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Effects of Human Maternal Placentophagy on Maternal Postpartum Iron-Status

Laura Kathleen Gryder
University of Nevada, Las Vegas

EFFECTS OF HUMAN MATERNAL PLACENTOPHAGY

ON MATERNAL POSTPARTUM IRON-STATUS

By

Laura Kathleen Gryder

Bachelor of Arts in Anthropology and German
University of Nevada, Las Vegas
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Laura Gryder

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Daniel C. Benyshek, Ph.D.
Examination Committee Chair

Kathryn Hausbeck Korgan, Ph.D.
Graduate College Interim Dean

Alyssa Crittenden, Ph.D.
Examination Committee Member

Peter B. Gray, Ph.D.
Examination Committee Member

James W. Navalta, Ph.D.
Graduate College Faculty Representative

ABSTRACT

Recently, human maternal placenta ingestion, known as placentophagy, has emerged as a rare but growing practice among postpartum mothers in industrialized societies, and is currently found in both home birth and hospital birth settings. The practice is purported to result in certain health benefits for postpartum mothers, some of which could be related to the iron content in full-term placenta (e.g., increased energy and an improved and more rapid postpartum recovery, among others). The aim of this research project was to investigate the effect of encapsulated placentophagy on maternal postpartum iron status via a randomized, double-blind, placebo-controlled pilot study (n=28). The majority of participants were Caucasian (79%), with at least some college education (88.7%), and married or in a domestic partnership (87%). Maternal iron status of women in the placenta supplement group (n=12), the placebo group (n=16), and an additional iron-supplement comparison group (n=3) was measured via hemoglobin, transferrin, and ferritin taken from blood samples at four time points; the 36th week of pregnancy, within 72 hours of parturition, between days five and seven postpartum, and during week three postpartum. All participants also completed a Willet Food Frequency Questionnaire in order to assess dietary iron intake during the study period. Results reveal no statistically significant differences in the maternal iron status of women in the placenta supplement and placebo (beef or vegetarian supplements) groups. While the small sample size of the additional (over-the-counter) oral iron supplement group did not allow for statistical comparison with the placenta supplement and placebo groups, maternal iron status of these participants varied only slightly from those of placenta supplement and placebo

group participants. The current study suggests that encapsulated placenta supplementation neither significantly improves, nor impairs postpartum maternal iron status for women consuming at least adequate amounts of dietary iron.

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TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

CHAPTER 1

INTRODUCTION AND STUDY AIMS

Introduction

Recently, the practice of human maternal placenta ingestion, known as placentophagy, has gained popularity among mothers in industrialized societies, who cite its many purported health benefits. As the practice has become more common, increasingly human placentophagy is spreading from natural home birth settings to hospital births. Advocates of this practice cite the ubiquity of placentophagy among mammals generally and note the hypothesized adaptive value of the behavior for mammalian mothers put forward by scientists (e.g., maternal pain reduction, increased milk production, and postpartum nutrition). Among human mothers, this practice is purported to result in many benefits including increased energy, and improved and more rapid postpartum recovery, among others. Such benefits may be related to the iron content in full-term placenta (Bradley, et al., 2004). Many pregnant and postpartum women are advised by medical professionals to take iron supplements in order to prevent or reverse iron-deficiency and iron-deficiency anemia (IDA) (Bodnar, Cogswell, & McDonald, 2005). Iron-deficiency and IDA are characterized mainly by fatigue, followed by additional symptoms of pallor, weakness, labored breathing, headache, palpitations, and dizziness (Pavord, Myers, Robinson, Allard, Strong, & Oppenheimer, 2012). Many new mothers choose to ingest their placenta as a "natural" alternative to hormone and nutrient supplements (PlacentaBenefits.info, 2015). Heme (i.e., animal sources) iron, in particular, may be an important component of these placenta supplements, potentially allowing

women an especially *rapid* recovery from deficiency, something currently unavailable to them with standard plant-based iron supplements or perhaps other forms of dietary iron (Pavord et al., 2012). Some placentophagic mothers have claimed their physicians noted an especially rapid postpartum iron rebound, and have attributed this recovery to placenta capsule ingestion (Young S.M., 2012).

The placenta is a highly vascularized, iron-rich organ (Bradley, et al., 2004). Placentophagy advocates propose that ingesting the organ in any form (e.g. raw, cooked, or dried and encapsulated) will provide all of the necessary iron required for postpartum mothers. This research project evaluates the efficacy of such claims by means of a clinically controlled comparison of postpartum maternal iron status. The project was conducted among 3 groups: women ingesting placenta supplements, women ingesting a vegetable or beef-based placebo, and women ingesting traditional ferrous sulfate iron supplements. The results of this study provide an objective evaluation of placentophagy advocates' claims of a more rapid iron rebound for postpartum women, as these claims have never been evaluated through rigorous scientific testing.

General Aims of the Study

Research on the emerging practice of human placentophagy can help inform pregnant and postpartum women about the potential benefits and risks of ingesting their placenta. The results of the current study may aide women in making more informed decisions about their own health and that of their offspring. The dominant narrative among placentophagy advocates revolves around the "natural" aspects and benefits of the practice (PlacentaBenefits.info, 2015), despite its conspicuous absence as a traditional

practice in contemporary human societies. Recent cross-cultural research reveals that while the ritual treatment/disposal of the placenta is central to many cultural beliefs and customs, consumption of the placenta is all but unknown. The single ethnographic case of placentophagy comes from Chicano culture, likely part of the recently emerging practice and not evidence of traditional consumption (Young & Benyshek, 2010).

It is important that women engaging in placentophagy know precisely what they are consuming. While women understand that they are ingesting placenta, there is only a vague idea of human placental composition. Most information women receive about this practice is anecdotal and provided by maternal human placentophagy proponents. Rigorous scientific studies can be used help to gain a clearer picture of precisely what, and how much they are ingesting. To date, the potential health benefits or risks of maternal placentophagy have not been scientifically assessed in humans. The public health implications may be significant. Discerning how placentophagy may effect maternal iron status immediately postpartum, and compared to standard over-the-counter oral iron supplements is especially important information for new mothers, and may be especially important for iron deficient or IDA women that choose to forego traditional over the counter (OTC) iron supplements in favor of placentophagy. For these women, iron uptake via placenta capsules (over the course of several weeks) must be sufficient to amend iron deficits. Because iron deficiency and IDA are associated with negative health outcomes for mother and child, this issue is particularly important and relevant.

CHAPTER 2

LITERATURE REVIEW

Mammalian Placentophagy

Maternal placentophagy, or the consumption of the placenta or afterbirth postpartum is a ubiquitous behavior among terrestrial mammal mothers. Nearly all of the more than 4,000 terrestrial mammalian species consume the placenta after birth: carnivore, herbivore, and omnivore alike. The only exceptions are camelids, and possibly, humans. Recently, a very small but growing number of women in industrialized countries are ingesting their placentas postpartum in the belief that it will improve recovery after delivery, increase lactation, and protect against postpartum mood disorders, amongst other purported benefits (Selander, Cantor, Young, & Benyshek, 2013). Many placentophagy advocates have cited non-human mammalian placentophagy to justify the practice among human mothers (PlacentaBenefits.info, 2015). Cross-cultural instances of this practice in human populations, however, are exceedingly rare to non-existent; a 2010 review of the cross-cultural literature regarding placenta treatment and disposal practices found no evidence of placentophagy as a longstanding traditional practice in any culture (Young & Benyshek, 2010).

Kristal and colleagues have described the most prevalent speculative explanations for placentophagy among mammals generally (Kristal, DiPirro, & Thompson, 2012). The first of these hypotheses states that, "the mother undergoes a shift in food preference toward carnivorousness at the time of parturition" (Kristal, 1980). A second proposes that, "mothers eat the afterbirth to maintain the cleanliness of the nest site and to avoid

attracting predators" (Kristal, 1980). While compelling, these hypotheses alone do not provide sufficient explanations for the practice across taxa.

Hypothesis 3 states "that mothers consume the afterbirth because of general hunger, i.e., that anorexia prior to parturition leads to placentophagy as a means of maintaining homeostatic food intake requirements" (Kristal, 1980); and a fourth hypothesis "that placentophagy is a response to specific hunger, i.e., a response to specific nutritional or hormonal needs which can be satisfied by consuming the afterbirth" (Kristal, 1980).

The hypothesis of general hunger applies to animals that fast prior to parturition, which may include humans, depending upon the culture studied. Maternal "anorexia prior to parturition leads to placentophagy as a means of maintaining homeostatic food intake requirements" (Kristal, 1980). There is also evidence of fasting among non-human animals (e.g. dogs, horses) prior to parturition (Kristal, 1980).

Finally, specific hunger occurs when nutritional deficiencies result in cravings for substances rich in those nutrients (e.g. iron, zinc, or calcium), hormones (e.g., oxytocin or progesterone) or endogenous opioid or opioid factors (e.g., placental opioid enhancing factor). It should be noted, however, that placentophagy among mammals is not strictly a maternal behavior, as the general and specific hunger hypotheses might imply. Some mammals, such as hamsters, consume the placenta crossing age, sex, and reproductive history lines (Gregg & Wynne-Edwards, 2005; Gregg & Wynne-Edwards, 2006). Because of the demands of pregnancy, however, postpartum mothers are more likely to suffer from nutritional deficiencies, and those found in the placenta may reestablish maternal nutritional sufficiency. Of specific interest to this study is the micronutrient

iron, which is depleted through pregnancy, blood loss during the birthing process, and demands on the postpartum mother through lactation.

Some women with iron deficiency or IDA experience *pica*, a craving for non-nutritive substances rich in certain micronutrients. In one study conducted in South Africa, 10 iron deficient women were engaging in pica by ingesting dry soil and paper, two of whom were pregnant (Walker, Walker, Sookaria, & Cannan, 1997). Another form of pica known as pagophagy (ice consumption) has been correlated with iron deficiencies in some women, wherein pagophagy either diminishes or disappears after iron treatment (Young S. L., 2010). In another analysis, Young and colleagues assessed over 482 cultural-level accounts for reports of human geophagy while additionally assessing accounts of 297 species that have engaged in geophagy across taxa. Ultimately they found that geophagy among humans is more closely associated with curbing illness from parasites or toxic substances, as opposed to compensating for any nutritional deficiencies (Young, Sherman, Lucks, & Pelto, 2011). Evidence supporting iron-related geophagy is controversial, as not all women engaging in the practice are iron deficient, and the practice among humans in general may have little to do with nutrition. Today most mothers in industrialized countries have easy access to iron supplements to prevent or treat iron deficiency during or immediately after parturition.

Iron

The placenta is a vascularized, iron-rich organ. Additionally, iron is a highly heat-stable nutrient. Combined, these two aspects may place the modern practice of human

placentophagy in the unique position of providing adequate iron supplies necessary for postpartum women.

Iron is also an essential nutrient. It is required for the formation of red blood cells (RBCs). The majority (~70%) of iron is found in hemoglobin bound to RBCs, while the remainder is found as transferrin produced by the liver and present in blood, or as ferritin stored in the liver, spleen, skeletal muscles, and bone marrow (Thompson & O'Donnell, 2010). Iron is essential to RBCs in: a) transporting oxygen from the lungs to tissues in the body, and b) transferring carbon dioxide waste back to the lungs for expulsion (Erickson, 1996). When sufficient amounts of iron stores have been depleted, iron deficiency and, eventually, the more severe, anemic form of iron deficiency, IDA, will result.

There are different causes of anemia, a condition characterized by a lack of healthy RBCs or hemoglobin in the blood. Iron deficiency is the most prevalent cause of anemia and is initiated through different mechanisms (e.g. chronic or rapid bleeding, insufficient dietary iron intake). Sickle cell trait, malaria, HIV, certain infectious diseases, and other nutritional deficiencies (e.g. folate, vitamin A) may also cause anemia (Stoltzfus, 2001). Regardless of its etiology, the result of anemia is the same: insufficient transport of oxygen to tissues.

The simplest, most accessible method for testing iron deficiency is that of hemoglobin concentration (for RBCs). The standard clinical cutoff value for hemoglobin for pregnant women is 11 g/dL, and 12 g/dL for non-pregnant women 15 years of age or older (World Health Organization, 2011). Blood donators are screened via this method for that reason; however, it has been shown that approximately 20% of donors with normal hemoglobin concentrations are nevertheless iron deficient (Ghosh, 2012). This is

known as ‘subclinical iron deficiency’. If multiple methods of measurement are not administered, an accurate view of iron status will not be obtained. For diagnostic and research purposes, medical practitioners and scientists may choose to test hemoglobin, serum iron, transferrin, transferrin saturation, ferritin, and total iron-binding capacity (TIBC), or any combination thereof (Khalafallah & Dennis, 2012). Ferritin is the storage form of iron, and transferrin is the transport form of iron when levels are low. Measuring hemoglobin, transferrin, and ferritin allows for multiple lines of evidence to determine iron status, as each analyte is affected by differing confounding factors. Additionally, hemoglobin, transferrin, and ferritin are commonly tested, due to the comparatively low cost of analysis and the wide availability and standardization of kits (Khalafallah & Dennis, 2012). One method used to assess iron deficiency is to measure three iron markers; if at least two of the three analytes are below expected levels, one can be classified as iron deficient (Cook, Finch, & Smith, 1976).

Iron and Reproduction

Pregnancy, birth, and the postpartum period are biologically demanding for women worldwide, across cultural and social categories. While all women are at risk, some are more likely to experience iron deficiency than others. For example, short inter-birth intervals (IBIs) and related multiparity lead to increased iron deficiency and IDA susceptibility. Women need time to replenish iron supplies, but short IBIs do not allow the time to do so. In such cases women will enter into new pregnancies already depleted, and by the end of the second pregnancy, iron deficiency is exacerbated (Allen, 2000). Additionally, obesity and a lack of exclusively breastfeeding infants at postpartum

checkup (meaning infants are partially or completely bottle fed) have both been identified as risk factors for postpartum iron deficiency and IDA, particularly among low-income women (Bodnar et al., 2002). The association between iron deficiency and not exclusively breastfeeding seems counterintuitive; however, hypotheses have been proposed to explain this phenomenon. Exclusive breastfeeding may extend lactational amenorrhea, thus preventing additional iron loss through menstruation, aiding in maintaining maternal iron supplies. Additionally, breastfeeding may be associated with women of higher Socio-Economic-Status (SES), engaging in healthier eating practices, or supplementation compliance (Bodnar et al., 2002).

