



University of Venda

**RELATIONSHIP OF FEEDING PRACTICES, ANTHROPOMETRIC  
STATUS AND GUT FUNCTION IN INFANTS IN DZIMAULI  
COMMUNITY IN SOUTH AFRICA**

**A mini dissertation submitted to the Department of Nutrition in partial fulfilment of the  
requirements for the award of Master of Science Degree in Public Nutrition, University  
of Venda.**

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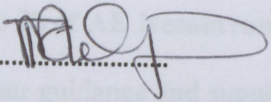
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## DECLARATION

I, **TJALE CLOUPAS MAHOPO** (Student NO 11551206), hereby declare that the dissertation, Titled “**Relationship of feeding practices, anthropometric status and gut function in infants in Dzimauli community in South Africa**”, submitted for the Master of Public Nutrition Degree at the University of Venda, has not been submitted previously for a degree at this or any other university. It is my own work in design and execution and all reference material contained therein has been duly acknowledged.

Signature.....



Date.....

08 May 14

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- > Charis Missionary Church members, these are the fruits of your prayers.

## DEDICATION

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2011-12	Malnutrition and Enteric Diseases Study
2015	Millennium Development Goals
2019	National Centre for Health Statistics
2012	National Food Consumption Survey
2007	Pubert Anamniotic Detection
2010	The American Health Organization
2011	Prevention of Mother to Child Transmission
PI	Principal Investigator
2014	South African Demographic Household Survey
SAVACG	South African Vitamin A Consultative Group
SPSS	Statistical Package for Social Science
UNICEF	United Nations Children's Fund
WAZ	Weight-for-age z-scores
WHO	World Health Organization
WHZ	Weight-for-height z-scores

## ABBREVIATIONS

BDHS	Bangladesh Demographic and Health Survey
BFHI	Baby-Friendly Hospital Initiative
DOH	Department of Health
DHS	Demographic Health Survey
FFQ	Food Frequency questionnaire
HAZ	Height- for- age z-scores
IRB	Institutional Review Board
LM	Lactulose:Mannitol
MAL-ED	Malnutrition and Enteric Diseases Study
MDG	Millennium Development Goals
NCHS	National Centre for Health Statistics
NFCS	National Food Consumption Survey
PAD	Pulsed Amperometric Detection
PAHO	Pan American Health Organization
PMTCT	Prevention of Mother to Child Transmission
PI	Principal Investigator
SADHS	South African Demographic Household Survey
SAVACG	South African Vitamin A Consultative Group
SPSS	Statistical Package for Social Science
UNICEF	United Nations Children's Fund
WAZ	Weight-for-age z-scores
WHO	World Health organization
WHZ	Weight-for-height z-scores

## DEFINITION OF TERMS

### **Anthropometric status**

Anthropometric status in this study refers to growth status of infants as determined by WHZ (Wasting), WAZ (underweight) and HAZ (stunting) indicators.

### **Complementary feeding**

Complementary feeding refers to age-appropriate, adequate and safe solid or semi-solid food in addition to breast milk or a breast milk substitute that the child receives (WHO, 2008).

### **Exclusive breastfeeding**

Exclusive breastfeeding refers to the infant's sole source of nutrition as breast milk with the complete absence of water, herbal tea or foods, except vitamin/mineral drops, syrups and medicine preparations (WHO, 2008).

### **Formula feeding**

Formula feeding refers to an act of giving a child who is not receiving any breast milk a suitable breast-milk substitute. The following terms: replacement feeding, non-breastfeeding or bottle feeding, are also used in the literature to indicate formula feeding (WHO, 2009).

### **Gut damage**

Gut damage involves the motility disorder in the GIT, alteration of the barrier function (increased permeability of the gut) and decreased absorption capacity of nutrients to the body (Berger et al., 2007).

### **Gut function**

Gut function, also referred to as intestinal permeability, represents a critical balance between absorption of nutritional agents and protection from pathogens (Taylor et al., 2010).

### **Infant feeding practices**

Infant feeding practices refer to complementary feeding, exclusive breastfeeding, formula feeding and predominant breastfeeding.

### **Intestinal permeability**

See gut function.

### **Lactulose**

Lactulose refers to a disaccharide that is not absorbed by healthy enterocytes (intestinal absorptive cells). The substantial absorption and consequent urinary excretion indicate damage to the integrity of the intestinal epithelium (Paroni et al., 2006).

### **Lactulose:Mannitol test**

Lactulose: Mannitol test is a quantitative assay for directly measuring the ability of two non-metabolised sugar molecules, mannitol and lactulose, to permeate the intestinal mucosa (Paroni et al., 2006).

### **Mannitol**

Mannitol refers to a monosaccharide that is absorbed passively by healthy enterocytes. The level of its absorption as detected from urinary excretion provides estimates of the functional surface area of intestinal epithelium (Paroni et al., 2006).

### **Mixed Feeding**

Mixed feeding refers to partial breastfeeding (WHO, 1996), giving the baby some breast milk regardless of how much is given and some artificial feeds, either milk or cereal or other foods.

### **Permeability value**

The permeability value is usually the maximum permeability or the maximum relative permeability (Lunn, 2000).

### **Predominant Breastfeeding**

Predominant breastfeeding refers to the feeding whereby the infant's predominant source of nutrition is breast milk accompanied by other liquids such as water or water-based drinks (including sweetened or flavoured water, teas, infusions and so on), fruit juice, oral

rehydration salts ORS) drop and syrup forms of vitamins, minerals, and medicines (WHO, 2008).

## **Weaning**

Weaning is a process from the introduction of the first foods until complete cessation of breastfeeding (Becquet et al., 2006).

## **Weaning food**

Weaning foods refer to all solid foods and/or any breast milk substitutes such as infant formula (Becquet et al., 2006).

## ABSTRACT

**Background and objectives:** Inappropriate feeding practices affect gut function in infants by damaging the mucosa of the small intestine. Feeding practices such as formula feeding and early introduction of weaning food can have profound consequences on growth, development, and survival chances of infants and young children. The purpose of this study was to determine the relationship between feeding practices and gut function in infants in Dzimauli community in South Africa.

*Key words: Feeding practices, gut function, lactulose:Mannitol, anthropometric status*

**Methods:** A prospective birth cohort study was carried out in Dzimauli community in Limpopo Province, South Africa. One hundred and thirty three children were recruited. Length and weight were measured monthly for the duration of 12 months for anthropometric assessment. Data on feeding practices was collected twice weekly using the 24 hour recall methods. Lactulose: mannitol data was obtained at 3, 6 and 9 months intervals. Bivariate analysis was used to correlate the relationships.

**Result:** Although almost all infants (92.5%) 123/133 were breastfed, none were breastfed exclusively for six months. On average, exclusively breastfeeding was only for 21 days. Water and formula milk were the first foods introduced to infants. Intestinal permeability was mostly poor at three months (67.6%) but improved with time as shown by 43.5% by month nine. No significant relationship was observed between any forms of feeding in exception of feeding colostrum which correlated positively with intestinal permeability. Children who were introduced to water and semi solids before three months had poorer anthropometric status at months 6 and 9. Mother's age had a negative influence on introduction of solid foods as older mothers introduced solids food earlier.

**Conclusion:** Poor trends of infant feeding still prevail in Dzimauli community shown in the current study. Infants fed colostrum showed better intestinal permeability as compared to their counterpart. Gut function was not associated with any of the reported feeding practices. Stunting was also observed as one of the challenges in the current study. Health education campaigns focusing on mothers to exclusively breastfeed up to six months in Dzimauli are needed. Studies to discover why mothers still introduce non-breast milk food earlier to their infants need to be conducted.

**Key words:** Feeding practices, gut function, lactulose:Mannitol, anthropometric status

## CHAPTER ONE: INTRODUCTION

### 1.1 Background and motivation of the study

The feeding practices of young children less than two years of age have a direct effect on their nutritional status (World Health Organisation (WHO), 2008a). Such practices may ultimately impact negatively on the child's quality of life with serious implications on the child's chances of survival. Infant feeding is a composite of behaviours that exist in a complex environment (Raphael, 1984). It is also a process that occurs over time and involves a range of behaviours including breastfeeding, formula feeding, introduction of supplementary foods and weaning practices. Inappropriate feeding practices such as bottle feeding and early introduction of weaning foods can have profound consequences on growth, development and survival of infants and children, particularly in developing countries (Butte et al., 2000; Kramer & Kakuma, 2001).

Pelletier et al. (1993) reported that inadequate feeding practices from birth through two years of age are the underlying causes of over half of childhood deaths from diseases. WHO (2008) reported that an estimated 9.5 million children died before their fifth birthday in 2006. Two thirds of these deaths occurred in the first year of life. Such death cases could be due to improper timing (too early or too late) introduction of complementary foods that are often nutritionally inadequate and unsafe (WHO, 2010). Although WHO/United Nations Children's Fund (UNICEF) (2003) recommends exclusive breastfeeding for the first 6 months of life, followed by complementary feeding with continued breastfeeding, it is estimated that only 34.8% of infants worldwide are exclusively breastfed for the first 6 months of life mostly in Southeast Asia (WHO, 2008a). Marques et al. (2001) reported that exclusive breastfeeding in Brazil is rarely

practised and often takes relatively short period of time as mothers tend to introduce complementary foods before the recommended time.

In Sri Lanka, almost 100% of mothers initiate breastfeeding (Agampodi et al., 2007) within 24 hours of birth. However, exclusive breastfeeding of up to four months was practised by 61.6% of mothers in Sri Lanka (Agampodi et al., 2007). In a study in Colombo city, Sri Lanka, Bundusena (2003) found that none of the participants practised exclusive breastfeeding up to 6 months. Furthermore, low rate of exclusive breastfeeding and increased inappropriate feeding practices (Agampodi et al., 2007) have been reported in Sri Lanka recently. Poor breastfeeding and complementary feeding practices are widespread worldwide posing risk to infants' growth and development of infants.

Children aged six to twenty three months and those living in rural areas are the most vulnerable to childhood malnutrition in South Africa (South African Vitamin A Consultative Group (SAVCG), 1995). South African Demographic Household Survey (SADHS, 2003) indicated that only 8.3% of infants below 6 months of age are exclusively breastfed. These were also supported by Mamabolo et al. (2004) and Mushaphi et al. (2008) who found that in the rural regions of Limpopo Province of South Africa infants are breastfed for longer periods, with more than 80% breastfed up to ninth month. However, exclusive breastfeeding during the first three months was uncommon. Mothers introduced other foods at an early age, with 56% of infants receiving some form of food by the end of the first month. The introduction of common non-breast milk foods comprised maize meal porridge and sorghum porridge (*mabella*), as well as teas and herbal drinks (Mamabolo et al., 2004).

Mushaphi et al. (2008) reported that the common early introduction of *tshiunza* (a traditional porridge) before the infants gut can digest foods other than breast milk interferes with exclusive breastfeeding and can severely reduce the positive effect of breastfeeding. It has been shown that poor infants feeding practises have negative impact on absorption and gut function in infants (Lunn et al., 1991). Moreover, growth faltering is reported to be common among infants weaned too early, starting at about three months of age, a period coinciding with the introduction of supplementary foods (Lunn et al., 1991; Goto et al., 2002; Goto et al., 2009).

Goto et al. (2002) indicated that the association between growth and gut damage is of great importance when determining the mechanism underlying child malnutrition and ill-health in less developed countries. Gut damage involves motility disorder, alteration of the barrier function (increased permeability) and decreased absorption capacity of nutrient to the body (Berger et al., 2007). In an earlier study, Goto et al. (2009) showed that infants are exposed to high level of infections associated with gut damage and growth faltering caused by inappropriate infants feeding practices and poor standards of hygiene. Solomons (2007) indicated that infectious diseases along with malnutrition are the leading causes of infant's mortality in African countries.

The present study was aimed at investigating the relationship of feeding practices, anthropometric status and gut function in infants to determine proper intervention and recommendation for improved infants and child feeding habits, and subsequent improvement in growth pattern. This study was part of the birth cohort project, Malnutrition and Enteric Diseases (MAL-ED) Study, in Dzimauli Community undertaken by the University of Venda in collaboration with University of Virginia.

## 1.2 Aim of the study

The aim of this study was to determine relationship of infant feeding practices, anthropometric status and gut function as measured by the lactulose: mannitol (LM) ratio in Dzimauli community in South Africa.

## 1.3 Objectives of the study

The objectives of the study were:

1.3.1 To determine the feeding practices of infants in Dzimauli community.

1.3.2 To determine gut (intestinal) permeability of infants in Dzimauli community.

1.3.3 To determine the association between feeding practices and gut function.

1.3.4 To determine the relationship between feeding practices, anthropometric status, and gut function in infants.

## 1.4 Research hypothesis

The study hypothesises that, infants introduced to non-breastmilk foods earlier than 6 months of age will have LM ratio of above 0.0864 (poor intestinal permeability) than those introduced in the recommended time.

## 1.5 Problem statement

The South African Millennium Development Goals (MDGs) report of 2005 indicated that under-five mortality rate has remained relatively constant since the 1998 estimates with slight decreases of 0.5% and 0.3% for infants and under-five mortality respectively. According to Lopez et al. (2000), half of the deaths from diarrheal disorders occur in children in the first year of life due to

poor breastfeeding practices. In South Africa, it is estimated that diarrheal disorders are the primary cause of death in children younger than five years of age leading to about 160-200 deaths per day (SADHS, 2003). Inappropriate feeding practices affect infants gut function by damaging the small intestinal mucosa through introduction of diarrheal pathogens such as *E.coli* and rotavirus in contaminated foods and water (Goto et al., 1999; Goto et al., 2002). Children with diarrhea present a greater LM ratio compared to children without diarrhea. Previous studies on child feeding practices conducted in the Limpopo Province in particular (Mamabolo et al., 2004; Mushaphi et al., 2008) indicated concerns about employed feeding practices. However, there is limited data on whether these feeding practices do affect gut function and permeability among young children in South Africa.

### **1.6 Significance of the study**

The study identified feeding practices that have an effect on gut function of infants in a rural set up in South Africa. The study provides additional information required for policy development and intervention studies to support child growth and development.

### **1.7 Dissertation outline**

Chapter one deals with the introduction of the study, the aim, objectives, hypothesis, problem statement as well as the significance of the study. Chapter two is the review of the literature pertaining to infant feeding practices, gut function and anthropometric assessments of infants including associations between these parameters. Chapter three elaborates on the methods in which information was obtained from the informants and infants. In chapter four, the results of the study are presented, analysed and interpreted. Chapter five and chapter 6 present discussions of the findings and conclusion with of the study recommendations respectively.

## CHAPTER TWO: LITERATURE REVIEW

### 2.1 Overview

This section introduces literature used to support the arguments made in the study about the feeding practices, gut function and growth status of infants. Relationships between feeding practices, gut functions and growth status of infants are also highlighted in this chapter.

### 2.2 Feeding practices

Adequate nutrition during infancy and early childhood is essential to ensure growth, health and development of children to their full potential. Black et al. (2008) reported that poor nutrition increases the risk of illness, and is responsible, directly or indirectly for the 9.5 million deaths that occurred in 2006 in children who are less than five years of age. Improved breastfeeding practices can contribute significantly to the achievement of the 4<sup>th</sup> goal of the Millennium Development Goals (WHO, 2002; 2008a). It was reported that optimal infants feeding practices rank among the most effective interventions to improve child health (WHO, 2002). Feeding practices are discussed below including breastfeeding practices, formula feeding and complementary feeding.

#### 2.2.1 Breastfeeding practices

Poor breastfeeding practices are still common both in developing and developed countries (WHO, 2008). Globally, 39% of children are exclusively breastfed for less than 6 months. Discussions on breastfeeding practices include breastfeeding initiation, exclusively breastfeeding and timing of the introduction of non-breastmilk foods with continued breastfeeding for two years and beyond. Black et al. (2008) indicated that early initiation of breastfeeding and

exclusive breastfeeding play a role in protecting infants against diarrhoeal diseases and reducing mortality among many infants in developing countries.

### 2.2.1.1 *Initiation of breastfeeding*

Initiation of breastfeeding within one hour after birth is highly recommended (WHO, 2008). The baby's suckling reflex is strong, and the baby is more alert half an hour after birth hence, an ideal time to initiate breastfeeding (Widstrom et al., 1990). Widstrom et al. (1990) recommends early breastfeeding for its potential to enhance bonding, improved chances of breastfeeding successes and its potential in lengthening the duration of breastfeeding. Delay in early initiation of breastfeeding (within one hour after delivery) increases the risk of death in neonatal stages (Edmond et al., 2007). Despite the benefits of early initiation of breastfeeding, most mothers do not initiate breast feeding even within 24 hours of birth due to lack of awareness (WHO, 2008a).

In South Africa, SADHS (2003) report indicates that although about 87% of infants were breastfed at least sometimes approximately only 45% of mothers initiated breastfeeding and they did so later than one hour. Limpopo Province showed a little better performance than national practices, 96% of mothers were reported to have initiated breastfeeding with 58% of them having initiated within the first hour and 42% within the first day.

### 2.2.1.2 *Exclusive breastfeeding*

The term exclusive breastfeeding is used when the infant's sole source of nutrition as breast milk with the possible exception of medicinal supplements Department of Health/United Nations Children's Funds (DOH/UNICEF, 2000). Current recommendations are that infants should be exclusively breastfed for the first 6 months of life (Greiner, 2001; WHO, 2007). Exclusive

breastfeeding remains uncommon in many countries (Scott et al., 2009; Doherty et al., 2012). There is quite a very small number of infants worldwide exclusively breastfed at the age of three months and older (WHO, 2009). According to Agampodi et al. (2007), similar findings were reported with few mothers (15.4%) breastfeeding for less recommended period. In a study conducted in Bangladesh, National Institute of Population Research and Training (NIPORT, 2007), mothers did breastfeed their infants, but exclusive breastfeeding was not widely practised. Furthermore, almost 50% of infants aged 2-3 months were exclusively breastfed and with 7% of infants exclusively breastfed at 6 months (NIPORT, 2007).

Exclusive breastfeeding in the early months of life is correlated strongly with increased infants' survival and lowered risk of illness, particularly from diarrheal diseases. Infants who are not breastfed in the first month of life may be as much as 25 times more likely to die than infants who are exclusively breastfed (WHO, 2007). Non-exclusive breastfeeding implies early introduction of non-breastmilk foods and therefore mixed feeding.

#### *2.2.1.3 Introduction of non-breastmilk foods*

Becquet et al. (2006) reported early introduction of non-breastmilk foods and/or complimentary (mixed) feeding at 4 months of age with only 8% of the infants being exclusively breastfed. It has been indicated that 65% of mothers gave something else other than breastmilk (pre-lacteal) in the first three days of life (NIPS and MI, 2008), a practice that is discouraged. This feeding type inhibits breastfeeding and introduces contaminants in infants. According to NFCS-FB-I (2007) breastfeeding is supplemented with other liquids earlier in South Africa. In other words, predominant breastfeeding is common in South Africa. Doherty et al. (2012) reported that early

cessation of breastfeeding is common amongst both HIV-negative and positive women in South Africa.

Madiba and Letsoalo (2013) indicated that in South Africa, mothers initiate mixed feeding as early as first months after birth. Moreover, mothers who do not know their HIV status showed a recurring trend of introducing other foods to their infants earlier as compared to mothers who knew their HIV status. Mother's perception of insufficient breastmilk were also reported as one of the factors that hinder compliance to recommended feeding practices in infants (Doherty et al., 2012). Similar findings were reported in South Africa (Kruger & Gericke, 2002). A study in the Gauteng province also showed that mothers believed that breastmilk alone cannot satisfy the child's hunger (MacIntyre et al., 2005). This belief of parents that breastmilk alone is inadequate for the growth and survival of the baby leading to early introduction of breastmilk substitutes and supplementation is reported in other developing countries such as Nigeria as well (Nwankwo & Brieger, 2002). This was also indicated in Australia where infants were introduced to solids before 4 months. Mothers reported that their infants either needed solids or they were ready for solids (Scott et al., 2009).

