MATERNAL WEIGHT AND DIET AS PROTECTIVE FACTORS AGAINST THE ADVERSE EFFECTS OF PRENATAL ALCOHOL EXPOSURE

Julie M. Hasken

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Approved by:

Linda S. Adair

Stephanie L. Martin

Amanda L. Thompson

Julie L. Daniels

Philip A. May

ABSTRACT

Julie M. Hasken: Maternal Weight and Diet as Protective Factors Against the Adverse Effects of Prenatal Alcohol Exposure
(Under the direction of Philip A. May)

Background: Alcohol is a known teratogen, and the prevalence of fetal alcohol spectrum disorders in the Western Cape Province of South Africa is estimated to be 17 - 28%. Yet the individual variation in child outcomes is not fully explained by the quantity, frequency, or gestational timing of prenatal alcohol exposure.

Methods: We examined the influence of maternal weight on the physical and neurocognitive development of infants with and without prenatal alcohol exposure. We compared the physical growth, dysmorphology, and neurocognitive trajectories of infants to understand similarities and differences in birth measurements and rate of change, from birth to 9 months, associated with alcohol exposure and maternal weight. We also examined the role of alcohol consumption and maternal dietary intake on infant physical development in early life.

Results: In this population where stunting remains a concern, higher maternal weight was associated with larger, less dysmorphic, infants with better neurodevelopmental outcomes. But the rate of change over time was similar among all infants regardless of maternal weight. Alcohol exposure consistently resulted in poorer growth and more dysmorphic infants. Most women in this population were not achieving adequate micronutrient intake for pregnant women and malnutrition remains a concern for this population. Alcohol had a direct adverse effect on maternal dietary intake.

Conclusion: This research attempted to better understand maternal weight and dietary intake as factors which may mitigate some of the adverse effects of prenatal alcohol exposure in the early infancy period. Alcohol was adversely associated with maternal dietary intake and infant outcomes. Maternal weight may be somewhat protective and may partially explain some of the individual variation in infant physical and neurocognitive outcomes, but higher maternal weight does not overcome the majority of the negative, teratogenic effects of prenatal alcohol exposure. These studies affirm that there is no known safe level of alcohol exposure during pregnancy.

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ARBD	Alcohol-Related Birth Defect
ARND	Alcohol-Related Neurodevelopmental Disorder
AUDIT	Alcohol Use Disorders Identification Test
BMI	Body Mass Index
BAC	Blood Alcohol Concentration
CDC	Centers for Disease Control and Prevention
DDD	Drinks per Drinking Day
DHA	Docosahexaenoic Acid
FAS	Fetal Alcohol Syndrome
FASD	Fetal Alcohol Spectrum Disorders
ICD	Inner Canthal Distance
IPD	Interpupillary Distance
NDSR	Nutrient Data System for Research
OFC	Occipitofrontal Circumference
PFAS	Partial Fetal Alcohol Syndrome
PFL	Palpebral Fissure Length
SD	Standard Deviation
TACE	Tolerance, annoyed, cut-down, eye-opener
WHO	World Health Organization

CHAPTER 1: INTRODUCTION

Overview

The prevalence of fetal alcohol spectrum disorders (FASD) has been estimated to be 1-5% in communities of the United States and 17 - 28% in the Western Cape Province of South Africa.[1–9] Yet there are many unanswered questions about maternal risk factors that affect the susceptibility and severity of infant outcomes associated with prenatal alcohol exposure.

We examined the influence of maternal weight and dietary intake during pregnancy on the development of infants with and without prenatal alcohol exposure. We compared the physical growth trajectories of infants to understand similarities and differences in birth measurements and the rate of change from birth to 9 months which is associated with alcohol exposure and maternal weight. Next, we compared the cognitive and behavioral abilities of infants to understand both the influence of prenatal alcohol exposure and maternal weight on infant neurodevelopmental outcomes. Finally, we examined the role of maternal dietary intake and alcohol consumption during pregnancy on the physical development of infants.

We hope this work will add to the current understanding of FASD etiology and the maternal risk factors associated with an FASD diagnosis. This work demonstrated that in a population, where alcohol consumption during pregnancy and undernutrition are commonplace, increased maternal weight was associated with better physical and neurodevelopmental outcomes. Alcohol consumption had a direct and negative impact on both maternal dietary intake and infant physical outcomes. Our findings, coupled with ongoing efforts to better understand the individual variation in outcomes in children with prenatal alcohol exposure, can help inform

future screening and intervention efforts to prevent and/or remediate the adverse effects of prenatal alcohol exposure.

Objectives and Specific Aims

Many children with FASD go unrecognized and/or undiagnosed due to a variety of reasons, including: the lack of reliable alcohol consumption information during pregnancy, few physicians who are willing or capable of recognizing or diagnosing FASD, the lack of consistent growth impairments over time, the timing of onset of observable neurocognitive delays, and the rate of change within and across diagnoses within the FASD continuum.[10,11] Both alcohol exposure and maternal obesity may independently predispose a child to poorer developmental trajectories. Severe maternal malnutrition can also adversely affect child development.[12] However, less is known about the subtle variations in maternal micronutrient intake, co-occurring with alcohol consumption during pregnancy, as it relates to infant outcomes. Since growth, dysmorphology, and neurodevelopmental abilities are key diagnostic features of an FASD diagnosis, it is necessary to understand how alcohol exposure, maternal weight, maternal micronutrient intake, and their possible interactions influence the developmental trajectories of children.

For over two decades, research on the prevalence and etiology of FASD has been ongoing in the Western Cape Province of South Africa. The 25-year history of working with this population has led to much of the literature on the physical and neurocognitive abilities of children with FASD being first described through these research efforts in South Africa. Several maternal risk factors associated with FASD, including the quantity, frequency, and timing of alcohol exposure as well as distal maternal risk factors such as maternal age and gravidity/parity, have been described in these communities. This work has resulted in a rich context for exploring etiological questions regarding the importance of maternal weight and dietary intake to the

severity and nature of infant outcomes in an alcohol-exposed population. The specific aims presented here are a natural progression from the previous years of work with this population.

Aim 1: Determine whether maternal weight, measured immediately following delivery, partially mitigates the effect of alcohol exposure on child growth and dysmorphology trajectories from birth to 9 months. We hypothesize that maternal weight mitigates some of the effects of alcohol exposure such that children with prenatal alcohol exposure born to heavier mothers will be larger (in length, weight, and head circumference) and be less dysmorphic compared to children with alcohol exposure born to lighter mothers.

Aim 2: Examine whether maternal weight, measured after delivery, moderates the relationship between alcohol consumption and infant neurodevelopmental outcomes from 6 weeks to 9 months of age. We hypothesize that postpartum maternal weight will mitigate some of the negative effects of alcohol exposure. We anticipate that children with prenatal alcohol exposure born to heavier mothers will perform better at 6 weeks and will have a greater (steeper) increase in the rate of change on neurocognitive abilities through 9 months, compared to children with alcohol exposure born to lighter mothers.

Aim 3: Determine whether the absolute grams of alcohol intake during pregnancy is associated with maternal micronutrient intake and whether the intake of micronutrients is associated with child physical outcomes at 6 weeks. We hypothesize that maternal micronutrient intake mediates the effect of alcohol on child physical outcomes such that alcohol intake leads to lower micronutrient intake which will be associated with poorer child physical outcomes.

The South African Context

The Western Cape Province of South Africa provides a unique population for an investigation into maternal weight, dietary intake, and FASD outcomes. In terms of alcohol consumption, prevalence of FASD, race/ethnicity, and social/cultural practices, South Africa has

provided an important venue in which to investigate this type of inquiry. In the Western Cape there is little stigma around drinking in general, even during pregnancy, with a high proportion (35-50%) of women of childbearing age regularly binge drinking most Friday and Saturday nights.[13,14] Approximately 55% of pregnant women reported drinking during pregnancy and 30% reported drinking in all trimesters.[15] This has led, in part, to the Western Cape having the highest documented prevalence of FASD in the world.[2–9] The high prevalence of FASD has been partially attributed to the 'Dop system' where farmworkers historically received alcohol as partial compensation for their labor. This practice has been outlawed for decades and is virtually nonexistent in present day, but the historical normative, drinking culture established under the Dop system remains common.[16,17]

In addition to the historical, normative patterns of alcohol consumption, the use of other illicit substances such as marijuana, methamphetamine, or heroin are virtually non-existent in these communities at this time. In recent, cross-sectional, FASD prevalence studies conducted in these communities, only 2-3% of women reported any illicit or other drug use during pregnancy, with marijuana ('dagga') or methamphetamine ('tik') being the most commonly used.[7,8] Tobacco use is a commonly reported behavior during pregnancy; however, the grams of tobacco per day has been relatively low at 2-3 grams or cigarettes per day in past investigations.[7,8] Therefore, unlike other populations which may have comorbid use of alcohol and other drugs, the teratogenic exposure in these communities is primarily alcohol.

In this study population, women have been found to be accurate at recalling and reporting alcohol consumption,[13,15,18–20] possibly due to the social norms around drinking.[21] Consuming alcohol with peers is one of the few forms of recreation for many women in these communities; therefore, drinking on weekends is a valued and substantial social and economic investment for

many.[13,15,19–23] Because most drinking occurs in a relatively structured pattern in small groups, and because individuals must allocate some of their very limited financial resources to purchase alcohol, most respondents can recall their alcohol use accurately. The high proportion of women who consume alcohol during pregnancy has led to the Western Cape Province having the highest documented FASD prevalence in the world: 17-28% of the general population of 1st grade children.[2–9]

Also, partially due to limited resources, the diet diversity and nutritional intake is similar and sub-optimal for many women of childbearing age.[24,25] National South African studies report 19% of households in South Africa and 11.6% of Western Cape households experience food insecurity.[26] The dietary staples include meat, stews (potatoes, onions, cabbage), tea with whole milk and sugar, porridge, white rice, and white bread with margarine. Many consume one main meal and have tea with slices of white bread with margarine as needed throughout the day. Moreover, many women of childbearing age in the Western Cape Province consume less than the US Institute of Medicine's Dietary Reference Intakes for virtually all micronutrients.[24,25] The Western Cape Province also has the highest obesity rate in South Africa.[27]

Therefore, the Western Cape Province of South Africa represents a unique population where alcohol consumption during pregnancy is common and the prevalence of FASD is high, which enables us to address etiologic questions about whether maternal weight and dietary intake are protective factors for lessening the severity of FASD. Such an inquiry would be difficult or nearly impossible in most other populations.

Previous FASD Prevalence and Prevention Efforts in the Western Cape Province

Since 1997, there have been ongoing studies on FASD etiology and prevention by a multidisciplinary, bi-national group of researchers. The first decade of research focused almost exclusively on determining the prevalence and characteristics of FASD through cross-sectional

studies. These studies included descriptions of child physical and neurobehavioral outcomes as well as identifying maternal risk factors associated with FASD. From these nine population-based, in-school studies of the prevalence of FASD, several distal maternal risk factors were identified. These risk factors include advanced maternal age, increased gravidity/parity, later birth order of the child, lower socioeconomic status, lower educational attainment, living in a rural environment, being unmarried, and less adherence to a formal religion.[13,19] Many of these, now widely accepted, maternal risk factors were first described in this Western Cape Province population,[8,19] but have since been shown to be maternal risk factors in other populations including Italy,[28,29] Ukraine,[30] Canada,[31] and the United States.[1]

The second decade of FASD research in the Western Cape focused on refining the criteria for diagnosing the full continuum of FASD, initiating prevention efforts, implementing early interventions for infants, and undertaking alcohol biomarker and nutrition research. The research attempted to identify modifiable behavioral and community-level determinants, drive down the age at which an accurate diagnosis of FASD can be made, and develop early interventions to help remediate the adverse effects of prenatal alcohol exposure. Through these previous and ongoing research endeavors, strategic partnerships have been cultivated with the prenatal clinics in these communities.

Sample

In 2014-2016, a cohort of pregnant women was recruited in antenatal clinics in five communities (and their surrounding rural areas) of the wine-growing region of the Western Cape Province of South Africa. The community populations range from 10,000 – 55,000 residents with approximately 90% living in formal dwellings.[32] All women seeking prenatal care in community clinics, including mobile clinics, were invited to participate in the study. After consent was obtained, pregnant women participated in a screening interview which included the

10-item Alcohol Use Disorders Identification Test (AUDIT)[33] and reported alcohol consumption for the previous 30 days of the pregnancy. Infants were assessed for physical growth and dysmorphology and neurodevelopmental abilities at 6 weeks and 9 months. The sample is predominately Afrikaans speaking of mixed race ('Cape Coloured') ancestry. The mother/infant dyads which did not remain in the study through 9 months were predominately women who abstained from alcohol during pregnancy. However, approximately 25% of the mothers in the final sample reported alcohol consumption during pregnancy and 75% of mothers were abstainers. These proportions of alcohol consumption and abstention are consistent with previous research in these communities.[2–9]

Maternal Height, Weight, and BMI in the Western Cape Province of South Africa

Because height and weight are used to calculate body mass index (BMI), height and weight are important indicators of population health. In nine cross-sectional, in-school studies of the prevalence of FASD, maternal height, weight, and BMI seven years postpartum have been measured. Maternal height, weight, and BMI seven years postpartum significantly differentiated women who gave birth to children with FASD from mothers who gave birth to children with typical development.[6–9,13,19] On average, women who gave birth to children with FASD were shorter, weighed less, and had a lower BMI compared to women who gave birth to children with typical development. Mothers of children with fetal alcohol syndrome (FAS) were generally the shortest, lightest, and had the lowest BMI compared to mothers of children with partial FAS (PFAS) and alcohol-related neurobehavioral disorders (ARND). The group mean difference in weight between mothers of children with FASD and mothers of children with typical development was greater than the group mean difference for height or BMI. Mothers of children with FASD were, on average, 154 -158 centimeters tall, weighed 53 -59 kilograms, and had a BMI of 21-26. On average, mothers of children with typical development were 157 -160

centimeters tall, weighed 67- 76 kilograms, and had a BMI of 26-28.[6–8,13,19] Women in the Western Cape, on average, may be slightly shorter (mean: 158 centimeters) and lighter (mean: 65 kilograms) compared to US female norms (162 centimeters for height and mean:77 kilograms in weight).[34] Regardless of a difference in scale, both maternal weight and BMI have consistently and significantly differentiated mothers of children with FASD and mothers of children with typical development in South Africa.

Outcome Measures: Growth and Dysmorphology

American pediatricians, who are board-certified as clinical geneticists/dysmorphologists, trained South African research staff to complete physical dysmorphology examinations of infants at 6 weeks and 9 months of age. The dysmorphology exam includes measuring a child's length/height, weight, and occipitofrontal (head) circumference (OFC). Length was measured using an infant length board with 0.1 centimeter precision and weight was measured using a digital scale with 0.01 kilogram precision. OFC was measured with a flexible tape measure and was measured to the nearest millimeter. OFC is the largest circumference of the head measured from the occiput (the most prominent point on the back of the head) to the supraorbital ridges (directly above the eyebrows). The presence or absence of the three cardinal facial features of FASD were also assessed. The three cardinal facial features of FASD are short palpebral fissure lengths (PFL, eye opening), smooth philtrum, and thin vermilion border of the upper lip. The presence or absence of 12 other minor anomalies were also assessed, in addition to measuring other facial features (e.g., inner canthal distance (ICD) and inner pupillary distance (IPD)).[35,36] PFL, ICD, and IPD were measured using a clear, plastic ruler held at a 45-degree angle to capture the full length of the measurement and allow for the natural curvature of the face. The ICD measures the distance between the left and right inner canthus (inner corner of the eye). The IPD measures the distance between the middle of the left and right pupil. The PFL measures

the distance between the inner and outer canthus (outer corner) of the eye. The evaluators were blinded to a child's in utero alcohol exposure history and findings from any previous study assessments (physical or developmental).

Following each dysmorphology exam, centiles were calculated for each growth measure. The South African government has previously adopted the CDC growth curves as national South African norms. Moreover, for children under the age of 2, the CDC has adopted the World Health Organization growth curves.[37] The sex-specific CDC/WHO growth curves were used to determine each infant's growth centile for length and weight. OFC measurements were plotted against growth charts developed by Nellhaus.[38] PFL measurements were plotted on curves developed by Thomas et al. with $\leq 10^{th}$ centile considered short.[39] A total dysmorphology score was also calculated based on the presence or absence of physical characteristics of the child. The total dysmorphology score ranges from 0-32 (Hoyme et al., 2005).[36] The total dysmorphology score has proven to be a useful research tool for differentiating children with FASD and children with typical development.[6–8] However, the total dysmorphology score is not intended to be diagnostic such that a certain total dysmorphology score denotes a specific diagnosis within the FASD continuum.

Outcome Measures: The Bayley Scales of Infant and Toddler Development, 3rd Edition

The Bayley Scales of Infant and Toddler Development is a standardized tool designed to assess development on four domains: cognitive, language, motor, and social/emotional for children aged 1 to 42 months.[40] The theoretical underpinnings of the Bayley are derived from classic themes of child development first put forth by Piaget, Vygotsky, and others, and the Bayley includes components of pretend play, novelty preference, number concepts, and preverbal intelligence. The Bayley is designed to identify strengths and weaknesses of a child and to identify children with developmental delays. The cognitive, language, and motor domains

are assessed through direct interaction with the child, whereas the social/emotional domain is assessed by the primary caregiver via a standardized questionnaire. Each domain is summarized by a raw score which can be converted into a composite score (mean=100, standard deviation=15) and a percentile rank (0-100).

The cognitive, language, and motor scales were normed using a stratified sample of 1700 children ranging from 1 to 42 months. The reference population was representative of the 2000 US Census Bureau population in terms of sex, race/ethnicity, geographic region, and parental education. Approximately 10% of the reference population was known to have mental, physical, or behavioral difficulties. The social/emotional scale was normed on a separate population of 456 children. The Bayley has been widely used internationally and has been shown to be a reliable tool, specifically with a South African population.[41]

The cognitive scale consists of 91 items. Initial items on the cognitive scale focus on response to external stimuli and object permanence and advance to imitation and pretend play. The cognitive domain assesses how infants think and respond to external stimuli. The language scale is comprised of two subscales: receptive and expressive communication. The receptive subscale, which contains 49 items, assesses the child's auditory acuity and ability to comprehend and respond to verbal stimuli. The expressive subscale with 48 items measures the child's ability to babble/use gestures (pre-verbal communication), vocalize, name objects, and communicate with others. The motor scale assesses both fine and gross motor function. The fine motor subscale contains 66 items and assesses skills associated with eye movement, perception-motor integration, and motor speed. The gross motor subscale has 72 items and measures limb and torso static positioning and movement. The social/emotional domain contains 35 items where the primary caregiver assesses whether the child does or does not demonstrate a specific behavior on

a 6-point scale ranging from 0 (‘cannot tell’), 1 (‘none of the time’) to 5 (‘all of the time’). On each scale, the examination begins at the designated starting point for the child’s chronological age, but the starting point may be adjusted to establish a basal level. The first three items administered must be answered correctly to establish the basal level. The ceiling is reached, and the assessment is stopped, when the child receives no credit on five consecutive items on each scale. The entire Bayley can be completed in approximately 50 minutes for children under 1 year of age. The test-retest correlation was found to be greater than 0.80 across all ages of the Bayley for the cognitive, language, and motor scales.

CHAPTER 2: LITERATURE REVIEW

Alcohol is a known teratogen. It is well established that alcohol can freely cross the placenta. Alcohol crossing the placenta has a direct effect on fetal tissue, physiology, function, and development. Alcohol exposure in the prenatal period is the necessary cause of fetal alcohol spectrum disorders (FASD). All children within the FASD continuum have physical dysmorphology and cognitive and/or behavioral impairments. The most severely physically affected children have fetal alcohol syndrome (FAS), followed by partial fetal alcohol syndrome (PFAS), alcohol-related neurobehavioral disorders (ARND), and alcohol-related birth defects (ARBD).[35,36,42] Conservative estimates have reported that 5% of children in the general US population fall within the FASD continuum.[1] South Africa has the highest documented prevalence of FASD, ranging from 17-28% in the general population.[2–9]

Clinical Diagnostic Guidelines for FASD

According to the most recent Updated Clinical Guidelines for Diagnosis of FASD (Hoyme et al., 2016),[35] children with FAS must have: A) growth deficiencies ($\leq 10^{th}$ centile) in length/height and/or weight; and B) small ($\leq 10^{th}$ centile) head circumference (OFC); and C) at least 2 of 3 cardinal facial features (smooth philtrum, narrow vermilion of the upper lip, short ($\leq 10^{th}$ centile) palpebral fissure lengths (PFL)); and D) documented neurocognitive and/or behavioral impairments. Children with PFAS must have: A) growth deficiencies; and C) at least 2 of 3 cardinal facial features; and D) neurocognitive and/or behavioral impairments. Due to the specificity of the cardinal facial features to FAS and PFAS and a number of other co-occurring minor anomalies, under the Updated Clinical Guidelines for Diagnosis of FASD, a diagnosis of

FAS and PFAS can be made by an experienced pediatrician without confirmed prenatal alcohol exposure. Children with ARND do not have 2 of the 3 cardinal facial features, but they must have documented prenatal alcohol exposure and demonstrate neurocognitive and/or behavioral impairments. Children with ARND can be, and often are, growth deficient or have a small OFC, but these are not required for a diagnosis of ARND. Children with ARBD have known prenatal alcohol exposure and have a major physical malformation, but they lack neurocognitive impairments. A diagnosis of ARBD is rare in most populations, because isolated morphological changes due to alcohol without neurocognitive and/or behavioral impairments do not occur frequently.[35]

Fetal Development with Prenatal Alcohol Exposure

The foundation and precursors of the central nervous system, heart, limbs, eyes, ears, and teeth/palate are formed within the first 8 weeks following fertilization. Prenatal alcohol exposure impairs neurogenesis (3-6 weeks post-fertilization) through altering induction, expansion, apoptosis, migration, and differentiation of the neural crest and its derivatives. These alterations can result in the 'classic' FASD face which is characterized by a smooth philtrum, thin vermilion of the upper lip, and short PFL. These neurogenesis alterations can also lead to other organ-specific defects.[43–45] Key brain structures (e.g., the hippocampus and cerebellum) do not fully form until the third trimester making the central nervous system vulnerable throughout pregnancy, and the fetus remains at risk for prenatal growth restrictions. Therefore, prenatal alcohol exposure can have consequences at any point during pregnancy, and there is no known safe level of alcohol consumption during pregnancy.[46]

Physical Dysmorphology Associated with Prenatal Alcohol Exposure

Early seminal work showed there was a significant positive association between maternal blood alcohol concentration (BAC) and developmental malformations.[47–50] In addition to the

growth deficiencies and the cardinal facial features of FASD, many other minor anomalies are associated with prenatal alcohol exposure. Depending on the quantity, frequency, and gestational timing of the prenatal alcohol exposure, physical malformations can occur in the cardiac, skeletal, renal, ophthalmic, auditory, and neurologic systems.[35] Common minor anomalies associated with FASD are: shorter ICD, shorter IPD, ptosis, epicanthal folds, flat nasal bridge, prognathism, long philtrum length, hypoplastic fingernails, clinodactyly of the fifth finger, camptodactyly, altered palmar creases, "railroad track" or "cupped" ears, and heart murmurs/malformations.[35,36,51–53] Children with prenatal alcohol exposure, but who do not meet criteria for an FASD diagnosis, have been shown to have altered physical features with reductions in ear length, facial depth, and frontal face width.[54] The observable facial changes correlate strongly with adverse neurobehavioral outcomes.[54]

Brain imaging studies have demonstrated that individuals with FASD have reduced total brain volume,[55] abnormal (thicker) cortices in the frontal, temporal, and parietal regions,[56,57] reduced callosal thickness,[58] reductions in white and grey matter,[55,59] reduced volume of the hippocampus,[60] reduced basal ganglia size,[61] and altered network connectivity.[55,62] These abnormalities are associated with altered cognitive abilities and are clinically relevant.[63–65] Using 3D imaging, facial asymmetry distinguished between individuals with FAS and unexposed controls.[66] Heavily alcohol-exposed children who do not meet criteria for an FASD diagnosis have facial depressions of the midface and lip/philtrum formations more similar to children with FAS/PFAS than compared to facial characteristics of unexposed controls.[67]

The total dysmorphology score, a weighted score of the presence or absence of observable minor anomalies during a routine clinical exam, has been developed as a research tool to distinguish among children with prenatal alcohol exposure. Multiple population-based

studies in the United States,[68–72] Italy,[28,73] and South Africa[2–9] have consistently shown that the total dysmorphology score significantly distinguishes between diagnostic categories within the FASD continuum and control children. Some studies have shown that the total dysmorphology score significantly differentiated between exposed and unexposed control children.[74]

Neurocognitive and Behavioral Characteristics Associated with Prenatal Alcohol Exposure

Children with prenatal alcohol exposure and/or FASD have been described as having a constellation of behavioral characteristics with individual variation in specific attributes manifested in any one child due to the variation in the quantity, frequency, and timing of alcohol exposure.[75] Using an ultrasound between 37 and 40 weeks of gestation, acute maternal alcohol consumption resulted in reductions in fetal heart rate, eye movement, breathing, and general body movement.[76] Neonates with prenatal alcohol exposure have been shown to have decreased arousal,[77,78] orientation,[79] habituation,[77] muscle tone,[80] and abnormal reflexes.[81] While higher quantities and more frequent alcohol exposure were associated with poorer outcomes in neonates,[78] poorer arousal was associated with even very low alcohol exposure.[82,83]

In the infancy period, global developmental delays and impairments among infants with prenatal alcohol exposure have been reported in Western[84–86] and non-Western[87,88] populations, including South Africa.[89,90] Poorer self-regulation behaviors such as irritability,[86,91] poor self-soothing/monitorting,[92] and sleeping problems[93] are also commonly reported among children with FASD and/or prenatal alcohol exposure. By 6 months of age, poorer visual acuity has been documented among infants with prenatal alcohol exposure compared to unexposed infants.[94,95] Gross and fine motor skills may also be delayed during infancy.[96] However, some studies in Western populations have not found an association between light, infrequent alcohol consumption and infant neurocognitive outcomes within the first year of life.[94,97,98] Yet other

studies have found that children with prenatal alcohol exposure perform within the normal range in early life but perform significantly more poorly in later years.[99]

By pre-school age, delays in cognitive and behavioral domains continue and present as lower scores in full IQ,[100] poor executive function,[101] impulsive behavior,[102] emotional dysregulation,[102] inattentiveness,[102] and motor difficulties.[103] As children age, the tools available to assess a child's development become more refined and sophisticated, thereby allowing for more specific identification of areas of delay. By elementary age, children with FASD and/or prenatal alcohol exposure may present as having a lower IQ,[7,8] poor executive function,[69–71] hyperactivity,[69–72] impulsivity,[69–72] emotional dysregulation,[104] peer-relationship challenges,[69–72] visual-motor deficits,[69,70] poor gross motor skills,[105] abnormal sleeping behaviors,[106] and abnormal eating patterns.[107] Many studies have suggested that these delays and deficits become more pronounced as a child ages into adolescence and adulthood.[108]

Heterogeneity in Outcome

Yet even within diagnostic categories within the FASD continuum, there is heterogeneity in child outcomes that is not fully explained by the quantity, frequency, or timing of alcohol consumption.[15] Similar quantities of alcohol exposure in the prenatal period can result in one child within a diagnostic category performing more poorly than another with the same diagnosis. Or given similar exposure, one child may be diagnosed on the spectrum while another child may be found developing within the normal range. Maternal weight has been suggested as a potential contributor to, or ameliorator of, the effects of prenatal alcohol exposure.[109–113]

Case-control comparisons in numerous cross-sectional studies have demonstrated that mothers of children with FASD had lower weight and body mass index (BMI) at 7 years postpartum compared to mothers of children with typical development. With the same quantity of alcohol consumed, women with greater body weight may achieve a lower peak blood alcohol

concentration (BAC) than women with lower body weight. Higher peak BAC, often achieved with drinking 3+ or 5+ drinks in a two-hour window, is associated with an increased likelihood and severity of an FASD diagnosis.[15,114] Moreover, in the human fetus, alcohol dehydrogenase (ADH) and catalase, the two major enzymes necessary for clearing alcohol from the body, have much lower enzyme levels than in adults.[115] The slower clearance of alcohol in the fetus causes higher and longer lasting concentration of alcohol in the fetal environment.

Maternal Weight as a Contributing Factor to Fetal Development

The metabolic profile of mothers who are overweight or obese is different than women with normal weight. Individuals with obesity have lower levels of adiponectin and higher levels of insulin, interleukin (IL)-6, and leptin compared to individuals with normal weight. With the lower adiponectin levels among individuals with obesity, there is an increase in placental nutrient transfer and increased fetal growth.[116,117] Higher IL-6 levels among individuals with obesity upregulate placental amino acid transport which may also promote fetal growth.[118] IL-6 also upregulates fatty acid uptake which may be a contributing factor to the excessive fat deposits and increased size associated with infants born to mothers with obesity.[119] Higher umbilical cord leptin levels, in both normally developing and intrauterine growth-restricted fetuses, are associated with increased adiposity,[120] birth weight,[121] birth length,[122] and birth OFC.[122] Taken together, higher maternal weight may contribute to increased fetal size at birth.

Maternal Weight as a Contributing Factor to Child Growth Trajectories

In non-alcohol exposed infants, postnatal growth occurs in a predictable fashion.[123] For infants with typical development, there are generally accepted growth expectations for weight gain (e.g., 30 grams per day for the first 3 months), length gain (e.g., 25 centimeters over the first year), and OFC.[123] Growth trajectories in the first year of life, especially rapid growth, has been associated with an increased risk of childhood obesity.[124,125] Risk factors associated with rapid

growth include short duration of breastfeeding[126] and maternal weight during the child's first year of life.[127] In non-alcohol-exposed pregnancies, higher maternal weight during the first year of life may predispose infants to poorer child outcomes.

In alcohol-exposed pregnancies, whether postnatal growth occurs in a predictable fashion is less clear. In prospective longitudinal cohorts, the long-term postnatal growth trajectory among children with prenatal alcohol exposure has been inconsistent, and this inconsistency may be linked to the quantity/frequency pattern of alcohol consumption.[128–134] In the prospective Avon Longitudinal Study carried out in England, heavy maternal prenatal alcohol consumption was associated with a significant reduction in birth weight and length compared to abstainers; but this reduction was not seen at age 2 or age 10.[135,136] However, a prospective cohort study recruited prenatally by Carter et al. in urban Cape Town, South Africa, reported that nearly 40% of children with heavy prenatal alcohol exposure continued to experience growth restriction into adolescence.[137] Carter et al. also demonstrated that alcohol exposure during pregnancy had a more negative effect (greater reduction) on the child's physical growth among children born to women with lower pre-pregnancy weights.[138] This suggests that maternal weight, in combination with the alcohol consumption pattern, may also be a significant factor in the inconsistent findings in postnatal growth trajectories of alcohol-exposed children.

It follows from the above evidence that excessive maternal weight at delivery and in the first year of life in non-alcohol exposed pregnancies may predispose children to excessive weight gain. However, in alcohol-exposed pregnancies growth restriction may persist into childhood, and higher maternal weight may be protective, resulting in less growth-restricted children. The effect of higher maternal weight and maternal alcohol consumption on child outcome may also present clinically as altered growth trajectories or altered body proportions (weight-for-length).

Maternal Weight as a Contributing Factor to Child Neurocognitive Trajectories

Many longitudinal studies have demonstrated that children born to mothers with severe obesity had a greater susceptibility to newborn regulatory problems,[139] developing cognitive impairments,[140–143] and neuropsychiatric and mood disorders.[144] But, the evidence of an association between maternal pre-pregnancy weight and child neurocognitive impairments is inconsistent.[143] Some studies have suggested there is a U-shaped or J-shaped association between maternal weight and infant cognitive outcomes with either low maternal weight or severe obesity being associated with cognitive impairment.[145,146] Other studies have found no association between maternal pre-pregnancy weight and cognitive outcomes in infants.[147]

In non-alcohol-exposed pregnancies, maternal pre-pregnancy weight or too little or excessive weight gain during pregnancy may predispose the child to poorer cognitive and behavioral outcomes. In alcohol-exposed pregnancies, fetuses are at risk for maladaptive brain formation (e.g., structural changes) and the infant is at a greater risk for neurocognitive delays due to the teratogenic effect of alcohol.[58,148,149] Therefore, alcohol exposure and maternal weight may both be independently associated with poorer behavioral and cognitive outcomes. However, increased maternal weight may result in less alcohol crossing the placenta and may lead to increased placental nutrient transfer and improved fetal development compared to alcohol-exposed pregnancies with lower maternal weight.

Postnatal Environmental Factors and Developmental Trajectories

Numerous animal and human studies have demonstrated the importance of early postnatal environmental factors that influence growth and cognitive development. The benefit of breastfeeding on reducing the risk for childhood obesity and improving child neurocognitive development is well documented.[150–152] However, alcohol can freely pass into breastmilk at concentrations corresponding roughly to that of maternal blood, with the peak in concentration

occurring approximately 30-60 minutes after alcohol consumption.[153] The literature is inconsistent as to whether the quantity of alcohol delivered via breastmilk has a lasting and independent effect from prenatal alcohol exposure on child development.[154,155] Yet one study in South Africa demonstrated that after controlling for alcohol consumption during pregnancy, mothers who reported consuming alcohol in the breastfeeding period were 6.4 times as likely to have a child diagnosed with FASD by age seven.[156] Therefore, alcohol exposure in the breastfeeding period may have adverse effects on child physical and neurocognitive development, independent from the adverse effects of prenatal alcohol exposure. Furthermore, there may be additive and adverse effects on child development from both prenatal exposure to alcohol in utero and postnatal exposure via alcohol in breastmilk.

In addition to the quantity, frequency, and timing of alcohol exposure,[15] other reported factors that contribute to the vulnerability and severity of an FASD diagnosis include: advanced maternal age,[5,6,8,19,20,157–159] smoking during pregnancy,[4,13] week of pregnancy recognition,[68,160] and socioeconomic status and/or educational attainment.[6,8,68,159,161] When considering a child's growth and neurocognitive trajectories, it is necessary to consider how postnatal factors influence development.

Maternal Weight is Not a Proxy for Adequate Nutrition

Increased body mass does not necessarily indicate more optimal nutrient intake. With urbanization and economic development, it is well documented that an individual may be nutrient deficient yet may be overweight or obese simultaneously.[162,163] The importance of maternal dietary intake during pregnancy has risen in prominence as more information about fetal programming and the developmental origins of health and disease have become known.[164,165] Since the diet consists of hundreds of biologically active components, examining the association between diet and disease can often not be done by examining individual nutrient

components separately. Moreover, micronutrient metabolism and utilization are dependent on nutrient balance, and there is an inter-relationship, and co-dependence, between micronutrients for appropriate metabolic function.

Maternal Micronutrient Status, Infant Outcomes, and Fetal Programming

All fetal nutrition comes from the mother. The transfer of nutrients from mother to fetus is dependent on availability of nutrients via the maternal diet, maternal nutrient stores, and the placenta's ability to transport the nutrients. The placenta is the intermediary between the maternal and fetal circulation, and its ability to provide nutrients to the fetus depends on the size, morphology, and vasculature of the placenta. Inadequate maternal diet can adversely affect placental development and function throughout pregnancy.[166]

During pregnancy, a small set of embryonic progenitor cells give rise to fetal organs and tissues following specific differentiation and proliferation pathways. This period of development is highly sensitive to an adverse fetal environment. Maternal deficiencies or abundancies of micronutrients can cause teratogenic effects on the fetus and may predispose the fetus to adverse effects when other teratogenic agents (e.g., alcohol) are present concurrently. Deficiencies of folate can lead to neural tube defects. Iodine deficiencies can cause cretinism. Vitamin A overload can lead to structural malformations in many organ systems. Maternal undernutrition has been associated with altered hormone regulation and epigenetic changes in the fetus.[167,168]

The consequences of suboptimal nutrition during pregnancy can present as intrauterine growth restriction and low birthweight. Birth size is a strong predictor of future growth. Infancy, a period with the maximum growth velocity, is vulnerable to inadequate nutrition. In high-risk environments, such as those where mothers may be undernourished, breastfeeding is beneficial to the infant in terms of immunity/reduced infections, preventing childhood morbidity, and improved cognitive development.[169,170] Even among undernourished mothers, the production

volume and macronutrient composition of breastmilk tends to be adequate for the infant; however, the micronutrient composition can still be inadequate.[170,171] Vitamin A, iodine, and the B-complex are among some of the micronutrients where poor maternal dietary intake and/or status can reduce the concentration in breastmilk.[172,173] This can place the infant at risk for further growth faltering and poor development.

Additionally, the "thrifty phenotype" hypothesis asserts that poor nutrition in utero can alter or program fetal metabolism to adapt to the nutrient environment with the expectation of a similarly nutrient inadequate environment in the future.[174,175] When the nutrient environment remains poor postnatally, the individual is metabolically well adapted; however, if the nutrient environment becomes adequate, the individual is metabolically maladaptive.[174,175] These changes in the fetus can be life-long and have been shown to predispose individuals exposed to nutrient-poor environments in utero to a wide range of diseases in adulthood including cardiovascular disease, diabetes, and cancer.[176,177] These changes occur independent of prenatal alcohol exposure but may be further complicated when undernutrition and alcohol exposure occur concurrently.[178]

The Teratogenic Effect of Alcohol on Micronutrient Intake, Absorption, and Utilization

Alcohol contains 7.1 kcal/g of energy. With regular consumption of alcohol, it can become a primary source of energy by displacing other macronutrients and cause primary malnutrition. The severity of the malnutrition depends on the quantity of alcohol consumed and quality of other foods consumed. Alcohol can dysregulate gastrointestinal function, impair placental function, and inhibit placental transfer of nutrients which all contribute to reduced bioavailability of nutrients for the fetus.[161,179] Alcohol also directly competes with or inhibits a number of metabolic pathways. Alcohol can deplete the maternal store of vitamin A leading to reduced availability and the disruption of normal cellular function in the fetus.[110,180] The

absorption of folic acid is reduced by alcohol which can lead to altered DNA and RNA synthesis.[181] Alcohol can adversely cause zinc sequestration in the maternal liver leading to reduced zinc availability for the fetus and can lead to fetal deficiencies.[111,182] Maternal regulation of calcium is impaired by alcohol which can result in reduced fetal skeletal ossification.[183] One-carbon metabolism is altered in the presence of alcohol and can lead to hyper-homocysteine and genome-wide hypomethylation in the fetus.[184,185] Because of the increased free radicals and reactive oxygen species caused by alcohol metabolism, alcohol increases the demand for antioxidant intake and reduces endogenous antioxidant levels (e.g., glutathione peroxidase).[110,186,187] Micronutrient deficiencies, especially of iron, zinc, and choline, can exacerbate the teratogenic effects of alcohol.[111,112,188] Collectively, alcohol can alter dietary intake and the absorption and utilization of micronutrients which can have negative consequences for fetal development.

