treatment, followed by two and three total IPTp doses. Women receiving one IPTp dose during pregnancy had the lowest weight gains. As far as dose timing, it was more important to consider total number of IPTp doses during pregnancy rather than the timing of the doses. Both in the 2nd and 3rd trimester, the data would suggest that the more doses received during either trimester had a positive effect on weight gain during treatment, but there was no statistical data in 2nd trimester when distinguishing between the effect of women receiving one, two, or three IPTp doses, and there were only statistically significant differences observed between three IPTp and one IPTp dose during the 3rd trimester.

A statistical difference was observed in the proportion of women achieving a MUAC ≥ 23 cm by total number of IPTp doses. The greatest proportion of women, after

adjustment, that achieved a MUAC ≥ 23 cm was seen in women receiving four (47.3 %) and three (37.8 %) total IPTp doses during pregnancy. Women receiving only one IPTp dose during pregnancy had the lowest proportion of women that achieved a MUAC ≥ 23 cm. Results would suggest that total number of IPTp doses correlates closely with achieving a MUAC ≥ 23 cm during pregnancy with recommendations being three or four total IPTp doses during pregnancy and one or two doses being received during the second trimester. IPTp doses received during the third trimester had a positive trendline correlating higher proportions of women that achieved a MUAC ≥ 23 cm with more IPTp doses received; however, there was no statistically significant evidence that there was any difference observed.

We also observed improvements in infant length, weight, head circumference, and MUAC at multiple time points by IPTp dosing. Most notably, the greatest improvements were seen in infant weight at birth, six weeks, and three months. The general trend observed was that women receiving three IPTp doses during pregnancy gave birth to infants with improved anthropometrics compared to infants born from women receiving one IPTp dose during pregnancy. A study done by Cates et al. showed similar results with women receiving three or more IPTp doses having a significantly reduced risk for LBW from 9.9 % to 6.9 % or roughly a 30 % decrease in incidence (Cates, 2018). Another study conducted in Malawi also showed that two or more IPTp doses reduced the incidence of LBW (Gutman, 2013).

Although the primary purpose behind IPTp dosing is to decrease the prevalence of malaria; it was not actually significantly different by IPTp dosing in the study, and we

observed improvements in maternal weight gain and achievement of MUAC ≥ 23 cm as well as infant outcomes, primarily birth weight.

Several studies have investigated the metabolic pathways for IPTp-SP dosing and its effect on outcomes unrelated to malaria, but the pathways remain largely undefined outside of the concept of the role of SP in inhibition of folate metabolism. A study done by Gutman et al. presumed that IPTp-SP improved LBW due to its antibacterial or anti-inflammatory properties (Gutman, 2017). Another possible mechanism involves SP causing alterations in the bacterial flora of the gut or reductions in the genitourinary tract organisms associated with adverse pregnancy outcomes, leading to effects on both maternal and infant weight gain (Dingens, 2016). A study done by Chico et al. showed SP to be an antibiotic that likely exerts an inhibitory effect against nonmalarial causes of LBW and pre-term delivery (Chico, 2015). There is significant research linking SP to reductions in many bacteria related to urinary tract infections, such as P. jiroveci pneumonia and Toxoplasma gondii (Chico, 2017), as well as SP was commonly used to treat N. gonorrhoeae before penicillin was developed (Schurmann, 2002). The theory behind the effectiveness of SP in reducing common bacteria associated with adverse pregnancy outcomes is women receiving a dose at each ANC decreases the overall bacteria load, reducing the incidence of adverse birth outcomes (Chico, 2017). There is also some evidence that SP exposure inhibits maternal inflammatory responses to infections that are known to trigger preterm delivery (von Linsingen, 2016).

Research is mixed of the effect of IPTp on anemia. In our sub-study there was no statistical difference observed in proportion of anemic women by IPTp dosing after adjusting for covariates. A study done by Kayentao et al. showed that IPTp dosing

decreased the prevalence of several maternal anemia (Kayentao, 2013), while another study showed that women receiving IPTp doses are 80 % less likely to be anemic (Wilson, 2011). Other studies correlated with our results showing no difference in the prevalence of anemia across IPTp doses (Harrington, 2011, and Mosha, 2014). No studies were found that showed IPTp dosing had a significant effect on hemoglobin values; however, a study done by Orish et al. showed an increase in hemoglobin values with women receiving three IPTp doses, but the values were not statistically significant (Orish, 2015).