Research suggests that, in order to meet the demands of pregnancy, women require 300 mg or more in iron stores prior to conception (Viteri & Berger, 2005). Women lose a total of 1000 mg of iron during pregnancy and lactation, resulting in the need for iron intake of 27 mg/day during pregnancy (Khalafallah & Dennis, 2012). The Recommended Dietary Allowance (RDA) for lactating women (age 19-50) is 9 mg/day (Institute of Medicine. Food and Nutrition Board, 2001). For women with short IBIs and multiple children, iron supplementation and the consumption of iron-rich and iron-fortified foods is of utmost importance.

Iron deficiency and IDA can cause problems not only for the mother; there are short- and long-term complications for children of iron deficient mothers. Immediate problems from maternal anemia occur during birth when hormone function is compromised, leading to preterm labor and pre-eclampsia which can compromise neonatal outcomes (Rasmussen, 2001). Rodent studies have shown reductions in offspring birth weight, as well as high blood pressure at a young age (Lewis, Forhead, Petry, Oxanne, & Hales,

2002). One study showed that female rhesus monkeys deprived of iron during pregnancy produced infants with significantly lower hemoglobin, mean corpuscular volume, and mean corpuscular hemoglobin as compared to controls (Golub, et al., 2006). Results from the same study showed that prenatally iron-deprived rhesus infants exhibited reduced activity levels and reduced responses to new environments compared to controls, and infants deprived of iron postnatally exhibited poor cognitive performance and greater emotional reactions compared to controls (Golub, Hogrefe, Germann, Capitanio, & Lozoff, 2006). In humans, infants born to anemic mothers are more likely to become iron deficient anemic themselves (Colomer, 1990), and IDA is known to negatively affect mental and motor development (Allen, 2000).

Iron deficiency is a worldwide problem, though the severity varies from country to country and from one ecological, political, and social context to another. According to data collected by the World Health Organization from 1993-2005, the global prevalence of anemia was 24.8% (World Health Organization, 2013). Among women of reproductive age, this figure increases (41.8% for pregnant women, 30.2% for non-pregnant women) (World Health Organization, 2013). While industrialized nations display a lower prevalence of iron deficiency, the incidence is nevertheless high and a major point of concern for health officials. Iron deficiency in Western societies ranges from 25-40% of pregnant women, that number increasing amongst lower SES populations (Alwan & Cade, 2011); however, exact figures are difficult to assess due to the common conflation of anemia and *iron deficiency* anemia, which has led several prominent publications to inflate cases of IDA (Stoltzfus, 2001).

Iron and Mood

A number of studies have linked low levels of iron to mood disorders. It is hypothesized that anemia, often caused by iron deficiency, increases the risk of experiencing mood disorders like anxiety and depression (Benton & Donohoe, 1999). One Australian study found a significant correlation between iron deficient women taking oral contraceptives and an increased probability of depression (Rangan, Blight, & Binns, 1998). Whether this was due to iron deficiency, oral contraceptives, or a combination of the two is unknown. A double-blind placebo-controlled study showed a statistically significant correlation between iron therapy and the improved mood of 16-17 year old high school females (Ballin, Berar, Rubinstein, Kleter, Hershkovitz, & Meytes, 1992). A 2003 study has linked anemia and postpartum depression: hemoglobin measurements taken at day seven postpartum identified eight women as anemic; by day 28, seven of those eight women showed signs of depression (Corwin, Murray-Kolb, & Beard, 2003).

The Evolutionary Theory of IDA

Denic and Argarwal (2007) hypothesize why iron deficiency is so prevalent among populations, crossing multiple social categories (e.g. countries, SES, sex, age, etc.) and surviving in the face of public health initiatives to eradicate iron deficiency. They begin by noting that iron deficiency became common in the Neolithic era with the changes agriculture and sedentism brought to diet. The authors then go on to propose that diseases humans had never been exposed to prior to sedentism and its associated crowding, caused iron deficiency to be favored by natural selection. Multiple lines of evidence have shown that iron deficiency protects against crowd diseases such as malaria, plague, and

tuberculosis. Sufficient iron stores are needed to prevent anemia and maintain overall health, however, iron can also be appropriated to fuel infectious agents. When epidemics occur, group selection favors those with lower levels of iron because those infectious agents have less nourishment from which to draw upon. In the face of such harrowing diseases, the authors argue, iron deficiency is the lesser evil with which the body can more easily cope. In a sedentary population, those who consumed iron deficient diets would survive more often than those who received adequate dietary iron. It is believed that, "multiple evolutionary factors have contributed in making iron deficiency a successful phenotype" (Denic & Agarwal, 2007, p. 603). Such a selective pressure might explain why iron deficiency and IDA are common, particularly amongst pregnant and postpartum women, the focus population of this study.

Where questions of evolutionary adaptation among human populations arise, anthropologists often look towards modern populations of hunter-gatherers as the closest analog to the ancestral conditions. If iron deficiency is a byproduct of a shift in food procurement methods (i.e. agricultural subsistence), then one might imagine that modern hunter-gatherers would not suffer from iron deficiency to the extent other populations do. One such study of iron deficiency among the !Kung Bushmen of the Kalahari desert, conducted in 1971, showed a very low incidence of iron deficiency and anemia, although these results may have been confounded by the use of cast iron cooking pots – known to provide nutritional sources of iron (Metz, Hart, & Harpending, 1971). Hunter-Gatherers are some of the fittest of populations, exercising regularly and with access to a broad-spectrum diet. In contrast, sedentary and semi-sedentary communities (e.g.

horticulturalists, agriculturalists) have far less dietary variety and suffer the most from nutritional deficiencies.

Iron and Encapsulated Human Placenta

In a study from 2000 conducted in Thailand, researchers analyzed 30 human (15 female, 15 male) heat-dried placentas for hormone and nutrient content. Results show that heat-treated and granulated placentas contain, on average, 1.01 mg/g of iron (Phuapradit et al., 2000). According to recommended dosages for the first week postpartum, women engaging in placentophagy ingest a maximum of 3300 mg/day of granulated encapsulated placenta (PlacentaBenefits.info, 2015). Recall that the daily recommended dietary iron intake for lactating females aged 19-50 is 9mg (Institute of Medicine. Food and Nutrition Board, 2001). Therefore, the amount of iron found in the placentas in the Phuapradit et al. study represents about 1/3 of the daily recommended dietary iron intake (3.3 mg of iron per day in placenta supplements).

Based on findings from the study by Phuapradit et al (2000), encapsulated placenta may provide 33% or more of the recommended dietary iron intake during the first week postpartum. Iron from meat sources (heme iron) comes in a more bioavailable form than vegetable sources (nonheme iron), meaning placental iron could have a greater impact upon dietary needs, compared to a simple nonheme iron supplement of the same dose. About half of the iron found in meat comes in heme form and has a higher absorption rate than nonheme iron. Nonheme iron is much more common in food generally, having a greater overall impact on iron supplies due to its sheer ubiquity. Additionally, nonheme iron absorption can be inhibited or increased by other dietary factors as well as existing

iron stores (Monson, 1988). The bioavailability of iron found in placenta may be further complicated by the hundreds of substances found in human placental tissue, including many micronutrients, peptide and steroid hormones, and endogenous opioid neuropeptides, among others. These substances may interact with iron and or otherwise affect the physiology of postpartum mothers in ways that inhibit or facilitate iron absorption.

CHAPTER 3

RESEARCH QUESTIONS AND HYPOTHESES

In order to determine whether the ingestion of placenta capsules affects iron levels of postpartum women, we compared iron measurements among three groups of women: one group ingesting typical non-heme iron tablet supplements (Iron Group), a second group ingesting a placebo made of either beef or vegetarian meat substitute (Placebo Group), and a third group ingesting their own dehydrated and encapsulated placenta (Experimental Group).

Null Hypothesis 1: Experimental, Placebo, and Iron Group participants will not show an increase in iron levels (assessed via hemoglobin, transferrin, and ferritin) measured during, (a) days five-seven postpartum, and (b) week three postpartum, as compared to baseline measure taken prior to supplement initiation (within 72 hours postpartum).

Hypothesis 1: Experimental, Placebo, and Iron Group participants will show an increase in iron levels (assessed via hemoglobin, transferrin, and ferritin) measured during, (a) days five-seven postpartum, and (b) week three postpartum, as compared to baseline measure taken prior to supplement initiation (within 72 hours postpartum).

Null Hypothesis 1a: Experimental Group participants will not show a greater interval increase in iron levels (assessed via hemoglobin, transferrin, and ferritin) as compared to Placebo Group and Iron Group participants from baseline (within 72 hours postpartum) to day five-seven postpartum.

Hypothesis 1a: Experimental Group participants will show a greater interval increase in iron levels (assessed via hemoglobin, transferrin, and ferritin) as compared

to Placebo Group and Iron Group participants from baseline (within 72 hours postpartum) to day five-seven postpartum.

Null Hypothesis 1b: Experimental Group participants will not show a greater interval increase in iron levels (assessed via hemoglobin, transferrin, and ferritin) as compared to Placebo Group and Iron Group participants from baseline to week three postpartum.

Hypothesis 1b: Experimental Group participants will show a greater interval increase in iron levels (assessed via hemoglobin, transferrin, and ferritin) as compared to Placebo Group and Iron Group participants from baseline to week three postpartum.

Null Hypothesis 1c: Experimental Group participants will not show a greater interval increase in iron levels (assessed via hemoglobin, transferrin, and ferritin) as compared to Placebo Group and Iron Group participants from days five-seven postpartum to week three postpartum.

Hypothesis 1c: Experimental Group participants will show a greater interval increase in iron levels (assessed via hemoglobin, transferrin, and ferritin) as compared to Placebo Group and Iron Group participants from days five-seven postpartum to week three postpartum.

The timing of each measurement is purposeful. The first measurement, taken during the 36th week of pregnancy, establishes a pre-supplementation iron baseline. As iron deficiency during pregnancy increases the likelihood of experiencing postpartum iron deficiency, this is an important measurement (Allen, 2000). The second measurement, taken within 72 hours of parturition, establishes the postpartum iron baseline, which can differ from the pregnancy iron baseline. Assessing iron status between days *five-seven* and during week three (days 21-27) respectively, assures that instances of rapid iron

uptake will be captured, as iron deficient women normally require four-six weeks to return to pre-pregnancy iron levels using standard iron supplementation, as well as capturing slower iron absorption that may occur during week three (Bodnar et al., 2002; Hyder, Persson, Chowdhury, Lonnerdal, & Ekstrom, 2003).

Hypotheses Rationale

Hypothesis 1 predicts that iron rebound will occur in all three groups; this is because postpartum iron status rebound is a normal occurrence and should be observed regardless of participant supplement group (SickKids, 2009). One postulated benefit of placenta ingestion relates to the organ's iron content (Selander et al., 2013). It is proposed that the bioavailability of iron found in placenta prepared in any form (e.g. raw, fried, baked, grilled, blended, encapsulated, etc.) is superior to standard ferrous sulfate (iron) supplementation. Ferrous sulfate is composed of non-heme iron, while the placenta contains heme iron. The body absorbs heme iron two-three times more readily than non-heme iron (15-35% of heme iron versus 2-20% of nonheme iron) (Pavord et al., 2012; Monson, 1988). The bioavailability of heme iron is generally not affected by other dietary factors, while diet can greatly affect the bioavailability of non-heme iron (Pavord et al., 2012; Young, et al., 2010). Meat of any form is a great source of heme iron and is accounted for in this study, along with other impacting dietary factors. Foods high in calcium can inhibit the bioavailability of both heme and nonheme iron. Phytates in cereals, and the tannins found in coffee and tea can inhibit iron absorption of nonheme iron. Eating foods with heme iron, as well as foods high in Vitamin C will enhance nonheme iron absorption (Pavord et al., 2012).

A majority of women engaging in placentophagy dehydrate, granulate, and encapsulate their placenta, resulting in a supply of placenta supplements to be ingested daily over a two to three week period, postpartum. One placenta encapsulation service recommends taking two capsules three times per day for two weeks, with each capsule containing 550 mg of dried placenta (PlacentaBenefits.info, 2015). One study has shown that dietary iron in cooked beef reduces the bioavailability of heme iron from 65% in raw beef, to 22% in beef cooked for 5.6 to 8.6 minutes. This study employed a fast, dry-heat cooking method wherein meat was prepared in a Silex clam cooker set at 200 degrees Celsius (392 degree Fahrenheit) (Purchas, Rutherford, Pearce, & Wilkinson, 2004). Placenta pills are typically prepared using low-heat methods of steaming and dehydration, which reach temperatures of 212 degrees Fahrenheit. As iron is a very heat-stable nutrient and is not exposed to high temperatures during encapsulation, iron degradation is limited with this method.

A recent study suggests that iron may have a significant effect upon women who engage in maternal placentophagy. The study reveals that 96% of nearly 200 women who had previously engaged in maternal placentophagy rated their experience as either "very positive" or "positive," 8% of participants listed restoring hormones/nutrients as a motivating factor for engaging in placentophagy, and 26% of participants reported increased energy, even though this was not a stated motivation for engaging in placentophagy (Selander et al., 2013). This is noteworthy, given that energy is associated with satisfactory iron stores, whereas iron deficiency is related to decreased energy, often manifested as reduced work capacity (Haas & Browlie, 2001).

CHAPTER 4

METHODS

Institutional Review Board and Institutional Biosafety Committee Approvals

This study was approved by UNLV's Institutional Review Board (IRB) Institutional Biosafety Committee (IBC). Active data collection for this study occurred between December 2013 and March 2015. This research project was conducted in conjunction with another study on the effects of human maternal placentophagy on postpartum recovery wherein Experimental and Placebo Groups, as well as Iron Group data were utilized for both studies.

Confidentiality

All information gathered in this study is kept as confidential as possible. All records (biological samples and written data) are being stored in a locked facility at UNLV for three years after completion of the study. Each participant was given a unique alphanumeric ID code. Questionnaires and biological samples are stored independent from the informed consent form to preserve confidentiality.

Participant Recruitment

Experimental and Placebo Groups

Participants were informed of the study by either the webpage or social media pages for Placenta Benefits (PBi; an organization that provides placenta encapsulation information and services), or by word of mouth through Jodi Selander, founder of PBi, or

midwives and physicians with whom she works regularly. Interested women were provided with contact information for a research team member who could then provide more information and answer any questions about the study, and who could schedule the first meeting with the research team.

Iron Group

Participants in the Iron Group were informed of the study by word of mouth, and distribution of study fliers to various pregnancy and postpartum support groups in the Las Vegas Valley (e.g. La Leche League, Well Rounded Momma, etc.). Flyer distribution did not yield any responses, however word of mouth successfully found participants for the Iron Group. All three participants in this group were women familiar with the researchers; during the course of socializing, each participant was told about the study and asked if they would like to participate.