Micronutrient Intake Among Women of Childbearing Age in South Africa

In certain segments of the South African population, food insecurity and undernutrition are common.[189] Despite national food fortification policies implemented in 2003, post implementation studies have shown that women and children continue to be micronutrient deficient.[190] Fortification did improve nutrient status for women who were lactating, but they were still deficient on fortified (vitamin A, riboflavin, B_6, and zinc) and non-fortified (calcium, vitamin B_{12}, C, and D) micronutrients.[191] Two studies in the Western Cape Province reported that the majority of women of childbearing age were likely inadequate (less than the US Institute of Medicine's Estimated Average Requirement) on most micronutrients.[24,25] Further, albeit the majority of mothers were likely deficient, mothers of children with FASD were reported to be consuming lower quantities of most micronutrients compared to mothers of

children with typical development.[24] South Africa, and specifically the Western Cape, is an area where undernutrition remains a concern and has a potential to adversely affect fetal and child development.

CHAPTER 3: THE INFLUENCE OF MATERNAL WEIGHT AND ALCOHOL EXPOSURE ON INFANT PHYSICAL CHARCATERISTICS

Overview

Background: Mothers of children with FASD tend to have lower weight compared to mothers of children with typical development, especially in developing countries. This phenomenon has also been demonstrated in Western populations. Yet how alcohol and maternal weight may independently predispose an infant to poorer physical growth trajectories is relatively unexplained in the literature.

*Method*s: Data originated from a longitudinal cohort of 406 South African mothers recruited in prenatal clinics. Their offspring were provided standardized dysmorphology examinations at 6 weeks and 9 months to assess growth and development. Linear mixed modeling was used to determine whether maternal weight and prenatal alcohol exposure exerted a significant influence on the infant's growth and dysmorphology within the first year of life.

Results: Controlling for six covariates, maternal weight accounted for 1-9% of the variance explained in child physical outcomes while alcohol exposure in the previous 30 days of pregnancy accounted for 2-5% of the variance by 9 months. Maternal weight was positively associated with birth length, weight, and head circumference (OFC) centile, but the rate of change over time was similar among all infants regardless of maternal weight. Maternal weight was inversely associated with the number of minor anomalies and total dysmorphology score. Alcohol was significantly associated with lower growth parameters and higher dysmorphology.

Conclusion: Infant physical outcomes were independently associated with maternal weight and prenatal alcohol exposure. Higher maternal body weight may be a protective factor but does not eliminate the adverse effects of alcohol on infant growth and dysmorphology. Regardless of maternal weight, alcohol remains a teratogen and can result in adverse infant development.

Introduction

Fetal alcohol spectrum disorders (FASD) is an umbrella term for the physical and neurocognitive delays/deficits associated with prenatal alcohol exposure. In the general US population, it is estimated that 5% of children fall within the FASD continuum.[1] The Western Cape Province of South Africa has the highest reported FASD prevalence in the world with an estimated 17 - 28% of children having FASD.[2–9,192] The individual variation in child growth, facial dysmorphology, and neurocognitive impairments among children with prenatal alcohol exposure is not fully explained by the quantity, frequency or timing of alcohol consumption or other known risk factors.[15] This results in some children developing within the normal range, while others have severe impairments despite similar exposure to alcohol in utero. Previous studies have demonstrated that children with FASD had significantly more minor anomalies and higher total dysmorphology scores than did children with typical development from birth to 60 months.[99] Children with fetal alcohol syndrome (FAS) were consistently the most dysmorphic and children developing within the typical range were the least dysmorphic. Yet within each FASD diagnostic category, there was individual variation in growth and dysmorphology, especially over time.

Maternal weight has long been suggested as a contributing factor to the risk and severity of an FASD diagnosis. From cross-sectional studies in South Africa, there is evidence that suggests that higher maternal weight may be protective against the adverse effects of prenatal

alcohol exposure.[7,19,193] Because women with higher weight have more body tissue to which alcohol is distributed, their blood alcohol concentration may be lower and the quantity of alcohol that crosses the placenta may be reduced, resulting in less severe growth deficiencies, dysmorphology, and neurobehavioral impairment. In longitudinal studies, mothers of children with FASD had significantly lower maternal weight and BMI when measured at 42 to 60 months postpartum.[99] This is consistent with previous findings from 5 separate cross-sectional studies in the Western Cape Province where maternal weight and BMI (7 years postpartum) has been found to be significantly lower among mothers of children with FASD. This is especially true for children with FAS when compared to mothers of children with typical development.[6–8,13,19] Therefore, among alcohol exposed pregnancies, increased maternal weight may be protective against some of the adverse effects of prenatal alcohol exposure. However, in non-alcohol-exposed pregnancies, increased pre-pregnancy maternal weight, and/or excessive weight gain during pregnancy, may increase the risk of obesity during early childhood and may place the child at risk for delay in neurocognitive and motor development.[194,195]

Because both alcohol exposure and maternal obesity may independently predispose a child to poorer physical developmental trajectories, it is necessary to understand how alcohol exposure and maternal weight influence the developmental trajectories of children. The Western Cape Province has the highest percentage of female adults who are overweight/obese in South Africa,[196] yet an estimated 40% of the Western Cape Province population is at risk for, or is, food insecure and, therefore, being underweight remains a concern.[189,196] There is also little stigma around drinking during pregnancy in the Western Cape, with a high proportion (35-50%) of women of childbearing age binge drinking regularly most Friday and Saturday nights.[13,14] The purpose of this study was to determine whether there was an influence of postpartum maternal

weight and prenatal alcohol exposure on growth and dysmorphology outcomes from birth to 9 months.

Methods

Study Design and Sample

Women seeking prenatal care in five communities in the highly agricultural region of the Western Cape Province of South Africa were invited to consent to, and participate in, a brief alcohol-screening, demographic, and health indicators questionnaire during the pregnancy. The brief questionnaire included the Alcohol Use Disorders Identification Test (AUDIT),[197] the Tolerance, Annoyed, Cut-down, and Eye-opener (TACE) screen,[198] and alcohol consumption in the previous 30 days of the pregnancy. The AUDIT assesses drinking behavior in the previous 12 months in order to identify risky or hazardous consumption patterns and/or alcohol use disorders.[197] The TACE assesses alcohol tolerance and a score of $\geq$2 is considered indication of at risk drinking.[198]

Of the 1,370 women who completed the brief questionnaire, 680 were able to be visited by study staff following the birth with 419 visits occurring within ten days postpartum. Maternal weight was measured and additional information about the pregnancy was obtained within ten days postpartum. Weight was measured with electronic scales with 0.01kg precision. Four hundred and six (406) mothers and their infants were followed until 9 months postpartum. The sample presented here was restricted to include only term (gestational age $\geq$ 37 weeks) and singleton births. Therefore, the final sample was 406 mother/infant dyads who were seen at least once during the follow-up period (see Figure 3.1).

Infant length, weight, and occipitofrontal (head) circumference (OFC) measurements were collected by the attending physician or nurse immediately following the birth and recorded on the infant's medical card. At 6 weeks and 9 months postpartum, study staff completed a

dysmorphology exam for each infant. The dysmorphology exam included measuring a child's length, weight, and OFC and assessing the presence or absence of 12 other minor anomalies.[35] The total dysmorphology score, a weighted summary measure, was determined for each infant following the dysmorphology exam.[35,36] A higher score indicates more dysmorphology. The examiners were blinded to the alcohol exposure history and any previous study assessments.

Statistical Analysis

Postpartum maternal weight was divided into tertiles, and one-way analysis of variance (ANOVA) tests were employed to compare maternal demographic and alcohol consumption patterns and child physical characteristics. Post-hoc analyses were performed using Dunnett's C pairwise comparisons with alpha= 0.05. Dunnett C comparisons control for alpha error (Type 1; false positive) produced when performing multiple comparisons of group means.[199] Categorical variables were examined using chi-square.

To test whether maternal weight mitigates the adverse effect of prenatal alcohol exposure, bivariate and partial correlations were undertaken. Partial correlations controlled for postpartum maternal weight. Stepwise regression analyses were also undertaken. Due to the common variance shared between the alcohol exposure variables, only one measure of drinking, the total number of drinks per drinking day (DDD) was included in the stepwise regression. DDD was selected as the predictor variable because it captures the drinking behavior during pregnancy, while the AUDIT score is based on alcohol behavior in the previous 12 months and lifetime. Data were transformed to correct for skewed distributions to meet the assumptions of linear regression. Logarithmic transformations were applied to DDD, gravidity, and maternal age. Step 1 adjusted for maternal prenatal alcohol consumption. Step 2 adjusted for tobacco use during pregnancy (yes/no), gravidity (log), maternal age (log), and trimester interviewed. Gestational age at birth, infant sex, maternal weight, and the number of days after birth when the

mother's weight was measured were each entered individually as subsequent steps. The final step added the interaction of DDD by maternal weight to test whether maternal weight modifies or attenuates the relationship between DDD and infant outcomes.

Finally, linear mixed models, which account for unbalanced data (varying number of repeated measures across children), unequal spacing of assessment timepoints, and the intra-individual correlation between repeated measures, were estimated. The association of maternal weight with infant length, weight, and OFC centiles were examined from birth to 9 months. The association of maternal weight with other child physical outcomes (number of minor anomalies and total dysmorphology score) was examined from 6 weeks to 9 months. A random effects model, with random intercept and slope for time, was utilized. The fixed effects were time (months), maternal weight, DDD, number of days following birth when maternal weight was measured, gestational age at birth, tobacco use during pregnancy, gravidity, and maternal age. Gestational age, maternal age, and maternal weight were centered on the sample mean. Models were estimated using restricted maximum likelihood. To account for the individual variation around repeated measures of the same phenomenon (e.g., length) at different timepoints, an autoregressive covariance matrix was used. Infants with at least one measurement (at birth, 6 weeks, or 9 months) were included in the analysis. To aid in the interpretation of parameter estimates, time (in months) was centered, thereby allowing the intercept to reflect the outcome at 6 weeks of age for dysmorphology outcomes (e.g., minor anomalies and total dysmorphology score). Since length, weight, and OFC were measured at birth, time was not centered in these models, and the intercept represents birth measurements. To address whether the association between physical outcomes and maternal risk factors change over time, a two-way age (time) interaction with each covariate was explored. Additionally, a 3-way interaction of maternal

weight, DDD, and time was explored as a fixed effect. All analyses were carried out in SPSS 26.[200]

Results

Maternal Characteristics

The maternal demographic and alcohol consumption information by maternal weight tertiles are displayed in Table 3.1. The tertiles approximate groups of 1) underweight-low normal weight, 2) high normal to overweight, and 3) overweight to obese individuals. Maternal age significantly distinguished the groups, with women in tertile 3 being significantly older than women in tertile 1. Approximately 20-25% of women in each group reported drinking in the previous 30 days of their pregnancy, and they were consuming 6.3 to 6.6 DDD on 3.1 to 4.4 days. There was no significant difference in quantity or frequency of alcohol consumption by maternal weight tertile.

Infant Characteristics

Infant physical growth and dysmorphology measurements at birth, 6 weeks, and 9 months by maternal weight tertile are displayed in Table 3.2. Stunting and wasting (length/weight centile <2.5 SD or equivalently < 3rd centile[201]) were present at birth and remained present for a proportion (approximately 20%) of the infants in this cohort. At birth, children born to mothers in tertile 1 were significantly shorter, lighter in weight, and had a smaller OFC than children born to heavier mothers.

By 6 weeks of age, in post-hoc comparisons, growth parameters (length, weight, and OFC) continued to be significantly different between infants of mothers in tertile 1 and tertile 3. Significantly more children born to mothers in tertile 1 were stunted, had true microcephaly (OFC $\leq$ 3rd centile), and had shorter inner canthal distance (ICD) at 6 weeks. The number of

minor anomalies and the total dysmorphology score were significantly higher among infants born to the lightest mothers (tertile 1) compared to heavier mothers.

By 9 months, children born to mothers in tertile 1 were significantly shorter, lighter, had a smaller OFC, ICD, and inner pupillary distance (IPD) and had more minor anomalies and a higher total dysmorphology score than the other two groups. Only two of the growth variables were not significantly different between groups by 9 months (OFC centile $<3^{rd}$ and palpebral fissure length (PFL) centile) and the former measurement approached significance.

Bivariate and Partial Correlations with Infant Physical Outcomes

Alcohol exposure by quantity and frequency was significantly and negatively associated with length and weight centile and positively associated with the number of minor anomalies and total dysmorphology score. Maternal weight was positively and significantly correlated with infant's length, weight, OFC, weight-for-length, ICD, IPD, and PFL while negatively correlated with the number of minor anomalies and total dysmorphology score at 9 months of age (Table 3.3). After controlling for maternal weight in partial correlations, DDD and the number of drinking days per week were negatively associated with infant length and weight centile and positively associated with the number of minor anomalies and total dysmorphology score (Table 3.4). Controlling for maternal weight attenuated the relationship between alcohol exposure and infant outcomes; however, all 4 of the 9 correlations remained significant. This suggests maternal weight may be protective and may partially explain some of the individual variation in infant outcomes, but higher maternal weight does not overcome the negative, teratogenic effects of prenatal alcohol exposure.

Stepwise Regression Predicting Infant Outcomes at 9 Months of Age

Table 3.5 summarizes the stepwise regression with adjusted R^2 and change statistics following each step. The regression coefficients are presented in the Appendix Tables

A.1-A.9. Each step was adjusted for all previous covariates. DDD (step 1) was negatively associated with length and weight centile and positively associated with the number of minor anomalies and total dysmorphology score. DDD explained 3.8% and 4.6% of the total variance for the number of minor anomalies and total dysmorphology score, respectively (adjusted R^2 was 0.038 and 0.046, see Table 3.5). In step 2, tobacco use during pregnancy, higher gravidity, and advanced maternal age was associated with poorer infant outcomes on length, weight-for-length, minor anomalies, and total dysmorphology score. Gestational age (step 3) predicted length, ICD, and PFL centile. Infant sex (step 4) was a significant predictor of ICD centile. Maternal weight (step 5) significantly added to the prediction of all infant's physical outcomes except for PFL centile. Maternal weight significantly added to the models with a change in R^2 ranging from 0.015 – 0.085, adding the most variance explained to infant weight centile. The number of days after delivery when maternal weight was measured added significantly to length centile only. The interaction term of maternal weight by DDD did not significantly add to the variance explained in any model.

Longitudinal Effect: Infant Length

In Table 3.6, maternal weight significantly predicted infant's birth length centile (B=.45, p<.001). Women with higher weight gave birth to longer infants. DDD was negatively associated with birth length centile (B=-5.42, p=.035). Gestational age significantly predicted higher birth length centile (B=2.99, p<.001) while tobacco use was associated with lower birth length centile (B=-4.44, p=.013). The maternal weight by time interaction was not significant indicating there was no difference in the rate of change (slope of growth trajectory) associated with maternal weight across time. Higher gravidity was associated with progressively lower length centile across time (B=-3.30, p=.023).

Longitudinal Effect: Infant Weight Centile and Weight-for-Length

Maternal weight and gestational age were significantly and positively associated with birth weight centile while DDD and trimester interviewed were negatively associated with birth weight centile (Table 3.7). The maternal weight by time interaction was not significant. In Table 3.8, maternal weight significantly predicted infant's weight-for-length at birth (B=0.14, p<.001) and across time (B=0.02, p =.030) (Figure 3.2). DDD significantly predicted birth weight-for-length (B=-3.47, p=.002) indicating that alcohol-exposed infants were thinner or weighed less (relative to their length) at birth than unexposed peers. Even though all infants were term (>37 weeks gestation), longer gestation was associated with higher weight centile and weight-for-length at birth. The trimester in which the mothers were interviewed was negatively associated with birth weight centile and weight-for-length.

Longitudinal Effect: Infant OFC Centile and Facial Measurement Centiles

Birth OFC centile was significantly predicted by maternal weight and trimester interviewed (Table 3.9). DDD in the previous 30 days was not associated with birth OFC centile. A negative association between tobacco use and birth OFC centile approached significance (p=.081). Similar analyses were performed on other facial measurements as the outcome but are not presented in table form. Maternal weight also significantly predicted ICD (B=.25, p<.001), IPD (B=.16, p=.025), and PFL centile (B=.22, p=.014) at 6 weeks. For OFC, ICD, IPD, and PFL, there was no significant difference in the rate of change across time by maternal weight.

Longitudinal Effect: Infant Minor Anomalies and Total Dysmorphology Score

In Tables 3.10 and 3.11, maternal weight was associated with a significant reduction in the number of minor anomalies (B=-0.03, p<.001) and total dysmorphology score (B=-0.05, p<.001) at 6 weeks. DDD was positively and significantly associated with the number of minor anomalies (B=1.95, p<.001) and total dysmorphology score (B=1.88, p=0.024) at 6 weeks. There

was a significant DDD by time interaction for total dysmorphology score (B=0.19, p=.026) (Figure 3.3). A significant DDD by sex interaction was observed for minor anomalies and approached significance for total dysmorphology score (B=-2.14, p=.051). Tobacco use and higher gravidity were associated with more minor anomalies and a higher total dysmorphology score.

Discussion

This study demonstrated that alcohol and maternal weight are important predictors of infant physical outcomes. While there was individual variation in growth parameters, on average, infants in this cohort were born and remained smaller than average, compared to WHO growth standards, on length, weight, and OFC through 9 months of age. Stunting and wasting, markers of an underlying inadequate early life environment,[202] were present at birth and remained present for nearly a quarter of the infants in this cohort at 9 months. Similarly, ten percent of infants had true microcephaly (OFC$\leq$3rd centile) at 6 weeks, and by 9 months of age the proportion of infants with microcephaly nearly doubled. This suggests potential stunting of brain development both in utero and postpartum. The greater percentage with microcephaly at 9 months suggests that some infants experienced growth faltering and were not maintaining similar brain volume development as were infants with typical development.

Our findings are consistent with previous work that indicates that alcohol consumption and maternal weight are associated with infant outcomes. Alcohol exposure was negatively associated with poorer infant physical outcomes. The stepwise regression indicated that even after controlling for DDD and other covariates, maternal weight still significantly added to all models of infant outcomes except PFL centile. Women with higher weight tended to produce larger children with fewer minor anomalies and lower total dysmorphology scores (less dysmorphic). This indicates that maternal weight plays a protective role in this population.

However, the women in this sample may have been more slender (mean weight = 64kg, SD=14.9) relative to Western norms during pregnancy.[203,204] It has been asserted there may be a U-shaped relationship between maternal weight and child outcomes.[145,146] Therefore, while higher maternal weight was beneficial in these communities, where undernutrition is common,[24,25] there may be an upper range where maternal weight is no longer protective and possibly harmful to the infant. Future studies will need to explore this possibility.

There was no statistical evidence for an interaction between maternal weight and DDD in the previous 30 days of pregnancy. Previous studies have indicated that alcohol quantity is associated with infant growth and dysmorphology outcomes.[2–9] Consuming alcohol in a binge fashion, which results in a higher blood alcohol concentration (BAC), may have a greater risk for adverse child outcomes compared to average absolute daily intake quantities.[205] However, others have suggested that estimating BAC is not necessary if alcohol consumption exceeded a binge of 4+ drinks per occasion.[206,207] Because the duration of the drinking episode was not assessed in this study, estimating BAC could not be undertaken, but BAC may partially explain why there was not a significant maternal weight by DDD interaction. Yet alcohol remains teratogenic regardless of maternal weight.

There was evidence of an interaction between maternal weight and time for infant's weight-for-length. This indicates that the rate of change, that is the amount of weight gained relative to length across time, differed from birth to 9 months by maternal weight. Mothers who were lighter had infants who were, on average, born with a lower body weight and gained less weight relative to their length, resulting in a more slender infant compared to peers. Interestingly, DDD by time was not significant, indicating that alcohol exposure may result in children who are small at birth but remain proportionally small through 9 months.

An interaction of DDD by time was observed for the total dysmorphology score. Higher DDD was associated with progressively higher total dysmorphology scores over time. Because the total dysmorphology score is a summary measure where cardinal features of FASD (e.g., smooth philtrum and narrow vermilion) are weighted more heavily than more frequently occurring minor anomalies (e.g., flat nasal bridge), the significant DDD by time interaction term suggests that minor anomalies associated with prenatal alcohol exposure may become more evident across time. While the total dysmorphology score is not intended to be a single indicator for diagnosis, the total dysmorphology score has been shown to correlate with prenatal alcohol exposure and with FASD diagnosis such that children with FAS, the most severe end of the FASD continuum, have higher total dysmorphology scores.[4–8] The total dysmorphology score at 9 months has been a significant predictor of an FASD diagnosis at 5 years of age.[99]

Several known maternal risk factors for FASD were included in this study. Tobacco use during pregnancy and higher gravidity were significantly, negatively associated with infant growth. The strong teratogenic, adverse effects of alcohol are similar in males and females.[74] Yet other effects of postnatal environmental factors were not included in this analysis. While declines of length centiles are typically seen in low- and middle- income countries following the introduction of complementary feeding, the environmental conditions (socioeconomic, cultural, sanitation/housing) were very similar among all women in these communities. Virtually all women in this study were of mixed-race ancestry (‘Cape Coloured’), who averaged 6-8 years of formal education, and they frequently worked in agricultural or agricultural support jobs. The majority (>80%) of women initiate and sustain breastfeeding through, on average, 18 months.[156] Alcohol can freely enter into the breastmilk, so the possibility of continued alcohol exposure via breastmilk adversely affecting child growth and development was not measured in this study.

There is growing evidence that higher maternal weight (obesity) can be a risk factor for adverse child outcomes. The Western Cape Province has the highest rate of maternal overweight/obesity in South Africa, which may predispose children to altered metabolic profiles and long-term consequences. In alcohol-exposed pregnancies, higher maternal weight may result in better physical outcomes;[137] however, it is also important to acknowledge that higher maternal weight may not be consistently beneficial. When health professionals are assessing the development of a child, careful consideration of the prenatal history, including alcohol exposure and maternal weight, may be important indicators to identify children at risk of growth faltering, the possibility of an FASD diagnosis, or other adverse health outcomes.

Strengths and Limitations

This paper has several strengths. The recruitment of women in antenatal clinics and providing standardized dysmorphology exams at fixed timepoints allow for the analysis of growth over time rather than limiting the analysis to a single time point. Previous studies in these communities have demonstrated that women are generally accurate in recalling and reporting alcohol consumption during pregnancy.[13,15,18–20] While <12% of child outcome data were missing at any timepoint, inevitably there are missing data in longitudinal studies. However, the linear mixed model approach employed here is robust in analyzing results with missing data and utilizes any available data for each infant.

There were also limitations to this study. Because this was a prospective cohort recruited in the prenatal clinic, maternal interviews were completed at the time when a woman sought prenatal care. At the time of interview, alcohol consumption was queried for the previous 30 days. However, most women were interviewed in the 2nd and 3rd trimesters. The lack of detailed information about alcohol exposure throughout pregnancy may have attenuated some of the findings. Second, while in severe cases a diagnosis within the FASD continuum can be made in

infancy, given the age of the infants, no formal diagnoses were yet assigned to any infants. Third, maternal weight was measured postpartum at varying times, but all times were within 10 days of birth. Maternal height was not measured; therefore, BMI could not be calculated. Fourth, because interviews occurred during pregnancy, information about infant feeding practices were not obtain and the influence of feeding practices could not be assessed. Fifth, this population is somewhat unique in terms of socioeconomic and nutritional environment (e.g., 22% experiencing poverty[208] and 30% overweight or obese[27]); therefore, findings here may not be readily applicable to other populations, particularly Western societies.

Conclusion

Other studies have suggested that pre-pregnancy maternal weight may be a protective factor in alcohol-exposed pregnancies, such that higher maternal weight may result in less severe effects, or outcomes, in the exposed child.[138] Overall, our findings suggest that infant growth, minor anomalies, and total dysmorphology are influenced both by maternal weight and prenatal alcohol exposure. Women with higher maternal weight produce larger, less dysmorphic infants. Prenatal alcohol exposure results in smaller, more dysmorphic infants. The rate of change in physical outcomes among infants differ across time by maternal weight and prenatal alcohol exposure. Higher maternal weight led to greater infant growth across time while alcohol exposure led to slower growth across time. Future studies will need to determine whether greater infant growth in infancy is associated with positive or adverse health outcomes later in life in this population. In alcohol exposed pregnancies, higher maternal body weight may be a significant protective factor for the fetus, but higher weight does not erase the totality of the adverse, teratogenic effects of alcohol on the fetus and infant. Alcohol exposure during pregnancy can adversely affect fetal development regardless of maternal weight.

Table 3.1 Maternal Characteristics as Reported During Pregnancy by Maternal Weight Tertile

	Tertile 1 (<56.0 kg) (n=135)		Tertile 2 (56.0 – 67.9 kg) (n=136)		Tertile 3 (≥68 kg) (n=135)		
	Mean	(SD)	Mean	(SD)	Mean	(SD)	p
Weight within 10 days of birth (kg)	48.5	(5.1)	61.7	(3.4)	80.6	(10.7)	<.001[A,B,C]
Age	26.2	(5.5)	27.1	(6.1)	28.2	(5.5)	.012[B]
Gravidity	2.7	(1.3)	2.8	(1.4)	2.9	(1.2)	.575
Parity	2.1	(1.3)	2.2	(1.2)	2.2	(1.1)	.654
Used tobacco (% Yes)	57.8		47.8		48.9		.197
AUDIT Total	10.4	(8.9)	9.2	(8.7)	8.9	(8.8)	.290
TACE Total	2.7	(0.6)	2.6	(0.7)	2.6	(0.6)	.176
Trimester interviewed							
First	5.9		10.3		8.1		
Second	23.0		22.8		25.2		
Third	71.0		66.9		66.7		.728
Drank in previous 30 days (% Yes)	25.2		23.5		20.0		.585
DDD – previous 30 days [1]	6.3	(5.7)	6.3	(5.2)	6.6	(5.0)	.973
Number of drinking days – previous 30 days[1]	4.3	(3.3)	4.4	(3.8)	3.1	(3.7)	.297
Number of 3+ binges – previous 30 days[1]	4.0	(3.5)	3.6	(3.9)	2.9	(3.8)	.517
AUDIT Total[1]	17.9	(6.2)	17.4	(7.0)	17.0	(6.2)	.887
TACE Total[1]	4.2	(.6)	3.8	(.9)	3.9	(.9)	.072

1. Among those who reported drinking in the previous 30 days.

AUDIT: Alcohol Use Disorders Identification Test; DDD: Drinks per drinking day

Post-hoc Dunnet C comparisons significant difference between:

A. Tertile 1 & Tertile 2;

B. Tertile 1 & Tertile 3;

C. Tertile 2 & Tertile 3.

Table 3.2 Child Physical Characteristics at Birth, 6 Weeks, and 9 Months by Maternal Weight Tertile

	Tertile 1 (≤53.0 kg)		Tertile 2 (53.1 – 64.0 kg)		Tertile 3 (≥64.1 kg)		
	Mean	(SD)	Mean	(SD)	Mean	(SD)	p
Birth		(n=135)		(n=136)		(n=135)	
Sex (% Male)		51.1		52.2		48.1	.788
Gestational Age	39.2	(1.1)	39.1	(1.1)	39.3	(1.0)	.354
Length Centile	26.8	(30.0)	36.0	(33.0)	41.8	(32.0)	<.001[A,B]
<3rd Centile[1]		27.4		16.2		13.3	.008
Weight Centile	14.9	(18.0)	22.5	(22.9)	28.4	(26.2)	<.001[A,B]
<3rd Centile[1]		36.3		21.3		8.1	<.001
Weight-for-Length	58.1	(7.4)	59.9	(7.8)	62.7	(8.7)	<.001[B,C]
Weight-for-Length Centile	26.8	(31.6)	26.0	(31.7)	31.5	(33.6)	.343
OFC Centile	15.6	(22.0)	22.9	(26.5)	26.9	(26.5)	.001[A,B]
6 weeks		(n=123)		(n=126)		(n=126)	
Age (in days)	44.3	(5.0)	43.5	(4.4)	44.6	(5.4)	.184
Length Centile	19.5	(23.7)	26.4	(26.3)	33.0	(27.9)	<.001[B]
<3rd Centile[1]		30.9		23.0		12.7	.002
Weight Centile	30.1	(24.4)	41.0	(30.8)	48.5	(29.3)	<.001[A,B]
<3rd Centile[1]		14.6		9.4		5.6	.055
Weight-for-Length	80.2	(9.7)	83.3	(11.8)	86.2	(10.4)	<.001[B]
Weight-for-Length Centile	65.2	(34.6)	65.0	(33.9)	65.7	(33.3)	.984
OFC Centile	23.8	(20.8)	32.2	(24.6)	33.6	(22.6)	<.001[A,B]
OFC ≤ 3rd Centile[1]		16.3		10.9		3.2	.003
OFC ≤ 10th Centile		33.3		23.4		16.7	.009
ICD Centile	39.1	(21.8)	43.0	(20.4)	47.0	(19.9)	.012[B]
IPD Centile	31.0	(25.9)	31.3	(26.6)	33.0	(27.4)	.809
PFL Centile	65.6	(31.2)	61.7	(31.1)	68.1	(32.3)	.265
# of minor anomalies	5.2	(3.0)	4.9	(2.7)	4.2	(2.6)	.019[B]
Total Dysmorphology Score	6.9	(4.5)	6.1	(4.4)	5.2	(4.0)	.008[B]
9 months		(n=117)		(n=121)		(n=125)	

Age (in days)	277.4	(8.2)	275.9	(8.0)	277.2	(9.6)	.361
Length Centile	24.5	(28.4)	33.8	(29.6)	36.5	(28.7)	.004[A,B]
<3rd Centile[1]		27.4		17.5		9.6	.002
Weight Centile	17.7	(23.0)	32.4	(30.4)	40.7	(31.5)	<.001[A,B]
<3rd Centile[1]		34.2		15.8		12.0	<.001
Weight-for-Length	113.2	(14.4)	120.8	(16.8)	123.7	(15.2)	<.001[A,B]
Weight-for-Length Centile	43.7	(32.9)	56.7	(32.5)	63.4	(31.4)	<.001[A,B]
OFC Centile	32.3	(30.2)	41.6	(33.0)	45.8	(31.7)	.004[B]
OFC ≤ 3rd Centile[1]		24.8		20.7		13.6	.084
OFC ≤ 10th Centile		35.0		29.8		20.0	.030
ICD Centile	38.8	(29.4)	46.5	(27.9)	46.9	(25.1)	.040
IPD Centile	17.5	(18.6)	28.3	(25.0)	26.7	(22.8)	<.001[A,B]
PFL Centile	49.5	(33.6)	51.2	(30.7)	55.5	(30.0)	.307
# of minor anomalies	7.0	(2.8)	6.0	(2.7)	5.8	(2.6)	.001[A,B]
Total Dysmorphology Score	9.2	(4.8)	7.6	(4.7)	7.2	(4.5)	.002[A,B]

OFC: occipitofrontal (head) circumference; ICD: Inner canthal distance;
IPD: Inner pupillary distance; PFL: Palpebral fissure length

1. Length <3rd centile is clinical definition for stunting; Weight < 3rd centile is clinical definition for wasting; OFC≤3rd centile is clinical definition for microcephaly.

Post-hoc Dunnet C comparisons significant difference between:

A. Tertile 1 & Tertile 2;
B. Tertile 1 & Tertile 3;
C. Tertile 2 & Tertile 3.

Table 3.3 Bivariate Correlations Between Child Characteristics at 9 Months of Age and Maternal Weight

Child Characteristic at 9 months	Drinks per Drinking Day (DDD) – 30 days prior (n=351)	Number of drinking days previous 30 days (n=351)	Maternal Weight (kg) (n=351)
Length Centile	-.136**	-.148**	.152**
Weight Centile	-.106*	-.147**	.304***
OFC Centile	-.009	-.060	.171***
Weight-for-Length	-.072	-.096	.273***
PFL Centile	-.061	-.105	.114*
ICD Centile	-.052	-.092	.110*
IPD Centile	.051	-.010	.126*
# of anomalies	.183***	.228***	-.158**
Total dysmorphology Score	.185***	.237***	-.152**

*p≤.05; **p≤.01; ***p≤.001

OFC: occipitofrontal (head) circumference; ICD: Inner canthal distance;
IPD: Inner pupillary distance; PFL: Palpebral fissure length

Table 3.4 Partial Correlations Between Child Characteristics at 9 Months of Age and Alcohol Consumption by Quantity and Frequency Controlling for Maternal Weight

Child Characteristic at 9 months	Drinks per Drinking Day (DDD) – 30 days prior (n=351)	Number of drinking days previous 30 days (n=351)
Length Centile	-.130*	-.134*
Weight Centile	-.094*	-.119*
OFC Centile	.000	-.042
Weight-for-Length	-.060	-.068
PFL Centile	-.055	-.094
ICD Centile	-.046	-.081
IPD Centile	.059	-.004
# of anomalies	.177***	.214***
Total dysmorphology Score	.179***	.224***

*p≤.05; **p≤.01; ***p≤.001

OFC: occipitofrontal (head) circumference; ICD: Inner canthal distance; IPD: Inner pupillary distance; PFL: Palpebral fissure length

Table 3.5 Stepwise Regression Predicting Infant Outcomes at 9 Months of Age

Infant Outcome	Step	R	R^2	Adjusted R^2	Std. Error of the Estimate	R Square Change	F Change	df1	df2	Sig. F Change
Length Centile	1	.193	.037	.034	28.790	.037	13.827	1	359	.000
	2	.293	.086	.073	28.207	.049	4.744	4	355	.001
	3	.314	.099	.083	28.050	.013	5.001	1	354	.026
	4	.314	.099	.081	28.089	.000	.015	1	353	.902
	5	.350	.122	.103	27.756	.024	9.530	1	352	.002
	6	.387	.149	.128	27.365	.027	11.130	1	351	.001
	7	.387	.150	.125	27.401	.000	.056	1	350	.813
Weight Centile	1	.161	.026	.023	29.765	.026	9.558	1	359	.002
	2	.217	.047	.034	29.608	.021	1.957	4	355	.101
	3	.235	.055	.039	29.523	.008	3.046	1	354	.082
	4	.235	.055	.037	29.562	.000	.065	1	353	.799
	5	.374	.140	.120	28.246	.085	34.646	1	352	.000
	6	.379	.144	.122	28.227	.004	1.484	1	351	.224
	7	.381	.145	.121	28.240	.002	.668	1	350	.414
OFC Centile	1	.051	.003	.000	32.045	.003	.929	1	360	.336
	2	.141	.020	.006	31.946	.017	1.560	4	356	.184
	3	.157	.025	.008	31.913	.005	1.729	1	355	.189
	4	.182	.033	.014	31.817	.009	3.157	1	354	.076
	5	.254	.065	.044	31.337	.032	11.917	1	353	.001
	6	.263	.069	.046	31.304	.005	1.753	1	352	.186
	7	.270	.073	.046	31.291	.003	1.294	1	351	.256
Weight-for-Length	1	.031	.001	-.002	33.2649	.001	.341	1	359	.560
	2	.167	.028	.014	32.9959	.027	2.469	4	355	.044
	3	.172	.029	.013	33.0180	.001	.524	1	354	.469
	4	.172	.029	.010	33.0647	.000	.001	1	353	.972
	5	.288	.083	.062	32.1828	.054	20.612	1	352	.000
	6	.292	.085	.062	32.1901	.002	.839	1	351	.360
	7	.294	.087	.060	32.2147	.001	.465	1	350	.496

Step 1: DDD; Step 2: Tobacco use during pregnancy, gravidity, maternal age; trimester of pregnancy when maternal interview occurred; Step 3: Gestational age at birth; Step 4: Sex of infant; Step 5: Maternal weight; Step 6: Number of days postpartum maternal weight was assessed; Step 7: DDD by Maternal Weight Interaction

Table 3.5 Stepwise Regression Predicting Infant Outcomes at 9 Months of Age (continued)

Infant Outcome	Step	R	R^2	Adjusted R^2	Std. Error of the Estimate	R Square Change	F Change	df1	df2	Sig. F Change
ICD Centile	1	.039	.002	-.001	27.6434	.002	.552	1	359	.458
	2	.148	.022	.008	27.5146	.020	1.842	4	355	.120
	3	.226	.051	.035	27.1417	.029	10.822	1	354	.001
	4	.255	.065	.047	26.9738	.014	5.421	1	353	.020
	5	.283	.080	.059	26.7991	.015	5.618	1	352	.018
	6	.289	.084	.060	26.7821	.004	1.447	1	351	.230
	7	.289	.084	.057	26.8203	.000	.001	1	350	.978
IPD Centile	1	.045	.002	-.001	22.8081	.002	.736	1	358	.391
	2	.127	.016	.002	22.7736	.014	1.271	4	354	.281
	3	.129	.017	.000	22.8020	.000	.120	1	353	.730
	4	.147	.022	.002	22.7746	.005	1.847	1	352	.175
	5	.198	.039	.017	22.6006	.018	6.441	1	351	.012
	6	.203	.041	.016	22.6120	.002	.648	1	350	.422
	7	.203	.041	.014	22.6435	.000	.028	1	349	.868
PFL Centile	1	.098	.010	.007	31.1768	.010	3.460	1	359	.064
	2	.118	.014	.000	31.2810	.004	.403	4	355	.806
	3	.205	.042	.026	30.8802	.028	10.274	1	354	.001
	4	.220	.048	.029	30.8193	.006	2.402	1	353	.122
	5	.239	.057	.036	30.7201	.009	3.284	1	352	.071
	6	.240	.058	.034	30.7536	.001	.232	1	351	.630
	7	.241	.058	.031	30.7885	.001	.206	1	350	.650
# of minor anomalies	1	.201	.040	.038	2.718	.040	14.892	1	355	.000
	2	.282	.079	.066	2.677	.039	3.721	4	351	.006
	3	.296	.088	.072	2.669	.008	3.206	1	350	.074
	4	.297	.088	.070	2.672	.000	.145	1	349	.704
	5	.334	.112	.091	2.641	.024	9.290	1	348	.002
	6	.335	.113	.090	2.643	.001	.302	1	347	.583
	7	.336	.113	.087	2.647	.000	.179	1	346	.673
Total Dysmor-phology Score	1	.221	.049	.046	4.634	.049	18.451	1	359	.000
	2	.319	.102	.089	4.530	.053	5.206	4	355	.000
	3	.319	.102	.086	4.536	.000	.044	1	354	.834
	4	.319	.102	.084	4.542	.000	.002	1	353	.962
	5	.351	.123	.103	4.494	.022	8.664	1	352	.003
	6	.351	.123	.101	4.500	.000	.034	1	351	.853
	7	.352	.124	.099	4.504	.001	.320	1	350	.572

Step 1: DDD; Step 2: Tobacco use during pregnancy, gravidity, maternal age; trimester of pregnancy when maternal interview occurred; Step 3: Gestational age at birth; Step 4: Sex of infant; Step 5: Maternal weight; Step 6: Number of days postpartum maternal weight was assessed; Step 7: DDD by Maternal Weight Interaction

OFC: occipitofrontal (head) circumference; ICD: Inner canthal distance;
IPD: Inner pupillary distance; PFL: Palpebral fissure length

Table 3.6 Linear Mixed Model Predicting Length Centile[1]

				95% CI	
	Estimate	SE	p	Lower	Upper
Fixed Effects					
Intercept	29.00	6.19	<.001	16.86	41.15
DDD – previous 30 days (log)	-5.42	2.56	0.035	-10.45	-0.38
Maternal Weight (kg)	0.45	0.08	<.001	0.31	0.60
Trimester Interviewed	-0.39	1.39	0.779	-3.13	2.35
Gestational Age at Birth	2.99	0.81	<.001	1.40	4.57
Sex (Males)	-1.44	1.71	0.400	-4.80	1.92
Sex (Female)	--	--			
Tobacco (Yes)	-4.44	1.77	0.013	-7.88	-0.94
Tobacco (No)	--	--			
Gravidity (log)	2.07	9.33	0.824	-16.23	20.37
Maternal Age (log)	-24.32	12.95	0.061	-49.73	1.09
Day Weight Measured	1.20	0.35	0.001	0.50	1.89
Time	1.79	0.84	0.033	0.15	3.44
Gravidity (log) * time	-3.30	1.46	0.023	-6.15	-0.45
Maternal weight * time	-0.01	0.01	0.501	-0.04	0.02

[1]Due to the Hessian matrix not converging with an autoregressive covariance matrix, an identify covariance matrix was used.

