

Nutritional status, feeding practices and motor development of 6-month-old infants

A.M.P Rothman

12978361

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Promoter: Dr. N.M Covic

Co-Promoters: Prof. M Faber

Prof. C.M Smuts

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PREFACE

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ABSTRACT

Background

Inadequate nutrition and development in the first thousand days have significant public health implications which include long-term effects on cognitive development and school achievement. Iron is one of the key nutrients needed for long-term growth and cognitive development. A strategy to address poor nutrition in infancy is the use of various home fortification products for example small-quantity lipid based nutritional supplements (SQ-LNS).

Aim

This study investigated the nutritional status, early feeding practices and psychomotor development of 6-month-old infants from a peri-urban community in Klerksdorp (North West Province), South Africa, as well as the acceptance of 2 novel SQ-LNS for complementary feeding by infants and their primary caregiver.

Design

The study used baseline data of a randomized controlled trial and had a cross-sectional design that included 750 6-month-old infants from a peri-urban community in the North West province in South Africa. Early feeding practices and dietary intake were investigated in association with nutritional status and psychomotor development.

For the acceptability trial, mother/infant pairs were enrolled in a two-part trial. Part 1 (n= 16) was a test-feeding trial with a cross-over, randomized design in which a five-point hedonic scale was used for sensory evaluation (disagree= 1, agree= 5). Part 2 (n= 38) was a two-week, home-use trial followed by focus group discussions.

Results

For 48.9% of the infants, exclusive breastfeeding was ceased at the age of 0–2 months. Semi-solid and/or solid foods were introduced mostly at the age of 3–4 months. At the age of 6 months, 70.1% of infants were still being breastfed. Frequently consumed complementary foods were mainly infant cereal (68.1%), formula milk (42.0%) and jarred infant foods (22.7%). Dietary intake data showed that for more than 80% of the infants, the nutrient density (amount of nutrient per 100 kcal) for iron, zinc, and calcium of the complementary diet was lower than the desired density.

With regards to nutritional status, 36.4% of the infants were anaemic (Hb < 11 g/dl), 16.1% were iron deficient (plasma ferritin concentration <12 µg/l), and 9.3% of the infants suffering from iron deficiency anaemia. The anthropometric data showed that 29.3% of the infants were stunted (LAZ < 2) and 10.1% were overweight (WLZ > 2).

Binary logistic regression analysis showed that male infants had a greater chance of being anaemic (OR= 1.388, p= 0.037). Infants who ceased being exclusively breastfed at the age of 0–2 months (OR=0.620, p=0.039) and infants who frequently consumed formula milk (OR=0.523, p=0.001) had a lower chance of being anaemic. Regarding iron deficiency (ID) status, male infants (OR=2.432, p=0.002) were found to have a greater chance to be ID; while infants with a higher birth weight (OR=0.417, p=0.005), infants who ceased being exclusively breastfed at the age of 0–2 months (OR=0.362, p=0.022), and infants who frequently consumed formula milk (OR=0.219, p=0.001), had a lower chance to be ID.

Multivariate linear regression analysis showed that haemoglobin concentration was positively related to eye-hand, locomotor and therefore also combined psychomotor activities [β = 0.677 (0.343, 1.011), p= < 0.001]; [β = 0.439 (0.164, 0.714), p= 0.002]; [β = 1.116 (0.586, 1.645), p= < 0.001], respectively. Frequent consumption of infant cereal (≥ 4 days a week) was positively related to locomotor development [β = 0.709 (0.043, 1.376), p= 0.037] and parent rating scores [β = 1.506 (0.912, 2.100); p= < 0.001]. Exclusive breastfeeding up to age 0–2 months was related to higher parent rating scores [β = 1.544 (0.673, 2.414), p = 0.001] compared to exclusive breastfeeding up to the age of 5–6 months. Male gender was associated with lower parent rating scores [β = -0.673 (-1.256, -0.089), p = 0.024], compared to female infants.

Findings from the acceptability trial showed that more than 70% of mothers reported a score of ≥ 4 on sensory attributes for both of the small-quantity lipid-based nutritional supplements, indicating that both supplements were well received. The mean reported consumption over the two week period was $65.3 \pm 34.2\%$ and $62.0 \pm 31.3\%$ of the 20 g daily portion for supplement A and B, respectively. Focus group discussions confirmed a positive attitude towards the supplements in the study population.

Conclusion

The study provides evidence that feeding practices at a very young age, including breast feeding practices, can have implications on nutritional status and/or iron status with consequences on infant development. Associations found between feeding practices and psychomotor development may be explained by the iron status of the infants as a consequence of feeding practices. It therefore emphasises the importance of adequate iron nutrition and iron status during early infancy.

Key terms: Infant, feeding practices, nutritional status, psychomotor development, acceptability, small-quantity lipid-based nutritional supplements

OPSOMMING

Agtergrond

Onvoldoende voeding en -ontwikkeling in die eerste duisend dae van lewe het beduidende implikasies vir die veld van openbare gesondheid en sluit 'n langtermyn-uitwerking op kognitiewe ontwikkeling en skoolprestasie in. Yster is een van die belangrikste voedingstowwe wat vir langtermyn-groei en kognitiewe ontwikkeling benodig word. Verskillende produkte word tans vir die verbetering van nutriëntinname in babas, vir gebruik voorgestel en 'n voorbeeld hiervan is lipied-gebaseerde voedingsaanvullings

Doel

Die studie het gepoog om die voedingstatus, vroeë voedingspraktyke en motoriese ontwikkeling van 6-maande-oue babas van 'n buitestedelike Suid-Afrikaanse gemeenskap in Jouberton (Noordwes provinsie) asook die aanvaarding van twee innoverende lipied-gebaseerde voedingsaanvullings vir aanvullende voeding deur die babas en hul primêre versorgers in die studie gemeenskap, bepaal.

Studie ontwerp

Hierdie studie het 750 ses-maande-oue babas van 'n buitestedelike gemeenskap in die Noordwes-provinsie in Suid-Afrika ingesluit. Voedingspraktyke, dieëtinname en nutriëntdigtheid is in verhouding tot die voedingstatus van die babas bepaal. Verder is die assosiasie van psigomotoriese ontwikkeling met voedingspraktyke en voedingstatus vasgestel.

Vir die aanvaarbaarheidsproef, is moeder-en-baba pare vir 'n tweeledige proef ingeskryf. Deel 1 (n= 16) was 'n toets-voedingsproef met 'n oorkruis, ewekansige ontwerp waarvoor 'n vyf-punt hedoniese skaal wat vir sensoriese evaluering (verskil= 1, saamstem= 5) gebruik is. Deel 2 (n= 38) was 'n twee weeklange proef vir tuisgebruik, gevolg deur fokusgroepbesprekings.

Resultate

Vir 48,9% van die babas is uitsluitlike borsvoeding op die ouderdom van 0–2 maande gestaak. Semi-soliede en/of vaste kos is meestal op die ouderdom van 3–4 maande bygevoeg. Op die ouderdom van ses maande het 70,1% van die babas steeds borsmelk ontvang. Aanvullende voedsel-items wat gereeld ingeneem is, het hoofsaaklik babagraankos (68,1%), formule melk (42,0%) en gebottelde babavoedsel (22,7%) ingesluit. By meer as 80% van die babas was die nutriëntdigtheid (hoeveelheid van nutriënt per 100 kcal) vir yster, sink en kalsium in hul aanvullende dieet laer as die gewenste digtheid.

Nutriënt status het gewys dat 36,4% van die babas anemies was, 16,1% van die babas het 'n ystertekort getoon (plasma ferritien konsentrasie < 12 µg/l) en 9,3% van die babas het aan ystertekort-anemie gely. Die antropometriese data het getoon dat 29,3% van die babas vertraagde groei gehad het (LAZ < 2) en 10,1% oorgewig was (WLZ > 2).

Ten opsigte van anemie-status, het manlike babas (OR= 1,388, p= 0,037) 'n groter kans getoon om anemies te wees. Babas vir wie uitsluitlike borsvoeding op die ouderdom van 0–2 maande gestaak is (OR= 0,620, p= 0,039;) en babas wat gereeld formule-melk ingeneem het (OR= 0,523, p= 0,001), het 'n laer risiko vir anemie getoon. Ten opsigte van ystertekort-status, het manlike babas (OR= 2,432, p= 0,002) 'n groter kans getoon om 'n ystertekort te hê; terwyl babas met 'n hoër geboortegewig (OR= 0,417, p=0,005), babas vir wie uitsluitlike borsvoeding op die ouderdom van 0–2 maande gestaak is (OR= 0,362, p= 0,022) en babas wat gereeld formule-melk ingeneem het (OR= 0,219, p= 0,001), 'n laer kans getoon het om 'n ystertekort te hê.

Hemoglobienstatus het positief bygedra tot oog-hand-koördinasie, lokomotoriese ontwikkeling en gekombineerde psigomotoriese ontwikkeling [β = 0.677 (0.343, 1.011), p= < 0.001]; [β = 0.439 (0.164, 0.714), p= 0.002]; [β = 1.116 (0.586, 1.645), p= < 0.001], onderskeidelik. Gereelde verbruik van kommersiële babagraankos (≥ 4 dae per week) het 'n positiewe bydra gemaak tot die lokomotoriese ontwikkeling [β = 0.709 (0.043, 1.376); p= 0.037]] en ouer-graderingstellingstellers [β = 1.506 (0.912, 2.100); p= < 0.001]. Ekslusiewe borsvoeding vir die ouderdom van 0–2 maande teenoor die ouderdom van van 5–6 maande, het positief bygedra tot ouer-graderingstellingstellers [β = 1.544 (0.673, 2.414), p = 0.001]]. Manlike geslag was geassosieer met laer ouer-graderingstellingstellers [β = -0.673 (-1.256, -0.089), p = 0.024] teenoor vroulike geslag.

Bevindinge vir die aanvaarbaarheidsproef het getoon dat, tydens Deel 1, meer as 70% van die moeders 'n telling van ≥ 4 vir sensoriese eienskappe by beide die klein-hoeveelheid lipied-gebaseerde voedingsaanvullings toegeken het. Hierdie resultaat dui aan dat beide aanvullings goed ontvang is. Vir Deel 2, was die gemiddelde verbruik vir aanvulling A en B wat vir die tydperk van twee weke aangemeld is, onderskeidelik $65,3 \pm 34,2\%$ en $62,0 \pm 31,3\%$ van die 20 g daaglikse porsie. Daar is tydens fokusgroepbesprekings 'n positiewe houding teenoor die aanvullings in die studiebevolking bevestig.

Gevolgtrekking

Hierdie studie het getoon dat voedingspraktyke tydens 'n vroeë ouderdom, (borsvoedingspraktyke ingesluit) implikasies het op nutriënt status en/of ysterstatus wat psigomotoriese ontwikkeling beïnvloed. Assosiasies wat gevind is tussen voedingspraktyke en psigomotoriese ontwikkeling kan deur die ysterstatus, wat deur voedingspraktyke beïnvloed word, verduidelik word. Hierdie studie beklemtoon die belangrikheid van voldoende voeding en ysterstatus tydens n vroeë ouderdom van ontwikkeling.

Sleuteltermes: Baba, voedingspraktyke, voedingstatus, psigomotoriese ontwikkeling, aanvaarbaarheid, klein-hoeveelheid lipied-gebaseerde voedingsaanvullings

TABLE OF CONTENTS

PREFACE	I
ABSTRACT	III
OPSOMMING	V
CHAPTER 1	1
1.1 Rationale of the study	1
1.2 Aim and objectives	3
1.2.1 Aim.....	3
1.2.2 Objectives.....	3
1.3 Study site and design	4
1.4 Structure	4
1.5 Research team	5
1.6 References	7
CHAPTER 2	10
LITERATURE REVIEW ON FEEDING PRACTICES, KEY SELECTED MICRONUTRIENT DEFICIENCIES, GROWTH AND DEVELOPMENT OF INFANTS AND YOUNG CHILDREN	10
2.1 Importance of breastfeeding	10
2.1.1 Nutrient content of breast milk	12
2.1.2 Other components and factors.....	14
2.2 Optimal complementary feeding	14
2.2.1 Selected key nutrients and their functions.....	14
2.2.2 Principles for complementary feeding	17

2.3	Breastfeeding and complementary feeding practices	19
2.3.1	Infant and young child feeding indicators	20
2.3.1.1	Global feeding practices	22
2.3.2	Feeding practices in South Africa	23
2.4	Prevalence of micronutrient deficiencies	26
2.4.1	Indicators for micronutrient deficiencies	27
2.4.1.1	Iron	27
2.4.1.2	Vitamin A	28
2.4.1.3	Zinc	29
2.4.1.4	Iodine	29
2.5	Growth faltering and overweight	30
2.5.1	Global prevalence of growth faltering and overweight	32
2.5.2	Growth faltering in South Africa	33
2.5.3	Child growth and development	35
2.6	Infant and child development	35
2.6.1	Child development assessment tools in low-income and middle-income countries.....	36
2.6.2	Micronutrient deficiencies and early childhood development.....	38
2.7	Actions needed to achieve optimum infant and child nutrition and development	40
2.7.1	Supporting environment.....	42
2.7.2	Nutrition-sensitive interventions and programmes	44
2.7.2.1	Food security	44
2.7.2.2	Water and sanitation.....	45

2.7.2.3	Integrated nutrition and early child development interventions.....	45
2.7.2.4	Health and planning services and nutrition education	46
2.7.3	Nutrition-specific interventions	47
2.7.3.1	Disease prevention and management	47
2.7.3.2	Single- and multiple nutrient interventions	48
2.7.3.3	Fortification.....	50
2.7.3.4	Home fortification.....	51
2.7.4	Key nutrition interventions in South Africa.....	53
2.8	Conclusion.....	56
2.9	References	58
CHAPTER 3.....		82
	FEEDING PRACTICES IN RELATION TO NUTRITIONAL STATUS OF 6-MONTH OLD INFANTS FROM A PERI-URBAN SETTING IN SOUTH AFRICA.	
CHAPTER 4.....		117
	ASSOCIATION OF PSYCHOMOTOR DEVELOPMENT WITH FEEDING PRACTICES AND NUTRITIONAL STATUS OF 6-MONTH-OLD INFANTS IN A PERI-URBAN COMMUNITY OF SOUTH AFRICA.	
CHAPTER 5.....		141
	ACCEPTABILITY OF NOVEL SMALL-QUANTITY LIPID-BASED NUTRIENT SUPPLEMENTS FOR COMPLEMENTARY FEEDING IN A PERI-URBAN SOUTH AFRICAN COMMUNITY	
CHAPTER 6.....		169
CONCLUSION		

ANNEXURE 1	178
ADDITIONAL POLICIES, PRINCIPLES AND INDICATORS	
ANNEXURE 2	183
CONTENT AND STYLE GUIDELINES FOR MATERNAL AND CHILD NUTRITION.....	
ANNEXURE 3	194
CONTENT AND STYLE GUIDELINES FOR THE FOOD AND NUTRITION BULLETIN	
ANNEXURE 4	199
PUBLISHED ARTICLE	

ABBREVIATIONS

AA	Arachidonic acid
AGP	Alpha-1 glycoprotein
AI	Adequate intake
AIDS	Acquired immunodeficiency syndrome
ANC	Antenatal care
APP	Acute phase response proteins
BANC	Basic antenatal care
BME	Breast milk energy
BSID	Bayley scales of infant development
CDATs	Child development assessment tools
CF	Correction factors
CHW	Community health workers
CI	Confidence Interval
CTC	Community therapeutic care
CRP	High-sensitivity C-reactive protein
DHA	Docosahexaenoic acid
DNA	Deoxyribonucleic acid
DOH	Department of Health
DPME	Department of Performance, Monitoring and Evaluation
DSD	Department of Social Development
EAR	Estimated average requirement
ECD	Early childhood development

EFA	Essential fatty acids
ELISA	Enzyme-linked immunosorbent assays
EPA	Eicosapentaenoic acid
FAO	Food and Agriculture Organization
FBDG	Food-based dietary guidelines
Fer	Plasma ferritin
GMP	Growth monitoring and promotion
HAZ	Height-for-age z-score
HIV	Human immunodeficiency virus
HSRC	Human Sciences Research Council
ID	Iron deficiency
IDA	Iron deficiency anaemia
IFPRI	International Food Policy Research Institute
IMCI	Integrated management of childhood illness
IQR	Interquartile range
IYCF	Infant and young child feeding
KDI	Kilifi developmental inventory
LC	Long-chain
LCPUFAS	Long-chain polyunsaturated fatty acids
LNS	Lipid-based nutritional supplements
LNSs	Lipid-based nutrient supplements
MAM	Moderate acute malnutrition
MBFI	Mother-baby friendly initiative

MDGs	Millennium development goals
MMN	Multiple micronutrient
MNP	Micronutrient powders
MCTC	Mother-to-child transmission
MUAC	Middle-upper-arm circumference
MUFAs	Mono-unsaturated fatty acids
NFCS-FB-1	National Food Consumption Survey
PHC	Public health care
pRBP	Retinol-binding protein
pROH	Plasma retinol
PUFAs	Polyunsaturated fatty acids
RNA	Ribonucleic acid
RUTF	Ready-to-use therapeutic food
SAM	Severe acute malnutrition
SD	Standard deviation
SDGs	Sustainable development goals
SF	Serum ferritin
SFAs	Saturated fatty acids
SQ-LNS	Small quantity lipid based nutritional supplements
SUN	Scaling up nutrition
TB	Tuberculosis
TfR	Transferrin receptor
UHT	Ultra high temperature

UIC	Urine iodine concentration
UNICEF	United Nations Children's Fund
VAD	Vitamin A deficiency
VAS	Vitamin A supplementation
WHA	World Health Assembly
WHO	World Health Organization
WHZ	Weight-for-height z-score
WLZ	Weight-for-length z-score

LIST OF TABLES

Chapter 1–2

Table 1	Research Team	6
Table 2	Criteria that define selected infant feeding practices (WHO 2008)	21
Table 3	Classification for assessing the severity of malnutrition by prevalence ranges among children under 5 years of age (WHO, 2015b)	32
Table 4	Age categories (months) (95% CI) for achievement for six gross motor milestones	36
Table 5	Global progress of stunting, exclusive breastfeeding and wasting toward the global WHA targets	41
Table 6	Comprehensive summary of key nutritional interventions in South Africa	54

Chapter 3

Table 1	Socio-economic characteristics of the primary caregivers	106
Table 2	Feeding practices for 6 month old infants	107
Table 3	Amount of iron (mg) contribution from main reported food groups, for consumers only	108
Table 4	Nutrient density (per 100 kcal) of the complementary diet (excluding breast milk) and percentage of infants with an intake below desired density	109
Table 5	Anaemia-, iron- , CRP- and AGP and anthropometric status of infants	110
Table 6	Total nutrient intake for anaemic and non-anaemic infants (median, IQR)	111
Table 7	Comparison of anaemic and non-anaemic infants regards birth weight, gender, anthropometry and early feeding practices	112
Table 8	Bivariate relationships between anaemia and various potential associated factors	114
Table 9	Bivariate relationships between ID based on plasma ferritin and various potential associated factors	115
Table 10	Bivariate relationships between ID based on TfR and various potential associated factors	116

Chapter 4:

Table 1	Nutritional status of 6-month-old infants and scores (mean, 95% CI) obtained in psychomotor development- and parent rating activities	136
Table 2	Comparison between development scores and caregiver's level of education, gender, early feeding practices, frequently consumed food items, anaemic- and iron status, growth status and birth weight of 6-month-old infants (mean, 95% CI)	137
Table 3	Multivariate linear regression analysis for psychomotor development	139
Table 4	Multivariate linear regression analysis for parent rating scores	140

Chapter 5:

Table 1	Nutrient content of the two SQ-LNS pastes	160
Table 2	Characteristics of the participants for Part 1 and Part 2 of the study	162
Table 3	Hedonic scores reported by the mother on her practical experience of using the two porridge-SQ-LNS-mixtures in Part 1 of the study	163
Table 4	Frequencies of the mother's own acceptance and perception of infant acceptance of the SQ-LNS-pastes after two weeks of home-use (Part 2 of the study) expressed as percentage	164

LIST OF FIGURES

<i>Chapter 2:</i>		
Figure 1	Global summary of some nutrition indicators for children younger than 2 years	21
Figure 2	Breastfeeding practices in South Africa in relation to Eastern and Southern Africa and the world	24
Figure 3	Prevalence of anaemia and vitamin A deficiencies in children younger than 5 years globally	26
Figure 4	Global prevalence of underweight, stunting, wasting and overweight for children younger than 5 years	33
Figure 5	The prevalence of stunting, wasting and underweight in children younger than 5 years at national and provincial level (below -2 SD)	34
Figure 6	Prevalence of underweight, stunting and wasting in the world, Eastern and Southern Africa and South Africa	34
Figure 7	Actions needed to achieve optimum infant and child nutrition and development	42

<i>Chapter 5</i>		
Figure 1	Representation of the research approach used for Part 1	165
Figure 2	Representation of the research approach used for Part 2.	166
Figure 3	Time used to consume SQ-LNS A and –B and control porridge respectively	167
Figure 4	Percentage of mothers who reported ‘Agree’ on both her own liking and her perception of the infant’s liking of the two porridge-SQ-LNS-mixtures and the control porridge in Part 1 of the study; *The term ‘Agree’ refers to the combination of ‘agree’ and ‘tend to agree	168

CHAPTER 1

1.1 Rationale of the study

Infants that are exclusively breastfed for the first 6 months of life have a fourteen times less chance to die than non-breastfed infants, and breastfed children in general have at least a 6 times greater chance for survival. Breastfeeding reduces the risk of major mortality risk factors such as acute respiratory infection and diarrhoea (Black *et al.*, 2008). Exclusive breastfeeding for the first 6 months can reduce stunting by protecting the infant against gastrointestinal infections (Kramer & Kakuma, 2012). Almost all stunted growth take place in the first thousand days after conception and the prevalence of stunting during this period may indicate poorer cognitive and educational outcomes later in life (Black & Alderman *et al.*, 2013). The first 6 months of life is important as it is part of the 'brain growth spurt' period (starting in the third trimester and lasting for the first few months of life) when nutrition has a particularly significant effect on brain development (Isaacs & Oates, 2008). Nutritional deficits during the first 2 years of life may cause long-term impairment in growth and intellectual performance (Prado & Dewey, 2014).