Participants

In order to participate in this study, volunteers were screened for either the Experimental and Placebo Groups, or the Iron Group. For the Experimental and Placebo Groups, volunteers were required to be healthy, pregnant females over the age of 18 who had decided to ingest their own placenta postpartum. Volunteers were excluded from participating in this study if they had experienced any serious pregnancy complications, ever ingested human placenta, or currently used drugs, cigarettes, or alcohol. Participants who had any current or chronic health conditions, or who were taking medications during pregnancy were required to discuss their decision to ingest their placenta postpartum with their medical practitioner and to confirm that the practitioner had not advised against

placentophagy in order to participate in the study. For the Iron Group, participants were required to be healthy, pregnant females over the age of 18 who had decided *not* to ingest their own placenta postpartum. Iron Group participants were excluded from participating in this study if they had experienced any pregnancy complications, were planning to ingest their placenta postpartum, currently used drugs, cigarettes, or alcohol, took any medications, or had a medical condition that affects iron status (e.g., hemochromatosis, thalassemia). Some participants had a medical condition that did not necessarily exclude them from the study. In such cases, participants signed a form indicating they had spoken with their medical doctor or midwife and that they were not advised against participating in the study.

Procedures

Experimental and Placebo Groups

After the initial screening and providing informed consent, participants in the Experimental and Placebo Groups met with two female research team members a total of four times in the home of the participant or in a few select cases, in a secure lab on the campus of UNLV.

The first meeting occurred during the 36th week of pregnancy where the participant completed a background information questionnaire and provided a blood sample. Approximately 600µl of blood was collected through a finger-stick blood draw into a Sarstedt Multivette 600 capillary blood collection lithium heparin tube. An additional 30µl (15µl x 2 measurements) of this blood was collected onto a STAT-Site MHgb test strip and placed in the STAT-Site MHgb meter for hemoglobin analysis. For biological

sample collection, researchers wore the following personal protective equipment: gloves, laboratory coat, and protective glasses.

The second meeting occurred within 96 hours postpartum where the participant's placenta was steamed, dehydrated, pulverized and encapsulated by a trained specialist, and the participant provided a blood sample. At this meeting, participants were instructed to ingest a postpartum supplement following a designated dosing schedule of two 550mg capsules three times per day for days 1-4, two 550mg capsules two times per day for days 5-12, and two 550mg capsules once per day for days 13-21. The supplement was either their own dehydrated and encapsulated placenta, or placebo capsules made of either organic, grass-fed beef, or soy-free vegetarian meat substitute (Quorn Meatless & Soy-Free Grounds). Prior to recruitment, identification codes were generated for each of the anticipated participants in the Experimental and Placebo Groups and group assignments were randomly generated for each participant using an online randomization generator (http://www.randomization.com/). This was done by the Principle Investigator, Dr. Daniel Benyshek, who also created sealed envelopes for each identification code that indicated which treatment they would receive, and was opened by the encapsulation specialist (Jodi Selander, Marcy Webb, or Romina Lizaso) after the placenta was prepared, indicating to her whether to leave the placenta or placebo capsules with the participant. Dr. Benyshek and the encapsulation specialists had no additional contact with participants once the group assignment had been revealed. None of the research team members who met with the participants throughout the study were aware of the participant's group assignment until the conclusion of that individual's participation. Information about group assignment was maintained by Dr. Benyshek and was not

accessible to other members of the research group. The assigned set of capsules (either placenta or placebo) remained with the participant, while the second set was returned by researchers to the lab at UNLV. A seal was created with a unique code sequence that would be easily recognized by the participant, while simultaneously preserving their anonymity, as this code was independent of the participant's ID code used for the study. The label with the unique code was affixed over the lid of the jar of capsules that was stored in the lab at UNLV. This was done in order to ensure that upon return of the placenta capsules to a participant in the Placebo Group, she would be able to identify that the capsules being returned were in fact her capsules. Labeled capsules were stored in a locked freezer, in a locked laboratory, in a locked hallway at UNLV.

The third meeting occurred between the fifth and seventh day postpartum where the participant completed a questionnaire about their dietary intake over the last year, and provided a blood sample.

The fourth and final meeting occurred during the third week postpartum (days 21-27) where participants provided a blood sample. At the conclusion of this meeting the participant was debriefed and her group assignment was revealed. If she was in the placebo group, her placenta capsules were returned to her at this time. Participants were able to identify their capsules by the unique coded sticker affixed over the lid and written in their handwriting.

Iron Group

Procedures for the Iron Group were identical to the Experimental and Placebo Group with the exception of the following: during the second meeting (between day 5 and 7 postpartum), participants were instructed to ingest one 65 mg postpartum iron supplement

(Nature's Bounty 325 mg Ferrous Sulfate) once a day in the morning with a meal (Stoltzfus & Dreyfuss, 1996; Hyder et al., 2003; Khalafallah & Dennis, 2012; Viteri & Berger, 2005) for the duration of their participation in the study.

Compensation

Experimental and Placebo Groups

Direct interaction between study participants and researchers occurred over the course of four separate occasions (approximately 60-90 minutes per meeting). Participants were compensated for their time with an $80 gift card to their choice of Target or Whole Foods at the conclusion of their participation in the study. To compensate participants for their time at the third meeting, they were provided with their choice of the following: housecleaning services, meal delivery, grocery delivery, doula services, or a consultation with a lactation counselor. Additionally, placenta encapsulation services were provided at no cost (approximate value of $300). When a participant withdrew prior to completion of the study, they received compensation on a pro-rated basis based on the number of meetings completed prior to withdrawal: a gift card in the amount of $20 per meeting completed. Participants were provided with their encapsulated placenta if the placenta had been processed prior to withdrawal. If withdrawal occurred after the third meeting, the selected service was also provided.

Iron Group

As with the Experimental and Placebo Groups, direct interaction between Iron Group participants and researchers occurred on four separate occasions (approximately 60 minutes per meeting). Participants were compensated for their time with an $80 gift card

to their choice of Target or Whole Foods. No participants withdrew prior to the completion of their participation in the study.

Questionnaires

A number of questionnaires were administered to evaluate changes in participants' feelings and experiences during pregnancy and postpartum, and are included in the broader study that evaluates the effects of placentophagy in postpartum women. The Background Questionnaire was administered at Meeting 1 (see appendix), and the Willet Food Frequency Questionnaire (FFQ) (Willet, et al., 1985) was administered at Meeting 3 (day five-seven postpartum) and collected at Meeting 4 (week three postpartum).

Willet Food Frequency Questionnaire

This questionnaire was administered to participants at the third meeting and collected at the fourth meeting. The extensive nature and lack of temporal sensitivity of the Willet FFQ compelled us to allow participants to complete the questionnaire at their leisure between Meetings 3 and 4. The Willet FFQ averages the overall dietary intake of the participant over the last year.

The iron content for each food item on the questionnaire was assessed in milligrams and obtained from the USDA National Nutrient Database for Standard Reference Release 27 (United States Department of Agriculture, 2015). The most accurate and acceptable food for each item was chosen. In some cases, multiple foods were listed for a single entry (e.g. peach, apricot, or plum); in such cases, each item's iron content was investigated and the food item with the highest iron content and controlling for portion size was chosen. It is important to note that dietary iron may be overcompensated using

this method. In a few rare cases, an item was not listed on the USDA Database. In such cases, the search engine google.com was utilized to find an appropriate representation of the item (e.g. the most popular or ubiquitous brand). For example, Coffee-Mate was selected to represent non-dairy coffee whitener. When possible, unfortified foods were chosen before fortified items. If a participant accidentally picked more than one frequency for a food item, the more frequent option was chosen. If she failed to select a frequency, the food was not included in the assessment.

The iron content for each supplement on the questionnaire was assessed in milligrams. Using the specific supplement information obtained from the Willet FFQ and the Background Questionnaire, exact supplementary iron intake was ascertained. In the few cases where a participant failed to provide a frequency for a particular supplement, a frequency of once per day was assumed. Utilizing these methods of assessment the average daily iron intake of each participant was ascertained.

Frequencies in the Willet FFQ were provided to participants as a range; for the purpose of accurate calculations, a single figure was chosen (e.g. *2-4 times per week* became *3 times per week*). To calculate a daily dietary iron intake, a total annual dietary iron intake was generated. The *x* amount of mg/Fe for each item was compared to its ingestion frequency, and an annual figure was generated in this way. For example, one egg contains 0.88 mg/Fe. The following is a formula for daily iron intake if a participant consumes one egg once per week: (*0.88* mg * 1 per week * 4 weeks * 13 months)/364 days = 0.12 mg/day. That participant, on average, receives 0.12 mg/day of iron from egg consumption. Equations include 13 months (as opposed to 12) to account for the days in excess of 28 missed every month using this formula. This provides an estimate up to 364

days, just one day short of a year. Each item's annual estimate was calculated in this manner, and each item was combined for an annual iron consumption total for each participant; that figure was then divided by 364 (days) in order to gain an average daily iron ingestion estimate.

The Willet FFQ aids in accounting for iron fluctuations due to dietary factors unrelated to iron, placenta, and placebo (beef or vegetarian) supplement ingestion. Categorizing the iron intake of participants in this way allows these factors to be cross-referenced with biological assays (hemoglobin, transferrin, and ferritin levels measured in participant blood and plasma), to assure that measured iron levels were not distorted by dietary factors.

Iron intake status was assigned to one of three groups based upon iron Recommended Dietary Intakes for lactating women between 19-50 years old (Trumbo, Yates, Schlicker, & Poos, 2001). Participants under the daily Recommended Dietary Allowance of 9 mg/day were categorized as *below*, participants between 9-45 mg/day were categorized as *adequate*, and participants exceeding the daily allowance of 45 mg/day were categorized as *exceeds*.

Placenta and Placebo Supplement Processing and Encapsulation

Jodi Selander is the founder of the Las Vegas based organization Placenta Benefits, LTD, a company that provides placenta encapsulation service to women in their own home after giving birth. Selander, or one of her encapsulation specialists (Webb and Lizaso), processed and encapsulated participants' placentas, in accordance with Occupational Safety and Health Administration (OSHA) and Environmental Protection

Agency (EPA) guidelines, in the home of the participant, using the Placenta Benefits proprietary method in which the placenta is steamed with herbs, dehydrated, pulverized, and encapsulated. The placenta was collected by the midwife or doctor at the birth and left with the participant if she delivered at home, or transported to the home by the participant if she delivered in a hospital. The encapsulation specialist met with the participant in her home where she rinsed the organ with water, steamed it with herbs added to the steaming water, and dehydrated the organ overnight. Once the placenta was completely dehydrated, it was then pulverized using a food processor and plant-based, non-gelatin capsules were filled with the dehydrated tissue to create the placenta capsules that were given to the participant. The technique follows OSHA and EPA guidelines for safe handling and disinfection and ensures that the placenta did not come into contact with another individual's placenta, as each placenta was processed and prepared in the participant's home using sanitized equipment.

The placebo capsules were prepared by a research team member, Sharon Young, at UNLV following identical methods to the processing and encapsulation of the placenta, with the exception of using herbs in the steaming water. Placebo capsules were prepared using either organic, free-range beef or a soy-free vegetarian ground beef substitute (Quorn brand Meatless & Soy-Free Grounds). Organic, free-range beef was used in order to reduce the presence of exogenous hormones in the meat-based placebo supplement, while maintaining a consistency that is similar to that of the placenta capsules. The vegetarian option was offered for participants who practiced a vegetarian diet or preferred a supplement that was not beef-based, and the meatless ground beef substitute was selected as the vegetarian option in order to maintain a similar consistency to that of the

placenta capsules. Three participants chose the vegetarian placebo, while the remainder (n=13) chose the beef placebo.

Assessment of Iron Status

Iron status was assessed via hemoglobin, transferrin, and ferritin levels in participant blood samples (Santo, 2012; Denic & Agarwal, 2007; Khalafallah & Dennis, 2012). Blood samples were typically collected in the late morning toward the end of each meeting, however the timing of sample collection varied for some participants. Blood samples were collected in the evening for three meetings (twice for one participant, once for another participant), and several samples were collected in the early morning or early afternoon.

Some studies have shown that blood samples collected via capillary action overestimate hemoglobin levels in the high range and underestimate hemoglobin levels in the low range, when compared to standard venous samples (Cable, et al., 2012). Other studies show more generally that capillary hemoglobin values are greater than venous levels (Patel, Wesley, Leitman, & Bryant, 2013). For this study, our use of the point of care (POC) tool, STAT-Site hemoglobin meter, prevents us from encountering this issue for hemoglobin. Expected Values for this POC instrument establish the expected values for adult females as 11.0-18.0 g/dL (Stanbio Laboratory, 2013). Hemoglobin was measured onsite using the portable STAT-Site MHgb meter, which has a total precision coefficient of variation (CV) of 2.9% - 4.2%, and a within run precision CV of 3.5% - 4.9% (Stanbio Laboratory, 2012). In the majority of cases, two blood spots were collected allowing us to calculate an average. In some cases the figures varied, but this

was normally due to collection error wherein drops of blood did not both meet the recommended 12μL required for the meter. Reference value ranges for this project are based upon the STAT-Site hemoglobin Expected Values of 11-18 g/dL.

ZRT Laboratory (Beaverton, Oregon) analyzed transferrin and ferritin via immunoassay of plasma taken from finger-stick capillary samples. Siemens ELISA kits for transferrin and ferritin were used, with the following expected values: transferrin: 202-364 mg/dL, and ferritin (Women 17-60 years): 13-150 mg/mL (Aquilani, et al., 2014; Eaton, 2013). The threshold values used in this analysis for transferrin and ferritin are based upon venous blood samples rather than capillary blood samples (capillary being the method of collection used in this study). For the purposes of this study we are comparing between groups, therefore minor (3-7%) fluctuations of the exact values will not affect the comparisons made (Mejia & Viteri, 1983; Lu, Lynch, Cook, Madan, & Bayer, 1987).

Statistical Measures

Using SPSS version 21, the following statistical measures were conducted: Mixed ANOVA, with sphericity adjustment where necessary (e.g., the Greenhouse-Geisser estimate), tested within- and between-subjects factors simultaneously. Mixed ANOVAs were also conducted controlling for dietary iron status (i.e., based on categories *below, adequate* or *exceeds* recommended intake).