Covariates without a time interaction term indicate the effect of the covariate on infant physical outcome at birth. The variable, time, indicates whether there is a change across time. Covariate by time interactions (covariate*time) indicate the rate of change (slope) of the trajectory attributable to the covariate.

DDD: Drinks per drinking day

Table 3.7 Linear Mixed Model Predicting Weight Centile

				95% CI	
	Estimate	SE	p	Lower	Upper
Fixed Effects					
Intercept	41.31	6.44	<.001	28.66	53.96
DDD – previous 30 days (log)	-7.18	2.97	0.016	-13.02	1.33
Maternal Weight (kg)	0.47	0.08	<.001	0.31	0.62
Trimester Interviewed	-3.67	1.61	0.023	-6.84	-0.51
Gestational Age at Birth	1.84	0.93	0.048	0.01	3.69
Sex (Males)	-2.26	1.98	0.253	-6.15	1.62
Sex (Female)	--	--			
Tobacco (Yes)	-2.30	2.05	0.262	-6.34	1.72
Tobacco (No)	--	--			
Gravidity (log)	-7.50	9.28	0.419	-25.73	10.73
Maternal Age (log)	-14.75	14.97	0.325	-44.16	14.66
Day Weight Measured	0.38	0.41	0.351	-0.42	1.18
Time	-0.03	0.19	0.879	-0.39	0.34
Maternal weight * time	0.02	0.01	0.189	-0.01	0.04

Covariates indicate the effect of the covariate on infant physical outcome at birth. The variable, time, indicates whether there is a change across time. Covariate by time interaction (covariate*time) indicates the rate of change across time (slope) of the trajectory attributable to the covariate.
DDD: Drinks per drinking day

Table 3.8 Linear Mixed Model Predicting Weight-for-Length

				95% CI	
	Estimate	SE	p	Lower	Upper
Fixed Effects					
Intercept	71.02	2.39	<.001	66.33	75.72
DDD – previous 30 days (log)	-3.47	1.09	0.002	-5.61	-1.32
Maternal Weight (kg)	0.14	0.03	<.001	0.08	0.20
Trimester Interviewed	-1.96	0.59	0.001	-3.12	-0.78
Gestational Age at Birth	1.99	0.34	<.001	1.31	2.67
Sex (Males)	1.27	0.85	0.136	-0.40	2.95
Sex (Female)	--	--			
Tobacco (Yes)	0.12	0.75	0.870	-1.36	1.61
Tobacco (No)	--	--			
Gravidity (log)	-2.34	3.41	0.493	-9.05	4.36
Maternal Age (log)	-4.22	5.51	0.445	-15.05	6.62
Day Weight Measured	-0.02	0.15	0.898	-0.32	0.28
Time	5.64	0.15	<.001	5.34	5.93
Maternal Weight * time	0.02	0.01	0.030	0.00	0.03
Sex (Male) * time	0.58	0.21	0.006	0.17	0.99

Covariates indicate the effect of the covariate on infant physical outcome at birth. The variable, time, indicates whether there is a change across time. Covariate by time interaction (covariate*time) indicates the rate of change across time (slope) of the trajectory attributable to the covariate.
DDD: Drinks per drinking day

Table 3.9 Linear Mixed Model Predicting OFC Centile

				95% CI	
	Estimate	SE	p	Lower	Upper
Fixed Effects					
Intercept	34.86	6.36	<.001	22.39	47.36
DDD – previous 30 days (log)	-2.05	2.94	0.486	-7.82	3.72
Maternal Weight (kg)	0.34	0.07	<.001	0.19	0.48
Trimester Interviewed	-3.97	1.59	0.013	-7.10	-0.84
Gestational Age at Birth	0.95	0.92	0.305	-0.86	2.76
Sex (Males)	-3.03	1.95	0.121	-6.87	0.80
Sex (Female)	--	--			
Tobacco (Yes)	-3.53	2.02	0.081	-7.50	0.44
Tobacco (No)	--	--			
Gravidity (log)	-7.74	9.17	0.399	-25.77	10.30
Maternal Age (log)	-9.82	14.79	0.507	-38.89	19.26
Day Weight Measured	0.76	0.40	0.058	-0.03	1.55
Time	1.66	0.19	<.001	1.28	2.03
Maternal weight * time	0.00	0.01	0.711	-0.02	0.03

Covariates indicate the effect of the covariate on infant physical outcome at birth. The variable, time, indicates whether there is a change across time. Covariate by time interaction (covariate*time) indicates the rate of change across time (slope) of the trajectory attributable to the covariate.
DDD: Drinks per drinking day

Table 3.10 Linear Mixed Model Predicting Number of Minor Anomalies

				95% CI	
	Estimate	SE	p	Lower	Upper
Fixed Effects					
Intercept	2.05	0.77	0.008	0.54	3.56
DDD – previous 30 days (log)	1.95	0.48	<.001	1.00	2.90
Maternal Weight (kg)	-0.03	0.01	0.001	-0.05	-0.01
Trimester Interviewed	-0.15	0.19	0.451	-0.52	0.23
Gestational Age at Birth	0.15	0.11	0.184	-0.07	0.37
Sex (Males)	0.49	0.27	0.067	-0.03	1.01
Sex (Female)	--	--			
Tobacco (Yes)	0.49	0.24	0.048	0.01	0.97
Tobacco (No)	--	--			
Gravidity (log)	3.68	1.10	0.001	1.50	5.85
Maternal Age (log)	-1.52	1.78	0.396	-5.02	1.99
Day Weight Measured	0.05	0.05	0.259	-0.04	0.15
Sex (Male) * DDD	-1.88	0.67	0.005	-3.21	-0.56
Time	0.20	0.027	<.001	0.15	0.26
Maternal weight * time	0.00	0.00	0.867	0.00	0.00

Covariates indicate the effect of the covariate on infant physical outcome at 6 weeks. The variable, time, indicates whether there is a change across time. The sex*DDD interaction indicates the sex difference at 6 weeks. Covariate by time interaction (covariate*time) indicates the rate of change across time from (slope) of the trajectory attributable to the covariate.
DDD: Drinks per drinking day

Table 3.11 Linear Mixed Model Predicting Total Dysmorphology Score

				95% CI	
	Estimate	SE	p	Lower	Upper
Fixed Effects					
Intercept	1.64	1.25	0.191	-0.82	4.10
DDD – previous 30 days (log)	1.88	0.83	0.024	0.25	3.51
Maternal Weight (kg)	-0.05	0.02	0.001	-0.08	-0.02
Trimester Interviewed	-0.08	0.32	0.801	-0.70	0.54
Gestational Age at Birth	0.04	0.18	0.808	-0.31	0.40
Sex (Males)	0.84	0.43	0.054	-0.013	1.69
Sex (Female)	--	--			
Tobacco (Yes)	1.20	0.40	0.003	0.41	1.98
Tobacco (No)	--	--			
Gravidity (log)	5.60	1.80	0.002	2.05	9.12
Maternal Age (log)	-1.53	2.90	0.597	-7.22	4.16
Day Weight Measured	0.06	0.08	0.451	-0.10	0.21
Sex (Male) * DDD	-2.14	1.09	0.051	4.29	0.01
Time	0.21	0.03	<.001	0.14	0.28
DDD * time	0.19	0.09	0.026	0.02	0.36
Maternal weight * time	0.00	0.00	0.951	0.00	0.00

Covariates indicate the effect of the covariate on infant physical outcome at birth. The variable, time, indicates whether there is a change across time. Covariate by time interaction (covariate*time) indicates the rate of change across time (slope) of the trajectory attributable to the covariate.
DDD: Drinks per drinking day

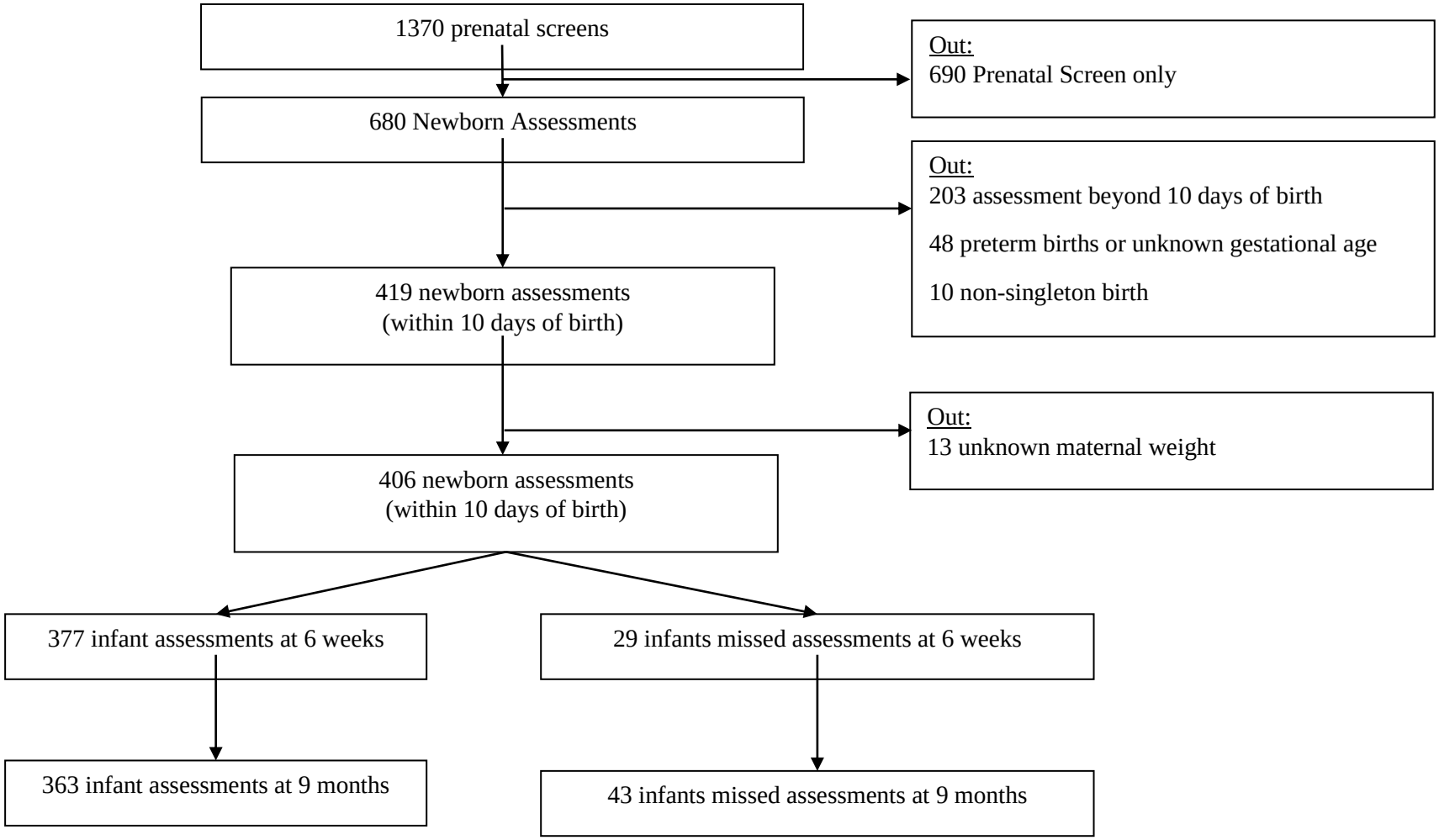

Figure 3.1 Consort Chart

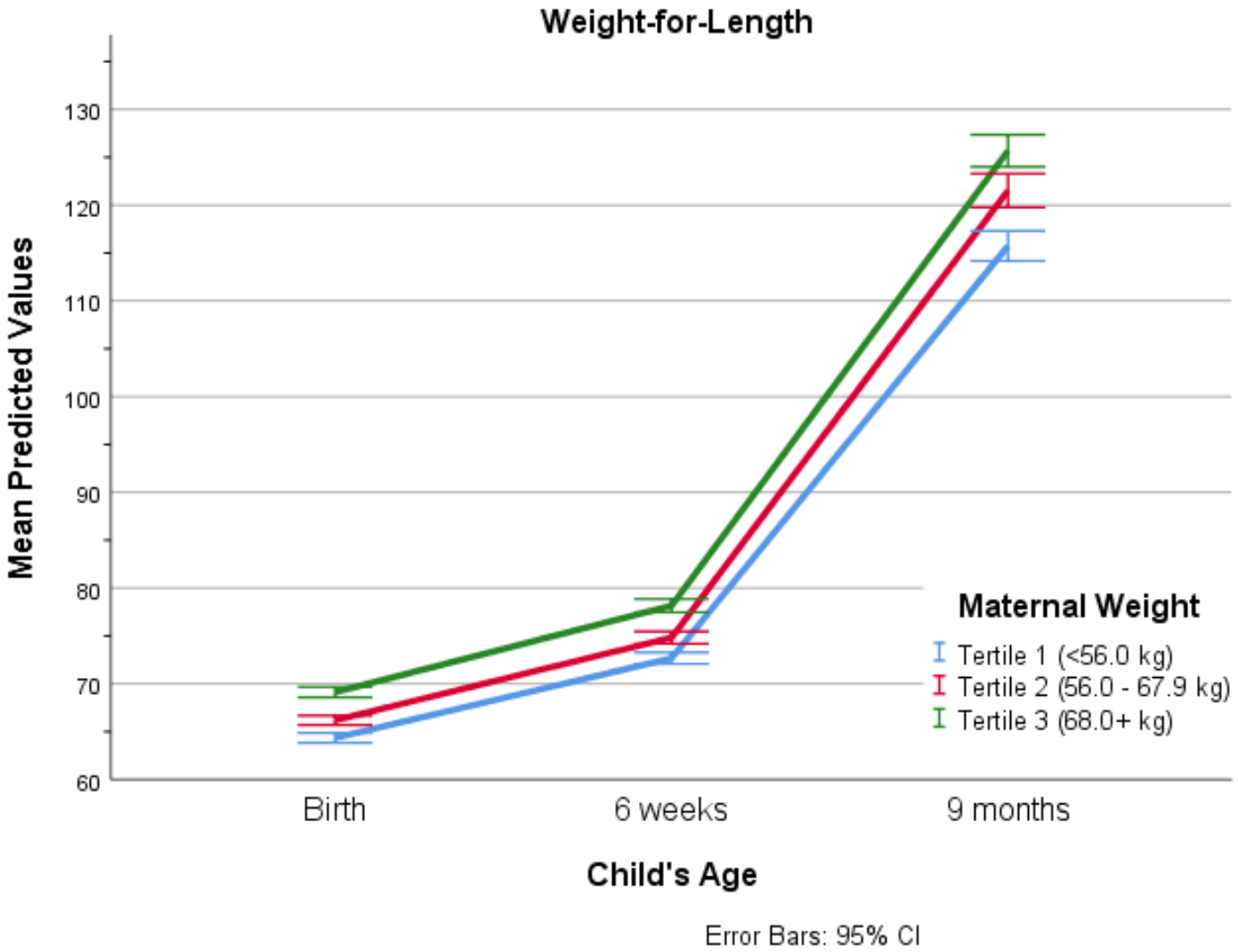

Figure 3.2 Maternal Weight Significantly Predicts Birth Weight-for-Length (B=0.14, p<.001) and there was Significant Difference in the Rate of Change (Slope) Across Time by Maternal Weight (B=0.02, P=0.030).

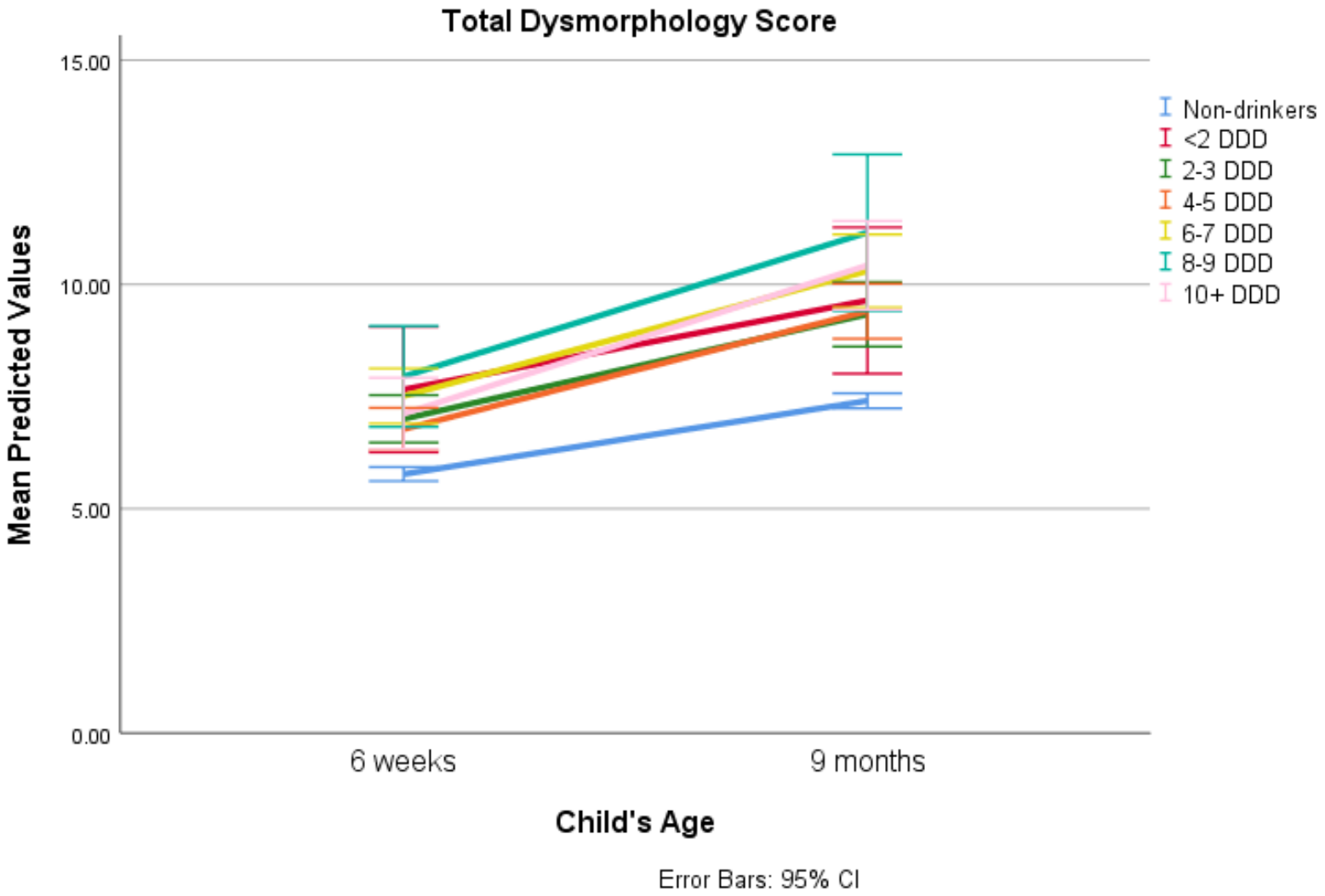

Figure 3.3 Drinks per Drinking Day (DDD) Differentiate Total Dysmorphology Score at 6 Weeks and there was Significant Difference in the Rate of Change (Slope) Across Time by DDD (B=0.19, P=0.26).

CHAPTER 4: THE INFLUENCE OF MATERNAL WEIGHT AND ALCOHOL EXPOSURE ON INFANT NEURODEVELOPMENTAL CHARACTERISTICS

Overview

Background: It has been asserted that among alcohol-exposed pregnancies, maternal weight may reduce the severity of infant neurocognitive impairments due to prenatal alcohol exposure. Few studies have investigated the possible effect of maternal weight and alcohol consumption within the first year of life.

*Method*s: Women seeking prenatal care were recruited into a longitudinal study to assess the neurocognitive development of their children. Maternal weight was measured within 10 days of birth. The Bayley Scales of Infant and Toddler Development, 3rd Edition were used to examine neurocognitive abilities among South African infants (*n*=406) at 6 weeks and 9 months postpartum. Linear mixed modeling was employed to determine whether maternal weight and prenatal alcohol exposure affect an infant's neurocognitive abilities within the first year of life.

Results: On average, infants were performing within the normal range on the cognitive, language, motor, and social/emotional domains. Yet 7% of children fell more than 2 standard deviations (SD) below the mean ('extremely low') and 25.6% of children fell 1.5 SD below the mean ('borderline') in at least one domain at 9 months. Higher maternal weight significantly predicted better cognitive and motor performance at 6 weeks; however, the rate of developmental growth was similar among all infants, regardless of maternal weight. Alcohol consumption in the previous 30 days of pregnancy did not predict any of the domains by 9 months of age.

Conclusion: Maternal weight is a significant predictor of some aspects of infant neurodevelopmental abilities within the first year of life. Additional studies are needed to determine whether this beneficial effect of higher maternal weight is sustained.

Introduction

In addition to poor global developmental outcomes,[89,209] specific deficits in motor,[103,210] learning,[211,212] attention,[213] language,[214,215] and executive function[216] have been documented among children with prenatal alcohol exposure.[217,218] Using the Revised Institute of Medicine Diagnostic Guidelines for fetal alcohol spectrum disorders (FASD), a score of 1.5 standard deviations (SD) below the mean on a standardized assessment is evidence for a developmental delay. All children diagnosed with the specific FASD diagnoses of fetal alcohol syndrome (FAS), partial FAS (PFAS), and alcohol-related neurodevelopmental disorder (ARND) must demonstrate at least one developmental delay in global cognitive ability or behavioral domain.[35,36]

The Bayley Scales of Infant and Toddler Development[40] is a widely used, standardized tool to assess an infant's development on cognitive, language, motor, and social/emotional abilities. There is no universally accepted definition of developmental delay for the Bayley; however, criteria based on standard deviations below the mean is a commonly used approach.[40] Previous studies have demonstrated that the Bayley is a suitable tool for assessing development in South Africa.[41,99,219,220]

Previous studies in South Africa using the Bayley, have demonstrated that alcohol-exposed infants performed worse on gross and fine motor function than unexposed infants, but showed no significant difference in language or cognitive performance at 6 months.[221] Alcohol-exposed, South African infants also performed worse than unexposed infants on the social/emotional domain at 6 months, albeit both groups were performing, on average, within the

normal range.[222] Another study found that children who were later diagnosed with FASD performed, on average, within the normal range at 9 months but fell to low average by the 42-month Bayley assessment. Furthermore, by 60 months 75% fell to $\leq 10^{th}$ centile on the Kaufmann Assessment Battery for Children,[223] an assessment of cognitive development.[99] In a population-based cohort in the Drakenstein municipality of the Western Cape Province, where only 15% of women reported alcohol consumption, 55.6% of infants had a delay (< 1 SD below the mean) in at least one domain on the Bayley and nearly 11% had delays on four domains by 24 months.[224] Taken together, the Bayley has been shown to differentiate child performance due to prenatal alcohol exposure or other circumstances; but many South African children may be at risk for developmental delays, whether exposed to alcohol prenatally or not.

Within FASD diagnostic categories there is individual variation. The severity of prenatal alcohol exposure on neurodevelopment cannot be fully explained by the quantity, frequency, and timing of alcohol exposure alone.[15] Other distal and environmental factors, such as maternal size, age, gravidity, socioeconomic status, and genetic factors have been identified in case control studies as contributing distal risk factors.[225] Cross-sectional studies in South Africa suggest that higher maternal weight may be protective against the adverse effects of prenatal alcohol exposure. Alcohol-exposed children born to heavier mothers performed better than alcohol-exposed children born to lighter mothers.[7,19,193] Yet in pregnancies where there is no in utero exposure to alcohol, many longitudinal studies have demonstrated that children born to mothers with severe obesity had a greater susceptibility to newborn regulatory problems,[139] cognitive impairments,[140–143] and neuropsychiatric/mood disorders.[144] The purpose of this study was to determine whether maternal weight and prenatal alcohol exposure influenced infant

neurocognitive abilities from 6 weeks to 9 months. As a secondary aim the influence of additional maternal risk factors on child neurodevelopmental abilities were explored.

Methods

Study Design

In five communities in the Western Cape Province of South Africa, women seeking prenatal care were invited to participate in a brief alcohol-screening questionnaire. This questionnaire consisted of the Alcohol Use Disorders Identification Test (AUDIT),[197] the Tolerance, Annoyed, Cut-down, Eye-opener (TACE) screen,[198] and questions ascertaining alcohol consumption for the previous 30 days of the pregnancy. The brief questionnaire was completed by 1370 women and 680 were visited by study staff following the birth with 419 visits occurring within ten days of delivery for term (gestational age≥37 weeks) and singleton births. Maternal weight was measured and additional information about the pregnancy and birth was obtained within ten days of delivery. Maternal weight was measured using an electric scale with 0.01kg precision. Mothers and their infants (*n*=406) were followed until 9 months postpartum (see Figure 4.1). At both 6 weeks and 9 months postpartum, the Bayley Scales of Infant and Toddler Development, 3rd Edition, assessment was carried out with each infant by study personnel. The examiners were blinded to the alcohol exposure history and any previous assessments. The sample presented in this paper was restricted to term (gestational age≥37 weeks) and singleton births, and the final sample included 406 mother/infant dyads.

Alcohol information was obtained via an in-person interview soon after the woman first sought prenatal care. Alcohol consumption was assessed by querying the participant on her usual number of drinks per drinking day (DDD) in the previous 30 days and on how many days in the previous 30 days she drank alcohol. The women also completed the AUDIT which assesses drinking behavior in the previous 12 months in order to identify risky or hazardous consumption

patterns and/or an alcohol use disorder.[197] A score of 8 or more is considered an indication of a hazardous drinking pattern. The TACE also identifies risky alcohol consumption with a score of 2 or more considered indication of risky drinking.[198]

Statistical Analysis

The postpartum maternal weight was collapsed into tertiles. The tertiles approximate groups of underweight-low normal weight, high normal to overweight, and overweight to obese. One-way analysis of variance (ANOVA) tests were used to compare maternal alcohol consumption patterns and child neurodevelopmental outcomes. Post-hoc analyses were performed using Dunnett's C pairwise comparisons with alpha= 0.05. Categorical variables were examined using chi-square. Consistent with the Revised Institute of Medicine diagnostic guidelines for FASD,[35] developmental delay was defined as a composite score $\leq$ 1.5 SD below the reference population mean.

Next, stepwise regression analyses were performed. Data transformations were undertaken to correct for skewed distributions. Logarithmic transformations were applied to DDD, gravidity, and maternal age. Step 1 adjusted for maternal prenatal alcohol consumption (DDD). Step 2 adjusted for tobacco use during pregnancy (yes/no), gravidity (log), maternal age (log), and the trimester when the interview occurred. In subsequent steps each variable was entered individually: gestational age at birth, sex of infant, maternal weight, and number of days post-delivery when maternal weight was measured. In the final step, an interaction term of DDD by maternal weight was included to determine if there was modification by maternal weight on infant neurocognitive outcomes.

To examine the effect of maternal weight and DDD on infant neurobehavioral abilities from six weeks to 9 months of age, linear mixed models with repeated measures were used. A random intercept and a random slope for time were included in the models. Time (months),

maternal weight, DDD, number of days post-delivery when maternal weight was measured, gestational age at birth, infant sex, tobacco use during pregnancy, gravidity, and maternal age were included as fixed effects. Gestational age, maternal age, and maternal weight were centered on the sample mean. Models were estimated using restricted maximum likelihood. An autoregressive covariance matrix was used to account for the individual variation around repeated measures of the same phenomenon at different timepoints. To determine whether the association between infant outcomes and maternal risk factors change over time, a two-way interaction with time and each covariate was explored. Infant sex interactions were also explored. To aid in the interpretation of parameter estimates, time (in months) was centered to have the intercept reflect the outcome at 6 weeks of age. All analyses were carried out using SPSS 26.[200]

Results

Maternal demographic and alcohol consumption information is shown in Table 4.1. Women in tertile 3 (the heaviest mothers) were significantly older than women in tertile 1. There was no significant difference in gravidity, parity, or tobacco use. Between 20-25% of women reported consuming alcohol in the previous 30 days. Women who drank consumed, on average, 6.3-6.6 DDD; however, there was no significant difference in DDD by maternal weight tertile. Among drinkers, although there was no difference between maternal groups, all three maternal groups had very high average AUDIT scores with means greater than 17 (range:2-31) meaning that high risk drinking was occurring regardless of maternal weight. Among drinkers, the TACE score approached significance (p=.072) in differentiating the maternal groups with women in tertile 1 having on average a higher (riskier) score.

Infant Neurodevelopmental Outcomes

Infant neurodevelopmental measures at 6 weeks and 9 months by maternal weight tertile are displayed in Table 4.2. At 6 weeks infants in this cohort performed, on average, within the "average" range (composite score 90-109) on all four domains. At 6 weeks, the cognitive, language, motor, and social/emotional domains were not different between the maternal tertile groups. Only the cognitive score approached significance (p=.094). By 9 months, the cohort performed in the "high average" range (composite score 110-119) on the cognitive domain and within the average range for language, motor, and social/emotional domains. Infants whose mothers were in tertile 1 performed significantly worse on the motor composite score and social/emotional composite score at 9 months compared to infants whose mothers were heavier (tertile 2 and 3). On average, infants in all maternal weight tertile groups were performing within the normal range, however, infants in tertile 1 were performing lower than infants in tertile 2 and 3 by 9 months of age.

Individual Performance: Percent Below the Mean

The percent of infants who scored $\leq$2 standard deviations (composite score <70, "extremely low") and $\leq$1.5 standard deviations (composite score $\leq$78, "borderline") below the mean are presented in Table 4.3. At 6 weeks, 1.6% of all infants were classified as borderline on cognitive, 9.3% on language, 0.5% on motor, and 1.6% on social/emotional domains. There was no significant difference by maternal weight tertile. By 9 months among all infants, 2.5% were borderline on cognitive, 9.9% on language, 11.6% on motor, and 7.5% on social/emotional domains. Significantly more infants in tertile 1 performed in the borderline and extremely low range on motor development compared to infants in tertile 2 and 3. At 9 months, a quarter (25.6%) of all infants fell within the borderline range and 6.7% were in the extremely low range.

Stepwise Regression: Outcomes at 9 Months

Summary of the stepwise regressions predicting infant outcomes at 9 months is shown in Table 4.4. Self-reported DDD in the previous 30 days of pregnancy (step 1) did not significantly predict any domain (cognitive, language, motor, or social/emotional). Step 2 (tobacco use, gravidity, maternal age, and trimester interviewed) accounted for 2.7% (change in R^2=0.27) of the variance in infant cognitive performance and 2.9% (change in R^2=0.29) of the variance in language percentile. See Appendix Tables B.1-B.4 for specific variable regression coefficients. Gestational age (step 3) accounted for 1.1% of the variance on the social/emotional domain while infant sex (step 4) accounted for 1.1% of the variance on the language domain. Maternal weight (step 5) significantly added to the model for the cognitive percentile only and accounted for 1% (change in R^2=0.11) of the variance. After controlling for alcohol consumption and all previous covariates, each additional kilogram of maternal weight was associated with a .186 increase in infant cognitive percentile (Appendix B.1, model 5, p=.047). There was no significant interaction between DDD and maternal weight (step 6) in any stepwise regression model.

Longitudinal Effects: Cognitive Performance

In Tables 4.5 through 4.8 the fixed effects of the linear mixed models are shown for each neurodevelopmental domain. In Table 4.5, maternal weight, infant sex, gravidity, and postpartum day when maternal weight was measured were significant predictors of infant cognitive abilities at 6 weeks. Maternal weight was significantly and positively associated with cognitive abilities at 6 weeks (B=0.16, p=.018) (Figure 4.2). There was a significant gravidity by sex interaction (B=43.23, p =.001). At lower gravidity, female infants perform significantly better than males; however, as gravidity increases the benefit of being female was no longer present. There was a significant difference in the rate of change across time by tobacco use during pregnancy. The number of days postpartum when maternal weight was measured was negatively associated with

infant cognitive performance at 6 weeks (B=-1.55, p=.004) and the rate of change was significantly different by the postpartum day when weight was measured (B=0.25, p=.008).

Longitudinal Effects: Language Performance

As seen in Table 4.6, neither DDD nor maternal weight predicted language abilities at 6 weeks or across time. Being interviewed later in pregnancy (a proxy for when prenatal care was first sought) was associated with lower language percentile rank at 6 weeks but the rate of change did not differ across time. Males performed worse at 6 weeks, but the rate of change across time was similar between males and females.

Longitudinal Effects: Motor Performance

Motor performance was predicted by maternal weight, gravidity, and time (Table 4.7). Higher maternal weight was associated with better infant motor performance at 6 weeks (B=0.27, p=.005). There was a significant interaction between maternal weight and infant sex (B=-0.26, p=.036). At lower maternal weight there was no significant difference between males and females, while at higher maternal weights female infants performed significantly better. Higher gravidity was negatively associated with motor performance at 6 weeks. The motor percentile rank significantly decreased over time (B=-3.89, p<.001) indicating the overall cohort performance followed a downward trajectory.

Longitudinal Effects: Social/Emotional Performance

Maternal weight did not significantly predict infant social/emotional performance. DDD was negatively associated with percentile rank at 6 weeks (Table 4.8). There was a significant interaction between DDD and tobacco use (B=15.00, p=.031) and higher performance at 6 weeks was predicted by higher maternal age. Time was a significant predictor of decreased performance; as infants aged, their performance followed a downward trajectory (B=-1.11, p<.001).

Discussion

On average, infants were performing within the normal range and continued to meet the developmental milestones as expected through 9 months. However, the downward trajectory observed for the motor and social/emotional domains may indicate the possibility of additional children falling within the at-risk performance categories. Furthermore, as the developmental milestones become more complex and require more integration of cognitive processes, the rate of change may become differentiated. Others have noted that among children in South Africa, Bayley domain scores decrease with advancing age;[41,99] therefore, the percent of children who may be at risk for experiencing a developmental delay may be underestimated by assessing children under one year of age. A 5-year longitudinal study completed in these communities found that the children with FASD and the children with typical development followed a downward developmental trajectory.[99] In early life assessments (<1 year of age), infants were performing within the normal range, but this was not sustained through 5 years; both children with FASD and children with typical development dropped, on average, below the 10th centile. As children age, more receptive and expressive language and visual-spatial motor integration is required and more advanced assessments may better differentiate children than was observed in this study.

In this sample, infants born to heavier mothers performed better than infants of mothers with lower weight on the cognitive and motor domains. Previously, the association between maternal pre-pregnancy weight and child neurodevelopmental impairments has been demonstrated to be inconsistent.[143] Therefore, it was not entirely unexpected that no associations between maternal weight and language or social/emotional domains were found in this sample at this young age. Previous studies have suggested maternal weight may have a U-shaped effect on infant developmental outcomes.[145,146] The women in this sample may have been more slender

(mean weight = 64kg, SD=14.9) relative to Western norms during pregnancy.[203,204] This sample may not have had a sufficiently wide distribution to fully capture the protective and/or at-risk ranges of maternal weight. Therefore, while higher maternal weight was beneficial in these communities, where undernutrition is common,[24,25] there may be an upper range where maternal weight is no longer protective and possibly harmful to the infant.

There was no significant interaction between maternal weight and time for any neurodevelopmental outcome assessed. This indicates that there was no difference in the developmental growth among infants in the first 9 months of life regardless of maternal weight. Any benefit derived from higher maternal weight was observable at 6 weeks, but the developmental growth (slope of the trajectory) among infants was similar across maternal weight.

The lack of an association between DDD and some infant outcomes in this sample may be, in part, because only alcohol consumption in the previous 30 days was assessed. Other researchers have found an association with alcohol consumption and poorer performance on the Bayley within the first year of life, but the alcohol consumption information covered the entire pregnancy.[221] Previous studies have shown that the quantity and timing of exposure predicts the development of cardinal FASD facial features and the severity of diagnosis within the FASD continuum.[15,226] Women in this sample may have stopped drinking upon pregnancy recognition which may have occurred prior to seeking prenatal care. Evidence suggests that the earlier in pregnancy alcohol cessation occurs, the better potential outcome for the child.[15] An alcohol measure which captures the entire pregnancy may better predict infant outcomes. Additionally, there may be other postnatal environmental factors (e.g., socioeconomic status, physical home

environment, sickness, or maternal affect) which affect neurodevelopmental outcomes within the first year of life.[227–230]

Previously we have demonstrated that compared to neurobehavior, dysmorphology correlates more strongly to heavy prenatal alcohol consumption.[193,231] Given that there is no one, specific neurocognitive and behavioral phenotype of children with FASD,[218,232] the differences between exposed and unexposed infants may be subtle within the first year of life. Even without a formal diagnosis on the FASD continuum, 25% of all infants in this sample fell within the borderline range for at least one domain and 6.7% were classified as extremely low in at least one domain by 9 months. In this sample, more infants fell within the lower extreme than would otherwise be expected.[40] Therefore, it may be more advantageous in this population and others to assess development at an older age or continue to follow infants into early childhood to track the developmental trajectory or change in trajectory across time, especially if prenatal alcohol exposure is suspected.

South Africa has the highest reported prevalence of FASD in the world with recent estimates ranging from 17-28%.[2–9] Many children with FASD go undiagnosed or are misdiagnosed in populations around the world.[11] There is a critical need for early evaluation and diagnosis for children with suspected prenatal alcohol exposure. Because the adverse effects of prenatal alcohol exposure are sustained throughout the lifetime,[233] it is imperative that diagnosis and developmental support begins as early as possible in order for them to receive the full benefit of interventions. While alcohol is the teratogenic agent leading to a diagnosis within the FASD continuum, other early life indicators may better help identify children at risk for an FASD diagnosis.[99] Prematurity, intimate partner violence, advanced maternal age, and socioeconomic factors of the child's environment may all be possible indicators to help clinicians identify

children at risk for an FASD diagnosis. Lower maternal weight seven years postpartum has repeatedly been associated with an FASD diagnosis for South African children in the first grade with a particularly strong association for children with a FAS diagnosis.[13,159] This study suggests higher maternal weight postpartum is a positive predictor of infant neurodevelopment within the first year of life. Higher maternal weight may have a biological effect (e.g., more body mass to distribute alcohol, thus lowering the concentration of fetal exposure) or higher maternal weight may be an indication of more favorable postnatal environmental factors. Studying children with alcohol exposure for longer time periods in longitudinal studies will be necessary to determine if the effect of higher maternal weight postpartum is sustained and beneficial as the child ages.