Globally, breastfeeding is poorly practiced. Only 44% of new-borns receive breast milk within 1 hour after birth and less than 40% of children under 6 months are fed only breast milk with no additional foods or liquids (including water) (IFPRI, 2014). Already in 2003, The Global Strategy for Infant and Young Child Feeding was developed by the World Health Organization and United Nations Children's Fund (UNICEF) to improve feeding practices globally (WHO & UNICEF, 2003). However, still, one of the World Health Assembly (WHA) nutrition targets is to increase exclusive breastfeeding by 50% in 2025 (IFPRI, 2014; UNICEF, 2014). The prevalence of exclusive breastfeeding in South Africa for infants younger than 6 months is very low (7.4%) (HSRC, 2013). Even though early initiation of breastfeeding is practiced by an estimated 60% of mothers and breastfeeding is a common practice in South Africa, mixed feeding rather than exclusive breastfeeding is practiced (HSRC, 2013). The estimated prevalence of breastfeeding up to 2 years is only 31% (UNICEF, 2014).

South Africa is a member of the United Nations and adopted the WHA International code of Marketing of Breast-milk Substitutes, which points to commitment to protect and promote breastfeeding in the country. The Tshwane Declaration for the Support of Breastfeeding (see Annexure 1, Box 1) was published in 2011 (DOH, 2013a) to call on stakeholders to mainstream breastfeeding in all relevant policies, legislation, strategies and protocols and to

provide services to support mothers to exclusively breastfeed their infants for 6 months, to start with appropriate complementary feeding and continue breastfeeding for up to 2 years of age and beyond. Recent studies have indicated that the implementation of this declaration on national level is not yet optimal as the introduction of semi-solid maize-based foods and formula milk before the age of 4 months is a common feeding practice (Du Plessis, 2013; Faber *et al.*, 2014; Goosen *et al.*, 2014). Nutrition-related indicators from South African District Health Information Systems indicate a significant under-reporting of the exclusive breastfeeding rate at 6 months and of growth monitoring, making it difficult to identify priority areas where intervention is needed. In terms of food availability, there is also a lack of data on the dietary diversity of pregnant women and children younger than 5 years (DOH, DSD, DPME, 2014). In most low-income communities, the first semi-solid foods given to infants are cereal-based gruels with low energy and nutrient density and low bioavailable iron due to the presence of phytic acid (Dewey, 2013). Nutrient density impacts micronutrient status (Troesch, *et al.* 2015). Globally, iron deficiency (ID) is the most common nutritional deficiency and more than 30% of the prevalence of anaemia is due to iron deficiency (Benoist *et al.*, 2008; WHO, 2015b). The risk of ID and anaemia is higher during infancy, which is a period of rapid growth and development where iron is an important component of the haemoglobin molecule and a key nutrient needed for neurodevelopmental processes of the brain during infancy (Tolentino & Friedman, 2007; Hermoso *et al.*, 2011; Prado & Dewey, 2014). Another nutrient of public health significance is vitamin A. Vitamin A deficiency weakens the immune system and increases the risk for morbidity and mortality (WHO, 2013a). Research on feeding practices in the age group 0–6 months associated with key nutrients like iron and vitamin A is lacking in a South African context.

Poor nutrition and development in the first few years of life have serious public health implications and contribute to early age morbidity and mortality (Bentley *et al.*, 2014). Currently, there is a strong focus on scaling up interventions to integrate nutrition and early development (Black & Dewey, 2014; Sabanathan *et al.*, 2015). Presently, many organisations and companies have shown interest in the development of small-quantity lipid-based nutrient supplements (SQ-LNS) and in their potential use in a variety of cultural and geographical settings (Arimond *et al.*, 2013). SQ-LNS is a type of home fortification and can contribute to energy, protein and essential fatty acids (EFA) needed for healthy development (Gibson, 2011; Arimond *et al.*, 2013). Two new SQ-LNS were developed for potential use in the South African setting. Both SQ-LNS are made from soy, which is less expensive than peanuts and which contains essential fatty acids and micronutrients needed for infant development

(Georgieff, 2007; Gibson, 2011). The effects of these 2 SQ-LNS on linear growth in infants enrolled at age 6 months and followed until age 12 months are currently being investigated in a randomised controlled trial, which is referred to as the Tswaka study (Tswaka meaning “mixing” in the Setswana language). The acceptance of SQ-LNS for infant feeding at a home use level is important to ensure large scale sustainable use. The acceptance of SQ-LNS has not yet been established in South Africa.

1.2 Aim and objectives

1.2.1 Aim

This study investigated the nutritional status, early feeding practices and psychomotor development of 6-month-old infants residing in the peri-urban Jouberton area in Klerksdorp in the North West province, South African. Since different home fortification products to improve nutrient intake for infants are being investigated, the acceptance of two novel SQ-LNS for complementary feeding by infants and their primary caregiver in the study community was determined.

1.2.2 Objectives

The objectives were the following:

1. Determine retrospectively infant feeding practices during the first 6 months of life by investigating the following,
 - a. Initiation, duration of and type of breastfeeding practice
 - b. Occurrence and age of introduction of milk feeding
 - c. Age of introduction of solid-, semi-solid or soft foods and other liquids.
2. Determine dietary intake of infants at the age of 6 months.
3. Determine the iron status and prevalence of anaemia in infants at 6 months taking into account plasma ferritin, plasma transferrin receptor and haemoglobin.
4. Determine the association of feeding practices, iron status and anaemia of 6-month-old infants.
5. Determine the association of psychomotor development, feeding practices, anaemia, iron and anthropometric status at 6 months.
6. Determine the acceptance of 2 novel SQ-LNS for complementary feeding by infants and their primary caregiver.

1.3 Study site and design

This study is embedded within a randomised controlled efficacy trial, the Tswaka study, which was designed to investigate the effects of two newly developed SQ-LNS on linear growth and motor development. The Tswaka study was done in the peri-urban Jouberton area in the greater Matlosana Municipality in Klerksdorp, South Africa. The study site is 200 km from the nearest metropolitan area (Johannesburg).

This study was done in 2 phases:

Phase 1: An acceptability trial to determine the acceptability of the two novel SQ-LNS preceded the randomised control trial. The acceptability trial consisted of a sensory evaluation part with a cross-over design, followed by a two-week home use trial and focus group discussions to provide information on practicality of use in the home environment.

Phase 2: A cross-sectional study used the baseline data of the Tswaka study involving 750, 6-month-old infants.

1.4 Structure

The structure of this thesis is in article format and it is divided into 6 chapters. The format and referencing style of the 3 articles (Chapters 3 – 5) are according to the respective journals' guidelines and these are indicated at the start of each relevant chapter.

Chapter 1 provides the background information, aim and objectives, structure of the thesis and information about the research team.

Chapter 2 summarises the relevant literature on infant and child feeding practices, key micronutrient deficiencies, motor development and strategies to improve infant and child feeding.

Chapter 3 presents the first article manuscript. The title of Manuscript 1 reads '**Feeding practices in relation to nutritional status of 6-month old infants from a peri-urban setting in South Africa.**' This manuscript documents early infant feeding practices in relation to anaemia, iron status and anthropometric measurements at 6 months in infants in a peri-urban South African community. This manuscript will be submitted to *Maternal and Child Nutrition*. A content and style guideline for Maternal and Child Nutrition is presented in Annexure 2 and is also applicable for Manuscript 2.

Chapter 4 presents the second article manuscript. The title of manuscript 2 is '**The association of psychomotor development with feeding practices and nutritional status**

of 6-month-old infants in a peri-urban community of South Africa'. In this manuscript the association of psychomotor development regards feeding practices and nutritional status of 6-month-old infants are investigated. Manuscript 2 will be submitted to *Maternal and Child Nutrition*.

Chapter 5 presents the third article manuscript. The title of manuscript 3 is '**Acceptability of novel small quantity lipid-based nutrient supplements for complementary feeding in a peri-urban South African community.**' This manuscript documents the research that determined the acceptability of two newly developed SQ-LNS pastes in a peri-urban South African community. This article has been published in the *Food and Nutrition Bulletin* (doi: 10.1177/0379572115616057). A content and style guideline for Food and Nutrition Bulletin is presented in Annexure 3. The published version of this manuscript can be found in Annexure 4.

Chapter 6 comprises a conclusion that summarises the essential findings of the study and provides recommendations.

1.5 Research team

The research team and their responsive roles in the study are listed in Table 1.

Table 1. Research Team

Members of the research team	Role
Prof. Marius Smuts	Co-promoter of PhD thesis. Principal Investigator of Tswaka trial. Overall responsibility for design and execution of Tswaka trial; quality control of laboratory analysis. Guidance regarding writing of dissertation and interpretation of results. Co-author of all manuscripts.
Prof. Mieke Faber	Co-promoter of PhD thesis. Co-Principal Investigator of Tswaka trial. Questionnaire development, fieldworker training, data coding and analysis for dietary data. Guidance regarding writing of dissertation and interpretation of results. Co-author of all manuscripts.
Dr. Namukolo Covic	Promoter of PhD thesis. Guidance regarding protocol development for of acceptability trial. Questionnaire development and fieldworker training for psychomotor assessment. Guidance regarding writing of dissertation and interpretation of results. Co-author of all manuscripts
Marinel Rothman	Ph.D. student. Protocol development, execution and data management of the acceptability trial. One of the study coordinators of the Tswaka randomized controlled trial. Involved in questionnaire development and fieldworker training for the Tswaka trial. Supervision of data collection and quality control of dietary data, feeding practices and psycho-motor development. Data analysis for acceptability study and (in consultation with M Cockeran) for cross sectional data. Interpretation of results. Leading author on all manuscripts.
Prof. Salome Kruger	Provided training on anthropometric measurements and analysis; standardization of anthropometry measurement.
Prof. Jane Kvalsvig	Psychologist who provided guidance on psychomotor testing.
Marike Cockeran	Statistician who provided guidance regarding statistical analysis for manuscripts.
Elleanor Rossouw	Coordination and management of laboratory procedures
Sr. Chrissie Lessing	Registered Nursing sister: Overall responsible for clinical procedures and blood sample collection.
Tonderayi Matsungu	One of the study coordinators of Tswaka trial. Supervision of data collection and quality control of anthropometric data. Co-author on 2 manuscripts.

1.6 References

- Arimond, M., Zeilani, M., Jungjohann, S., Brown, K.H., Ashorn, P., Allen, L.H., Dewey, K.G., 2013. Considerations in developing lipid-based nutrient supplements for prevention of undernutrition: experience from the International Lipid-Based Nutrient Supplements (iLiNS) Project. *Maternal and child nutrition*, May 6. doi: 10.1111/mcn.12049.
- Benoist, B.D., McLean, E., Egll, I. & Cogswell, M. 2008. Worldwide prevalence of anaemia 1993-2005: WHO global database on anaemia. Geneva: World Health Organization.
- Bentley, M.E., Johnson, S.L., Wasser, H., Creed-Kanashiro, H., Shroff, M., Fernandez Rao, S., *et al.* 2014. Formative research methods for designing culturally appropriate, integrated child nutrition and development interventions: an overview. *Annals of the New York academy of sciences*, 1308(1):54-67.
- Black, M.M. & Dewey, K.G. 2014. Promoting equity through integrated early child development and nutrition interventions. *Annals of the New York academy of sciences*, 1308(1):1-10.
- Black, R.E., Allen, L.H., Bhutta, Z.A., Caulfield, L.E., De Onis, M., Ezzati, M., *et al.* 2008. Maternal and child undernutrition: global and regional exposures and health consequences. *The lancet*, 371(9608):243-260.
- Black, R.E., Alderman, H., Bhutta, Z.A., Gillespie, S., Haddad, L., Horton, S., *et al.* 2013. Maternal and child nutrition: building momentum for impact. *The lancet*, 382(9890):372-375.
- Black, R.E., Victora, C.G., Walker, S.P., Bhutta, Z.A., Christian, P., De Onis, M., *et al.* 2013. Maternal and child undernutrition and overweight in low-income and middle-income countries. *The lancet*, 382(9890):427-451.
- Dewey, K.G. 2013. The challenge of meeting nutrient needs of infants and young children during the period of complementary feeding: an evolutionary perspective. *The journal of nutrition*, 143(12):2050-2054.
- DOH (Department of health) see South Africa.
- Du Plessis, L.M. 2013. Commitment and capacity for the support of breastfeeding in South Africa: a paediatric food-based dietary guideline. *South African journal of clinical nutrition*, 26(3):S120-S128.

Faber, M., Laubscher, R. & Berti, C. 2014. Poor dietary diversity and low nutrient density of the complementary diet for 6- to 24-month-old children in urban and rural KwaZulu-Natal, South Africa. *Maternal and child nutrition*, doi: 10.1111/mcn.12146. Date of access: 19 Aug. 2014.

Georgieff, M.K. 2007. Nutrition and the developing brain: nutrient priorities and measurement. *The American journal of clinical nutrition*, 85(2):614S-620S.

Gibson, R. 2011. Strategies for preventing multi-micronutrient deficiencies: a review of experiences with food-based approaches in developing countries. In: Thompson, B., & Amoroso, L., (eds). *Combating micronutrient deficiencies: food-based approach*. CABI and FAO, Oxfordshire: UK:1-92.

Goosen, C., McLachlan, M. & Schübl, C. 2014. Infant feeding practices during the first 6 months of life in a low-income area of the Western Cape Province. *South African journal of child health*, 8(2):50-54.

Hermoso, M., Vollhardt, C., Bergmann, K. & Koletzko, B. 2011. Critical micronutrients in pregnancy, lactation, and infancy: considerations on vitamin D, folic acid, and iron, and priorities for future research. *Annals of nutrition and metabolism*, 59(1):5-9.

HSRC (Human Sciences Research Council). 2013. The South African National Health and Nutrition Examination Survey (SANHANES-1). Data analysis on infant feeding practices, and anthropometry in children under five years of age: South Africa 2012. Report for UNICEF. Cape Town.

IFPRI (International Food Policy Research Institute). 2014. *Global nutrition report 2014: actions and accountability to accelerate the world's progress on nutrition*. Washington, DC.

Isaacs, E. & Oates, J. 2008. Nutrition and cognition: assessing cognitive abilities in children and young people. *European journal of nutrition*, 47(3):4-24.

Kramer, M.S. & Kakuma, R. 2012. Optimal duration of exclusive breastfeeding. *Cochrane database dystematic review*, 8:1-131.

Prado, E.L. & Dewey, K.G. 2014. Nutrition and brain development in early life. *Nutrition reviews*, 72(4):267-284.

Sabanathan, S., Wills, B. & Gladstone, M. 2015. Child development assessment tools in low-income and middle-income countries: How can we use them more appropriately? *Archives of disease in childhood*, 100:482-488.

South Africa. Department of Health. 2013a. South African infant and young child feeding policy. Pretoria.

South Africa. Department of Health. 2013b. Roadmap for nutrition in South Africa 2013-2017. Pretoria.

Tolentino, K. & Friedman, J.F. 2007. An update on anaemia in less developed countries. *The American journal of tropical medicine and hygiene*, 77(1):44-51.

Troesch, B., Biesalski, H.K., Bos, R., Buskens, E., Calder, P., Saris, W.H.M. 2015. Increased of foods with high nutrient density can help break the intergenerational cycle of malnutrition and obesity. *Nutrients*, 7(7):6016-6037.

UNICEF (United Nations Children's Fund - Division of communication). 2014. Statistical tables: economic and social statistics on the countries and areas of the world, with particular reference to children's well-being. New York, USA.

WHO (World Health Organization). 2013. Essential nutrition actions: Improving maternal, newborn, infant and young child health and nutrition. Geneva, Switzerland.

WHO (World Health Organization). 2014. Maternal, infant and young child nutrition in East and Southern African countries: moving to national implementation. Report of a World Health Organization workshop, Entebbe, Uganda, 26–28 November 2013.

WHO (World Health Organization). 2015. Micronutrient deficiencies: iron deficiency anaemia. <http://www.who.int/nutrition/topics/ida/en/> Date of access: 2 July 2015.

WHO & UNICEF (World Health Organization United Nations Children's Fund). 2003. Global strategy for infant and young child feeding. Geneva, Switzerland: World Health Organization.

CHAPTER 2

Literature review on feeding practices, key selected micronutrient deficiencies, growth and development of infants and young children

Malnutrition, including undernutrition and micronutrient deficiencies, causes stunting and wasting, which are major risk factors for neonatal disorders, pneumonia and diarrhoea. These diseases cause the death of nearly eight million children each year (Bentley *et al.*, 2014). Undernutrition can be reduced by improving exclusive breastfeeding practices and establishing optimal complementary feeding practices that include continuing breastfeeding for up to 2 years or beyond. This could save the lives of 1.5 million children younger than 5 years annually (WHO, 2013a).

This literature review firstly focuses on the importance of breastfeeding as a cornerstone for healthy growth and development. Secondly, optimal complementary feeding practices are discussed, followed by current international and national breastfeeding and complementary feeding practices. Against the background of current breastfeeding and complementary feeding practices, undernutrition in terms of micronutrient malnutrition, growth and development is discussed. Finally, some general actions needed to achieve optimum infant and child nutrition and development are reported.

2.1 Importance of breastfeeding

The 2003 Lancet series on child survival highlighted breastfeeding as the most important preventative approach for saving child lives (Jones *et al.*, 2003). Breast milk is the best food source for optimal nutrient intake and it provides immunological protection. Breastfeeding has different benefits for both the mother and infant, including reducing the risk of major mortality risk factors like acute respiratory infection and diarrhoea (Black *et al.*, 2008; Kramer & Kakuma, 2012).