The Baby Friendly Hospital Initiative review indicated the main reasons provided by mothers for the early introduction of complementary foods as perceived inadequate production of breast milk and the belief that breast milk alone was not enough to satisfy the infants. The timing of its introduction is an individual decision however, six months of exclusive breastfeeding is recommended for most healthy term infants (WHO, 2008).

### 2.2.2 Formula feeding

Formula feeding refers to an act of giving a child who is not receiving any breast milk a suitable breast-milk substitute (WHO, 2008b). Formula feeding during the first 6 months of life is discouraged because it deprives infants of the disease prevention and nutritious content of breastmilk. Without the external immune support provided by breastmilk, infants are effectively immune compromised (Riordan, 1999). This increases the risk of formula-fed infants developing diarrhoea and the risk that the illness may become severe. The major concerns of formula feeding are hygiene and the use of feeds which are either too dilute, with the associated risks of under nutrition, or too concentrated, possibly resulting in hypernatraemic dehydration and over nutrition in infants (Egemen et al., 2002). This is also reflected in the potential of bottle feeding resulting in an increasing morbidity because of unsafe water and preparation facilities used at home (Mihirshahi et al., 2010).

Some of the factors reported to contribute towards formula feeding include the mothers' employment status and education level. Sanarath et al. (2010) found that in Sri Lanka, working mothers were the ones who mostly gave their infants formula when compared to those who were not working. They also reported that mothers with lower education breastfed their infants for longer periods and delayed the introduction of non-breastmilk.

### 2.2.3 Complementary feeding

The inclusion of any feeding in addition to breast milk into the diet constitutes complementary feeding (WHO, 2010). Complementary food is needed when breastmilk (or infants formula) alone is no longer sufficient for both nutritional and developmental reasons (Przyrembel, 2012). Inappropriate introduction of complementary foods can be disadvantageous because it reduces

the duration of breastfeeding. Furthermore, it interferes with the absorption of important nutrients from breastmilk and it increases the risks of contamination and allergic reactions (Saldiva et al., 2007). Any liquids given other than giving breast to the child should be fed by cup rather than by bottle (WHO, 2008). Feeding-bottles with artificial nipples and pacifiers may cause nipple confusion and infants may refuse to breastfeed after their use. Feeding-bottles are more difficult to keep clean than cups, and the ingestion of pathogens can lead to illnesses and even death. Pacifiers can also easily become contaminated leading to infections (WHO, 2010).

Nutritious foods in addition to breast milk from the age of 6 months are necessary for babies. Locally available and affordable foods that enrich the baby's diet with additional energy and micronutrients should be offered (Saldiva et al., 2007). Such food can either be taken being soft or mashed and in small quantities several times a day. These complementary foods should gradually increase in amount and frequency as the baby grows. Breastfeeding, on demand, should continue until the age of two years or beyond (Przyrembel, 2012). Late introduction of complementary foods is also disadvantageous because children's energy requirements will not be met resulting in deceleration in growth and increased risks of malnutrition and micronutrient deficiencies (Przyrembel, 2012).

Appropriate complementary feeding helps promote growth, prevent stunting, and increase a child's chances for a healthy, productive life as an adult. Improving complementary feeding requires a combination of strategies (Saldiva et al., 2007). Strategies for achieving adequate nutrition for infants and young children in modern times must address the challenges of meeting nutrients needs (Dewey, 2013).

A study in Pakistan (Shamim, 2005) indicated that poor weaning practices including underfeeding and lack of nutritionally balanced diet are major problems especially during the second 6 months of an infants' life in low-socioeconomic groups. Complementary foods are introduced within the seventh month of life, with the exception of semi-solid food and infant's formula that are introduced early than the recommended time (Bacquet et al., 2006).

Studies on infants feeding (Kruger & Gericke, 2002; Mamabolo et al., 2004; Mushaphi et al., 2008) showed that complementary feeding starts very early in South Africa, at an average age of two to three months of age. According to the SADHS (2003), approximately 70% of children are reported to have received complementary feeds before the age of 6 months.

Weaning is a process from the introduction of the first foods until complete cessation of breastfeeding (Becquet et al., 2006). It is recommended that weaning process start at 6 months. At 6 months, infants iron stores become depleted and the infant iron requirement remains higher. Badenhorst (1993) indicated that introduction and selection of appropriate weaning food is critical at this stage in life. Weaning foods refer to all solid foods and/or any breast milk substitutes such as infant's formula (Becquet et al., 2006). According to Coutsoudis et al. (1999) early introduction of food may cause damage to the infants gut. Furthermore, infants receiving mixed feeding are more likely to have poor gut function than those breastfed exclusively. In South Africa, infants are introduced to a mixed family diet by the age of 7-9 months with reduced frequencies of breastfeeding (Steyn et al., 1993; Coutsoudis et al., 1999; Kruger & Gericke 2002; Mamabolo et al., 2004; Mushaphi et al., 2008).

A study conducted nationally in Pakistan indicated that commercial infant's formula and other milk (fresh, tinned, and powdered cow or other animal milk) are introduced as early as the first month after birth (NIPS and MI, 2008). Steyn et al. (1993) indicated that infants in the rural Limpopo Province are weaned on a refined maize meal soft porridge, followed brown bread dipped tea with sugar as the first semi-solid foods. It is a common practice in South Africa to introduce maize meal soft porridge as the first weaning food (Faber et al., 2004). However, a study conducted by Kruger and Gericke (2002) indicated that commercial infants cereals were used more often in urban than in rural areas of low socioeconomic status. Faber (2004) reported that various energy rich foods such as sugar, margarine and peanut butter, were also added to the porridge of most infants, thereby increasing the energy density. Faber and Benade (2007) further indicated that, foods introduced to children was found to be bulky with low-nutrient density and limited variety of food also given that predisposes the children to low micronutrient intake.

In South Africa animal products are not widely used in infants feeding (Faber & Benade, 2001; Kruger & Gericke, 2002). Cow and goat milk were being perceived as having unsuitable nutritional composition and sometimes being used unpasteurised (Kruger & Gericke, 2002). In Mpumalanga, a province with similar characteristic with Limpopo Province, mashed pumpkin was given to infants as a weaning food for its easier digestibility (Ukpe et al., 2009).

### **2.3 Gut function**

The small intestine is a major organ performing digestion and absorption of nutrients. It allows passage of digested nutrients into the blood stream. Moreover, it prevents entry of toxic substances and undigested food macromolecules from getting into the blood stream (Jayalakshmi et al., 2009). The immaturity of young children's gastrointestinal tract, neuromuscular coordination, and immunological function limits the types of foods that children are able to

consume. This limitation exposes them to an elevated risk of food-borne infections and food allergies (Brown, 2007). Normal intestinal permeability is required for physiological function of the small intestine hence, food malabsorption is a significant cause of malnutrition resulting from abnormal function of the small intestinal mucosa (Lunn, 2000).

Mucosal integrity loss is likely to permit increased absorption of lactulose because of the poor intercellular tight junctions. Furthermore, loss of absorptive surface area as a result of associated villous shortening would be expected to reduce total absorption of mannitol (Reyes et al., 2006). Mucosal function can be assessed using the dual-sugar intestinal permeability test, which is used in clinical investigation of small bowel disease (Lunn, 1991). Dual-sugar intestinal permeability test is valuable at community level because of its non-invasive property.

The methods used to test intestinal permeability depend on the excretion of sugars in the small intestine (Black, 1984). LM ratio measurement is a proven, effective index of intestinal permeability (Taylor et al., 2010). LM test ratios for investigation of mucosal damage have shown that food mal-absorption is associated with early growth retardation (Goto et al., 2002). According to Jayalakshmi et al. (2009) high intestinal permeability indicates damage to the small intestinal mucosa. Poor food mal-absorption had greater effect on infant's growth status (Lunn (2000). Moreover, infants indicated with low weight-for-age have a significantly increased LM ratio.

Most studies (Goto et al., 2002; Campbell et al., 2003; Lung, 2000) relate growth faltering to either food intake or infection. Lunn (2000) reported two ways that lead to growth faltering in children. First, partial villus atrophy results in a loss of enzymes that are required for the normal

digestion and absorption of food. Second, the mucosal damage compromises the barrier function of the small intestine, allowing the translocation of antigenic macromolecules into the mucosal tissues and into blood. In the study conducted by Campbell et al. (2003), it was reported that the mucosal damage was the cause of malnutrition in children rather the consequence. Furthermore, mucosal damage in children has been reported as one of the major contributors to growth impairment and malnutrition worldwide.

Taylor et al. (2010) indicated that, feeding infants with breastmilk promote intestinal closure earlier and more consistently than formula feeding during the first postnatal months. Poor nutritional practices such as giving water and solids before 6 months could have a direct influence on intestinal permeability of infants gut. According to Boaz et al. (2013) infants with concurrent malnutrition and diarrhoea always show poor gut performance indicating an increased or leaky gut.

## 2.4 Child growth

A child's growth is the most important indicator of health, measured by the length, weight and head circumference based on age standards (WHO, 2006). In developing countries, poor growth faltering of infants has been shown as a consequence of chronic or recurrent exposure to infection brought about by living in unhygienic and unsanitary environments (Campbell et al., 2003). However, infant feeding practices are also indicated as the determining factor of infants' weight and length gain during early postnatal life, particularly those in the first year of life (Simondom et al., 2001). Infant feeding practices are very often dependent on the mother's attitude and perception regarding breastfeeding and its nutritional value and satiety (Simondom et al., 2001).

Anthropometry is a practical and immediately applicable technique for assessing children's development patterns during the first years of life (WHO, 2006). An evaluation of the growth of children also provides useful insights into the nutrition and health situation of entire population groups. Anthropometric indicators are less accurate than clinical and biochemical techniques when it comes to assessing individual nutritional status (Gorstein & Akre, 1988). A number of factors have been shown to be associated with infant's growth faltering in developing countries (Kessler & Dawson, 1999) including suboptimal weaning and inappropriate complementary feeding (Shrimpton et al., 2001) as well as health factors relating to infectious diseases like diarrhoea (Assis et al., 2005).

Growth faltering is common in many developing countries starting at an early age with weight-for-age z-scores (WAZ) and weight-for-height z-scores (WHZ) falling from about 3 months of age until about 12 months of age (Shrimpton et al., 2001). Furthermore, the decline in height-for-age starts after three months and continues until about 24 months of age. In a Bangladesh national study, malnutrition was reported to be very high in children less than five years olds, 40.8% stunted, 47.1% underweight and 11.9% wasted (BDHS, 2007). Shrimpton et al. (2001) indicated that growth faltering is also a problem in children from 0 to 2 years of life in the developing countries. Similarly in Bangladesh, Goto et al. (2009) indicated that growth faltering was mostly reported at the ages of 3 to 4 months following the introduction of supplementary food. Lunn (2000) suggested that poor infants feeding can lead to infants losing weight due to the partial villus atrophy that reduces the absorption and digestion of lactose and probably other nutrients important for child growth and development.

The onset of stunting is often associated with the introduction of complementary feeding (Brown et al., 1995). In the rural Limpopo Province, Mamabolo et al. (2004) indicated that infants weaned earlier than 6 months had a poor growth status. South African researchers currently refer to the three national studies done, the SAVACG (1995), National Food Consumption Survey (NFCS) (Labadarios et al., 2000) and the National Food Consumption Survey-Fortification Baseline (2007), for reporting the anthropometric status of South African children. The three studies indicated that nutritional status can be assessed by dietary, anthropometric, biochemical and clinical methods (Faber and Wenhold, 2007). A number of key indicators are useful in determining the level of malnutrition in children.

## 2.5 Conclusion

Out of three ways used to feed infants, exclusive breastfeeding is the one that supports adequate growth (Onayade et al., 2004). Exclusive breastfeeding is portrayed in the literature as a factor that enables adequate gut function thus allowing intake of sufficient nutrients and energy by the infant's body leading to adequate growth and development. Conversely, formula and complementary feeding if improperly practices are believed to be practices that promote damage on the gut and therefore affecting its proper functioning. Infectious diseases are however also associated with gut damage and growth faltering (Goto et al., 2009). Therefore, both inappropriate feeding practices and poor standards of hygiene have been reported as some of the causes of poor gut function and growth faltering in infants (Solomons, 2007). Inappropriate feeding practices can also be a source of infection through bottle and pacifier uses (WHO, 2010).

## CHAPTER THREE: METHODOLOGY

### 3.1 Study design

Descriptive and exploratory study designs were used to determine the effect of feeding practices, gut function and growth indicators. Quantitative methods were used to collect and analyse data. A birth cohort was followed for one year to determine feeding practices, food intake, markers for gut function and anthropometric assessment. The LM test was used to measure gut permeability in infants.

### 3.2 Study area and population

The study was carried out in the Dzimauli community in the Mutale Municipality, about 47 km from Thohoyandou. It is a poor and underdeveloped rural area of the Limpopo province with poor sanitation and inadequate potable water supply. Housing is mainly of mud bricks. Terrain is hilly and it is surrounded by streams flowing towards the Mutale River. The inhabitants use the streams for domestic activities such as washing, cooking, drinking water and other household activities. Most of the households practise subsistence farming and grow various kinds of vegetables and fruits such as paw-paw, mango, banana, guava and lemons.

Figure 3.1 shows the study area. Nine settlements of the Rambuda community area were selected for the study. The settlements selected for the study were Tshandama, Pile, Tshapasha, Tshibvumo, Matshavhawe, Thongwe, Madadani, Mbahela and Baimoro. These villages are adjacent to each other and they are stretched to a distance of approximately 8 km from Tshandama to Baimoro. The study participants were infants born to women in the settlement of the study area. The estimated population size of the study area is approximately 8000 residents.

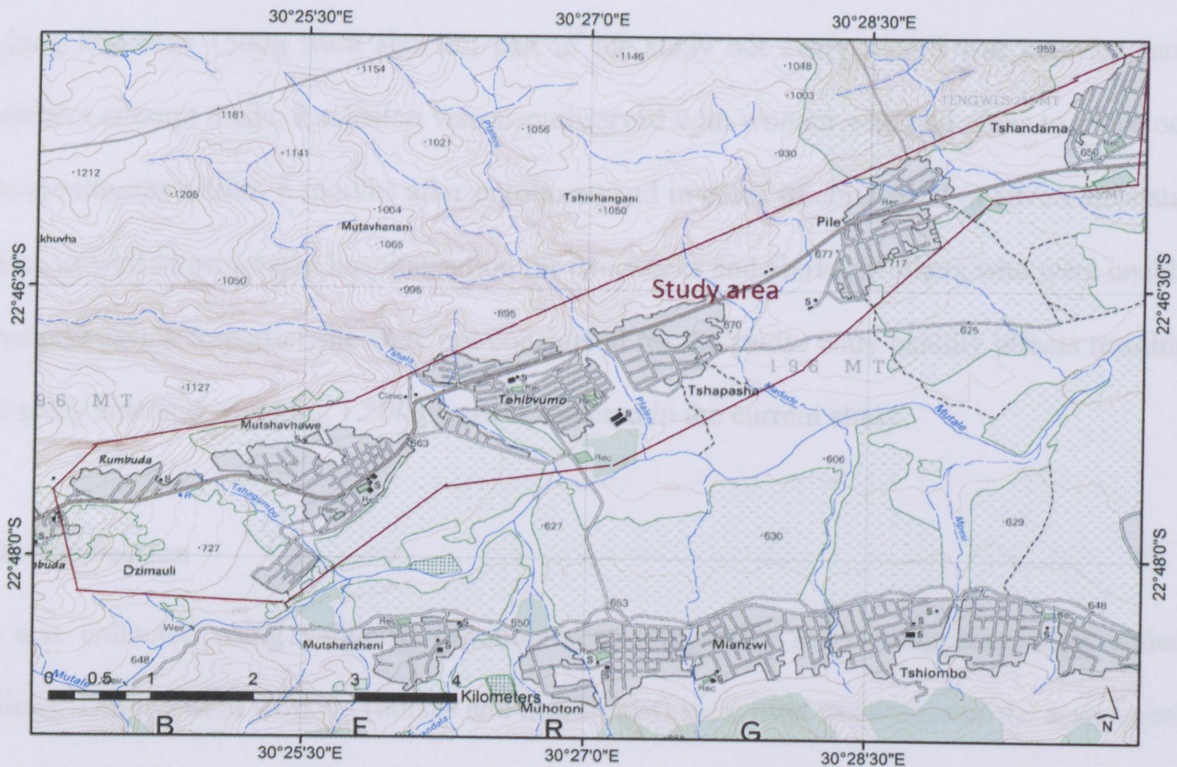


Figure 3.1. Map showing the study area

### 3.3 Sampling

All infants born at the area from October 2009 to May 2011 were approached and recruited to participate in the study. The participants were identified from the health centres, clinics, hospitals and community liaison officers. Pregnant women residing at Tshandama to Rambuda were approached and consented to be participants before they gave birth and enrolled within 17 days of child's birth. A sample size of 133 infants was used for the purpose of this study and followed up for a period of one year. All infant born from October 2009 and were one year at the end of May 2011 were included in the study for ease of data analysis preparation. Data was generated in different time points of follow up. Mother/caregivers provided data captured in case reporting forms.

Infants born to teenage mothers of less than sixteen years, twins and those born at the study area but not residing at Rambuda community were excluded from the study. Furthermore, infants weighing less than 1500g were also not part of the study but every record was kept for each recruitment attempt made. Exclusion was also observed with women who had plans to move out of the community within 6 months after enrolment and in cases of congenital or severe neonatal diseases as well as prolonged hospitalisation for two weeks and above. Mothers who were unable to give permission to participate were not considered for the study. Only healthy infants residing at the study side and weighing 1.500g were considered in the current study.

### **3.4 Data collection**

Data was collected using validated questionnaires (Appendix A-F) comprising close ended questions. Trained field workers (nurses and nutritionist) were used to administer the assessment forms. Local language (Tshivenda) was used to administer assessment forms to ensure understanding and accuracy of responses.

#### **3.4.1 Stages of data collection**

Once the study participants were enrolled in the study, on the day of enrolment, the study researcher began the baseline surveys on infant and mothers' demographics and infants' feeding practices from the previous 24 hours. The information on infants feeding practices was collected twice weekly by the study nurses and nutritionist for a period of one year. A monthly assessment form was administered comprising feeding practices and food frequencies until the child was 8 months old. For anthropometric measurements a record sheet was filled monthly from the age of nine months until the age of 12 months. At months 3, 6 and 9 urine samples were collected.

### 3.4.2 Instruments

The instruments were developed by the MAL-ED collaborators (University of Virginia, USA) site subcommittees including South Africa and adapted for the purpose of this study.

During recruitment, three assessment forms, namely Screening Form (Appendix A), Child Assessment Form (Appendix B), and Baseline Demographic Questionnaire (Appendix C) were used. The Child Assessment Form and Screening form were used to screen the mother and the infant's eligibility for the study. The Baseline Demographic Questionnaire used for the information of the mother, age, educational qualifications, marital status and parity were also determined during screening. Infant's early feeding practices were also determined at birth using the Child Assessment Form.