Strengths and Limitations

This paper has strengths. First, this was a prospective study which recruited pregnant women seeking prenatal care in five communities known to report alcohol consumption reliably and accurately in prenatal clinics.[234] Second, neurocognitive assessments were completed using the Bayley Scales of Infant and Toddler Development which have been widely used internationally and validated for South African populations. Third, the amount of missing data was low. Inevitably there are missing data in longitudinal studies; however, the linear mixed model approach utilized in this analysis is robust and utilized all available data for each infant thereby capitalizing on all participants recruited into this study. Additionally, the longitudinal nature of this study allowed for assessing the rate of change over time rather than assessing a single point in time.

There were limitations as well. First, alcohol consumption information was obtained for the 30 days prior to the interview. While women in these communities have been shown to be forthright in their reporting of alcohol consumption in the prenatal clinic,[234] some women may have stopped drinking prior to the interview. However, the trimester interviewed was not a

strong predictor of any of the neurocognitive outcomes. Second, because interviews occurred during pregnancy, information about postnatal factors were not obtained in this study. Similarly, information about educational attainment and household income were not collected in this study. Developmental delays can be caused by many factors including alcohol and socioeconomic and household environment. Future studies which capture aspects of the socioeconomic and household environment are needed. Third, information about breastfeeding was not obtained. In these communities the majority (>90%) of women breastfeed.[156] Breastfeeding is known to be positively associated with child outcomes.[151] But, consuming alcohol during the breastfeeding period has been demonstrated to be both common and harmful to the developing infant in these populations where this study was carried out.[156] However, the beneficial effect of breastfeeding or the possibility of a harmful effect of alcohol exposure via breastmilk cannot be determined in this study.

Conclusion

While other studies have suggested that maternal weight may be a protective factor in alcohol-exposed pregnancies and result in less severe effects of prenatal alcohol exposure,[138] to our knowledge, this is the first study which examined differences in infant neurocognitive abilities by maternal weight. Overall, the findings suggest that in this South African population, higher maternal weight may be beneficial to some neurodevelopmental abilities. Higher maternal weight contributed to higher initial performance, but the developmental growth across time may be similar among all infants regardless of maternal weight. Longitudinal studies are warranted to determine whether maternal weight remains protective in alcohol-exposed pregnancies.

Table 4.1 Maternal Characteristics as Reported During Pregnancy by Maternal Weight Tertile

	Tertile 1 (<56.0 kg) (n=135)		Tertile 2 (56.0 – 67.9 kg) (n=136)		Tertile 3 (≥68 kg) (n=135)		p
Weight within 10 days of birth (kg)	48.5	(5.1)	61.7	(3.4)	80.6	(10.7)	<.001[A,B,C]
Age	26.2	(5.5)	27.1	(6.1)	28.2	(5.5)	.012[B]
Gravidity	2.7	(1.3)	2.8	(1.4)	2.9	(1.2)	.575
Parity	2.1	(1.3)	2.2	(1.2)	2.2	(1.1)	.654
Used tobacco (% Yes)	57.8		47.8		48.9		.197
AUDIT Total	10.4	(8.9)	9.2	(8.7)	8.9	(8.8)	.290
TACE Total	2.7	(0.6)	2.6	(0.7)	2.6	(0.6)	.176
Trimester interviewed							
First	5.9		10.3		8.1		
Second	23.0		22.8		25.2		
Third	71.0		66.9		66.7		.728
Drank in previous 30 days (% Yes)	25.2		23.5		20.0		.585
DDD – previous 30 days[1]	6.3	(5.7)	6.3	(5.2)	6.6	(5.0)	.973
Number of drinking days – previous 30 days[1]	4.3	(3.3)	4.4	(3.8)	3.1	(3.7)	.297
Number of 3+ binges – previous 30 days[1]	4.0	(3.5)	3.6	(3.9)	2.9	(3.8)	.517
AUDIT Total[1]	17.9	(6.2)	17.4	(7.0)	17.0	(6.2)	.887
TACE Total[1]	4.2	(.6)	3.8	(.9)	3.9	(.9)	.072

1. Among those who reported drinking in the previous 30 days.

DDD: Drinks per drinking day; AUDIT: Alcohol Use Disorders Identification Test

Post-hoc Dunnet C comparisons significant difference between:

A. Tertile 1 & Tertile 2;

B. Tertile 1 & Tertile 3;

C. Tertile 2 & Tertile 3.

Table 4.2 Child Neurocognitive Abilities Measured by the Bayley Scales of Infant Development at 6 Weeks and 9 Months by Maternal Weight Tertile

	Tertile 1 (≤53.0 kg)		Tertile 2 (53.1 – 64.0 kg)		Tertile 3 (≥64.1 kg)		
	Mean	(SD)	Mean	(SD)	Mean	(SD)	p
Birth	(n=135)		(n=136)		(n=135)		
Sex (% Male)	51.1		52.2		48.1		.788
Gestational Age	39.2	(1.1)	39.1	(1.1)	39.3	(1.0)	.354
APGAR – 1 minute	9.1	(1.4)	8.9	(1.1)	9.0	(1.1)	.466
APGAR – 5 minutes	9.7	(0.9)	9.7	(0.6)	9.7	(0.5)	.886
6 weeks	(n=123)		(n=126)		(n=126)		
Age (in days)	44.3	(5.0)	43.5	(4.4)	44.6	(5.4)	.184
Cognitive Percentile	63.7	(27.4)	70.7	(23.2)	67.0	(25.3)	.094
Composite Score	106.7	(13.8)	109.8	(12.5)	108.1	(13.3)	.162
Language Percentile	37.8	(23.2)	41.5	(22.6)	40.3	(22.7)	.427
Composite Score	93.9	(11.7)	95.5	(11.3)	94.9	(11.6)	.508
Motor Percentile	67.0	(20.9)	70.5	(21.5)	68.5	(23.4)	.455
Composite Score	107.9	(9.9)	109.2	(10.2)	108.8	(11.5)	.569
Social-Emotional Percentile	60.7	(22.2)	65.1	(22.4)	63.3	(24.0)	.303
Composite Score	104.6	(10.9)	107.0	(11.1)	106.2	(12.0)	.569
9 months	(n=117)		(n=121)		(n=125)		
Age (in days)	277.4	(8.2)	275.9	(8.0)	277.2	(9.6)	.361
Cognitive Percentile	70.3	(28.4)	69.6	(26.2)	75.3	(23.9)	.186
Composite Score	110.4	(16.5)	109.5	(14.4)	113.2	(13.3)	.120
Language Percentile	35.1	(22.6)	35.3	(22.0)	41.2	(23.9)	.060
Composite Score	92.6	(12.1)	92.7	(11.9)	95.7	(12.1)	.060
Motor Percentile	34.3	(26.4)	40.8	(25.8)	40.5	(25.6)	.099
Composite Score	90.7	(15.5)	95.5	(12.8)	95.3	(12.6)	.011[A,B]
Social-Emotional Percentile	48.9	(31.6)	60.0	(30.0)	53.5	(31.2)	.022[A]
Composite Score	99.3	(16.3)	105.0	(15.3)	101.2	(16.1)	.019[A]

Post-hoc Dunnet C comparisons significant difference between:

A. Tertile 1 & Tertile 2;

B. Tertile 1 & Tertile 3;

C. Tertile 2 & Tertile 3.

Table 4.3 Percent of Infants 2 and 1.5 Standard Deviations (SD) Below the Mean on the Bayley Scales of Infant Development at 6 Weeks and 9 Months by Maternal Weight Tertile

Composite Score	All infants %	Tertile 1 (≤53.0 kg) %	Tertile 2 (53.1 – 64.0 kg) %	Tertile 3 (≥64.1 kg) %	P[1]
6 weeks		(n=123)	(n=128)	(n=125)	
Cognitive <70	0.8	0.8	0.8	0.8	.999
Cognitive ≤78	1.6	1.6	1.6	1.6	.999
Language <70	2.9	2.4	3.9	2.4	.716
Language ≤78	9.3	8.9	9.4	9.5	.987
Motor <70	0.3	0.0	0.0	0.8	.368
Motor ≤78	0.5	0.0	0.8	0.8	.615
Social/Emotional <70	0.0	0.0	0.0	0.0	--
Social/Emotional ≤78	1.6	2.4	0.8	1.6	.577
Any domain <70	3.2	3.3	3.9	2.4	.792
Any domain ≤78	10.9	12.2	9.4	11.2	.767
9 months		(n=117)	(n=120)	(n=125)	
Cognitive <70	1.4	2.6	1.7	0.0	.220
Cognitive ≤78	2.5	3.4	2.5	1.6	.662
Language <70	3.9	4.3	5.0	2.4	.552
Language ≤78	9.9	11.1	10.8	8.0	.667
Motor <70	3.3	7.7	1.7	0.8	.005[A,B]
Motor ≤78	11.6	17.9	6.7	10.4	.022[B]
Social/Emotional <70	0.6	0.9	0.0	0.8	.604
Social/Emotional ≤78	7.5	9.5	4.2	8.9	.233
Any domain <70	6.7	8.6	7.5	4.0	.328
Any domain ≤78	25.6	30.4	21.7	25.0	.300

Note: a composite score ≤70 is 2 standard deviations below the mean. A composite score of ≤78 is 1.5 standard deviations below the mean. Per Hoyme et al., 2016 diagnostic guidelines for FASD,[35] a neurocognitive score 1.5 SD below the mean meets criteria for evidence of developmental delay.

1. Chi-square with the 'all infants' column excluded from the analysis.

Post-hoc z-test of proportions significant difference between: [A]Tertile 1 & Tertile 2; [B]Tertile 1 & Teritle 3.

Table 4.4 Stepwise Regression Predicting Outcomes as Measured by the Bayley Scales of Infant Development Infant at 9 Months of Age

Infant Outcome	Step	R	R^2	Adjusted R^2	Std. Error of the Estimate	R Square Change	F Change	df1	df2	Sig. F Change
Cognitive Percentile	1	.016	.000	-.003	26.311	.000	.097	1	359	.755
	2	.164	.027	.013	26.104	.027	2.425	4	355	.048
	3	.164	.027	.010	26.141	.000	.008	1	354	.929
	4	.178	.032	.012	26.114	.005	1.725	1	353	.190
	5	.206	.042	.021	26.005	.011	3.959	1	352	.047
	6	.209	.043	.019	26.027	.001	.406	1	351	.524
	7	.218	.048	.020	26.009	.004	1.488	1	350	.223
Language Percentile	1	.040	.002	-.001	22.9945	.002	.587	1	359	.444
	2	.175	.031	.017	22.7842	.029	2.665	4	355	.032
	3	.177	.031	.015	22.8095	.001	.212	1	354	.646
	4	.205	.042	.023	22.7140	.011	3.985	1	353	.047
	5	.221	.049	.027	22.6681	.007	2.431	1	352	.120
	6	.224	.050	.026	22.6802	.002	.624	1	351	.430
	7	.225	.050	.023	22.7112	.000	.042	1	350	.838
Motor Percentile	1	.082	.007	.004	25.9365	.007	2.444	1	359	.119
	2	.175	.030	.017	25.7687	.024	2.173	4	355	.072
	3	.178	.032	.015	25.7881	.001	.466	1	354	.495
	4	.183	.033	.014	25.8024	.002	.608	1	353	.436
	5	.198	.039	.018	25.7598	.006	2.167	1	352	.142
	6	.203	.041	.016	25.7741	.002	.611	1	351	.435
	7	.203	.041	.014	25.8090	.000	.051	1	350	.822
Social/ Emotional Percentile	1	.007	.000	-.003	31.2327	.000	.018	1	357	.894
	2	.113	.013	-.001	31.2086	.013	1.138	4	353	.338
	3	.153	.024	.007	31.0821	.011	3.879	1	352	.050
	4	.163	.027	.007	31.0778	.003	1.096	1	351	.296
	5	.165	.027	.005	31.1125	.001	.219	1	350	.640
	6	.201	.040	.016	30.9454	.013	4.789	1	349	.029
	7	.211	.044	.017	30.9255	.004	1.450	1	348	.229

Step 1: DDD; Step 2: Tobacco use during pregnancy, gravidity, maternal age; trimester of pregnancy when maternal interview occurred; Step 3: Gestational age at birth; Step 4: Sex of infant; Step 5: Maternal weight; Step 6: Number of days postpartum maternal weight was assessed; Step 7: DDD by Maternal Weight Interaction

Table 4.5 Linear Mixed Model Predicting Bayley Cognitive Percentile Rank

				95% CI	
	Estimate	SE	p	Lower	Upper
Fixed Effects					
Intercept	99.82	7.67	<.001	84.74	114.89
DDD – previous 30 days (log)	-2.91	2.92	0.319	-8.65	2.83
Maternal Weight	0.16	0.07	0.018	0.03	0.29
Trimester Interviewed	-2.95	1.59	0.065	-6.08	0.19
Gestational Age at Birth	-0.69	0.92	0.452	-2.49	1.11
Sex (Male)	-29.01	7.64	<.001	-44.03	-14.00
Sex (Female)	--	--			
Tobacco (Yes)	-0.22	2.59	0.933	-5.32	4.88
Tobacco (No)	--	--			
Gravidity (log)	-31.41	11.22	0.005	-53.49	-9.34
Maternal Age (log)	14.93	14.68	0.310	-13.95	43.80
Day Weight Measured	-1.55	0.53	0.004	-2.58	-0.51
Gravidity * sex (male)	43.23	13.21	0.001	17.19	69.15
Time	-1.16	0.56	0.037	-2.25	-0.07
Tobacco (Yes) * time	1.07	0.46	0.020	0.17	1.97
Day Weight Measured * time	0.25	0.09	0.008	0.07	0.44

Covariates without a time interaction term indicate the effect of the covariate on infant physical outcome at 6 weeks of age. The variable, time, indicates whether there is a change in the slope of the trajectory across time among all individuals. Covariate by time interactions (covariate*time) indicate the rate of change (slope) of the trajectory attributable to the covariate.
DDD: drinks per drinking day

Table 4.6 Linear Mixed Model Predicting Bayley Language Percentile Rank

				95% CI	
	Estimate	SE	p	Lower	Upper
Fixed Effects					
Intercept	54.17	5.82	<.001	42.72	65.63
DDD – previous 30 days (log)	-2.32	2.68	0.387	-7.58	2.94
Maternal Weight	0.10	0.06	0.090	-0.02	0.22
Trimester Interviewed	-2.87	1.46	0.049	-5.74	-0.01
Gestational Age at Birth	-0.80	0.84	0.344	-2.45	0.86
Sex (Male)	-3.81	1.79	0.034	-7.33	-0.29
Sex (Female)	--	--			
Tobacco (Yes)	-2.45	1.85	0.186	-6.08	1.18
Tobacco (No)	--	--			
Gravidity (log)	-10.27	8.32	0.218	-26.63	6.10
Maternal Age (log)	5.14	13.46	0.703	-21.32	31.60
Day Weight Measured	-0.07	0.37	0.843	-0.80	0.65
Time	-0.35	0.20	0.080	-0.75	0.04

Covariates without a time interaction term indicate the effect of the covariate on infant physical outcome at 6 weeks of age. The variable, time, indicates whether there is a change in the slope of the trajectory across time among all individuals. No two-way time interactions were significant.
DDD: drinks per drinking day

Table 4.7 Linear Mixed Model Predicting Bayley Motor Percentile Rank

				95% CI	
	Estimate	SE	p	Lower	Upper
Fixed Effects					
Intercept	80.05	5.88	<.001	68.49	91.62
DDD – previous 30 days (log)	-3.04	2.70	0.261	-8.36	2.27
Maternal Weight	0.27	0.10	0.005	0.08	0.45
Trimester Interviewed	0.03	1.48	0.982	-2.87	2.93
Gestational Age at Birth	1.29	0.85	0.128	-0.37	2.96
Sex (Male)	-1.99	1.81	0.272	-5.54	1.56
Sex (Female)	--	--			
Tobacco (Yes)	1.68	1.86	0.367	1.98	5.35
Tobacco (No)	--	--			
Gravidity (log)	-17.79	8.40	0.035	-34.31	-1.27
Maternal Age (log)	-10.06	13.59	0.459	-36.77	16.65
Day Weight Measured	-0.22	0.37	0.559	-0.95	0.52
Maternal Weight * Sex (Male)	-0.26	0.12	0.036	-0.50	-0.02
Time	-3.89	0.22	<.001	-4.32	-3.47

Covariates without a time interaction term indicate the effect of the covariate on infant physical outcome at 6 weeks of age. The variable, time, indicates whether there is a change in the slope of the trajectory across time among all individuals. The maternal weight by sex interaction (maternal weight*sex) indicates as maternal weight increases males perform significantly worse than females.
No two-way time interactions were significant.
DDD: drinks per drinking day

Table 4.8 Linear Mixed Model Predicting Bayley Social/Emotional Percentile Rank

				95% CI	
	Estimate	SE	p	Lower	Upper
Fixed Effects					
Intercept	83.05	7.01	<.001	69.26	96.84
DDD – previous 30 days (log)	-15.34	5.90	0.010	-26.94	-3.73
Maternal Weight	0.08	0.07	0.294	-0.07	0.22
Trimester Interviewed	-3.05	1.75	0.082	-6.50	0.39
Gestational Age at Birth	-1.46	1.01	0.149	-3.45	0.53
Sex (Male)	0.41	2.15	0.850	-3.82	4.63
Sex (Female)	--	--			
Tobacco (Yes)	-3.14	2.42	0.196	-7.89	1.62
Tobacco (No)	--	--			
Gravidity (log)	-16.78	10.01	0.094	-36.46	2.90
Maternal Age (log)	35.02	16.17	0.031	3.23	66.82
Day Weight Measured	-0.82	0.44	0.064	-1.69	0.05
DDD * Smoking (Yes)	15.00	6.92	0.031	1.40	28.60
Time	-1.11	0.22	<.001	-1.54	-0.68

Covariates without a time interaction term indicate the effect of the covariate on infant physical outcome at 6 weeks of age. The variable, time, indicates whether there is a change in the slope of the trajectory across time among all individuals. No two-way time interactions were significant.
DDD: drinks per drinking day

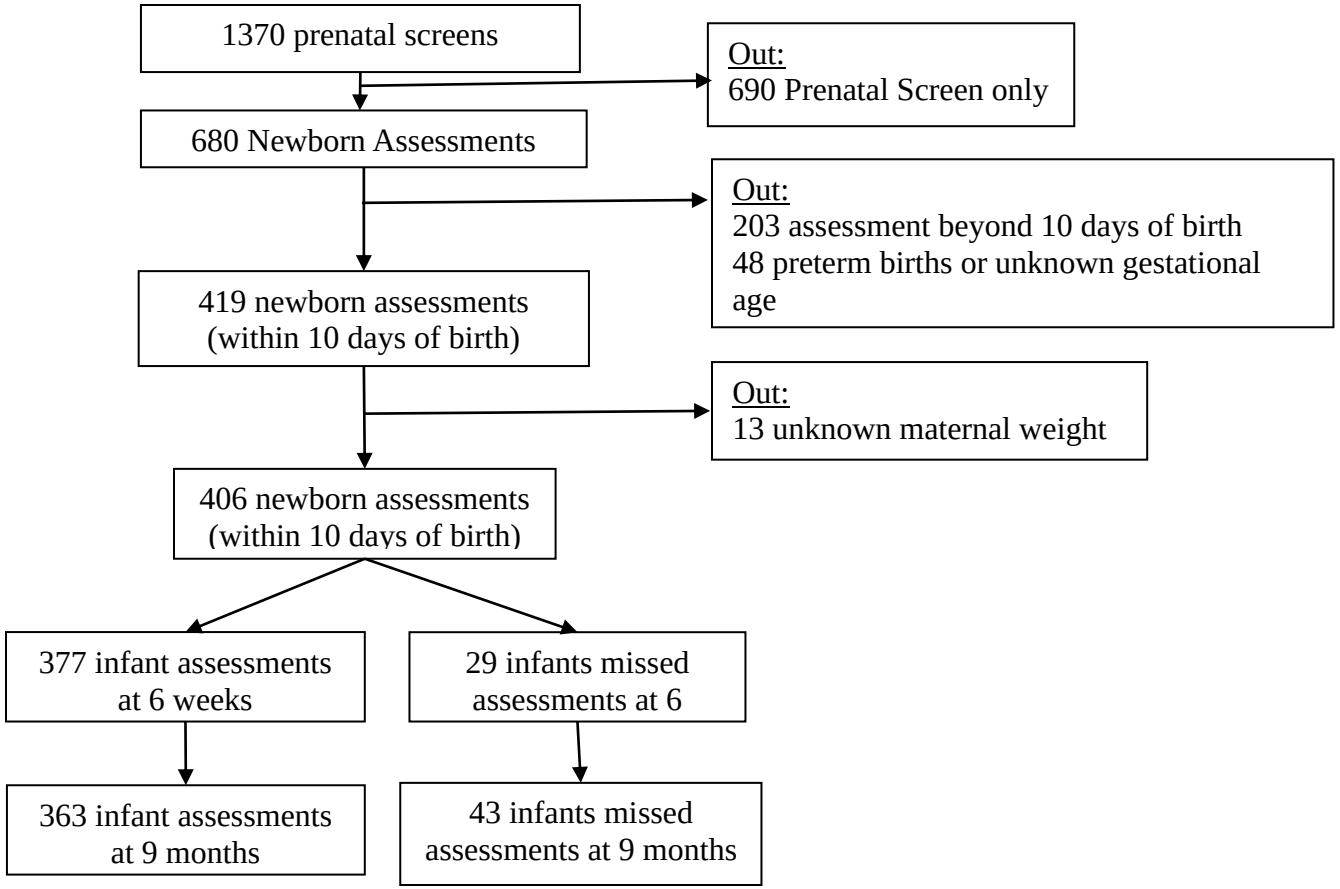

Figure 4.1 Study Consort Chart

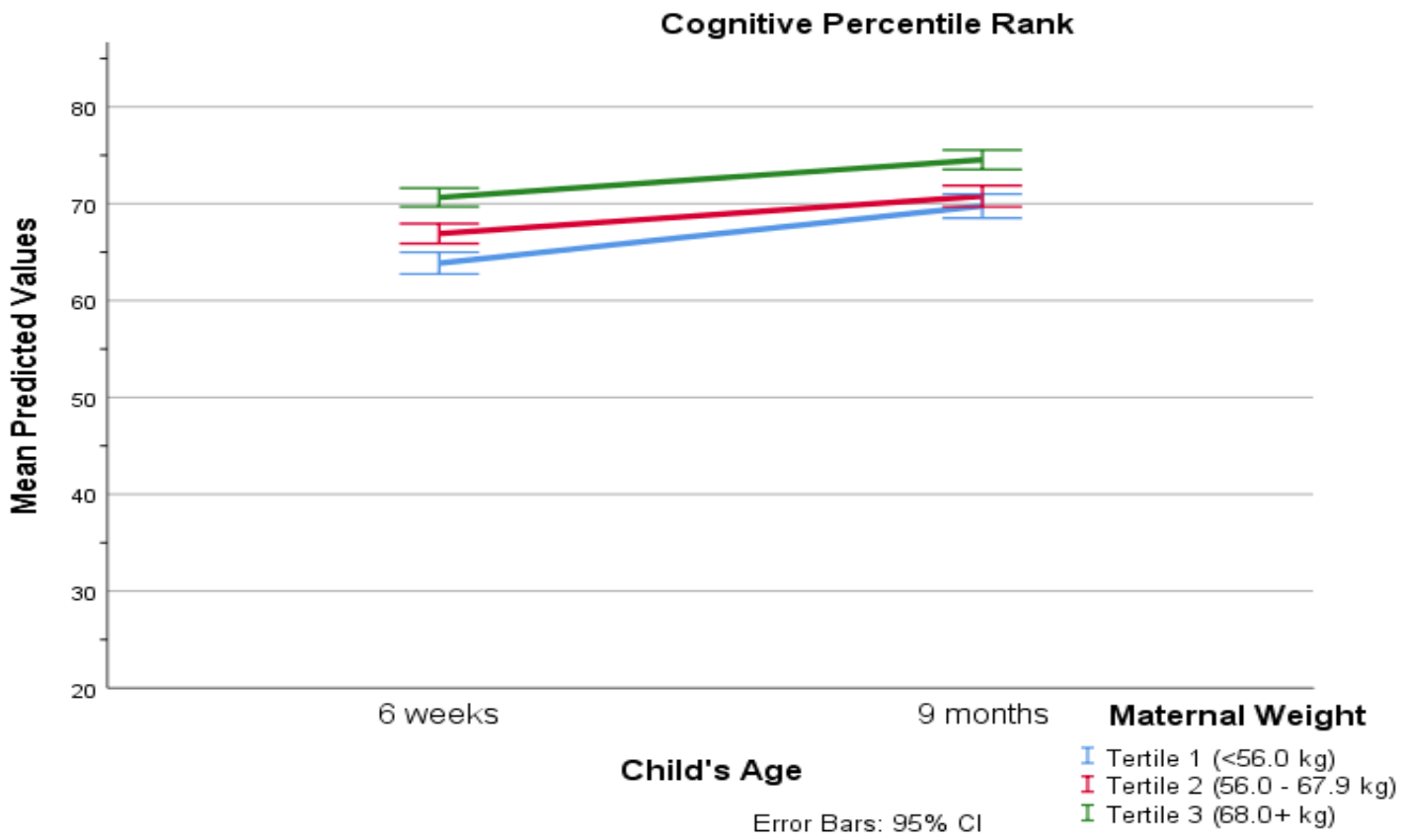

Figure 4.2 Maternal Weight Significant Predicts 6 Weeks Cognitive Percentile Rank (B=0.16, p=.018). There is no Significant Difference in the Rate of Change (Slope) by Maternal Weight.

CHAPTER 5: THE EFFECT OF ALCOHOL AND MATERNAL DIETARY INTAKE ON INFANT PHYSICAL OUTCOMES AT SIX WEEKS OF AGE

Overview

Background: All pregnant women should strive for an optimal dietary intake. Optimal dietary intake during pregnancy has multiple benefits and affects both short- and long-term outcomes in women and their children. Dietary intake is known to be poor in individuals who consume heavy quantities of alcohol. Both maternal diet and alcohol intake can adversely affect fetal and child physical outcomes.

*Method*s: Two, 24hr dietary recalls from pregnant women in the Western Cape Province of South Africa (*n*=178) were utilized to assess alcohol consumption and dietary intake on infant outcomes. A structural equation model was constructed to determine whether there was a direct and/or indirect effect of alcohol and maternal dietary intake on child outcomes at 6 weeks of age.

Results: Virtually all women were likely deficient (intake < Estimated Average Requirement) on most micronutrients. Controlling for six covariates, alcohol was significantly and negatively correlated with six micronutrients: vitamin K, thiamin (B_1), phosphorus, zinc, sodium, and selenium as well as the total number of micronutrient deficiencies. There was a significant, direct effect of alcohol on infant physical outcomes. There was also a significant and direct effect of alcohol on maternal micronutrient intake as measured by the number of micronutrient intakes below the Recommended Dietary Allowance. There was no significant indirect effect of alcohol via maternal micronutrient intake on infant outcomes at six weeks of age.

Conclusion: Many women had deficient intake of most micronutrients regardless of alcohol consumption during pregnancy. Yet alcohol had a negative effect on dietary intake and had a direct, negative effect on infant outcomes at six weeks of age. Although no significant indirect effect of alcohol via micronutrient intake was demonstrated, additional studies utilizing maternal plasma concentrations of nutrients may better elicit whether there is an indirect effect of alcohol via maternal nutrition on infant physical outcomes at 6 weeks of age.

Introduction

Alcohol, a known teratogen, crosses the placenta and can affect fetal tissue, physiology, function, and development. Prenatal alcohol exposure can have life-long implications for an individual, especially for individuals who fall within the fetal alcohol spectrum disorders (FASD) continuum. Many pregnancies are unplanned with alcohol exposure occurring before pregnancy recognition, resulting in many alcohol-exposed pregnancies annually.[235–240] In the United States the estimated prevalence of FASD in the general population is 1-5%.[1] The Western Cape Province of South Africa has the highest reported FASD prevalence in the world with an estimated 17-28% of children in the general population falling within the FASD continuum.[2–9]

Although alcohol is known to dysregulate fetal development, there is tremendous variation in the physical outcomes of individuals prenatally-exposed to alcohol which cannot be explained by the quantity, frequency, and gestational timing of the alcohol exposure.[15] Other maternal factors which have been reported to contribute to the vulnerability and severity of FASD diagnosis include: advanced maternal age,[5,6,8,19,20,157–159] smoking during pregnancy,[4,13] socioeconomic status and/or educational attainment,[6,8,68,159,161] and maternal body mass index (BMI).[6,8,68,159,161] Because alcohol is known to interact with many nutrients, maternal nutrient intake has also been suggested as an important determinant of the risk and severity of having a child with an FASD diagnosis.

It is essential for all pregnant women, whether alcohol consuming or not, to have optimal maternal dietary intake. Micronutrient deficiencies are common globally among women of childbearing age.[241] Alcohol contributes 7.1 kcal/g of energy, and the continued use of alcohol can replace other macronutrients as a primary energy source. Alcohol is primarily metabolized via the alcohol dehydrogenase (ADH) pathway and to a lesser extent via the microsomal ethanol oxidizing system (MEOS) and catalase mechanisms. All three mechanisms oxidize ethanol to highly toxic acetaldehyde before acetaldehyde dehydrogenase (ALDH) further oxidizes acetaldehyde to acetate (non-toxic). The toxic acetaldehyde leads to increased reactive oxygen species, free radicals, and cell damage.[225] Because alcohol is a toxic substance, the body prioritizes the metabolism of alcohol over other macronutrients in all three mechanisms.[242–244]

In addition to the production of toxic by-products during alcohol metabolism, alcohol can compromise the maternal nutritional status through the displacement or malabsorption and utilization of essential nutrients. The severity of the malnutrition depends on the quantity of alcohol consumed and the quality of food consumed. Alcohol can dysregulate gastrointestinal function, impair placental function, and inhibit placental transfer of nutrients which all contribute to a reduction in the bioavailability of nutrients for the fetus.[161,179] A compromised nutritional environment can result in suboptimal fetal growth and development.[110] Several micronutrients have been suggested as likely contributors or ameliorators of the effects of prenatal alcohol exposure; yet there is limited research on nutrition and FASD in humans.[109–113] The interaction of alcohol with micronutrients may partially explain the variation in infant outcomes. Alcohol competes in the metabolism of vitamin A and depletes maternal stores and reduces availability to the fetus, therefore, leading to interruptions of normal cellular processes in the fetus.[110,180] Folic acid absorption is reduced by alcohol consumption and deficiencies in folate can lead to altered

DNA and RNA synthesis resulting in inappropriate cellular apoptosis.[181] In the presence of alcohol, when there is an acute zinc deficiency, zinc is maladaptively sequestered in the maternal liver leading to reduced bioavailability for placental transfer and fetal zinc deficiency.[111,182] Maternal regulation of calcium is impaired following alcohol consumption and fetal skeletal ossification can be reduced in alcohol-exposed fetuses.[183] Alcohol alters one-carbon metabolism which involves choline, betaine, folate, methionine, vitamin B_6, vitamin B_{12}, and homocysteine.[184] The alteration in one-carbon metabolism results in genome-wide hypomethylation in the fetus.[185] The maternal status of docosahexaenoic acid (DHA) and the placental transfer of DHA to the fetus is reduced by alcohol consumption.[245] Alcohol increases the demand for antioxidants (vitamin A, vitamin E, and selenium). Additional antioxidants are needed to neutralize the free radicals and reactive oxygen species produced by the metabolism of alcohol and because alcohol also reduces the endogenous antioxidant levels (e.g., glutathione peroxidase).[110,186,187]

Independent of prenatal alcohol exposure, poor maternal nutrition during pregnancy has been associated with low birth weight, poor fetal growth, and congenital malformations.[246] Although the exposure to poor nutrition during gestation is short in duration, the maternal diet and nutritional status can have profound effects on the health of the child into adulthood.[176,177] The suboptimal availability of nutrients to the fetus during gestation can lead to alterations of cellular and tissue function which can be maladaptive and detrimental in the long-term. While prenatal alcohol exposure is the sole cause of an FASD diagnosis, it is plausible that maternal nutrient intake mediates the effect of alcohol on child outcomes. Prenatal alcohol exposure may lead to poor maternal nutrient intake which may place a fetus at greater risk for poor birth outcomes and at risk for being diagnosed on the FASD continuum. The purpose of this paper

was to determine the direct and indirect effect of alcohol and maternal dietary intake on infant physical outcomes at six weeks of age.

Methods

Study Design and Sample

In five communities of the Western Cape Province of South Africa, all women who were seeking prenatal care were invited to complete a brief alcohol screening assessment. The assessment included the Alcohol Use Disorders Identification Test (AUDIT) which is a widely-used tool that identifies risky drinking in the previous 12 months.[197] Women were further invited to participate in a nutrition sub-study, if at the time of enrollment into the sub-study, the women were between 5-36 weeks gestation. Both women who reported consuming alcohol during pregnancy and non-drinkers were recruited.

Two hundred and eighty-one (281) women agreed to participate to complete 2, 24hr dietary recalls and complete an extensive, in-person maternal interview which assessed demographics, childbearing history, and alcohol consumption during the pregnancy. Each woman completed a 24hr dietary recall for one weekday and one weekend day (Friday or Saturday) during pregnancy. The recalls were completed in-person with trained interviewers using a multi-pass method which elicits increasingly more detail about the foods consumed with each pass. Information about oral dietary supplements was also ascertained during the recalls. The dietary recalls were entered into the Nutrition Data System for Research (NDSR) version 2014.[247] NDSR is a comprehensive nutrient calculation software which contains more than 20,000 foods and over 8,000 brand-name items. For foods that are specific to South Africa (e.g., Nutri-Mil®, a complete soy-based powder mixed with water for adults), user recipes were created and matched to the publicly-available formulations provided by the manufacturer.

NDSR estimated total micronutrient intakes via dietary intake and oral supplement intake for each day.

During the index pregnancy, expectant mothers also completed an in-depth in-person maternal interview which assessed demographic traits, childbearing history, and alcohol consumption before and during the pregnancy. Maternal height, weight, occipitofrontal (head) circumference (OFC), and left upper arm circumference was also measured at the time of the interview. Alcohol consumption was assessed via the AUDIT as well as questions about the quantity (drinks per drinking day) and frequency of drinking (the number of drinking days per week) in the 3 months prior to pregnancy.

Immediately following the birth, the infant's length, weight, and OFC were measured by the attending physician or nurse and was recorded on the infant's medical card. The physical characteristics and dysmorphology of the infants were assessed by grant-funded study staff at 6 weeks of age for 178 infants. The six week physical examination included measuring the infant's length, weight, and OFC and assessing the presence or absence of 12 other minor anomalies (e.g., strabismus, ptosis, epicanthal folds, flat nasal bridge, anteverted nares, "railroad track" and "cupped" ears, and altered palmer creases).[35] The philtrum and the vermilion of the upper lip were assessed using the mixed-race lip/philtrum guide developed previously for this particular population.[248] The South African study staff were trained by American pediatricians, who are fellowship-trained and board-certified in medical genetics/dysmorphology, on the appropriate methods and techniques for completing the dysmorphology exam. The examiners were blinded to the alcohol exposure and all other knowledge of the infant's background.

Statistical Analysis

Dietary data from the two separate recall days were combined into a single intake amount for each woman. For each recall the micronutrient intake from supplements was added to the

micronutrient intake from food sources for each woman. Both a simple average and a weighted average of the weekday and the weekend recall were estimated. The weighted average was estimated by multiplying the weekday recall by .714 (5/7) and the weekend day recall by .286 (2/7). Using both the simple average and the weighted average, women were classified as likely inadequate by the US Institute of Medicine's Estimated Average Requirements (EAR) and Recommended Dietary Allowance (RDA) for pregnant women.[249] The EAR for a given micronutrient is defined as the intake needed to meet the nutritional needs for 50% of individuals in a specific sex and life stage. The RDA is defined as the necessary intake to meet the nutritional needs of 97-98% of healthy individuals in a specific sex and life stage. When there is insufficient scientific evidence to establish a RDA, an Adequate Intake (AI) is determined.

Women were divided into three maternal groups: 1) women who reported drinking during pregnancy and had any alcohol in the dietary recalls (any alcohol in recalls); 2) women who reported drinking previously during the pregnancy but did not have any alcohol in the dietary recalls (quitter/less frequent drinkers); and 3) women who did not drink during pregnancy. Maternal demographic, maternal alcohol consumption patterns, and child physical characteristics were compared using one-way analysis of variance (ANOVA) with post-hoc Dunnett C pairwise comparisons (alpha= 0.05). Dunnett C comparisons control for Type 1 error (false positives) produced when performing multiple comparisons of group means.[199] Categorical variables were examined using chi-square. To assess the relationship between alcohol intake and dietary intake, bivariate and partial correlations were undertaken.

Finally, a structural equation model was utilized to examine the direct and indirect effect of alcohol intake on the infant physical outcomes at six weeks of age. The indicators for the latent variable of alcohol were the weekday and weekend day alcohol consumption quantities

from the 24hr dietary recalls. Indicators of the maternal latent variable were known maternal risk factors that have been associated with FASD diagnosis in several cross-sectional studies in the Western Cape Province.[6–9] These indicators included: educational attainment, tobacco use during pregnancy, gravidity, and maternal BMI. Indicators of the infant latent variable were total dysmorphology score, number of minor anomalies, and OFC centile. The number of minor anomalies has been shown to be correlated with an FASD diagnosis, and total dysmorphology score at 9 months has been shown to be predictive of a future FASD diagnosis at 5 years of age.[6,7,99] OFC centile is the differentiating diagnostic criteria for children with FAS and PFAS.[35] OFC centile is also considered a proxy for brain volume and growth.[250] The number of micronutrient deficiencies, a measured variable, was selected as the summary maternal nutrition variable. Treating maternal nutrition as a latent variable was also explored, but model fit did not improve and the results did not differ. The alcohol and maternal latent variables were scaled to have a variance of one. Maximum likelihood estimation was utilized in the structural equation model. There were no missing data in the model. All analyses were carried out in SPSS (version 26) and AMOS (version 27).[200]

Results

The maternal demographic, childbearing, and alcohol consumption information is displayed in Table 5.1. Maternal weight, BMI, years of education, marital status, tobacco use, number of drinks per week before pregnancy, number of drinking days per week before pregnancy, estimated drinks per drinking day before pregnancy, and the total AUDIT score significantly distinguished the maternal groups. Women who had any alcohol in the recalls (group 1) had significantly lower weight, BMI, and educational attainment, were least likely to be married, were more likely to use tobacco during pregnancy, and reported a higher quantity and frequency of alcohol consumption before pregnancy compared to women who reported

quitting or less frequent drinking (group 2) and non-drinkers (group 3). The number of drinking days per week prior to pregnancy and total AUDIT score significantly distinguished all three maternal groups in post-hoc Dunnett C comparisons. Although not statistically significant between maternal groups, 14% - 28% reported being hungry during the pregnancy due to lack of available food in the home.