Pearson's r calculated correlations among dependent variables (i.e. hemoglobin, transferrin, and ferritin) within each time period (Meetings 1 through 4). Fluctuations were analyzed within the Experimental Group and the Placenta Group and then compared

between groups. Given the small sample size of the Iron Group (n=3), data for that condition are assessed as a case study, as they cannot be statistically evaluated alongside Experimental and Placebo Group data.

For the small number of data points missing, data from a participant's previous time period were carried forward to fill in missing data (i.e. Meeting 2 for participant PL-1012, and Meeting 4 for participant PL-1017).

CHAPTER 5

RESULTS

Initial recruitment of the study had three participants in the Iron Group and 36 participants in the Experimental and Placebo Groups. Eight participants withdrew or were dropped from the study, and one participant switched from the Experimental and Placebo Group to the Iron Group after the first meeting. Only one participant withdrew from the study after initiation of supplementation, due to fear of a bad reaction to the placenta capsules (e.g. participant felt weepy and hormonal), however after withdrawal, it was discovered that she was a participant in the Placebo Group. This means that there was no group-based bias in dropouts due to placenta or placebo ingestion.

The following is a breakdown of participants included in final analyses for each test group: (1) Experimental Group: (n=12), and (2) Placebo Group: (n=16). Iron Group (n=3) will be examined separately.

Participant Demographics

The women who participated in this study were an average age of 30 years old, with the majority Caucasian (77.4%). Most participants had an education of at least some college or more (83.9%); with almost equal amounts of women having attended some college, and earning Bachelors and Masters degrees. Fifty-five point two percent had a household income of at least $50,000 a year; 17.2% of participants earned between $20,000 and $30,000 (the lowest household income bracket selected by participants). According to a 2014 Demographic Report, the median household income for Las Vegas

was $45,670, and $47,270 for Clark County, Nevada (City of Las Vegas Economic and Urban Development Department & Redevelopment Agency, 2014). The majority of participants were married or in a domestic partnership (87%) (see Table 1). The demographic profile of participants in the current study is largely consistent with that of other published studies of women who have engaged in maternal placentophagy in the past (Selander et al., 2013).

Demographic Characteristics			
			Frequency %
Age (N = 31; mean = 30)			
Ethnicity (N = 31)			
	African American	2	6.5%
	Caucasian	24	77.4%
	Hispanic/Latina	4	12.9%
	Mixed: American Indian/Alaska Native, Caucasian	1	3.2%
Education (N = 31)			
	High School or equivalent	1	3.2%
	Vocational/technical school	4	12.9%
	Some college	9	29.0%
	Bachelor's degree	8	25.8%
	Master's degree	8	25.8%
	Doctoral degree	1	3.2%
Household Income (N = 29)			
	$20,001 - 30,000	5	17.2%
	$30,001 - 40,000	5	17.2%
	$40,001 - 50,000	3	10.3%
	$50,001 - 60,000	2	6.9%
	$60,001 - 70,000	3	10.3%
	$70,001 - 80,000	2	6.9%
	Over $80,000	9	31.0%
Marital Status (N = 31)			
	Single, never married	3	9.6%
	Married or domestic partnership	27	87.0%
	Divorced	1	3.2%

Table 1: Demographic Characteristics

Dietary Iron RDA at Meeting One

Recall that the potential impact of dietary iron unrelated to capsule ingestion upon iron status is controlled for via the Willet FFQ. Participants generally split evenly into one of two categories: *adequate* or *exceeds* RDA for lactating women (9mg/day), with only one participant *below* RDA. However, 9mg/day is much lower than the recommendations for pregnant women of 27 mg/day (Khalafallah & Dennis, 2012). For this reason it was important to determine the annual average dietary iron intake of participants during week 36 of pregnancy (Meeting 1) based upon the recommended daily allowance for pregnant women. Table 2 shows that an additional three participants were below RDA for pregnant women, splitting almost evenly among the three conditions: Experimental, Placebo, and Iron Groups.

Week 36 of Pregnancy (Meeting 1) RDA for pregnant women (27 mg/day)				
	Experimental (Placenta) Group	Placebo Group	Iron Group	Total
Below (<27 mg/day)	1	2	1	4
Adequate (27-45 mg/day)	5	4	1	10
Exceeds (>45mg/day)	5	10	1	16
Total	n = 11*	n = 16	n = 3	n = 30

Table 2: Week 36 of Pregnancy (Meeting 1): RDA for pregnant women (27 mg/day)
*One participant from the Experimental Group did not complete the Willet FFQ.

Iron Status Analytes

The reference ranges for each iron analyte (hemoglobin, transferrin, and ferritin respectively) are examined below, separated according to participant condition: Experimental (Placenta) Group, Placebo Group, and Iron Group. Reference ranges

classify participants into three analyte status categories: *below*, *expected*, and *exceeds* reference range (see Table 3). It is important to note that these categories do not represent 'iron deficient' clinical diagnosis categories in the context of this study. Such diagnoses should only be made by a licensed health care provider using standardized clinical procedures and laboratory methods. Rather, these categories are presented here for heuristic and comparative purposes only – based on manufacturer POC meter and laboratory assay reference ranges. For hemoglobin ranges, recall that the STAT-Site Hgb Meter expected values for adult females are 11.0-18.0 g/dL (Stanbio Laboratory, 2013). Expected transferrin values range between 202-364 mg/dL, with levels greater than 364 indicating transferrin deficiency and levels lower than 202 indicating transferrin excess (Aquilani, et al., 2014). Expected ferritin values for women 17-60 years are 13-150 mg/mL (Eaton, 2013).

	HGB	**Transferrin**	**Ferritin**
Below	<11 g/dL	>364 mg/dL	<13 ng/mL
Expected	11-18 g/dL	202-364 mg/dL	13-150 ng/mL
Exceeds	>18 g/dL	<202 mg/dL	>150 ng/mL

Table 3: Expected Ranges for Iron Analytes

Figures 1-3 categorize the Experimental (Placenta) Group (n=12), the Placebo Group (n=16), and the Iron Group (n=3) by hemoglobin, transferrin, and ferritin expected values (*below* and *expected*), as well as provide the number of participants for each expected value category and meeting. Tables 4-6 provide us with additional information on

averages for each meeting, subdivided by the status of the analyte (*below*, *expected*, and *exceeds*). Please note that no participant hemoglobin samples exceeded expected values, and therefore the *exceeds* category was not included in Figure 1 and Table 4.

Hemoglobin

From the data in Figure 1 and Table 4, we can see that *below* participants in the Experimental (Placebo) Group increase from Meeting 1 to 2. Following placenta supplement initiation, *below* values decrease across the next two meetings. No participant hemoglobin levels fall within the *exceeds* range for the Experimental Group.

Participants from The Placebo Group with values *below* the reference range increase from Meeting 1 (two participants) to Meeting 2 (nine participants). Following placebo supplement initiation, values *below* the reference range decrease to one participant at Meeting 3, and zero participants at Meeting 4. No participant hemoglobin ranges fall within the *exceeds* range for this Placebo Group.

All three participants from the Iron Group are within the *expected* hemoglobin range over the course of the four meetings, with no participants experiencing *below* or *exceeds* hemoglobin

Transferrin

For all three groups (Experimental, Placebo, and Iron) we can see that participants with values *below* the reference range decrease across the four meetings, participants with values in the *expected* reference range increases across the four meetings, and only two participants from the Placebo Group *exceeds* transferrin at Meeting 4 (Figure 2 and Table 5).

Ferritin

For all three groups, Figure 3 and Table 6 show that participants with ferritin values *below* the reference range generally decrease across all four meetings, excluding a slight uptick of one participant at Meeting 4 for the Experimental Group. The Iron Group had a single case of ferritin values *below* the reference range at Meeting 1, which resolved thereafter. The number of participants within the *expected* range generally increases across meetings for all groups; once again, however, the Experimental Group has a decrease by one participant from Meeting 3 (ten participants) to Meeting 4 (nine participants). Only one participant (PL-1032, from the Placebo Group) recorded an *exceeds* value for transferrin, which occurs across all four meetings.

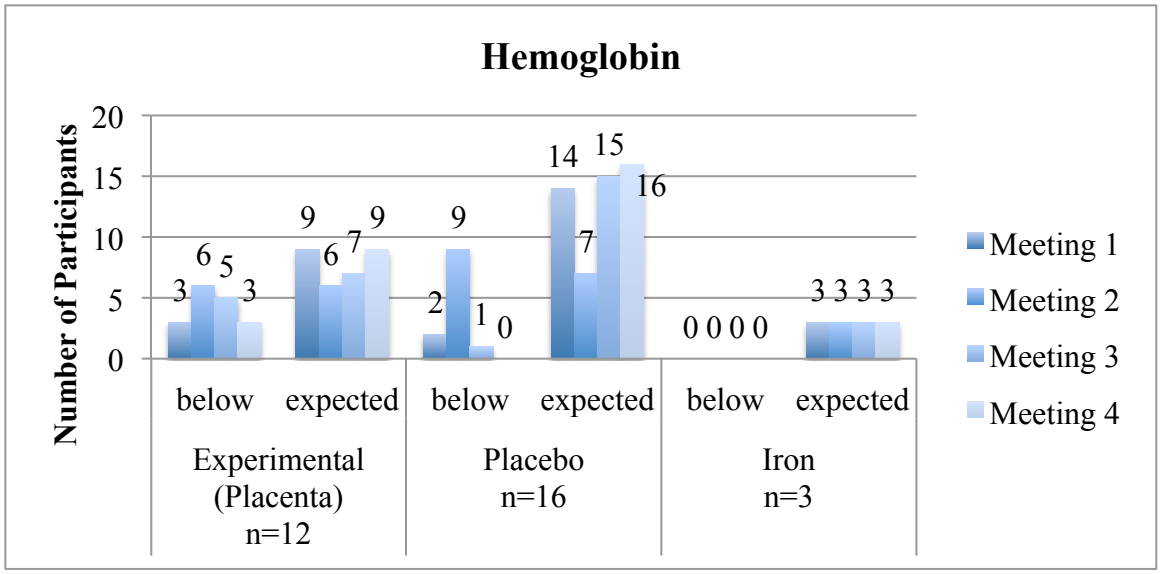

Figure 1: Number of participants with hemoglobin levels in *below* and *expected* ranges, broken up by Condition. Note that no participant hemoglobin samples exceeded the expected range.

Hemoglobin Group	Analyte Status	Meeting 1		Meeting 2		Meeting 3		Meeting 4	
		N	Mean	N	Mean	N	Mean	N	Mean
Experimental (Placenta)	**below**	3	10.38	6	8.88	5	9.44	3	9.53
	expected	9	12.38	6	11.65	7	12.62	9	13.34
Placebo	**below**	2	9.05	9	10.41	1	10.85	0	N/A
	expected	14	12.65	7	12.50	15	12.77	16	13.45
Iron	**below**	0	N/A	0	N/A	0	N/A	0	N/A
	expected	3	11.98	3	12.33	3	12.75	3	12.50

Table 4: Number of participants and means of hemoglobin levels in *below* and *expected* ranges, broken up by Condition. Note that no participant hemoglobin samples exceeded the expected range.

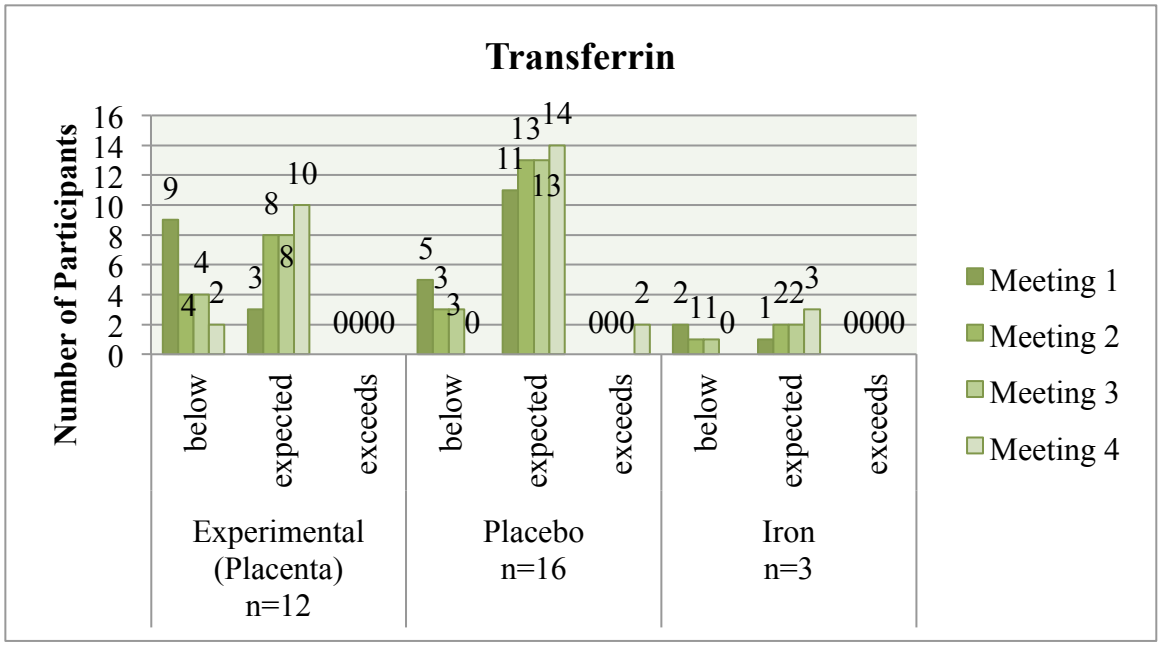

Figure 2: Number of participants with transferrin levels in *below*, *expected*, and *exceeds* ranges, broken up by Condition.

Transferrin Group	Analyte Status	Meeting 1		Meeting 2		Meeting 3		Meeting 4	
		N	Mean	N	Mean	N	Mean	N	Mean
Experimental (Placenta)	below	9	416.33	4	414.25	4	409.50	2	393.00
	expected	3	352.66	8	333.75	8	327.75	10	281.60
	exceeds	0	N/A	0	N/A	0	N/A	0	N/A
Placebo	below	5	435.60	3	420.00	3	408.66	0	N/A
	expected	11	329.09	13	303.61	13	295.38	14	272.57
	exceeds	0	N/A	0	N/A	0	N/A	2	188.50
Iron	below	2	485.50	1	510.00	1	511.00	0	N/A
	expected	1	347.00	2	317.00	2	309.50	3	278.33
	exceeds	0	N/A	0	N/A	0	N/A	0	N/A

Table 5: Number of participants and means of transferrin levels in *below*, *expected*, and *exceeds* ranges, broken up by Condition.