Surveillance assessment form (Appendix D) was used to assess the infant's general health. The form consisted of questions on child's current health status, gastrointestinal and respiratory illness questions. Structured infants feeding practices and a 24 hour recall assessment questions were also asked. Urine samples were collected using the Urine Collection Form (Appendix F) at the child age of 3, 6 and 9 months. Monthly Assessment Form (Appendix E) was also used to record weight, height and head of the infants from 1-12 month, while urine collection form was used record samples collected from the field.

### 3.4.3 Surveillance assessment

Data on child illness and intake of specific foods was collected twice weekly for twelve months from date of enrolment using Surveillance assessment form (Appendix D). Data on child illnesses and general health was not reported in this current study and was not used because it is beyond the purpose of this study. In addition a 24 hour recall on breastfeeding and food consumption practices were recorded. Questions on food included presence and frequency of breastfeeding, infant's formula, fruit juices and various liquids. Mothers were also asked if semi-solid food such as porridge or soft porridge, yoghurt, banana and fermented food were given to the infants.

### 3.4.4 Monthly assessment

Data on infants feeding patterns including breastfeeding frequencies, formula milk and animal feeds given to the child were recorded from months one to eight. Information on various liquids given to the infants such as water, tea, juices and home prepared solutions were collected. On monthly basis information on dietary diversity of semi-solids food pertaining to gut function was also collected. Intake of foods such as porridge, green leafy vegetable and foods made with beans, eggs, cheese and sugary food fruits such as bananas and tomatoes were also recorded. The infant's weight and length were assessed and recorded for a period of 12 months in the Monthly Assessment Form (Appendix E). All the measurements were taken twice and the average value was computed. The following procedures were considered during anthropometric measurements:

#### 3.4.4.1 Weight

Three persons weighed the infants using a digital scale once every month. The measurements were taken monthly between or  $\pm 2$  days of the infant's day of anniversary at the infant's home. The infants were put at the centre of the scale tray and weighed nude. One person maintained the safety of the infants and the other read the weight aloud while the third person wrote on the form. The infants were weighed twice to the nearest 10 g or 0.01 kg and an average value was recorded.

#### 3.4.4.2 Length

A measuring board, Seca 417 (baby measuring board, 0-100 cm), was used to measure the infant's recumbent length once every month from 0 to 12 months. The measuring board was placed on a flat surface and the infant was put down with the face up on the board with the crown of the head touching the headboard. Two persons weighed the infant with one maintaining the head position during measurement. The other one kept the infant's legs extended at the hips and knees and made him or her to remain flat on the board while adjusting the movable board against the heels. Two length measurements were taken and the average length was recorded to the nearest 0.1 cm.

#### 3.4.4.3 Urine collection

Urine sample was collected at 3, 6 and 9 months of age intervals for intestinal permeability using Urine collection form (Appendix F). Infants were given L: M solution as early as six in the morning to avoid mothers feeding the infants. Fieldworkers visited the household of the infants and collected the urine samples. Sterile water was used for the L:M solution preparation. An amount of 1.0 ml mannitol was added to water and mixed for about 15 minutes using magnetic

stir bag. A measured lactulose (5g) was added to the beaker and stirred for another 15 minutes to dissolve. L:M solution was aliquoted into labelled sterile vials according to the weight of each subject (at a dose of 2 ml/Kg of weight or a maximum 20 ml). Final concentration of 20 ml Lactulose: Mannitol = 1 g mannitol + 5 g lactulose was given to the infant for the assessment.

Infants were required to fast for at least two hours before ingesting the prepared L:M solution. Regular diet resumed 30 minutes after ingestion of the test solution. The infants emptied their bladders before ingesting the test sugar solution. A paediatric urine bag was used to collect urine samples for five hours attached to the infant's genitals. Urine sample was put in appropriate flasks containing one ml of chlorhexidine. The samples were therefore put in an ice cooler just after collection and transported to the University of Venda laboratory for analysis. Total urine volume was measured and recorded and a sample of 5 ml was stored in -80 °C. in a freezer at the University laboratory.

### **3.5 Training of field workers**

One week training for fieldworkers was arranged by the researcher (PI) Principal Investigator of the MALED project at the University of Venda. All assessment forms were in English however, field workers were trained in Venda. Field workers were grouped in groups of two to go through all the assessment forms and interview each other regarding the forms. The process continued for a week until all the fieldworkers were familiar with the assessment forms.

### **3.6 Pilot study**

The instrument was piloted before commencement of the research project. The pilot results were used for training of fieldworkers and to improve the understanding and accuracy of responses. A household with an infant aged 9 days was chosen for piloting in the same area. The infant did not

form part of the main study. Due to the extent of the training and credibility of the instrument one fieldworker was given an opportunity to interview a household during the pilot study to see if the forms were understood and the time each fieldworker will take for all the assessment in one household.

### 3.7 Validity and Reliability

The study was validated by using trained field workers from specific fields such as nutritionist and assistant nurses. Study nurses were used for the collection of urine samples in the study. Instruments were adopted from credible studies. Several instrument reviews were conducted by experts (Nutritionists) to ensure all instruments will collect the information intended to collect.

Weekly meetings and trainings were held with the fieldworker to go through the challenging forms and solve any problem they encounter with data collection. A pilot study was also done to test if the instrument was suitable for the study area and whether it would be able to measure what it was intended for. Local language (Tshivenda) was used to ensure that participants will understand the questions and will answer appropriately.

For reliability, various instruments were used in the study and same data was collected more than once. The surveillance form was administered twice on a weekly basis to capture all the necessary data about child illnesses and the twice weekly 24 hour recall. Food frequency questionnaire was administered once a month for the period of 1 to 8 months to accommodate the expanding diet as the child grows. Mothers were provided with feeding record sheets to record all the food given to the child to prevent or minimise guess reporting of food intake. The anthropometric measurements were taken twice and the mean value was computed to verify data

accuracy. Digital weight scale was used to measure weight and length board to measure recumbent length.

Frequency distributions were determined to describe demographic data as well as feeding. Quality was done assurance to ensure true reflection of responses given by the responded and measurement taken by the fieldworkers. Quality assurance was done by the researcher at randomly selected 5 to 10% of the sample every month. Researcher visited 5-10% of the assessment done by the fieldworkers on a monthly basis to check consistency of the measurement and procedures followed when doing anthropometric measurement. If the measurement had 4-5% difference the field worker will be requested to re measure the child in the presence of the researcher.

### **3.8 Ethical considerations**

This study was conducted within the birth cohort project (Mal-ED) at the University of Venda coordinated by the Department of Microbiology, in collaboration with the University of Virginia, USA. The study protocol received approval from the Health, Safety and Research Ethics Committee of the University of Venda (SMNS/09/MBY/004) and the Institute Review Board for Health Sciences Research of the University of Virginia (HSR#/4269). Permission to carry out the study was obtained from the Limpopo Provincial Department of Health and the Dzimauli authorities. Signed informed consent was obtained from all the participants prior to study engagement. Signed consent forms were kept under lock and kept by principal investigator (PI) of the Mal-ED South Africa.

### 3.9 Data analysis

Statistical Package for Social Science (SPSS), version 21, was used to analyse the data. Frequency distributions were determined to describe demographic data as well as feeding practices to indicate progressive transition of the infants from breastfeeding to complementary feeding. The infants were divided into groups of exclusive breastfeeding, formula feeding and complimentary feeding according to how they were fed.

Anthropometric measurements were analysed by calculating standard deviation from the median of the National Centre for Health Statistics/WHO references growth curves (z-scores) to determine weight-for-age, length-for-age and weight-for-length variables in order to describe the growth status of study children. The z-scores were calculated using computer software (WHO Anthro version 2.0). The association between feeding practices and gut function was determined using pearsons correlations coefficient. Feeding practices and anthropometry results were correlated with L:M ratio scores. The initiation age of various feeding practices were explored in relation to anthropometry indicators and concentration of L:M scores. The L:M permeability test is considered abnormal or positive for comparison purposes if the L:M ratio is  $\geq 0.0864$ . These values equal to normal mean plus two standard deviations as previously reported (Barboza et al., 1999). Urine specimens were used to assess the ratio of lactulose: mannitol excretion. The L:M test is considered a consistent and sensitive method to measure small intestinal epithelial area, paracellular and transcellular transport, damage and permeability (Berger et al., 2007). The ratio of lactulose:mannitol excretion in urine after administration of these non-metabolized sugars has been used to evaluate the extent of malabsorption and intestinal permeability disruption in several infections and nutritional diseases, including human immunodeficiency virus (HIV) infection.

L:M concentration was determined by high performance liquid chromatography (HPLC) employing pulsed amperometric detection (PAD). In the sugar absorption test, the urinary LM ratio was measured after oral ingestion of a solution of lactulose and mannitol. Using chromatographic peaks, the quantified mannitol and lactulose in infant's urine were used to calculate the percentage excreted in relation to the ingested amounts of sugar. The calculation was used to determine the lactulose-to-mannitol ratio (L/M).

Two-tailed Student *t* test was used for continuous and categorical variables in comparisons between the groups, using  $P < 0.05$  and  $P < 0.01$  for statistically significant difference. Two-sided significance tests were used throughout. For analysis of outcome variables, values of mean  $\pm$  SEM at 3, 6, 9 and 12 months were presented for both groups.

Table 4.1. Socio-demographic characteristics of children and mothers (n=133)

Socio-demographic variables	No.	%	Total
<b>CHILD CHARACTERISTICS</b>			
Gender			
Male	63	(47.4)	
Female	70	(52.6)	
Age groups at recruitment (in days)			
0-5	34	(25.6)	
6-10	41	(30.8)	
11-15	58	(43.6)	
<b>MOTHERS CHARACTERISTICS</b>			
Age groups			
15-19	4	(3.0)	
20-25	60	(45.1)	
26-35	46	(34.6)	
36-42	23	(17.3)	
Never use a child birth			257

#### 4.1 Socio-demographic characteristics

Table 4.1 shows the socio-demographic characteristics of the mothers and infants. A total of 133 infants and their mothers aged 16 to 42 years participated in the study. Just above half of the infants 52.6% (70/133) in the study were females. Almost half of the study participants (48.8%) 65/133 were in the age range of 19 to 25 years. All the mothers (100%) had at least one year of formal school training. Reasonably, about two thirds of the mothers (64.7%) 86/133 has completed 8 to 11 years of schooling and only few had tertiary education (11.3%) 15/133. There were about 40% (52/133) of single mothers at the time of enrolment and less than two percent (1.5%) 2/133 of the mothers were divorced. Age at first marriage ranged from 16 to 30 with 34.6% (46/133) of mothers married at the age 19-24 years. Mothers (58.6%) 78/133 had an average of 2 to 4 children at the time of enrolment in the study.

Table 4.1. Socio-demographic characteristics of children and mothers (n=133)

Socio-demographic variables	No.	%	Mean
<b>CHILD CHARACTERISTICS</b>			
<b>Child sex</b>			
Males	63	(47.4)	
Females	70	(52.6)	
<b>Age groups at recruitment (in days)</b>			
0-5	34	(25.6)	
6-10	41	(30.8)	
11-17	58	(43.6)	
<b>MOTHERS CHARACTERISTICS</b>			
<b>Age groups</b>			
16-18	9	(6.8)	
19-25	65	(48.8)	
26-35	46	(34.6)	
36-42	13	(9.80)	
*Mean age at child birth			25.7

Socio-demographic variables	No.	%	Mean
<b>Education level</b>			
Primary education	10	(7.5)	
Secondary education			
Grade 8-11	76	(57.1)	
Grade 12	32	(24.1)	
Tertiary	15	(11.3)	
<b>Marital status</b>			
Never married	52	(39.1)	
Married	79	(59.4)	
Divorced	2	(1.5)	
<u>Mean marital age (n=81)</u>			20.7
<b>Age at first marriage (in years)</b>			
16-18	20	(24.7)	
19-24	46	(56.8)	
25-30	15	(18.5)	
<b>Parity</b>			
1	50	(37.6)	
2-4	78	(58.7)	
≥ 5	5	(3.7)	

\* Mean age at child birth refers to the first-born child.

## 4.2 Infant Feeding Practices

Table 4. 2 show results of early feeding experiences of mothers in Dzimauli. Almost all (93%) infants in the study were breastfed and given colostrum. More than half of infants (55.6%) were breastfed within the first hour after birth, while 34.6% were breastfed later than the first hour but the same day. Quite few of the infants (2.3%) were breastfed after a day.

Results for infant feeding practices including, exclusive breastfeeding, predominant feeding, formula and complementary feeding, are presented below.

### 4.2.1 Exclusive Breastfeeding

Duration of exclusive breastfeeding is presented in Figure 2. Exclusive breastfeeding did not exceed three months of age. Less than half (48%) of infants were still breastfed exclusively by

the end of their first month of life. By the end of second month of age, very few of them, 22/123 (14.6%), were continued on exclusive breastfeeding and only 8.1% remained breastfed exclusively until three months of age.

Table 4.2. Early feeding experiences of mothers in Dzimauli (n=133)

Breastfeeding initiation	No.	(%)
<b>Ever breastfed</b>		
Yes	123	(92.5)
No	10	(7.50)
<b>Infants fed colostrum</b>		
Yes	123	(92.5)
No	10	(7.50)
<b>Time infants first breastfed (n=123)</b>		
Within one hour	74	(55.6)
1-24 hours	46	(34.6)
1-3 days	3	(2.30)

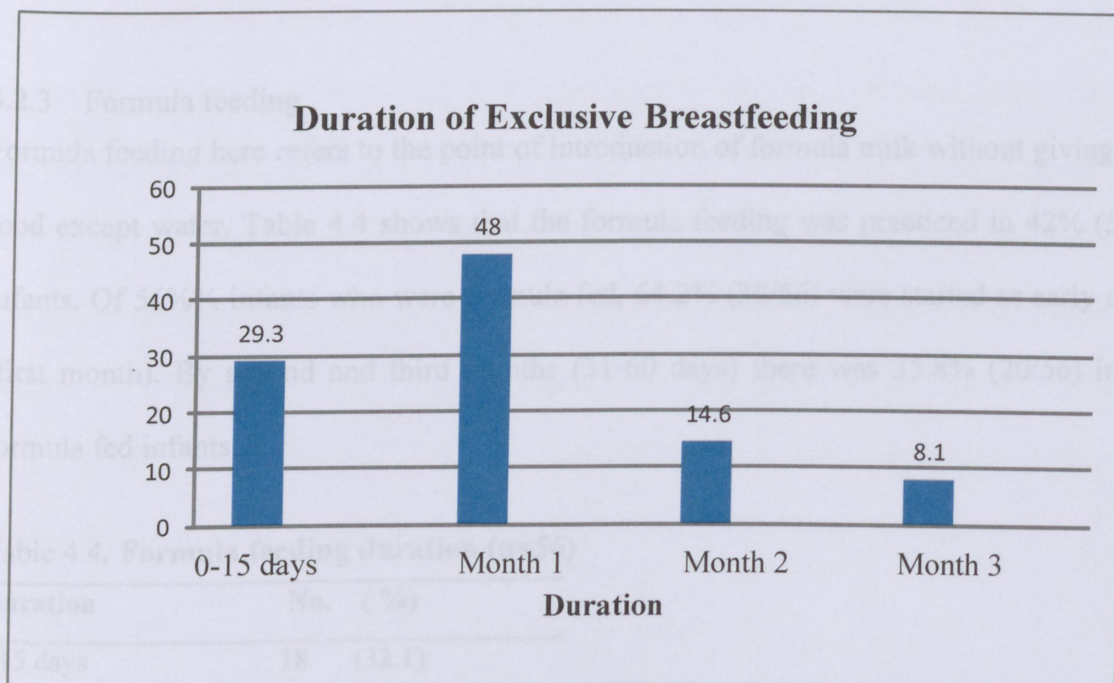


Figure 4.1 Duration of exclusive breastfeeding of infants in Dzimauli

#### 4.2.2 Predominant feeding and duration

Table 4.3 presents predominant feeding which is the practice of introduction of water while breastfeeding until other foods are introduced. The predominant feeding results indicate that 55% (73/133) of infants were given water with breast milk only up to 150 days of their lives. Of the predominantly fed children, about 6.8% (5/73) infants were introduced to water earlier than 15 days of life, 41.1% (30/73) between 16-30 days, 16.4% (12/73) in 61-90 days and less than ten percent of infants (6.8%) (5/73) were added to predominantly fed infants in 91-150 days.

Table 4.3. **Predominant feeding duration (n=73)**

Duration	No.	(%)
0-15 days	5	(6.80)
16-30 days	30	(41.1)
31-60 days	21	(28.8)
61-90 days	12	(16.5)
91-150 days	5	(6.80)

#### 4.2.3 Formula feeding

Formula feeding here refers to the point of introduction of formula milk without giving any other food except water. Table 4.4 shows that the formula feeding was practiced in 42% (56/133) of infants. Of 56%% infants who were formula fed, 64.2% (36/56) were started as early as 30 days (first month). By second and third months (31-60 days) there was 35.8% (20/56) increase of formula fed infants.

Table 4.4. **Formula feeding duration (n=56)**

Duration	No.	(%)
≤15 days	18	(32.1)
16-30 days	18	(32.1)
31-60 days	20	(35.8)

#### 4.2.4 Complementary feeding

Table 4.5 shows introduction of complementary foods in infants in terms of type and starting time. Foods were categorized according to semi-solid, tea, fruit juices, other liquids and grains. Semi-solids were given to 99.2% (131/133) of infants before the age of 6 months (180 days). The practice of giving semi-solid foods to infants was mostly common 64.4% (85/133) during the second and third months (60-90 days). Less than 2 % of children were introduced to solids during the first 15 days of life. Although tea was also given to infants in the first 6 months of age, it was mostly introduced after 6 months to 68.2% (75/110) of the children. Less than 10 % of infants were introduced to tea within the first 90 days of life.

Fruit juices were also introduced late (after 6 months) to 70.2% (52/80) of infants with just 30.1% (24/80) given solids before 6 months. There were also other liquids, including glucose water and traditional medicines (*Tshiunza*) that were reported by 89.5% (119/133) participants besides tea and juices. Such liquids were introduced as early as within the first 15 days by 5.3% (6/119) and increased by 32.4% (32/119) by the end of third month. Those given these liquids after 6 months were 47.3% (53/119). Grains such as fermented soft porridge (fortified and unfortified maize meal), instant porridge and maltabella were also given to infants throughout the first year of life starting from 15 days of birth (2.3%). By end of 60 days, more (42.9%) children were also fed grains. Majority of the children (70%) 93/133 were introduced to grains in their third month of life and the number increased to 91.8% (122/133) by 90 days.

Table 4.5. Types of solid and semi-solid foods and the time of their introduction in infants

Foods	No.	(%)
<b>Semi-solid food (n=133)</b>		
≤15 days	2	(1.50)
16-30 days	35	(26.5)
31-90 days	85	(64.4)
91-150 days	10	(7.60)
<b>Tea (n=110)</b>		
≤90 days	6	(5.40)
91-180 days	29	(26.4)
210-270 days (7-9 months)	54	(49.1)
300-360 days (10-12 months)	21	(19.1)
<b>Juices (n=80)</b>		
≤90 days	3	(3.80)
91-180 days	21	(26.0)
210-270 days (7-9 months)	28	(35.1)
300-360 days (10-12 months)	28	(35.1)
<b>Other liquids(n=119)</b>		
0-15 days	6	(5.30)
16-30 days	32	(32.4)
31-180 days	17	(14.9)
210-270 days (7-9 months)	32	(28.1)
300-360 days (10-12 months)	21	(19.3)
<b>Grains (n=133)</b>		
1-15 days	3	(2.30)
16-30 days	33	(24.8)
31-60 days	57	(42.8)
61-90 days	29	(21.7)
91-180 days	1	(8.40)

### 4.3 Intestinal permeability of infants in the Dzimauli area.

Table 4.6 presents intestinal permeability of infants at three (n=108), six (n=85) and nine (n=62) months of age. Out of 108 infants assessed at month three, 67.6% (73/108) had poor intestinal permeability (>0.085). At month 6 also, about half of the 85 assessed infants, 55.3% (47/85), had poor intestinal permeability (>0.085). Conversely, at month nine, the more than half of infants, 56.5% (35/62), had normal intestinal permeability (≤0.085).