Group mean intakes are presented individually for the weekday (Appendix Table C.1) and weekend (Appendix Table C.2). For the weekday recall, only alcohol and sodium differentiated the maternal groups. For the weekend recall, alcohol, animal protein, vitamin B_{12}, and choline were significantly different between groups. Women who had any alcohol in the recalls consumed, on average, 47 grams of alcohol or approximately 3 standard drinks. Table 5.2 presents the estimated weighted average of macro- and micro- nutrient intake by alcohol consumption. As expected, grams of alcohol significantly differentiated the maternal groups. Excluding alcohol and sodium, no other macro- or micro- nutrient significantly distinguished between the groups using the weighted average. Appendix Table C.3 displays the identical table for the simple average and no macro- or micro- nutrients other than alcohol and choline were significantly different between groups.

The percent of women who were inadequate by EAR is presented in Table 5.3. While none of the comparisons were statistically significant between the groups, nine micronutrients had greater than 50% of the women not meeting EAR. Six micronutrients (vitamin A, D, E, B_6, magnesium, and zinc) had greater than 85% of the women not meeting EAR. Table 5.4 shows the percent of women who were less than the RDA for pregnant women. Similar to the EAR comparisons, the proportion of women less than RDA did not differ by maternal group. Eleven micronutrients had more than 85% of the women not meeting the RDA and 17 micronutrients

had 50% of the women not meeting the RDA. On average, women were deficient on 15 to 16 micronutrients in each maternal group; however, the range of deficiencies was from 3-25. Appendix Tables C.4 and C.5 show the identical tables using the simple mean of the recalls to estimate the percent less than EAR and RDA. The results were virtually identical. No micronutrient had all women meeting the RDA. Regardless of alcohol consumption, most women in this sample were not meeting the EAR or RDA for pregnant women.

Table 5.5 presents the infant physical characteristics at birth and 6 weeks of age by maternal drinking group. Length and weight centile significantly differentiated infants between the maternal groups. Infants exposed to alcohol (groups 1 and 2) were significantly shorter than infants born to abstainers. Infants born to women with any alcohol in the recalls (group 1) weighed significantly less than infants born to quitter/less frequent drinkers (group 2) and abstainers (group 3). At 6 weeks, length, weight, weight-for-length, OFC centile, inner canthal distance (ICD), palpebral fissure length (PFL) centile, philtrum ranking, number of minor anomalies, and total dysmorphology score significantly differentiated the groups. Infants born to group 1 (any alcohol in the recalls) had, on average, lower centile scores on length, weight, OFC, ICD, and PFL and had higher (poorer) philtrum ranking, more minor anomalies, and higher average total dysmorphology score. Length, weight, OFC, weight-for-length, PFL, and philtrum ranking significantly differentiated group 1 from group 3 in post-hoc analyses. Length and OFC centile differentiated quitter/less frequent drinkers (group 2) from abstainers (group 3) with women in group 3 having infants with higher length and OFC centiles at six weeks of age. The number of minor anomalies and total dysmorphology score significantly distinguished between group 1 from group 2 and group 3 in post-hoc comparisons. The number of minor anomalies and

total dysmorphology score did not differentiate group 2 (less frequent drinkers) from group 3 (abstainers) in post-hoc comparisons.

Using the weighted dietary recall data, bivariate and partial correlations between grams of alcohol and nutrients are displayed in Table 5.6. After controlling for maternal BMI, age, gravidity, tobacco use during pregnancy, educational attainment, and total energy consumed, alcohol consumption was negatively associated with total carbohydrate, total protein, vegetable protein, and total dietary fiber intake. Six micronutrients were significantly and negatively associated with grams of alcohol consumed. These nutrients were vitamin K, thiamin, phosphorus, zinc, selenium, and sodium. Only one micronutrient (vitamin B_6) was positively associated with the intake of alcohol.

Figure 5.1 is the hypothesized model linking alcohol intake, micronutrient intake, maternal characteristics, and infant outcomes. The ovals represent latent variables, rectangles represent measured variables, and circles are error terms. The hypothesized directionality of predicted relationships are represented by single arrow lines. Double arrow arcs represent hypothesized correlations. It was hypothesized that alcohol has a direct effect on infant physical outcomes and an indirect effect on infant physical outcomes via nutrition (dietary intake). A correlation was hypothesized between maternal characteristics and alcohol intake. Model fit was deemed adequate with a comparative fit index (CFI) of .934 and a Root Mean Square Error Approximation (RMSEA) of .070. All measured variables had significant loadings for their respective factors. The standardized coefficients are shown in Figure 5.2. Alcohol had a direct, negative effect on infant physical outcomes (standardized coefficient = -0.79, p=.027). Alcohol also had a direct effect on the number of micronutrient deficiencies as measured by RDA or AI (standardized coefficient = 0.22, p=.013). The number of micronutrient deficiencies did not have

a direct effect on infant physical outcomes (p=.918). There was no indirect effect of alcohol via the number of micronutrient deficiencies on infant physical outcomes. Appendix Figure C.1 is a structural equation model with maternal nutrition as a latent variable using micronutrient intake values as the indicator variables. The results did not change. Alcohol has a direct effect on maternal dietary intake and infant outcomes but no indirect effect via maternal micronutrient intake.

Discussion

Women in this sample reported consuming, on average, fewer calories than is generally recommended for pregnant women as well as non-pregnant women of childbearing age with sedentary lifestyles. While there was no overt disease associated with micronutrient deficiencies present in this sample, many women were deficient on several essential vitamins and minerals. Even with supplements, the majority of women were not consuming the recommended intake for many micronutrients including vitamin A, C, D, E, K, B_6, zinc, calcium, magnesium, potassium, and choline. While some micronutrients can be stored in the body, water soluble vitamins (e.g., vitamin C, B_6, and B_{12}) are not stored in the body and an individual should attempt to consume adequate quantities each day. In contrast to water soluble vitamins, fat soluble vitamins (A, D, E, and K) can be stored in the body; therefore, intake may not represent actual micronutrient status in the body. Because the body can store fat soluble vitamins, meeting the RDA for fat soluble vitamins may not be necessary everyday.[251] But since >90% of the sample did not meet the EAR for vitamin A, D, and E, it is likely that a large proportion of the sample is inadequate in fat soluble vitamins. Vitamin D can also be endogenously synthesized by the body following sun (ultraviolet B) exposure through the conversion of 7-dehydrocholesterol to cholecalciferol (vitamin D_3); therefore, not all vitamin D may need to be ingested if there is adequate sun exposure.[252] Nevertheless, individuals should strive to consume vitamin D quantities as

recommended by the Institute of Medicine. Trace elements and minerals such as iron and calcium are highly metabolically regulated to maintain homeostasis and cellular function. While plasma levels of minerals may not dramatically fluctuate with changes in dietary intake, it is still essential to consume adequate daily amounts of minerals to meet the nutritional demands of the mother and fetus without depleting the maternal nutrient stores. Iron is stored and easily mobilized from the liver. If there is inadequate intake to meet the demands of pregnancy, other mineral stores (calcium, magnesium, and zinc) located in the skeleton can be mobilized, but at the expense of skeletal integrity.[253] Beginning pregnancy with depleted mineral stores and/or depletion during pregnancy has been associated with poorer infant outcomes and lower infant nutrient stores.[253,254]

Individual micronutrient deficiencies can have an adverse effect on health, yet nutrients rarely work in isolation and optimal quantities of all micronutrients are needed to meet the additional demands of the mother, placenta, and fetus during pregnancy. The dietary staples of meat, stews (cabbage, potatoes, onions), rice, porridge (grits/polenta), white bread with margarine, tea, and instant coffee with cream and sugar in this population were not meeting the dietary requirements for most. In this population there was virtually no difference whether percent likely deficient was assessed by EAR or RDA. This indicates that those who were deficient were inadequate by a substantial amount and were classified as insufficient by non-trivial amounts. This sample is fairly similar to national South African studies where 19% of households in South Africa and 11.6% of Western Cape households experience food insecurity[26] and women and children were deficient on several micronutrients.[196] Despite several national policies designed to address nutritional status and food insecurity (e.g., food fortification, food supplementation, and school feeding programs) that have been enacted in South Africa,

nutritional deficiencies remain a concern for the majority of women. Additional supplementation may be warranted for this population, especially if pregnant. However, even with the national mandate of providing iron and folate supplements (ferrous sulphate of 170 mg and folate of 5mg to be consumed daily) to all pregnant women upon pregnancy recognition, 15% of women were not meeting dietary recommendations for these two nutrients. Increasing access to food and reducing food insecurity may also be warranted in these communities.

There was also virtually no difference in percent likely deficient by alcohol intake. Because percent likely deficient is a dichotomous variable, the relative intake among women may still be different even if women were classified as deficient. Although none met statistical significance, the mean intakes were consistently higher among women who did not drink compared to those who reported drinking during pregnancy. Moreover, alcohol can disrupt nutrient utilization by the mother and fetus through abnormal absorption,[255] altered composition and function of the microbiome,[256] altered renal function/reabsorption of nutrients,[255] and altered placental transport.[257,258] The availability and utilization of nutrients may be different between women who consume alcohol and those who abstain despite similar nutrient intake. Future studies assessing the plasma concentration of key nutrients will be necessary to determine whether the maternal and fetal availability of nutrients differ by alcohol intake.

Infant characteristics at birth and 6 weeks differentiated infants by alcohol exposure. Infants exposed to alcohol were smaller at birth and remained smaller at 6 weeks compared to unexposed children. PFL centile and vermilion border of the upper lip, two of the three cardinal facial features of FASD, also significantly distinguished the alcohol-exposed children from unexposed children. Consistent with other studies of older children with FASD, in post-hoc analysis the number of minor anomalies and total dysmorphology score at 6 weeks differentiated

infants in group 1 from both women who quit drinking/drank less frequently and non-drinkers.[99] To our knowledge, our study is the second study to show the total dysmorphology score differentiated alcohol exposed infants from unexposed infants as young as 6 weeks of age. While the total dysmorphology score is not intended to be diagnostic, this study suggests that the total dysmorphology score is a useful measure to capture the physical effect of alcohol exposure on children even in very early life.

It has been well established that alcohol is negatively associated with nutrient intake among individuals who consume heavy quantities of alcohol. Among women who had any alcohol in their recalls, they reported consuming, on average, approximately 3 standard drinks (47 grams of alcohol) per occasion. While this does not meet the National Institute of Alcohol Abuse and Alcoholism definition of a 'binge' for a female (4 standard drinks per occasion), in this sample, many micronutrients were significantly and negatively associated with alcohol consumption. This suggests that even light to moderate drinking may have a negative impact on dietary intake.

Although total grams of alcohol consumed had a direct, negative effect on maternal nutrient intake, the total grams of alcohol consumed did not have an indirect effect via maternal nutrient intake on infant physical outcomes at 6 weeks of age. Because maternal intake does not equal maternal nutrient status, these findings do not negate the possibility that maternal nutrient status (and not maternal intake) mediates the relationship between alcohol and infant physical outcomes. It is known that alcohol disrupts the absorption and utilization of several micronutrients. This analysis did not account for the metabolic inhibitions of alcohol. Further studies which assess the mediation of maternal nutrient status using plasma concentrations of

nutrients are warranted to better elicit whether there is an indirect effect of alcohol via maternal nutrition.

While there was no direct effect of maternal nutrient intake on infant outcomes, in this population, virtually all mothers were deficient on several micronutrients. Regardless of alcohol exposure status, there was little change in the percent of mothers who were likely deficient whether measured by EAR or RDA. This suggests that women were vastly under consuming many micronutrients. It is possible that mothers with a marginal increase in intake were still likely deficient and no biologically significant improvements were gained in physical outcomes for the infant at 6 weeks of age. Moreover, maternal nutrient intake contributes to the development of fetal nutrient stores which develop rapidly in the 2nd and 3rd trimester. For some nutrients (e.g., iron), the fetal/infant nutrient stores are a major source for the infant in the first 6 months after birth.[259] If infants lack adequate nutrient stores to support growth postnatally, it is possible that an assessment at a later age may demonstrate an effect or a stronger association of maternal dietary intake on infant physical outcomes. For example, shorter adult stature has been linked to growth faltering in the first 2 years of life.[260] The reductions in maternal dietary intake of specific micronutrients (e.g., methyl donors of choline, vitamin B_{12}, and folate) may have also led to long-term epigenetic changes which may increase the infant's susceptibility to later morbidity (obesity or insulin resistance).[261] It is also possible that infant neurodevelopmental outcomes may have been adversely affected by micronutrient deficiencies during pregnancy. Specific brain structures, function, and neurochemistry have been shown to be altered following in utero micronutrient deficiencies,[262] and infant micronutrient deficiencies have been linked to lower educational attainment and poorer human capital.[260] Future studies will be needed to explore these possibilities.

Strengths and Limitations

Strengths of this study include it is a prospective study design which recruited pregnant women seeking prenatal care in five communities. Dietary information was collected via a widely used multi-pass technique which increases the granularity of food quantity and preparation with each pass. Additionally, dietary oral supplement information was obtained from each woman which allowed for a more complete assessment of total dietary intake. Physical and dysmorphology assessments were completed using a standardized tool which has repeatedly been used in this South African population, in the United States, and elsewhere internationally. Because the mothers completed in-depth maternal demographic and childbearing interviews while pregnant, several maternal characteristics which can influence fetal outcomes were controlled in these analyses.

There were limitations as well. Because women were enrolled shortly after seeking prenatal care, the timing of the dietary recalls varied between women, yet most were interviewed in 2nd and 3rd trimester. Because few women were interviewed during the first trimester, neither morning sickness nor appetite suppression associated with the first trimester was likely a factor in this sample. Second, it is known that for some nutrients, such as iron, intake is not a good indicator of status. Without plasma concentrations of nutrients, definitive conclusions about nutrient sufficiency cannot be made. However, a diet that is consistently deficient in nutrients is more likely to have low plasma concentrations despite the endogenous mechanisms at play to maintain nutrient homeostasis. Third, although the outcome data were empirically derived with standard physical assessments, the infant data were collected within the first 6 weeks of life. Early infant physical characteristics are not yet fully stabilized and temperament and tolerance to brief physical exams are variable. Assessing children at an older age may reduce the inherent variability and artificial noise possibly associated with early infant assessments.

Conclusion

In this sample, many of the women were likely deficient on many micronutrients. Light to moderate alcohol consumption was significantly and negatively associated with many micronutrients, although no overt micronutrient deficiency diseases/syndromes were present in this sample. Infants born to mothers who consumed alcohol during pregnancy were significantly smaller and more dysmorphic than unexposed infants. Alcohol had a significant and direct teratogenic effect on infant physical outcomes. Although maternal dietary intake did not mediate the relationship between alcohol and physical outcomes at 6 weeks of age, this does not negate the possibility of long term, adverse health effects and poor outcomes for the child or that maternal nutrient status may have a direct effect on infant physical outcomes.

Table 5.1 Maternal Characteristics by Alcohol Exposure Status

Maternal Characteristic at time of interview (during pregnancy)	Drank during pregnancy with alcohol in recalls (*n*=47)		Drank during pregnancy without alcohol in recalls (*n*=76)		Did not drink during pregnancy (*n*=55)		P
	Mean	(SD)	Mean	(SD)	Mean	(SD)	
Height (cm)	156.6	(6.2)	157.3	(6.1)	156.0	(6.1)	.514
Weight (kg)	59.2	(11.5)	64.2	(19.4)	68.5	(17.8)	.025[B]
Occipitofrontal circumference (OFC) (cm)	53.7	(1.6)	54.2	(2.1)	54.6	(1.8)	.071
Upper arm circumference (cm)	25.3	(3.5)	26.1	(4.8)	27.4	(4.7)	.055
Body mass index (BMI)	24.2	(5.0)	25.9	(7.2)	28.0	(6.6)	.013[B]
Age	28.0	(6.1)	26.2	(6.0)	28.5	(7.4)	.100
Years of education	8.4	(2.3)	9.9	(2.2)	10.1	(2.1)	<.001[A,B]
Legally married (% Yes)	14.9		15.8		32.7		.032
Gravidity	3.4	(1.6)	3.0	(1.5)	1.5	(1.1)	.274
Parity	2.0	(1.5)	1.6	(1.4)	.4	(.8)	.196
Miscarriages	.3	(.7)	.2	(.6)	.4	(.8)	.453
Week of pregnancy when interviewed	19.4	(6.3)	19.3	(8.2)	18.9	(7.6)	.936
Use tobacco (% Yes)	93.6		78.9		40.0		<.001
Hungry in pregnancy because lack of food (% Yes)	27.7		17.1		14.5		.206
# of drinks per week – before pregnancy	14.3	(10.9)	12.2	(13.1)	1.1	(4.0)	<.001[B,C]
# of drinking days per week – before pregnancy	2.2	(1.0)	1.6	(1.0)	.2	(.5)	<.001[A,B,C]
Estimated # of drinks per drinking day (DDD) – before pregnancy	6.1	(3.2)	6.6	(4.3)	.8	(2.3)	<.001[B,C]
Total AUDIT score	17.8	(6.3)	14.5	(6.9)	2.8	(6.1)	<.001[A,B,C]

Post-hoc Dunnet C comparisons significantly different between: [A.] Group 1 & Group 2; [B.] Group 1 & Group 3; [C.]Group 2 & Group 3

Table 5.2 Maternal Dietary Intake During Pregnancy: Weighted[1] Average

	Drank during pregnancy with alcohol in recalls (*n*=47)		Drank during pregnancy without alcohol in recalls (*n*=76)		Did not drink during pregnancy (*n*=55)		
	Mean	(SD)	Mean	(SD)	Mean	(SD)	p
Total Grams	2342.7	(795.4)	2192.4	(827.4)	2259.0	(803.2)	.607
Energy (kcal)	1638.7	(638.7)	1609.6	(623.5)	1428.4	(559.2)	.148
Total Fat (g)	49.6	(31.9)	52.3	(26.8)	43.5	(24.1)	.196
Total Carbohydrate (g)	220.2	(94.2)	228.6	(90.1)	213.3	(83.9)	.620
Total Protein (g)	52.8	(26.0)	58.2	(27.0)	48.3	(24.7)	.102
Animal Protein (g)	31.4	(20.8)	33.5	(22.3)	26.6	(19.2)	.179
Vegetable Protein (g)	21.4	(10.0)	24.7	(11.0)	21.7	(9.4)	.132
Alcohol (g)	15.8	(16.1)	.0	(.0)	.0	(.0)	<.001[A,B]
Cholesterol (mg)	188.2	(131.4)	170.6	(129.1)	175.3	(191.7)	.820
Total Saturated Fatty Acids (g)	13.6	(9.5)	14.4	(7.4)	12.2	(6.6)	.290
Total Monounsaturated Fatty Acids (g)	17.3	(11.4)	17.9	(9.3)	14.6	(8.6)	.145
Total Polyunsaturated Fatty Acids (g)	14.7	(10.7)	15.7	(10.4)	13.1	(8.9)	.369
Total Dietary Fiber (g)	12.3	(6.8)	14.5	(8.7)	13.1	(6.3)	.275
Total Vitamin A (RAE) (mcg)	261.8	(209.1)	312.1	(250.1)	434.1	(999.1)	.304
Vitamin D (calciferol) (mcg)	2.9	(3.5)	3.5	(3.5)	3.4	(3.2)	.639
Vitamin E (Total a-Tocopherol) (mg)	5.5	(4.6)	5.5	(4.0)	5.1	(3.5)	.794
Vitamin K (phylloquinone) (mcg)	40.3	(42.1)	46.2	(34.3)	43.0	(35.5)	.674
Vitamin C (ascorbic acid) (mg)	65.1	(66.4)	79.8	(79.2)	86.5	(89.2)	.386
Thiamin (vitamin B_1) (mg)	1.4	(.7)	1.6	(.7)	1.6	(.8)	.253
Riboflavin (vitamin B_2) (mg)	1.2	(.5)	1.2	(.5)	1.3	(.9)	.378
Niacin (vitamin B_3) (mg)	19.5	(7.9)	20.2	(8.6)	19.5	(10.7)	.888
Pantothenic Acid (mg)	3.6	(1.6)	3.7	(1.7)	4.1	(3.2)	.435
Vitamin B_6 (mg)	.9	(.4)	.8	(.4)	.7	(.5)	.160
Dietary Folate Equivalents (mcg)	2084.3	(1212.7)	2283.6	(1032.2)	2148.0	(1067.8)	.584
Vitamin B_{12} (cobalamin) (mcg)	3.2	(4.6)	4.1	(5.3)	3.3	(5.6)	.541

Calcium (mg)	563.0	(283.4)	611.5	(248.9)	633.6	(258.0)	.385
Phosphorus (mg)	686.1	(305.2)	735.3	(328.7)	674.5	(301.4)	.501
Magnesium (mg)	189.8	(71.2)	189.9	(84.4)	177.3	(78.3)	.620
Iron (mg)	46.5	(19.9)	54.1	(28.5)	47.6	(35.6)	.278
Zinc (mg)	6.4	(3.7)	6.7	(3.1)	6.3	(2.9)	.695
Copper (mg)	1.9	(.8)	2.1	(.9)	1.9	(1.2)	.508
Manganese (mg)	4.7	(2.0)	5.2	(2.3)	4.8	(2.8)	.478
Selenium (mcg)	85.6	(41.9)	99.6	(47.9)	84.5	(45.6)	.112
Sodium (mg)	2518.8	(1243.1)	2849.3	(1323.3)	2310.7	(1023.8)	.041
Potassium (mg)	1702.7	(808.3)	1741.9	(929.3)	1670.9	(866.4)	.900
Choline (mg)	252.5	(131.0)	220.4	(112.3)	209.5	(148.8)	.224
Betaine (mg)	166.0	(79.5)	172.9	(81.7)	152.5	(78.4)	.355
Linoleic acid (g)	13.2	(9.7)	13.8	(9.4)	11.7	(7.9)	.440
Alpha-linolenic acid (g)	1.0	(.8)	1.2	(.9)	.9	(.7)	.219
Eicosapentaenoic acid (EPA) (g)	.1	(.1)	.1	(.2)	.1	(.1)	.189
Docosapentaenoic acid (DPA) (g)	.0	(.0)	.0	(.1)	.0	(.0)	.141
Docosahexaenoic acid (DHA) (g)	.1	(.2)	.2	(.4)	.1	(.2)	.188

Post-hoc Dunnet C comparisons significantly different between: [A.] Group 1 & Group 2; [B.] Group 1 & Group 3; [C.]Group 2 & Group 3

1. Weekday recall values were weighted by .714 (5/7) and weekend recall values were weighted by .286 (2/7).

Table 5.3 Percent of Women Less Than Estimated Average Requirement (EAR) – Weighted Mean

	% Less than EAR				
	EAR	Drank during pregnancy with alcohol in recalls (*n*=47)	Drank during pregnancy without alcohol in recalls (*n*=76)	Did not drink during pregnancy (*n*=55)	p
Vitamin A (RAE) (mcg)[1]	550	87.2	88.2	87.3	.984
Vitamin D (mcg)	10	97.9	94.7	92.7	.494
Vitamin E (mg)	12	89.4	92.1	96.4	.386
Vitamin C (mg)	70	66.0	60.5	50.9	.286
Thiamin (mg)	1.2	42.6	23.7	30.9	.089
Riboflavin (mg)	1.2	57.4	57.9	58.2	.997
Niacin Equivalents (mg)[2]	14	29.8	22.4	30.9	.487
Vitamin B_6 (mg)	1.6	96.6	94.7	92.7	.893
Dietary Folate Equivalents (mcg)	520	23.4	17.1	20.0	.692
Vitamin B_{12} (mcg)	2.2	51.1	42.1	52.7	.422
Calcium (mg)	800	78.7	77.6	80.0	.948
Phosphorus (mg)	580	42.6	36.8	43.6	.695
Magnesium (mcg)	290	91.5	89.5	92.7	.806
Iron (mg)	22	12.8	15.8	18.2	.755
Zinc (mg)	9.5	85.1	84.2	85.5	.979
Copper (mg)	0.8	12.8	9.2	14.5	.628
Selenium (mcg)	49	23.4	11.8	20.0	.216
# of micronutrients below EAR	--	9.9 (3.6)	9.2 (3.0)	9.7 (3.5)	.511

Estimated Average Requirement (EAR) for pregnant women, aged 19–30, used for: vitamin A, C, D, E, thiamin, riboflavin, niacin, vitamin B_6, folate, vitamin B_{12}, calcium, phosphorus, magnesium, iron, zinc, and selenium.

[1] Retinol Activity Equivalents

[2] Niacin Equivalents (1 niacin equivalent = 1 mg of Niacin or 60 mg of tryptophan).

Table 5.4 Percent of Women Less Than Recommended Dietary Allowance (RDA) – Weighted Mean

		% Less than RDA or AI			
	RDA/AI	Drank during pregnancy with alcohol in recalls (*n*=47)	Drank during pregnancy without alcohol in recalls (*n*=76)	Did not drink during pregnancy (*n*=55)	p
Vitamin A (RAE) (mcg)[1]	770	97.9	96.1	92.7	.437
Vitamin D (mcg)	15	97.9	98.7	100.0	.584
Vitamin E (mg)	15	93.6	97.4	98.2	.398
Vitamin C (mg)	85	68.1	64.5	58.2	.569
Vitamin K (mcg)	90^	93.5	90.7	85.5	.372
Thiamin (mg)	1.4	57.4	36.8	41.8	.077
Riboflavin (mg)	1.4	70.2	75.0	63.6	.373
Niacin Equivalent (mg) [2]	18	48.9	43.4	54.5	.451
Pantothenic acid (mg)	6^	89.4	88.2	87.3	.948
Vitamin B_6 (mg)	1.9	97.9	97.4	94.5	.582
Dietary Folate Equivalents (mcg)	600	23.4	17.1	20.0	.692
Vitamin B_{12} (mcg)	2.6	63.8	47.4	87.3	.948
Calcium (mg)	1000	91.5	92.1	94.5	.811
Phosphorus (mg)	700	57.4	51.3	54.5	.798
Magnesium (mcg)	350	97.9	96.1	98.2	.727
Iron (mg)	27	14.9	15.8	21.8	.580
Zinc (mg)	11	91.5	88.2	92.7	.653
Copper (mcg)	1000	12.8	13.2	18.2	.664
Manganese (mg)	2.0^	8.5	7.9	16.4	.257
Selenium (mcg)	60	39.5	23.8	25.5	.433
Sodium (g)	1.5^	21.3	10.5	18.2	.235
Potassium (mg)	4700^	100.0	97.4	100.0	.257
Choline (mcg)	450^	91.5	98.7	92.7	.139

Linoleic Acid (g)	13^	61.7	60.5	72.7	.315
Alpha-Linolenic Acid (g)	1.4^	78.7	75.0	81.8	.643
# of micronutrients below RDA/AI	--	16.6 (4.3)	15.8 (3.7)	16.5 (4.4)	.491

Recommended Dietary Allowance (RDA) for pregnant women, aged 19–30, used for: vitamin A, C, D, E, thiamin, riboflavin, niacin, vitamin B_6, folate, vitamin B_{12}, calcium, phosphorus, magnesium, iron, zinc, and selenium.
Adequate Intake for pregnant women, aged 19-30, used for: pantothenic acid, manganese, sodium, potassium, choline, linoleic acid, and alpha-linolenic acid.
^ denotes Adequate Intake
[1] Retinol Activity Equivalents
[2] Niacin Equivalents (1 niacin equivalent = 1 mg of Niacin or 60 mg of tryptophan).

Table 5.5 Infant Characteristics at Birth and 6 Weeks by Alcohol Exposure

	Drink during pregnancy with alcohol in recalls		Drink during pregnancy without alcohol in recalls		Did not drink during pregnancy		
	Mean	(SD)	Mean	(SD)	Mean	(SD)	p
Birth	(*n*=43)		(*n*=73)		(*n*=50)		
Sex (% Male)	42.6		44.7		52.7		.537
Length Centile	31.0	(29.9)	35.9	(31.9)	53.7	(34.1)	.001[B,C]
Weight Centile	15.5	(16.2)	24.9	(24.0)	32.4	(25.1)	.002[A,B]
OFC Centile	24.2	(24.9)	26.2	(28.1)	34.3	(29.7)	.164
Weight-for-Length	55.3	(8.6)	58.2	(8.6)	59.9	(10.1)	.040[B]
Weight-for-Length Centile	21.7	(28.8)	27.2	(32.0)	22.3	(28.8)	.593
Body Mass Index	11.6	(1.6)	12.1	(2.0)	12.2	(2.9)	.369
Ponderal Index	2.4	(.4)	2.5	(.7)	2.5	(.9)	.779
Gestational Age	38.1	(2.2)	38.3	(1.9)	38.4	(2.3)	.868
6 Weeks	(*n*=47)		(*n*=76)		(*n*=55)		
Age (days)	49.6	(18.0)	49.7	(13.1)	52.3	(17.9)	.606
Length Centile	17.9	(25.6)	25.5	(27.2)	38.2	(29.0)	.001[B,C]
Weight Centile	33.4	(28.6)	38.2	(31.3)	49.3	(28.4)	.020 [B]
OFC Centile	24.5	(22.8)	27.7	(22.9)	40.3	(25.9)	.002 [B,C]
Weight-for-Length	80.9	(10.0)	83.4	(11.3)	86.4	(8.6)	.025[B]
Body Mass Index	15.6	(1.7)	15.6	(1.9)	15.8	(1.5)	.783
Ponderal Index	3.0	(.4)	2.9	(.4)	2.9	(.3)	.239
ICD Centile	40.0	(18.8)	49.2	(21.3)	47.5	(20.4)	.048[A]
IPD Centile	32.1	(24.8)	36.1	(28.0)	41.9	(28.3)	.188
PFL Centile	56.1	(33.0)	66.8	(29.0)	72.5	(24.5)	.017[B]
Philtrum Ranking	3.5	(.7)	3.3	(.6)	3.1	(.8)	.025[B]

Vermilion Ranking	3.3	(.6)	3.2	(.6)	3.1	(.7)	.106
Number of Minor Anomalies	7.2	(2.8)	5.9	(3.2)	5.0	(2.9)	.001[A,B]
Total Dysmorphology Score	10.0	(4.9)	7.7	(4.9)	6.5	(4.0)	.001[A,B]

ICD: Inner canthal distance; IPD: Inner pupillary distance; OFC: occipitofrontal circumference; PFL: palpebral fissure length

Post-hoc Dunnet C comparisons significantly different between: [A.] Group 1 & Group 2; [B.] Group 1 & Group 3; [C.]Group 2 & Group 3

Table 5.6 Zero-Order and Partial Correlations of Alcohol with Weighted Average Macro- and Micro-Nutrients

	Weighted Average of Grams of Alcohol			
	Zero order		Partial[1]	
	r	p	r	p
Total Grams	.133	.077	.148	.053
Energy (kcal)	-.052	.488	--	--
Total Fat (g)	-.142	.059	-.128	.093
Total Carbohydrate (g)	-.162	.031	-.268	<.001
Total Protein (g)	-151	.044	-.227	.003
Animal Protein (g)	-.113	.132	-.134	.081
Vegetable Protein (g)	-.154	.040	-.164	.031
Cholesterol (mg)	.014	.854	.002	.975
Total Saturated Fatty Acids (g)	-.163	.030	-.143	.062
Total Monounsaturated Fatty Acids (g)	-.133	.077	-.111	.148
Total Polyunsaturated Fatty Acids (g)	-.099	.190	-.045	.554
Total Dietary Fiber (g)	-.193	.010	-.225	.003
Total Vitamin A (RAE) (mcg)	-.067	.371	-.006	.935
Vitamin D (calciferol) (mcg)	-.122	.103	-.117	.127
Vitamin E (Total a-Tocopherol) (mg)	-.082	.276	-.057	.455
Vitamin K (phylloquinone) (mcg)	-.209	.005	-.201	.008
Vitamin C (ascorbic acid) (mg)	-.138	.066	-.094	.222
Thiamin (vitamin B_1) (mg)	-.197	.008	-.217	.004
Riboflavin (vitamin B_2) (mg)	-.099	.189	-.074	.336
Niacin (vitamin B_3) (mg)	-.092	.222	-.113	.139
Pantothenic Acid (mg)	-.104	.166	-.086	.264
Vitamin B_6 (mg)	.113	.133	.187	.014
Dietary Folate Equivalents (mcg)	-.117	.119	-.122	.111
Vitamin B_{12} (cobalamin) (mcg)	-.106	.159	-.105	.171
Calcium (mg)	-.180	.016	-.122	.112
Phosphorus (mg)	-.133	.078	-.179	.019
Magnesium (mg)	-.041	.583	-.031	.684
Iron (mg)	-.119	.113	-.045	.561
Zinc (mg)	-.187	.013	-.223	.003
Copper (mg)	-.115	.127	-.043	.574
Manganese (mg)	-.136	.070	-.051	.506
Selenium (mcg)	-.167	.026	-.205	.007
Sodium (mg)	-.155	.039	-.208	.006
Potassium (mg)	-.091	.227	-.102	.183
Choline (mg)	.091	.225	.111	.148
Betaine (mg)	-.030	.687	.014	.885
Linoleic acid (g)	-.085	.258	-.020	.795

Alpha-linolenic acid (g)	-.161	.031	-.122	.111
Eicosapentaenoic acid (EPA) (g)	-.073	.333	-.072	.346
Docosapentaenoic acid (DPA) (g)	-.082	.276	-.095	.216
Docosahexaenoic acid (DHA) (g)	-.076	.317	-.095	.214
# of micronutrient less than RDA	.173	.021	.235	.002

1. Adjusted for maternal BMI, age, gravidity, tobacco use, education, and total energy

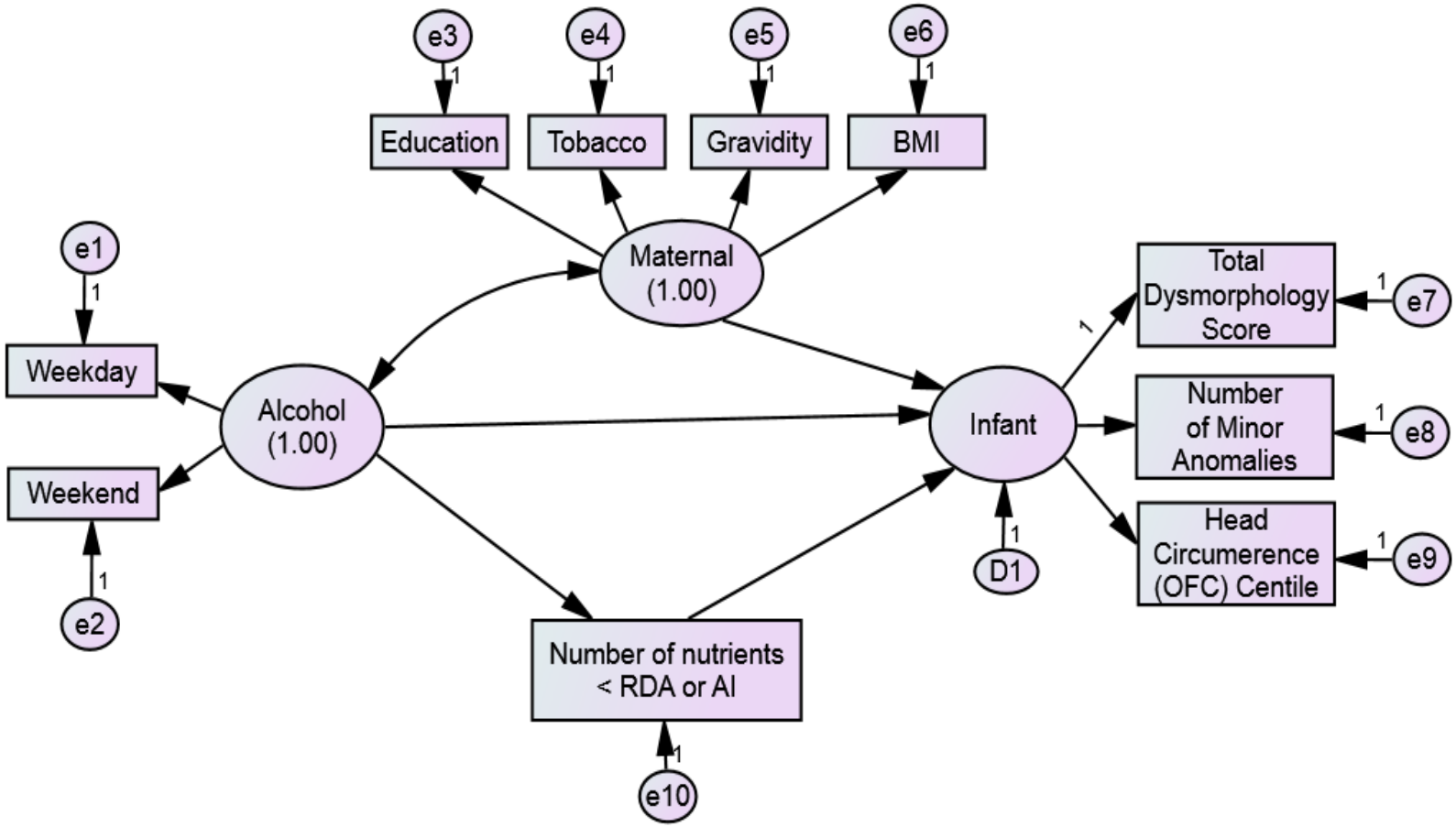

Figure 5.1 Hypothesized Association Between Alcohol, Maternal Dietary Intake, and Infant Physical Outcomes

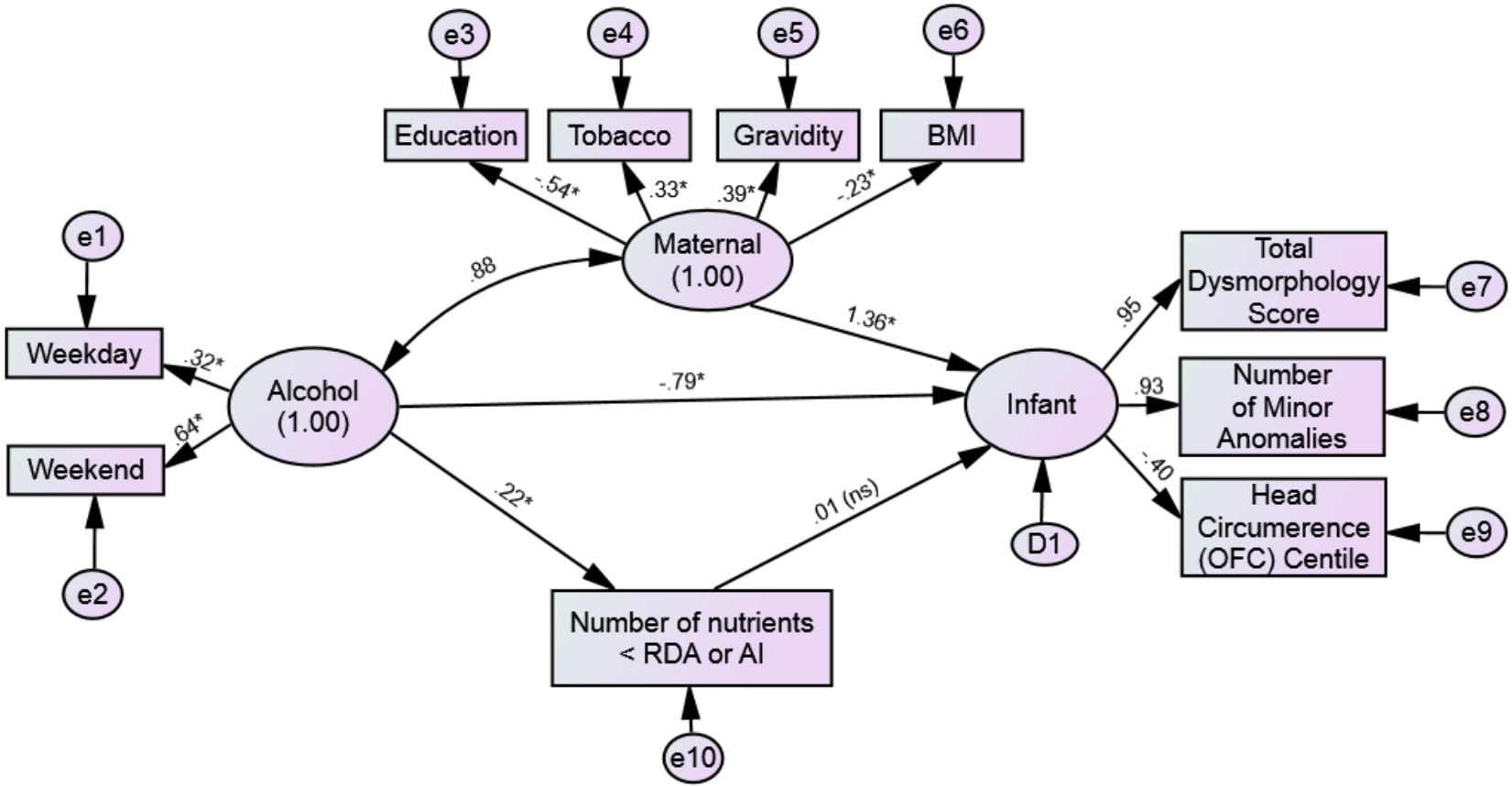

Figure 5.2 Standardized Coefficients: Alcohol, Maternal Dietary Intake, and Infant Physical Outcomes
*p<.05; ns: Not Significant

CHAPTER 6: SYNTHESIS

While FASD is completely preventable if a mother does not consume alcohol during pregnancy, many pregnancies are unplanned and alcohol exposure frequently occurs before the woman is aware that she is pregnant.[263] A number of social, cultural, and behavioral factors influence a woman's decision to drink before and/or after pregnancy recognition which results in thousands of fetuses exposed to alcohol prenatally.[23,238–240,264,265] In the United States, 53.6% of women of childbearing age reported consuming alcohol in the past 30 days and 10% of pregnant women reported alcohol consumption.[240] Thus the question remains regarding how to mitigate the adverse effects of prenatal alcohol exposure. Because of the individual variation in the manifestations of prenatal alcohol exposure, diagnosing children with FASD in early childhood remains difficult. This research has attempted to explore maternal weight and nutrition as factors which may mitigate some of the adverse effects of prenatal alcohol exposure in the early infancy period.