The WHO recommends initiation of breastfeeding within the first hour after birth, exclusive breastfeeding until 6 months of age, and introduction of complementary foods at age 6 months with continued breastfeeding until the age of 2 years or older (WHO & UNICEF, 2014). The South African paediatric Food-based Dietary Guidelines Working Group formulated key messages with regard to infants younger than 6 months to be tested in a field setting. These messages are: 1) “give your baby only breast milk for the first 6 months. No other food or drink is needed at this age. If a baby is given other food and drink, he or she will consume less breast milk, and thereby receive less nutrition. Babies are protected against infection when

they are breastfed; 2) hold your baby against your chest, skin-to-skin, within one hour of birth. Start breastfeeding at this time; 3) feed your baby several times during the day and night. This will help your body to make more milk; 4) breast milk contains substances that help to protect your baby against illness. If your baby does not get breast milk, he or she is at a greater risk of developing serious illnesses; 5) ask for help if your baby is having difficulty breastfeeding” (Du Plessis, 2013). A systematic review of evidence for breastfeeding initiation practices and neonatal outcomes has indicated that early initiation of breastfeeding (within one hour) decreased the risk of mortality by 44% (Debes *et al.*, 2013). Initiation of breastfeeding within one hour after birth stimulates the production of breast milk from the mother and protects the infant from infection. The colostrum is an important source of nutrition and immune protection of the infant. Colostrum is special milk rich in white cells and antibodies, protein, minerals and fat-soluble vitamins A, D, E and K. The yellowish colour of colostrum is due to the presence of vitamin A. It also provides important immune protection for the infant’s first exposure to the micro-organisms in the environment. On the third day after delivery, the intake of breast milk increases to 300–400 ml per 24 hours, and then on the fifth day to 500–800 ml. Transitional milk is known as the kind of milk secreted from day 7–14 and the kind of milk secreted after 2 weeks is called mature milk (WHO, 2009b). Emotional bonding occurs between the infant and mother during breastfeeding. Early skin-to-skin care in neonates showed to increase the breastfeeding rate by 27% of infants aged 1–4 months and also has the potential to increase the duration of breastfeeding (Moore *et al.*, 2012). Results of a systematic review showed that exclusive breastfeeding for the first 6 months, in comparison to exclusive breastfeeding for 3–4 months, reduces gastrointestinal infection, increases maternal weight loss and has no adverse effect on growth. A reduced level of iron was observed, but delayed cord clamping and maternal iron supplementation can treat and rectify the situation (Kramer & Kakuma, 2012).

Breastfed infants are less likely to become overweight. A systematic review done by Horta and Victora (2013) showed that a longer duration of breastfeeding in children may reduce the prevalence of overweight or obesity later in life, and has a protective effect against type-2 diabetes (Horta & Victora, 2013). Breastfeeding was also shown to be associated with an increased intelligence in childhood and adolescence. All of the above findings must take residual confounding factors into consideration as breastfeeding duration was higher in more educated mothers with a higher income (Horta & Victora, 2013). A study conducted in the Honduras in different communities on poor income, less educated mother-infant pairs also

showed that exclusively breastfed infants crawled sooner than those who were mixed-fed (Dewey *et al.*, 2001).

In developing countries, bottle-fed infants are at a greater risk of illness, especially when considering diarrhoeal infections and pneumonia (Bahl *et al.*, 2005) as the utensils used to prepare bottle feeding may be dirty and contaminated (Lanigan *et al.*, 2001). An infant fed with an artificial teat may also have difficulties to learn to attach well at the breast, and feeding with an artificial teat has been associated with early cessation of breastfeeding (WHO, 2009b).

2.1.1 Nutrient content of breast milk

The nutrient content of breast milk is highly variable between individuals (Gidrewics *et al.*, 2014) and derives from 3 sources: 1) the synthesis in the lactocyte; 2) dietary in origin, and 3) the maternal stores, which are especially important for fatty acid composition. The macronutrient composition of breast milk varies within mothers and across populations. The mean macronutrient composition is estimated to be 3.2–3.6 g/dl for fat, 6.7–7.8 g/dl for lactose and 0.9–1.2 g/dl for protein. The energy content is highly correlated with the fat content of breast milk and ranges from 65–70 kcal/dl (Ballard & Morrow, 2013). Unless the mother is deficient, breast milk is a sufficient source of most vitamins and minerals needed by the infant (WHO, 2009b). The composition of breast milk is discussed in short below.

Fat: An estimated 50% of energy in breast milk is provided by fats (German & Dillard, 2006). Brenna *et al.* (2007) reported from a descriptive meta-analysis that the mean concentrations of long-chain polyunsaturated fatty acids (LCPUFAs), namely docosahexaenoic acid (DHA) and arachidonic acid (AA) in breast milk are $0.32 \pm 0.22\%$ and $0.47 \pm 0.13\%$ respectively. These fats are both important for neural development (Brenna *et al.*, 2007). The average breast milk intake of 750 ml for the first 6 months after birth seems to correspond with these concentrations (Cunnane *et al.*, 2000). There is a higher concentration of DHA found in breast milk when compared to formula milk (Heaton *et al.*, 2013). However, the availability of these LCPUFAs depend on the LCPUFAs stores at birth, available LCPUFAs from the diet and the ability of the infant to synthesize LCPUFAs from their shorter-chain precursors (Innis, 1991). Low-birth-weight preterm infants have limited stores of the LCPUFAs needed for rapid development at birth, and they need preformed dietary LCPUFAs (Innis, 2003; Larque *et al.*, 2002). Maternal LCPUFA body stores are closely related to the LCPUFA content in breast milk (Del Prado, 2001). Mothers of preterm infants transfer less LCPUFAs to the foetus during the shorter pregnancy and provide a smaller volume of human milk for preterm infants and therefore might have higher LCPUFA stores than mothers of full-term infants (Kovács *et al.*, 2005). Therefore,

preterm infants need their own mother's milk, which might have a higher LPUFA content (Bokor *et al.*, 2007).

Lactose: Milk sugar lactose is the main carbohydrate in breast milk and breast milk contains about 7 g lactose per 100 ml. Oligosaccharides present in breast milk provide important protection against infection (WHO, 2009b).

Protein: Many proteins in breast milk are digested and a good source of well-balanced amino acids. Breast milk contains adequate amounts of essential amino acids for the developing infant. Lipase, amylase, β -casein, lactoferrin, haptocorrin, helps in the digestion and utilisation of macro- and micronutrients from breast milk (Lönnerdal, 2003). The proteins in breast milk have many protective factors against infection, including immunoglobulin, which covers the intestinal mucosa and prevents the entry of bacteria into the cells; white blood cells, which protect against micro-organisms; and whey proteins, which protect against bacteria, viruses and fungi (WHO, 2009b).

Calcium: About half of the calcium found in breast milk is bound to proteins. Infants need a small amount of calcium for bone mineralisation because of the slow growth rate over a 3-month-period (Kent *et al.*, 2009).

Choline: Breast milk is rich in highly bioavailable choline needed for cognitive development (Ilcol *et al.*, 2005).

Vitamin A: Breast milk is an important source of vitamin A, which is needed by new-born infants with low liver vitamin A stores (Engle-Stone *et al.*, 2014).

Iron and zinc: Iron and zinc are present in breast milk in low concentrations, but both are highly bioavailable (WHO, 2009b). During the third trimester of pregnancy, the term infant accretes 80% of iron. Iron requirements during the first 6 months of infancy are dependent on the iron stores at birth and infants born prematurely are deficient in total body iron (Baker & Greer, 2010). Maternal iron status (before and during pregnancy), gestational age, birth weight and the timing of the umbilical cord clamping all influence the iron status of the new-born infant at birth. A breastfed infant with a normal birth weight born from a mother with an adequate iron status during pregnancy and who experienced delayed umbilical cord clamping, will have sufficient iron for the first 6 months (Chaparro *et al.*, 2006; Dewey & Chaparro, 2007; Hutton & Hassan, 2007).

2.1.2 Other components and factors

Breast milk also contains a variety of bioactive components with medicinal qualities. Factors like growth factors (epidermal-, neuronal-, insulin-like-, vascular endothelial- and erythropoietin growth factors), growth-regulating hormones and immunological factors for the protection against infection and inflammation can also be found in breast milk (Ballard & Morrow, 2013).

2.2 Optimal complementary feeding

Complementary feeding is defined by WHO (2003) as the process starting when breast milk alone is no longer sufficient to meet the nutritional requirements of infants and therefore other foods and liquids are needed along with breast milk (WHO, 2003). During the introduction of complementary feeding, infants are particularly vulnerable and the nutrient adequacy of the diet is essential. The most challenging age group for meeting micronutrient needs falls within the second 6 months of life (Dewey, 2013). An adequately diverse diet that includes breast milk and a variety of plant and animal source foods is needed to provide approximately 40 different known nutrients in different amounts for optimal growth and development of the infant (Bloem, 2013). In developing countries, it is a challenge to feed nutrient dense complementary food to infants and young children in order to meet the nutritional requirements of especially iron, vitamin A, zinc, iodine and DHA (Black & Alderman *et al.*, 2013; Black *et al.*, 2008). These nutrients are discussed in more detail along with some key nutrients like choline, folate and vitamin B12 needed for optimal brain development (Prado & Dewey, 2014).

2.2.1 Selected key nutrients and their functions

Iron: Iron is essential for the functioning of many biochemical processes, which include electron transfer reactions, gene regulation, binding and transport of oxygen, as well as the regulation of cell growth and differentiation (Beard, 2001). Rapid growth along with increased haemoglobin needs occurs during infancy. Iron is an important component of the haemoglobin molecule and a key nutrient needed to transport oxygen to all organs in the body (Hermoso *et al.*, 2011; Prado & Dewey, 2014). The extent to which iron is absorbed from the diet and used for normal body functions is known as the bioavailability of iron. There are 2 types of dietary iron, namely nonheme iron, which is present in both plant foods and animal tissues, and heme iron, which comes from haemoglobin and myoglobin in animal source foods. Heme iron is more bioavailable and may contribute to $\geq 40\%$ of total absorbed iron, while nonheme iron is much less well absorbed than heme iron. Inhibitors of iron absorption include phytate in plant-

based foods (main inhibitor of iron absorption); polyphenols found in plant foods and beverages such as tea (restricts nonheme and heme iron absorption); and proteins such as milk proteins, eggs and albumin. Ascorbic acid is an enhancer of iron absorption (Hurrell & Egli, 2010). Good sources of heme iron are meat, poultry, fish and eggs (WHO, 2005).

Vitamin A: Vitamin A is required in small amounts for the normal functioning of the visual systems, maintenance of cell function for growth, epithelial cellular integrity, immune function and reproduction (WHO, 2009a). Vitamin A is involved in the regulation of neurodevelopment by promoting cell differentiation and regulating the expression of genes and is also involved in dopamine-regulated cognitive and motor activity (Anjos *et al.*, 2013). Dietary requirements for vitamin A are provided by a mixture of preformed vitamin A (retinol) present in animal source foods and provitamin A carotenoids, which are found in plant source foods and that need to be converted into retinol in the intestinal mucosa and the liver to be taken up by cells. Good sources of retinol include organ meat, dairy products, egg yolk, whereas β -carotene (pro-vitamin A) can be found in yellowish fruits and vegetables like papaya and pumpkin (WHO, 2009a).

Zinc: As the fourth most abundant ion in the brain; zinc contributes to brain structure and function through: 1) DNA and RNA synthesis; 2) the metabolism of macronutrients and 3) the synthesis of co-enzymes for biogenic-amine synthesis and metabolism (Sandstead *et al.*, 2000). Zinc plays a central role in cellular growth, differentiation and metabolism (Brown, Wuehler *et al.*, 2001) and is vital for nucleic acid and protein synthesis, gastrointestinal and immune function for healthy growth (Moynahan, 1974; MacDonald, 2000). Sources of zinc include red meat, poultry and fortified cereals (WHO & FAO, 2006).

Iodine: Iodine is seen as the primary potentially preventative micronutrient in cognitive disabilities during childhood (Aburto *et al.*, 2014). Iodine is used for the synthesis of thyroid hormones needed for the development of the central nervous system through neurogenesis, neuronal migration, axon and dendrite growth, synaptogenesis and myelination (Prado & Dewey, 2014). Iodine deficiency causes poor growth, poor cognitive development and low urinary iodine excretion and in severe cases, iodine deficiency may cause death. Iodine deficiency disorders include a spectrum of growth, developmental and functional abnormalities and the swelling of the thyroid gland is the most common manifestation of iodine deficiency. Severe iodine deficiency during pregnancy results in foetal death or cretinism, whereas mild and moderate deficiency during pregnancy causes poor foetal development and a high risk of

speech and hearing defects, as well as impaired motor development and growth (Aburto *et al.*, 2014). Iodine can be found in iodised salt and fortified cereals (WHO & FAO, 2006).

Choline, folate and vitamin B12: Choline is critical during the period of growth and development and is interrelated with folate (Zeisel, 2006). During periods of rapid growth and cell division, especially during pregnancy, folate's role for normal neural tube closure is well known and it is vital for brain development through nucleotide synthesis, methylation processes, DNA integrity and transcription (Hermoso *et al.*, 2011; Anjos *et al.*, 2013). Folate (vitamin B9) plays a central role in the synthesis and methylation of nucleotides needed for cell multiplication and tissue growth. Folate is closely interrelated to vitamin B12 and the combination of severe folate deficiency and vitamin B12 deficiency may cause megaloblastic anaemia (Aslinia *et al.*, 2006). After 6 months of age, infants may be vulnerable to vitamin B12 deficiency if the breastfeeding mother is vitamin B12 deficient and/or the infant consumes a low amount of animal source food (Black, 2008). Choline, folate and vitamin B12 are found in animal source foods, legumes and dairy products (WHO & FAO, 2006).

Fats: Fats are the main source of energy for healthy growth and development during infancy. Key functions of fat with regard to the small stomach size of infants are that fat exhibit slow gastric emptying, prolong satiety, facilitate the absorption of lipid-soluble vitamins and provide EFAs (Uauy *et al.*, 2000). Animal-derived foods, full-cream milk, avocados and spreads made from groundnuts, other nuts or seeds are good sources of fat (WHO, 2005)

Fatty acids: Dietary fatty acids are classified according to the degree of unsaturation namely: 1) saturated fatty acids (SFAs) with no double bonds; 2) mono-unsaturated fatty acids (MUFAs) with one double bond, and 3) long chain polyunsaturated fatty acids (LCPUFAs) with two or more double bonds in the carbon chain. Humans are unable to synthesise the essential polyunsaturated fatty acids (PUFAs) and must acquire it from their diet. LCPUFAs are subdivided into 12 families according to the location of double bonds, ranging from the n-1 to n-12 position countered from the terminal carbon of the fatty acid chain. The n-6 and n-3 family are the most important families in terms of human health and nutrition (FAO, 2010). The two most abundant LCPUFAs found in the brain are DHA and AA (Agostoni, 2008). Arachidonic acid (AA) is part of the n-6 PUFAs. Meat, eggs and fish are good sources of AA (FAO, 2010). Eicosapentaenoic acid (EPA) and DHA are part of the n-3 PUFAs and good sources of n-3 fatty acids are marine fish like mackerel, salmon, and sardine, as well as fish oils (FAO, 2010). LC PUFAs play an essential role in the developing brain where most of the PUFAs are concentrated. The development of the infant's brain lays the foundation for the structure of the

adult's brain and therefore, the optimal intake of LPUFAs is very important. Studies indicated that there is strong evidence that n-3 fatty acids are essential for healthy development from the time in utero during pregnancy throughout life (Connor, 2000). Among LPUFAs, DHA plays the primary role in brain development from birth throughout infancy and is also concentrated in some retinal cells to support visual activity. DHA deficiencies may cause neurodevelopment deficits (McNamara & Carlson, 2006). It was found that DHA deficiency during brain growth may be related to neurological or neurodegenerative diseases later in life (Farquharson *et al.*, 1995). Therefore, the intake of oily fish during pregnancy and breastfeeding is important for the developing foetus and infant (Heaton *et al.*, 2013). AA and DHA are vital for the cerebral cortex during the last trimester of gestation and for the first 18 months of life, which is also known as the "growth spurt" of brain development when the developing brain is vulnerable to micronutrient deficiencies too (Nyaradi *et al.*, 2013).

When comparing cognitive development in breastfed versus formula-fed infants, results of a meta-analysis showed that, after adjustment for appropriate key cofactors, breastfeeding was associated with significantly higher scores for cognitive development. The presence of long-chain polyunsaturated fatty acids (LPUFAs) in breast milk was given as possible reason for the higher cognitive scores in breastfed infants (Anderson *et al.*, 1999).

2.2.2 Principles for complementary feeding

Children younger than 2 years have high nutrient needs to support growth and development. Therefore, complementary foods should be high in nutrient density as breast-fed infants consume relatively small amounts of foods in addition to breast milk (Dewey, 2013). The WHO published guiding principles for complementary feeding of both the breastfed and non-breastfed child (WHO, 2003; WHO, 2005). The principles are summarised in Table 1 in Annexure 1. These guidelines can be adapted to local feeding practices and conditions. Some of the most important terms with regard to optimal complementary feeding include energy and nutrient density and dietary diversity. Responsive feeding, good hygiene and proper food handling is also important in terms of how, when, where and by whom the child is fed (WHO, 2003).

Energy and nutrient density: For breastfed infants aged 6–8 months, the daily total energy required from the complementary diet depends on the breast milk energy (BME) intake, which varies from 552 kcal for low- to 160 kcal for high BME intake. From the age of 9 to 23 months, energy requirements from the complementary diet increase from 701 kcal to 1028 kcal for low BME intake, and from 257 kcal to 516 kcal for high BME intake (Dewey & Brown, 2003). During

the first 6 months of life, dietary total fat should contribute 40–60% of energy (%E) for healthy growth and, depending on the physical activity of the child, and estimated 35% for age 6–24 months (FAO, 2010). The energy density of the available local foods guides the number of meals required for the non-breastfed child and is calculated from the energy requirements (by assuming a gastric capacity of 30 g/kg body weight/day). Five to six meals/day are needed to meet the energy requirements when energy density of foods is 0.6 kcal/g, 4 meals a day are needed when energy density is 0.8 kcal/g and 3 meals are needed when the energy density is 1.0 kcal/g (WHO, 2005).

Breastfed infants consume a small amount of complementary food in addition to breast milk. Complementary food should be high in nutrient density (amount of nutrient per 100 kcal of food). Iron and zinc are generally problematic nutrients (Dewey, 2013), because of the low concentrations in human milk. For infants in developing countries, the minimum target nutrient density for iron and zinc is 4.5 mg/100 kcal and 1.14 mg/100 kcal respectively at the age of 6–8 months (when assuming that the average energy need from complementary food is 200 kcal/d with an average intake of breast milk). The minimum iron and zinc densities of complementary foods are significantly lower in the second year of life (1.0 and 0.46 mg/100 kcal, respectively) (Dewey, 2013).

Dietary diversity: Daily dietary intake of at least four of seven food groups like 1) grains, roots and tuber; 2) legumes and nuts; 3) dairy products (milk yogurt, cheese), 4) flesh foods (meat, fish, poultry and liver/organ meats, 5) eggs, 6) vitamin A rich fruit and vegetables and 7) other fruits and vegetables is a good indication that the infant most probably consumed at least one animal source food and at least one fruit or vegetable with a staple food. Non-breastfed infants need a daily intake of approximately 200–400 ml of milk if adequate animal source foods are consumed, or 300-500 ml of milk if animal source foods are not consumed regularly. Milk sources include full-cream animal milk, Ultra High Temperature (UHT) milk, reconstituted evaporated (not condensed) milk, fermented milk or yoghurt and expressed milk (heat treated if mother is HIV-positive) (WHO, 2005).

Responsive feeding: Mothers or the primary caregiver are the main responsible persons to ensure the adequate nutrient intake for their infant or young child (IFPRI, 2014). Social interactions during feeding between caregivers and infants, like smiling and verbalisations, also promote the intake of food. Caregivers must be encouraged to participate in the development of the infant with simple activities such as play and communication, also during

feeding times (Grantham-McGregor & Baker-Henningham, 2005; Galler, Bryce & Waber *et al.*, 2012; Galler, Bryce & Zichlin *et al.*, 2012).