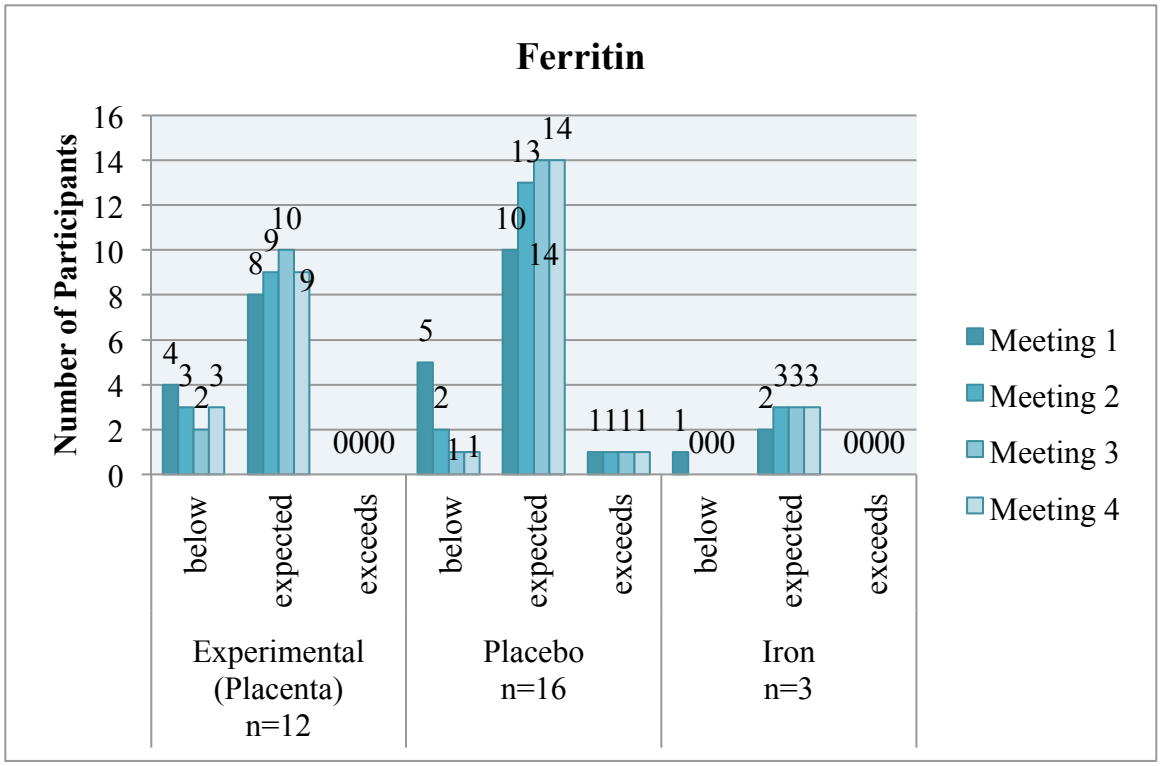

Figure 3: Number of participants with ferritin levels in *below*, *expected*, and *exceeds* ranges, broken up by Condition.

Ferritin Group	Analyte Status	Meeting 1		Meeting 2		Meeting 3		Meeting 4	
		N	Mean	N	Mean	N	Mean	N	Mean
Experimental (Placenta)	below	4	6.67	3	7.35	2	6.17	3	6.93
	expected	8	25.67	9	55.53	10	51.18	9	66.44
	exceeds	0	N/A	0	N/A	0	N/A	0	N/A
Placebo	below	5	8.60	2	9.11	1	8.89	1	12.00
	expected	10	23.45	13	40.02	14	41.12	14	54.46
	exceeds	1	160.00	1	236.00	1	347.00	1	289.00
Iron	below	1	7.86	0	N/A	0	N/A	0	N/A
	expected	2	21.68	3	39.63	3	48.73	3	67.26
	exceeds	0	N/A	0	N/A	0	N/A	0	N/A

Table 6: Number of participants and means of ferritin levels in *below*, *expected*, and *exceeds* ranges, broken up by Condition.

Deficiencies of Multiple Analytes at Corresponding Time Points

Table 7 supplies a breakdown of participants who recorded sample values below the expected reference range for more than one analyte at corresponding time points. The Experimental (Placenta) Group had a greater amount of participants that began with values below the expected reference range for multiple analytes than did the Experimental (Placenta) Group. Twenty-five percent of Experimental Group participants were below the expected reference range in two out of three analytes at Meeting 1, compared to 13% of Placebo Group Participants. While 33% of Iron Group participants were below the expected reference range in two out of three analytes at Meeting 1, it is important to note that this percentage comprises a single participant (with a total of three Iron Group participants), and making comparisons to the other larger groups difficult.

Seventeen percent of Experimental (Placenta) Group participants were below the expected reference range in three out of three analytes at Meeting 1, compared to 6% of Placebo Group Participants, and 0% of Iron Group participants.

This initial disparity in iron analytes among groups will be important when further analyzing our results.

			Meeting 1		Meeting 2		Meeting 3		Meeting 4	
		Analytes: Hemoglobin, Transferrin, Ferritin	**Number of values below the expected reference range**							
			2 of 3	**3 of 3**	**2 of 3**	**3 of 3**	**2 of 3**	**3 of 3**	**2 of 3**	**3 of 3**
Total Group N	**N = 12**	Experimental (Placenta) Group N	3	2	1	2	1	2	0	2
		Experimental (Placenta) Group Percentage	25%	17%	8%	17%	8%	17%	0%	17%
	N = 16	Placebo Group N	2	1	1	1	0	0	0	0
		Placebo Group Percentage	13%	6%	6%	6%	0%	0%	0%	0%
	N = 3	Iron Group N	1	0	0	0	0	0	0	0
		Iron Group Percentage	33%	0%	0%	0%	0%	0%	0%	0%

Table 7: Participants with sample values below the expected reference range for 2 of 3, or 3 of 3 analytes at corresponding time points.

The following three sections analyze iron levels assessed via hemoglobin, transferrin, and ferritin, whereas above, participants were separated into dietary iron status groups.

Hemoglobin Results

Analysis of hemoglobin data for Experimental (Placenta) and Placebo Groups, not controlling for dietary iron status, showed a statistically significant ($p=0.000$) change over time (Figure 4). There was a decrease in hemoglobin at Meeting 2, followed by an improvement in hemoglobin at Meeting 3 and a further improvement at Meeting 4.

Hemoglobin pairwise comparisons for the Experimental Group using Bonferroni corrected post hoc tests show there were statistically significant differences between Meetings: 1 to 2 ($p=0.048$), 2 to 3 ($p=0.005$), 2 to 4 ($p=0.000$), and 3 to 4 ($p=0.001$). Likewise, hemoglobin pairwise comparisons for the Placebo Group using Bonferroni corrected post hoc tests show there were statistically significant differences between measures: 2 to 3 ($p=0.001$), and 2 to 4 ($p=0.000$).

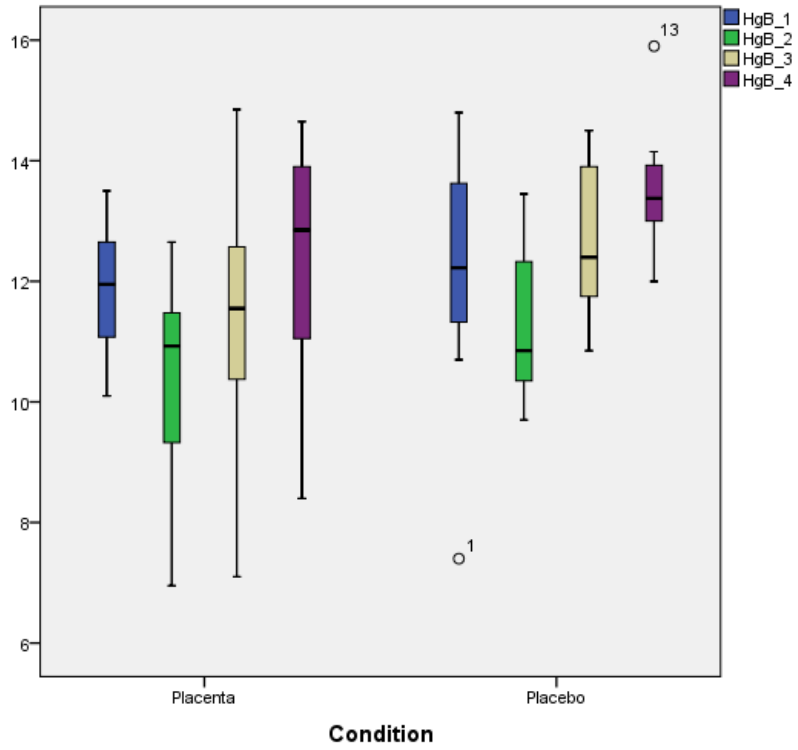

Figure 4: Hemoglobin boxplots comparing Experimental (Placenta) Group and Placebo Group across meetings 1-4.

However, the interaction of hemoglobin and condition was not significant (p=0.276). In other words, the hemoglobin of Experimental and Placebo Groups changed over time, but changed in similar ways. Figure 5 shows the parallel movement of hemoglobin status over time. We see that the groups have parallel lines that decrease from Meetings 1 to 2, and then increase steadily afterward (indicating hemoglobin status improvement).

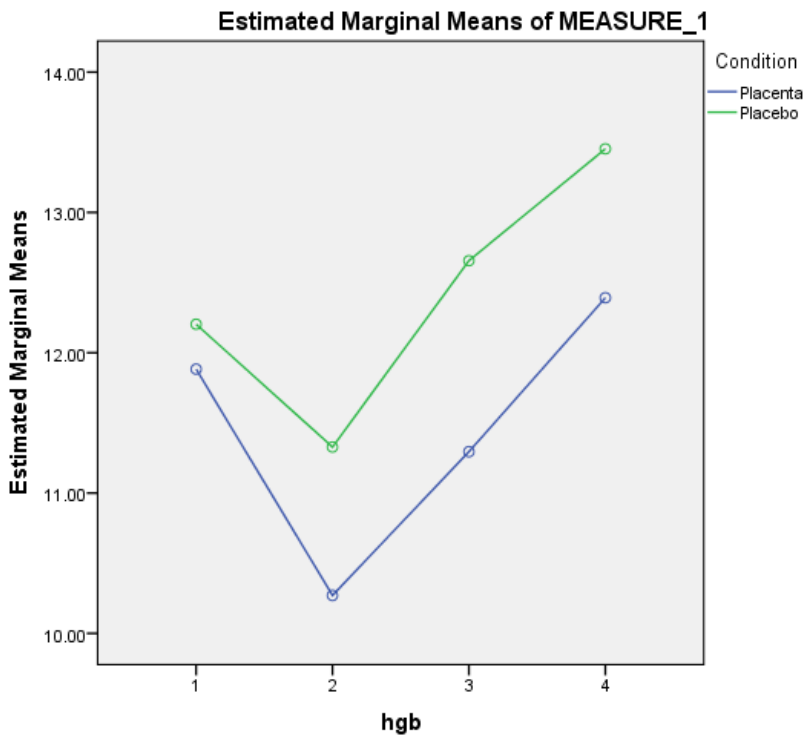

Figure 5: Hemoglobin Estimated Marginal Means comparing Experimental (Placenta) Group and Placebo Group across meetings 1-4.

The difference in mean hemoglobin values between the Experimental and Placebo Groups at any time point simply represents the different average baseline levels for the two groups. In addition, between-subjects effects show that the average difference between Experimental and Placebo Groups was not statistically significant (p=0.059).

The majority of Experimental and Placebo Group participants were grouped into *adequate* dietary iron status or *exceeds* RDA recommendations for iron intake among lactating women (19 – 50 y) based upon the results of their Willet FFQ responses. Eleven participants had a daily average iron intake between 9-45 mg/day, or *adequate* dietary iron intake. Fifteen participants had a daily average iron intake over 45 mg/day, or *exceeds* dietary iron intake. Four participants fell below the RDA iron intake recommendations (Table 2). A Mixed ANOVA was completed controlling for these

dietary groups, with no statistical differences shown between dietary controls and the hemoglobin results.

Transferrin Results

Analysis of transferrin data for Experimental (Placenta) and Placebo Groups, not controlling for dietary iron status, showed a statistically significant ($p=0.000$) change over time (Figure 6). There was a steady improvement of iron status as estimated from plasma transferrin, over time (note that lowering transferrin levels indicate improved iron status).

Transferrin pairwise comparisons for the Experimental Group using Bonferroni corrected post hoc tests show there were statistically significant differences between Meetings: 1 to 3 ($p=0.04$), 1 to 4 ($p=0.000$), 2 to 4 ($p=0.005$), and 3 to 4 ($p=0.001$). Transferrin pairwise comparisons for the Placebo Group using Bonferroni corrected post hoc tests show there were statistically significant differences between measures: 1 to 2 ($p=0.002$), 1 to 3 ($p=0.000$), 1 to 4 ($p=0.000$), 2 to 4 ($p=0.000$), and 3 to 4 ($p=0.000$).

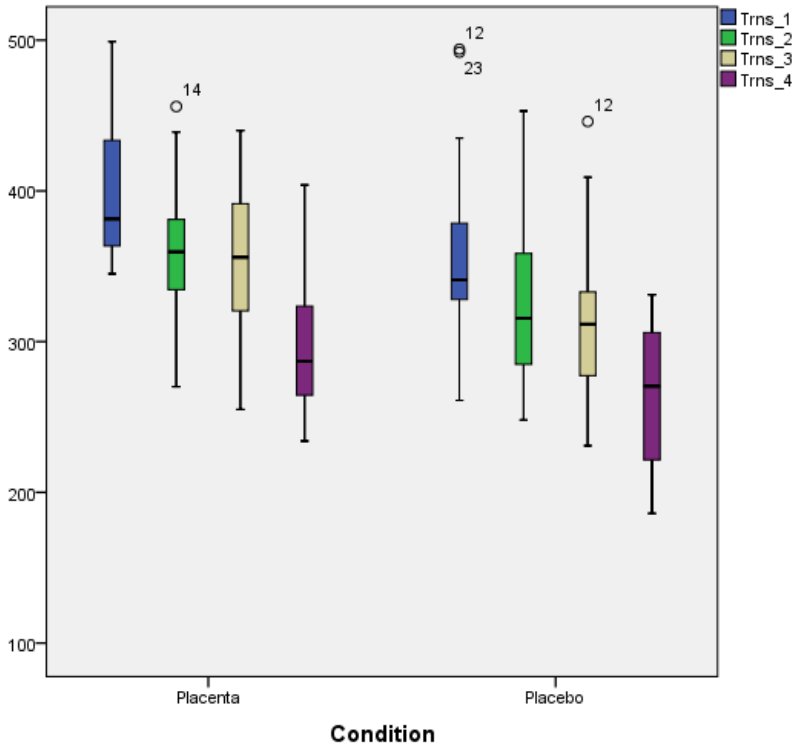

Figure 6: Transferrin boxplots comparing Experimental (Placenta) Group and Placebo Group across meetings 1-4.