Alcohol Consumption During Pregnancy Remains a Concern in the Western Cape Province

Approximately a quarter (25%) of women in these studies reported alcohol consumption during pregnancy. This percent mirrors the estimated prevalence of FASD in the Western Cape Province of 17-28%.[2–9] Consistent with previous cross-sectional[2–9] and prospective longitudinal cohorts[99] in the Western Cape, we found women who reported alcohol consumption during pregnancy, reported consuming high quantities of alcohol, mostly on two days each week.

For some women, consuming alcohol with friends is one of the few forms of recreation available in these communities; therefore, drinking on Friday and Saturday nights is a valued and substantial social and economic investment for many.[13,15,19–23] Because most alcohol consumption occurs in a structured pattern in small groups, and scarce funds must be allocated to purchase alcohol, most respondents can recall their alcohol use accurately.

In these communities, qualitative studies have indicated that many women believe alcohol consumption during pregnancy is not harmful to the fetus despite receiving alcohol abstinence messaging while pregnant.[21] Women continue to drink while pregnant due to: having an unplanned pregnancy, utilizing alcohol as a coping mechanism to deal with stress and abuse/trauma, needing the socialization that accompanies weekend drinking, and feelings of personal invincibility.[236] Therefore, while some women will quit or reduce drinking while pregnant, others will continue to drink throughout their pregnancy. This has been shown in South Africa[6–8,15] and in at least three recent American samples.[69–71] The percent of women who report drinking decreases in each trimester of pregnancy, but those who continue to drink may continue to consume alcohol in high quantities.

Unlike other populations, where there may be concurrent use of alcohol and other drugs, alcohol remains the primary substance of use. Few women (2-3%) in these communities reported any drug use during pregnancy with marijuana (‘dagga’) or methamphetamine (‘tik’) being the drug of choice.[7,8] This results in a population where the teratogenic exposure is solely alcohol. This allows for inquiries to explore the etiology and developmental trajectories of FASD that would be difficult, to impossible, in other populations where alcohol consumption and drug use may occur concurrently. However, alcohol consumption during pregnancy remains a concern in these communities.

Alcohol Exposure Influences Infant Growth and Development

The studies presented here demonstrated that increased alcohol consumption was associated with poorer physical growth and more dysmorphology. While threshold analyses were not undertaken here, increasing drinks per drinking day (DDD) was associated with poorer physical outcomes: smaller length, weight, head circumference (OFC), and facial measurements. To our knowledge, this was only the second study which demonstrated that at 6 weeks of age, the total dysmorphology score can differentiate among children with and without prenatal alcohol exposure. There is also some evidence in these studies that the effect of prenatal alcohol exposure may become more pronounced as the child ages.

While alcohol in the previous 30 days of pregnancy was not associated with poorer neurodevelopmental outcomes, an association may become evident as the cognitive integration and processing required becomes more complex. Others have shown differences between alcohol-exposed and unexposed infants in the first year of life with other developmental measures (e.g., visual acuity and eye blink conditioning);[54,94,95] therefore, it is possible that assessing other aspects of development may have demonstrated an association in this cohort. However, there is not a single neurocognitive profile for children with FASD.[218,232,266] It is also possible that as the children age, the developmental assessment tools become more sensitive to specific behavior delays/deficits and an association may become apparent. Previous longitudinal studies in these communities have demonstrated that Bayley scores did not differentiate children with FASD and children with typical development until 3-4 years of age.[99] Previously we have shown that dysmorphology, relative to neurocognitive outcomes, correlated more strongly with alcohol exposure.[193,231,266] Yet, consistent with previous public health recommendations, these studies affirm that there is no known safe level of alcohol exposure during pregnancy.[42,46]

Maternal Weight Influences Infant Growth and Development

The link between maternal obesity and infant birth length and weight has been well established in previous research.[267] Severe obesity can lead to negative consequences for the fetus/infant.[268] Yet in alcohol-exposed pregnancies, maternal weight has been found to be positively associated with better outcomes, and it is therefore likely a protective factor. Infants in our study communities were smaller than average when compared to WHO growth standards. Because most children were smaller than average in these communities, increased maternal weight was associated with improved (larger) infant physical growth outcomes (length, weight, and OFC centile) as well as other eye measurements (ICD, IPD, and PFL centile). OFC centile reflects the brain volume and is widely considered a proxy for neural growth and overall brain development.[250,269] Because the eyes are created from outgrowths of the brain, the eyes also provide an indication of overall brain development.

The effect of maternal weight on infant growth was observable when first measured in this study (either at birth or six weeks of age, depending on the specific measurement). But there was no difference in the growth trajectory through 9 months of age for nearly all physical measurements. The only exception was that maternal weight significantly influenced the rate of change across time for the infant's weight-for-length. Infants of heavier mothers gained more weight (relative to their length) compared to their peers born to lighter mothers. Additional follow-up studies will be necessary to determine if this additional infant weight-for-length observed in infants born to heavier mothers will be beneficial, detrimental, or indifferent in the child's overall health and physical development. Rapid growth in early life has been shown to be associated with childhood obesity.[124,125] Yet in this study population where stunting remains a public health concern, there may be a beneficial effect of being born to heavier mothers.

The diagnostic criteria for fetal alcohol syndrome (FAS), the most severe end of the FASD continuum, requires length/height and/or weight and OFC to be $\leq 10^{th}$ centile. The increase in growth measurements observed in infants born to heavier mothers may result in children being diagnosed on the less severe end of the FASD continuum or potentially developing within the normal range, even if exposed to alcohol prenatally.

In these communities, higher maternal weight was also associated with better infant performance on cognitive and motor domains. Yet the developmental growth rate was similar among all infants regardless of maternal weight. This suggests that infants were born at a given performance ability, but there was no acceleration or lag in development associated with maternal weight from birth to 9 months. However, a downward trajectory of neurocognitive abilities was demonstrated in this entire cohort. Continued neurodevelopmental assessments are needed.

With the same quantity of alcohol consumed over the same duration of time, women with greater body weight will achieve a lower blood alcohol concentration than women with lower body weight. In alcohol-exposed pregnancies, additional maternal weight may lower the concentration of alcohol that crosses the placenta and therefore, reduce the alcohol exposure to the fetus. The reduction in alcohol crossing the placenta may lead to less alcohol-induced oxidative stress, cell apoptosis, disruption in cell migration, differentiation and adhesion, and/or alteration in insulinlike growth factors function on cell proliferation and survival.[270] There is no single mechanism of alcohol-induced damaged that explains all of the physical and neurodevelopmental characteristics observed in children with prenatal alcohol exposure.[270–272] Yet even at a specific fetal developmental stage, different mechanisms of cell death exist and respond differently to varying levels of alcohol exposure.[270] Higher maternal weight may reduce

the fetal blood alcohol concentration and reduce the susceptibility and severity of alcohol-induced damage on physical and neurocognitive (brain) development. Higher maternal weight, possibly through lowering the fetal blood alcohol concentration, can lead to larger, less dysmorphic infants and to better average initial neurodevelopment which was demonstrated in this study.

Initial Growth and Development Parameters and Later Growth and Performance

In these studies, maternal weight and alcohol exposure independently influenced infant birth parameters and 6-week neurodevelopmental performance. Maternal weight led to an increase in initial measures of growth and development while alcohol consumption led to a decrease in initial measurements. For nearly all growth parameters, the effect of maternal weight and alcohol was present at first assessment. Birth measures were a strong predictor of future growth, and, on average, infants continued to develop within their percentile channel across time through 9 months.

Although maternal weight was associated with increased initial performance on two domains of neurodevelopmental performance at 6 weeks, the postnatal environment and lack of infant stimulation resulted in nearly all infants following a downward trajectory through 9 months. Similar downward trajectories have been previously documented in this population.[99] Initial performance on early life neurodevelopmental assessments is likely not predictive of future neurodevelopmental abilities in this population. There is a generalized high risk for developmental delays in this population, whether alcohol exposed or not.

Maternal Dietary Intake during Pregnancy Remains a Concern in the Western Cape

Despite many national policies (e.g., food fortification) designed to improve the health and nutritional status of women and children in South Africa, women in our sample were deficient on several key micronutrients. While alcohol consumption was negatively associated

with dietary intake, all women (alcohol consuming and non-consuming) in this sample were deficient. This is placing all infants, whether alcohol exposed or not, at risk for poor growth and future adverse health consequences. The thrifty phenotype hypothesis asserts that a poor fetal nutrient environment can program fetal metabolism to adapt to the nutrient environment with the expectation of a similarly nutrient inadequate environment postnatally.[174,175] These alterations in the fetus have been shown to predispose individuals exposed to nutrient-poor environments in utero to a wide range of diseases in adulthood.[176,177] These metabolic changes occur independent of prenatal alcohol exposure, but these metabolic changes may be further complicated when undernutrition and alcohol exposure occur concurrently.[178] Additional studies will need to examine this possibility.

While some micronutrients are highly metabolically regulated to ensure that necessary cellular processes are maintained, other micronutrients (e.g., water soluble vitamins) are not highly regulated and require daily consumption to meet the Institute of Medicine's dietary recommendations. Although no significant indirect effect of alcohol via micronutrient intake was demonstrated in this study, because of nutrient metabolic regulation and the known alcohol-induced malabsorption of micronutrients, dietary intake may not be a sufficient proxy for certain micronutrient concentrations in the blood. Additional studies utilizing maternal plasma nutrient concentrations may better elicit whether there is an indirect effect of alcohol via maternal nutrition. However, our study clearly demonstrates that alcohol has a direct and negative effect on infant outcomes, even if not mediated through maternal dietary intake.

Applicability to Other Populations

The Western Cape Province of South Africa is somewhat unique in terms of the historical drinking patterns, cultural norms, nutrition, and socioeconomic conditions. Malnutrition and challenging socioeconomic conditions are present for a large portion of these community

members. This may make the conclusions drawn here less applicable to other populations where less challenging nutritional and socioeconomic conditions are present. The women in the studies were representative of the general population in these communities; a substantially large proportion of the population consumed alcohol during pregnancy and those who continued to consume alcohol later in pregnancy continued to drink in high quantities: a mean of 6 drinks per drinking day, 1-2 times per week. These communities also have a significant proportion of women who are both underweight and micronutrient deficient, and there are also women who are overweight/obese and micronutrient deficient. Both alcohol consumption and nutritional challenges may have contributed to the high prevalence of FASD in the Western Cape yet the findings from this work can lead to translatable implications for public health practice in these communities and possibly elsewhere.

Implications for Diagnosis and Public Health

While there is substantial variation in the manifestation of the effects of prenatal alcohol exposure, alcohol is a known teratogen and can result in adverse consequences in fetal development. The Western Cape Province has the highest documented prevalence of FASD in the world,[5–8] yet FASD and the effects of prenatal alcohol exposure on fetal growth and development are not unique to South Africa. The United States has an estimated FASD prevalence of 1-5%,[1] and globally, thousands of infants are exposed to alcohol prenatally.[273,274]

Children with FASD go unrecognized and undiagnosed due to a variety of reasons including: the lack of reliable alcohol consumption history during pregnancy, the timing of onset of neurocognitive delays, and few physicians and interdisciplinary teams that are willing or capable of recognizing or diagnosing FASD.[10,11] The American Academy of Pediatrics found only 50% of their members felt prepared to make a diagnosis within the FASD continuum[275] likely due, in part, to the perceived stigma towards addressing alcohol consumption by pregnant

women and the lack of general awareness about considering prenatal alcohol exposure in the differential diagnosis.[276] Yet most physicians believed that having an FASD diagnosis would be beneficial for the child.[275] Additional efforts to train medical providers in early identification and referrals of infants with suspected prenatal alcohol exposure and/or poor nutrient environments remains essential to prevent and/or remediate FASD.[277] This becomes especially important when the mother is underweight herself.

A population-based study in 4 regions of the United States reported that of the 222 identified children with FASD in 1st grade, only two children had previously received an FASD diagnosis from a physician.[1] In a population of foster and adoptive children in the United States, another study found 80.1% of children who met criteria for a diagnosis within the FASD continuum were never diagnosed by an average age of eight years old and 6.4% were misdiagnosed.[11] This results in many infants and children who are not being adequately identified as "at risk" and intervention services are not considered and/or implemented. Because the first 1000 days are a unique developmental period where life course trajectories are being established, it is during this period when benefits of interventions are most powerful. If prenatal alcohol exposure is known or is suspected, maternal weight following delivery or maternal malnutrition may be used as one of many tools that clinicians can use to help identify that an infant may be at higher risk for postnatal onset growth deficiencies and a possible FASD diagnosis. Combined with other early life indicators such as prematurity and/or small-for-gestational age, physicians and early interventionists may be able to use a constellation of indicators to determine whether an infant should be followed more closely to apply or to rule out a possible FASD diagnosis.

While there is a proportion of the population that continues to consume alcohol during pregnancy, there are microenvironments that exist within these communities which support abstinence from alcohol in general and especially during pregnancy. Due to the historical norms that are partially in place due to the 'Dop' system, many of the participants in these studies live on the farms for which they work. Over time, each farm developed cultural norms and expectations with regards to alcohol consumption. Certain farm owners have made, and continue to make, efforts to uplift the working conditions and lives of their workers.[17] These efforts have included providing opportunities for adult training in technical skills, providing or co-signing for financial assistance for their workers, and providing opportunities (creche/pre-schools) for the children of workers on the farm itself. Farm owners who prioritize investing in their workers often understand the adverse effects of prenatal alcohol exposure and do not condone drinking, especially excessive drinking on their farm or any alcohol consumption during pregnancy. In addition to the social support and norms that may be present in a specific microenvironment of a particular farm, many women who previously consumed alcohol report complete abstinence upon converting to a formal religion. Many churches and formal religious organizations in South Africa understand the adverse effects of prenatal alcohol exposure and have active programs to prevent prenatal alcohol exposure. Therefore, while alcohol consumption during pregnancy remains a concern, there are some positive social determinants of health in these communities which help reduce alcohol consumption during pregnancy. Public health interventions which amplify alcohol abstaining social norms and improve living conditions in general may help reduce the prevalence of FASD in these communities. Moreover, in these communities, the generalized high risk for smaller than average infants and developmental delays suggest that

family-level and community-level improvements in nutrition, food quality, living conditions, and overall socioeconomic status may lead to better outcomes for all children in these communities.

Limitations

There were limitations to these studies, and the results should be interpreted with appropriate caution. First, these studies were carried out in South Africa which may have different cultural, socioeconomic, and nutritional environments; therefore, these results may be less applicable to other populations or contexts. Second, while these studies focused on early life, because this period is not well characterized among children with prenatal alcohol exposure, these analyses did not continue to follow the infants beyond 9 months. It remains unknown if the effects observed in these studies will be maintained into childhood. Third, because these were prospective cohorts recruited in the prenatal clinics, information about the postnatal environment was not ascertained in these studies. While the social and environmental factors were similar among all participants in these studies, the influence of postnatal environmental factors could not be included in these analyses. Fourth, all maternal demographic, alcohol consumption, and dietary intake information was self-reported. Previous studies with pregnant women in these communities have indicated that women are accurate reporters of alcohol consumption.[234] However, some recall bias may be present. Fifth, because maternal height was not measured in Aim 1 and Aim 2, BMI could not be calculated. Without both height and weight measurements, maternal blood alcohol concentration could not be estimated. Much of the underlying rationale for this inquiry was based on higher maternal weight may lead to lower maternal and fetal blood alcohol concentration, and therefore, less alcohol-induced damage, but this could not be directly assessed in this study. Sixth, while the physical and neurobehavioral measures were assessed using standardized techniques, measurement error cannot be ruled out. Finally, as with all studies, the power of these analyses was dependent on sample size. The associations identified in

these studies were assumed to be true associations. The lack of association does not negate the possibility of a true, underlying association which was not observable due to the limited statistical power attributed to small sample sizes in these studies.

Significance

This work helped to clarify how maternal weight and nutrition influence infant outcomes among alcohol-exposed pregnancies. This work adds to a growing literature which is attempting to identify early life indicators, both maternal risk factors and infant characteristics, to drive down the age at which a diagnosis on the FASD continuum can be made accurately. Through early identification, appropriate and early intervention services can be implemented for children who meet criteria for FASD and for children at risk for developmental delays associated with prenatal alcohol exposure. It should be noted that all children with FASD are unique in their physical and neurocognitive and behavioral presentation, abilities, and strengths. The adverse outcomes of prenatal alcohol exposure manifest in each affected child in similar, yet unique ways. Therefore, when determining if intervention or close monitoring of a child is warranted, the possibility of alcohol exposure, maternal weight, and maternal dietary intake should all be included among the many factors considered in the child evaluation.

Future Directions

Further research which explicitly examines maternal weight and/or BMI and dietary intake on the physical and neurocognitive trajectories of children with prenatal alcohol exposure will determine if the differentiation seen at 9 months observed in these cohorts persists into later life. More specifically, assessing maternal dietary status through known, valid biomarkers of circulating nutrient concentration and adequacy of nutrient stores will be critical to understanding the maternal nutritional contributions to an FASD diagnosis. Future studies are warranted to determine if metabolic changes observed in children exposed to prenatal alcohol

exposure differ based on maternal metabolic profiles (overweight/obese vs not) and nutritional status (malnourished vs not). While this work attempted to better explain the individual variation in infant outcomes associated with prenatal alcohol exposure and maternal weight and nutrition, there remains unexplained individual variation in outcomes. Studies of the maternal and paternal genetic, epigenetic, and nutrigenetic influences on FASD are also warranted.

Future studies are needed which focus on the development and implementation of theory-informed interventions to optimize pre-pregnancy maternal weight and maternal nutrition during pregnancy. This is especially poignant for women who consume moderate to heavy amounts of alcohol. This population had both women who were underweight and had several micronutrient deficiencies as well as women who were overweight and had several micronutrient deficiencies. Future studies which examine whether maternal micronutrient supplementation during the prenatal and/or postnatal/breastfeeding period are needed to determine whether improved maternal dietary intake may result in better infant development.

Finally, with the ultimate goal of preventing all FASD, secondary prevention interventions which focus on alcohol cessation during pregnancy are warranted for this population. Tertiary interventions such as one-on-one case management should be continued, for they have been shown to reduce alcohol consumption and improve child outcomes in these communities.[278,279] Moreover, interventions which promote infant-caregiver bonding and provide additional infant stimulation may enrich the lives of all infants, whether alcohol exposed or not.[210] Improving overall general development of all infants and children in these communities is a warranted primary or universal prevention goal for this population.

APPENDIX A: CHAPTER 3 STEPWISE REGRESSION

Table A.1 Stepwise Regression Coefficients Predicting Length Centile at 9 Months

		Unstandardized		Standardized			95% CI for B	
		B	Std. Error	Beta Coeff.	t	Sig.	Lower	Upper
1	(Constant)	34.679	1.698		20.421	.000	31.340	38.019
	DDD – 30 days prior	-16.024	4.309	-.193	-3.719	.000	-24.499	-7.550
2	(Constant)	12.550	28.219		.445	.657	-42.948	68.048
	DDD – 30 days prior	-11.599	4.452	-.139	-2.606	.010	-20.354	-2.845
	Tobacco Use (Yes)	-5.930	3.082	-.101	-1.924	.055	-11.992	.131
	Gravidity (log)	-41.283	13.859	-.209	-2.979	.003	-68.539	-14.027
	Maternal Age (log)	26.864	22.234	.083	1.208	.228	-16.863	70.592
	Trimester Interviewed	3.469	2.414	.075	1.437	.152	-1.278	8.216
3	(Constant)	-110.168	61.635		-1.787	.075	-231.386	11.050
	DDD – 30 days prior	-11.618	4.427	-.140	-2.625	.009	-20.324	-2.912
	Tobacco Use (Yes)	-5.782	3.066	-.099	-1.886	.060	-11.812	.247
	Gravidity (log)	-41.707	13.783	-.211	-3.026	.003	-68.813	-14.600
	Maternal Age (log)	27.354	22.111	.085	1.237	.217	-16.131	70.840
	Trimester Interviewed	3.253	2.402	.071	1.354	.177	-1.472	7.977
	Gestational Age at Birth	3.131	1.400	.113	2.236	.026	.377	5.885
4	(Constant)	-110.737	61.892		-1.789	.074	-232.461	10.987
	DDD – 30 days prior	-11.636	4.435	-.140	-2.624	.009	-20.359	-2.913
	Tobacco Use (Yes)	-5.778	3.070	-.099	-1.882	.061	-11.816	.260
	Gravidity (log)	-41.772	13.812	-.211	-3.024	.003	-68.937	-14.608
	Maternal Age (log)	27.555	22.201	.086	1.241	.215	-16.108	71.218
	Trimester Interviewed	3.231	2.412	.070	1.340	.181	-1.512	7.974
	Gestational Age at Birth	3.145	1.407	.113	2.236	.026	.378	5.912
	Sex of Infant (male)	-.369	2.984	-.006	-.124	.902	-6.237	5.498
5	(Constant)	-107.711	61.166		-1.761	.079	-228.006	12.585
	DDD – 30 days prior	-10.349	4.402	-.124	-2.351	.019	-19.007	-1.691
	Tobacco Use (Yes)	-5.612	3.034	-.096	-1.850	.065	-11.580	.355
	Gravidity (log)	-40.407	13.655	-.204	-2.959	.003	-67.263	-13.550
	Maternal Age (log)	18.817	22.120	.058	.851	.396	-24.686	62.321
	Trimester Interviewed	3.682	2.388	.080	1.542	.124	-1.014	8.378
	Gestational Age at Birth	2.823	1.394	.102	2.025	.044	.082	5.565
	Sex of Infant (male)	-.309	2.948	-.005	-.105	.916	-6.108	5.489
	Maternal Weight (kg)	.309	.100	.157	3.087	.002	.112	.506
6	(Constant)	-94.605	60.432		-1.565	.118	-213.459	24.249
	DDD – 30 days prior	-9.814	4.343	-.118	-2.260	.024	-18.357	-1.272
	Tobacco Use (Yes)	-5.582	2.991	-.095	-1.866	.063	-11.466	.301
	Gravidity (log)	-40.516	13.463	-.205	-3.009	.003	-66.995	-14.038
	Maternal Age (log)	13.283	21.871	.041	.607	.544	-29.732	56.297
	Trimester Interviewed	3.144	2.360	.068	1.332	.184	-1.497	7.784
	Gestational Age at Birth	2.423	1.380	.087	1.756	.080	-.291	5.136
	Sex of Infant (male)	-.151	2.907	-.003	-.052	.958	-5.869	5.566
	Maternal Weight (kg)	.344	.099	.175	3.468	.001	.149	.539
	Day Weight Assessed	1.994	.598	.167	3.336	.001	.818	3.169
7	(Constant)	-93.695	60.635		-1.545	.123	-212.950	25.560
	DDD – 30 days prior	-5.617	18.251	-.068	-.308	.758	-41.512	30.278

	Tobacco Use (Yes)	-5.623	3.000	-.096	-1.874	.062	-11.524	.278
	Gravidity (log)	-40.538	13.482	-.205	-3.007	.003	-67.053	-14.023
	Maternal Age (log)	12.943	21.947	.040	.590	.556	-30.222	56.108
	Trimester Interviewed	3.052	2.394	.066	1.275	.203	-1.657	7.761
	Gestational Age at Birth	2.402	1.384	.087	1.735	.084	-.320	5.125
	Sex of Infant (male)	-.219	2.925	-.004	-.075	.940	-5.971	5.534
	Maternal Weight (kg)	.354	.109	.180	3.261	.001	.141	.568
	Day Weight Assessed	1.993	.598	.167	3.330	.001	.816	3.170
	DDD*Maternal Weight	-.068	.286	-.052	-.237	.813	-.630	.495

Step 1: DDD; Step 2: Tobacco use during pregnancy, gravidity, maternal age; trimester of pregnancy when maternal interview occurred; Step 3: Gestational age at birth; Step 4: Sex of Infant Step 5: Maternal weight; Step 6: Number of days postpartum maternal weight was assessed; Step 7: DDD by Maternal weight interaction

Table A.2 Stepwise Regression Coefficients Predicting Weight Centile at 9 Months

		Unstandardized		Standardized			95% CI for B	
		B	Std. Error	Beta Coeff.	t	Sig.	Lower	Upper
1	(Constant)	33.036	1.756		18.816	.000	29.583	36.488
	DDD – 30 days prior	-13.774	4.455	-.161	-3.092	.002	-22.536	-5.012
2	(Constant)	8.993	29.620		.304	.762	-49.260	67.245
	DDD – 30 days prior	-14.241	4.673	-.166	-3.048	.002	-23.430	-5.052
	Tobacco Use (Yes)	-.324	3.235	-.005	-.100	.920	-6.687	6.038
	Gravidity (log)	-35.859	14.547	-.176	-2.465	.014	-64.468	-7.250
	Maternal Age (log)	37.372	23.338	.113	1.601	.110	-8.526	83.270
	Trimester Interviewed	-3.472	2.533	-.074	-1.371	.171	-8.455	1.510
3	(Constant)	-91.807	64.872		-1.415	.158	-219.390	35.776
	DDD – 30 days prior	-14.256	4.659	-.167	-3.060	.002	-23.419	-5.093
	Tobacco Use (Yes)	-.203	3.227	-.003	-.063	.950	-6.549	6.143
	Gravidity (log)	-36.207	14.507	-.178	-2.496	.013	-64.737	-7.677
	Maternal Age (log)	37.774	23.272	.114	1.623	.105	-7.994	83.543
	Trimester Interviewed	-3.650	2.528	-.077	-1.444	.150	-8.622	1.323
	Gestational Age at Birth	2.572	1.474	.090	1.745	.082	-.326	5.470
4	(Constant)	-93.040	65.137		-1.428	.154	-221.146	35.067
	DDD – 30 days prior	-14.296	4.668	-.167	-3.063	.002	-23.476	-5.116
	Tobacco Use (Yes)	-.194	3.231	-.003	-.060	.952	-6.549	6.160
	Gravidity (log)	-36.349	14.536	-.179	-2.501	.013	-64.938	-7.760
	Maternal Age (log)	38.210	23.365	.115	1.635	.103	-7.743	84.162
	Trimester Interviewed	-3.697	2.538	-.078	-1.456	.146	-8.689	1.295
	Gestational Age at Birth	2.603	1.481	.091	1.758	.080	-.309	5.515
	Sex of Infant (male)	-.800	3.140	-.013	-.255	.799	-6.976	5.375
5	(Constant)	-87.167	62.247		-1.400	.162	-209.589	35.256
	DDD – 30 days prior	-11.798	4.480	-.138	-2.633	.009	-20.610	-2.987
	Tobacco Use (Yes)	.128	3.088	.002	.041	.967	-5.945	6.201
	Gravidity (log)	-33.699	13.897	-.166	-2.425	.016	-61.031	-6.368
	Maternal Age (log)	21.255	22.511	.064	.944	.346	-23.017	65.527
	Trimester Interviewed	-2.822	2.430	-.060	-1.161	.246	-7.601	1.957
	Gestational Age at Birth	1.978	1.419	.069	1.394	.164	-.812	4.768
	Sex of Infant (male)	-.684	3.000	-.011	-.228	.820	-6.584	5.217
	Maternal Weight (kg)	.599	.102	.297	5.886	.000	.399	.799
6	(Constant)	-82.230	62.336		-1.319	.188	-204.829	40.369
	DDD – 30 days prior	-11.597	4.480	-.136	-2.588	.010	-20.408	-2.785
	Tobacco Use (Yes)	.139	3.086	.002	.045	.964	-5.929	6.208
	Gravidity (log)	-33.741	13.887	-.166	-2.430	.016	-61.054	-6.428
	Maternal Age (log)	19.170	22.560	.058	.850	.396	-25.200	63.540
	Trimester Interviewed	-3.025	2.434	-.064	-1.243	.215	-7.812	1.762
	Gestational Age at Birth	1.827	1.423	.064	1.284	.200	-.972	4.626
	Sex of Infant (male)	-.624	2.999	-.010	-.208	.835	-6.522	5.273
	Maternal Weight (kg)	.612	.102	.303	5.986	.000	.411	.814
	Day Weight Assessed	.751	.617	.061	1.218	.224	-.461	1.964
7	(Constant)	-78.992	62.491		-1.264	.207	-201.897	43.914
	DDD – 30 days prior	3.339	18.810	.039	.177	.859	-33.655	40.333
	Tobacco Use (Yes)	-.006	3.092	.000	-.002	.999	-6.087	6.076
	Gravidity (log)	-33.820	13.894	-.166	-2.434	.015	-61.146	-6.493
	Maternal Age (log)	17.962	22.619	.054	.794	.428	-26.525	62.448

	Trimester Interviewed	-3.351	2.468	-.071	-1.358	.175	-8.205	1.502
	Gestational Age at Birth	1.755	1.427	.062	1.230	.219	-1.051	4.561
	Sex of Infant (male)	-.864	3.014	-.014	-.287	.774	-6.793	5.064
	Maternal Weight (kg)	.650	.112	.322	5.799	.000	.429	.870
	Day Weight Assessed	.748	.617	.061	1.213	.226	-.465	1.961
	DDD*Maternal Weight	-.241	.295	-.180	-.818	.414	-.820	.339

Step 1: DDD; Step 2: Tobacco use during pregnancy, gravidity, maternal age; trimester of pregnancy when maternal interview occurred; Step 3: Gestational age at birth; Step 4: Sex of Infant Step 5: Maternal weight; Step 6: Number of days postpartum maternal weight was assessed; Step 7: DDD by Maternal weight interaction

Table A.3 Stepwise Regression Coefficients Predicting OFC Centile at 9 Months

		Unstandardized		Standardized			95% CI for B	
		B	Std. Error	Beta Coeff.	t	Sig.	Lower	Upper
1	(Constant)	40.980	1.887		21.718	.000	37.269	44.691
	DDD – 30 days prior	-4.622	4.795	-.051	-.964	.336	-14.051	4.808
2	(Constant)	51.764	31.958		1.620	.106	-11.086	114.614
	DDD – 30 days prior	-4.120	5.039	-.045	-.818	.414	-14.030	5.790
	Tobacco Use (Yes)	-2.422	3.484	-.038	-.695	.487	-9.274	4.429
	Gravidity (log)	-28.202	15.693	-.130	-1.797	.073	-59.065	2.660
	Maternal Age (log)	8.633	25.180	.024	.343	.732	-40.887	58.154
	Trimester Interviewed	-2.397	2.733	-.048	-.877	.381	-7.771	2.978
3	(Constant)	-29.598	69.633		-.425	.671	-166.544	107.348
	DDD – 30 days prior	-4.104	5.034	-.045	-.815	.415	-14.005	5.796
	Tobacco Use (Yes)	-2.364	3.481	-.037	-.679	.498	-9.209	4.482
	Gravidity (log)	-28.427	15.678	-.131	-1.813	.071	-59.260	2.406
	Maternal Age (log)	8.996	25.156	.026	.358	.721	-40.478	58.469
	Trimester Interviewed	-2.551	2.733	-.051	-.934	.351	-7.925	2.823
	Gestational Age at Birth	2.074	1.578	.069	1.315	.189	-1.029	5.178
4	(Constant)	-37.913	69.581		-.545	.586	-174.756	98.930
	DDD – 30 days prior	-4.385	5.021	-.048	-.873	.383	-14.260	5.490
	Tobacco Use (Yes)	-2.324	3.470	-.036	-.670	.504	-9.149	4.501
	Gravidity (log)	-29.455	15.641	-.136	-1.883	.060	-60.216	1.306
	Maternal Age (log)	12.272	25.148	.035	.488	.626	-37.185	61.730
	Trimester Interviewed	-2.908	2.732	-.058	-1.064	.288	-8.280	2.465
	Gestational Age at Birth	2.284	1.577	.076	1.448	.149	-.819	5.386
	Sex of Infant (male)	-5.993	3.373	-.094	-1.777	.076	-12.626	.641
5	(Constant)	-34.716	68.538		-.507	.613	-169.510	100.078
	DDD – 30 days prior	-2.772	4.968	-.030	-.558	.577	-12.542	6.998
	Tobacco Use (Yes)	-2.098	3.418	-.033	-.614	.540	-8.821	4.625
	Gravidity (log)	-27.758	15.413	-.128	-1.801	.073	-58.071	2.555
	Maternal Age (log)	1.239	24.974	.004	.050	.960	-47.877	50.356
	Trimester Interviewed	-2.336	2.696	-.046	-.867	.387	-7.638	2.965
	Gestational Age at Birth	1.894	1.558	.063	1.216	.225	-1.170	4.957
	Sex of Infant (male)	-5.932	3.322	-.093	-1.786	.075	-12.466	.602
	Maternal Weight (kg)	.390	.113	.181	3.452	.001	.168	.612
6	(Constant)	-29.614	68.573		-.432	.666	-164.479	105.251
	DDD – 30 days prior	-2.547	4.965	-.028	-.513	.608	-12.312	7.218
	Tobacco Use (Yes)	-2.062	3.415	-.032	-.604	.546	-8.779	4.654
	Gravidity (log)	-27.844	15.397	-.129	-1.808	.071	-58.125	2.437
	Maternal Age (log)	-1.265	25.019	-.004	-.051	.960	-50.471	47.940
	Trimester Interviewed	-2.577	2.699	-.051	-.955	.340	-7.885	2.731
	Gestational Age at Birth	1.734	1.561	.058	1.111	.267	-1.335	4.804
	Sex of Infant (male)	-5.881	3.319	-.092	-1.772	.077	-12.408	.646
	Maternal Weight (kg)	.405	.113	.189	3.575	.000	.182	.628
	Day Weight Assessed	.903	.682	.069	1.324	.186	-.438	2.244
7	(Constant)	-34.444	68.676		-.502	.616	-169.512	100.624
	DDD – 30 days prior	-25.570	20.835	-.281	-1.227	.221	-66.547	15.408
	Tobacco Use (Yes)	-1.843	3.419	-.029	-.539	.590	-8.567	4.882
	Gravidity (log)	-27.716	15.391	-.128	-1.801	.073	-57.985	2.554
	Maternal Age (log)	.595	25.062	.002	.024	.981	-48.696	49.885

	Trimester Interviewed	-2.074	2.734	-.041	-.759	.449	-7.451	3.303
	Gestational Age at Birth	1.841	1.563	.061	1.178	.240	-1.233	4.915
	Sex of Infant (male)	-5.507	3.334	-.086	-1.652	.099	-12.063	1.050
	Maternal Weight (kg)	.348	.124	.162	2.807	.005	.104	.592
	Day Weight Assessed	.908	.682	.070	1.332	.184	-.432	2.249
	DDD*Maternal Weight	.371	.326	.260	1.138	.256	-.271	1.013

Step 1: DDD; Step 2: Tobacco use during pregnancy, gravidity, maternal age; trimester of pregnancy when maternal interview occurred; Step 3: Gestational age at birth; Step 4: Sex of Infant Step 5: Maternal weight; Step 6: Number of days postpartum maternal weight was assessed; Step 7: DDD by Maternal weight interaction

Table A.4 Stepwise Regression Coefficients Predicting Weight-for-Length Centile at 9 Months