Developmental milestones are closely related to eating behaviour of infants and children. For example, during the first 6 months of life, the infant must be able to suck/swallow normally and also regulate feeding periods by demand by turning away from the breast/bottle. By the age of 6 months the infant must be able to touch food with fingers and be able to lean toward food, open the mouth when hungry and turn the head when full. Children in the age group 6–12 months must be able to explore with a spoon to self-feed and must also be able to eat finger foods. Children aged 12–24 months must be able to self-feed and imitate and explore during family meals. This is also the stage where children are "picky eaters" (Carruth *et al.*, 2004). Infant and children's food preferences are influenced by regulatory processes such as hunger, satiety, imitation, like looking at what the rest of the family members are eating, exploration, like the desire for autonomy, and the environment, like mealtime settings and distractions. Mealtimes must be a pleasant time. The caregiver must pay attention to responsive feeding by recognising and guiding the infant/child's cues of hunger and satiety. Non-responsive feeding is driven by a lack of reciprocity between the parent and infant/child. Examples of non-responsive feeding are force feeding, indulgent feeding by the infant/child, involvement of the parent during meals or the restriction of food intake by the parent (Black & Aboud, 2011).

Good hygiene and proper food handling: Infants in families of poor socio-economic status are exposed to restricted food intake, fewer household resources, and poor psychosocial stimulation in crowded households (Hackman & Farah, 2009). The intake of poor nutrient-dense foods served under poor hygienic conditions is reflected in the high morbidity and mortality rates in developing countries (Black *et al.*, 2008). During diarrhoea and fever, non-urinary water losses can be 2–3 times greater than normal and the intake of extra fluid is important with a nutrient dense diet (WHO, 2005).

2.3 Breastfeeding and complementary feeding practices

The largest part of the "first thousand days", which is the period from conception to the child's second birthday, falls within the time frame from the introduction of complementary foods to 24 months of age. This time frame is important for the prevention of undernutrition in disadvantaged populations where growth faltering and high incidence of infection often occurs (Dewey, 2013). Therefore, optimal feeding practices especially for children under 2 years of age are important for healthy growth and development. Infant and young child feeding

practices can be assessed by using indicators that focus on selected food-related aspects of child feeding and are amendable to population-level measurements (WHO, 2008).

2.3.1 Infant and young child feeding indicators

Certain criteria used to define infant and young child feeding practices were set by the WHO (2008) so that a child can be classified according to his/her feeding practice. The criteria used to define exclusive breastfeeding, predominant breastfeeding and complementary feeding are shown in Table 2.

Table 2. Criteria that define selected infant feeding practices (WHO 2008)

	Breast milk ¹	ORS, drops, syrups ²	Water, water-based drinks, fruit juice	Non-human milk	Food-based fluids	Semi-solid / solid food
Exclusive breastfeeding	Yes	Yes	No	No	No	No
Predominant breastfeeding	Yes	Yes	Yes	No	No	No
Complementary feeding	Yes	Yes	Yes	Yes	Yes	Yes

¹includes milk expressed or from a wet nurse

ORS: Oral Rehydration Solutions made at home (six level teaspoons of sugar and half teaspoon of salt dissolved in one litre of clean water) (WHO & UNICEF, 2006).

²includes vitamins, minerals and medicines

Infant and young child feeding practices in developing countries can be measured by using simple, valid and reliable population-level indicators (See Annexure 1, Box 2). The primary functions of these indicators are 1) assessment of feeding practices; 2) targeting populations at risk, and 3) monitoring and evaluation of interventions. The minimum meal frequency indicator is a proxy for energy intake from foods other than breast milk (includes only non-liquid feeds for breastfed children and for non-breastfed children includes both milk feeds and solid/semi-solid feeds). The minimum acceptable diet includes both minimum dietary diversity and minimum meal frequency. For non-breastfed children, milk feeds are excluded from the dietary diversity score for the calculation of the minimum acceptable diet, as milk feeds are considered a separate and required element for non-breastfed infants (WHO, 2008). Studies conducted in low income countries on infants 6–24 months old indicated that increased dietary diversity is positively associated with height-for-age, and the infant and young child feeding indicators indicated that the consumption of a minimum acceptable diet has the potential to reduce the risk of stunting and underweight (Arimond & Ruel, 2004; Marriott *et al.*, 2012). A recent study published on feeding practices of 0 to 24-month-old children in Nepal used the infant and young child feeding indicators and showed that 83% of children younger than 2 years received age-appropriate breastfeeding, but only 48% were breastfed within one hour after birth. Exclusive breastfeeding for the first 6 months followed by feeding a minimum meal frequency to children older than 6 months were associated with a higher weight-for-age Z-score, and higher dietary diversity was associated with higher height-for-age Z-scores (Lamichhane *et al.*, 2015).

2.3.1.1 Global feeding practices

Suboptimal feeding practices are found in West and Central Africa, which is the region with the highest neonatal mortality rate and the highest mortality rate of infants younger than 12 months (35 and 72 per 1000 live births, respectively). Suboptimum breastfeeding together with vitamin A and zinc deficiency cause 3.1 million child deaths annually (Black & Victora *et al.*, 2013). In this region, the early initiation of breastfeeding is 39%; 25% of mothers practice exclusive breastfeeding until 6 months and 44% of mothers still breastfeed their children at the age of 2 years. The minimum acceptable diet is reported to be only 10% for children 6–24 months old (UNICEF, 2014). Figure 1 shows a summary of some feeding practice indicators for children younger than 2 years globally.

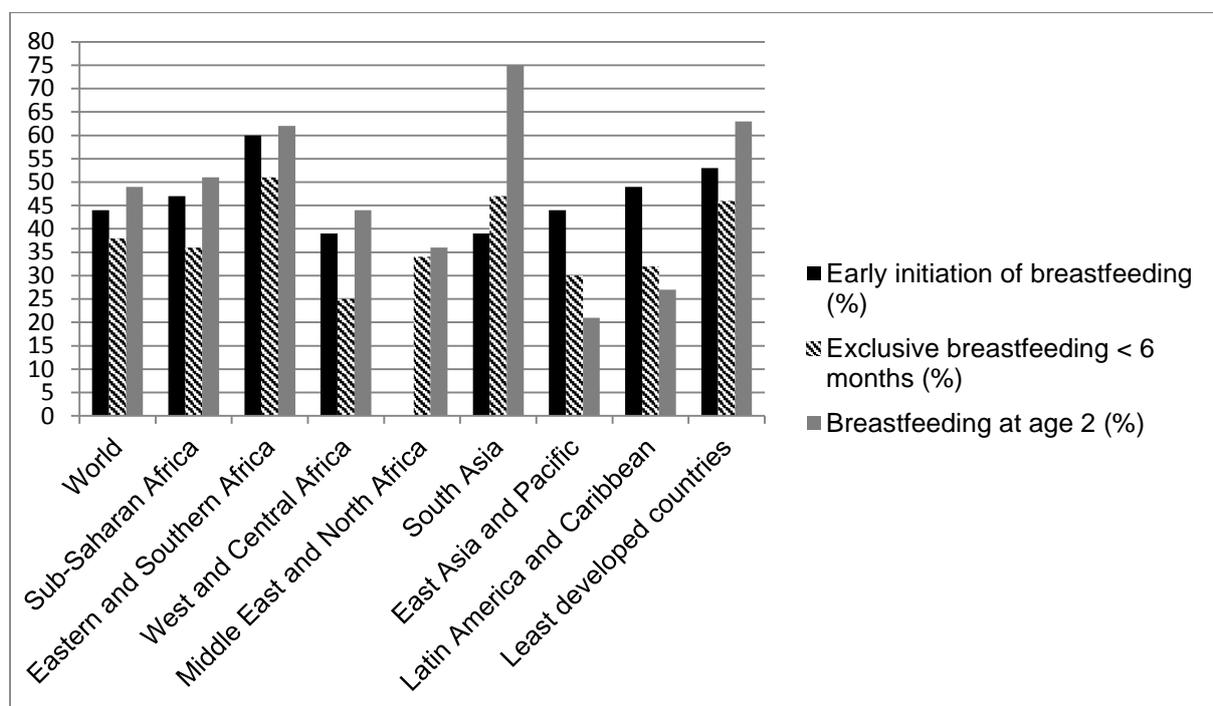


Figure 1. Global summary of some nutrition indicators for children younger than 2 years (UNICEF 2014)

East Asia and the Pacific reported to have the lowest percentage of mothers who still practiced breastfeeding until their children are 2 years old (21%) (UNICEF, 2014). China, with the largest population in the world and more than 70% of inhabitants living in rural areas, reported in 2012 that only 24% of mothers practiced exclusive breastfeeding for the first 6 months and 32% continued breastfeeding until one year post-partum (Zhou *et al.*, 2012).

Early initiation of breastfeeding is less than 50% in most regions, except for Eastern and Southern Africa (60%) (UNICEF, 2014). However, only 51% of mothers in Eastern and

Southern Africa practice exclusive breastfeeding for the first 6 months (UNICEF, 2014). In countries like Uganda and Mozambique, mothers already start in the first month with mixed feeding by introducing various liquids like water and food items like commercial cereal, vegetables and fruit (Engebretsen *et al.*, 2007; Arts *et al.*, 2010). Grain-based gruels are often given to African infants before 6 months (Cherop *et al.*, 2010; Engebretsen *et al.*, 2007; Kalanda *et al.*, 2006).

South Asia has a high percentage of mothers who still breastfeed their children at the age of 2 years (75%) (UNICEF, 2014). However, in spite of the high breastfeeding rates, an analysis of recent national survey data by South Asia Infant Feeding Research Network showed that, the percentage of breastfed children aged 6 to 23 months who achieved the WHO recommendations for minimum dietary diversity ranged from 15% (India) to 71% (Sri Lanka), and for minimum acceptable diet from 9% (India) to 68% (Sri Lanka) (Senarath *et al.*, 2012).

It is reported that only 34% of infants in the Middle East and North African region are exclusively breastfed for 6 months and only 36% of children receive breast milk at the age of 2 years (UNICEF, 2014). Infants in this region are often introduced to herbs and herbal tea before the age of 6 months, and coffee being given to infants in Indonesia (Nasreddine *et al.*, 2012).

Some factors contributing to low rates of exclusive breastfeeding are a poor knowledge of breastfeeding techniques and a lack of information regarding the dangers of starting with complementary feeding before the age of 6 months, maternal leave legislation and other work place policies that hinder women's ability to breastfeed. Culture and beliefs may also influence exclusive breastfeeding, and the mother's belief that her breast milk is inadequate may result in mixed feeding (Caroli *et al.*, 2012; WHO, 2014). Poor maternal education and lower household wealth were associated with inappropriate complementary feeding practices in South Asian countries (Senarath *et al.*, 2012)

2.3.2 Feeding practices in South Africa

The 2012 SANHANES reported that for infants below 6 months of age, 17.5% were never been breastfed, 7.4% were exclusively breastfed, and 75.1% were breastfed but not exclusively. For those infants below 24 months of age who have ever been breastfed, breastfeeding was initiated within the first hour after birth for 83% of infants (HSRC, 2013). Breastfeeding up to the age of 2 years is low (31%) (UNICEF, 2014). In relation to Eastern and Southern Africa, South Africa has a lower level of exclusive breastfeeding (< 6 months of

age) and breastfeeding up to the age of 2 years (7.4% versus 51% and 31% versus 62%) (Fig. 2) (UNICEF, 2014). National data on the number of South African children younger than 2 years who do not receive a minimum acceptable diet is lacking (DOH, 2013b; HSRC, 2013).

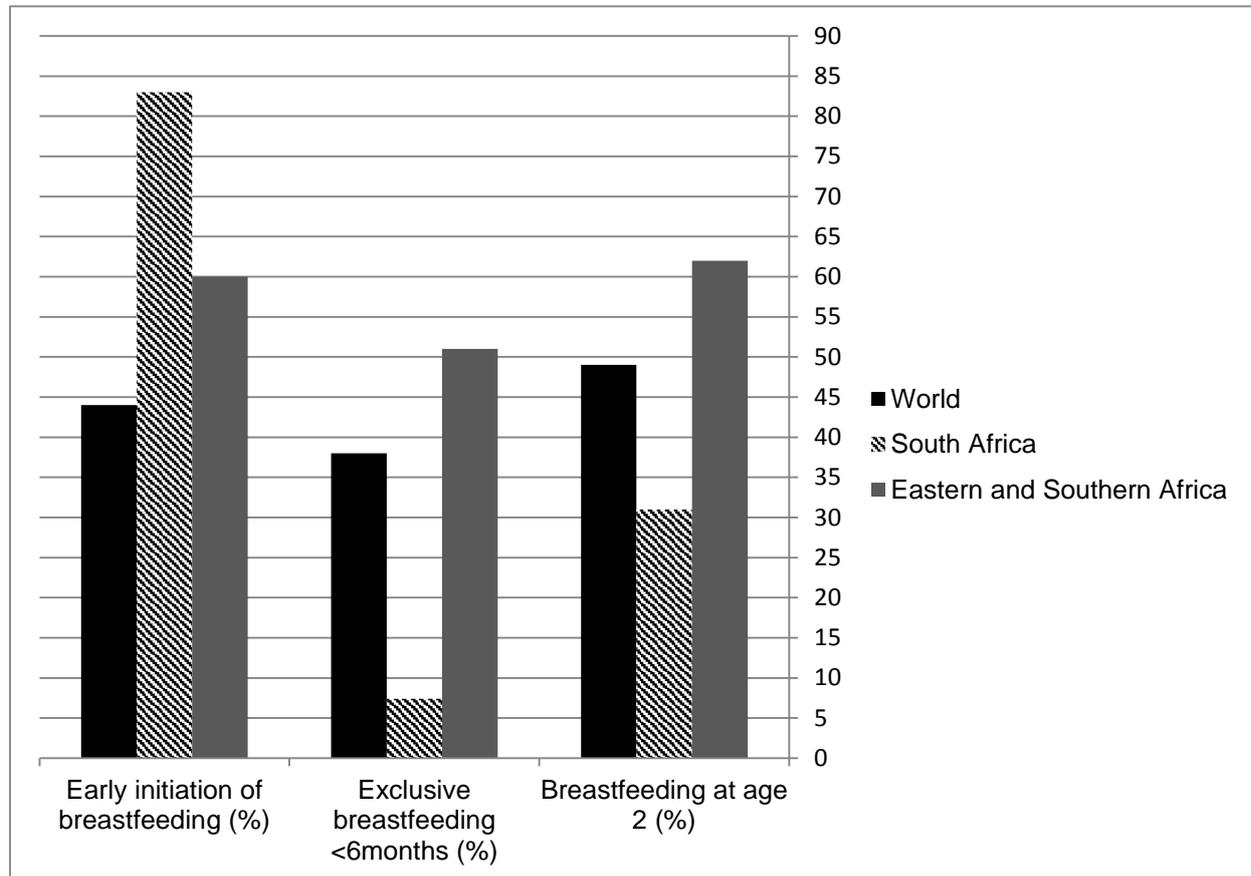


Figure 2. Breastfeeding practices in South Africa in relation to Eastern and Southern Africa and the world (UNICEF, 2014)

A longitudinal study in KwaZulu-Natal (1999–2000) that followed infants from birth for 16 weeks found that 46% of infants received non-breast milk fluids or solid food within 48 hours of birth, and that only 10% of infants were exclusively breastfed for 6 weeks and 6% for 16 weeks (Bland *et al.*, 2002). More than ten years later, a study conducted in the Western Cape (2011) on infant feeding practices during the first 6 months of life showed that 77% of mothers initiated breastfeeding, but only 6% of the infants were exclusively breastfed; 83% of infants were introduced to water in the first 4 weeks of life and 75% of infants received food or formula milk before the age of 3 months (Goosen *et al.*, 2014).

The early cessation of breastfeeding may be due to cultural beliefs that breast milk alone is inadequate and also because of the perception of HIV+ mothers that HIV can be transmitted

through breastfeeding (Nor *et al.*, 2012). Some reasons behind this perception may be the fact that HIV-infected mothers were advised to exclusively breastfeed or formula feed their infants for the first 6 months and they could receive free formula milk through primary health care centres (DOH, 2007). This situation was changed when South Africa adopted the 2010 WHO guidelines on HIV and infant feeding. These guidelines recommend that HIV-infected mothers should practice breastfeeding and use antiretroviral drugs to prevent HIV transmission (DOH, 2013a). Cadegan *et al.* (2012) argued that another reason for the low exclusive breastfeeding rate is the aggressive marketing of breast milk substitutes by the infant feeding industry (Cadegan *et al.*, 2012).

In a review article, including papers published from January 2004 until April 2012, it was concluded that South African infants are generally introduced to solids before the age of 4 months, and that breastfeeding and complementary feeding practices are poor (Du Plessis, 2013). National data collected in 2012 reported that the average age of introduction of semi-solid or solid foods was 4.5 months and 63.5% of children were given semi-solid or solid food before 6 months of age; while 30.7% and 5.8% of infants were introduced to solid/semi-solid foods between 6–8 months and 9–18 months, respectively. It was reported that children living in urban informal areas were introduced to semi-solid or solid foods at a younger mean age, namely 3.9 months compared to 4.5–4.6 months in other localities (HSRC, 2013).

National data collected in 2012 show that the most common complementary food introduced were commercial infant cereal/porridge (51.2%) and homemade cereal/porridge (29%). Children in urban formal areas were more likely to receive commercial cereal/porridge than those living in urban informal and rural informal areas (60.5% versus 41.7% and 41.1%) and less likely to receive home-made cereal/porridge (20.7%) than those living in rural informal (37%) and rural formal areas (39.8%). Less than 5% of infants were given pureed or mashed vegetables or fruit as first complementary food (HSRC, 2013). A study conducted in a rural and an urban area of KwaZulu-Natal reported that the mean age of introduction of solids was 3.5 months in the rural area and 4.2 months in the urban area, with maize meal porridge as the most common food introduced in both areas (70% and 60% respectively), followed by commercial infant cereals (20% and 23% respectively). Animal derived food, vegetables and fruit were not reported for the rural area, whereas only 3% of mothers reported to give potatoes, butternut or rice as a first introductory food. A low dietary diversity was recorded for more than 70% of children in both areas (Faber *et al.*, 2014).

2.4 Prevalence of micronutrient deficiencies

The interpretation of the effect of a single micronutrient deficiency is difficult to interpret. Micronutrients that are obtained from the same food source, for example iron, zinc and vitamin B-12 that are all found in meat, fish and poultry, are often deficient simultaneously (Black, 2003).

According to literature, key nutrients with high public health importance are iron, vitamin A, zinc and iodine (Black *et al.*, 2013). Vitamin A and zinc deficiency have the greatest effect among the micronutrients by causing an estimated 3.1 million child deaths annually (Black *et al.*, 2013). There is strong evidence that zinc, vitamin A and iron deficiency affects growth faltering (Rivera *et al.*, 2003) and iodine and iron deficiency together with stunting may lead to underdeveloped children (Black & Victora *et al.*, 2013).

Figure 3 indicates the global prevalence of anaemia and vitamin A deficiency of children younger than 5 years (Black & Victora *et al.*, 2013). Globally, more than 30% of children younger than 5 years are vitamin A deficient, and the highest percentage of children suffering from vitamin A deficiency are found in Africa. The prevalence of anaemia is the highest in Africa and Asia (between 15 and 20%) (Black & Victora *et al.*, 2013). For zinc, South and Southeast Asia, Sub-Saharan Africa and Central America are identified as being at highest risk of inadequate zinc intake for children younger than 5 years (Wessells & Brown, 2012).