Table 4.6. Intestinal permeability of infants in the Dzimauli area

*L/M ratio	Month 3 n=108		Month 6 n=85		Month 9 n=62	
	No.	(%)	No.	(%)	No.	(%)
Low ( $\leq 0.085$ )	35	(32.4)	38	(44.7)	35	(56.5)
High ( $> 0.085$ )	73	(67.6)	47	(55.3)	27	(43.5)

\*LM ratio above 0.085 indicates poor intestinal permeability

Figure 4.2 shows linear relationship between intestinal permeability (LM ratio) and the age in months (at 3, 6 and 9 months) of Dzimauli children. Results show that as the child grows the intestinal permeability works better. Intestinal permeability was poorest at month three, 67.6% (73/108) and 56.5% (35/62) showed improvement at month nine.

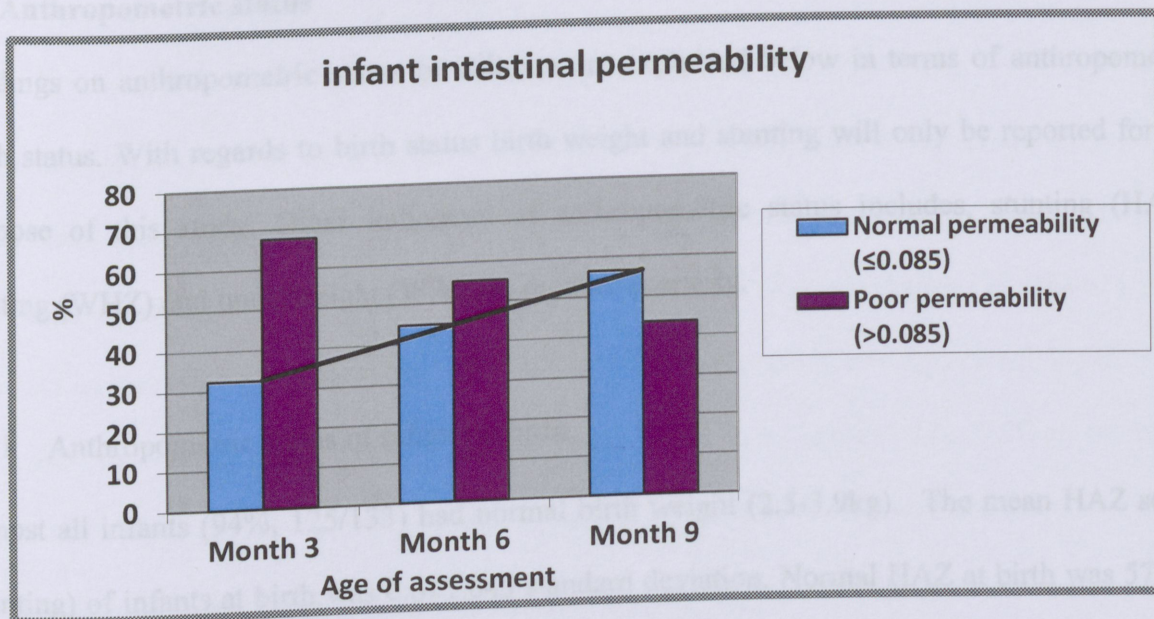


Figure 4.2. Linear relationship of intestinal permeability with age of infants in their first nine months of life

#### 4.4 Infant feeding practices and gut function correlations at birth (Dzimauli), n=133

There were no significant correlations observed between the different types of feeding practices and LM ratios (intestinal permeability) except with the introduction of colostrum irrespective of the delay. Inverse relationship was observed between infants who were breastfed the first milk (colostrum) showed and those not given the first milk with intestinal permeability at three months ( $r=-0.202$ ,  $p<0.05$ ) compared to those who were not given. However, timing of semi-solids had a significant relationship. The type of solid foods showed relationship at different age of introduction with LM, water ( $r=0.035$ ,  $p=0.003$ ), tea ( $r=0.201$ ,  $p=0.003$ ) and solids ( $r=0.220$ ,  $p=0.023$ ) at the age of three months.

#### 4.5 Anthropometric status

Findings on anthropometric status of children are explained below in terms of anthropometric birth status. With regards to birth status birth weight and stunting will only be reported for the purpose of this study. Other indicators of anthropometric status includes, stunting (HAZ), wasting (WHZ) and underweight (WAZ) at months quarterly.

##### 4.5.1 Anthropometric status of infants at birth

Almost all infants (94%, 125/133) had normal birth weight (2.5-3.9kg). The mean HAZ score (stunting) of infants at birth was  $0.6\pm 1.043$  standard deviation. Normal HAZ at birth was 57.1% (76/133) of infants based on the WHO (2009) standard. About 18.0% and 24.8% of infants were stunted and mildly stunted respectively. Anthropometric characteristics of study infants at birth (Dzimauli, South Africa) are shown in Table 4.7.

Table 4.7. Anthropometric characteristics of study infants at birth (Dzimauli), n=133

Birth weight	No.	(%)
Low	8	(6.0)
Normal	125	(94.0)
Stunting at birth		
Stunted	24	(18.1)
Mildly stunted	33	(24.8)
Normal	76	(57.1)

#### 4.5.2 Weight-for-age (WAZ) (underweight) of infants over a 12 month period

Table 4.8 presents the WAZ in children 0-12 months in Dzimauli community using WHO (2009) z-scores classifications. Most of the children (67.2%) (80/119) at month one had normal WAZ (-1SD to  $\leq$  +2SD), while less than one fifth (10.9 %) (13/119) were severely underweight ( $<$  -3SD). Mild (-2SD to  $<$  -1SD) and moderate underweight (3SD to  $<$  -2SD) was 17.6% (21/119) and 4.2% of infants at month one respectively.

At month three, most of the children (64.9%) (74/114) had normal WAZ (-1SD to  $\leq$  +2SD), while about one fifth (14%) of infants were underweight. Less than a quarter (21.1%) of infants was mildly underweight at months three of age in the study. About a quarter (38.1%) of infants was also underweight at month nine. Just above 50% were reported having normal WAZ scores at 12 months of age. There were no children with WAZ score greater than 2 SD in the study samples. Underweight decreased with age but from six months to nine months there is a slight improvement then drops again after nine months.

Table 4.8. Weight-for-age profile of infants over 0-12 months in Dzimauli community

Z-score classification	Interpretation	Month 1 *n=119	Month 3 *n=114	Month 6 *n=96	Month 9 *n=97	Month 12 *n=99
		No. (%)	No. (%)	No. (%)	No. (%)	No. (%)
< -3SD	Severely underweight	13 (10.9)	12 (10.5)	11 (11.5)	4 (4.1)	7 (7.3)
-3SD to < -2SD	Underweight	5 (4.2)	4 (3.5)	7 (7.3)	8 (8.2)	11 (11.5)
-2SD to < -1SD	Mild underweight	21 (17.6)	24 (21.1)	20 (20.8)	25 (25.8)	22 (22.9)
-1SD to +1SD	Normal WAZ	80 (67.2)	74 (64.9)	58 (60.4)	60 (61.9)	56 (58.3)

\* n was not always the same because some participant were not available at the time of assessment  $\pm 2$  days of assessment. Some participants dropped out before they reached 12 months.

#### 4.5.3 Weight-for-height (Wasting) (WHZ) of infants over a 12 month period

Table 4.9 presents weight-for-height (WHZ) of the infants using WHO (2009) z-scores classification. More than half 58.8% (70/119) of infants at month one had normal WHZ (-1SD to  $\leq +2$ SD), while nearly one third 29.9% were severely wasted (< -3SD). Mildly wasted (-2SD to < -1SD) and wasted (3SD to < -2SD) were reported at less than 10% and 3.4% at month one respectively.

The pattern of wasted infants was observed during the first three months with 58.8% infants wasted at months one and three. Approximately one fifth (13.5%) of the infants were at risk of wasting at months six (18.6%) and twelve (11.5%) respectively. Most severe wasting cases were observed at the first month and decreased as the infants aged at third (28.9%) and sixth (26%) months.

**Table 4.9. Weight-for-height (wasting) profile of infants over 12 months in Dzimauli**

Z-score classification	Interpretation	Month 1 *n=119 No. (%)	Month 3 *n=114 No. (%)	Month 6 *n=96 No. (%)	Month 9 *n=97 No. (%)	Month 12 *n=99 No. (%)
< -3SD	Severely wasted	35 (29.4)	33 (28.9)	25 (26.0)	19 (19.6)	23 (24)
-3SD to < -2SD	Wasting	4 (3.4)	6 (5.3)	4 (4.2)	4 (4.1)	6 (6.3)
-2SD to < -1SD	Mildly wasted	10 (8.4)	8 (7.0)	13 (13.5)	18 (18.6)	11 (11.5)
-1SD to +1SD	Normal WHZ	70 (58.8)	67 (58.8)	54 (56.3)	56 (57.7)	56 (58.3)

\* n was not always the same because some participant were not available at the time of assessment  $\pm 2$  days of assessment. Some participants dropped out before they reached 12 months.

#### 4.5.4 Height-for-age (HAZ) (stunting) of infants over a 12 months period

Table 4.10 presents height for age (HAZ) of the study participants using WHO (2009) z-scores classification. Most of the infants at first (61.3%) and third (59.6%) months had normal HAZ (-1SD to  $\leq$  +2SD). One fifth (18.1%) of infants were stunted at the first month of age. Just above half of the infants at six (55.2%) and nine (52.6%) also had normal HAZ scores. However, a number of infants at twelve months were mildly stunted (34%) (-2SD to < -1SD) of stunting and/or stunted (-3SD to < -2SD) (23.7%). An alarming number of stunted infants (29.9%) were mostly affected at twelve months of age.

**Table 4.10. Height-for-age (Stunting) profile of infants over 12 months in Dzimauli**

Z-score classification	Interpretation	Month 1 *n=119 No. (%)	Month 3 *n=114 No. (%)	Month 6 *n=96 No. (%)	Month 9 *n=97 No. (%)	Month 12 *n=99 No. (%)
< -3SD	Severely stunted	8 (6.7)	8 (7.0)	2 (2.1)	6 (6.2)	6 (6.2)
-3SD to < -2SD	Stunted	11 (9.2)	11 (9.6)	13 (13.5)	14 (14.4)	17 (23.7)
-2SD to < -1SD	Mild stunting	27 (22.7)	27 (23.7)	28 (29.2)	26 (26.8)	33 (34)
-1SD to +1SD	Normal height	73 (61.3)	68 (59.6)	53 (55.2)	51 (52.6)	41 (42.3)

\* n was not always the same because some participant were not available at the time of assessment  $\pm 2$  days of assessment. Some participants dropped out before they reached 12 months.

#### 4.6 Relationship between growth and gut function in infant

There was no relationship between L: M ratio at three months and anthropometric status during the first year of life. Significant correlations were observed between stunted children and L:M ratios at 6 month of age. Moreover, inverse correlations were observed between stunted children at three ( $r=-.381$ ,  $p=.003$ ) and nine ( $r=-.278$ ,  $p=.037$ ) months respectively. Further correlations were observed between weights-for-length at three months and L: M ratio at nine months of life. Association between growth and gut function is shown in Table 4.11.

Table 4.11. Association between growth and gut function in infants Dzimauli, SA

	LM excretion at M3	LM excretion at M6	LM excretion at M9	HAZ at M 3	WHZ at M 3	HAZ at M 6	HAZ at M 9
LM excretionM3	1	.032	-.031	.020	-0.47	-.051	-.010
LM excretionM6	108	85	.811	.838	.640	.621	.922
LM excretionM9	0.32	1	.105	-.084	-.036	<b>-.238*</b>	-.208
HAZ at M 3	.772	.416	.455	.751	<b>.036</b>	.065	
WHZ at M 3	85	85	62	81	81	<b>78</b>	80
HAZ at M 6	-.031	.105	1	<b>-.381**</b>	<b>.287*</b>	-.225	<b>-.278*</b>
HAZ at M 9	.811	.416	.003	<b>.029</b>	.087	<b>.037</b>	
	62	62	61	<b>58</b>	<b>58</b>	59	57

Height-for-age at month 3-HAZ at M3; weight –for- height at month 3-WHZ at M3; Height- for- age at month 6-HAZ at M 6; length –for- age at month 9-HAZ at M9; LM excretion ratios at month 3, 6 and 9.

#### 4.7 Relationship between solids and growth, solids and maternal characteristic

This section reports the result of the correlations between maternal characteristic, child growth and the timing of introduction of solid foods to the children diet. The variables are determined at the first three months, when the child is four to six months old and at seven to nine and when the child was introduced new solids foods at ten to twelve month.

#### 4.7.1 Relationship between introduction of solids and growth, solids and maternal characteristics at three months.

Correlations were observed between HAZ and WAZ ( $r=0.5$ ,  $p=0.000$ ), HAZ and mothers' age ( $r=0.196$ ,  $p=0.037$ ) and between HAZ and number of children the mother has ( $r=0.315$ ,  $p=0.001$ ). Inverse correlations were observed between WHZ and HAZ ( $r=0.33$ ,  $p=0.000$ ) and also WAZ and WAZ ( $r=-0.57$ ,  $p=0.000$ ). There were also weight for length correlations observed between number of years of schooling ( $r=0.27$ ,  $p=0.003$ ). Older mothers introduce solid foods earlier to their infants ( $r=0.227$ ,  $p=0.012$ ). Correlation was therefore shown by an increased weight gain, children who are given solid foods within three months of their life ( $r=0.192^*$ ,  $p=0.050$ ). Grains were first food mostly given by older mothers ( $r=0.258^*$ ,  $p=.004$ ). Parity showed positive correlation with mother's age ( $r=0.809^{**}$ ,  $p=0.600$ ). However, a negative correlation was observed with marital age ( $r=-0.364$ ,  $p=0.00$ ) and years of schooling ( $r=-0.294$ ,  $p=0.00$ ). Table 4.12 shows detailed results of relationship between variables.

Table 4.12. Relationship between introduction of solids, growth and maternal characteristics at three (3) months

		HAZ	WAZ	WHZ	Water	Solids	Other liquids	grains	Mother age	Yrs schooling	Parity
HAZ	R	1	<b>.574*</b>	<b>-.332*</b>	-.087	.066	<b>-.341*</b>	.049	<b>.196*</b>	-.108	<b>.315*</b>
	P		<b>.000</b>	<b>.000</b>	.393	.053	<b>.039</b>	.622	<b>.037</b>	.825	<b>.001</b>
	N	114	<b>114</b>	<b>114</b>	99	105	<b>37</b>	105	<b>114</b>	114	<b>114</b>
WAZ	R	<b>.574**</b>	1	<b>.577*</b>	-.018	<b>.192*</b>	-.176	.189	.172	.144	<b>.197*</b>
	P	<b>.000</b>		<b>.000</b>	.861	<b>.050</b>	.299	.053	.068	.126	<b>.036</b>
	N	<b>114</b>	114	<b>114</b>	99	<b>105</b>	37	105	114	114	<b>114</b>
WHZ	R	<b>-.332*</b>	<b>.577**</b>	1	.073	.144	.200	.158	-.002	<b>.273**</b>	-.086
	P	<b>.000</b>	<b>.000</b>		.475	.142	.236	.108	.982	<b>.003</b>	.362
	N	<b>114</b>	<b>114</b>	114	99	105	37	105	114	<b>114</b>	114
Water	R	-.087	-.018	.073	1	.109	<b>.334*</b>	.096	.064	-.018	.060
	P	.393	.861	.475		.260	<b>.044</b>	.322	.496	.846	.521
	N	99	99	99	114	108	<b>37</b>	108	117	117	117
Solids	R	.066	<b>.192*</b>	.144	.109	1	-.185	<b>.993**</b>	<b>.227*</b>	-.101	.087
	P	.503	<b>.050</b>	.142	.260		.260	<b>.000</b>	<b>.012</b>	.267	.341
	N	105	<b>105</b>	105	108	122	39	<b>119</b>	<b>122</b>	122	122
Other liquids	R	<b>-.341*</b>	-.176	.200	<b>.334*</b>	-.185	1	.042	.147	-.123	.222
	P	<b>.039</b>	.299	.236	<b>.044</b>	.260		.801	.348	.434	.152
	N	<b>37</b>	37	37	<b>37</b>	39	43	38	43	43	43
Grains	R	.049	.189	.158	.096	<b>.993**</b>	.042	1	<b>.258**</b>	-.102	.112
	P	.622	.053	.108	.322	<b>.000</b>	.801		<b>.004</b>	.266	.218
	N	105	105	105	108	<b>119</b>	38	122	<b>122</b>	122	122
Mother age	R	<b>.196*</b>	.172	-.002	.064	<b>.227*</b>	.147	<b>.258**</b>	1	-.093	<b>.809**</b>
	P	<b>.037</b>	.068	.982	.496	<b>.012</b>	.348	<b>.004</b>		.286	<b>.000</b>
	N	<b>114</b>	114	114	117	<b>122</b>	43	<b>122</b>	133	133	<b>133</b>
Yrs schooling	R	-.108	.144	<b>.273**</b>	-.018	-.101	-.123	-.102	-.093	1	<b>-.294**</b>
	P	.251	.126	<b>.003</b>	.846	.267	.434	.266	.286		<b>.001</b>
	N	114	114	<b>114</b>	117	122	43	122	133	133	<b>133</b>
Parity	R	<b>.315**</b>	<b>.197*</b>	-.086	.060	.087	.222	.112	<b>.809**</b>	<b>-.294**</b>	1
	P	<b>.001</b>	<b>.036</b>	.362	.521	.341	.152	.218	<b>.000</b>	<b>.001</b>	
	N	<b>114</b>	<b>114</b>	114	117	122	43	122	<b>133</b>	<b>133</b>	133

R-Pearson correlation; P-sig.(2-tailed) \*\*P<0.01; \*P<0.05; N-sample size; HAZ-stunting; WAZ-underweight; WHZ- wasting; Foods introduced before or at three months (Solids; Other liquids including traditional medicine; Grains) ;Mother age- mothers age at enrolment; Yrs schooling-mothers year of schooling; Parity-number of children mother has at enrolment.

#### 4.7.2 Relationship between introduction of solids and growth, solids and maternal characteristics at six (6) months.

At first six months of life, correlations were observed between HAZ and WAZ ( $r=0.567$ ,  $p=0.000$ ). Z-score (WAZ and WHZ) were significantly correlated to each other ( $r=0.567$ ,  $p=0.000$ ), ( $r=0.863$ ,  $p=0.000$ ). As number of years of schooling increases, WAZ showed increase ( $r=0.567$ ,  $p=0.000$ ). The older the mother the likely it became that at 6 months of age semi-solids and liquids would be introduced to the infants. Correlations were observed between mothers' age and introduction of solids ( $r=0.236$ ,  $p=0.007$ ). Tea ( $r=0.339$ ,  $p=0.46$ ) and grains ( $r=0.215$ ,  $p=0.013$ ) were mostly introduced by older mothers. Mothers with more years of schooling were inversely correlated with parity ( $r=-0.294$ ,  $p=0.001$ ). Mothers who had more years of schooling had fewer children. Mother's marital age was also inversely correlated with their age (her age) ( $r=-0.308$ ,  $p=0.000$ ) and parity ( $r=0.809$ ,  $p=0.000$ ). Detailed correlations are shown in Table

4.13.