		Unstandardized		Standardized			95% CI for B	
		B	Std. Error	Beta Coeff.	t	Sig.	Lower	Upper
1	(Constant)	55.354	1.962		28.211	.000	51.495	59.213
	DDD – 30 days prior	-2.907	4.979	-.031	-.584	.560	-12.699	6.885
2	(Constant)	62.055	33.010		1.880	.061	-2.865	126.974
	DDD – 30 days prior	-7.167	5.207	-.076	-1.376	.170	-17.408	3.074
	Tobacco Use (Yes)	5.220	3.605	.079	1.448	.149	-1.871	12.310
	Gravidity (log)	-21.095	16.212	-.094	-1.301	.194	-52.978	10.788
	Maternal Age (log)	15.988	26.009	.044	.615	.539	-35.162	67.138
	Trimester Interviewed	-7.543	2.823	-.145	-2.671	.008	-13.095	-1.990
3	(Constant)	15.281	72.552		.211	.833	-127.407	157.969
	DDD – 30 days prior	-7.174	5.211	-.076	-1.377	.169	-17.422	3.073
	Tobacco Use (Yes)	5.276	3.609	.079	1.462	.145	-1.821	12.373
	Gravidity (log)	-21.257	16.224	-.095	-1.310	.191	-53.164	10.651
	Maternal Age (log)	16.175	26.027	.044	.621	.535	-35.013	67.362
	Trimester Interviewed	-7.625	2.828	-.146	-2.697	.007	-13.186	-2.064
	Gestational Age at Birth	1.193	1.648	.038	.724	.469	-2.048	4.435
4	(Constant)	15.469	72.856		.212	.832	-127.817	158.756
	DDD – 30 days prior	-7.168	5.221	-.076	-1.373	.171	-17.437	3.100
	Tobacco Use (Yes)	5.275	3.614	.079	1.460	.145	-1.833	12.382
	Gravidity (log)	-21.235	16.259	-.095	-1.306	.192	-53.212	10.742
	Maternal Age (log)	16.108	26.134	.044	.616	.538	-35.290	67.506
	Trimester Interviewed	-7.618	2.839	-.146	-2.683	.008	-13.202	-2.034
	Gestational Age at Birth	1.189	1.656	.038	.718	.473	-2.069	4.446
	Sex of Infant (male)	.122	3.512	.002	.035	.972	-6.785	7.029
5	(Constant)	20.630	70.922		.291	.771	-118.854	160.114
	DDD – 30 days prior	-4.973	5.105	-.053	-.974	.331	-15.013	5.066
	Tobacco Use (Yes)	5.558	3.518	.084	1.580	.115	-1.361	12.477
	Gravidity (log)	-18.906	15.834	-.084	-1.194	.233	-50.046	12.234
	Maternal Age (log)	1.209	25.648	.003	.047	.962	-49.233	51.651
	Trimester Interviewed	-6.849	2.769	-.131	-2.474	.014	-12.294	-1.404
	Gestational Age at Birth	.639	1.617	.020	.396	.693	-2.540	3.819
	Sex of Infant (male)	.225	3.418	.003	.066	.948	-6.499	6.948
	Maternal Weight (kg)	.527	.116	.236	4.540	.000	.298	.755
6	(Constant)	16.396	71.088		.231	.818	-123.416	156.209
	DDD – 30 days prior	-5.146	5.109	-.055	-1.007	.315	-15.195	4.903
	Tobacco Use (Yes)	5.548	3.519	.084	1.577	.116	-1.373	12.469
	Gravidity (log)	-18.871	15.837	-.084	-1.192	.234	-50.019	12.277
	Maternal Age (log)	2.997	25.728	.008	.116	.907	-47.603	53.597
	Trimester Interviewed	-6.675	2.776	-.128	-2.405	.017	-12.134	-1.216
	Gestational Age at Birth	.769	1.623	.024	.474	.636	-2.423	3.961
	Sex of Infant (male)	.173	3.420	.003	.051	.960	-6.552	6.899
	Maternal Weight (kg)	.515	.117	.231	4.416	.000	.286	.745
	Day Weight Assessed	-.644	.703	-.048	-.916	.360	-2.027	.739
7	(Constant)	19.477	71.286		.273	.785	-120.726	159.679
	DDD – 30 days prior	9.059	21.457	.096	.422	.673	-33.141	51.260
	Tobacco Use (Yes)	5.410	3.527	.082	1.534	.126	-1.527	12.348
	Gravidity (log)	-18.946	15.850	-.084	-1.195	.233	-50.119	12.227
	Maternal Age (log)	1.847	25.803	.005	.072	.943	-48.900	52.595

	Trimester Interviewed	-6.986	2.815	-.134	-2.482	.014	-12.523	-1.450
	Gestational Age at Birth	.701	1.627	.022	.431	.667	-2.500	3.901
	Sex of Infant (male)	-.055	3.439	-.001	-.016	.987	-6.818	6.708
	Maternal Weight (kg)	.551	.128	.247	4.309	.000	.299	.802
	Day Weight Assessed	-.647	.704	-.048	-.919	.359	-2.031	.737
	DDD*Maternal Weight	-.229	.336	-.155	-.682	.496	-.890	.432

Step 1: DDD; Step 2: Tobacco use during pregnancy, gravidity, maternal age; trimester of pregnancy when maternal interview occurred; Step 3: Gestational age at birth; Step 4: Sex of Infant Step 5: Maternal weight; Step 6: Number of days postpartum maternal weight was assessed; Step 7: DDD by Maternal weight interaction

Table A.5 Stepwise Regression Coefficients Predicting ICD Centile at 9 Months

		Unstandardized		Standardized			95% CI for B	
		B	Std. Error	Beta Coeff.	t	Sig.	Lower	Upper
1	(Constant)	44.786	1.631		27.466	.000	41.579	47.992
	DDD – 30 days prior	-3.074	4.138	-.039	-.743	.458	-11.211	5.064
2	(Constant)	35.964	27.526		1.307	.192	-18.171	90.099
	DDD – 30 days prior	-5.390	4.342	-.069	-1.241	.215	-13.930	3.149
	Tobacco Use (Yes)	-.610	3.006	-.011	-.203	.839	-6.523	5.303
	Gravidity (log)	5.638	13.519	.030	.417	.677	-20.948	32.225
	Maternal Age (log)	14.080	21.688	.046	.649	.517	-28.573	56.733
	Trimester Interviewed	-5.257	2.354	-.121	-2.233	.026	-9.887	-.627
3	(Constant)	210.648	59.640		3.532	.000	93.355	327.942
	DDD – 30 days prior	-5.364	4.283	-.068	-1.252	.211	-13.788	3.060
	Tobacco Use (Yes)	-.821	2.966	-.015	-.277	.782	-6.655	5.013
	Gravidity (log)	6.241	13.337	.033	.468	.640	-19.988	32.470
	Maternal Age (log)	13.383	21.395	.044	.625	.532	-28.695	55.460
	Trimester Interviewed	-4.949	2.324	-.114	-2.129	.034	-9.521	-.378
	Gestational Age at Birth	-4.457	1.355	-.171	-3.290	.001	-7.122	-1.792
4	(Constant)	220.921	59.435		3.717	.000	104.030	337.813
	DDD – 30 days prior	-5.032	4.259	-.064	-1.181	.238	-13.409	3.345
	Tobacco Use (Yes)	-.893	2.948	-.016	-.303	.762	-6.691	4.906
	Gravidity (log)	7.426	13.264	.040	.560	.576	-18.660	33.512
	Maternal Age (log)	9.756	21.320	.032	.458	.648	-32.174	51.686
	Trimester Interviewed	-4.558	2.316	-.105	-1.968	.050	-9.113	-.003
	Gestational Age at Birth	-4.717	1.351	-.180	-3.491	.001	-7.374	-2.060
	Sex of Infant (male)	6.670	2.865	.121	2.328	.020	1.036	12.305
5	(Constant)	223.165	59.058		3.779	.000	107.015	339.315
	DDD – 30 days prior	-4.078	4.251	-.052	-.959	.338	-12.438	4.282
	Tobacco Use (Yes)	-.770	2.930	-.014	-.263	.793	-6.531	4.992
	Gravidity (log)	8.438	13.185	.045	.640	.523	-17.493	34.370
	Maternal Age (log)	3.279	21.357	.011	.154	.878	-38.725	45.283
	Trimester Interviewed	-4.223	2.305	-.097	-1.832	.068	-8.758	.311
	Gestational Age at Birth	-4.956	1.346	-.190	-3.681	.000	-7.603	-2.308
	Sex of Infant (male)	6.715	2.847	.122	2.359	.019	1.117	12.313
	Maternal Weight (kg)	.229	.097	.124	2.370	.018	.039	.419
6	(Constant)	218.541	59.145		3.695	.000	102.217	334.865
	DDD – 30 days prior	-4.267	4.251	-.054	-1.004	.316	-12.627	4.094
	Tobacco Use (Yes)	-.780	2.928	-.014	-.267	.790	-6.538	4.978
	Gravidity (log)	8.477	13.177	.045	.643	.520	-17.438	34.392
	Maternal Age (log)	5.232	21.405	.017	.244	.807	-36.867	47.331
	Trimester Interviewed	-4.034	2.309	-.093	-1.747	.082	-8.575	.508
	Gestational Age at Birth	-4.814	1.350	-.184	-3.565	.000	-7.470	-2.158
	Sex of Infant (male)	6.659	2.845	.121	2.341	.020	1.064	12.255
	Maternal Weight (kg)	.217	.097	.117	2.231	.026	.026	.407
	Day Weight Assessed	-.704	.585	-.062	-1.203	.230	-1.854	.447
7	(Constant)	218.438	59.349		3.681	.000	101.712	335.164
	DDD – 30 days prior	-4.739	17.864	-.060	-.265	.791	-39.873	30.395
	Tobacco Use (Yes)	-.776	2.937	-.014	-.264	.792	-6.552	5.000
	Gravidity (log)	8.480	13.196	.045	.643	.521	-17.473	34.432
	Maternal Age (log)	5.270	21.482	.017	.245	.806	-36.980	47.520

	Trimester Interviewed	-4.023	2.344	-.093	-1.717	.087	-8.633	.586
	Gestational Age at Birth	-4.812	1.355	-.184	-3.551	.000	-7.477	-2.147
	Sex of Infant (male)	6.667	2.863	.121	2.329	.020	1.036	12.297
	Maternal Weight (kg)	.215	.106	.116	2.024	.044	.006	.425
	Day Weight Assessed	-.704	.586	-.062	-1.201	.231	-1.856	.449
	DDD*Maternal Weight	.008	.280	.006	.027	.978	-.543	.558

Step 1: DDD; Step 2: Tobacco use during pregnancy, gravidity, maternal age; trimester of pregnancy when maternal interview occurred; Step 3: Gestational age at birth; Step 4: Sex of Infant Step 5: Maternal weight; Step 6: Number of days postpartum maternal weight was assessed; Step 7: DDD by Maternal weight interaction

Table A.6 Stepwise Regression Coefficients Predicting IPD Centile at 9 Months

		Unstandardized		Standardized			95% CI for B	
		B	Std. Error	Beta Coeff.	t	Sig.	Lower	Upper
1	(Constant)	23.792	1.348		17.654	.000	21.142	26.442
	DDD – 30 days prior	2.930	3.415	.045	.858	.391	-3.786	9.647
2	(Constant)	37.568	22.783		1.649	.100	-7.240	82.375
	DDD – 30 days prior	1.451	3.596	.022	.404	.687	-5.620	8.523
	Tobacco Use (Yes)	-1.400	2.493	-.031	-.561	.575	-6.303	3.504
	Gravidity (log)	7.582	11.194	.049	.677	.499	-14.433	29.597
	Maternal Age (log)	-4.793	17.951	-.019	-.267	.790	-40.097	30.512
	Trimester Interviewed	-3.917	1.949	-.110	-2.009	.045	-7.750	-.083
3	(Constant)	53.015	50.139		1.057	.291	-45.593	151.624
	DDD – 30 days prior	1.455	3.600	.022	.404	.686	-5.625	8.536
	Tobacco Use (Yes)	-1.420	2.497	-.031	-.569	.570	-6.332	3.491
	Gravidity (log)	7.640	11.209	.050	.682	.496	-14.405	29.685
	Maternal Age (log)	-4.856	17.975	-.019	-.270	.787	-40.207	30.495
	Trimester Interviewed	-3.890	1.953	-.109	-1.992	.047	-7.731	-.049
	Gestational Age at Birth	-.394	1.139	-.018	-.346	.730	-2.634	1.846
4	(Constant)	57.957	50.211		1.154	.249	-40.794	156.707
	DDD – 30 days prior	1.611	3.598	.025	.448	.655	-5.464	8.687
	Tobacco Use (Yes)	-1.445	2.494	-.032	-.579	.563	-6.351	3.461
	Gravidity (log)	8.202	11.203	.053	.732	.465	-13.832	30.236
	Maternal Age (log)	-6.638	18.001	-.027	-.369	.713	-42.041	28.765
	Trimester Interviewed	-3.694	1.956	-.103	-1.888	.060	-7.541	.153
	Gestational Age at Birth	-.519	1.141	-.024	-.455	.650	-2.764	1.726
	Sex of Infant (male)	3.292	2.422	.072	1.359	.175	-1.472	8.056
5	(Constant)	60.126	49.834		1.207	.228	-37.885	158.138
	DDD – 30 days prior	2.482	3.587	.038	.692	.489	-4.572	9.536
	Tobacco Use (Yes)	-1.347	2.476	-.030	-.544	.587	-6.216	3.522
	Gravidity (log)	9.143	11.124	.059	.822	.412	-12.735	31.021
	Maternal Age (log)	-12.494	18.012	-.050	-.694	.488	-47.919	22.931
	Trimester Interviewed	-3.396	1.945	-.095	-1.746	.082	-7.221	.429
	Gestational Age at Birth	-.738	1.136	-.034	-.650	.516	-2.973	1.496
	Sex of Infant (male)	3.322	2.404	.073	1.382	.168	-1.406	8.049
	Maternal Weight (kg)	.207	.081	.135	2.538	.012	.047	.367
6	(Constant)	57.621	49.956		1.153	.250	-40.631	155.874
	DDD – 30 days prior	2.383	3.591	.037	.664	.507	-4.679	9.445
	Tobacco Use (Yes)	-1.364	2.477	-.030	-.551	.582	-6.236	3.508
	Gravidity (log)	9.187	11.130	.060	.825	.410	-12.702	31.077
	Maternal Age (log)	-11.395	18.072	-.046	-.631	.529	-46.940	24.149
	Trimester Interviewed	-3.291	1.950	-.092	-1.688	.092	-7.127	.544
	Gestational Age at Birth	-.661	1.141	-.031	-.580	.563	-2.905	1.582
	Sex of Infant (male)	3.282	2.405	.072	1.364	.173	-1.449	8.013
	Maternal Weight (kg)	.200	.082	.131	2.440	.015	.039	.361
	Day Weight Assessed	-.399	.496	-.043	-.805	.422	-1.374	.576
7	(Constant)	58.157	50.130		1.160	.247	-40.437	156.752
	DDD – 30 days prior	4.822	15.092	.075	.319	.750	-24.861	34.505
	Tobacco Use (Yes)	-1.388	2.485	-.030	-.559	.577	-6.275	3.499
	Gravidity (log)	9.176	11.145	.060	.823	.411	-12.744	31.097
	Maternal Age (log)	-11.592	18.136	-.046	-.639	.523	-47.263	24.078

	Trimester Interviewed	-3.345	1.979	-.094	-1.690	.092	-7.237	.548
	Gestational Age at Birth	-.673	1.144	-.031	-.588	.557	-2.924	1.578
	Sex of Infant (male)	3.242	2.421	.071	1.339	.181	-1.519	8.003
	Maternal Weight (kg)	.206	.090	.135	2.293	.022	.029	.383
	Day Weight Assessed	-.400	.496	-.043	-.805	.421	-1.376	.577
	DDD*Maternal Weight	-.039	.236	-.039	-.166	.868	-.504	.425

Step 1: DDD; Step 2: Tobacco use during pregnancy, gravidity, maternal age; trimester of pregnancy when maternal interview occurred; Step 3: Gestational age at birth; Step 4: Sex of Infant Step 5: Maternal weight; Step 6: Number of days postpartum maternal weight was assessed; Step 7: DDD by Maternal weight interaction

Table A.7 Stepwise Regression Coefficients Predicting PFL Centile at 9 Months

		Unstandardized		Standardized			95% CI for B	
		B	Std. Error	Beta Coeff.	t	Sig.	Lower	Upper
1	(Constant)	53.934	1.839		29.328	.000	50.317	57.550
	DDD – 30 days prior	-8.681	4.667	-.098	-1.860	.064	-17.858	.497
2	(Constant)	72.993	31.294		2.332	.020	11.448	134.538
	DDD – 30 days prior	-9.921	4.937	-.112	-2.010	.045	-19.629	-.212
	Tobacco Use (Yes)	-.732	3.418	-.012	-.214	.830	-7.455	5.990
	Gravidity (log)	3.001	15.369	.014	.195	.845	-27.225	33.227
	Maternal Age (log)	-8.228	24.657	-.024	-.334	.739	-56.720	40.264
	Trimester Interviewed	-3.226	2.677	-.066	-1.205	.229	-8.490	2.038
3	(Constant)	-120.658	67.855		-1.778	.076	-254.108	12.791
	DDD – 30 days prior	-9.950	4.873	-.112	-2.042	.042	-19.534	-.366
	Tobacco Use (Yes)	-.499	3.375	-.008	-.148	.883	-7.137	6.139
	Gravidity (log)	2.333	15.174	.011	.154	.878	-27.509	32.175
	Maternal Age (log)	-7.455	24.342	-.022	-.306	.760	-55.328	40.419
	Trimester Interviewed	-3.567	2.645	-.073	-1.349	.178	-8.768	1.634
	Gestational Age at Birth	4.941	1.541	.167	3.205	.001	1.909	7.972
4	(Constant)	-112.845	67.908		-1.662	.097	-246.401	20.711
	DDD – 30 days prior	-9.698	4.866	-.109	-1.993	.047	-19.269	-.127
	Tobacco Use (Yes)	-.554	3.369	-.009	-.164	.870	-7.179	6.071
	Gravidity (log)	3.234	15.155	.015	.213	.831	-26.571	33.039
	Maternal Age (log)	-10.213	24.359	-.030	-.419	.675	-58.120	37.695
	Trimester Interviewed	-3.270	2.646	-.067	-1.236	.217	-8.474	1.935
	Gestational Age at Birth	4.743	1.544	.160	3.073	.002	1.707	7.779
	Sex of Infant (male)	5.073	3.274	.081	1.550	.122	-1.365	11.511
5	(Constant)	-110.879	67.698		-1.638	.102	-244.023	22.265
	DDD – 30 days prior	-8.862	4.873	-.100	-1.819	.070	-18.445	.722
	Tobacco Use (Yes)	-.446	3.358	-.007	-.133	.894	-7.051	6.159
	Gravidity (log)	4.121	15.114	.020	.273	.785	-25.604	33.846
	Maternal Age (log)	-15.889	24.482	-.046	-.649	.517	-64.039	32.260
	Trimester Interviewed	-2.977	2.643	-.061	-1.126	.261	-8.174	2.221
	Gestational Age at Birth	4.534	1.543	.153	2.938	.004	1.499	7.569
	Sex of Infant (male)	5.112	3.263	.082	1.567	.118	-1.305	11.530
	Maternal Weight (kg)	.201	.111	.096	1.812	.071	-.017	.418
6	(Constant)	-108.753	67.916		-1.601	.110	-242.326	24.821
	DDD – 30 days prior	-8.775	4.881	-.099	-1.798	.073	-18.375	.826
	Tobacco Use (Yes)	-.441	3.362	-.007	-.131	.896	-7.053	6.171
	Gravidity (log)	4.104	15.131	.019	.271	.786	-25.654	33.862
	Maternal Age (log)	-16.787	24.580	-.049	-.683	.495	-65.129	31.555
	Trimester Interviewed	-3.064	2.652	-.062	-1.155	.249	-8.279	2.152
	Gestational Age at Birth	4.469	1.551	.151	2.882	.004	1.419	7.519
	Sex of Infant (male)	5.138	3.267	.082	1.573	.117	-1.288	11.563
	Maternal Weight (kg)	.206	.111	.098	1.851	.065	-.013	.426
	Day Weight Assessed	.324	.672	.025	.482	.630	-.998	1.645
7	(Constant)	-110.713	68.130		-1.625	.105	-244.709	23.283
	DDD – 30 days prior	-17.816	20.507	-.201	-.869	.386	-58.148	22.516
	Tobacco Use (Yes)	-.353	3.371	-.006	-.105	.917	-6.984	6.277
	Gravidity (log)	4.151	15.148	.020	.274	.784	-25.641	33.944
	Maternal Age (log)	-16.056	24.660	-.047	-.651	.515	-64.557	32.445

	Trimester Interviewed	-2.866	2.690	-.058	-1.065	.287	-8.157	2.425
	Gestational Age at Birth	4.513	1.555	.152	2.901	.004	1.454	7.572
	Sex of Infant (male)	5.283	3.286	.085	1.608	.109	-1.181	11.747
	Maternal Weight (kg)	.184	.122	.088	1.505	.133	-.056	.424
	Day Weight Assessed	.325	.672	.026	.484	.629	-.997	1.648
	DDD*Maternal Weight	.146	.321	.105	.454	.650	-.486	.777

Step 1: DDD; Step 2: Tobacco use during pregnancy, gravidity, maternal age; trimester of pregnancy when maternal interview occurred; Step 3: Gestational age at birth; Step 4: Sex of Infant Step 5: Maternal weight; Step 6: Number of days postpartum maternal weight was assessed; Step 7: DDD by Maternal weight interaction

Table A.8 Stepwise Regression Coefficients Predicting Number of Minor Anomalies at 9 Months

		Unstandardized		Standardized			95% CI for B	
		B	Std. Error	Beta Coeff.	t	Sig.	Lower	Upper
1	(Constant)	5.966	.161		37.020	.000	5.649	6.283
	DDD – 30 days prior	1.580	.410	.201	3.859	.000	.775	2.386
2	(Constant)	7.194	2.690		2.675	.008	1.904	12.485
	DDD – 30 days prior	1.337	.425	.170	3.143	.002	.500	2.173
	Tobacco Use (Yes)	.479	.294	.087	1.632	.104	-.098	1.057
	Gravidity (log)	3.927	1.317	.211	2.982	.003	1.337	6.517
	Maternal Age (log)	-2.553	2.118	-.084	-1.206	.229	-6.718	1.612
	Trimester Interviewed	.010	.230	.002	.044	.965	-.442	.463
3	(Constant)	-2.155	5.870		-.367	.714	-13.699	9.390
	DDD – 30 days prior	1.336	.424	.170	3.151	.002	.502	2.170
	Tobacco Use (Yes)	.490	.293	.089	1.674	.095	-.086	1.066
	Gravidity (log)	3.903	1.313	.210	2.973	.003	1.321	6.485
	Maternal Age (log)	-2.539	2.111	-.084	-1.203	.230	-6.691	1.613
	Trimester Interviewed	-.006	.230	-.001	-.024	.981	-.457	.446
	Gestational Age at Birth	.239	.134	.092	1.790	.074	-.024	.502
4	(Constant)	-2.329	5.895		-.395	.693	-13.922	9.265
	DDD – 30 days prior	1.332	.425	.169	3.135	.002	.496	2.167
	Tobacco Use (Yes)	.492	.293	.089	1.677	.094	-.085	1.069
	Gravidity (log)	3.886	1.315	.209	2.954	.003	1.299	6.472
	Maternal Age (log)	-2.483	2.119	-.082	-1.172	.242	-6.650	1.684
	Trimester Interviewed	-.011	.230	-.003	-.049	.961	-.464	.442
	Gestational Age at Birth	.244	.134	.093	1.815	.070	-.020	.508
	Sex of Infant (male)	-.108	.285	-.020	-.380	.704	-.670	.453
5	(Constant)	-2.594	5.827		-.445	.656	-14.054	8.866
	DDD – 30 days prior	1.215	.422	.154	2.882	.004	.386	2.044
	Tobacco Use (Yes)	.475	.290	.086	1.638	.102	-.095	1.045
	Gravidity (log)	3.743	1.301	.201	2.878	.004	1.185	6.302
	Maternal Age (log)	-1.629	2.113	-.054	-.771	.441	-5.784	2.527
	Trimester Interviewed	-.059	.228	-.014	-.260	.795	-.508	.389
	Gestational Age at Birth	.273	.133	.105	2.054	.041	.012	.535
	Sex of Infant (male)	-.104	.282	-.019	-.367	.714	-.658	.451
	Maternal Weight (kg)	-.029	.010	-.157	-3.048	.002	-.048	-.010
6	(Constant)	-2.383	5.845		-.408	.684	-13.879	9.113
	DDD – 30 days prior	1.224	.422	.155	2.898	.004	.393	2.055
	Tobacco Use (Yes)	.475	.290	.086	1.638	.102	-.095	1.046
	Gravidity (log)	3.742	1.302	.201	2.874	.004	1.181	6.302
	Maternal Age (log)	-1.717	2.121	-.057	-.809	.419	-5.888	2.455
	Trimester Interviewed	-.067	.229	-.016	-.294	.769	-.517	.383
	Gestational Age at Birth	.267	.134	.102	1.994	.047	.004	.530
	Sex of Infant (male)	-.102	.282	-.018	-.360	.719	-.657	.454
	Maternal Weight (kg)	-.029	.010	-.154	-2.962	.003	-.048	-.010
	Day Weight Assessed	.032	.058	.028	.550	.583	-.082	.146
7	(Constant)	-2.234	5.863		-.381	.703	-13.765	9.296
	DDD – 30 days prior	1.953	1.776	.248	1.100	.272	-1.539	5.446
	Tobacco Use (Yes)	.469	.291	.085	1.611	.108	-.104	1.041

	Gravidity (log)	3.739	1.304	.201	2.869	.004	1.176	6.303
	Maternal Age (log)	-1.780	2.129	-.059	-.836	.404	-5.966	2.407
	Trimester Interviewed	-.083	.232	-.019	-.356	.722	-.539	.374
	Gestational Age at Birth	.264	.134	.101	1.964	.050	.000	.527
	Sex of Infant (male)	-.115	.284	-.021	-.403	.687	-.674	.445
	Maternal Weight (kg)	-.027	.011	-.144	-2.525	.012	-.048	-.006
	Day Weight Assessed	.032	.058	.028	.548	.584	-.082	.146
	DDD*Maternal Weight	-.012	.028	-.095	-.423	.673	-.066	.043

Step 1: DDD; Step 2: Tobacco use during pregnancy, gravidity, maternal age; trimester of pregnancy when maternal interview occurred; Step 3: Gestational age at birth; Step 4: Sex of Infant Step 5: Maternal weight; Step 6: Number of days postpartum maternal weight was assessed; Step 7: DDD by Maternal weight interaction

Table A.9 Stepwise Regression Coefficients Predicting Total Dysmorphology Score at 9 Months

		Unstandardized		Standardized			95% CI for B	
		B	Std. Error	Beta Coeff.	t	Sig.	Lower	Upper
1	(Constant)	7.456	.273		27.275	.000	6.918	7.994
	DDD – 30 days prior	2.980	.694	.221	4.295	.000	1.616	4.344
2	(Constant)	8.578	4.533		1.892	.059	-.336	17.492
	DDD – 30 days prior	2.404	.715	.178	3.363	.001	.998	3.810
	Tobacco Use (Yes)	1.170	.495	.123	2.366	.019	.197	2.143
	Gravidity (log)	6.955	2.226	.217	3.124	.002	2.577	11.333
	Maternal Age (log)	-3.736	3.573	-.072	-1.046	.296	-10.762	3.290
	Trimester Interviewed	-.060	.388	-.008	-.156	.876	-.823	.702
3	(Constant)	10.420	9.899		1.053	.293	-9.048	29.888
	DDD – 30 days prior	2.404	.716	.178	3.358	.001	.996	3.812
	Tobacco Use (Yes)	1.169	.495	.123	2.359	.019	.194	2.143
	Gravidity (log)	6.960	2.229	.217	3.122	.002	2.575	11.344
	Maternal Age (log)	-3.743	3.577	-.072	-1.046	.296	-10.779	3.293
	Trimester Interviewed	-.057	.388	-.008	-.147	.884	-.821	.707
	Gestational Age at Birth	-.047	.224	-.011	-.209	.834	-.488	.394
4	(Constant)	10.452	9.936		1.052	.294	-9.089	29.994
	DDD – 30 days prior	2.405	.717	.178	3.353	.001	.994	3.816
	Tobacco Use (Yes)	1.169	.496	.123	2.355	.019	.193	2.144
	Gravidity (log)	6.963	2.234	.217	3.117	.002	2.570	11.357
	Maternal Age (log)	-3.755	3.592	-.072	-1.046	.296	-10.819	3.308
	Trimester Interviewed	-.056	.390	-.007	-.143	.887	-.823	.712
	Gestational Age at Birth	-.048	.225	-.011	-.212	.832	-.491	.395
	Sex of Infant (male)	.023	.482	.002	.047	.962	-.925	.971
5	(Constant)	10.106	9.831		1.028	.305	-9.228	29.441
	DDD – 30 days prior	2.212	.713	.164	3.104	.002	.810	3.613
	Tobacco Use (Yes)	1.136	.491	.120	2.314	.021	.170	2.101
	Gravidity (log)	6.743	2.211	.211	3.049	.002	2.394	11.092
	Maternal Age (log)	-2.383	3.584	-.046	-.665	.506	-9.432	4.665
	Trimester Interviewed	-.127	.387	-.017	-.329	.743	-.888	.633
	Gestational Age at Birth	-.002	.224	.000	-.008	.994	-.441	.438
	Sex of Infant (male)	.020	.477	.002	.042	.966	-.918	.958
	Maternal Weight (kg)	-.048	.016	-.150	-2.943	.003	-.080	-.016
6	(Constant)	10.210	9.860		1.035	.301	-9.183	29.602
	DDD – 30 days prior	2.216	.714	.164	3.104	.002	.812	3.621
	Tobacco Use (Yes)	1.137	.492	.120	2.312	.021	.170	2.103
	Gravidity (log)	6.741	2.214	.210	3.044	.003	2.386	11.096
	Maternal Age (log)	-2.433	3.599	-.047	-.676	.499	-9.511	4.645
	Trimester Interviewed	-.132	.388	-.018	-.340	.734	-.895	.631
	Gestational Age at Birth	-.005	.225	-.001	-.022	.982	-.447	.437
	Sex of Infant (male)	.021	.478	.002	.044	.965	-.918	.961
	Maternal Weight (kg)	-.047	.016	-.149	-2.905	.004	-.080	-.015
	Day Weight Assessed	.018	.098	.009	.185	.853	-.175	.211
7	(Constant)	10.552	9.888		1.067	.287	-8.896	30.000
	DDD – 30 days prior	3.864	2.999	.287	1.288	.198	-2.035	9.763
	Tobacco Use (Yes)	1.121	.493	.118	2.275	.024	.152	2.091
	Gravidity (log)	6.733	2.217	.210	3.038	.003	2.373	11.092
	Maternal Age (log)	-2.568	3.610	-.049	-.711	.477	-9.669	4.532

	Trimester Interviewed	-.168	.394	-.023	-.426	.670	-.942	.606
	Gestational Age at Birth	-.012	.225	-.003	-.055	.956	-.455	.430
	Sex of Infant (male)	-.006	.481	-.001	-.012	.990	-.951	.939
	Maternal Weight (kg)	-.043	.018	-.136	-2.421	.016	-.079	-.008
	Day Weight Assessed	.018	.098	.009	.181	.857	-.175	.211
	DDD*Maternal Weight	-.027	.047	-.126	-.566	.572	-.119	.066

Step 1: DDD; Step 2: Tobacco use during pregnancy, gravidity, maternal age; trimester of pregnancy when maternal interview occurred; Step 3: Gestational age at birth; Step 4: Sex of Infant Step 5: Maternal weight; Step 6: Number of days postpartum maternal weight was assessed; Step 7: DDD by Maternal weight interaction

APPENDIX B: CHAPTER 4 STEPWISE REGRESSION

Table B.1 Stepwise Regression Coefficients Predicting Bayley Cognitive Percentile Rank at 9 Months

		Unstandardized		Standardized			95% CI for B	
		B	Std. Error	Beta Coeff.	t	Sig.	Lower	Upper
1	(Constant)	71.976	1.552		46.378	.000	68.924	75.028
	DDD – 30 days prior	-1.228	3.938	-.016	-.312	.755	-8.973	6.517
2	(Constant)	83.863	26.471		3.168	.002	31.804	135.922
	DDD – 30 days prior	-4.815	4.120	-.065	-1.169	.243	-12.917	3.287
	Tobacco Use (Yes)	-7.898	2.850	-.150	-2.771	.006	-13.503	-2.293
	Gravidity (log)	-7.767	12.828	-.044	-.605	.545	-32.996	17.462
	Maternal Age (log)	9.392	20.592	.032	.456	.649	-31.107	49.891
	Trimester Interviewed	-3.276	2.234	-.079	-1.466	.144	-7.670	1.119
3	(Constant)	79.339	57.128		1.389	.166	-33.013	191.691
	DDD – 30 days prior	-4.814	4.125	-.065	-1.167	.244	-12.927	3.299
	Tobacco Use (Yes)	-7.901	2.854	-.151	-2.768	.006	-13.514	-2.288
	Gravidity (log)	-7.780	12.847	-.044	-.606	.545	-33.047	17.486
	Maternal Age (log)	9.411	20.622	.032	.456	.648	-31.147	49.969
	Trimester Interviewed	-3.284	2.240	-.080	-1.466	.143	-7.689	1.120
	Gestational Age at Birth	.116	1.293	.005	.089	.929	-2.426	2.658
4	(Constant)	74.281	57.199		1.299	.195	-38.213	186.774
	DDD – 30 days prior	-4.994	4.123	-.067	-1.211	.227	-13.104	3.115
	Tobacco Use (Yes)	-7.935	2.851	-.151	-2.783	.006	-13.543	-2.327
	Gravidity (log)	-8.377	12.842	-.047	-.652	.515	-33.634	16.879
	Maternal Age (log)	11.464	20.661	.040	.555	.579	-29.169	52.097
	Trimester Interviewed	-3.495	2.243	-.085	-1.558	.120	-7.907	.916
	Gestational Age at Birth	.242	1.295	.010	.187	.852	-2.305	2.788
	Sex of Infant (male)	-3.641	2.773	-.069	-1.313	.190	-9.095	1.812
5	(Constant)	76.032	56.968		1.335	.183	-36.008	188.071
	DDD – 30 days prior	-4.222	4.125	-.057	-1.024	.307	-12.334	3.890
	Tobacco Use (Yes)	-8.042	2.840	-.153	-2.832	.005	-13.628	-2.457
	Gravidity (log)	-7.568	12.795	-.043	-.591	.555	-32.732	17.596
	Maternal Age (log)	6.181	20.745	.021	.298	.766	-34.619	46.981
	Trimester Interviewed	-3.223	2.238	-.078	-1.440	.151	-7.624	1.179
	Gestational Age at Birth	.055	1.293	.002	.043	.966	-2.487	2.598
	Sex of Infant (male)	-3.612	2.761	-.069	-1.308	.192	-9.042	1.819
	Maternal Weight (kg)	.186	.094	.106	1.990	.047	.002	.371
6	(Constant)	78.168	57.114		1.369	.172	-34.161	190.497
	DDD – 30 days prior	-4.123	4.131	-.055	-.998	.319	-12.248	4.001
	Tobacco Use (Yes)	-8.048	2.842	-.153	-2.832	.005	-13.639	-2.458
	Gravidity (log)	-7.623	12.806	-.043	-.595	.552	-32.809	17.563
	Maternal Age (log)	5.119	20.829	.018	.246	.806	-35.847	46.085
	Trimester Interviewed	-3.323	2.245	-.081	-1.480	.140	-7.740	1.093
	Gestational Age at Birth	-.008	1.298	.000	-.007	.995	-2.561	2.544
	Sex of Infant (male)	-3.582	2.764	-.068	-1.296	.196	-9.018	1.854
	Maternal Weight (kg)	.193	.094	.109	2.044	.042	.007	.378
	Day Weight Assessed	.363	.569	.034	.637	.524	-.757	1.483
7	(Constant)	74.242	57.165		1.299	.195	-38.188	186.672
	DDD – 30 days prior	-24.643	17.321	-.330	-1.423	.156	-58.709	9.422
	Tobacco Use (Yes)	-8.245	2.845	-.157	-2.898	.004	-13.841	-2.650

	Gravidity (log)	-7.505	12.798	-.042	-.586	.558	-32.675	17.665
	Maternal Age (log)	6.788	20.860	.023	.325	.745	-34.239	47.814
	Trimester Interviewed	-2.874	2.274	-.070	-1.264	.207	-7.346	1.598
	Gestational Age at Birth	.086	1.299	.004	.067	.947	-2.469	2.641
	Sex of Infant (male)	-3.250	2.775	-.062	-1.171	.242	-8.708	2.209
	Maternal Weight (kg)	.142	.103	.080	1.374	.170	-.061	.344
	Day Weight Assessed	.367	.569	.034	.645	.519	-.752	1.486
	DDD*Maternal Weight	.331	.271	.283	1.220	.223	-.203	.865

Step 1: DDD; Step 2: Tobacco use during pregnancy, gravidity, maternal age; trimester of pregnancy when maternal interview occurred; Step 3: Gestational age at birth; Step 4: Sex of Infant Step 5: Maternal weight; Step 6: Number of days postpartum maternal weight was assessed; Step 7: DDD by Maternal weight interaction

Table B.2 Stepwise Regression Coefficients Predicting Bayley Language Percentile Rank at 9 Months