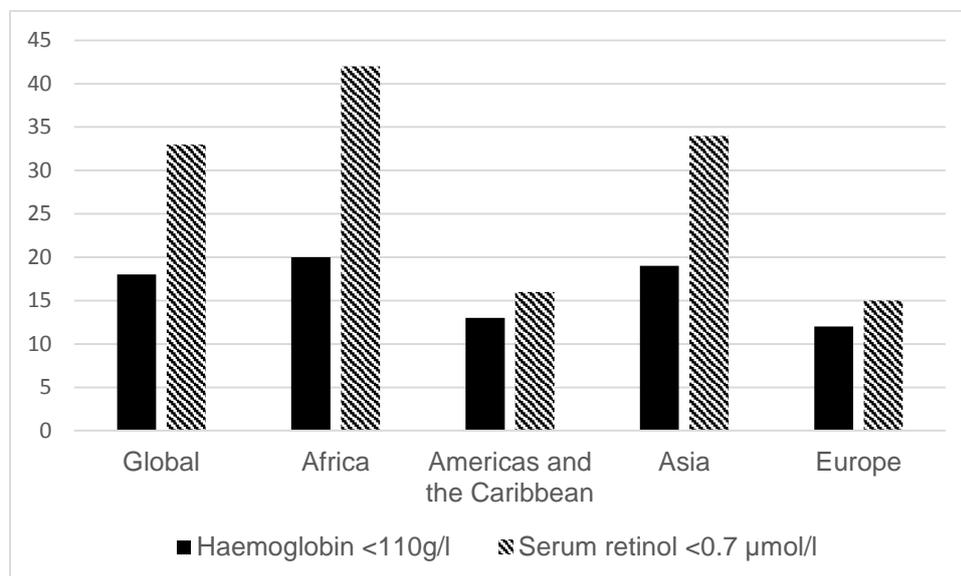


Figure 3. Prevalence of anaemia and vitamin A deficiencies in children younger than 5 years globally (Black & Victora *et al.*, 2013).

#UIC= urine iodine concentration

A systematic review done on iron status of European children aged 6–36 months reported that 2–25% of infants aged 6–12 months were iron deficient. The prevalence of IDA was below 5% for infants and young children in Northern and Western Europe (Eussen *et al.* 2015).

In South Africa, limited recent data is available on the prevalence of micronutrient deficiencies in children younger than 5 years. The 2005 National Food Consumption Survey (NFCS-FB-1) showed that 38.7% of 1–3 year old children were anaemic, 2 out of 3 children aged 1–9 years had poor vitamin A status, 45.3% of children aged 1–9 years had an inadequate zinc status and 5 out of 10 children had an excessive urinary iodine concentration (Labadarios *et al.*, 2008). The 2012 SANHANES data showed that 43.6% of 2–4 year old children were vitamin A deficient and 10.7% were anemic (it should however be noted that the sample size was small, n=437 and n=551, respectively) (Shisana *et al.*, 2014). Baseline data of 2 intervention studies conducted more than ten years ago in KwaZulu-Natal on children aged 6–12 months, showed that 40–46% of the children were anaemic, 18.3–59.5% were iron deficient, 16.1–18% were vitamin A deficient, and 43–48% were zinc deficient (Smuts, Dhansay *et al.*, 2005; Faber *et al.*, 2005).

2.4.1 Indicators for micronutrient deficiencies

Vitamin and mineral deficiencies can only be reduced by and controlled through on-going programmes. Insufficient dietary intake is the primary cause of micronutrient deficiencies. Other conditions like infectious and parasitic diseases, chronic illness and certain genetic conditions may also affect vitamin and mineral status. It is useful to be informed about vitamin and mineral deficiencies to develop appropriate intervention strategies. The WHO recommended the use of indicators to classify the public health significance of vitamin and mineral deficiencies and to monitor progress in the treatment of the vitamin and/or mineral deficiency (Gorstein *et al.*, 2007).

2.4.1.1 Iron

Iron deficiency can be classified by 3 stages of increasing severity: 1) depletion of iron stores (Stage 1), iron deficiency without anaemia – ID (Stage 2), and iron deficiency anaemia – IDA (Stage 3). Risk for iron deficiency is increased during infancy and early childhood when rapid growth occurs, women in reproductive age when iron is lost through menstrual blood and during pregnancy when an increase in red cell mass and growth occurs (Scholl, 2011). Indicators of iron status include red blood cell parameters, free erythrocyte protoporphyrin, plasma iron, total iron binding capacity, transferrin saturation and transferrin receptor (TfR)

and ferritin. In terms of population iron status, plasma ferritin and soluble transferrin receptor may be used in areas where inflammation is not prevalent. In areas of widespread infection or inflammation, acute phase response proteins (APPs) like C-reactive protein and α_1 -acid-glycoprotein can assist in the interpretation of plasma ferritin values as it is also a positive acute-phase protein and is elevated in the presence of infection or inflammation (WHO, 2011b). Plasma ferritin is assessed by enzyme-linked immunosorbent assays (ELISA) or enzyme immunoassays after the collection of venous blood. Plasma ferritin less than 10–12 $\mu\text{g/l}$ is an indication of depletion of iron stores (WHO, 2011b). Nel *et al.* (2015) compared 4 methods that can be used to indicate the prevalence of iron deficiency by adjusting for inflammation, which includes 1) excluding infants with inflammation; 2) using a higher cut-off (SF<12 $\mu\text{g/l}$); 3) using different cut-offs for infants with versus without inflammation (SF<30 $\mu\text{g/l}$ vs. SF<12 $\mu\text{g/l}$); and 4) Adjusting SF concentrations with correction factors (CFs) as described by Thurnham *et al.* (2015). The study concluded that, when SF as only indicator for iron deficiency and both APPs are available, it is recommended to adjust SF by using CFs (Nel *et al.*, 2015).

The prevalence of anaemia is an important public health indicator and may be used with SF to indicate the severity of iron deficiency. Anaemia is a condition where there is insufficient red blood cells to carry oxygen for the body's physiologic needs. Recommended haemoglobin levels to diagnose anaemia at sea level for children 6–59 months of age are 100–109 g/l for mild anaemia, 70–99 g/l for moderate anaemia and lower than 70 g/l for severe anaemia. Residential elevation above sea level may increase haemoglobin concentrations and anaemia may consequently be underestimated in residents at high altitudes. The WHO (2011a) set specific adjustments in place for people living at altitudes higher than 1000 metres above sea level (WHO, 2011a). The population prevalence of anaemia can be indicated in surveys by the recommended cyanmethemoglobin and the HemoCue® system (WHO, 2011a).

2.4.1.2 Vitamin A

Vitamin A deficiency (VAD) is a major cause of morbidity and mortality from infection in especially pregnant women and children younger than 5 years. Vitamin A deficiency is classified as serum retinol <0.70 $\mu\text{mol/l}$ and may be worsened by high rates of infection, especially during diarrhoea and measles (WHO, 2009a). According to WHO (2011c), the prevalence of VAD (serum retinol <0.70 $\mu\text{mol/L}$) in populations indicates the level of public health significance (mild: 2–9%; moderate: 10–19% and severe: 20% or more) (WHO, 2011c). Population vitamin A status can be measured by plasma retinol (pROH) and retinol-binding

protein (pRBP), but inflammation can complicate the interpretation as it lowers both pROH and pRBP (Engle-Stone *et al.*, 2014). The adjustment of pROH with CFs can also be used to indicate the prevalence of VAD (Thurnham *et al.*, 2015). Breast milk vitamin A can also be a good indicator for assessing the vitamin A status of the lactating mother and her breastfed infant (Stoltzfus & Underwood 1995) and is also less affected by inflammation (Engle-Stone *et al.*, 2014).

2.4.1.3 Zinc

The recommended biochemical indicator for zinc is the prevalence of serum zinc concentration below the age/sex/time of day-specific cut-offs. Intervention is necessary when the prevalence of zinc deficiency is more than 20%. The recommended cut-offs represent the 2.5th percentile of the serum zinc distribution from a healthy reference population and are age and sex specific. The time of day of sampling must also be considered. The recommended cut-off is 65 µg/dl for children younger than 10 years when samples are collected in the morning. For dietary indicators, estimated average requirement (EAR) should be used and intervention is needed with a prevalence of inadequate intake greater than 25% (de Benoist *et al.*, 2007). Adequate intake (AI) based on the breast milk content of zinc at different ages and the average milk consumed are used for infants. During the first few months after birth the total zinc transferred through milk falls from about 2.5 mg/day at 1 month to approximately 0.8 mg/day at 6 months. An AI intake of 2.0 mg/day is recommended for 0 to 5 months old infants (Hotz & Brown, 2004). Serum or plasma zinc is the best available biomarker of the risk of zinc deficiency in populations. Low height-for-age may also indicate the prevalence of zinc deficiency. A low height-for-age prevalence of more than 20% may indicate zinc deficiency (de Benoist *et al.*, 2007). There is a lack of reliable, widely accepted indicators of zinc status. A systematic review concluded that plasma, urinary, and hair zinc are biomarkers of zinc status, but there is still a need for high-quality studies using these biomarkers. Urinary zinc excretion (24h) and hair zinc can provide information on zinc status in zinc-supplemented individuals, but zinc status in depleted individuals is not certain. Enzymes and zinc-binding proteins to indicate zinc deficiency must be further investigated (Lowe *et al.*, 2009).

2.4.1.4 Iodine

Iodine can mainly be found in the thyroid gland in minute amounts (WHO & FAO, 2006). Although there is currently a worldwide awareness to reduce iodine deficiency, it is estimated that worldwide, 35% of people still have insufficient intake of iodine (Aburto *et al.*, 2014). Salt iodine content is a useful proxy for exposure in areas where the iodine intakes are low in poor

communities, and iodized salt used in the household may be used as an indicator of iodine nutrition. More than 90% of dietary iodine is excreted in the urine; therefore urinary iodine concentration can be used as an indicator of recent iodine intake. Thyroid-stimulating hormone is mainly used to determine the concentrations of circulating thyroid hormone, which reflects iodine intake. Serum thyroid hormone T3 and T4 can also be used in iodine-deficient populations. The measurement of the goitre may also be used by neck inspection, palpation and thyroid ultrasonography. The WHO recommends that the total goitre rate be used to define the severity of iodine deficiency in populations (< 5% - iodine sufficiency; 5–19.9% - mild deficiency, 20–29.9% - moderate deficiency and > 30% severe deficiency). However, in areas of mild iodine deficiency, the palpation of goitre has poor sensitivity and specificity (Rohner *et al.*, 2014).

2.5 Growth faltering and overweight

Micronutrient deficiencies and undernutrition contribute to the global burden of disease. Inadequate dietary intakes and infectious diseases lead to growth faltering and childhood overweight leads to adult obesity, diabetes and non-communicable diseases (Black & Victora *et al.*, 2013).

Growth status can be assessed by anthropometric indices, namely weight-for-height, height-for-age and weight-for-age (WHO & UNICEF, 2009b). Percentiles and Z-scores in anthropometric measures can be used to assess nutritional status and growth. (Whang & Chen, 2012). Mid-upper-arm circumference (MUAC) is also used as an independent diagnostic criterion to indicate malnutrition (MUAC <115 mm) in children aged 6 to 60 months old) (WHO & UNICEF, 2009b).

Length/height-for-age: Linear growth has a part based on length (length-for-age, 0–24 months) and another on height (height-for-age, 2 years and older) (WHO, 2006a).

Low height-for-age: Stunting is defined as below -2 standard deviations from the median height-for-age of the reference population and is seen as linear growth faltering (UNICEF, 2014). Stunting reflects chronic malnutrition. Childhood stunting correlates with non-communicable diseases, reduced productivity later in life and impaired neurocognitive development (WHO, 2014). Growth failure can already begin in utero and continues after birth as 20% of childhood stunting is caused by intrauterine growth restriction due to maternal undernutrition (Black, Alderman *et al.*, 2013)

Low weight-for-height: Wasting, defined by WHO (2014) as a 'reduction or loss of body weight in relation to height'. A child is classified as wasted when he or she has fallen below -2 SD of the median of the WHO growth standard population weight-for-height (WHO/UNICEF/WFP, 2014). Weight-for-height below -3 SD may be used as a criterion to identify severe acute malnutrition in infants and children (WHO & UNICEF, 2009b).

Wasting is a condition in which children lose weight rapidly and is caused by a combination of infection and a diet of poor nutrient. Some underlying causes of wasting are poor access to appropriate and timely health care, inadequate feeding practices, poor food security and a lack of a hygienic and clean environment (WHO/UNICEF/WFP, 2014). Severe infection at an early age like measles, pneumonia, meningitis and malaria can cause severe wasting and also have long-term effects on linear growth (Black, Victora *et al.*, 2013).

High weight-for-height: According to WHZ-scores, a value of ≥ 2 SD indicates high weight-for-height (overweight) in children (Wang & Chen 2012). Body mass index-for-age [Body mass index is the ratio weight (in kg)/recumbent length or standing height (in m²)] is a measurement that combines weight and height data and can be used to indicate overweight in children (Wang & Chen 2012).

Low weight-for-age: Underweight is seen as a low weight-for-age (of less than -2 SD) and is influenced by the height-for-age and weight-for-height of a person (Wang & Chen, 2012).

Growth charts are useful in the assessment of stunting, wasting and overweight in children and consequently indicate the general well-being of populations. The WHO published new child growth standards for attained weight and height in 2006 based on breastfed infants and appropriately fed children of different ethnic origins (WHO, 2006a).

In order to classify levels of stunting, underweight and wasting at population level, WHO developed prevalence ranges, which are shown in Table 3 (WHO, 2015b)

Table 3. Classification for assessing the severity of malnutrition by prevalence ranges among children under 5 years of age (WHO, 2015b)

Indicator	Severity of malnutrition by prevalence ranges (%)			
	Low	Medium	High	Very high
Stunting	<20	20–29	30–39	≥40
Wasting	<5	5–9	10–14	≥15
Underweight	<10	10–19	20–29	≥30

2.5.1 Global prevalence of growth faltering and overweight.

Currently, the global stunting rates are dropping, between 1990 and 2014 the prevalence of stunting declined from 39.6% to 23.8%, but 159 million children around the world are still affected by stunting. There are 10 million more children overweight than 2 decades ago, the prevalence of overweight increased slightly from 4.8%–6.1% between 1990 and 2014. It was reported in 2014 that 1 out of 13 children in the world was wasted (50 million children under the age of 5 years). Almost a third of all wasted children were severely wasted (WHZ < -3 SD) with a global prevalence of 2.4%. In Africa, when considering children younger than 5 years, the prevalence of stunting is rising and the prevalence of overweight nearly doubled since 1990. Nearly all wasted children under the age of 5 years are living in Asia and Africa (68% in Asia and 28% in Africa) (UNICEF, WHO, World Bank, 2015).

Figure 4 illustrates the global prevalence of underweight, stunting, wasting and overweight for children younger than 5 years by region. According to the WHO (2015b) classification for the severity of malnutrition within in a population (see table 3), the prevalence of underweight is high in South Asia (32%); the prevalence of stunting is high in Eastern and Southern Africa (39%), South Asia (38%), Sub-Saharan Africa (37%) and least developed countries (37%); while the prevalence of wasting is high in South Asia (15%) and West and Central Africa (11%). The highest prevalence of overweight is reported to be in Middle East and North Africa (10%) (UNICEF, 2014).

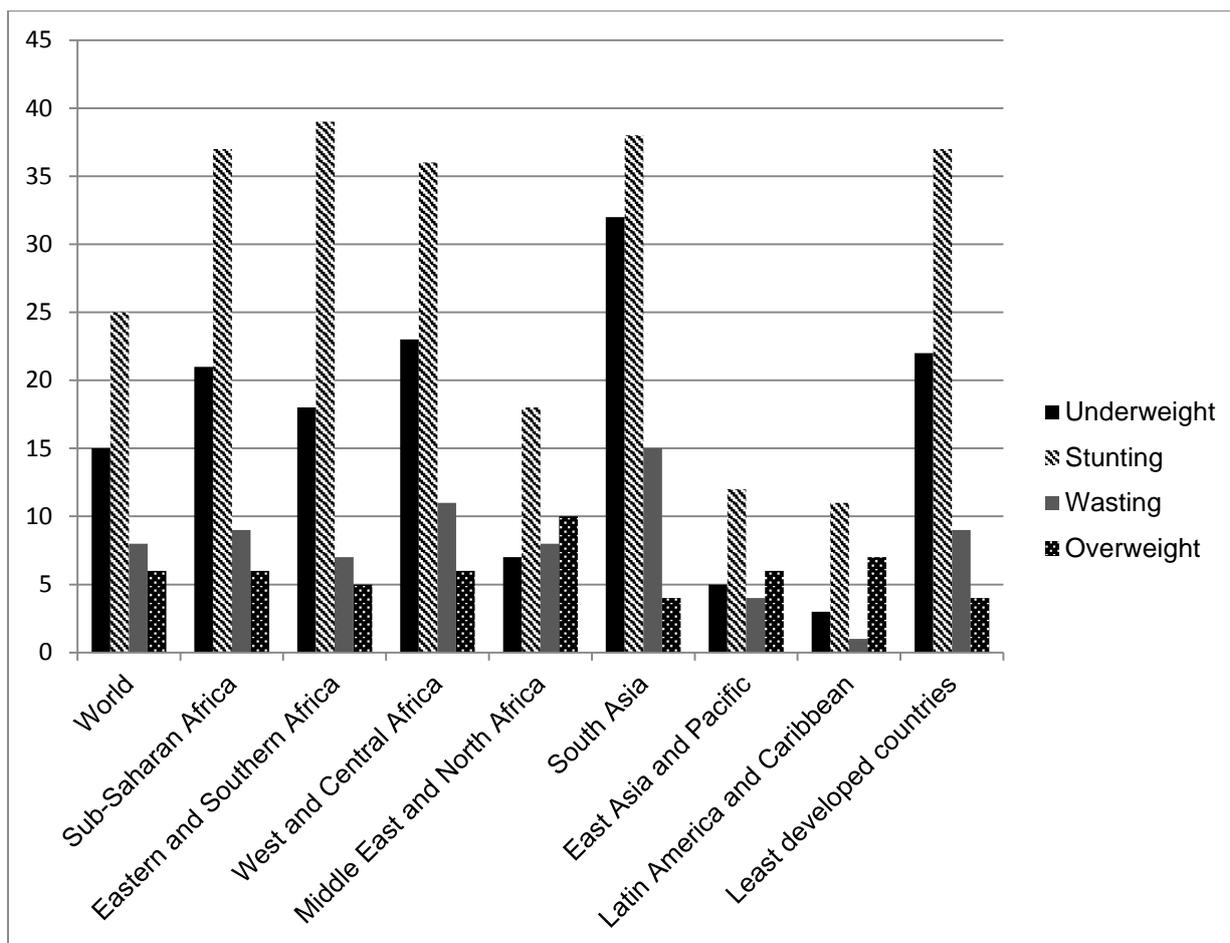


Figure 4. Global prevalence of underweight, stunting, wasting and overweight for children younger than 5 years (UNICEF, 2014)

2.5.2 Growth faltering in South Africa

Undernutrition, stunting and micronutrient deficiencies with the co-existence of increasing incidence of overweight, indicates that South Africa is in a nutrition transition (DOH, 2013b). Figure 5 indicates the prevalence of stunting, wasting and underweight in children younger than 5 years at national and provincial level (below -2 SD) as reported in 2012. Nationally, for children under the age of 5 years, 22% were stunted and 7% were severe stunted, 2.5% were wasted and 1.2% were severe wasted. The highest rate of stunting can be found in Mpumalanga (30%), followed by the Northern Cape, Free State and the North West (28%). The prevalence of wasting and underweight are the highest in the Northern Cape (11% and 15% respectively). It is reported that the highest percentage of severe wasting (8%) was also found in the Northern Cape (HSRC, 2013). The 2012 SANHANES showed that at national level for children aged 2–5 years, the prevalence of overweight and obesity for boys was 17.5% and 4.4% respectively, and for girls 18.9% and 4.9% respectively (Shisana et al. 2014)

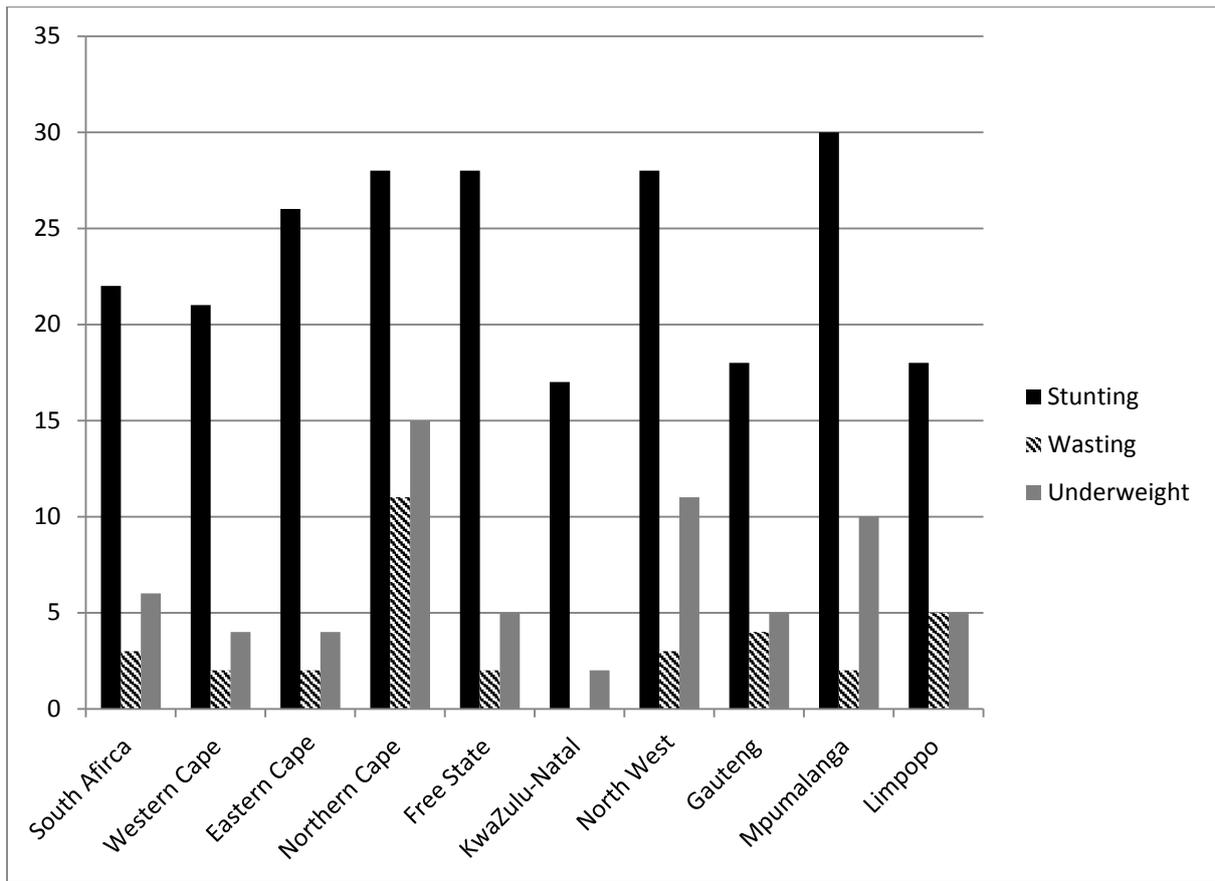


Figure 5. The prevalence of stunting, wasting and underweight in children younger than 5 years at national and provincial level (below -2 SD) (HSRC, 2013).