5.2 Hemoglobin and Anemia

Maternal weight gain correlated closely with hemoglobin at enrollment (p-value = 0.0111). The range for hemoglobin at enrollment was between 7.0 and 15.5 g/dL and the higher hemoglobin values were found to correlate to higher weight gains during treatment. Hemoglobin after ten weeks of study participation did not have a statistical effect on weight gain during treatment, but the difference between first and second hemoglobin measurements did. We observed that women with a lower difference between first and second hemoglobin measurement (greater negative Δ Hgb value) had the highest weight gains.

Anemia status, defined by hemoglobin less than 11.0 g/dL, was not found to predict maternal weight gain. Women, both anemic and non-anemic at enrollment had a 2.97 kg, on average, weight gain during treatment (p-value= 0.9754). Similarly, anemia status at the second measurement did not predict maternal weight gain. We examined the relationship between the severity of anemia, defining mild (Hgb between 7 and 9.9 g/dL)

and moderate anemia (greater than or equal to 9.9 g/dL, but less than 11.0 g/dL, and did not observe any statistically significant results.

The change in hemoglobin was a significant predictor of achievement of MUAC ≥ 23 cm. Women with a higher change in hemoglobin were 1.08 times more likely to achieve a MUAC ≥ 23 cm (p-value = 0.0271). This is consistent with previous studies, which showed increased hemoglobin values led to increased MUAC (Laghari, 2017). Other studies confirm our findings, showing anemia to be associated with low MUAC (Ghosh, 2019), and a study done in Ethiopia showed that a MUAC > 23 cm reduced the odds of anemia during pregnancy by 41 %. While we were unable to find a direct association between anemia status and achievement of MUAC ≥ 23 cm, we did observe a relationship between anemia after ten weeks and MUAC at delivery. Women that were anemic after ten weeks had a lower MUAC at delivery (p-value = 0.0024).

Low pre-pregnancy BMI or fast gestational weight gain (GWG) was been correlated to higher risk of iron deficiency anemia (Tan, 2018). In our study, we found lower hemoglobin to be correlated with lower weight gain, which is opposite of findings by Campos et al. showing low hemoglobin and high prevalence of anemia to be associated with excessive weekly weight gain (Campos, 2019). In their study, the reasoning was attributed to hemodilution, leading to an increased ratio of plasma volume to erythrocytes, or a physiological anemia. Lao et al. had results similar to ours, showing that iron deficiency anemia is associated with a decreased weight gain during pregnancy (Lao, 2004).

There was no observed effect of maternal anemia status at either time point on any infant outcomes monitored. Our research does not include women that were

classified as having severe anemia, hemoglobin less than 7.0 g/dL, therefore, we only examined the relationship between mild and moderate maternal anemia and infant outcomes. We found that moderate maternal anemia was a predictor of infant length, weight, and head circumference at six weeks and three months, but mild maternal anemia at enrollment was not a significant predictor. The association between maternal anemia and LBW is largely controversial. Some previous research has shown no association among maternal anemia and infant anemia (Shao, 2012, and Solange, 2015). Maternal anemia has been linked to increased morbidity and risk of fetal demise (Abu-Ouf, 2015) and a study in Beninese showed severe anemia to increase the risk of LBW (Bodeau-Livinec, 2011).

A study done in Pakistan and India showed that only severe anemia was able to statistically predict differences in infant outcomes (Parks, 2019). Another piece of research suggests that anemia, regardless of severity, is an important predictor of infant outcomes (Tunkyi, 2018). A study conducted by Menon et. al in India showed that it was important to consider the trimester the mother was anemic and found that anemia in the second trimester had the greatest statistical predicting power for infant outcomes (Menon, 2016). A study in Benin, West Africa, strongly linked the prevalence of malaria in pregnant women to reduced infant hemoglobin through the first year of life (Accrombessi, 2015). Another study done in Malawi showed that use of iron supplementation as well as antimalarial procedures during pregnancy improved infant hemoglobin. (Le Chessie, 2002).

5.3 Strengths and Limitations

This study had a few major strengths, which include the method of data analysis, through its randomization. The study included a large sample number and comprehensive data collection for several different variable of interest, which helped rule out any confounding variables that may be at play.