		Unstandardized		Standardized			95% CI for B	
		B	Std. Error	Beta Coeff.	t	Sig.	Lower	Upper
1	(Constant)	37.703	1.356		27.797	.000	35.036	40.371
	DDD – 30 days prior	-2.636	3.442	-.040	-.766	.444	-9.405	4.133
2	(Constant)	60.885	23.104		2.635	.009	15.447	106.323
	DDD – 30 days prior	-4.274	3.596	-.065	-1.189	.235	-11.346	2.797
	Tobacco Use (Yes)	1.388	2.488	.030	.558	.577	-3.504	6.280
	Gravidity (log)	-11.774	11.197	-.076	-1.052	.294	-33.795	10.246
	Maternal Age (log)	-2.659	17.974	-.010	-.148	.882	-38.007	32.689
	Trimester Interviewed	-5.614	1.950	-.156	-2.878	.004	-9.449	-1.778
3	(Constant)	81.193	49.848		1.629	.104	-16.842	179.228
	DDD – 30 days prior	-4.279	3.600	-.066	-1.189	.235	-11.358	2.800
	Tobacco Use (Yes)	1.402	2.490	.031	.563	.574	-3.496	6.300
	Gravidity (log)	-11.717	11.210	-.075	-1.045	.297	-33.763	10.330
	Maternal Age (log)	-2.746	17.995	-.011	-.153	.879	-38.135	32.644
	Trimester Interviewed	-5.575	1.954	-.155	-2.853	.005	-9.418	-1.731
	Gestational Age at Birth	-.519	1.128	-.024	-.460	.646	-2.737	1.699
4	(Constant)	74.506	49.752		1.498	.135	-23.341	172.354
	DDD – 30 days prior	-4.517	3.587	-.069	-1.259	.209	-11.571	2.537
	Tobacco Use (Yes)	1.357	2.480	.030	.547	.585	-3.521	6.235
	Gravidity (log)	-12.506	11.170	-.080	-1.120	.264	-34.474	9.462
	Maternal Age (log)	-.032	17.971	.000	-.002	.999	-35.375	35.311
	Trimester Interviewed	-5.854	1.951	-.162	-3.000	.003	-9.691	-2.017
	Gestational Age at Birth	-.352	1.126	-.016	-.313	.755	-2.567	1.863
	Sex of Infant (male)	-4.814	2.412	-.105	-1.996	.047	-9.557	-.071
5	(Constant)	75.703	49.657		1.525	.128	-21.960	173.365
	DDD – 30 days prior	-3.989	3.595	-.061	-1.110	.268	-11.060	3.082
	Tobacco Use (Yes)	1.284	2.476	.028	.519	.604	-3.585	6.153
	Gravidity (log)	-11.953	11.153	-.077	-1.072	.285	-33.888	9.982
	Maternal Age (log)	-3.641	18.083	-.014	-.201	.841	-39.205	31.924
	Trimester Interviewed	-5.667	1.951	-.157	-2.905	.004	-9.504	-1.831
	Gestational Age at Birth	-.479	1.127	-.022	-.425	.671	-2.696	1.737
	Sex of Infant (male)	-4.794	2.407	-.104	-1.992	.047	-9.528	-.060
	Maternal Weight (kg)	.127	.082	.083	1.559	.120	-.033	.288
6	(Constant)	78.010	49.770		1.567	.118	-19.874	175.894
	DDD – 30 days prior	-3.883	3.600	-.059	-1.079	.281	-10.962	3.197
	Tobacco Use (Yes)	1.277	2.477	.028	.516	.606	-3.594	6.149
	Gravidity (log)	-12.013	11.159	-.077	-1.076	.282	-33.960	9.935
	Maternal Age (log)	-4.788	18.151	-.019	-.264	.792	-40.486	30.910
	Trimester Interviewed	-5.776	1.957	-.160	-2.952	.003	-9.625	-1.928
	Gestational Age at Birth	-.548	1.131	-.025	-.485	.628	-2.772	1.676
	Sex of Infant (male)	-4.762	2.409	-.104	-1.977	.049	-9.499	-.025
	Maternal Weight (kg)	.134	.082	.087	1.633	.103	-.027	.296
	Day Weight Assessed	.392	.496	.042	.790	.430	-.584	1.368
7	(Constant)	78.586	49.917		1.574	.116	-19.589	176.760
	DDD – 30 days prior	-.873	15.124	-.013	-.058	.954	-30.619	28.873
	Tobacco Use (Yes)	1.306	2.484	.028	.526	.599	-3.580	6.192

	Gravidity (log)	-12.030	11.175	-.077	-1.077	.282	-34.008	9.948
	Maternal Age (log)	-5.033	18.215	-.020	-.276	.782	-40.858	30.792
	Trimester Interviewed	-5.842	1.986	-.162	-2.942	.003	-9.748	-1.937
	Gestational Age at Birth	-.562	1.134	-.026	-.495	.621	-2.793	1.669
	Sex of Infant (male)	-4.810	2.423	-.105	-1.985	.048	-9.577	-.044
	Maternal Weight (kg)	.142	.090	.092	1.573	.117	-.035	.319
	Day Weight Assessed	.391	.497	.042	.788	.431	-.586	1.369
	DDD*Maternal Weight	-.049	.237	-.048	-.205	.838	-.514	.417

Step 1: DDD; Step 2: Tobacco use during pregnancy, gravidity, maternal age; trimester of pregnancy when maternal interview occurred; Step 3: Gestational age at birth; Step 4: Sex of Infant Step 5: Maternal weight; Step 6: Number of days postpartum maternal weight was assessed; Step 7: DDD by Maternal weight interaction

Table B.3 Stepwise Regression Coefficients Predicting Bayley Motor Percentile Rank at 9 Months

		Unstandardized		Standardized			95% CI for B	
		B	Std. Error	Beta Coeff.	t	Sig.	Lower	Upper
1	(Constant)	39.567	1.530		25.862	.000	36.558	42.575
	DDD – 30 days prior	-6.069	3.882	-.082	-1.563	.119	-13.704	1.566
2	(Constant)	77.896	25.825		3.016	.003	27.108	128.685
	DDD – 30 days prior	-5.284	4.067	-.072	-1.299	.195	-13.282	2.713
	Tobacco Use (Yes)	.133	2.813	.003	.047	.962	-5.400	5.666
	Gravidity (log)	-16.006	12.664	-.091	-1.264	.207	-40.911	8.899
	Maternal Age (log)	-21.742	20.328	-.076	-1.070	.286	-61.720	18.237
	Trimester Interviewed	.530	2.206	.013	.240	.810	-3.808	4.868
3	(Constant)	43.768	56.277		.778	.437	-66.911	154.448
	DDD – 30 days prior	-5.277	4.070	-.071	-1.297	.196	-13.281	2.727
	Tobacco Use (Yes)	.157	2.816	.003	.056	.956	-5.381	5.695
	Gravidity (log)	-16.103	12.674	-.092	-1.271	.205	-41.028	8.823
	Maternal Age (log)	-21.596	20.344	-.075	-1.062	.289	-61.607	18.415
	Trimester Interviewed	.465	2.209	.011	.210	.833	-3.880	4.810
	Gestational Age at Birth	.870	1.275	.036	.683	.495	-1.637	3.378
4	(Constant)	40.760	56.440		.722	.471	-70.241	151.762
	DDD – 30 days prior	-5.382	4.074	-.073	-1.321	.187	-13.395	2.630
	Tobacco Use (Yes)	.177	2.817	.003	.063	.950	-5.364	5.718
	Gravidity (log)	-16.453	12.689	-.094	-1.297	.196	-41.408	8.502
	Maternal Age (log)	-20.392	20.414	-.071	-.999	.319	-60.540	19.757
	Trimester Interviewed	.341	2.216	.008	.154	.878	-4.018	4.700
	Gestational Age at Birth	.944	1.279	.039	.738	.461	-1.572	3.460
	Sex of Infant (male)	-2.137	2.740	-.041	-.780	.436	-7.525	3.251
5	(Constant)	41.887	56.352		.743	.458	-68.943	152.716
	DDD – 30 days prior	-4.816	4.086	-.065	-1.179	.239	-12.852	3.219
	Tobacco Use (Yes)	.256	2.813	.005	.091	.928	-5.277	5.788
	Gravidity (log)	-15.860	12.674	-.090	-1.251	.212	-40.787	9.067
	Maternal Age (log)	-24.264	20.549	-.085	-1.181	.239	-64.679	16.152
	Trimester Interviewed	.541	2.217	.013	.244	.807	-3.819	4.901
	Gestational Age at Birth	.808	1.281	.033	.631	.529	-1.711	3.326
	Sex of Infant (male)	-2.115	2.735	-.041	-.773	.440	-7.494	3.265
	Maternal Weight (kg)	.137	.093	.078	1.472	.142	-.046	.319
6	(Constant)	39.306	56.480		.696	.487	-71.775	150.388
	DDD – 30 days prior	-4.936	4.091	-.067	-1.207	.228	-12.982	3.109
	Tobacco Use (Yes)	.248	2.815	.005	.088	.930	-5.288	5.784
	Gravidity (log)	-15.793	12.682	-.090	-1.245	.214	-40.734	9.149
	Maternal Age (log)	-22.973	20.627	-.080	-1.114	.266	-63.541	17.595
	Trimester Interviewed	.664	2.224	.016	.298	.766	-3.710	5.037
	Gestational Age at Birth	.885	1.285	.036	.689	.492	-1.643	3.412
	Sex of Infant (male)	-2.151	2.737	-.041	-.786	.432	-7.534	3.232
	Maternal Weight (kg)	.129	.093	.074	1.381	.168	-.055	.313
	Day Weight Assessed	-.441	.564	-.042	-.782	.435	-1.550	.668
7	(Constant)	40.096	56.665		.708	.480	-71.352	151.544
	DDD – 30 days prior	-1.185	17.187	-.016	-.069	.945	-34.988	32.619
	Tobacco Use (Yes)	.212	2.823	.004	.075	.940	-5.340	5.765

	Gravidity (log)	-15.814	12.699	-.090	-1.245	.214	-40.791	9.162
	Maternal Age (log)	-23.278	20.699	-.081	-1.125	.262	-63.989	17.433
	Trimester Interviewed	.582	2.256	.014	.258	.797	-3.856	5.019
	Gestational Age at Birth	.868	1.289	.036	.673	.501	-1.668	3.403
	Sex of Infant (male)	-2.212	2.754	-.043	-.803	.422	-7.628	3.205
	Maternal Weight (kg)	.138	.102	.079	1.352	.177	-.063	.340
	Day Weight Assessed	-.442	.565	-.042	-.782	.435	-1.552	.669
	DDD*Maternal Weight	-.061	.269	-.052	-.225	.822	-.590	.469

Step 1: DDD; Step 2: Tobacco use during pregnancy, gravidity, maternal age; trimester of pregnancy when maternal interview occurred; Step 3: Gestational age at birth; Step 4: Sex of Infant Step 5: Maternal weight; Step 6: Number of days postpartum maternal weight was assessed; Step 7: DDD by Maternal weight interaction

Table B.4 Stepwise Regression Coefficients Predicting Bayley Social/Emotional Percentile Rank at 9 Months

		Unstandardized		Standardized			95% CI for B	
		B	Std. Error	Beta Coeff.	t	Sig.	Lower	Upper
1	(Constant)	54.255	1.842		29.447	.000	50.632	57.879
	DDD – 30 days prior	-.635	4.745	-.007	-.134	.894	-9.968	8.697
2	(Constant)	2.522	31.351		.080	.936	-59.136	64.181
	DDD – 30 days prior	-.721	5.003	-.008	-.144	.885	-10.560	9.118
	Tobacco Use (Yes)	-1.614	3.409	-.026	-.474	.636	-8.318	5.090
	Gravidity (log)	-17.552	15.362	-.083	-1.143	.254	-47.765	12.661
	Maternal Age (log)	47.104	24.654	.137	1.911	.057	-1.384	95.592
	Trimester Interviewed	-1.866	2.683	-.038	-.696	.487	-7.144	3.411
3	(Constant)	121.436	67.972		1.787	.075	-12.246	255.117
	DDD – 30 days prior	-.842	4.983	-.009	-.169	.866	-10.642	8.958
	Tobacco Use (Yes)	-1.707	3.395	-.027	-.503	.615	-8.385	4.971
	Gravidity (log)	-17.288	15.301	-.082	-1.130	.259	-47.380	12.804
	Maternal Age (log)	46.642	24.556	.135	1.899	.058	-1.652	94.936
	Trimester Interviewed	-1.666	2.674	-.034	-.623	.534	-6.926	3.593
	Gestational Age at Birth	-3.032	1.539	-.104	-1.970	.050	-6.059	-.004
4	(Constant)	126.477	68.133		1.856	.064	-7.523	260.477
	DDD – 30 days prior	-.657	4.985	-.007	-.132	.895	-10.463	9.148
	Tobacco Use (Yes)	-1.737	3.395	-.028	-.512	.609	-8.414	4.940
	Gravidity (log)	-16.765	15.307	-.079	-1.095	.274	-46.869	13.340
	Maternal Age (log)	44.578	24.631	.129	1.810	.071	-3.865	93.022
	Trimester Interviewed	-1.465	2.681	-.030	-.547	.585	-6.738	3.807
	Gestational Age at Birth	-3.151	1.543	-.108	-2.042	.042	-6.186	-.116
	Sex of Infant (male)	3.467	3.311	.056	1.047	.296	-3.045	9.979
5	(Constant)	127.054	68.220		1.862	.063	-7.118	261.227
	DDD – 30 days prior	-.441	5.012	-.005	-.088	.930	-10.299	9.418
	Tobacco Use (Yes)	-1.706	3.399	-.027	-.502	.616	-8.392	4.980
	Gravidity (log)	-16.564	15.330	-.078	-1.081	.281	-46.715	13.586
	Maternal Age (log)	43.024	24.881	.125	1.729	.085	-5.911	91.960
	Trimester Interviewed	-1.390	2.689	-.028	-.517	.606	-6.678	3.898
	Gestational Age at Birth	-3.205	1.549	-.110	-2.069	.039	-6.252	-.158
	Sex of Infant (male)	3.487	3.315	.056	1.052	.294	-3.032	10.007
	Maternal Weight (kg)	.053	.113	.025	.468	.640	-.169	.274
6	(Constant)	118.696	67.961		1.747	.082	-14.968	252.361
	DDD – 30 days prior	-.901	4.990	-.010	-.180	.857	-10.715	8.914
	Tobacco Use (Yes)	-1.738	3.381	-.028	-.514	.608	-8.388	4.913
	Gravidity (log)	-16.394	15.248	-.077	-1.075	.283	-46.383	13.595
	Maternal Age (log)	47.357	24.827	.137	1.907	.057	-1.472	96.186
	Trimester Interviewed	-.995	2.680	-.020	-.371	.711	-6.266	4.277
	Gestational Age at Birth	-2.952	1.545	-.101	-1.910	.057	-5.991	.087
	Sex of Infant (male)	3.370	3.298	.054	1.022	.308	-3.116	9.855
	Maternal Weight (kg)	.027	.113	.013	.242	.809	-.194	.249
	Day Weight Assessed	-1.482	.677	-.117	-2.188	.029	-2.815	-.150
7	(Constant)	123.249	68.022		1.812	.071	-10.537	257.036
	DDD – 30 days prior	23.567	20.921	.263	1.126	.261	-17.582	64.715
	Tobacco Use (Yes)	-1.978	3.385	-.032	-.584	.559	-8.636	4.679

	Gravidity (log)	-16.423	15.238	-.078	-1.078	.282	-46.393	13.546
	Maternal Age (log)	45.737	24.847	.133	1.841	.067	-3.133	94.606
	Trimester Interviewed	-1.533	2.716	-.031	-.564	.573	-6.874	3.808
	Gestational Age at Birth	-3.060	1.547	-.105	-1.978	.049	-6.102	-.018
	Sex of Infant (male)	2.913	3.317	.047	.878	.381	-3.612	9.437
	Maternal Weight (kg)	.086	.123	.041	.700	.484	-.155	.327
	Day Weight Assessed	-1.486	.677	-.117	-2.195	.029	-2.817	-.155
	DDD*Maternal Weight	-.395	.328	-.282	-1.204	.229	-1.040	.250

Step 1: DDD; Step 2: Tobacco use during pregnancy, gravidity, maternal age; trimester of pregnancy when maternal interview occurred; Step 3: Gestational age at birth; Step 4: Sex of Infant Step 5: Maternal weight; Step 6: Number of days postpartum maternal weight was assessed; Step 7: DDD by Maternal weight interaction

APPENDIX C: CHAPTER 5 SUPPLEMENTAL TABLES AND FIGURE

Table C.1 Maternal Dietary Intake Including Supplements During Pregnancy: Weekday

	Drank during pregnancy with alcohol in recalls (*n*=47)		Drank during pregnancy without alcohol in recalls (*n*=76)		Did not drink during pregnancy (*n*=55)		
	Mean	(SD)	Mean	(SD)	Mean	(SD)	p
Total Grams	2296.6	(896.2)	2229.7	(883.1)	2319.8	(884.9)	.833
Energy (kcal)	1574.7	(759.6)	1596.3	(721.8)	1384.6	(581.8)	.194
Total Fat (g)	48.8	(36.9)	50.0	(31.4)	39.9	(24.7)	.162
Total Carbohydrate (g)	228.0	(108.6)	232.6	(106.1)	211.0	(90.2)	.475
Total Protein (g)	52.5	(30.4)	56.2	(29.8)	47.9	(28.3)	.287
Animal Protein (g)	31.0	(24.2)	30.1	(23.3)	26.0	(23.1)	.498
Vegetable Protein (g)	21.5	(11.4)	26.1	(13.7)	22.0	(10.1)	.062
Alcohol (g)	3.4	(11.2)	.0	(.0)	.0	(.0)	.002
Cholesterol (mg)	183.1	(164.3)	160.4	(161.4)	171.3	(252.9)	.820
Total Saturated Fatty Acids (g)	13.3	(11.2)	13.9	(9.2)	11.0	(6.7)	.193
Total Monounsaturated Fatty Acids (g)	16.9	(13.1)	16.8	(10.9)	13.2	(9.0)	.136
Total Polyunsaturated Fatty Acids (g)	14.7	(12.6)	15.3	(12.7)	12.4	(9.5)	.373
Total Dietary Fiber (g)	13.4	(8.6)	15.6	(11.5)	13.7	(6.9)	.355
Vitamin A (RAE) (mcg)	277.0	(234.7)	330.0	(313.6)	334.1	(300.8)	.543
Vitamin D (calciferol) (mcg)	2.9	(4.4)	3.0	(3.9)	3.4	(3.8)	.800
Vitamin E (Total □-Tocopherol) (mg)	5.8	(5.4)	5.3	(4.7)	5.2	(4.4)	.786
Vitamin K (phylloquinone) (mcg)	40.4	(52.1)	48.7	(40.8)	45.7	(44.7)	.616
Vitamin C (ascorbic acid) (mg)	69.8	(72.7)	80.9	(96.5)	89.8	(96.0)	.539
Thiamin (vitamin B_1) (mg)	1.5	(.8)	1.7	(.8)	1.6	(.9)	.413
Riboflavin (vitamin B_2) (mg)	1.2	(.6)	1.2	(.6)	1.4	(1.1)	.237
Niacin (vitamin B_3) (mg)	19.0	(8.9)	19.6	(9.7)	20.2	(12.6)	.868
Pantothenic Acid (mg)	3.7	(1.9)	3.8	(2.1)	4.3	(3.8)	.401
Vitamin B_6 (mg)	1.3	(.8)	1.3	(.9)	1.5	(1.6)	.479

Dietary Folate Equivalents (mcg)	2127.8	(1306.0)	2299.3	(1050.1)	2119.8	(1123.1)	.599
Vitamin B_{12} (cobalamin) (mcg)	3.0	(5.8)	3.4	(5.7)	3.4	(6.2)	.918
Calcium (mg)	594.3	(352.4)	626.0	(324.4)	649.8	(320.0)	.699
Phosphorus (mg)	669.6	(380.3)	740.0	(394.7)	682.6	(350.5)	.535
Magnesium (mg)	184.5	(91.6)	192.7	(100.0)	182.2	(82.0)	.790
Iron (mg)	46.4	(21.5)	54.2	(29.5)	47.3	(38.9)	.299
Zinc (mg)	6.5	(4.4)	6.6	(3.5)	6.3	(3.3)	.860
Copper (mg)	1.9	(.9)	2.1	(1.0)	1.9	(1.3)	.517
Manganese (mg)	4.7	(2.2)	5.3	(2.6)	4.9	(3.0)	.478
Selenium (mcg)	85.6	(49.1)	97.1	(55.9)	84.9	(52.4)	.338
Sodium (mg)	2612.0	(1436.0)	2900.9	(1663.5)	2226.7	(1016.8)	.031[C]
Potassium (mg)	1701.8	(1016.5)	1782.8	(1147.9)	1753.0	(1004.3)	.920
Choline (mg)	229.5	(165.8)	213.0	(130.1)	209.1	(185.5)	.790
Betaine (mg)	154.0	(97.7)	174.7	(101.7)	153.9	(94.0)	.380
Linoleic acid (g)	13.3	(11.6)	13.6	(11.3)	11.1	(8.7)	.382
Alpha-linolenic acid (g)	.9	(.8)	1.2	(1.2)	.8	(.7)	.091
Eicosapentaenoic acid (EPA) (g)	.0	(.1)	.1	(.2)	.1	(.2)	.707
Docosapentaenoic acid (DPA) (g)	.0	(.0)	.0	(.0)	.0	(.0)	.813
Docosahexaenoic acid (DHA) (g)	.1	(.2)	.1	(.3)	.1	(.3)	.779

No significant post-hoc Dunnet C comparisons.

Table C.2 Maternal Dietary Intake During Pregnancy: Weekend Day

	Drank during pregnancy with alcohol in recalls (*n*=47)		Drank during pregnancy without alcohol in recalls (*n*=76)		Did not drink during pregnancy (*n*=55)		
	Mean	(SD)	Mean	(SD)	Mean	(SD)	p
Total Grams	2457.8	(986.9)	2099.1	(927.4)	2107.2	(942.7)	.091
Energy (kcal)	1798.6	(653.8)	1642.8	(752.9)	1537.9	(726.6)	.191
Total Fat (g)	51.6	(33.4)	58.0	(38.3)	52.6	(33.8)	.549
Total Carbohydrate (g)	200.7	(88.5)	218.8	(101.1)	219.0	(119.6)	.590
Total Protein (g)	53.5	(28.5)	63.0	(44.1)	49.3	(23.6)	.073
Animal Protein (g)	32.6	(23.6)	41.9	(42.1)	28.2	(18.4)	.044[C]
Vegetable Protein (g)	20.9	(11.9)	21.1	(10.2)	21.2	(14.1)	.995
Alcohol (g)	46.7	(48.2)	.0	(.0)	.0	(.1)	<.001[A,B]
Cholesterol (mg)	201.2	(174.3)	196.2	(189.8)	185.5	(167.9)	.900
Total Saturated Fatty Acids (g)	14.3	(9.7)	15.6	(10.8)	15.2	(9.8)	.792
Total Monounsaturated Fatty Acids (g)	18.4	(12.4)	20.8	(14.8)	18.1	(12.8)	.466
Total Polyunsaturated Fatty Acids (g)	14.7	(11.1)	16.7	(12.5)	15.0	(11.5)	.598
Total Dietary Fiber (g)	9.4	(6.3)	11.5	(6.5)	11.7	(8.4)	.195
Vitamin A (RAE) (mcg)	223.7	(288.4)	267.5	(304.0)	230.9	(173.4)	.608
Vitamin D (calciferol) (mcg)	3.0	(3.6)	4.8	(7.2)	3.4	(4.2)	.156
Vitamin E (Total a-Tocopherol) (mg)	4.8	(4.3)	5.9	(5.5)	4.6	(3.6)	.286
Vitamin K (phylloquinone) (mcg)	39.9	(37.1)	40.2	(36.7)	36.3	(33.8)	.813
Vitamin C (ascorbic acid) (mg)	53.5	(72.7)	77.0	(105.5)	78.4	(100.0)	.339
Thiamin (vitamin B_1) (mg)	1.3	(.7)	1.5	(.8)	1.6	(1.0)	.101
Riboflavin (vitamin B_2) (mg)	1.2	(.6)	1.2	(.8)	1.2	(.7)	.985
Niacin (vitamin B_3) (mg)	20.7	(10.6)	21.6	(14.6)	17.9	(9.4)	.223
Pantothenic Acid (mg)	3.5	(1.8)	3.3	(1.7)	3.6	(2.7)	.747
Vitamin B_6 (mg)	1.5	(.7)	1.3	(.8)	1.2	(.9)	.160
Dietary Folate Equivalents (mcg)	1975.3	(1260.9)	2244.6	(1053.3)	2218.7	(1106.4)	.402
Vitamin B_{12} (cobalamin) (mcg)	3.6	(4.2)	5.8	(8.6)	3.0	(5.1)	.038

Calcium (mg)	485.0	(262.1)	575.1	(340.6)	592.9	(289.3)	.166
Phosphorus (mg)	727.3	(288.7)	723.4	(375.8)	654.4	(308.3)	.431
Magnesium (mg)	203.2	(75.2)	182.8	(90.4)	165.0	(98.3)	.102
Iron (mg)	46.8	(23.8)	53.9	(29.0)	48.3	(30.6)	.326
Zinc (mg)	6.2	(3.6)	7.0	(4.8)	6.3	(3.2)	.484
Copper (mg)	1.8	(.8)	2.0	(.9)	1.8	(1.0)	.619
Manganese (mg)	4.5	(2.0)	4.9	(2.1)	4.7	(2.6)	.630
Selenium (mcg)	85.8	(48.1)	105.8	(71.1)	83.5	(42.6)	.054
Sodium (mg)	2285.6	(1377.7)	2720.4	(1735.2)	2520.5	(1660.2)	.353
Potassium (mg)	1705.0	(809.3)	1639.5	(909.9)	1465.5	(977.1)	.373
Choline (mg)	310.1	(166.9)	238.8	(181.0)	210.4	(141.9)	.009[B]
Betaine (mg)	195.9	(120.9)	168.6	(99.1)	149.1	(82.5)	.067
Linoleic acid (g)	12.9	(9.7)	14.2	(11.5)	13.3	(10.0)	.779
Alpha-linolenic acid (g)	1.1	(1.1)	1.1	(.8)	1.2	(.9)	.924
Eicosapentaenoic acid (EPA) (g)	.1	(.3)	.2	(.7)	.0	(.1)	.096
Docosapentaenoic acid (DPA) (g)	.0	(.1)	.1	(.2)	.0	(.0)	.094
Docosahexaenoic acid (DHA) (g)	.2	(.5)	.4	(1.2)	.1	(.2)	.080

Post-hoc Dunnet C comparisons significantly different between: [A.] Group 1 & Group 2; [B.] Group 1 & Group 3; [C.]Group 2 & Group 3

Table C.3 Maternal Dietary Intake During Pregnancy: Simple Average

	Drank during pregnancy with alcohol in recalls (*n*=47)		Drank during pregnancy without alcohol in recalls (*n*=76)		Did not drink during pregnancy (*n*=55)		
	Mean	(SD)	Mean	(SD)	Mean	(SD)	p
Total Grams	2377.2	(787.4)	2164.4	(821.7)	2213.5	(793.7)	.353
Energy (kcal)	1686.7	(589.4)	1619.6	(604.9)	1461.2	(579.0)	.136
Total Fat (g)	50.2	(30.0)	54.0	(27.0)	46.2	(25.5)	.279
Total Carbohydrate (g)	214.3	(87.4)	225.7	(85.2)	215.0	(87.8)	.704
Total Protein (g)	53.0	(24.6)	59.6	(29.2)	48.6	(23.0)	.058
Animal Protein (g)	31.8	(19.8)	36.0	(25.8)	27.1	(17.4)	.075
Vegetable Protein (g)	21.5	(11.4)	26.1	(13.7)	21.6	(9.9)	.324
Alcohol (g)	25.1	(24.9)	.0	(.0)	.0	(.0)	<.001[A,B]
Cholesterol (mg)	192.1	(124.9)	178.3	(126.7)	178.4	(158.8)	.839
Total Saturated Fatty Acids (g)	13.8	(8.8)	14.8	(7.3)	13.1	(7.1)	.472
Total Monounsaturated Fatty Acids (g)	17.7	(10.8)	18.8	(9.7)	15.7	(9.3)	.209
Total Polyunsaturated Fatty Acids (g)	14.7	(9.9)	16.0	(9.8)	13.7	(9.1)	.409
Total Dietary Fiber (g)	11.4	(6.0)	13.6	(7.0)	12.7	(6.4)	.212
Total Vitamin A (Retinol Equivalents) (mcg)	250.4	(213.0)	298.7	(231.5)	373.1	(718.5)	.363
Vitamin D (calciferol) (mcg)	2.9	(3.1)	3.9	(4.1)	3.4	(3.1)	.346
Vitamin E (Total a-Tocopherol) (mg)	5.3	(4.2)	5.6	(4.0)	4.9	(3.1)	.633
Vitamin K (phylloquinone) (mcg)	40.1	(36.8)	44.4	(31.9)	41.0	(31.1)	.742
Vitamin C (ascorbic acid) (mg)	61.6	(64.9)	78.9	(76.5)	84.1	(88.3)	.316
Thiamin (vitamin B_1) (mg)	1.4	(.7)	1.6	(.6)	1.6	(.8)	.164
Riboflavin (vitamin B_2) (mg)	1.2	(.5)	1.2	(.5)	1.3	(.8)	.615
Niacin (vitamin B_3) (mg)	19.9	(7.9)	20.6	(9.3)	19.0	(9.8)	.619
Pantothenic Acid (mg)	3.6	(1.5)	3.6	(1.6)	4.0	(2.9)	.497
Vitamin B_6 (mg)	1.5	(.7)	1.3	(.8)	1.2	(.9)	.160
Dietary Folate Equivalents (mcg)	2051.6	(1183.4)	2271.9	(1028.7)	2169.2	(1052.3)	.544
Vitamin B_{12} (cobalamin) (mcg)	3.3	(4.0)	4.6	(5.8)	3.2	(5.3)	.221

Calcium (mg)	539.6	(249.2)	600.5	(232.0)	621.4	(235.9)	.203
Phosphorus (mg)	698.5	(269.0)	731.7	(308.4)	668.5	(281.9)	.467
Magnesium (mg)	193.8	(62.5)	187.8	(78.8)	173.6	(80.3)	.369
Iron (mg)	46.6	(19.9)	54.1	(28.2)	47.8	(33.5)	.275
Zinc (mg)	6.4	(3.4)	6.8	(3.3)	6.3	(2.8)	.569
Copper (mg)	1.9	(.7)	2.0	(.9)	1.9	(1.1)	.519
Manganese (mg)	4.6	(1.9)	5.1	(2.2)	4.8	(2.7)	.498
Selenium (mcg)	85.7	(40.0)	101.5	(49.0)	84.2	(42.3)	.052
Sodium (mg)	2448.8	(1185.0)	2810.7	(1248.6)	2373.6	(1144.4)	.085
Potassium (mg)	1703.4	(715.0)	1711.2	(829.5)	1609.3	(827.3)	.747
Choline (mg)	269.8	(122.0)	225.9	(118.4)	209.8	(131.2)	.044
Betaine (mg)	175.0	(79.8)	171.6	(75.9)	151.5	(72.4)	.217
Linoleic acid (g)	13.1	(8.9)	13.9	(8.9)	12.2	(8.0)	.544
Alpha-linolenic acid (g)	1.0	(.8)	1.2	(.8)	1.0	(.7)	.513
Eicosapentaenoic acid (EPA) (g)	.1	(.2)	.1	(.4)	.1	(.1)	.107
Docosapentaenoic acid (DPA) (g)	.0	(.0)	.0	(.1)	.0	(.0)	.081
Docosahexaenoic acid (DHA) (g)	.1	(.3)	.3	(.6)	.1	(.2)	.095

Post-hoc Dunnet C comparisons significantly different between: [A.] Group 1 & Group 2; [B.] Group 1 & Group 3; [C.]Group 2 & Group 3

Table C.4 Percent of Women Less Than Estimated Average Requirement EAR Using Simple Mean

	% Less than EAR				
	EAR	Drank during pregnancy with alcohol in recalls (*n*=47)	Drank during pregnancy without alcohol in recalls (*n*=76)	Did not drink during pregnancy (*n*=55)	p
Vitamin A (RAE) (mcg)[1]	550	87.2	88.2	89.1	.959
Vitamin D (mcg)	10	97.9	94.7	92.7	.494
Vitamin E (mg)	12	91.5	96.1	96.4	.451
Vitamin C (mg)	70	66.0	63.2	54.5	.450
Thiamin (mg)	1.2	44.7	26.3	32.7	.109
Riboflavin (mg)	1.2	51.1	57.9	58.2	.712
Niacin Equivalents (mg)[2]	14	27.7	21.1	30.9	.422
Vitamin B_6 (mg)	1.6	61.7	72.4	76.4	.247
Dietary Folate Equivalents (mcg)	520	23.4	17.1	20.0	.692
Vitamin B_{12} (mcg)	2.2	46.8	39.5	58.2	.106
Calcium (mg)	800	78.7	76.3	80.0	.875
Phosphorus (mg)	580	38.3	36.8	45.5	.590
Magnesium (mcg)	290	95.7	93.4	92.7	.805
Iron (mg)	22	10.6	15.8	18.2	.560
Zinc (mg)	9.5	85.1	85.5	90.9	.594
Copper (mg)	0.8	8.5	7.9	16.4	.257
Selenium (mcg)	49	21.3	10.5	16.4	.260
# of micronutrients below EAR	--	9.4 (3.5)	9.0 (3.0)	6.7 (3.6)	.522

Estimated Average Requirement (EAR) for pregnant women, aged 19–30, used for: vitamin A, C, D, E, thiamin, riboflavin, niacin, vitamin B_6, folate, vitamin B_{12}, calcium, phosphorus, magnesium, iron, zinc, and selenium.

1 Retinol Activity Equivalents

2 Niacin Equivalents (1 niacin equivalent = 1 mg of Niacin or 60 mg of tryptophan).

Table C.5 Percent of Women Less Than Recommended Dietary Allowance (RDA) Using Simple Mean

	% Less than RDA or AI				
	RDA/AI	Drank during pregnancy with alcohol in recalls (*n*=47)	Drank during pregnancy without alcohol in recalls (*n*=76)	Did not drink during pregnancy (*n*=55)	p
Vitamin A (RAE) (mcg)[1]	770	95.7	96.1	94.5	.915
Vitamin D (mcg)	15	97.9	97.4	100.0	.494
Vitamin E (mg)	15	95.7	97.4	98.2	.753
Vitamin C (mg)	85	74.5	65.8	61.8	.387
Vitamin K (mcg)	90^	93.2	94.4	92.7	.893
Thiamin (mg)	1.4	57.4	44.7	41.8	.246
Riboflavin (mg)	1.4	66.0	72.4	69.1	.749
Niacin Equivalent (mg) [2]	18	46.8	44.7	54.5	.525
Pantothenic acid (mg)	6^	89.4	92.1	89.1	.809
Vitamin B_6 (mg)	1.9	74.5	81.6	81.8	.572
Dietary Folate Equivalents (mcg)	600	23.4	17.1	20.0	.692
Vitamin B_{12} (mcg)	2.6	53.2	46.1	60.0	.285
Calcium (mg)	1000	97.9	96.1	96.4	.855
Phosphorus (mg)	700	55.3	50.0	58.2	.635
Magnesium (mcg)	350	97.9	97.4	98.2	.951
Iron (mg)	27	17.0	17.1	20.0	.895
Zinc (mg)	11	91.5	92.1	92.7	.973
Copper (mcg)	1000	12.8	10.5	20.0	.292
Manganese (mg)	2.0^	6.4	6.6	16.4	.118
Selenium (mcg)	60	40.5	19.4	25.5	.104
Sodium (g)	1.5^	21.3	11.8	20.0	.300
Potassium (mg)	4700^	100.0	98.7	100.0	.509
Choline (mcg)	450^	89.4	96.1	94.5	.314
Linoleic Acid (g)	13^	66.0	63.2	67.3	.880
Alpha-Linolenic Acid (g)	1.4^	78.7	73.7	80.0	.661
# of micronutrients below RDA	--	16.3 (4.3)	15.8 (3.8)	16.5 (4.4)	.564

Recommended Dietary Allowance (RDA) for pregnant women, aged 19–30, used for: vitamin A, C, D, E, thiamin, riboflavin, niacin, vitamin B_6, folate, vitamin B_{12}, calcium, phosphorus, magnesium, iron, zinc, and selenium.

Adequate Intake for pregnant women, aged 19-30, used for: pantothenic acid, manganese, sodium, potassium, choline, linoleic acid, alpha-linolenic acid.

^ denotes Adequate Intake

[1] Retinol Activity Equivalents

[2] Niacin Equivalents (1 niacin equivalent = 1 mg of Niacin or 60 mg of tryptophan).

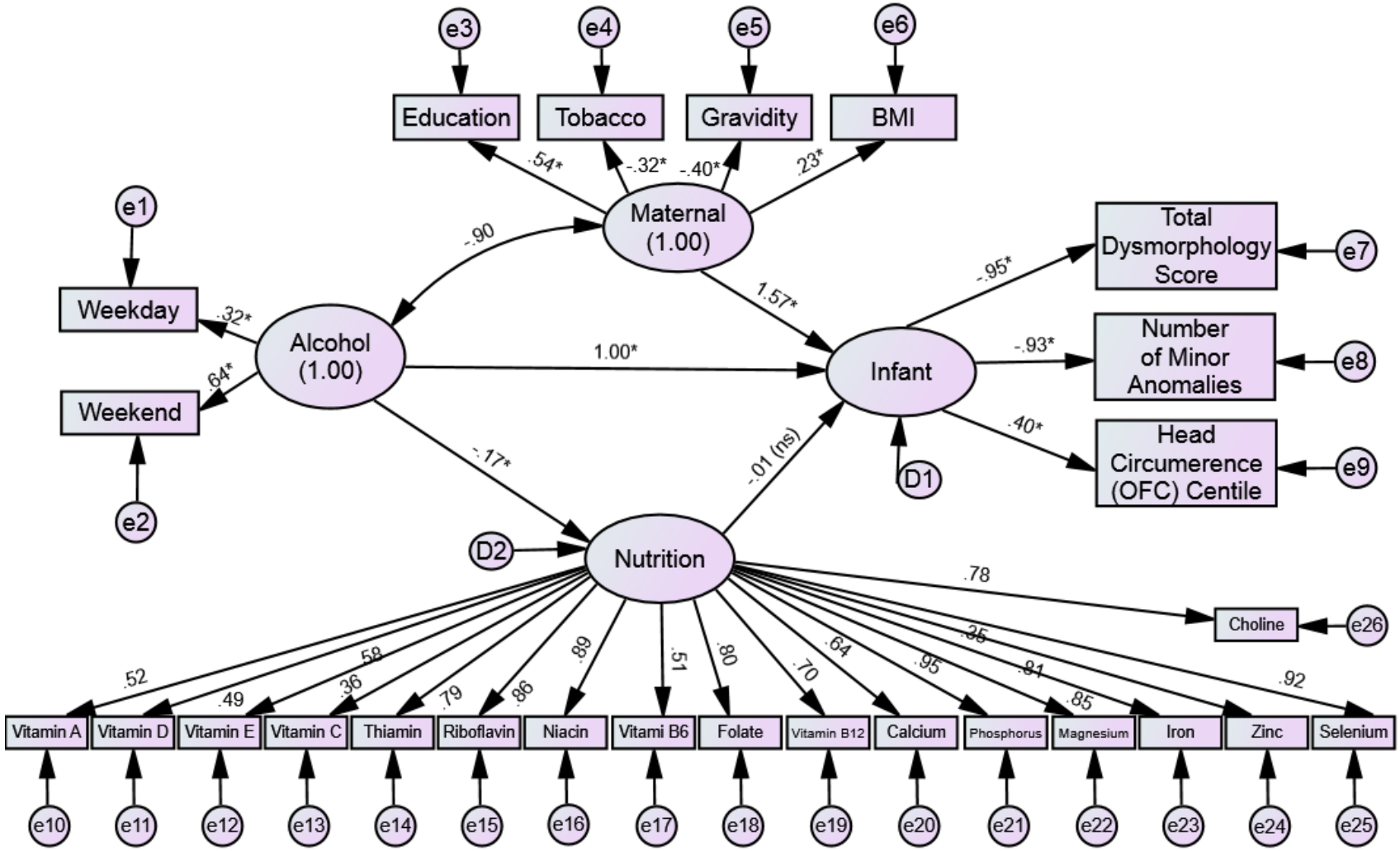

Figure C.1 Structural Equation Model Predicting Infant Physical Outcomes at 6 Weeks of Age
CFI=.777, RMSE = .115

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