The interaction of transferrin and condition was not significant (p=0.986). As with hemoglobin, the transferrin values of Experimental and Placebo Groups changed over time, but changed in similar ways. Figure 7 shows the parallel movement of transferrin status over time. We see that the groups have parallel lines that decrease steadily through Meetings 1 to 4 (indicating transferrin status improvement). Once again, differences in transferrin values for the Experimental and Placebo Groups reflected different baseline levels which were not affected by either the experimental or placebo conditions. Between-subjects effects show that the average difference between Experimental and Placebo Groups was not statistically significant (p=0.052).

As with hemoglobin, a Mixed ANOVA was used to control for iron dietary groups (*adequate* and *exceeds*). There was only one difference shown between transferrin (n=28),

and transferrin_adequate (n=11). The between-groups test for transferrin of all dietary groups indicates that the variable condition (Experimental vs. Placebo) is not significant (p=0.052), consequently, in Figure 7 we see that the lines for the two groups are close together. The between-groups test for transferrin_adequate indicates that the variable condition (Experimental vs. Placebo) is significant (p=0.037), consequently in Figure 8 we see that the lines for the two groups are a bit apart (especially in comparison to Figure 7). This means that transferrin_adequate conditions (Experimental and Placebo) started off at statistically significant different points. Recall that Mixed ANOVAs showed no statistical differences between dietary controls and hemoglobin results. This underlines the importance of assessing multiple analytes (in this study: hemoglobin, transferrin, and ferritin) to garner a more accurate understanding of iron status.

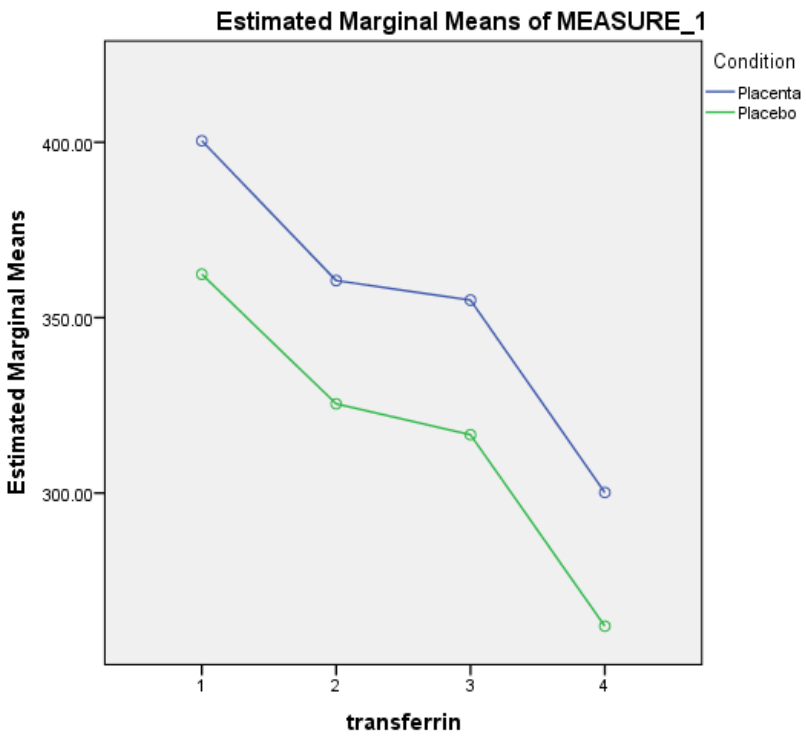

Figure 7: Transferrin Estimated Marginal Means comparing Experimental (Placenta) Group and Placebo Group across meetings 1-4, for all dietary transferrin groups (n=28).

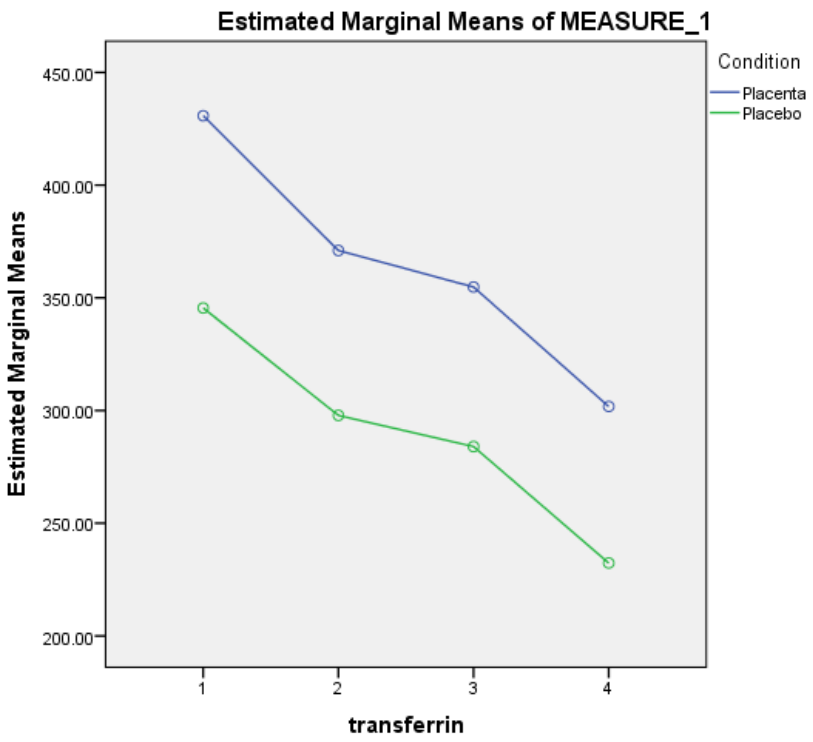

Figure 8: Transferrin Estimated Marginal Means comparing Experimental (Placenta) Group and Placebo Group across meetings 1-4, for Adequate Dietary Group (n=11)

Ferritin Results

Analysis of ferritin data for Experimental (Placenta) and Placebo Groups, not controlling for dietary iron status, required a Log10 transformation to normalize the data. Figure 9 shows the ferritin boxplots prior to Log10 transformation, and Figure 10 shows the normalized data.

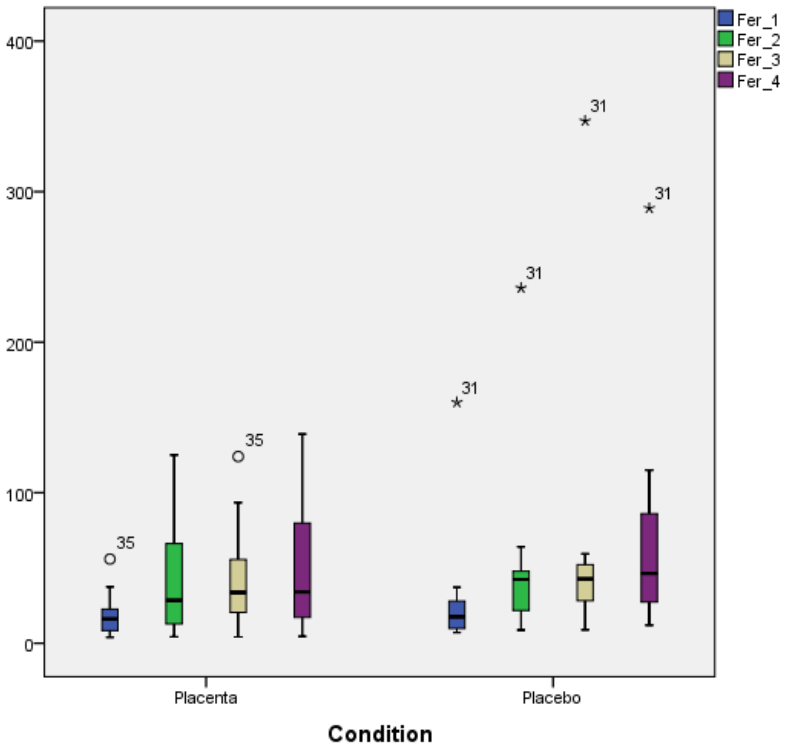

Figure 9: Ferritin boxplots comparing Experimental (Placenta) Group and Placebo Group across meetings 1-4, prior to Log10 transform.

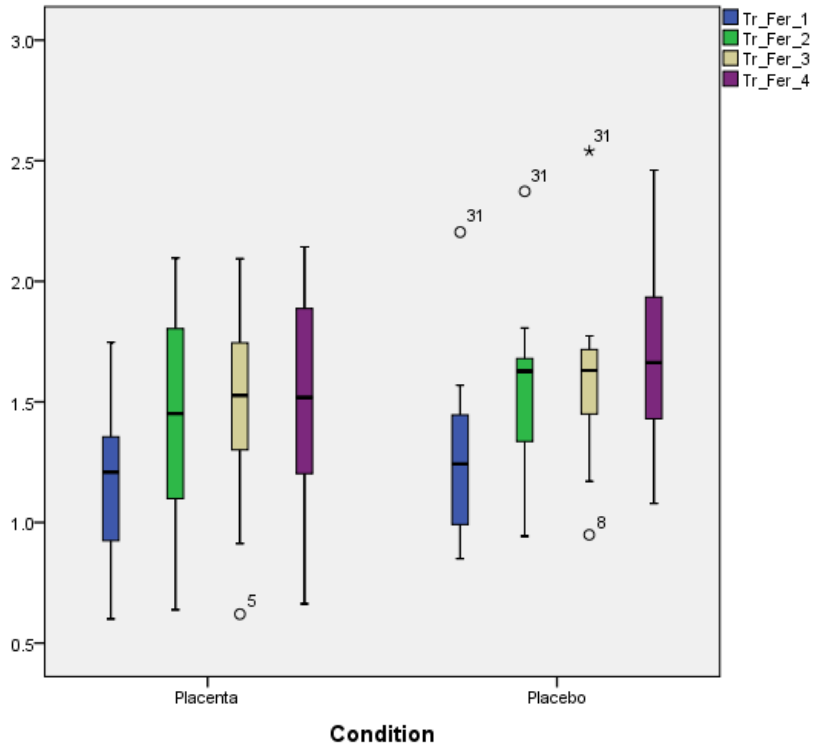

Figure 10: Ferritin boxplots comparing Experimental (Placenta) Group and Placebo Group across meetings 1-4, after Log10 transform.

Normalized ferritin data reveals a statistically significant (p=0.000) change (Figure 10). There was a steady increase of ferritin over time, indicating improved iron status. However, the interaction of ferritin and condition was not significant (p=0.751). As with the two other biomarkers for iron status in the study, ferritin of Experimental and Placebo Groups changed over time, but changed in similar ways. Figure 11 shows the parallel movement of ferritin status. We see that the groups have parallel lines that increase steadily from Meetings 1 to 4 (indicating ferritin status improvement). In addition, between-subjects effects reveal that the average difference in ferritin values between Experimental and Placebo Groups was not statistically significant (p=0.357).

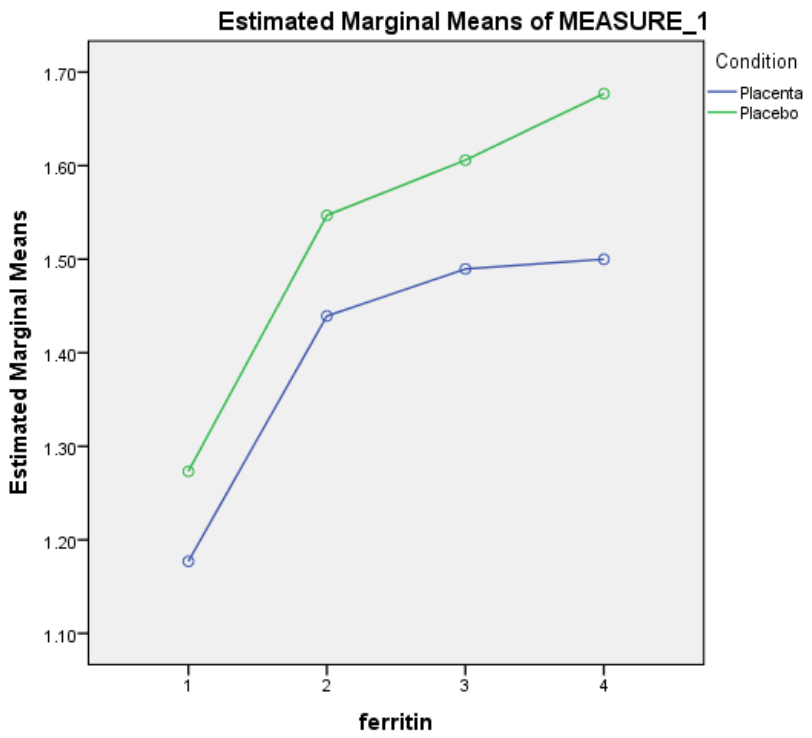

Figure 11: Ferritin Estimated Marginal Means comparing Experimental (Placenta) Group and Placebo Group across meetings 1-4.

Ferritin pairwise comparisons for the Experimental Group using Bonferroni corrected post hoc tests show there were statistically significant differences between Meetings: 1 to 2 (p=0.026), and 1 to 3 (p=0.003). Pairwise comparisons for the Placebo Group using Bonferroni corrected post hoc tests show there were statistically significant differences between Meetings: 1 to 2 (p=0.000), 1 to 3 (p=0.000), and 1 to 4 (p=0.000).

Once again, a Mixed ANOVA was used to control for iron dietary groups (*adequate* and *exceeds*). No statistical differences were shown between iron dietary groups and overall ferritin results.

Pearson's r Correlation

Pearson's r Correlation was utilized to calculate correlations among variables within each time period. The following are statistically significant ($p<0.10$) correlations from both conditions (Experimental and Placebo Groups), broken up by meeting time.