**Table 4.13. Relationship between introduction of solids and growth, solids and maternal characteristics at six (6) months**

		Mother age	Marital age	Yrs schooling	Parity	HAZ	WAZ	WHZ	Water	Solids	Tea	Grains
Mother age	R	1	<b>-.308**</b>	-.093	<b>.809**</b>	.053	.089	.075	.113	<b>.236**</b>	<b>.339*</b>	<b>.215*</b>
	P		<b>.000</b>	.286	<b>.000</b>	.610	.387	.466	.207	<b>.007</b>	<b>.046</b>	<b>.013</b>
	N	133	<b>133</b>	133	<b>133</b>	96	96	96	127	<b>132</b>	<b>35</b>	<b>133</b>
Marital age	R	<b>-.308**</b>	1	.079	<b>-.364**</b>	-.005	.097	.120	.050	.026	-.088	.048
	P	<b>.000</b>		.365	<b>.000</b>	.964	.348	.243	.575	.771	.615	.583
	N	<b>133</b>	133	133	<b>133</b>	96	96	86	127	132	35	133
Yrs schooling	R	-.093	.079	1	<b>-.294**</b>	.111	<b>.239*</b>	<b>.224*</b>	-.049	-.055	.162	-.075
	P	.286	.365		<b>.001</b>	.283	<b>.019</b>	<b>.028</b>	.583	.532	.352	.392
	N	133	133	133	<b>133</b>	96	<b>96</b>	<b>96</b>	127	132	35	133
Parity	R	<b>.809**</b>	<b>-.364**</b>	<b>-.294**</b>	1	.092	.021	-.034	.118	.104	.193	.133
	P	<b>.000</b>	<b>.000</b>	<b>.001</b>		.374	.840	.741	.186	.236	.267	.126
	N	<b>133</b>	<b>133</b>	<b>133</b>	133	96	96	96	127	132	35	133
HAZ	R	.053	-.005	.111	.092	1	<b>.567*</b>	.080	.033	.033	.066	-.016
	P	.610	.964	.283	.374		<b>.000</b>	.440	.755	.753	.749	.881
	N	96	96	96	96	96	<b>96</b>	96	93	96	26	96
WAZ	R	.089	.097	<b>.239*</b>	.021	<b>.567**</b>	1	<b>.863**</b>	.050	.145	.016	.179
	P	.387	.348	<b>.019</b>	.840	<b>.000</b>		<b>.000</b>	.633	.157	.938	.081
	N	96	96	<b>96</b>	96	<b>96</b>	96	<b>96</b>	93	96	26	96
WHZ	R	.075	.120	<b>.224*</b>	-.034	.080	<b>.863**</b>	1	.038	.139	-.019	<b>.210*</b>
	P	.466	.243	<b>.028</b>	.741	.440	<b>.000</b>		.716	.177	.928	<b>.040</b>
	N	96	96	<b>96</b>	96	96	<b>96</b>	96	93	96	26	<b>96</b>
Water	R	.113	.050	-.049	.118	.033	.050	.038	1	.172	<b>.365*</b>	.130
	P	.207	.575	.583	.186	.755	.633	.716		.054	<b>.037</b>	.145
	N	127	127	127	127	93	93	93	127	126	<b>33</b>	127
Solids	R	<b>.263**</b>	.026	-.055	.104	.033	.145	.139	.172	1	.247	<b>.823**</b>
	P	<b>.007</b>	.771	.532	.236	.753	.157	.177	.054		.153	<b>.000</b>
	N	<b>132</b>	132	132	132	96	96	96	126	132	35	<b>132</b>
Tea	R	<b>.339*</b>	-.088	.162	.193	.066	.016	-.019	<b>.365*</b>	.247	1	.239
	P	<b>.046</b>	.615	.352	.267	.749	.938	.928	<b>.037</b>	.153		.167
	N	<b>35</b>	35	35	35	26	26	26	<b>33</b>	35	35	35
Grains	R	<b>.215*</b>	.048	-.075	.133	-.016	.179	<b>.210*</b>	.130	.823	.239	1
	P	<b>.013</b>	.583	.392	.126	.881	.081	<b>.040</b>	.145	.000	.167	
	N	<b>133</b>	133	133	133	96	96	<b>96</b>	127	132	35	133

R-Pearson correlation; P-sig.(2-tailed) \*\*P<0.01; \*P<0.05; N-sample size; HAZ-stunting; WAZ-underweight; WHZ- wasting; Foods introduced at six months (Water, Solids, Tea, Grains). Mother age-mother's age at enrolment; Yrs schooling-mothers year of schooling; Parity-number of children mother has at enrolment.

### 4.7.3 Relationship between introduction of solids, growth and maternal characteristics at nine months.

Anthropometric z-score at nine months were correlated between HAZ and WAZ ( $r=0.638$ ,  $p=0.00$ ) and HAZ and WHZ ( $r=0.207$ ,  $p=0.042$ ). WHZ was positively correlated with WAZ ( $r=0.877$ ,  $p=0.000$ ). Mother's age was a contributing factor to introduction of solids even at nine months ( $r=0.236$ ,  $p=0.007$ ), fruit juice ( $r=0.287$ ,  $p=0.039$ ) and grain ( $r=0.215$ ,  $p=0.013$ ). Wasted infants were introduced to tea ( $r=0.228$ ,  $p=0.049$ ) and other liquids ( $r=0.303$ ,  $p=0.11$ ) earlier. Correlation was observed between families with more children and introduction of other liquids ( $r=0.295$ ,  $p=0.034$ ) and fruits ( $r=0.295$ ,  $p=0.034$ ). Detailed correlations are shown in Table 4.14.

	P	900	908	901		954	918	905	236	906	934	126
	N	133	133	133	133	97	97	97	133	88	52	133
HAZ	R	0.76	0.36	0.30	0.61	1	0.68**	0.67*	0.33	0.57	-0.07	0.16
	P	0.00	0.01	0.04	0.00		0.00	0.00	0.00	0.01	0.11	0.29
	N	97	97	97	97	97	97	97	97	75	44	97
WAZ	R	0.28	0.67	0.37	0.64	0.68**	1	0.77**	0.57	-0.08	-0.34	0.04
	P	0.01	0.00	0.01	0.00	0.00		0.00	0.01	0.04	0.17	0.81
	N	97	97	97	97	97	97	97	97	75	44	97
WHZ	R	-0.10	0.92	0.25	0.17	0.67*	0.77**	1	0.10	-0.28*	-0.70	0.01
	P	0.73	0.00	0.07	0.63	0.00	0.00		0.24	0.05	0.07	0.06
	N	97	97	97	97	97	97	97	97	75	44	97
Solids	R	0.24**	0.05	-0.25	0.04	0.13	0.67	0.10	1	0.13	0.52	0.23**
	P	0.07	0.71	0.02	0.36	0.86	0.14	0.23		0.04	0.01	0.00
	N	133	133	133	133	97	97	97	133	88	52	133
Tea	R	-0.23	0.53	0.03	-0.79	0.07	-0.45	0.28*	0.15	1	0.56	0.74
	P	0.03	0.30	0.84	0.00	0.00	0.04	0.00	0.19		0.01	0.04
	N	97	97	97	97	97	97	97	97	75	44	97
Fruit juice	R	-0.27*	-0.09	0.02	-0.29	-0.05	-0.29	-0.21	0.12	0.58	1	-0.35
	P	0.05	0.78	0.91	0.04	0.71	0.07	0.07	0.21	0.01		0.07
	N	97	97	97	97	97	97	97	97	75	44	97
Grain	R	0.21*	0.04	-0.04	0.15	0.18	0.09	0.05	0.48**	0.14	-0.33	1
	P	0.03	0.63	0.92	0.06	0.06	0.10	0.24	0.00	0.04	0.07	
	N	133	133	133	133	97	97	97	133	88	52	133

R-Pearson correlation; P-significance; \*\*P<0.01, \*P<0.05. Example: HAZ=stature; WAZ=underweight; WHZ=weight; Foods introduced at nine months: Fruit, Grains, Tea and Juice, Grains; Mother age=mother's age at enrolment; Yrs schooling=years year of schooling; First-born=if children number has at enrolment.

**Table 4.14. Relationship between introduction of solids and growth, solid and maternal characteristics nine (9) months**

		Mother age	Marital age	Yrs of schooling	Parity	HAZ	WAZ	WHZ	Solids	Tea	Fruit juice	Grains
Mother age	R	1	<b>-.308**</b>	.093	<b>.809**</b>	0.76	.022	-.030	<b>.236**</b>	-.083	<b>.287*</b>	<b>.215*</b>
	P		<b>.000</b>	.286	<b>.000</b>	.459	8.33	.773	<b>.007</b>	.445	<b>.039</b>	<b>.013</b>
	N	133	<b>133</b>	133	<b>133</b>	97	97	97	<b>132</b>	88	<b>52</b>	<b>133</b>
Marital age	R	<b>-.308**</b>	1	.079	<b>-.364**</b>	.056	.067	.077	.026	.152	-.069	.048
	P	<b>.000</b>		.365	<b>.000</b>	.583	.514	.454	.771	.158	.628	.583
	N	<b>133</b>	133	133	<b>133</b>	97	97	97	132	88	52	133
Yrs of schooling	R	-.093	.079	1	<b>-.294**</b>	.189	.187	.135	-.055	.023	.002	-.075
	P	.286	.365		<b>.001</b>	.064	.067	.187	.532	.834	.991	.392
	N	133	133	133	<b>133</b>	97	97	97	132	88	52	133
Parity	R	<b>.809**</b>	<b>-.364**</b>	<b>-.294**</b>	1	.061	.024	-.017	.104	-.179	<b>.295*</b>	.133
	P	<b>.000</b>	<b>.000</b>	<b>.001</b>		.554	.818	.865	.236	.096	<b>.034</b>	.126
	N	<b>133</b>	<b>133</b>	<b>133</b>	133	97	97	97	132	88	<b>52</b>	133
HAZ	R	.076	.056	.189	.061	1	<b>.638**</b>	<b>.207*</b>	.133	.097	-.037	.116
	P	.459	.583	.064	.554		<b>.000</b>	<b>.042</b>	.196	.408	.811	.260
	N	97	97	97	97	97	<b>97</b>	<b>97</b>	97	75	44	97
WAZ	R	.022	.067	.187	.024	<b>.638**</b>	1	<b>.877**</b>	.067	-.148	-.234	.094
	P	.833	.514	.067	.818	<b>.000</b>		<b>.000</b>	.514	.204	.127	.361
	N	97	97	97	97	<b>97</b>	97	<b>97</b>	97	75	44	97
WHZ	R	-.030	.077	.135	.017	<b>.207*</b>	<b>.877**</b>	1	.010	<b>-.228*</b>	-.270	.053
	P	.773	.454	.187	.865	<b>.042</b>	<b>.000</b>		.923	<b>.049</b>	.077	.606
	N	97	97	97	97	<b>97</b>	<b>97</b>	97	97	<b>75</b>	44	97
Solids	R	<b>.236**</b>	.026	-.055	.104	.133	.067	.010	1	.175	.152	<b>.823**</b>
	P	<b>.007</b>	.771	.532	.236	.196	.514	.923		.104	.281	<b>.000</b>
	N	<b>132</b>	132	132	132	97	97	97	132	88	52	<b>132</b>
Tea	R	-.083	.152	.023	-.179	.097	-.148	<b>-.228*</b>	.175	1	.258	.074
	P	.445	.158	.834	.096	.408	.204	<b>.049</b>	.104		.091	.494
	N	88	88	88	88	75	75	<b>75</b>	88	88	44	88
Fruit juice	R	<b>.287*</b>	-.069	.002	<b>.295*</b>	-.037	-.234	-.270	.152	.258	1	-.035
	P	<b>.039</b>	.628	.991	<b>.034</b>	.811	.127	.077	.281	.091		.807
	N	<b>52</b>	52	52	<b>52</b>	44	44	44	52	44	52	52
Grains	R	<b>.215*</b>	.048	-.075	.133	.116	.094	.053	<b>.823**</b>	.074	-.035	1
	P	<b>.013</b>	.583	.392	.126	.260	.361	.606	<b>.000</b>	.494	.807	
	N	<b>133</b>	133	133	133	97	97	97	<b>132</b>	88	52	133

R-Pearson correlation; P-sig.(2-tailed) \*\*P<0.01; \*P<0.05; N-sample size; HAZ-stunting; WAZ-underweight; WHZ- wasting; Foods introduced at nine months (Water, Solids, Tea, fruit juice, Grains). Mother age- mother's age at enrolment; Yrs schooling-mothers year of schooling; Parity-number of children mother has at enrolment.

#### 4.7.4 Relationship between introduction of solids and growth, solids and maternal characteristics at twelve months.

Correlation was observed between underweight infants and mother's years of schooling ( $r=0.203$ ,  $p=0.047$ ). Wasted infants were introduced to formula milk ( $r=-0.374$ ,  $p=0.010$ ) early as compared to their counterpart. Positive relationship was also observed with WHZ ( $r=-0.374$ ,  $p=0.023$ ) and marital age ( $r=0.264$ ,  $p=0.032$ ). Mothers who got married at a later age continued with introduction of formula. Detailed correlations are shown in Table 4.15.

Marriage	R	0.203	0.185	0.154	0.187	0.181	0.171	0.166	0.131
	N	133	133	133	97	96	96	110	131
HAZ	R	0.234	0.141	0.145	0.1	0.004	0.141	0.229	0.153
	N	97	97	97	97	96	96	95	96
WAZ	R	0.23	0.169	0.203*	0.296**	1	0.00	-0.374*	-0.052
	N	96	96	96	96	96	96	95	96
WHZ	R	0.20	0.129	0.179	0.161	0.020*	1	-0.374*	0.144
	N	96	96	96	96	96	96	95	96
Waste	R	0.131	0.062	0.009	0.185	-0.00	-0.052	0.057	0.607**
	N	131	131	131	96	95	95	95	100
Solids	R	0.236**	0.026	-0.053	0.129	0.119	0.067	0.051	0.200*
	N	132	132	132	97	96	96	95	110
Formula	R	-0.008	-0.264*	0.053	-0.238	-0.279*	-0.374*	1	-0.008
	N	96	96	96	46	46	46	65	31
Tea	R	0.07	0.125	0.050	0.157	-0.052	-0.144	0.108	1
	N	110	110	110	96	95	95	95	110
Grains	R	0.13*	0.048	-0.073	0.147	0.121	0.074	0.053	0.08
	N	133	133	133	97	96	96	95	110

R-Pearson correlation; P- $\chi^2$ -test; \*\*P<0.01, \*P<0.05, N-sample size; HAZ-meaning; WAZ-underweight; WHZ-wasting; Food introduced in first month (Water, Solids, Tea, fruit juice, Grains); Mother age- mother's age at enrollment; Yrs schooling- mother's year of schooling; Party- number of children mother has at enrollment

Table 4.15. Relationship between introduction of solids and growth, solids and maternal characteristics at twelve (12) months

		Mother age	Marital age	Yrs schooling	HAZ	WAZ	WHZ	Formula	Tea	Grains
Mother age	R	1	<b>-.308*</b>	-.093	.024	.023	.020	-.008	.007	.215
	P		<b>.000</b>	.286	.813	.827	.845	.948	.944	.013
	N	133	<b>133</b>	133	97	96	96	66	110	133
Marital age	R	<b>-.308**</b>	1	.079	.141	.169	.129	<b>-.264*</b>	.125	.048
	P	.000		.365	.168	.101	.209	<b>.032</b>	.192	.583
	N	1333	133	1333	97	96	96	<b>66</b>	110	133
Yrs schooling	R	-.093	.079	1	.146	<b>.203*</b>	.179	.053	.050	-.075
	P	.286	.365		.154	<b>.047</b>	.081	.671	.606	.392
	N	1333	1333	133	97	<b>96</b>	96	66	110	133
HAZ	R	.024	.141	.146	1	<b>.596**</b>	.161	-.229	.153	.147
	P	.813	.168	.154		<b>.000</b>	.118	.125	.136	.151
	N	97	97	97	97	<b>96</b>	96	46	96	97
WAZ	R	.023	.169	<b>.203*</b>	<b>.596**</b>	1	<b>.885**</b>	<b>-.374*</b>	-.052	.131
	P	.827	.101	.047	.000		<b>.000</b>	<b>.010</b>	.617	.205
	N	96	96	96	96	96	<b>96</b>	<b>46</b>	95	96
WHZ	R	.020	.129	.179	.161	<b>.885**</b>	1	<b>-.334*</b>	-.144	.074
	P	.845	.209	.081	.118	.000		<b>.023</b>	.165	.472
	N	96	96	96	96	96	96	<b>46</b>	95	96
Water	R	.155	.062	.000	.108	-.003	-.052	.057	<b>.360**</b>	.196
	P	.076	.482	.997	.294	.981	.616	.650	<b>.000</b>	.025
	N	131	131	131	96	95	95	65	<b>109</b>	131
Solids	R	<b>.236**</b>	.026	-.055	.139	.119	.067	.054	<b>.238*</b>	<b>.823**</b>
	P	.007	.771	.532	.176	.248	.517	.665	<b>.012</b>	<b>.000</b>
	N	132	132	132	97	96	96	66	<b>110</b>	<b>132</b>
Formula	R	-.008	<b>-.264*</b>	.053	-.229	<b>-.374*</b>	<b>-.334*</b>	1	-.108	.011
	P	.946	.032	.671	.125	.010	.023		.447	.927
	N	66	66	66	46	46	46	66	52	66
Tea	R	.007	.125	.050	.153	-.052	-.144	-.108	1	.128
	P	.944	.192	.606	.136	.617	.165	.447		.181
	N	110	110	110	96	95	95	52	110	110
Grains	R	.215*	.048	-.075	.147	.131	.074	.011	.128	1
	P	.013	.583	.392	.151	.205	.472	.927	.181	
	N	133	133	133	97	96	96	66	110	114

R-Pearson correlation; P-sig.(2-tailed) \*\*P<0.01; \*P<0.05; N-sample size; HAZ-stunting; WAZ-underweight; WHZ- wasting; Foods introduced at nine months (Water, Solids, Tea, fruit juice, Grains). Mother age- mother's age at enrolment; Yrs schooling-mothers year of schooling; Parity-number of children mother has at enrolment

## CHAPTER FIVE: DISCUSSION

### 5.1 Overview

This chapter discusses the findings regarding feeding practices, gut function, anthropometric status and the interaction between these parameters in infants in Dzimauli community.

### 5.2 The discussion of findings

There was no relationship observed between the type of feeding practices and gut function in infants. Exclusive breastfeeding was practiced by few mothers and for a relatively short duration and therefore its relationship with the gut function could not be deduced. However, breastfeeding initiation as well as its timing showed a positive association with gut function. Gut function was associated with poor anthropometric status of children at month three, where type of feeding is crucial as compared to months six and nine. Infants introduced to non breastmilk foods as early as within the first six months had poor height-for-age, weight-for-height and weight-for-age in the study. Maternal characteristics such as age, educational level and marital status were amongst the factors influencing infant feeding in the current study.