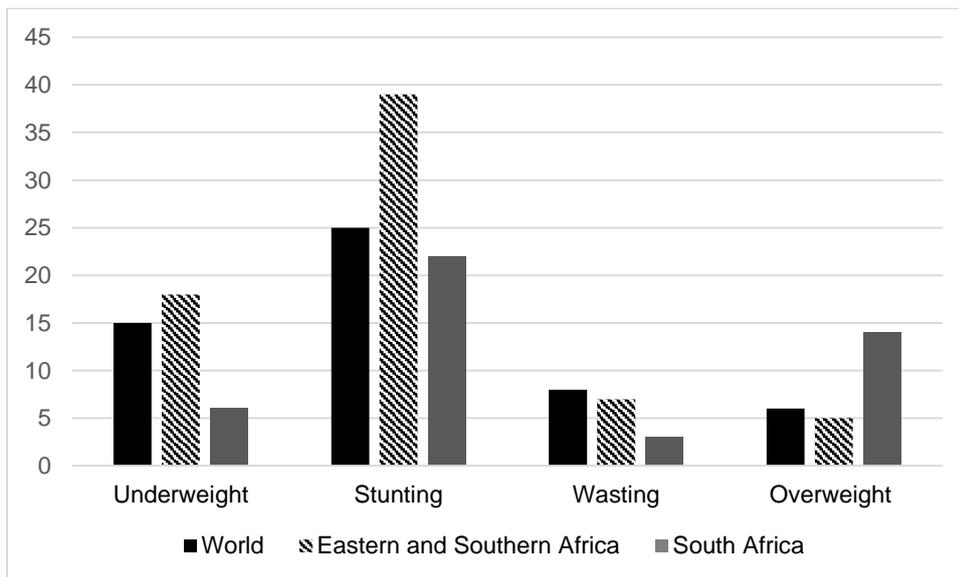


Figure 6. Prevalence of underweight, stunting, wasting and overweight in the world, Eastern and Southern Africa and South Africa for children aged 0-5 years (HSRC, 2013; WHO, 2015c).

Globally, the prevalence of underweight, stunting and wasting are 15%, 25% and 8%, respectively for children younger than 5 years. In South Africa, the prevalence of underweight, stunting and wasting for children younger than 5 years are 6%, 22% and 3% respectively. (Fig. 6). Compared to the Eastern and Southern African region, South Africa has a lower prevalence of underweight, stunting and wasting, but a higher prevalence of overweight (HSRC, 2013; WHO, 2015c).

2.5.3 Child growth and development

Poor maternal nutrition and infections may cause intrauterine growth restriction during a critical period for brain development (Walker *et al.*, 2007). Infants born small for gestational age have an increased risk of death throughout infancy and a risk for growth faltering in the first 2 years of life and it is also estimated that 20% of stunting is due to foetal growth restriction (Black & Victora *et al.*, 2013). Significant associations were found between stunting and delayed cognitive development (Fernald *et al.*, 2009). Stunting can be used as a risk marker of poor child development, as stunting before the first thousand days may indicate poorer cognitive and educational outcomes later in life (Black & Alderman *et al.*, 2013). De Onis (2006) studied the relationship between motor development and physical growth and found that reaching motor milestones is mostly independent of variations in physical growth in healthy populations (de Onis, 2006).

2.6 Infant and child development

Critical foundations for healthy child development are: 1) stable, responsive and nurturing caregiving with potential to learn, 2) safe, supportive physical environments, and 3) appropriate nutrition (Chan, 2014). Developmental domains include the cognitive, language, executive function/self-regulatory, social-emotional and motor domains. Cognitive skills for infants and toddlers include problem-solving with objects by understanding what is meant when asked 'one' or 'two' of an object (Jurado & Rosselli, 2007). Language development already starts at babbling and gesturing in the first months after birth followed by first words and sentences between 2 and 3 years. Social-emotional development consists of preferential attachments to caregivers, willingness to explore new objects and spaces and enjoy the introducing and interacting with people. Motor development can be classified in large motor skills like rolling-over, sitting, walking, running and jumping while fine motor skills refer to drawing and writing, which involves eye-hand coordination and muscle control (Fernald *et al.*, 2009). The 6 gross-motor milestones according to the WHO (2006b) are sitting with support, standing with assistance, hands-and-knees crawling, walking with assistance, standing alone,

walking alone. The age of achievement for these motor milestones are also illustrated in Table 4.

Table 4. Age categories (months) for achievement for six gross motor milestones (WHO, 2006b)

Motor milestone	Mean (SD) in months	Months (95% CI)	
		1 st Percentile	99 th Percentile
Sitting without support	6.0 (1.1)	3.8 (3.7, 3.9)	9.2 (8.9, 9.4)
Standing with assistance	7.6 (1.4)	4.8 (4.7, 5.0)	11.4 (11.2, 11.7)
Hand-and-knees crawling	8.5 (1.7)	5.2 (5.0, 5.3)	13.5 (13.1, 13.9)
Walking with assistance	9.2 (1.5)	5.9 (5.8, 6.1)	13.7 (13.4, 14.1)
Standing alone	11.0 (1.9)	6.9 (6.8, 7.1)	16.9 (16.4, 17.4)
Walking alone	12.1 (1.8)	8.2 (8.0, 8.4)	17.6 (17.1, 18.0)

2.6.1 Child development assessment tools in low-income and middle-income countries

A comprehensive assessment of child development is needed for infants and children from birth to 3 years, because of intense development and interconnections among domains. Direct observation of the child by the assessor is important and the primary caretaker can be helpful by reporting the abilities in the different domains of development of his/her infant (Fernald *et al.*, 2009). Infant and young child development can be assessed by 1) direct testing; 2) obtaining ratings or reports of the child's behaviours or skills by informants like parents or primary caretaker, and 3) observation of the child in daily or structured activities. The reliability and validity of the measurement are increased by measuring multiple domains and the use of multiple tests and methodologies to measure both within and across domains like using parent rating, direct child testing and/or observation (Sabanathan *et al.*, 2015).

Brain imaging markers are also successful in the reflection of changes in brain and cognitive function and may also be used for sensitive measures of long-term effects (Sizonenko *et al.*, 2013). The WHO Care for Development guidelines were developed to support health professionals in giving guidance on the stimulation of child development (Bentley *et al.*, 2014).

Child development assessment tools (CDATs) were developed to evaluate developmental domains, namely cognitive, language, motor, adaptive and socio-emotional to guide future interventions. Each domain of development can be assessed by looking at certain activities that must be achieved by a certain age. Some of the limitations of CDATs is that there are limited data available on a single CDAT that can be used in all populations and therefore a variety of tools have been developed worldwide (Sabanathan *et al.*, 2015).

Challenges for the assessment of child development include intensive training for the assessors by considering test/re-test reliability and consistency of timing during assessment (Fernald *et al.*, 2009). The test used for assessment must be fair by considering demographic characteristics (including familiarity), type of material, cultural relevance items, testing situation and the importance in the type of responding. Child development can be formed by cultural ideas (Fernald *et al.*, 2009). Parents have different cultural child-rearing practices, which influence gross motor development. A study based on the development of 3–9 month-old Cameroonian and German infants showed that there are differences in motor and language development between these different cultures (Vierhaus *et al.*, 2011).

The Bayley Scales of Infant Development (BSID) is the world's most common assessment tool used for individual assessment for infants and includes language, cognitive and social-emotional measurements (Fernald *et al.*, 2009). The second edition of the Bayley Scales of Infant Development was recently adapted by Biasini *et al.* (2015) for use by paediatric professionals in low-and-middle income countries with limited resources. Findings were that it can be successfully used to determine child development (Biasini *et al.*, 2015).

Only a few CDATs are available in low and middle income countries and most are applied in research settings. The Kilifi Developmental Inventory (KDI) was developed in Africa by adapting a range of CDATs and is a continuous measure used for the assessment of psychomotor development in a resource-poor setting (Fernald *et al.*, 2009; Sabanathan *et al.*, 2015). This inventory was designed to be used by assessors with little experience in child development and is an affordable tool for use in field studies. The child's performance is assessed in 2 main domains, namely locomotor development and fine motor skills by a parental report and direct observation. Early executive function and emotional control are also measured. Items used for assessment are based on Griffiths Mental Development Scale, the Kenyan Screening Test for Children aged 6 months to 6 years, The Portage Early Education Programme and the Movement Assessment Battery for Children (Fernald *et al.*, 2009). The KDI has been evaluated for reliability and validity in normal and disease exposed populations

in coastal Kenya (Abubakar *et al.*, 2008). The Parent Rating Scales of Motor and Language Development measures gross motor and language milestones for children 6–59 months and was used in Tanzania and Nepal (Fernald *et al.*, 2009). The Parent Rating Scales of Motor Development was used in a South African randomised controlled trial that assessed developmental outcomes in infants at age 6-12 months and again at age 12-18 months (Faber *et al.*, 2005). Abubakar *et al.* (2007) assessed the developmental outcomes in children from Kenya by using both the KDI and Parental Rating scales. They reported a significant positive relationship between most KDI scores and the Parental Rating scores (Abubakar *et al.*, 2007).

The Malawi Developmental Assessment Tool was developed by Gladstone *et al.* (2010) and is useful to assess socio-emotional items for 0–72 month old children (Gladstone *et al.*, 2010). A Guide for monitoring child development in children 0–24 months was developed in Turkey and is based on an open-ended precoded interview with the parent with no specific cognitive domain questions. This guide may be useful for monitoring "at risk" populations (Ertem *et al.*, 2008). Rapid neurodevelopmental assessment was developed in Bangladesh for children 0–2 years and can be used in small high risk populations (Khan *et al.*, 2010). Recently, the INTERGROWTH-21st Neurodevelopment Assessment was developed to assess children older than 2 years and can be used by non-specialists in low-, middle- and high-income countries. This tool measures vision, cortical auditory processing, cognition, language skills, behaviour, motor skills, attention and sleep-wake patterns (Fernandes *et al.*, 2014).

There are therefore a number of tools available for assessing cognitive and motor development in infants and young children. Some of these tools have specifically been adapted to low resource settings and can be used in nutrition-related research to assess motor milestone development and other cognitive functions. Using these types of tools, a number of studies have demonstrated the effect of micronutrient interventions on motor milestone development (Fernald *et al.*, 2009).

2.6.2 Micronutrient deficiencies and early childhood development

The relationship between early motor development and micronutrient deficiency is inseparable as micronutrients are related to motor development (Prado & Dewey, 2014). The effect of micronutrient deficiencies on motor development is effected by the stage of development, the degree of deficiency and the duration of inadequate intake (Anjos *et al.*, 2013). Deficiencies of key nutrients like fatty acids, iron, zinc, iodine, choline and B-vitamins have an effect on the 5 key neurodevelopmental processes, namely 1) neuron proliferation, 2) axon and dendrite growth, 3) synapse formation, pruning and function, 4) myelination and 5) neuron apoptosis

(Prado & Dewey, 2014) The effects of some key nutrient deficiencies on psychomotor-development are discussed next.

Iron: Iron deficiency is most prevalent during the first 2 years of life when many parts of the brain are becoming myelinated and deficiency may influence the central nervous system (Walter, 2003). Fifty percent of anaemia is caused by iron deficiency whereas vitamin A, vitamin B12 and folate deficiency, malaria, HIV and other infectious diseases also cause anaemia. A number of observational studies reported that anaemic infants showed lower academic performance during their school-age years, even after the treatment of the anaemia (Grantham-McGregor and Ani, 2001; Black, 2003). A study done by Shafir *et al.* (2008) in the USA reported that iron deficient infants (9–10 months) with or without anaemia have poorer motor development (Shafir *et al.*, 2008).

Zinc: Even though zinc is the fourth most abundant ion in the brain and contributes to brain structure and function, evidence from human studies are lacking. Animal studies showed that zinc deficiency caused poor attention, learning and memory (Prado & Dewey, 2014). Black (2003) concluded from a review of 6 zinc-supplementation trials that zinc supplementation was associated with more activity and improved the quality of motor development in 3 of the trials, whereas 3 found no impact on motor development (Black, 2003). Low maternal intakes of zinc during pregnancy and lactation were shown to be associated with less focused attention in neonates and decreased motor development at the age of 6 months (Bhatnagar & Taneja, 2001).

Iodine: Iodine deficiency is seen as the most common preventable cause of mental retardation and may cause congenital hypothyroidism and irreversible mental retardation (Walker *et al.*, 2007). Chronic iodine deficiency results in poor intelligence. Results from a meta-analysis showed that children with iodine deficiency compared to individuals with healthy iodine levels have a lower IQ (Qian *et al.*, 2005). The effect of iodine deficiency on neurodevelopment may already start during pregnancy. Mild iodine deficiency in the first trimester of pregnancy can have a negative effect on children's cognition 8 years later (Bath *et al.*, 2013).

Fatty acids: Brain fatty acid composition is dependent on dietary fatty acid availability, therefore fatty acid imbalance and deficiencies may lead to impaired development (Wainwright, 2002). Animal studies showed that DHA deficiency resulted in reduced neuron proliferation and fatty acid deficiency reduces the amount of and alters the composition of myelin (Prado & Dewey, 2014).

B-vitamins: Vitamin B12, folate and choline: Vitamin B12, folate and choline play an important part in the developing brain. Folic acid deficiency may cause neural tube defects and vitamin B12 deficiency disrupts early development through myelination and dendrite formation (Black, 2008). Vitamin B12 deficiency affects the central nervous system and may cause brain atrophy. It is seldom found that vitamin B12 deficiency occurs before 6 months of age, because infants store vitamin B12 in the liver, even if the infant is born to a vitamin B12-replete mother. However, after 6 months of age, infants may be vulnerable to vitamin B12 deficiency if the breastfeeding mother is vitamin B12 deficient and/or the infant consumes a small amount of animal source food (Black, 2008). Vitamin B12 deficiency may cause long-term neurologic and cognitive impairment like irritability, anorexia, developmental regression and poor intellectual progress (Graham *et al.*, 1992; von Schenck *et al.*, 1997; van Dusseldorp *et al.*, 1999). Folate and choline metabolism are interrelated. The hippocampus area of the brain (integral part for learning and memory) is mostly affected by choline deficiency. Choline deficiency increases the rate of neuronal cell death (Zeisel, 2006a & 2006b).

2.7 Actions needed to achieve optimum infant and child nutrition and development

World leaders adopted the Millennium Declaration and set 8 Millennium Development Goals (MDGs) to be met by September 2015. The first MDG focuses on undernutrition, which includes the prevalence of underweight and the proportion of the population below minimum level of dietary energy consumption. Minimal focus was placed on nutrition or specific methods on how to achieve the nutrition goals. Following the MDGs, the post-2015 Sustainable Development Goals (SDGs) will be launched at the end of 2015 according to planning. Six global nutrition targets are embedded in the SDGs developed by the WHA (Korenromp, 2015). The 6 nutrition targets are:

1. 40% reduction in the number of children under 5 who are stunted
2. 50% reduction of anaemia in women of reproductive age (pregnant and non-pregnant)
3. 30% reduction in low birth weight
4. No increase in childhood overweight
5. Increase the rate of exclusive breastfeeding in the first 6 months up to at least 50%
6. Reduce and maintain childhood wasting to less than 5%

The progress towards reaching these goals are presented in Table 5. Globally, there has been a 20% reduction in stunting in children younger than 5 years (the target is 40%), a 10%

reduction in low birth weight (the target is 30%); and a 41% increase in EBF (the target is 50%). There has been no progress towards the reduction of childhood wasting (IFPRI, 2014; UNICEF, WHO & World Bank, 2014).

Table 5. Global progress of stunting, exclusive breastfeeding and wasting toward the global WHA targets (IFPRI, 2014; UNICEF, WHO and World Bank, 2014)

WHA target for 2025	Progress towards 2025 target
40% reduction in the number of children under 5 who are stunted	20% reduction noted in 2012
30% reduction in low birth weight	10% noted in 2012
No increase in childhood overweight	Increased from 4.8%–6.1% as noted in 2014
Increase the rate of exclusive breastfeeding in the first 6 months up to at least 50%	A rate of 41% was reported in 2012
Reduce and maintain childhood wasting to less than 5%	No progress (was 8% globally in 2013)

In order to reach the WHA targets, it is important to address malnutrition by focussing on poverty, maternal health, child mortality and education (World Bank, 2005). Potential benefits of some proven interventions to manage malnutrition include: 1) Empowering women to practice optimal nutrition during the first thousand days, which includes exclusive breastfeeding, antenatal supplements, appropriate complementary feeds from age 6 months and food-related hygiene; 2) enabling adequate intake of vitamins and minerals through diverse diets, fortified foods and supplements, and 3) ensuring that those who are at risk of malnutrition have access to and benefit from nutrient dense food through nutritional management of infections and therapeutic feeding (Bhutta *et al.*, 2008).

Figure 7 illustrates actions needed to achieve optimum infant and child nutrition and development (Black & Victora *et al.*, 2013). First, the importance of a supporting environment needed for optimum infant and child nutrition and development is discussed, followed by some aspects of nutrition-sensitive interventions and programmes. For nutrition-specific interventions, some aspects of disease prevention and management are discussed as well as micronutrient supplementation and fortification.