Meeting 1				
	Transferrin to Ferritin		Hemoglobin to Ferritin	
	Effect	p-value	Effect	p-value
Experimental (Placenta)	Strong, negative (r = -0.680)	0.015		
Placebo			Positive, moderate (r = 0.533)	0.033

Table 8: Meeting 1 Pearson's r Correlation by Condition

Meeting 2		
	Transferrin to Ferritin	
	Effect	p-value
Experimental (Placenta)	Negative, strong (-0.762)	0.004
Placebo	Negative, moderate (-0.449),	0.081

Table 9: Meeting 2 Pearson's r Correlation by Condition

Meeting 3						
	Transferrin to Ferritin		Transferrin to Hemoglobin		Hemoglobin to Ferritin	
	Effect	p-value	Effect	p-value	Effect	p-value
Experimental (Placenta)	Negative, strong (-0.837)	0.001	Positive, strong (0.667)	0.018		
Placebo	Negative, strong (-0.589)	0.016			Positive, moderate (0.495)	0.051

Table 10: Meeting 3 Pearson's r Correlation by Condition

Meeting 4						
	Transferrin to Ferritin		Transferrin to Hemoglobin		Hemoglobin to Ferritin	
	Effect	p-value	Effect	p-value	Effect	p-value
Experimental (Placenta)	Negative, strong (-0.868),	0.000	Negative, strong (-0.821)	0.001	Positive, strong (0.799),	0.002
Placebo	Negative, strong (-0.572),	0.021				

Table 11: Meeting 4 Pearson's r Correlation by Condition

Analytes from Meetings 2 through 4 correlate similarly across conditions. The effect size and correlation of transferrin to ferritin is similar for each condition: negatively correlated with a moderate to strong effect. As transferrin decreases, ferritin increases. This makes sense, as transferrin will lower and ferritin will rise with improved iron status. This is further indication that a great deal of difference between Experimental and Placebo Groups may not exist.

Iron Group

The small sample size (n=3) of the Iron Group will not enable comparative statistical analyses to be conducted with the Experimental and Placebo Groups. Instead, descriptive statistics for this data are examined to garner a general idea of Iron Group results and how they might compare given a more robust sample size.

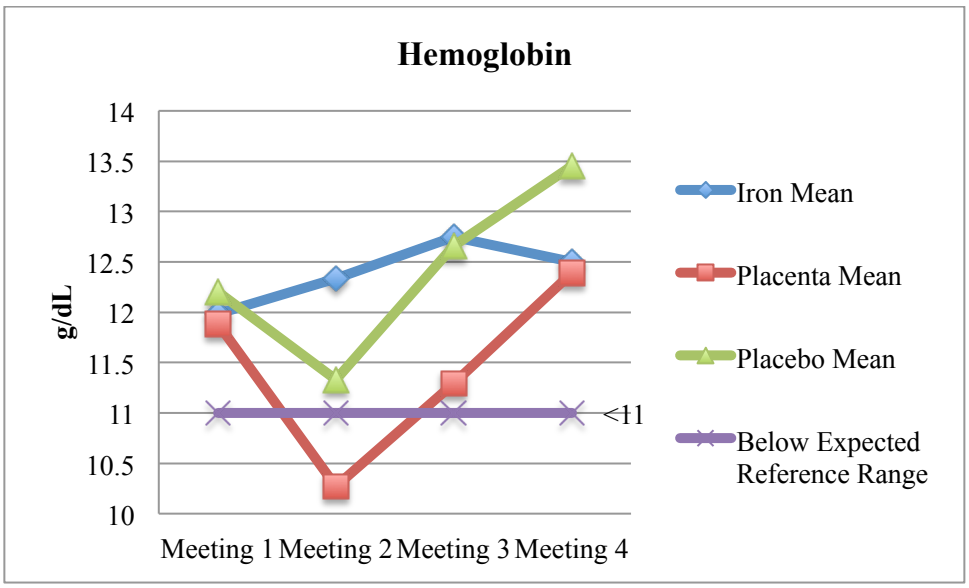

Figure 12: Hemoglobin means for meetings 1-4 by Condition.

Hemoglobin								
Condition	Meeting 1		Meeting 2		Meeting 3		Meeting 4	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Iron	11.98	0.80	12.33	0.93	12.75	1.00	12.50	0.93
Placenta	11.88	1.11	10.27	1.84	11.29	2.11	12.39	1.97
Placebo	12.20	1.79	11.33	1.17	12.66	1.20	13.45	0.90

Table 12: Hemoglobin means and Standard Deviations for meetings 1-4 by Condition.

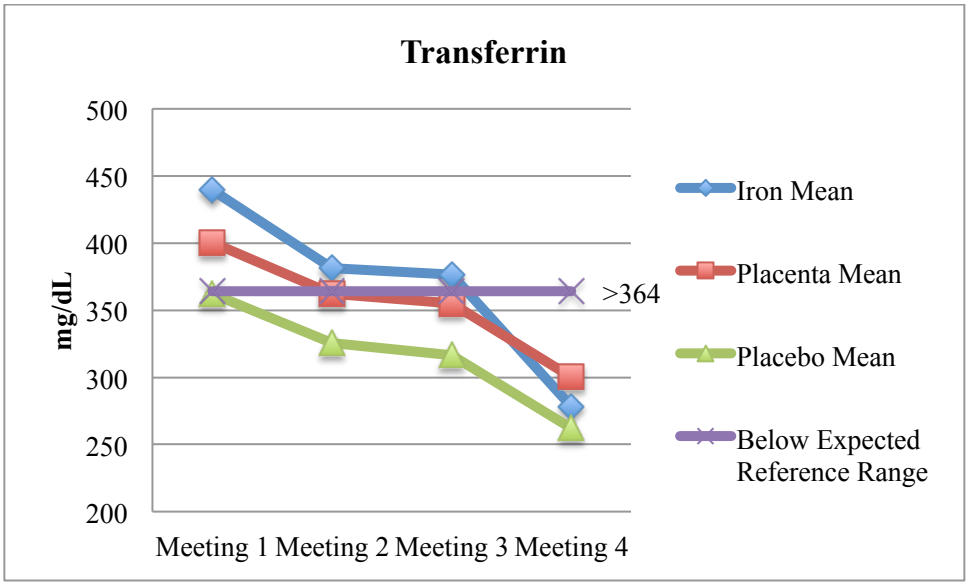

Figure 13: Transferrin means for meetings 1-4 by Condition. Note that transferrin deficiency is indicated by greater than values.

Transferrin								
Condition	Meeting 1		Meeting 2		Meeting 3		Meeting 4	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Iron	439.33	129.20	381.33	113.00	376.67	118.00	278.33	57.18
Placenta	400.42	49.65	362.00	52.00	355.00	51.60	300.16	51.71
Placebo	362.37	62.60	325.44	58.40	316.62	56.20	262.06	46.02

Table 13: Transferrin means and Standard Deviations for meetings 1-4 by Condition.

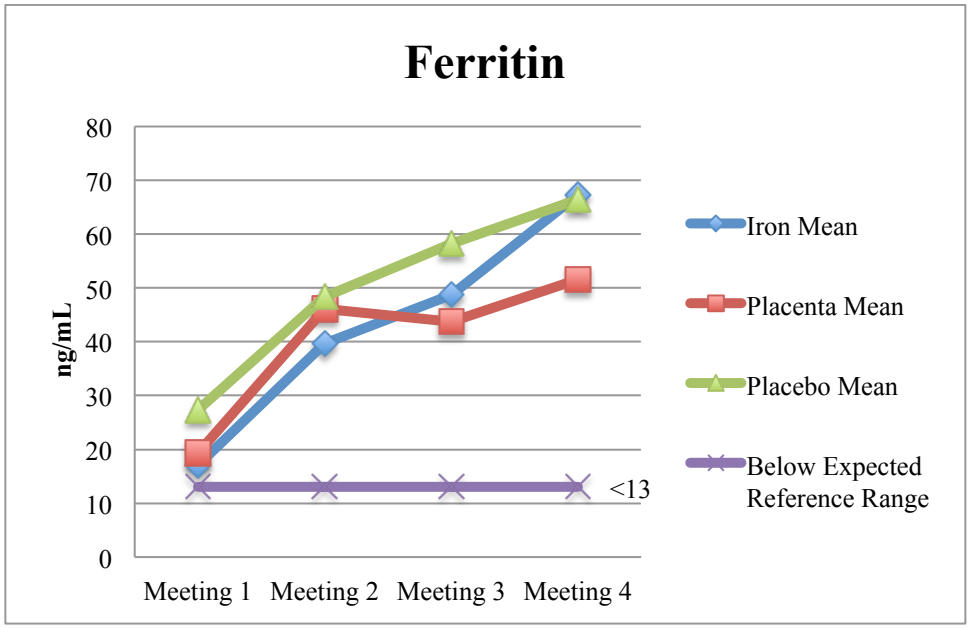

Figure 14: Ferritin means for meetings 1-4 by Condition.

Ferritin								
Condition	Meeting 1		Meeting 2		Meeting 3		Meeting 4	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Iron	17.08	14.68	39.63	25.00	48.73	17.40	67.26	61.36
Placenta	19.34	14.75	46.05	40.70	43.68	35.10	51.57	47.00
Placebo	27.34	36.52	48.41	52.60	58.23	78.40	66.47	66.82

Table 14: Ferritin means and Standard Deviations for meetings 1-4 by Condition.

For the Iron Group, excluding hemoglobin, the means of transferrin and ferritin for each meeting generally trend the same as the Experimental Group and the Placebo Group. Due to the Iron Group's small sample size, it is not possible to draw any conclusions based on statistical analysis. However, from these iron supplement group comparisons,

the data are consistent with this study's null hypotheses (*1a*, *1b*, and *1c*), that the Experimental Group would not show a greater interval increase in iron levels (assessed via hemoglobin, transferrin, and ferritin) as compared to Placebo and Iron Group participants from: (*1a*) baseline (within 72 hours postpartum) to day five-seven postpartum, (*1b*) baseline to week three postpartum, and (*1c*) days five-seven postpartum to week three postpartum.

CHAPTER 6

DISCUSSION

Hypotheses Revisited

Hypothesis 1: Supported

Experimental, Placebo, and Iron Group participants showed an increase in iron levels (assessed via hemoglobin, transferrin, and ferritin) measured during, (a) days five-seven postpartum, and (b) week three postpartum, as compared to baseline measure taken prior to supplement initiation (within 72 hours postpartum).

Hypothesis 1a: Rejected

Experimental Group participants did not show a greater interval increase in iron levels (assessed via hemoglobin, transferrin, and ferritin) as compared to Placebo Group and Iron Group participants from baseline (within 72 hours postpartum) to day five-seven postpartum.

Hypothesis 1b: Rejected

Experimental Group participants did not show a greater interval increase in iron levels (assessed via hemoglobin, transferrin, and ferritin) as compared to Placebo Group and Iron Group participants from baseline to week three postpartum.

Hypothesis 1c: Rejected

Experimental Group participants did not show a greater interval increase in iron levels (assessed via hemoglobin, transferrin, and ferritin) as compared to Placebo Group and Iron Group participants from days five-seven postpartum to week three postpartum.

Data analyses support Hypothesis 1 and fail to support the Null Hypothesis. The statistical analyses conducted to test Hypotheses *1a*, *1b*, and *1c* support the Null Hypotheses and fail to support Hypotheses *1a*, *1b*, and *1c*. The Iron Group failed to recruit a sufficient amount of participants to address this group's role in Hypotheses *1a*, *1b*, and *1c* via statistical analysis.

Public Health Implications

Placentophagy is a growing practice in the Las Vegas Valley and has been spreading within industrialized countries since the 1960s (Young & Benyshek, 2010). High profile celebrities are engaging in this practice, and television programs have featured placentophagy, thereby increasing its public visibility and introducing millions of women to a practice that has not been subjected to rigorous scientific evaluation with respect to its potential health benefits and/or risks.

If some women forgo traditional postpartum iron supplementation in favor of placenta supplementation, it is necessary that any iron present in those supplements be in a bioavailable form and in sufficient amounts to meet the increased iron demands associated with pregnancy and nursing. Iron deficiency and IDA are associated with negative health outcomes for both mother and child, making this issue particularly important. Unfortunately, this study was not able to assess the effect of encapsulated placenta supplementation on the iron status of postpartum mothers suffering from clinically diagnosed iron deficiency or IDA. The current study's results do suggest, however, that encapsulated placenta supplementation neither significantly improves, nor

significantly impairs postpartum maternal iron status for women consuming at least adequate amounts of dietary iron postpartum[1].

Limitations

Given the recruitment limitations associated with a double-blind placebo-controlled study centered upon the ingestion of a participant's own placenta, the participant sample was, as a matter of necessity, self-selecting; not every pregnant women will want to ingest their placenta postpartum. Therefore, the Experimental and Placebo Groups used in this study consisted of a self-selecting population, making true randomization impossible.

Samples sizes for Experimental and Placebo Groups could have been more robust, however the sample sizes were large enough to conduct statistical analyses and draw conclusions about differences and similarities between the two groups. The Iron Group's low sample size (n=3) prevented similar statistical comparisons among all three groups. Instead, Iron Group data were examined as case studies. Due to this we were unable to address portions of this study's research questions related to the efficacy of placenta supplementation in comparison to standard iron supplements regarding recovery from postpartum iron deficiency.

The gold-standard for hemoglobin measurement is the direct cyanmethaemoglobin method analyzed via spectrophotometer. This method, however, requires processing of samples within a few hours of collection as well as access to an appropriately equipped laboratory (Sari, et al., 2001). Given these constraints, the POC STAT-Site Hemoglobin

[1] Only one participant from our sample had an average daily dietary intake of iron below RDA recommendations during lactation.

Meter is an excellent alternative. POC tools are essential for immediate bedside care of patients, and are extremely convenient when used to conduct medical anthropological field research. The accuracy of the STAT-Site Hemoglobin meter is dependent upon the use of blood drops of a particular volume, however. The STAT-Site meter will give erroneous results for drops smaller than 10µL and larger than 30µL (Stanbio Laboratory, 2013). Both slight variation in blood collection technique among project researchers, and variation in the hydration of participants effecting ease of blood flow may lead to differences in the size of blood drops collected and therefore hemoglobin results. In order to reduce this error, whenever possible, two drops were collected and averaged.

For this study, blood samples were collected via finger-stick lancet. Generally, venous blood draws are a more commonly used collection method, and most ranges for analytes are based upon venous blood as opposed to finger-stick capillary blood samples. As mentioned in Chapter 4, capillary blood differs from venous blood in a number of ways and affects some, but not all, analytes. In the case of this study, ranges for expected hemoglobin status were included with the STAT-Site Hemoglobin meter, which are already adapted for capillary blood samples. Transferrin and ferritin ranges, however, are not adapted to the collection method used in this study. Lu et al. (1987) reports that capillary ferritin values may be about 7% higher than venous values. Another study shows that capillary plasma overestimates ferritin by 8.9% in adults when compared to venous plasma (Mejia & Viteri, 1983). All blood samples from this study were collected via finger-stick capillary method. Given that iron status was not included in the participant selection criteria, and that one of the primary goals of this study was to make

relative comparisons between groups, obtaining laboratory values precise enough for diagnostic purposes (e.g., IDA), were not necessary.