Feeding practices showed high breastfeeding initiation rate which is a good infant feeding practice (WHO, 2010) but subsequent feeding practices were generally below optimal as indicated by the timing of initiation and introduction of solids. Even with good initiation rate, timing of initiation (within an hour of birth) was mainly not done properly. Most mothers started breastfeeding after an hour surpassed but still within twenty four hours. This delay in breastfeeding initiation could be encouraged by the mothers giving birth at homes where mothers visit health facilities after long hour of birth and initial breastfeeding established late (Tampah-Naah and Kumi-Kyereme, 2013). Conversely, introduction of solids was too early, as early as

the first month of life, compared to the recommended time, six months, of exclusive breastfeeding (WHO, 2010). In a recent study conducted by Madiba and Letsoalo (2013) in South Africa, reported that mixed feeding at an early age is still a concern as mothers give their infants semi-solids foods as early as the first months after birth. Scott et al. (2009) also indicated that early introduction was common in Australia with mothers giving non breastmilk foods as early as the fourth month.

With regards to the delay in initiation, the observed poor feeding practices in this study seem to follow a trend reported in various parts of the world including South Africa and Limpopo Province in particular. For instance, WHO (2008) declared poor feeding practices as a common concern in both developed and developing countries. In 2009, Wiljndaele et al. (2009) was still reporting lack of improvement particularly in the timing of initiation in both the developing and developed countries. The delay in the initiation of breastfeeding of infants in the developing countries was also reported by Mhrshahi et al. (2010), indicating less than 30% of mothers' breastfeeding within the first hour after birth. Breastfeeding delay in South Africa has been reported by SADHS (2003). Five years later, Mushaphi et al (2008) still reported this poor practice in Limpopo Province. WHO (2010) pinpointed lack of awareness of the benefits of early initiation by mothers as the possible reason for breastfeeding delay. Of great concern is that, suboptimum breastfeeding in young children has been documented in observational studies as the risk of morbidity and mortality (Arifeen et al., 2001).

Early introduction of non breastmilk foods and the compliance of exclusive breastfeeding for six months is another experienced challenge with regards to feeding practices of infants (WHO, 2002). Patel et al. (2010) and Mihrshahi et al. (2010) indicated poor exclusive breastfeeding practices worldwide citing, 46.4% of infants exclusively breastfed in India and 42.5% in Bangladesh. In rural Malawi, non-nutritive liquids (mostly water) other than breastmilk were given to infants before they were four months old (Kerr et al., 2007). Scott et al. (2009) also reported high level of non-compliance related to the timing of solids food amongst Australian mothers. In South Africa, there has been a concern of even low rate of exclusive breastfeeding problem with only 8% of infants aged less than six months exclusively breastfed despite 87% of breastfed infants (SADHS, 2003).

Labadarios et al. (2000) indicated that in South Africa, mothers give their infants other liquids too early whilst breastfeeding. This was also indicated recently by Madiba and Letsoalo (2013). In the current study, less than 10% of infants were exclusively breastfed within the first three months. Similar findings of poor exclusive breastfeeding were reported by other studies conducted in Limpopo Province. Mamabolo et al. (2004) reported introduction of solids foods to infants during their first 3 months of life in Polokwane rural areas. According to Mamabolo et al. (2004), more than half of the mothers (64.4%) had introduced solids foods to their infants before three months of age. In fact, 56% of infants had received some form of food by the end of the first month. Mushaphi et al. (2008) reported a 7.6% of exclusive breastfeeding in Mutale Municipality in the Vhembe District. These findings were not much different with a report from the North West Province of South Africa where 10.4% of children were exclusively breastfed (Kruger and Gericke, 1999). In this study, water was the first food introduced to infants (47.9%) in the first months of life. This has been a trend observed in the Limpopo whereby infants were

given water with or without sugar or salt (Mamabolo et al., 2004; Mushaphi et al., 2008). The other common non-breastmilk foods in the province included maize meal porridge, sorghum porridge (*mabella*), tea and herbal drinks (Mamabolo et al., 2004). These foods also featured in the current study indicating that the feeding practices have not changed in a decade.

Several factors that may contribute towards poor feeding practices of infant's have been identified in the study. There are programs in place to improve infants feeding practices in South Africa however, several factors impact negatively on compliance with the recommended guidelines. A number of socio-demographic variables were associated with the initiation and duration of breastfeeding in the current study. Age, marital status and educational level of the mother as well as cultural practices are discussed below.

In this study, older mothers introduced solids foods to infants as early as fifteen days after birth. This practice was also observed in rural in Kwazulu Natal (Faber and Benade 2007) and Limpopo Provinces (Mushaphi et al., 2008) as well as in urban setting of Western Cape Province (Sibeko et al., 2005) in South Africa. These results were contrary to the study conducted by Wiljndaele et al. (2009) in America where it were younger mothers who practiced poor feeding practices. In South Africa, older mothers may be reluctant to change from old cultural practices irrespective of the nutrition education given.

With regards to marital status, married mothers in this study initiated formula feeding early as compared to single mothers. This probably is an indication that spouses and in-laws of mothers have an inappropriate influence on choices made for infant feeding at home. In an unpublished focus group conducted in the same area Mothers recruited in the study were still relatively young

(less than 26 years old). Most of them were still going to school but some were employed. The association between breastfeeding and education has been investigated in other studies (Doherty et al., 2012; Radwan, 2013). In the developing countries, higher education was associated with the adoption of modern ideas of infant feeding (Radwan, 2013). Modern ideas of breastfeeding include choosing use of formula milk and other solids over breastfeeding. Formula milk was strongly associated with education, mothers in this study also introduced solids foods and formula milk as early as the first months after birth. One of the influencing factors could have been leaving their children with their parents (children's grandmothers) or other relatives or caregivers.

Lutter et al. (2011) identified cultural beliefs and knowledge as contributing factors towards poor feeding practices. Mushaphi et al. (2008) showed a cultural practice of introducing *tshiunza* immediately after birth in the Venda culture. Doherty et al. (2011) indicated that long standing cultural practice and the support of formula milk by government through the Prevention to Mother to Child Transmission (PMTCT) and protein energy malnutrition programme were among the factors that contributed to the low rate of exclusive breastfeeding in South Africa.

Goto et al. (2002) reported a high number (92%) of Nepali children aged from 0-60 months with poor L: M ratios (indicating poor gut function). Intestinal permeability in infants represents a critical balance between absorption of nutritional agents and protection from dangerous pathogens (Goto et al., 2009; Mondal et al., 2012). In this study most impairment of the gut in infants occurred at three months and got better with time. At nine months, most children showed normal intestinal permeability. This improvement in the intestinal permeability is expected at later months because the gut of the infant from six months is physiologically ready for the kinds

of foods fed to them. According to previous studies (Goto et al., 2002; Taylor et al., 2010), the damage of the gut in infants occurs as a result of early introduction of foods other than breast milk before six months of age.

Although the current study reports no relationship between introduction feeding practices and gut function as defined by the high absorption of the lactulose and mannitol sugars. Boaz et al. (2013) in his study increased mannitol absorption is an outcome of increased gut permeability in children. Introduction of semi-solids/solids was associated with gut permeability Gambian (Lunn, 2000) and Nepali (Goto et al., 2009) infant. Furthermore, early introduction of non-breast milk is associated with an increased risk of diarrhoea in infancy and increased body fat (Scott et al., 2009). The current study did not look at the association between infection and with any of the variable in the study.

Poor gut function often results in stunted growth and poor development. Lunn (2000) indicated that poor infant feeding can lead to infants losing weight due to the partial villus atrophy that reduces the absorption and digestion of lactose and probably other nutrients important for child growth and development. Campbell et al. (2003) also discovered in their study that the mucosal damage was the cause of malnutrition in children rather than the consequence. Furthermore, mucosal damage in children has been reported as one of the major contributors to growth impairment and malnutrition worldwide (Goto et al., 2009). Lutter et al. (2011) reported that in 2010 an estimated 9% and 27% of children under the age of five globally were wasted and stunted respectively. Similar observations were made in the present study with majority of infants being wasted (19.6%-24.6%) and stunted (15.6%-29.9%). According to WHO (2013) a prevalence of 10-14% of wasting needs a serious consideration. Growth faltering is common in

many developing countries starting at an early age with underweight-scores (WAZ) and wasting z-scores (WHZ) falling from about 3 months of age until about 12 months of age (Shrimpton et al., 2001).

Undernutrition in the first year of life is still a serious challenge in rural South Africa (SADHS, 2003). Anthropometric assessment in this study indicated that wasting and stunting were the main growth problems in infants with the severity of wasting (19.6%-29.4%) throughout the first year of life. Infants introduced to solid foods as early as the first six months of age had poor weight for age and weight for height z-scores. Of great concern is the observation made in this study that poor anthropometric status in infants was as early as just after birth. In the Limpopo Province, Mamabolo et al. (2008) reported that infants who were weaned earlier than six months had poor growth status. This trend has been documented in both developing (Kerr et al., 2007) and developed (Mihirshahi et al., 2010) countries with poor feeding practices. However, Goto et al. (2002) and Campbell et al. (2003) related growth faltering to either food intake or infection.

In a Malawian study, Kerr et al. (2007) also indicated that infant introduced to non breastmilk food before six month were given food that could pose risk on introducing pathogens to the child. Therefore infection could also be a contributing factor in the poor growth of the children in this study. Types of food introduced showed some effect in poor growth of children with age. Similar observations were made in Malawi, whereby infant introduced to foods such as porridge and water early than six months had poor anthropometric status (Kerr et al., 2007). Introduction of grains showed positive relationship from six months of age with wasting decreasing as grains were introduced. Infants introduced to tea and other liquids early had poor weight for height at

nine months. Formula milk was associated with poor weight for height at the age of twelve months.

## CHAPTER SIX: CONCLUSIONS AND RECOMMENDATIONS

### 6.1 Introduction

This chapter reports the conclusions and recommendations made from the findings on the relationship between feeding practices, anthropometric status and gut function in infants in Drimau community.

### 6.2 Conclusion

The aim of the study was to determine the relationship between infant feeding practices, anthropometric status and gut function in Drimau community. This study confirmed the importance of giving colostrum to infants for better intestinal permeability. Relationship between exclusive breastfeeding and gut function could not be concluded. However, there was no relationship observed between different feeding practices with gut function. These observations could have been due to other unexpected factors such as age when the association was observed. Early introduction (before the age of 6 months) of non-breastmilk foods affected the growth status of infants. Non-breastmilk foods that were introduced before 3 months of age resulted in infants with poor weight-for-age (underweight) and weight-for-height (wasting), as well as stunting.

Mothers still do not comply with the recommended infant feeding guidelines, so poor feeding practices of infants in Drimau have been observed as major focus area that needs more attention possibly to other rural areas of South Africa as well. There was no optimal and exclusive feeding in the current study. Foods such as water, maize meal soft porridge and formula milk were amongst the first introduced foods in the study.

## CHAPTER SIX: CONCLUSIONS AND RECOMMENDATIONS

### 6.1 Introduction

This chapter reports the conclusions and recommendations made from the findings on the relationship between feeding practices, anthropometric status and gut function in infants in Dzimauli community.

### 6.2 Conclusion

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Mothers still do not comply with the recommended infant feeding guidelines, so poor feeding practices of infants in Dzimauli have been observed as major focus area that needs more attention possibly to other rural areas of South Africa as well. There was no optimal and exclusive feeding in the current study. Foods such as water, maize meal soft porridge and formula milk were amongst the first introduced foods in the study.

Poor intestinal permeability was observed as early as the first three months of life in the current study and improved as the infants grow. Nevertheless, associations between feeding practices and anthropometric status of infants were observed as early as the first six months of life. Underweight, wasting and stunting were found prevalent in Dzimauli as early as first three months after birth and continue until the child is twelve months of age. Wasting was more prevalent than stunting and underweight in the current study. Maternal characteristics such as level of education, age and marital status influence decision on infant feeding.

### 6.3 Recommendations

It is recommended that studies to discuss why mothers still introduce non breastmilk food earlier to their infants need to be conducted and reasons for introduction of such food before 6 months should be investigated. Furthermore, studies should focus on timing of introduction on solids food and child gut performance and the effect on child growth and development. Breastfeeding promotion strategies should be more emphasised especially in rural communities. Working mothers should be given a long maternity leave to have enough time to be spent nursing their infants. Breastfeeding campaigns should focus in all age groups rather than mothers. Family members should be encouraged also in the support of breastfeeding mothers and be educated on the importance of supporting breastfeeding mothers.

Research focusing on infant intestinal permeability and its effect on nutritional status should be considered in the near future. The association of stunting and wasting observed in the study and child development should be investigated.

## 7. REFERENCES

Agampodi, S.B., Agampodi, T., and Piyaseeli, U.K.P. (2007). Breastfeeding practices in a public health field practice in Sri Lanka: a survival analysis. *International Breastfeeding Journal*, 2:13. Available at: <http://www.internationalbreastfeedingjournal.com/content/2/1/>. Accessed 23 May 2011.

Arifeen, S., Black, R.E., Antelman, G., Baqui, A., Caulfield, L., and Becker, S. (2001). Exclusive Breastfeeding Reduces Acute Respiratory Infection and Diarrhea Deaths Among Infants in Dhaka Slums. *Pediatrics*, **108** (4):67, Available at <http://www.internationalbreastfeedingjournal.com/content/2/1/>. Accessed 20 August 2012

Assis, A.M., Barreto, M.L., Santos, L.M., Fiaccone, R., and da Silva, G.G.S. (2005). Growth faltering in childhood related to diarrhea: a longitudinal community based study. *European Journal of Clinical Nutrition* **59**, 1317–1323.

Barboza Junior, M.S., Silva, T.M., Guerrant, R.L., and Lima, A.A (1999). Measurement of intestinal permeability using mannitol and lactulose in children with diarrheal diseases. *Brazil Journal of Medical Biology Res*, **32**:1499-1504.

Badenhorst, C.J., Steyn, N.P., and Jooste, P.L. (1993). Nutritional status of Pedi school children aged 6–14 years in two rural areas in Lebowa: a comprehensive nutritional survey of dietary intake, anthropometric and clinical measurements. *South African Journal of Food Science and Nutrition*, **5**:112–5.

Becquet, R., Leroy, V., Ekouevi, D.K., Viho, I., Castetbon, K., Fassinou, P., Dabis, F., Timite-Konan, M., and ANRS 1201/1202 Ditrane Plus study group. (2006). Complementary feeding

adequacy in relation to nutritional status among early weaned breastfed children who are born to HIV-infected mothers. *Paediatrics*, **117** (4):701-710.

Berger, M.M., Alexander, L.S., and Chiolero, R.L. (2007). Gut absorption capacity in the critically ill. Intestinal failure: definitions and clinical consequences. *Intensive Care and Emergency Medicine*, **13**: 627-639.

Black, R.E., Brown, K.H., and Becker, S. (1984). Effects of diarrhea associated with specific enteropathogens on the growth of children in rural Bangladesh. *Paediatrics*, **73**: 799–805.

Black, R.E., Allen, L.H., Bhutta, Z.A., Caufield, L.E., de Onis, M., Ezzati, M., Mathers, C., and Rivera, J;Maternal and child undernutrition study group.(2008). Maternal and child undernutrition: global and regional exposures and health consequences. *Lancet*, **371**: 243-260.

Boaz, R. T., Joseph A. J., Kang, G., and Bose, A. (2013). Intestinal permeability in normally nourished and malnourished children with and without diarrhoea. *Indian Pediatrics*, **50** (1): 152-153.

Bundusena, A.S.L. (2003). Selected determinants and sequelae of exclusive breastfeeding up to six month in infants attending hospital and field well baby clinics. MSc dissertation, Post Graduate Institute of Medicine, University of Colombo.

Butte, N.F., Wong, W.W., Hopkinson, J.M., Heinz, C.J., Mehta, N.R., and Smith, E.O.B. (2000). Energy requirements derived from total energy expenditure and energy deposition during the first 2 years of life. *American Journal of Clinical Nutrition*, **72**:1558-69.

Campbell, D.I., Elia, M., and Lunn, P.G. (2003). Growth faltering in rural Gambian infants is associated with impaired small intestinal barrier function, leading to endotoxemia and systemic inflammation. *Journal of Nutrition* **133**:1332-1338.

Coutsoudis, A., Coovasia, H., Pillay, K., Spooner, B., and Kuhn, L. (1999). Infants-feeding pattern and HIV-1 transmission. *Lancet*, **354** (9177):471.

Department of Health/ United Nations Children's Fund (2000). South African Breastfeeding Guidelines for Health Workers. Pretoria: Government Printer.

Dewey, K.G. (2013). The Challenge of Meeting Nutrient Needs of Infants and Young Children during the Period of Complementary Feeding: An Evolutionary Perspective. *Journal of Nutrition*, **143** (12):2050-4.

Doherty, T., Sanders, D., Jackson, D., Swanevelder, S., Lombard, C., Zembe, W., Chopra, M., Goga, A., Colvin, M., Fadnes, L.T., Engebretsen, I.M., Ekström, E.C., Tylleskär, T., and PROMISE EBF study group. (2012). Early cessation of breastfeeding amongst women in South Africa: an area needing urgent attention to improve child health. *BioMed Central Pediatric*, **12**:105. Available at: <http://www.biomedcentral.com/1471-2431/12/105>

Edmond, K.M., Kirkwood, B.R., Amenga-Etego, S., Owusu-Agyei, S., and Hurt, L.S. (2007). Effect of early infants feeding practices on infection-specific neonatal mortality: an investigation of the causal links with observational data from rural Ghana. *American Journal of Clinical Nutrition*, **86**: 1126-31.

Egemen, A., Kusin, N., Aksit, S., Emek, M., and Kuugol, Z. (2002). A generally neglected threat in infant's nutrition: incorrect preparation of infant's formulae. *Turkish Journal of Pediatrics*, **44**: 298-303.

Faber, M. (2004). Complementary foods consumed by 6-12 months old rural infants in South Africa are inadequate in micronutrients. *Public Health Nutrition*, **8** (4): 373-381.

Faber, M., and Benade, A.J.S. (2001). Perception of infants cereal and dietary intakes of children aged 4-24 months in rural South Africa community. *International Journal of Food Science and Nutrition*. **52**: 359-365.

Faber, M., and Wenhold, F. (2007). Nutrition Contemporary in South Africa. *Water South Africa*, **33** (3): 393-400.

Faber, M., and Benade A.J.S. (2007). Complementary feeding and nutritional status of 6-12 months old infants in rural Kwazulu Natal. *South African Journal Clinical Nutrition*, **20** (1):16-24.

Greiner, T. (2001). International recommendations: Exclusive breastfeeding for six months: Feedback on United Nations ACC/SCN annual meeting. Available at: <http://www.who.int/inf-pr-2001/en/note2001-07.html>. Accessed 23 February 2010

Gorstein, J., and Akre, J. (1988). The use of anthropometry to assess nutritional status. *World Health Statistics*, **41**(2):48-58.

- Goto, K., Chew, F., Torun, B., Peerson, J.M., and Brown, K.H. (1999). Epidemiology of altered intestinal permeability to lactulose and mannitol in Guatemalan infants. *Journal of Pediatric Gastroenterology and Nutrition*: **28**:282–290.
- Goto, R., Panther-Brick, C., Northrop-Clewes, C.A., Manahdhar, R., and Tuladhar, N.R. (2002). Poor intestinal permeability in mildly stunted Nepali children: Association with weaning practices and *Giardia lamblia* infection. *British Journal of Nutrition*: **88**: 141-149.
- Goto, R., Mascie-Taylor, N., and Lunn, P.G. (2009). Impact of intestinal permeability status and parasitic infections on growth faltering in rural Bangladesh. *British Journal of Nutrition*: **101**: 1509-1516.
- Jayalakshmi, K., Ghoshal, U.C., Kumar, S., Misra, A., Roy, R., and Khetrapal, C.L. (2009). Assessment of small intestinal permeability using <sup>1</sup>H-NMR spectroscopy. *Journal of Gastrointestinal Liver Disease*, **18** (1):27-32.
- Kerr, R.B., Berti, P.R., and Chirwa, M. (2007). Breastfeeding and mixed feeding practices in Malawi: Timing, reasons, decision makers, and child health consequences. *Food and nutrition Bulletin*, **28** (1): 90-99.
- Kruger, R., and Gericke, G.J. (2002). A qualitative exploration of rural feeding and weaning practices, knowledge and attitude on nutrition. *Public Health Nutrition*: **6** (2): 217-223.