When trying to observe the effects of no IPTp or four IPTp doses, our study lacked power due to a low sample size. While we found significant improvements for both maternal and infant health outcomes when three IPTp doses were given during pregnancy, we were unable to have the consistent ability to see any effect four IPTp doses had; similarly, no IPTp doses had several observed outcomes that were unable to be fully interpreted.

The average weeks spent enrolled in the study was around nine weeks, which explains why the second hemoglobin sample size was smaller than hemoglobin at enrollment. The average gestational age at enrollment of women in the study was twenty-four weeks; however, fundal height was used to determine gestational age, which women often delivered with fundal heights well below 40 cm. Well-nourished and severely malnourished women were excluded from the study, which limited the scope of inference for our results. It would have been beneficial to observe health outcomes of severely malnourished women as previous research has indicated benefits of IPTp dosing in severely malnourished populations. Human error, misreporting, and lack of adherence also serve as limitations to this study.

CHAPTER 6:

CONCLUSION

All the data analyzed showed that women receiving three or four IPTp doses had statistically improved maternal and infant outcomes or at least a positive trend towards improved outcomes if not statistically significant. Maternal hemoglobin was also found to be a statistically significant predictor of maternal and some infant outcomes.

We hypothesized women receiving less than three total IPTp doses during pregnancy or having low hemoglobin would have lower weight gain during treatment as well as a lower proportion of women that achieve a MUAC ≥ 23 cm. We found that women receiving three of four IPTp doses had the biggest improvements in weight gain and achievement of MUAC ≥ 23 cm. When considering the timing of IPTp doses, three IPTp doses had improvements in maternal weight gain during treatment over one IPTp dose during the third trimester. Women receiving any number of IPTp doses during the second trimester had a higher proportion of women achieve a MUAC ≥ 23 cm. We found that hemoglobin was directly proportional, with a positive trendline, to weight gain during treatment. This is the first research, to our knowledge, that examined the predictability of maternal weight gain during treatment and achievement of MUAC ≥ 23 cm by IPTp dosing or maternal hemoglobin.

Another key analysis we performed examined if maternal hemoglobin or number of IPTp doses had any predicting power for infant outcomes at birth, six weeks, and three months. The relationship between maternal hemoglobin or IPTp dosing had been studied extensively in the past and our findings are consistent with previous research.

IPTp dosing and maternal hemoglobin is important to consider when predicting both maternal and infant outcomes. These results indicate that further research is needed. IPTp dosing is closely related to illness or malaria and while receiving IPTp limits malaria in a population, illness should be considered when predicting maternal and infant outcomes. It would also be important to consider previous history of illness as we found many outcomes were statistically significantly affect by hemoglobin at enrollment, or a piece of the woman's baseline health entering the study. Further research should also be done to address whether there are improved outcomes with women receiving four IPTp doses compared to three IPTp doses.

Implications of this research are consistent with previous research in that IPTp-SP doesn't just improve maternal and infant health outcomes by reducing the prevalence of malaria as we did not observe a difference in prevalence of malaria by IPTp-SP dosing. Our results are consistent with guidelines issued by the WHO that women should receiving at least three doses of IPTp during pregnancy, beginning early in the second trimester. Research should be done in the future to assess if there are biological adaptions over time that interfere with undernourished populations' ability to become replete or if global criteria apply to undernourished populations in Sub-Saharan Africa.

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APPENDIX:

Appendix Table A: Nutrient Composition and Comparison by Treatment

Nutrient	RUSF Amount (%RDA)	CSB+UNIMMAP[1] Amount (%RDA)	CSB-IFA[2] Amount (%RDA)
Energy (kcal)	920	893	893
Protein (g)	36	33	33
α-linolenic acid (g)	2.26 (161)	0	0
Linoleic acid (g)	13.96 (107)	0	0
Vitamin A (µg)	2628 (341)	3210 (417)	2410 (312)
Vitamin B1 (Thiamin; mg)	3.2 (228)	1.7 (121.5)	0.3 (20)
Vitamin B2 (Riboflavin; mg)	3.8 (270)	4.7 (355)	3.29 (235)
Niacin (B3; mg)	35 (194)	36.8 (204)	18.8 (104)
Vitamin B6 (mg)	4.0 (210)	5.9 (198)	4.0 (210)
Vitamin B_{12} (µg)	5.5 (262)	7.3 (253)	4.7 (181)
Folic acid (µg)	574 (143)	659 (165)	659 (163)
Vitamin C (mg)	170 (200)	281 (331)	211 (249)
Vitamin D (µg)	30 (200)	20.9 (206)	25.4 (169)
Vitamin E (mg)	39.2 (261)	29.5 (197)	19.5 (130)
Vitamin K (µg)	192 (213)	70.5 (78)	70.5 (78)
Iodine (µg)	300 (136.4)	244 (170)	94 (43)
Copper (µg)	2400 (240)	2000 (200)	0
Iron (mg)	45 (170)	45.3 (180.7)	79.2 (292)
Zinc (mg)	24.6 (223)	26.8 (243)	11.8 (107)
Magnesium (mg)	327 (93)	0	0
Calcium (mg)	1830 (183)	851 (85)	851 (85)
Selenium (µg)	123 (205)	65 (108)	0

[1] Estimates a daily portion of 235 g CSB+ per day plus UNIMMAP supplement

[2]Estimates a daily portion of 235 g CSB per day plus iron (60 mg) and folic acid (400 µg)

Appendix Table B: Univariate Analysis of IPTp Doses and Categorical Covariates.

Characteristics	Total Number of IPTp Doses (n = 1648)						P-value
	0 n (%)	1 n (%)	2 n (%)	3 n (%)	4 n (%)	Overall n (%)	
Intervention							0.0461
RUSF	19 (1.2)	117 (7.1)	246 (14.9)	152 (9.2)	19 (1.2)	553 (33.6)	
CSB-IFA	15 (0.9)	126 (7.7)	230 (14.0)	169 (10.3)	16 (1.0)	550 (33.4)	
CSB-UNIMMAP	9 (0.6)	120 (7.3)	269 (16.3)	121 (7.3)	20 (1.2)	545 (33.1)	
Seasonality							0.3524
Rainy	13 (0.8)	130 (7.9)	240 (14.6)	130 (7.9)	15 (0.9)	528 (32.0)	
Dry	30 (1.8)	233 (14.1)	505 (30.6)	312 (18.9)	40 (2.4)	1120 (68.0)	
Clinic Region							< 0.0001
Blantyre	8 (0.5)	62 (3.8)	118 (7.2)	90 (5.5)	11 (0.7)	289 (17.5)	
Chikhwawa	27 (1.6)	173 (10.5)	305 (18.5)	147 (8.9)	13 (0.8)	665 (40.4)	
Mulange	3 (0.2)	17 (1.0)	54 (3.3)	9 (0.6)	1 (0.1)	84 (5.1)	
Zomba	5 (0.3)	111 (6.7)	268 (16.3)	196 (11.9)	30 (1.8)	610 (37.0)	
Education							0.0007
None	5 (0.3)	54 (3.3)	79 (4.8)	28 (1.7)	1 (0.1)	167 (10.1)	
1-3 years	10 (0.6)	66 (4.0)	104 (6.3)	54 (3.3)	4 (0.2)	238 (14.5)	
4-6 years	17 (1.0)	129 (7.8)	271 (16.5)	174 (10.6)	21 (1.3)	612 (37.2)	
7-8 years	7 (0.4)	75 (4.6)	198 (12.0)	126 (7.7)	24 (1.5)	430 (26.1)	
Secondary	4 (0.2)	37 (2.3)	88 (5.3)	57 (3.5)	5 (0.3)	191 (11.6)	
Tertiary	0 (0)	2 (0.1)	4 (0.2)	3 (0.2)	0 (0)	9 (0.6)	

[1]P-values calculated using Chi-squared analysis.

Appendix Table C: Univariate Analysis of IPTp Doses and Continuous Covariates

	Total Number of IPTp Doses (n = 1648)						
	0 n = 43	1 n = 363	2 n = 745	3 n = 442	4 n = 55	Overall	P-value
Hemoglobin at enrollment (g/dL)	10.03±0.3[ab]	9.89±0.08[b]	10.24±0.05[a]	10.22±0.07[a]	10.20±0.2[ab]	10.15±0.05	0.0044
Weeks on Treatment	7.80±0.8[bc]	7.48±0.3[c]	8.92±0.2[b]	10.47±0.3[a]	12.2±0.8[a]	9.1±0.1	<0.0001