Some methods used to analyze data gathered from the Willet FFQ favored the possibility of overestimation of participant dietary iron status. For example, some questions on the Willet FFQ presented participants with a selection of similar foods, but required a single answer for that grouping; one entry asked the frequency of consumption of peaches, apricots, or plums. In cases such as these, each item's iron content was investigated using the USDA National Nutrient Database. The food item with the highest iron content, controlling for portion size, was always chosen. The use of such methods may lead to the overcompensation of dietary iron, however, it is preferable to overestimate dietary iron contributions rather than potentially falsely attribute the fluctuations of hemoglobin, transferrin, and ferritin to supplement ingestion.

CHAPTER 7

CONCLUSION AND RECOMMENDATIONS

Pregnancy, birth, and the postpartum period present considerable physiological challenges to women. Iron deficiency is often a byproduct of the demands associated with human reproduction. Iron deficiency can be particularly problematic for women with high IBIs, those who do not exclusively breastfeed, are obese, or are low income individuals (Allen, 2000; Bodnar, Siega-Riz, Miller, Cogswell, & McDonald, 2002). Iron deficiency can also negatively affect the children of iron deficient mothers (Rasmussen, 2001; Lewis et al., 2002; Golub, Hogrefe, Germann, Capitanio, & Lozoff, 2006; Colomer, 1990). Currently, many maternal placentophagy advocates claim that encapsulated placenta is an excellent source of iron for postpartum mothers (Selander et al., 2013). To date, however, there have been no rigorous scientific studies that have investigated this claim.

The results of this study help fill major gaps of knowledge in the practice of human maternal placentophagy. The current study suggests that encapsulated placenta supplementation neither significantly improves, nor impairs postpartum maternal iron status for women consuming at least adequate amounts of dietary iron. It is possible that participants' placenta was not necessarily a superior source of iron than a beef or soy-free vegetarian placebo supplement of the same daily intake. This may mean that, among women with adequate dietary iron intake, the bioavailability of iron present in the placebo supplement is roughly equal to the bioavailability of iron in the placenta supplement.

OTC oral iron supplements are commonly used by women around the world, and are of particular importance for pregnant and postpartum women experiencing iron deficiency and IDA. One of the original goals of this study was to examine how placenta supplements compared to standard oral iron supplements in order to deduce whether placenta supplements are a viable option for the restoration of women's iron status postpartum, as this alternative form of iron supplementation may appeal to many mothers looking for a more "natural" way to manage postpartum iron deficiency. While the small size of the OTC iron supplement group served only as case studies for comparison to the study's Placenta and Placebo groups, it appears that, among women with adequate dietary iron intake, encapsulated placenta supplements offer neither a superior, nor inferior, source of oral iron supplementation. Unfortunately, the question of whether placenta supplements are superior, equal, or inferior to standard iron supplements for women with postpartum iron deficiency or IDA was beyond the scope of this study.

Future research might include focusing upon clinically diagnosed iron deficient and IDA participants. Such studies would allow researchers to examine how well encapsulated placenta supplements compare to traditional iron supplements among women whose iron status is the most severely compromised. In addition to such clinical studies, the research of placentophagy would benefit from the ethnographic study of women engaging in placentophagy as well as placentophagy advocates. Such ethnographies could examine topics such as women's motivations for engaging in placentophagy, and maternal health decision-making.

APPENDIX

Background Information Questionnaire

Biomedical IRB – Full Board Review Approval Notice

Biomedical IRB – Expedited Review Continuing Review Approved

Background Information Questionnaire

An Investigation of the Effects of Human Maternal Placentophagy in Postpartum Maternal Health and Recovery

University of Nevada, Las Vegas

Please answer the following questions as accurately as possible

Age: _____ Height: _____ ft _____ in Weight: _____ lbs

With which ethnicity do you most closely identify?

__ American Indian/Alaska Native __ Caucasian __ Middle Eastern

__ Asian __ Hispanic/Latina __ Other _____

__ African American __ Hawaiian/Pacific Islander

What is the highest level of education you have completed?

__ Grammar School __ Bachelor's degree __ Doctoral degree

__ High School or equivalent __ Master's degree __ Professional degree (MD, JD, etc.)

__ Some college __ Vocational/technical school

Which of the following income groups includes your total annual family income:

__ Under $10,000 __ $30,001—$40,000 __ $60,001—$70,000

__ $10,000—$20,000 __ $40,001—$50,000 __ $70,001—$80,000

__ $20,001—$30,000 __ $50,001—$60,000 __ Over $80,000

Please indicate your marital status:

__ Single, never married __ Married or domestic partnership

__ Separated __ In a committed relationship, not cohabiting

__ Divorced __ Widowed

Total number of people in the household: _____

Zip Code: _____

Do you currently consume alcoholic beverages? Yes No

If yes, how often (circle one)? daily frequently occasionally rarely

Do you currently smoke cigarettes? Yes No

In the 3 months before your most recent pregnancy, did you take a multivitamin containing folic acid or a folic acid supplement?

Never Rarely Occasionally Almost every day Daily

Is this your first pregnancy? Yes No

If this is not your first pregnancy, how many times have you given live birth? ___

Please list the dates that you have given live birth:

MM / DD / YYYY . MM / DD / YYYY . MM / DD / YYYY .

MM / DD / YYYY . MM / DD / YYYY . MM / DD / YYYY .

MM / DD / YYYY . MM / DD / YYYY . MM / DD / YYYY .

Did you have complications with any of your pregnancies? Yes No

If yes, please explain briefly:

_____

_____

Have you ever ingested placenta, in any form, after any of your pregnancies? Yes No

If yes, after which pregnancy did you ingest placenta? _____

Did you experience any effects that you attributed to placentophagy? Yes No

Please describe: _____

_____

Have you been diagnosed with, or do you suspect you may have an iron-related disorders (*e.g.*, hemochromatosis, iron deficiency, iron deficiency anemia)? Yes No

If yes, was this condition diagnosed by a medical practitioner? Yes No

If yes, please describe: _____

_____

Do you have any chronic health conditions? Yes No

If yes, please describe them here: _____

_____

_____

Have you received a blood transfusion within the past year? Yes No

If yes, when did you receive the most recent transfusion? MM / YY .

Are you currently consuming a special diet (e.g., vegan, vegetarian, etc.)? Yes No

If yes, please explain: _____

_____

Did your diet changed after learning that you were pregnant? Yes No

If yes, please explain how?

_____

_____

_____

How much has your diet changed since pre-pregnancy (circle one)?

drastically moderately minimally none

Are you currently taking any medications? Yes No

If yes, please list the medication(s) you are taking: _____

_____

_____

Are you currently taking any nutritional supplements (including multivitamins): Yes No

If yes, please list them here, and indicate the brand and dose: _____

_____

_____

_____

Please mark any of the following conditions that you have experienced, and indicate the severity and whether the condition was diagnosed by a medical professional:

___	Depression	Mild	Moderate	Severe	___	Diagnosed
___	Antenatal (pregnancy) depression	Mild	Moderate	Severe	___	Diagnosed
___	Postnatal depression	Mild	Moderate	Severe	___	Diagnosed
___	Maternity blues	Mild	Moderate	Severe	___	Diagnosed
___	Premenstrual dysphoric disorder (PMDD)	Mild	Moderate	Severe	___	Diagnosed
___	Anxiety	Mild	Moderate	Severe	___	Diagnosed
___	Bipolar disorder	Mild	Moderate	Severe	___	Diagnosed
___	Schizophrenia	Mild	Moderate	Severe	___	Diagnosed
___	Anemia	Mild	Moderate	Severe	___	Diagnosed
___	Insomnia/sleep disorder	Mild	Moderate	Severe	___	Diagnosed
___	Thyroid disorder	Mild	Moderate	Severe	___	Diagnosed
___	Endocrine disorder	Mild	Moderate	Severe	___	Diagnosed
___	Reproductive disorder (e.g., PCOS, endometriosis)	Mild	Moderate	Severe	___	Diagnosed

Do you have an allergy or sensitivity to Ferrous sulfate (iron)? Yes No

What is the starting date of your last menstrual period? _MM_/_DD_/_YY_

What is your projected due date? _MM_/_DD_/_YY_

Where do you plan to give birth?

__ In a hospital with a physician __ At home

__ In a hospital with a midwife __ At a birthing center

__ Other Please describe: _____

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Biomedical IRB – Full Board Review
Approval Notice

NOTICE TO ALL RESEARCHERS:

Please be aware that a protocol violation (e.g., failure to submit a modification for any change) of an IRB approved protocol may result in mandatory remedial education, additional audits, re-consenting subjects, researcher probation, suspension of any research protocol at issue, suspension of additional existing research protocols, invalidation of all research conducted under the research protocol at issue, and further appropriate consequences as determined by the IRB and the Institutional Officer.

DATE: July 23, 2013

TO: **Dr. Daniel Benyshek**, Anthropology

FROM: Office of Research Integrity - Human Subjects

RE: Notification of IRB Action
Protocol Title: **An Investigation of the Effects of Human Maternal Placentophagy on Postpartum Health and Recovery**
Protocol #: 1305-4465M
Expiration Date: July 22, 2014

This memorandum is notification that the project referenced above has been reviewed and approved by the UNLV Biomedical Institutional Review Board (IRB) as indicated in Federal regulatory statutes 45 CFR 46 and UNLV Human Research Policies and Procedures.

The protocol is approved for a period of one year and expires July 22, 2014. If the above-referenced project has not been completed by this date you must request renewal by submitting a Continuing Review Request form 60 days before the expiration date.

PLEASE NOTE:
Upon approval, the research team is responsible for conducting the research as stated in the protocol most recently reviewed and approved by the IRB, which shall include using the most recently submitted Informed Consent/Assent forms and recruitment materials. The official versions of these forms are indicated by footer which contains approval and expiration dates.

Should there be *any* change to the protocol, it will be necessary to submit a **Modification Form** through ORI - Human Subjects. No changes may be made to the existing protocol until modifications have been approved by the IRB. Modified versions of protocol materials must be used upon review and approval. Unanticipated problems, deviations to protocols, and adverse events must be reported to the ORI – HS within 10 days of occurrence.

If you have questions or require any assistance, please contact the Office of Research Integrity - Human Subjects at IRB@unlv.edu or call 895-2794.

Office of Research Integrity - Human Subjects
4505 Maryland Parkway • Box 451047 • Las Vegas, Nevada 89154-1047
(702) 895-2794 • FAX: (702) 895-0805

UNLV
UNIVERSITY OF NEVADA LAS VEGAS

Biomedical IRB – Expedited Review
Continuing Review Approved

NOTICE TO ALL RESEARCHERS:

Please be aware that a protocol violation (e.g., failure to submit a modification for any change) of an IRB approved protocol may result in mandatory remedial education, additional audits, re-consenting subjects, researcher probation suspension of any research protocol at issue, suspension of additional existing research protocols, invalidation of all research conducted under the research protocol at issue, and further appropriate consequences as determined by the IRB and the Institutional Officer.

DATE: June 19, 2014
TO: **Dr. Daniel Benyshek**, Anthropology
FROM: Office of Research Integrity – Human Subjects
RE: Notification of IRB Action
Protocol Title: **An Investigation of the Effects of Human Maternal Placentophagy on Postpartum Health and Recovery**
Protocol #: 1305-4465M
Expiration Date: June 18, 2015

Continuing review of the protocol named above has been reviewed and approved.

This IRB action will reset your expiration date for this protocol. The protocol is approved for a period of one year from the date of IRB approval. The new expiration date for this protocol is June 18, 2015. If the above-referenced project has not been completed by this date you must request renewal by submitting a Continuing Review Request form 30 days before the expiration date.

PLEASE NOTE:
Upon approval, the research team is responsible for conducting the research as stated in the protocol most recently reviewed and approved by the IRB, which shall include using the most recently submitted Informed Consent/Assent forms and recruitment materials. The official versions of these forms are indicated by footer which contains current approval and expiration dates.

Should there be *any* change to the protocol, it will be necessary to submit a **Modification Form** through ORI - Human Subjects. No changes may be made to the existing protocol until modifications have been approved by the IRB. Modified versions of protocol

materials must be used upon review and approval. Unanticipated problems, deviations to protocols, and adverse events must be reported to the ORI – HS within 10 days of occurrence.

If you have questions or require any assistance, please contact the Office of Research Integrity - Human Subjects at IRB@unlv.edu or call 895-2794.

Office of Research Integrity - Human Subjects
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(702) 895-2794 • FAX: (702) 895-0805

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CURRICULUM VITAE

Graduate College
University of Nevada, Las Vegas

Laura Kathleen Gryder

Home Address:
1142 Hunters Peak ct.
Henderson, NV 89052

Degrees:
Bachelor of Arts, Anthropology and German, 2008
University of Nevada, Las Vegas

Special Honors and Awards:

Spring 2014	UNLV Graduate & Professional Student Association (GPSA) Grant
Spring 2014	UNLV Patricia Rocchio Memorial Scholarship
Fall 2013	UNLV Anthropology Society Scholarship
Fall 2013	Edwards and Olswang Scholarship
2013-2014	Patricia Sastaunik Scholarship
2013-2014	UNLV Alumni Association Scholarship
May 2013	UNLV's First Annual Anthropology Research Forum (ARF) Honorable Mention: graduate student paper presentation
Summer 2013	UNLV Graduate & Professional Student Association (GPSA) Grant
Fall 2012	Edwards and Olswang Scholarship
2012-2013	Patricia Sastaunik Scholarship
2012-2013	UNLV Alumni Association Scholarship
Fall 2012	UNLV Angela Nicole Peterson Scholarship
Spring 2008	UNLV Outstanding German Major
Fall 2008	Dee Smith Endowment Scholarship
Fall 2003	Millennium Scholarship

Thesis Title:
Effects of Human Maternal Placentophagy on Maternal Postpartum Iron-Status

Thesis Examination Committee:
Chairperson, Dr. Daniel C. Benyshek, Ph.D.
Committee Member, Dr. Alyssa Crittenden, Ph.D.
Committee Member, Dr. Peter Gray, Ph.D.
Graduate Faculty Representative, Dr. James Navalta, Ph.D.