- Kruger, R., and Gericke, G.J. (1999). Breast-feeding practices of children (0–2 years) in a rural area in the North West Province in South Africa. *South African Medical Journal*: **89** (9): 1019–1020.
- Kramer, M.S., and Kakuma, R. (2001). The optimal duration of exclusive breastfeeding: a systematic review. World Health Organisation, Geneva.
- Labadarios, D., Steyn, N., Maunder, E., Macintyre, U., Swart, R., Gericke, G., Hukisson, J., Dannhausser, A., Vorster, H.H., and Nesamvuni, E.A. (2000). The National Food Consumption Survey (NFCS): children aged 1-9 years, South Africa, 1999. Department of Health: Directorate of Nutrition, Pretoria. 1046.
- Lopez, A.D., Salomon, J., Ahmad, O., and Murray, C.J.L. (2000). Life table for countries: data, methods and results. World Health Organization, Geneva: 9.
- Lunn, P.G., Northrop-Clewes, C.A., and Downes, R.M. (1991). Intestinal permeability, mucosal injury and growth faltering in Gambian infants. *Lancet* **338**: 907–910.
- Lunn, P.G. (2000). The impact of infection and nutrition on gut function and growth in childhood. *Proceedings of the Nutrition Society*, **59**:147-154.
- Lutter, C.K., Daelmans, B.M., de Onis, M., Kothari, M.T., Ruel, M.T., Arimond, M., Deitchler, M., Dewey, K.G., Blössner, M., and Borghi, E. (2011). Undernutrition, poor feeding practices, and low coverage of key nutrition interventions. *Pediatrics*, **128** (6):1418-27.

- Madiba, S., and Letsoalo, R. (2013). HIV disclosure to partners and family among women enrolled in prevention of mother to child transmission of HIV program: implications for infant feeding in poor resourced communities in South Africa. *Global Journal of Health Sciences*, **5** (4):1-13.
- Mamabolo, R.L., Albert, M., Mbenyane, G.X., Steyn, N.P., and Nthangeni, N.G. (2004). Feeding practices and growth of infants from birth to 12 month in the central region of the Limpopo province of South Africa. *Nutrition in Africa*, **20**:327-333.
- Marques, N.M., Lira, P.I.C., Lima, M.C., da Silva, N.L., Filho, M.B., Huttly, S.R.A., and Ashworth, A. (2001). Breastfeeding and early weaning practices in Brazil: a longitudinal study. *Pediatrics*, **108**: 66.
- Mehrshahi, S., Kabir, I., Roy, S.K., Agho, K.E., Senarath, U., Dibley, M.J., and South Asia Infant Feeding Research Network. (2010). *Food Nutrition Bulletin*, **31** (2): 295-313.
- Millennium Development Goals Report. (2005). United Nations, New York. Available at: millenniumindicators.un.org. Accessed 2009.
- Mondal, D., Minak, J., Alam, M., Liu, Y., Dai, J., Korpe, P., Liu, L., Haque, R., and Petri Jr, W.A. (2012). Contribution of enteric infection, altered intestinal barrier function, and maternal malnutrition to infant in Bangladesh. *Clinical Infectious Diseases*, **54** (2): 185-92.

- Mushaphi, L.F., Mbhenyane, X.G., Khoza, L.B., and Amay, A.K.A. (2008). Infants feeding practices of mother and the nutritional status in the Vhembe District of Limpopo Province. *South African Journal of Clinical Nutrition*, **21** (2):36-41.
- NFCS-FB-I. (2007). Executive Summary of the National Food Consumption Survey fortification Baseline, South Africa, 2005.
- National Institute of Population Research and Training (NIPORT), Mitra and Associates (MA), and ORC Macro (ORCM). 2007. Bangladesh Demographic and Health Survey 2007. Daka, Bangladesh and Calverton, Maryland (USA): *National Institute of Population Research and Training*.
- Nwankwo, B.O., and Brieger, W.R. (2002). Exclusive breastfeeding is undermined by use of other liquids in rural south western. *Nigeria Journal of Tropical Paediatrics*, **48**:109-11.
- Onayade, A., Abiona, T. C., Abayomi, I.O., and Makanjuola, R.O. (2004). The first six month growth and illness of exclusively and non-exclusively breast-fed infants in Nigeria; **81**(3):146-53.
- Patel, A., Badhoniya, N., Khadse, S., Senarath, U., Agho, K. E., and Dibley, M. J. (2010). Infant and young child feeding indicators and determinants of poor feeding practices in India: Secondary data analysis of National Family Health Survey 2005–06. *Food and Nutrition Bulletin*. **31**(2): 314-333(20).

- Pelletier, D. L., Frongillo, E.A., and Jr. Habicht, J.P. (1993). Epidemiologic evidence for potential effect of malnutrition on child mortality. *American Journal of Public Health*, 83: 130-1133.
- Przyrembel, H. (2012). Timing of introduction of complementary food: short and long term health consequences. *Annals of Nutrition and Metabolism*, 60 (2): 8-20.
- Radwan, H. (2013). Patterns and determinants of breastfeeding and complementary feeding practices of Emirati Mothers in the United Arab Emirates. *BioMed Central Public Health Journal*. 13:171.
- Raphael, D. (1984). Weaning is always: The anthropology of breastfeeding behaviour. *Ecology of Food and Nutrition*, 15: 203-213.
- Reyes, H., Zapata, R., Hernández, I., Gotteland, M., Sandoval, L., Jirón, M.I., Palma, J., Almuna, R., and Silva, J.J. (2006). Is a leaky gut involved in the pathogenesis of intrahepatic cholestasis of pregnancy? *Hepatology*, 43 (4):715-22.
- Riordan, J. (1999). The biological specificity of breast milk: breast milk and human lactation. (Eds). Riordan, J and Bartlet Publishers: Boston.
- Saldiva, S.R.M., Escuder, M.M., Mondini, L., Levy, R.B., and Venancio, S.I. (2007). Feeding habits of children aged 6-12 months and associated maternal factors. *Journal of Paediatrics*, 83 (1): 53-8.

- Senarath, U., Dibley, M.J., Godakandage, S.S., Jayawickrama, H., Wickramasinghe, A., Agho, K.E., and South Asia Infant Feeding Research Network. (2010). Determinants of infant and young child feeding practices in Sri Lanka: secondary data analysis of Demographic and Health Survey 2000. *Food and Nutrition Bulletin*, 31(2):352-65.
- Scott, J.A., Binns, C.W., Graham, K.I., and Oddy, W.H. (2009). Predictors of early introduction of solids in infants: results of a cohort study. *BioMed Central Pediatrics*, 9:60. Available at: <http://www.biomedcentral.com/1471-2431/9/60>. Accessed 23 May 2013.
- Shamim, S. (2005). Weaning practices in peri-urban low-socioeconomic groups. *Journal of the College of Physicians and Surgeons- Pakistan*, 15 (3): 129-32.
- Shrimpton, R., Victora, C.G., de Onis, M., Lima, R.N.C., Blossner, M., and Clugston, G. (2001). Worldwide timing of growth faltering: implications for nutritional interventions. *Paediatrics*, 107 (75): 1-7.
- Sibeko, L., Dhansay, M.A., Charlton, K.E., Johns, T., and Gray-Donald, K. (2005). Beliefs, attitudes, and practices of breastfeeding mothers from a periurban community in South Africa. *Journal of Human Lactation*, 21(1):31-8.
- Solomons, N.W. (2007). Malnutrition and infection: an update. *British Journal of Nutrition*, 98 (1): 5-10.
- South African Demographic and Health Survey. (2003). South African Demographic and Health Survey. Pretoria.

South African Vitamin A Consultative Group. (1995). Children aged 6 to 71 months in South Africa, 1994 their anthropometric, vitamin A, iron and immunisation coverage status. Isando. Available at: <http://www.sahealthinfo.org.za>. Accessed 23 February 2010,

Steyn, N.P., Badenhorst, C.J., Nel, J.H., and Ladzani, R. (1993). Breastfeeding and weaning practices of Pedi mothers and the dietary intakes of their preschool children. South African *Journal of Food Sciences and Nutrition*, **5** (1): 10-3.

Taylor, S.N., Basile, L.A., Ebeling, M., and Wagner, C.L. (2010). Intestinal permeability in preterm infants by feeding type: mother's milk versus formula. *Breastfeeding Medicine*, **4** (1):11-15.

Tampah-Naah, M., and Kumi-Kyereme, A. (2013). Determinants of exclusive breastfeeding among mothers in Ghana: a cross-sectional study. *International Breastfeeding Journal*, **8**:13

Ukpe, I.S., Blitz, J., Hugo, J., and Theledi, M. (2009). The Infants-feeding practices of mothers enrolled in the prevention of other-to- child transmission of HIV programme at a primary health care clinic in the Mpumalanga Province, South Africa. *South African Family Practice*, **51**(4) 337-339.

Widstom, A.M, Wahlberg, V., Matthiesen, A.S., Eneroth, P., Uvnas-Moberg, K., and Werner, S. (1990). Short-term effect of early suckling and touch of the nipple on maternal behaviour. *Early Human Development*, **21**:153-63.

Wijndaele, K., Lakshman, R., Landsbaugh, J.R., Ong, K.K., Ogilvie, D. (2009). Determinants of early weaning and use of unmodified cow's milk in infants: a systematic review. *Journal of American Diet Association*, **109** (12):2017-28.

World Health Organization. (2002). Breastfeeding and Replacement Feeding Practices in the Context of Mother-to-child Transmission of HIV: An Assessment Tool for Research. Available at: WHO/RHR/01.12,WHO/CAH/01.2.1. Accessed 12 February 2010.

World Health Organisation Child Growth Standard. (2006). Available at: [www.who.int/childgrowth/en](http://www.who.int/childgrowth/en). Accessed 20 December 2013.

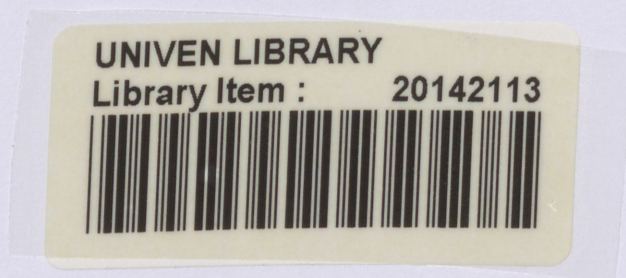
World Health Organization. (2007). Evidence on the long-term effects of breastfeeding: systematic reviews and meta-analyses. Geneva. Available at: [http://www.who.int/child-adolescent-health/New\\_Publications/NUTRITION/](http://www.who.int/child-adolescent-health/New_Publications/NUTRITION/). Accessed 10 January 2010.

World Health Organization. (2008a). Indicators for assessing infants and young child feeding practices: Part 1 definitions. Geneva, World Health Organization.

World Health Organization. (2008b). The International Code of Marketing of Breastmilk Substitutes: Frequently asked questions; (updated version 2008). Geneva, Switzerland. Available at: <http://www.who.int/child-adolescent-health/>. Accessed 12 February 2010.

World Health Organisation. (2010). Guidelines on HIV and infant feeding 2010: Principles and recommendation for infant feeding in the context of HIV and a summary of evidence. Geneva, World Health organisation.

## 8. APPENDICES



8.1 Appendix A

Participant ID:

If no response for any question, write 'NA' in the response column.

#	Question	Code	Response
81	Fieldworker ID		<input type="checkbox"/>
82	Today's date (DD/MM/YYYY)		<input type="checkbox"/>
83	Are there plans to start a committee of the community within the month?		<input type="checkbox"/>
84	Is the number >10 members age?		<input type="checkbox"/>
85	Does the number have number >10 members in ID study?		<input type="checkbox"/>
86	Was this a meeting program?		<input type="checkbox"/>
87	Is the child healthy? (Does not have respiratory disease / acute or chronic diarrhea, malnutrition, prolonged fever, convulsion)		<input type="checkbox"/>
88	Is the mother able to give informed consent?		<input type="checkbox"/>

**8. APPENDICES**

## 8.1 Appendix A

Participant ID:

### SCREENING FORM (SCR)

If no response for any question, write NA as a response.

#	Question	Code	Response
01	Fieldworker ID		<input type="text"/> <input type="text"/>
02	Today's date (DD/MMM/YY)	Yes = 01 No = 00	<input type="text"/> <input type="text"/> / <input type="text"/> <input type="text"/> <input type="text"/> / <input type="text"/> <input type="text"/>
03	Are there plans to move outside of the community within six months?	Yes = 01 No = 00	<input type="text"/> <input type="text"/>
04	Is the mother <16 years of age?	Yes = 01 No = 00	<input type="text"/> <input type="text"/>
05	Does the mother have another child in the MAL-ED study?	Yes = 01 No = 00	<input type="text"/> <input type="text"/>
06	Was this a multiple pregnancy?	Yes = 01 No = 00	<input type="text"/> <input type="text"/>
07	Is the child healthy? (does not have congenital diseases / severe neonatal disease requiring prolonged hospitalization)	Yes = 01 No = 00	<input type="text"/> <input type="text"/>
08	Is the mother able to give informed consent?	Yes = 01 No = 00	<input type="text"/> <input type="text"/>

## 8.2 Appendix B

CHILD ASSESSMENT FORM (CAF)			
If no response for any question, write NA as a response.			
#	Question	Code	Response
01	Fieldworker ID		<input type="text"/> <input type="text"/> <input type="text"/>
02	Today's date (DD/MMM/YY)		<input type="text"/> <input type="text"/> / <input type="text"/> <input type="text"/> <input type="text"/> / <input type="text"/> <input type="text"/>
03	Screen complete and Consent obtained	Yes = 01 No = 00	<input type="text"/> <input type="text"/>
04	Date of birth (DD/MMM/YY)		<input type="text"/> <input type="text"/> / <input type="text"/> <input type="text"/> <input type="text"/> / <input type="text"/> <input type="text"/>
05	Sex of a child	Male=01 Female=02	<input type="text"/> <input type="text"/>
06	Birth weight (kg) * (from birth record, if available)		<input type="text"/> <input type="text"/> . <input type="text"/> <input type="text"/>
07	How long after childbirth did the mother begin breastfeeding him/her?	00 = within 1 hour 01 = 1-24 hours; 02 = 1-3 days 03 = 4+ days later NA = never breastfed	<input type="text"/> <input type="text"/>
08	Was the child fed the first milk (colostrum)?	Yes = 01 No = 00	<input type="text"/> <input type="text"/>
09	Was there pre-lacteal feeding of the child?	Yes = 01 No = 00	<input type="text"/> <input type="text"/>
10	Current weight (kg) *		<input type="text"/> <input type="text"/> . <input type="text"/> <input type="text"/>
11	Current length (cm)		<input type="text"/> <input type="text"/> <input type="text"/>
12	Current head circumference (cm)		<input type="text"/> <input type="text"/> <input type="text"/>

### 8.3 Appendix C

#### BASELINE DEMOGRAPHIC QUESTIONNAIRE (DAF)

If no response for any question, write NA as a response.

#	Question	Code	Response
01	Fieldworker ID		<input type="text"/> <input type="text"/> <input type="text"/>
02	Date (DD/MMM/YY)		<input type="text"/> <input type="text"/> / <input type="text"/> <input type="text"/> <input type="text"/> / <input type="text"/> <input type="text"/>
03	What is your age?	10-99 (years)	<input type="text"/> <input type="text"/>
04	(Record sex)	Male= 01; Female=02	<input type="text"/> <input type="text"/>
05	What is your relationship to [child's name]?	Father= 01; Mother= 02; Grandmother= 03; Grandfather=04; Sibling=05; Other=06	<input type="text"/> <input type="text"/>
06	Are you currently married, divorced, widowed, or never married? <i>If never married, skip to question 8.</i>	Never married=01 Married=02 Divorced=03 Widowed=04	<input type="text"/> <input type="text"/>
07	How old were you when you got married for the first time?	08-50 (years)	<input type="text"/> <input type="text"/>
08	Have you ever attended school? If no, skip to question 11.	Yes = 01, No = 00	<input type="text"/> <input type="text"/>
09	How many years of schooling have you completed?	00-20	<input type="text"/> <input type="text"/>
10	If younger than 25 years old: are you currently attending school or college?	Yes = 01, No = 00	<input type="text"/> <input type="text"/>
<i>The remaining questions are for the mother of the child. If the mother is living but unavailable, return to the home at a later time to complete this questionnaire.</i>			
11	How old are you?	Yes = 01, No = 00	<input type="text"/> <input type="text"/>
12	Are you currently married, divorced, widowed, or never married? <i>If never married, skip to question 8.</i>		<input type="text"/> <input type="text"/>
13	How old were you when you got married		<input type="text"/> <input type="text"/>





SAF/SAR/v2.3(03)MAY10

Child ID:

MM/YY:  /



Date	M A I J J																																					
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31							
<b>What liquids are the child consuming? (ask about past 24 hours and fill in column corresponding to yesterday's date) (Yes=1, No=0, NA)</b>																																						
28	Breast milk																																					
29	# times during night																																					
30	# times during day																																					
31	Infant formula																																					
32	# times during night																																					
33	# times during day																																					
34	Animal milk - Tinned, powdered, or fresh																																					
35	# times during night																																					
36	# times during day																																					
37	Plain water																																					
38	Tea, coffee, local examples																																					
39	Fruit juices																																					
40	Other liquids																																					
<b>What solid foods are the child consuming? (ask about past 24 hours and fill in column corresponding to yesterday's date) (Yes=1, No=0, NA)</b>																																						
41	Semi-solid, mashed or solid foods? If NO, then stop																																					
42	Porridges, breads, other foods/drinks made from wheat, oat, barley, soy?																																					
43	Rice?																																					
44	White potatoes, white yams, manioc, other roots?																																					
45	Peanuts, spreads with peanuts?																																					
46	Beans, peas, lentils?																																					
47	Bananas?																																					
48	Yogurt, kefir?																																					
49	Onions, leeks, shallots, garlic?																																					
50	Fermented foods or drinks?																																					
51	Supervisor check (initial)																																					