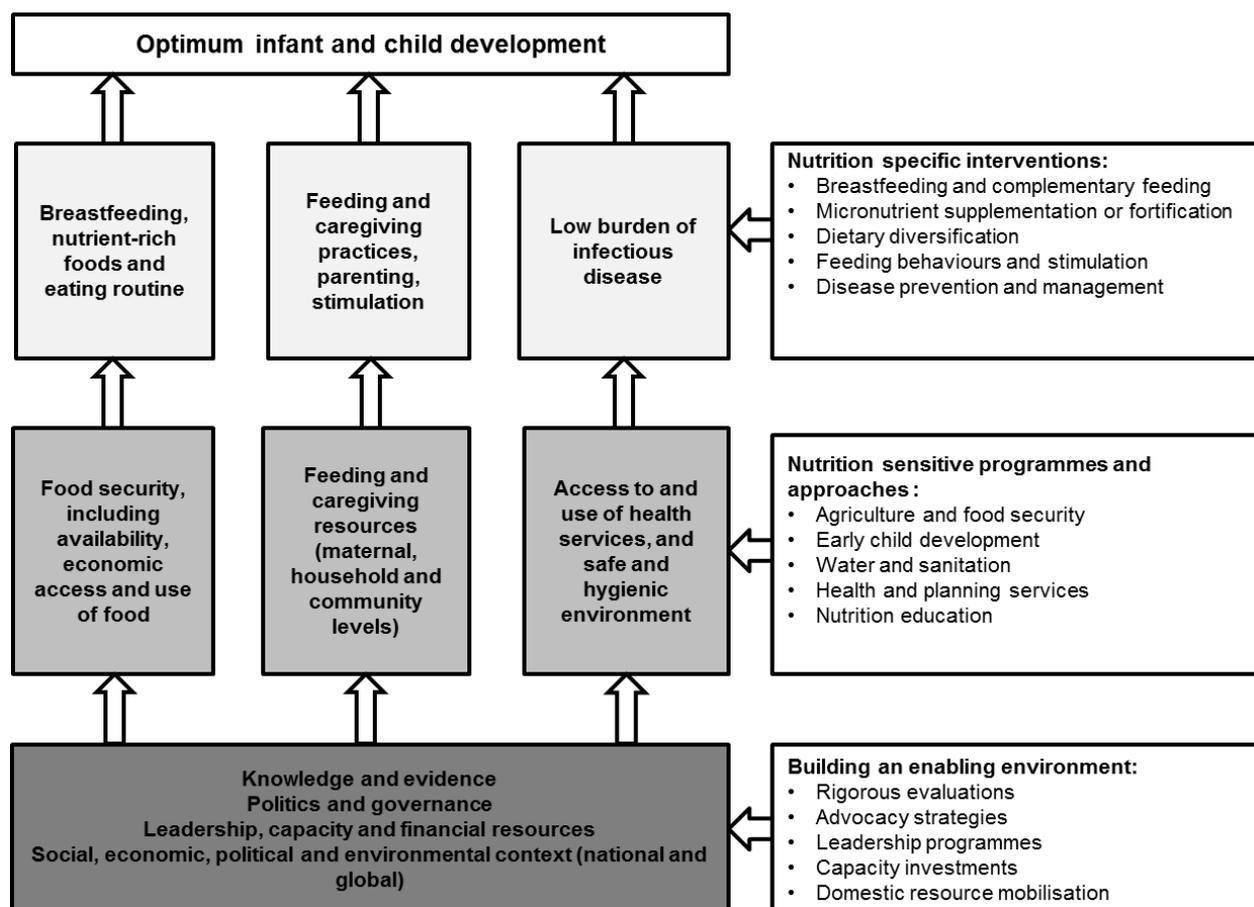


Figure 7. Actions needed to achieve optimum infant and child nutrition and development (Source: Black & Victora *et al.*, 2013).

2.7.1 Supporting environment

The success of any strategy for combating malnutrition depends on the country's political and policy leadership for enabling the environment in terms of policies, leadership, institutional and individual capacities in addition to a strong commitment to develop and implementation of acceptable, sustainable solutions (Gillespie *et al.*, 2013). There has been a lack of clear policies to address micronutrient deficiencies in young children living in resource-poor countries (Neufeld & Ramakrishnan, 2011) although the recent Scaling up Nutrition (SUN) movement has galvanised efforts towards policy development. The primary purpose of the SUN Movement Civil Society Network is to encourage the formation of civil society alliances in each SUN country to ensure that nutrition remains a high priority at global and national agendas. Thirty-three countries have already established the SUN civil society alliances. Countries like Bangladesh, Uganda, Peru, Pakistan, Cameroon and Kenya, which form part of the SUN movement, have showed that a multi-sector approach can be successfully

implemented to scale up nutrition (SUN, 2014). The most recent Global Nutrition Report has recommended that there must be a better political involvement regarding infant and young child feeding globally with special focus on breastfeeding practices to increase new-born survival (IFPRI, 2014).

Over the years there have been efforts by organizations like WHO, UNICEF and other advocacy organizations to encourage an enabling environment for optimal infant and young child feeding. WHO and UNICEF developed the Global Strategy for Infant and Young Child Feeding (See Annexure 1, Box 3). This strategy aims to improve the nutritional status, growth and development, health and the survival of infants and young children through optimal feeding (WHO & UNICEF, 2003). The Global Strategy for Infant and Young Child Feeding is based on the Baby-friendly Hospital Initiative (renamed as the Mother-Baby Friendly Initiative) (WHO & UNICEF, 2009a) (See Annexure 1, Box 4), the International Code of Marketing of Breast milk Substitutes (1981), the Innocenti Declaration on the protection, promotion and support of breastfeeding (1990) (See Annexure 1, Box 5), the overall context of national policies and programmes on nutrition and child health, and it is consistent with the World Declaration and Plan of action for Nutrition. The improvement of health practices at health facilities and at home can be based on the Integrated Management of Childhood Illness (IMCI) strategy that was developed by the WHO and UNICEF (Ahmed *et al.*, 2010).

South Africa is a member of the United Nations and adopted the WHA International code of Marketing of Breast milk Substitutes and agreed to remove non-compliant products from the market by 6 December 2015 (DOH, 2013c). South Africa also agreed on the Innocenti Declarations part of the Convention on the Rights of the Child. Therefore, South Africa is committed to protect and promote breastfeeding. However South Africa has struggled with very low levels of exclusive breastfeeding in particular. Earlier policies provided HIV-positive mothers with formula through public institutions, but the Tshwane Declaration for the Support of breastfeeding that was published in 2011 (See Annexure 1, Box 1) was a turn-around. The National Breastfeeding Consultative Meeting which led to the Tshwane Declaration was held in August 2011. There South Africa declared to actively promote, protect and support exclusive breastfeeding as a public health intervention to optimise child survival. The declaration also recommended the cessation of provision of formula milk at public health facilities, encouraging exclusive breastfeeding even for HIV-positive mothers in accordance with WHO guidelines. Another initiative, the Campaign on Accelerated Reduction of Maternal and Child Mortality in South Africa (CARRMA) was launched in 2012 to guide the efforts and implementation of

existed plans and strategies. The National Infant and Young Child Feeding Policy (IYCF) was released in 2013 and is aligned with several global initiatives, including the Convention on the Right of the child, the International Code of Marketing of Breast milk Substitutes, the Innocenti Declaration, the Mother-Baby Friendly Initiative (MBFI), the United Nations Joint Guidelines on HIV and Infant Feeding 2010, CARMMA, Roadmap for Nutrition in South Africa and the Strategic Plan for Maternal, Newborn, Child and Women's Health (MNCWH) and Nutrition (DOH, 2013a). Recently, a 5-year roadmap for Nutrition for South Africa 2013-2017 was set in place to provide a framework for nutrition and nutrition-related issues in health care systems like the Strategic Plan for Maternal, Newborn, Child and Women's Health (MNCWH) and Nutrition in South Africa. The roadmap focuses on increasing life expectancy; decreasing maternal and child mortality, combating HIV and AIDS and decreasing Tuberculosis (TB) (DOH, 2013b). However, there has also been missed opportunities, one of which is the lack of a nutrition component to the recent community-based Primary Health Care Re-engineering Programme (DOH et al. 2014).

2.7.2 Nutrition-sensitive interventions and programmes

Nutrition-sensitive interventions and programmes address the underlying determinants of child nutrition and development, namely food security, adequate caregiving resources at the maternal, household and community levels, access to health services and a safe and hygienic environment (Ruel & Alderman, 2013).

2.7.2.1 Food security

As shown in Figure 7, household food security, care and a healthy environment are the underlying determinants influencing the nutritional intake, health and status of an infant and young child (Black *et al.*, 2013). Infants and young children are dependent on the primary caretaker's feeding practices and feeding styles (de Groot *et al.*, 2015). Global trends show that just over 1 in 9 people is undernourished because of a lack of the food they need for an active and healthy life (FAO *et al.*, 2015).

Two studies conducted in rural areas of 2 provinces in South Africa showed that poverty was prevalent in a large proportion of the study area and approximately 30% of households reported that they do have enough food for consumption. The local production of food may potentially contribute to the availability of food at household level by selective crop production of nutrient dense foods (like vitamin A rich vegetables). Also, many households lacked electricity, used an open fire to cook food and used river water for consumption. Health

services were poorly utilised. Even though the major source of income was child support and pension or disability grants, a large percentage of households did not receive a child support grant (Schoeman *et al.*, 2010; Smuts *et al.*, 2008). Some social protection agendas include social transfers, programmes ensuring access to social services, social support and care services (Ruel *et al.*, 2013). Cash transfer programmes deliver cash directly to households and focus on poor and vulnerable groups. These programmes are shown to be successful in increasing resources to food, health and care in especially sub-Saharan Africa (de Groot *et al.*, 2015).

A recent example of a country that faces poor food security is Kenya. Kenya continues to face high levels of drought, floods and cross-border civil strife. Due to below average rains, there are constrained food access and high food prices. Malnutrition amongst children are high and access to safe water is low. Access to routine immunisations, maternal and neonatal care and nutritional services are hindered by violence, leading to further vulnerabilities. Kenya also remains susceptible to the current Ebola outbreak due to weak health systems. UNICEF and partners support more than 1.2 million children affected by food insecurity, malnutrition and disease outbreaks (UNICEF, 2015).

2.7.2.2 Water and sanitation

In sub-Saharan Africa, some of the leading risk factors for mortalities with child and maternal undernutrition are unsafe water, poor sanitation and lack of handwashing (Forouzanfar *et al.*, 2015). Interventions to improve water quality were associated with a 42% relative reduction in diarrhoea, and sanitation interventions decreased diarrhoea morbidity by 37%. Hygiene interventions reduced the incidence of diarrhoea by 31% (Bhutta, Das & Walker *et al.*, 2013). A meta-analysis indicated that solar disinfection of water, provision of soap and the improvement of water quality improved height-for-age Z-scores in children younger than 5 years (Dangour *et al.*, 2013; De Onis *et al.*, 2013).

2.7.2.3 Integrated nutrition and early child development interventions

Recently, the focus has been on scaling up interventions to integrate nutrition and early development (Black & Dewey, 2014; Sabanathan *et al.*, 2015). A meta-analysis of 30 early childhood programmes reported that better cognition and behaviour can be met when educational and mixed (stimulation/care with nutrition) programmes were applied versus nutrition-only programmes (Nores & Barnett, 2010). Therefore, integrated interventions have the potential to strengthen both the nutritional status and development of infants and children

(Walker *et al.*, 2007; Black, *et al.*, 2008; Walker *et al.*, 2011; Ruel & Alderman, 2013). Evaluations of integrated programmes (health and nutrition services) were done on children from birth to 8 years in one of the poorest countries of the world, namely Bangladesh, and it was found that integrated interventions were effective (Black & Dewey, 2014). Programmes based on child development and nutrition in Malawi, Angola and South Sudan also proved to be successful (Perry *et al.*, 2014).

In India and Pakistan, a sustainable programme incorporating nutrition and games (SPRING) was developed to provide opportunities and encouragement for the caregivers. Caring, feeding and playing were integrated by using caregiver beliefs and practices to develop a culturally appropriate intervention (Lingam *et al.*, 2014). Grow Smart was another integrated micronutrient supplementation and early learning intervention conducted in India, which focused on infants and pre-school age children. This study indicated the need to collaborate with stakeholders (including village leaders and administrators), the national programme that provides early child nutrition and pre-school education in the villages (Fernandez-Rao *et al.*, 2014).

2.7.2.4 Health and planning services and nutrition education

Community-based interventions are delivered by health care personnel or individuals in local homes, villages or communities. Some interventions include the provision of antenatal, natal and postnatal care, breastfeeding counselling and the development of skills (Bhutta, Das & Rizvi, 2013). It is important that education must focus on uneducated adolescents who are more likely to get pregnant. Complications from pregnancy and childbirths for girls aged 15-19 years account for an estimate 70 000 deaths annually. Educated mothers are more likely to immunise their children and to be better informed about nutrition (UNICEF, 2008). Maternal education and income growth are some of the main determinants in lowering child mortality worldwide (Wang *et al.*, 2014). The exclusive breastfeeding rate can be increased by 90% in infants of age 1–5 months by combined individual and group counselling, which also has the potential to increase the breastfeeding rate (Bhutta, Das & Rizvi, 2013). A systematic review shows that educational or counselling interventions increased the exclusive breastfeeding rate by 43% at day one, by 30% up to 1 month and by 90% from 1–5 months, with significant reductions in rates of no breastfeeding in all three time intervals (Haroon *et al.*, 2013). Breast milk intake can also be increased by human donor milk. A systematic review done on the effectiveness of donor breast milk indicated that preterm infants can be protected from necrotising enterocolitis by consuming donor breast milk (Arslanoglu *et al.*, 2013). However,

there is a need for scientific evidence based on the health and economic benefits of donor breast milk banks as concerns are raised on the risk of possible transmission of HIV, cytomegalovirus and Creutzfeldt-Jakob disease (Modi, 2006).

Complementary feeding promotion in children 6–24 months of age showed significant effects on the increase in height gain in food secure populations and significant effects on the reduction of stunting in food insecure populations (Lassi *et al.*, 2013).

2.7.3 Nutrition-specific interventions

Nutrition-specific interventions and programmes refer to interventions or programmes that address the immediate determinants of foetal and child nutrition and development (Ruel & Alderman, 2013). Optimum breastfeeding and complementary feeding practices with dietary diversification are already discussed in sections 2.1 and 2.2. Some aspects of disease prevention and management will be discussed followed by micronutrient supplementation.

2.7.3.1 Disease prevention and management

A report by UNICEF (2004) stated that, in Africa, malaria caused approximately 20% of all child deaths per year and malaria during pregnancy was the mayor cause of low birth weight in sub-Saharan Africa (UNICEF, 2004). However, according to a recent publication of Lancet (2014) on the Global Burden of Disease, Injuries and Risk Factor Study 2013, a low of 0% for neonatal disorders and neglected tropical diseases and malaria were recorded. Some diseases reported accounted for deaths in 2013 ranged from diarrhoea, lower respiratory diseases (62.4%), HIV/AIDS and TB (59.8%), nutritional deficiencies (56.8%) and communicable maternal, neonatal and nutritional diseases (44.1%). The global under-5 mortality rate as reported in 2013, ranges from 31.9% in the neonatal period, 9.7% in late neonatal period, 29.4% in the post-neonatal period and 28.9% between the ages of 1–4 years. As more than 60% of the Global Burden of Disease are caused by diarrhoea and pneumonia (Forouzanfar *et al.*, 2015), these 2 adverse events were also reported to be the leading cause of under-5 child deaths (29%) worldwide and is concentrated in poor regions in sub-Saharan Africa and South Asia (UNICEF, 2012). The incidence of diarrhoea and pneumonia can decrease in the event of optimal breastfeeding practices. An increase of 165% in diarrhoea incidence was noted in non-breastfed infants aged 0–5 months and a 32% increase was noted in 6–11 month-old infants (Lamberti *et al.*, 2011). Diarrhoea is most commonly caused by the Rotavirus and pneumonia by *H influenzae* type b, which causes pneumococcal pneumonia. New vaccines developed showed to be effective to treat these viruses (Bhutta, Das & Walker

et al., 2013). The use of an oral rehydration solution also showed to be successful in treating diarrhoea (Munos *et al.*, 2010). Sub-Saharan Africa is also heavily affected by HIV and accounts for almost 70% of the global total of new HIV infections. In low- and middle-income countries, paediatric coverage of antiretroviral therapy is low. Less than one in four children living with HIV had access to ART in 2013 (WHO, 2015a). The prevalence of HIV is found to be the highest in South African women and the second highest in South African children younger than 5 years (UNICEF, 2014). An intervention cohort study conducted in KwaZulu-Natal indicated that the mortality rate of 3-month-old infants born to HIV-infected mothers who initiated exclusive breastfeeding was 6.1%, versus 15.1% in infants given replacement feeds from birth. The study further showed that breastfed infants receiving solids before the age of 6 months were 11 times more likely to become infected compared to exclusively breastfed infants (Coovadia *et al.*, 2007). The number of deaths of HIV-infected children younger than 5 years in Botswana increased by about 20 times in a year's time by 2006 and almost all of these infants were not breastfed (Coovadia *et al.*, 2007). The estimated risk of transmission in Zimbabwean infants who were exclusively breastfed for at least 3 months was 1.3% compared with 3.9% in infants receiving mixed feeding (Iliff *et al.*, 2005). Promotion in the Era of HIV (PROMISE-EBF) reported that individual breastfeeding peer counselling could be used to effectively increase the exclusive breastfeeding prevalence in sub-Saharan Africa (Tylleskär *et al.*, 2011). Tuberculosis is also one of the top ten causes of child mortality worldwide and the prevalence of early TB infection may be found in infants born to HIV-1 infected mothers (Cranmer *et al.*, 2014). The childhood TB roadmap was launched in 2013 to assist in the elimination of childhood TB (WHO, 2013b).

Cholera infection has the potential to kill rapidly in outbreak settings. Vaccines are shown to successfully reduce the risk of cholera infection by 52% (Ali *et al.*, 2005; Longini *et al.*, 2007)

2.7.3.2 Single- and multiple nutrient interventions

The WHO acknowledges that the provision of micronutrients, especially during the first thousand days, have a significant impact on reducing death and disease. Scaling-up refers to deliberate efforts to increase the impact of successfully tested vitamin and mineral interventions for the benefit of more people and to foster policy and programme development. The following micronutrient supplementation for infants and young children is guided by the WHO: 6-monthly large dose vitamin A supplementation for children younger than 5 years; daily iron supplementation for low-birth weight infants and young children, and iron and folic acid supplementation for children living in malaria-endemic settings (Pena-Rosas *et al.*, 2012)

Intermittent iron supplementation can reduce the risk of anaemia by 49% and iron deficiency by 76% in children younger than 24 months. There is a lack of convincing evidence to indicate that iron treatment is associated with mental development in children younger than 27 months (Bhutta, Das & Rizvi, 2013).

Vitamin A supplementation has the potential to reduce the risk of mortality at the age of 6 months by 14% and in children aged 6–59 months by 24%. Vitamin A supplementation can also reduce the diarrhoea-related mortality rate in children aged 6–59 months by 28% (Haider & Bhutta, 2011).

Results of a meta-analysis showed that preventative zinc supplementation may increase linear growth and weight gain in infants who are at risk of zinc deficiency (Imdad & Bhutta, 2011). However, a systematic review by Yakoob *et al.* (2011) showed that preventative zinc supplementation did not significantly reduce the all-cause mortality rate (Yakoob *et al.*, 2011). Findings on the impact of zinc supplementation on infant development and behaviour have not been consistent (Black *et al.*, 2004).

A systematic review showed that early intake of long-chain polyunsaturated omega-3 fatty acids (n-3 LC-PUFAs) through breast milk or fortified foods enhanced child development and visual acuity (Campoy *et al.*, 2012) and recent studies indicated that supplementation with n-3 LC-PUFAs contributes to better cognitive development later in life (De Jong *et al.*, 2012; Colombo *et al.*, 2013; Koletzko *et al.*, 2014). Fatty acid intervention can also improve immune function such as inflammatory disorders in children living in low-income countries (Prentice & van der Merwe, 2011).