## 8.5 Appendix E

### MONTHLY ASSESSMENT FORM A (MOA)

*If no response for any question, write NA as response*

#	Question:	Code:	Response:
<b>Urine collection</b>			
01	Study researcher/fieldworker ID:	###	<input type="text"/> <input type="text"/> <input type="text"/>
02	Today's date	DD/MMM/YY	<input type="text"/> <input type="text"/> / <input type="text"/> <input type="text"/> <input type="text"/> / <input type="text"/> <input type="text"/>
<b>Nutrition</b>			
03	Are you breastfeeding <Child>? If No, then skip to Q.6	Yes=01, No=00	<input type="text"/> <input type="text"/>
04	How many times did you breastfeed<Child> last night?	00-99	<input type="text"/> <input type="text"/>
05	During the day yesterday, how many times did you breastfeed<Child>?	00-99	<input type="text"/> <input type="text"/>
06	Do you give<Child>infant formula? If No, then skip to Q.9	Yes=01, No=00	<input type="text"/> <input type="text"/>
07	How many times did you feed <Child> formula from sunset to sunrise?	00-99	<input type="text"/> <input type="text"/>
08	Yesterday, during the day, how many times did you feed<Child> formula?	00-99	<input type="text"/> <input type="text"/>
09	Do you give <Child> other milk, such as tinned, powdered or fresh animal milk? If No, then skip to Q.12	Yes=01, No=00	<input type="text"/> <input type="text"/>
10	Last night, how many times did you feed <Child> animal milk from sunset to sunrise	00-99	<input type="text"/> <input type="text"/>
11	Yesterday, during the day, how many times did you feed <Child> animal milk?	00-99	<input type="text"/> <input type="text"/>
Yesterday, during the day or last night, did <Child> have			
12	Plain water	Yes=01, No=00	<input type="text"/> <input type="text"/>
13	Tea, coffee <local example>?	Yes=01, No=00	<input type="text"/> <input type="text"/>
14	Fruit or vegetable juice?	Yes=01, No=00	<input type="text"/> <input type="text"/>
15	Any other liquids, such as sugar water, thin soup or broth, carbonated drinks <local examples>	Yes=01, No=00	<input type="text"/> <input type="text"/>
16	IS <Child> eating any semi-solid, mashed or	Yes=01,	<input type="text"/> <input type="text"/>

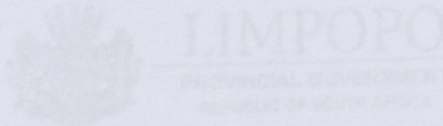
	solids> If No, go to Q.32	No=00	
Thinking about yesterday, during the day and at night, did<Child> have any of these foods, even if they were in combination with other foods?			
17	Rice, porridge, bread, noodles or other foods made from grains?	Yes=01, No=00	<input type="checkbox"/> <input type="checkbox"/>
18	White potatoes, white yams, manioc, or other food made from roots?	Yes=01, No=00	<input type="checkbox"/> <input type="checkbox"/>
19	Carrot, squash, or sweet potatoes that are yellow or orange inside?	Yes=01, No=00	<input type="checkbox"/> <input type="checkbox"/>
20	Any green leafy vegetables such as spinach?	Yes=01, No=00	<input type="checkbox"/> <input type="checkbox"/>
21	Food made with beans, lentils, peas, corn, ground nuts?	Yes=01, No=00	<input type="checkbox"/> <input type="checkbox"/>
22	Ripe mangoes, papayas, or other sweet yellow/orange or red fruit?	Yes=01, No=00	<input type="checkbox"/> <input type="checkbox"/>
23	Any other fruit or vegetable such as banana, apple, orange, tomatoes, avocados?	Yes=01, No=00	<input type="checkbox"/> <input type="checkbox"/>
24	Liver, kidney, heart, or organ meats?	Yes=01, No=00	<input type="checkbox"/> <input type="checkbox"/>
25	Fresh or dried fish or shellfish?	Yes=01, No=00	<input type="checkbox"/> <input type="checkbox"/>
26	Cheese, yogurt, or other dairy products?	Yes=01, No=00	<input type="checkbox"/> <input type="checkbox"/>
27	Any sugary, foods such as pastries, cakes or biscuit?	Yes=01, No=00	<input type="checkbox"/> <input type="checkbox"/>
28	Any commercial available food for infants or young children?	Yes=01, No=00	<input type="checkbox"/> <input type="checkbox"/>
29	Yesterday, counting meals and snacks, how many times did you feed <Child>?	00-909	<input type="checkbox"/> <input type="checkbox"/>
30	How would you describe your child's appetite? Would you say it is:	01=poor 02=fair 03=good, 04=very good	<input type="checkbox"/> <input type="checkbox"/>
31	Does the salt that you cook with have iodine in it?	Yes=01, No=00	<input type="checkbox"/> <input type="checkbox"/>
32	Do you wash the salt before using it?	Yes=01, No=00	<input type="checkbox"/> <input type="checkbox"/>
33	Does <Child> routinely eat things that are non-food items like ash, clay. Paint chips, dirt, starch, cigarettes or cigarette butts?	Yes=01, No=00	<input type="checkbox"/> <input type="checkbox"/>
<b>Anthropometry (from 1-12 months)</b>			
34	Weight (kg)		<input type="checkbox"/> <input type="checkbox"/> . <input type="checkbox"/> <input type="checkbox"/>
35	Length /height (cm)		<input type="checkbox"/> <input type="checkbox"/> <input type="checkbox"/> <input type="checkbox"/>

8.7 Appendix G: Ethical Clearance Certificate

8.6 Appendix F

URINE COLLECTION FORM (UCF)			
<i>If no response for any question, write NA as response</i>			
#	Question:	Code:	Response:
<b>Urine collection</b>			
01	Fieldworker ID:	###	<input type="text"/> <input type="text"/> <input type="text"/>
02	Date	DD/MMM/YY	<input type="text"/> <input type="text"/> <input type="text"/> <input type="text"/> <input type="text"/> <input type="text"/> <input type="text"/> <input type="text"/> <input type="text"/>
03	Start time (solution ingestion completion)	Time (24 Hr Scale; HH:MM)	<input type="text"/> <input type="text"/> : <input type="text"/> <input type="text"/>
04	End time of urine collection	Time (24 Hr Scale; HH:MM)	<input type="text"/> <input type="text"/> : <input type="text"/> <input type="text"/>
05	Total urine volume (ml)	###	<input type="text"/> <input type="text"/> <input type="text"/>
06	Time of departure from field	Time (24 Hr Scale; HH:MM)	<input type="text"/> <input type="text"/> : <input type="text"/> <input type="text"/>
<b>Urine transport and receiving</b>			
07	Lab technician ID	###	<input type="text"/> <input type="text"/> <input type="text"/>
08	Was a 2mL aliquot of urine without chlorhexidine obtained	Yes=01, No=00	<input type="text"/> <input type="text"/>
09	Was a 5-15mL aliquot of urine with chlorhexidine obtained	Yes=01, No=00	<input type="text"/> <input type="text"/>
10	Time of arrival at field research lab	Time (24 Hr Scale; HH:MM)	<input type="text"/> <input type="text"/> : <input type="text"/> <input type="text"/>
11	Sample ID		<input type="text"/> <input type="text"/> <input type="text"/> <input type="text"/> <input type="text"/> <input type="text"/> <input type="text"/> <input type="text"/> <input type="text"/> <input type="text"/>
Observations			

## 8.7 Appendix G: Ethical Clearance Certificate



RESEARCH AND INNOVATION

OFFICE OF THE DIRECTOR

### Ethical Clearance Certificate

Name of Researcher/Investigator: Dr. P. Bessong

**TITLE OF PROJECT:** "Etiology, Risk Factors, and Interactions of Enteric Infections and Malnutrition and its Consequences for Child Health and Development."

Project Number: SMNS/09/MBY/004

### SUPERVISORS/CORESEARCHERS/COINVESTIGATORS

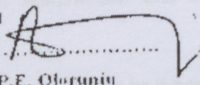
NAME	INSTITUTION & DEPARTMENT	ROLE
Dr. P. Bessong	University of Venda	Principal Investigator
Prof. Tholene Sudi	University of Venda	Researcher
Ms. Cebisa Nesamang	University of Venda	Researcher
Dr. Aaron Emyun-huyu	Songazwi Medical Center	Researcher
Dr. Amidou Samie	University of Venda	Researcher
Prof. Yhaurani Netshandama	University of Venda	Researcher
Mr. Richard Muliyil	University of Venda	Researcher
Dr. Rebecca Billingham	University of Virginia	Co-Principal Investigator
Prof. Richard L. Guerrant	University of Virginia	Principal Investigator
Ms Leah Barrett	University of Virginia	Researcher

### ISSUED BY:

University of Venda, Health, Safety and Research Ethics Committee

Date Considered: 24 June 2009

Decision by Committee: Ethical Clearance granted

Signature of Chairperson of the Committee: 

Name of the chairperson of the Committee: Prof. P.E. Olorunju



UNIVERSITY OF VENDA  
PRIVATE BAG X1550, THOHoyANDOU 0950, LIMPOPO PROVINCE, SOUTH AFRICA  
TELEPHONE 018 962 8504 8313 7-24 FAX 018 962 8435 4715

## 8.8 Appendix H: Department of Health Provincial Approval



**LIMPOPO**  
PROVINCIAL GOVERNMENT  
REPUBLIC OF SOUTH AFRICA

### DEPARTMENT OF HEALTH AND SOCIAL DEVELOPMENT

Enquiries: Ramalivhana NJ

Ref: 4/2/2

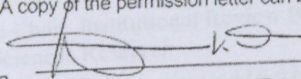
8 July, 2009  
Dr. Bessong .P  
University of Venda  
P/Burg x 5050  
THOHOYANDOU  
0950

Dear Dr. Bessong .P

"Etiology, Risk Factors, and Interactions of Enteric Infections and Malnutrition and the Consequences for Child Health and Development.

Permission is hereby granted to Dr Bessong P to conduct a study in Limpopo Province, South Africa

- The Department of Health and Social Development will expect a copy of the completed research for its own resource centre after completion of the study.
- The researcher is expected to avoid disrupting services in the course of his study
- The Researcher/s should be prepared to assist in interpretation and implementation of the recommendations where possible
- The Institution management where the study is being conducted should be made aware of this,
- A copy of the permission letter can be forwarded to Management of the Institutions concerned

7  10/09/09  
HEAD OF DEPARTMENT  
HEALTH AND SOCIAL DEVELOPMENT  
LIMPOPO PROVINCE

Private Bag X9302 Polokwane  
18 College Str., Polokwane 0700 • Tel: 015 293 6000 • Fax: 015 293 6211 • Website: <http://www.limpopo.gov.za>

*The heartland of Southern Africa – Development is about people!*

## 8.9 Appendix I: Institutional Review Board Approval

<p>University of Virginia  <b>Institutional Review Board for Health Sciences Research</b>          Protection of Human Subjects Approval          Assurance Identification/Certification/Declaration          (Common Federal Rule)</p>		
<p><b>HSR # 14269</b></p>		
<p>Event: Approval Grant Continuation - Expedited</p>	<p>Type: Grant</p>	<p>Sponsor: Foundation for the National Institutes of Health, FNIH</p>
<p>Principal Investigator: Richard Guerrant, MD</p>		
<p>Title: Etiology, Risk Factors and Interactions of Enteric Infections and Malnutrition and the Consequences for Child Health and Development (FNIH)</p>		
<p>Assurance: Federal Wide Assurance (FWA)#: 00006183</p>		
<p>Certification of IRB Review: The HSR abides by 21CFR50, 21CFR56, 45CFR46, 45CFR160, 45CFR164 and ICH guidelines. This activity has been reviewed and approved by the IRB in accordance with these regulations.</p>		
<p>Approval Date: 02/15/11          Grant Expiration Date: 02/14/12          Approved to Enroll a total of 0 subjects.</p>	<p>Comments: 9. Continuing review of research, not conducted under an investigational new drug application or investigational device exemption where categories (2) through (8) do not apply but the IRB has determined and documented at a convened full IRB meeting that the research involves no greater than minimal risk and no additional risks have been identified</p>	
<p>Current Status: Funded - Active Protocol(s) with Human Participant</p>		
<p>Consent Version Dates:</p>		
<p>Committee Members (did not vote):</p>		
<p>The official signing below certifies that the information provided above is correct and that, as required, future reviews will be performed and certification will be provided.</p>		
<p>Name: Lynn R. Noland, RN PhD          Title: Vice Chair, Institutional Review Board for Health Sciences Research          Phone: 434-924-9634 Fax: 434-924-2932</p>	<p>Name and Address of Institution:          Institutional Review Board for Health Sciences Research          PO Box 800483          University of Virginia          Charlottesville, VA 22908</p>	
<p>Signature: <i>Lynn R. Noland</i></p>		<p>Date: FEB 15 2011</p>

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## 8.10 Appendix J: Participants Informed Consent Form

<b>Enteric Infections and Malnutrition</b>		<b>Study Site: South Africa</b>	
IRB-HSR # 14595	<b>COHORT Study</b>	Version #5	11Sep09
INTERNATIONAL RESEARCH INFORMED CONSENT (ICF)		Page 1 of 4	



**Participant names:**

**Infant:** \_\_\_\_\_ **PID #** \_\_\_\_\_

**Mother:** \_\_\_\_\_ **PID #** \_\_\_\_\_

### —CONSENT TO PARTICIPATE IN A RESEARCH STUDY—

#### What is this study about?

Doctors and faculty at the University of Venda, South Africa and at institutions in the United States are trying to learn more about the causes of malnutrition in young children in our country and other countries around the world, including Bangladesh, Brazil, India, Nepal, Pakistan, Peru and Tanzania. Malnutrition is a disease where inadequate amounts of nutrients are obtained from the food that we eat. It has several causes. One cause is the lack of access to adequate amounts of food. In addition, malnutrition is associated with infections of our intestines that can change our use of nutrients. It is also possible that genes make some children more susceptible to malnutrition than others. Genes are made up of material known as DNA. They are inherited from our parents, and humans also carry genes from the trillions of friendly microbes that live on and in our bodies. This effort to learn more about the causes of malnutrition in young children is called a research study. Research ethics committees in the United States (at the University of Virginia, Johns Hopkins, Washington University in St. Louis and the University of Colorado at Boulder) as well as in your community have approved this study.

The reason to do this research study is that malnutrition is a great problem in our country and your child's participation in this study may lead to new ways of preventing or treating malnutrition. In this sense, one possible benefit of participating may be to improve the health of those who live in our country and other countries.

You are asked to be a participant in this research study because your child's participation may help us identify what causes malnutrition in children during the first 2 years of life. This is a time when malnutrition can be very common and can lead to long-term or life-long disabilities if not treated properly.

The researchers in charge of this study are Dr Pascal Bessong and Dr Richard Guerrant, University of Virginia.

This study will take place at the University of Venda.

#### What will happen during the study?

If you agree that your child can participate in this study, we will do the following:

- Visit you two times a week for 24 months to monitor your child for diarrhea and general health.
- Visit you (mother) at the beginning of the study to measure your height and weight and collect a small amount of saliva and/or blood (equivalent to a teaspoon) to determine your blood group and to isolate your human genes (DNA). (Your decision to participate in this part of the study is optional and won't affect your child's participation.)

APPROVAL DATE 8/10/10  
EXPIRATION DATE 8/9/11

<b>Enteric Infections and Malnutrition</b>		<b>Study Site: South Africa</b>	
IRB-HSR # 14595	<b>COHORT Study</b>	Version #5	11Sep09
INTERNATIONAL RESEARCH INFORMED CONSENT (ICF)		Page 2 of 4	



- Visit once per month for months 0-24 (25 times) to:
  1. Collect child feeding practices
  2. Measure his/her height, weight, skin-fold, arm and head circumference
  3. Collect your child's stool samples at each of these visits and whenever he/she suffers from diarrhea to identify and/or isolate microbes and, on certain occasions, study the stool's microbial genes (DNA) and their products.
- When your child is about 7 and 15 months of age (2 times):
  1. Collect a small amount of saliva and/or blood (equivalent to a teaspoon) to isolate your child's genes (DNA) and to test the level of nutrients and antibodies that your child produced in response to childhood vaccines.
- When your child is about 3, 6, 9 and 15 months of age (4 times):
  1. Have your child drink a sugar solution and collect his/her urine for 5 hours afterwards.
- Periodically perform interviews:
  1. Assessing you (mother) and your household:
    - Interview you in order to gather information about your household and about your mood, when your child is about 1, 6, 15, and 36 months of age.
    - In a private room at a community center/clinic, we will measure your ability to reason and solve problems when your child is about 9 months of age.
  2. Assessing your child:
    - Interview you (mother) to ask about your child's language abilities (when your child is about 9, 15, and 36 months of age) and your child's personality and home environment (when your child is about 6 and 36 months of age).
    - Will take you and your child to a private room at a community center/clinic where we will measure your child's mental and motor development as well as record your child's behavior when your child is about 6, 15, and 36 months of age (and possibly at 5-7 years old).

There is a possibility that these assessments will be video recorded so that we can check the quality of the test administration.
- If your child is referred to a health center, hospital or physician, we may obtain your child's medical records/test results.

We will store your and your child's saliva and/or blood, urine, stool and DNA collected as a part of this study in secure storage chambers, such as freezers, that are located in rooms that can only be accessed by members of our scientific team. Some of these samples collected may be shipped to a laboratory in another country for specialized testing. All specimens will be coded with a number so no identifying information (ie. your name or address) will be stored with your specimens or test results. If you change your mind about having your specimens saved for further testing, you may contact us and the samples will be destroyed and not saved.

### **Could the research help me?**

People might have good things happen to them because they are in research studies. These are called benefits. The benefits to you of being in this study might be that we will be able to determine the cause of your child's diarrhea, should it occur, and will refer you to local health care providers for appropriate treatment. Your child will also benefit from the growth monitoring and promotion that will take place with each month's weighing. The doctor and/or the researcher will inform you of any relevant information found from the conduct of this study that is important to your personal medical care or situation.

<b>Enteric Infections and Malnutrition</b>		<b>Study Site: South Africa</b>	
IRB-HSR # 14595	<b>COHORT Study</b>	Version #5	11Sep09
INTERNATIONAL RESEARCH INFORMED CONSENT (ICF)		Page 3 of 4	



### **Could the research hurt me?**

Sometimes things happen to people in research studies that may hurt them or make them feel bad. These are called risks. The risks of this study are that you may feel uncomfortable answering some of the questions on the mood questionnaire and that you and your child may have mild, temporary pain when the needle is inserted during the blood collection and/or have a bruise where the blood is taken. Since there is a slight risk that the information we collect in this study could be used to identify you and your child, we will take great care to protect your personal information.

### **How will my privacy be protected?**

Study records that identify you (ie. your name and address) will be kept confidential and locked in a secure place under the responsibility of the local principal investigator. As noted above, your child's specimens and test results will be coded and stored separately for analysis by the researchers who are working together on this project at the different study sites. In other words, by using this approach we will help ensure that none of these researchers at these different study sites or elsewhere will be able to identify your child as the donor of these specimens. Other persons who may have access to your test results and consent form include groups which oversee the safety of a study including accrediting agencies, South Africa and United States federal, state and local agencies having oversight of this research, for example, the University of Virginia, Washington University, University of Colorado, and Johns Hopkins University Research Compliance staff and Institutional Review Board (IRB) members or designates. The IRB is a special committee at these Universities that reviews all medical research studies involving human participants and is similar to the Health, Safety and Research Ethics Committee of the University of Venda. In addition, other authorized researchers (profit or non-profit) may have access to these data. Because of the potential need to release information to these parties and because the genetic information we are collecting is potentially identifying, we cannot guarantee absolute confidentiality but we will make every effort that we can to preserve confidentiality.

In the future, we may wish to perform additional studies that measure the molecules present in blood and stool specimens that already have been collected. As with all tests on all samples collected, the individuals measuring these molecules will not know who has donated the samples because they will be coded.

If you sign this form, you have given us permission to release information to authorized researchers and the safety committee. There is no expiration date to this permission. This information, like all of the information generated from this study, will be coded so it is unlikely that anyone will be able to trace it to you or your child: in other words, your and his/her privacy will be protected.

In addition, we also ask your permission to publically release information about the bugs (bacteria) in your child's gut. This would be released only with your permission and with no information about you or your child associated with it.

Your participation in this research study is voluntary. However, you will not be allowed to participate in this research if you do not sign this form.

### **Do I have to participate?**

You do not have to be in this study if you do not want to. This means your participation is voluntary. It is up to you to decide whether or not being in the study is in your best interest.