A meta-analysis of single and multiple nutrient interventions done on children younger than 5 years concluded that there were no significant benefits when iron and vitamin A are used as single-micronutrient interventions, whereas multiple micronutrient interventions improved linear growth (Ramakrishnan *et al.*, 2009). A systematic review done on multiple micronutrient (MMN) supplementation concluded that MMN supplementation increased length and weight as well as cognitive development in children aged 6 months to 16 years (Bhutta, Das & Rizvi, 2013). A systematic review of randomised controlled trials done on children aged 6 months to 11 years indicated that MMN supplementation reduced anaemia, iron deficiency anaemia and retinol deficiency and improved haemoglobin concentrations. However, MMN supplementation may lead to an increase in diarrhoea incidents and showed no significant effects on serum ferritin, zinc deficiency, stunting, wasting, underweight, fever height-for-age, weight-for-age and weight-for-height z-scores (Bhutta, Das & Rizvi, 2013).

Infants from rural Bangladesh showed better motor development when supplemented weekly with both iron (20 mg) and zinc (20 mg). The study also indicated no additional benefit in terms of motor development was observed when the infants received a micronutrient supplement containing 16 vitamins and minerals (Black *et al.*, 2004). Recently, Colombo *et al.* (2014) found that Peruvian infants supplemented with zinc (10 mg/d), iron (10 mg/day) and copper (0.5 mg/day) for 12 months showed normative neurodevelopment when consuming a diet low in zinc (Colombo *et al.*, 2014). A meta-analysis and systematic review done by Dewey *et al.* (2009) concluded that micronutrient supplementation in addition to the added energy (including fat and protein) of complementary food has a significant effect on increased weight and height (Dewey *et al.*, 2009).

2.7.3.3 Fortification

The WHO guided fortification in terms of wheat flour fortification with micronutrients, corn/maize flour fortification with micronutrients, the fortification of edible oils and fats with micronutrients, sugar fortification with micronutrients, salt iodisation and the fortification of condiments and seasonings (Pena-Rosas *et al.*, 2012). Globally, the Codex Alimentarius of the Food and Agriculture Organization (FAO) and the WHO established general principles for the addition of vitamins and minerals to food and today more than 67 countries fortify certain staple foods (de Lourdes Samaniego-Vaesken *et al.*, 2012). Folic acid fortification of wheat or maize is also one of the most successful public health initiatives in the past 50–75 years as shown in countries like the US where studies showed a decrease of 19–32% in the prevalence of neural tube defects (Honein *et al.*, 2001; Williams *et al.*, 2002; Mathews 2007; Boulet *et al.*, 2008) as well as Canada, Costa Rica, Chile, Argentina and Brazil and South Africa, which reported a decline of 19–55% (Persad *et al.*, 2002; Ray *et al.*, 2002; Chen & Rivera 2004; Hertrampf & Cortés 2004; De Wals *et al.*, 2007; Sayed *et al.*, 2008; López-Camelo *et al.*, 2010),

Das *et al.* (2013) concluded from a systematic review on micronutrient fortification that fortification has a significant impact on increasing serum micronutrient concentrations and haemoglobin concentrations for children when food was fortified with vitamin A, iron and multiple micronutrients (Das *et al.*, 2013). A systematic review done on all age groups by Gera *et al.* (2012) stated that iron-fortified foods may increase haemoglobin and serum ferritin, but there is a lack of evidence of an effect on infections, anthropometric measures or motor development (Gera *et al.*, 2012).

The effects of micronutrient fortified milk and cereal food for infants and children younger than 5 years were reviewed by Eichler *et al.* (2012) and it was concluded that the fortification of milk and cereal products may be effective for reducing anaemia in this age group in developing countries, but there is still a gap in the evidence for functional health outcomes (Eichler *et al.*, 2012). Full-fat soybean flour mixed with micronutrients were developed for home fortification in Vietnam and China (Dewey *et al.*, 2009)

In South Africa, the legislation on fortifying all white and brown bread flour and maize meal with vitamins A, B1, B2, B6, niacin and folic acid and minerals like iron and zinc came into effect in 2003 (DOH & UNICEF, 2007). However, the amount of staple food eaten by children in the age group 6–12 months may not be sufficient to contribute to a significant increase in micronutrient intake (Dewey *et al.*, 2009). A fortified maize-meal porridge, developed specifically for babies, was shown to reduce anaemia and improve iron status in a randomised controlled trial in South Africa (Faber *et al.*, 2005).

Biofortification is a relatively new approach for combatting micronutrient deficiencies as a public health problem (Unnevehr *et al.*, 2007). A study done in Uganda by Hotz *et al.* (2012) indicated that β -carotene-rich orange sweet potatoes increased the vitamin A intake of women and children aged 3–5 years (Hotz *et al.*, 2012). Genetic engineering has led to the development of Golden Rice and a study conducted in Boston revealed that the β -carotene derived from Golden Rice can be effectively converted to vitamin A in humans (Tang *et al.*, 2009).

2.7.3.4 Home fortification

Home fortification of complementary foods has been proposed as an innovative intervention for improving micronutrient intake in children younger than 2 years. Products that can be used for home fortification include nutritabs, micronutrient powders and lipid-based nutritional supplements (LNSs) (Gibson, 2011). Dewey concluded from a systematic review done in 2009 that micronutrient powders is most likely to be used for home fortification. Micronutrient powders can be sprinkled onto a portion of complementary food before consumption (Dewey *et al.*, 2009). De-Regil *et al.* (2013) reported from a systematic review done on home fortification of foods with multiple micronutrient powders (MNP) for health and nutrition in children under 2 years of age that MNP can be effectively used to reduce anaemia and iron deficiency, but evidence is lacking to indicate a decrease of other vitamin and mineral deficiencies as well as data on child survival strategy and developmental outcomes (De-Regil *et al.*, 2013).

Crushable/chewable tablets are multiple micronutrient tablets and dissolve in a small amount of liquid without difficulty. These tablets can also be crushed and added to complementary food (Dewey *et al.*, 2009). The International Research on Infant Supplementation (IRIS) trial examined the efficacy of a multi-micronutrient supplementation in the form of a food-like chewable tablet on infants 6–12 months old in 4 countries (South Africa, Peru, Vietnam, Indonesia). Results indicated that daily use of the tablets decreased weight growth faltering in all 4 countries and reduced the prevalence of anaemia, iron deficiency and zinc deficiency (Smuts, Lombard *et al.*, 2005).

Lipid-based nutritional supplements are produced in a food base and provide energy, protein and EFA required for growth. Vitamins found in LNS are protected from oxidation because it is embedded in fat and therefore prolongs the shelf life of the products. The ration size for infants is designed to avoid displacement of breast milk and also to allow for dietary diversity. Most LNS products intended for home fortification of local diets have a daily ration size of 20 g (Arimond *et al.*, 2013)

Ready-to-use therapeutic food (RUTF) or large quantity LNS are used for the treatment of severe acute malnutrition (SAM) and is designed to provide 80–280 g/day (100–1500 kcal/day) and 100% of energy needed from food other than breast milk (Arimond *et al.*, 2013). Ready-to-use therapeutic food or medium quantity LNS are used for the treatment of moderate acute malnutrition (MAM) and is designed to provide 45–90 g/day (250–500 kcal/day) and to provide 50–100% of energy needed from food other than breast milk (Arimond *et al.*, 2013). Steenkamp and Lategan (2013) conducted a small study in a clinical setting in South Africa to determine the acceptability of RUTF for children between the ages of 12 and 60 months, 18% were HIV-infected and 39% presented with moderate acute malnutrition. The study concluded that the RUTF were accepted by more than 80% of the children, but questioned the recommended portion sizes of 20 g, specifically for young children with poor appetites (Steenkamp & Lategan, 2013).

Small-quantity lipid-based nutrient supplements (SQ-LNS) are used to prevent undernutrition and promote growth and development. It provides <50% of the energy needed from food other than breast milk. The typical ration is 20 g/day (110 kcal/day) (Arimond *et al.*, 2013).

SQ-LNS are easy to use and require minimum change in dietary behaviours (Arimond *et al.*, 2013; Siega-Riz *et al.*, 2014). Presently, many organisations and companies have shown interest in the development of SQ-LNS and their potential use in a variety of cultural and geographical settings (Arimond *et al.*, 2013). Recently, a study conducted in the Honduras

indicated that SQ-LNS improved micronutrient status in non-malnourished children aged 6–18 months (Siega-Riz *et al.*, 2014). Iannotti *et al.* (2014) showed that daily consumption of SQ-LNS resulted in a significant increase in linear growth in healthy infants aged 6–11 months in Haiti (Iannotti *et al.*, 2014). However, Huybregts *et al.* (2012) supplemented children aged 6–36 months in central Africa with a daily dose of 46 g LNS for 4 months and results showed that adding LNS to household food did not reduce the prevalence of wasting (Huybregts *et al.*, 2012). Pulakka *et al.* (2014) found that daily doses of SQ-LNS (10–40 g) did not increase the physical activity of Malawian toddlers (Pulakka *et al.*, 2014).

It is important to assess the sensory acceptability of SQ-LNS pastes within the given socio-cultural context to explore their potential for usage (Berti *et al.*, 2014). Moreover, if the sensory attributes are perceived as acceptable, but the product is inconvenient to use, the product will not be sustainable (Pelto *et al.*, 2013). Acceptability trials on SQ-LNS were conducted in Ghana, Guatemala, Burkino Faso on infants aged 6–18 months and it found that the pastes tested were acceptable. All 4 studies used a kind of Nutributter® with different combinations of peanuts and soy (Adu-Afarwuah *et al.*, 2011; Hess *et al.*, 2011; Matias *et al.*, 2011; Phuka *et al.*, 2011). To my knowledge, there is no literature available on the acceptability of soy-based SQ-LNS for infants aged 6–12 months in the South African context.

2.7.4 Key nutrition interventions in South Africa

In South Africa, various delivery modes of nutrition interventions are in place as indicated in Table 6. Especially in the rural areas of South Africa, the Primary Health Care (PHC) clinic services are an imperative delivery platform to improve infant and young child feeding. PHC need support from the government to provide the resources needed, like growth monitoring, deworming, vitamin A and therapeutic zinc supplementation. Preventative services like VAS, deworming and growth monitoring linked to counselling on IYCF are needed to ensure healthy growth and development. Mothers are supported by PHC services and visited at home by the community health workers (CHW) to be informed about practicing early initiation and exclusive breastfeeding. Early Childhood Development (ECD) facilitators are used to ensure healthy development of the infant and young child by combining education of development with a healthy diet (DOH, 2013b). Recently, the Department of Social Development launched a draft national early childhood development policy of the Republic of South Africa. This policy entails the commitment of the government to ensure universal availability of, and equitable access to, a comprehensive package of quality early childhood development services for all young

children from conception until they enter formal schooling (Grade R) or until the age of 8 years (in the case of children with developmental difficulties or disabilities) (DSD, 2015).

Table 6. Comprehensive summary of key nutritional interventions in South Africa (DOH, 2013b)

Intervention	Target population	Delivery platform	Existing policies, frameworks, guidelines
<i>Exclusive breastfeeding promotion</i>	Pregnant women and families of children younger than 6 months	Community nutrition programmes ANC (BANC) through community outreach, PHC clinic services and hospital services Communication campaigns School curricula for grade 10 to 12	PMTCT 2010 clinical guidelines BANC; BFHI; IYCF policy IMCI Draft regulations on marketing of infant foods Nutrition and HIV School Health Services policy Health Promoting Schools initiative
<i>Improved complementary feeding with continued breastfeeding and targeted supplementary feeding where needed</i>	Pregnant women and families of children younger than 24 months; populations with high percentage of children aged 6–23 months with WAZ <2 (underweight)	Community nutrition programmes; outreach (CHW) and PHC services; communication campaigns	IYCF, IMCI, GMP
<i>Healthy eating for optimal weight management during pregnancy and lactation</i>	Pregnant women and breastfeeding mothers	Community nutrition programmes; outreach (CHW) and PHC services; communication campaigns, pre-schools and communities	BANC, FBDG, School Health Services policy, Health promoting Schools initiative
<i>Implementation of evidence-based interventions for detection of malnutrition during pregnancy</i>	Pregnant women	ANC – at PHC clinic and hospital services	2007 Guidelines for Maternity Care in SA
<i>Improved hygiene practices, including hand washing</i>	Caregivers and families	Community outreach, PHC, hospital services and communication campaigns	IMCI, IYCF, Guidelines, Health promoting schools initiatives

Intervention	Target population	Delivery platform	Existing policies, frameworks, guidelines
<i>Nutrition education and information on healthy eating and health risks associated with poor diets</i>	Entire population and individuals with chronic conditions	Community outreach, PHC, hospital services and communication campaigns	Guidelines on chronic diseases (hypertension, diabetes) FBDG, Food Guide
<i>Vitamin A supplementation</i>	Twice yearly doses for children 6–59 months of age	Hospital services, Child health weeks. Routine PHC services and outreach (CHW)	IMCI, GMP, VAS protocol, 2007 Guidelines for Maternity care in SA
<i>Therapeutic zinc supplementation</i>	Children aged 6–59 months with diarrhoea	Diarrhoea treatment – PHC, hospitals	IMCI Guidelines
<i>Fortification of staple foods</i>	Entire population	Market-based strategy	Food fortification regulations
<i>Salt iodization</i>	Entire population	Market-based strategy	Salt iodization regulations
<i>Deworming (situational)</i>	Children 6–59 months of age	Child Health weeks, routine PHC services and outreach	IMCI
<i>Multiple micronutrient supplements and targeted supplementary feeding to undernourished individuals</i>	Individuals with chronic conditions like HIV and TB	Community nutrition programmes, clinics, PHC and hospitals	Nutrition and HIV and AIDS guidelines
<i>Treatment of severe acute malnutrition</i>	Children 6–59 months of age with <-3 WHZ (severely wasted) (with or without oedema or MUAC <115mm	PHC and district and regional hospitals	WHO 10 steps to manage severe acute malnutrition
<i>Prevention and treatment for moderate undernutrition</i>	Children 6–59 months of age with <-2 WHZ	Community nutrition programmes, PHC services	WHO CTC Guidelines

ANC: Antenatal care; BANC: Basic antenatal care; PHC: Primary health care; BFHI: Baby friendly hospital initiative; IYCF: Infant and young child feeding; IMCI: Integrated management of childhood illness; CHW: Community health workers; GMP: Growth monitoring and promotion; FBDG: Food-based dietary guidelines; VAS: Vitamin A supplementation; CTC: Community therapeutic care.

The effectiveness of these key interventions are, however, questioned. A report presenting the national evaluation on nutrition interventions for children from conception to the age of 5 years, reported that a well-defined-nutrition plan is needed across all sectors that invest in nutrition by specifically focussing on infants and children under the age of 5 years. Stronger coordination and monitoring is needed during the implementation of interventions by government departments. There should be more focus on nutrition promotion, exclusive breastfeeding, complementary feeding, dietary diversity and hygiene education where food security and agriculture (like food gardens) should be integrated with the nutrition in a substantive way (DOH *et al.*, 2014).

2.8 Conclusion

The importance of nutrition to improve child survival is critical to accelerate progress towards the achievement of the post-2015 SDGs. Globally, infant feeding practices are poorly practiced and not according to the WHO recommendations for children 0–24 months (Caroli *et al.*, 2012; Zhou *et al.*, 2012; Du Plessis *et al.*, 2013; IFPRI, 2014). This despite the fact that early initiation of breastfeeding decreases the risk of mortality by 44% (Debes *et al.*, 2013) and the lives of 1.5 million children younger than 5 years can be saved by exclusive breastfeeding and optimal complementary feeding practices with continuing breastfeeding for up to 2 years or beyond (WHO, 2013a). Feeding practices have an impact on the nutritional status of infant and young children and also an indirectly impact on public health indicators like child survival, health and development (WHO, 2008). As described, the first thousand days are a critical period of vulnerability to infections and diseases, where especially iron, zinc and vitamin A deficiency lead to infections, poor immune system and impaired growth and development. Therefore, the effect of physical growth rates and stunting on cognitive abilities and psychological functioning in children younger than 5 years is a global research priority (Black *et al.*, 2013) and the focus is on scaling up interventions to integrate nutrition and early development (Black & Dewey, 2014; Sabanathan *et al.*, 2015). International and national fortification programmes are in place. Unfortunately, infants starting with complementary foods consume a small amount of food at a time and nutrient density is important for healthy growth and development (Dewey, 2013). Animal-source foods (meat, fish, eggs and dairy products) are often not affordable and are not frequently consumed in poor households. Therefore, the use of SQ-LNS to be used at home for the provision of macro- and micronutrients currently receives the interest of many organisations and companies (Arimond *et al.*, 2013). The acceptability and efficacy of SQ-LNS has not been tested in a South African environment.

Provincial data in South Africa reflected that there is a lack of large sample infant studies on feeding practices and growth. Available studies indicated that a high percentage of South African infants receive complementary foods and liquids already at 4 months of age (HSRC, 2013). Against the background of poor infant feeding practices in South Africa and a lack of South African literature available on; 2) feeding practices in the age group 0–6 months associated with iron 2) growth rates of infants aged 6 months; 3) the association of psychomotor development and nutrient status of infants 0–6 months; and 4) the acceptability of SQ-LNS for infants aged 6–12 months, this study is valuable by providing evidence on feeding practices, nutritional status and psychomotor development of 6-month-old South African infants.

2.9 References

- Abubakar, A., Van De Vijver, F.J., Mithwani, S., Obiero, E., Lewa, N., Kenga, S., *et al.* 2007. Assessing developmental outcomes in children from Kilifi, Kenya, following prophylaxis for seizures in cerebral malaria. *Journal of health psychology*, 12(3):417-430.
- Abubakar, A., Holding, P., Van Baar, A., Newton, C. & van de Vijver, F.J. 2008. Monitoring psychomotor development in a resource limited setting: an evaluation of the Kilifi Developmental Inventory. *Annals of tropical paediatrics: international child health*, 28(3):217-226.
- Aburto, N., Abudou, M., Candeias, V. & Wu, T. 2014. Effect and safety of salt iodization to prevent iodine deficiency disorders: a systematic review with meta-analyses: WHO eLibrary of Evidence for Nutrition Actions (eLENA). Geneva: World Health Organization.
- Adu-Afarwuah, S., Lartey, A., Zeilani, M. & Dewey, K.G. 2011. Acceptability of lipid-based nutrient supplements (LNS) among Ghanaian infants and pregnant or lactating women. *Maternal and child nutrition*, 7(4):344-356.
- Agostoni, C. 2008. Role of long-chain polyunsaturated fatty acids in the first year of life. *Journal of pediatric gastroenterology and nutrition*, 47:S41-S44.
- Ahmed, H.M., Mitchell, M. & Hedt, B. 2010. National implementation of Integrated Management of Childhood Illness (IMCI): policy constraints and strategies. *Health policy*, 96(2):128-133.
- Ali, M., Emch, M., von Seidlein, L., Yunus, M., Sack, D.A., Rao, M., *et al.* 2005. Herd immunity conferred by killed oral cholera vaccines in Bangladesh: a reanalysis. *The lancet*, 366(9479):44-49.
- Anderson, J.W., Johnstone, B.M. & Remley, D.T. 1999. Breast-feeding and cognitive development: a meta-analysis. *The American journal of clinical nutrition*, 70(4):525-535.
- Anjos, T., Altmäe, S., Emmett, P., Tiemeier, H., Closa-Monasterolo, R., Luque, V., *et al.* 2013. Nutrition and neurodevelopment in children: focus on NUTRIMENTHE project. *European journal of nutrition*, 52(8):1825-1842.
- Arimond, M. & Ruel, M.T. 2004. Dietary diversity is associated with child nutritional status: evidence from 11 demographic and health surveys. *The journal of nutrition*, 134(10):2579-2585.

- Arimond, M., Zeilani, M., Jungjohann, S., Brown, K.H., Ashorn, P., Allen, L.H., *et al.* 2013. Considerations in developing lipid-based nutrient supplements for prevention of undernutrition: experience from the International Lipid-Based Nutrient Supplements (iLiNS) Project. *Maternal and child nutrition*, doi: 10.1111/mcn.12049.
- Arslanoglu, S., Moro, G.E., Bellù, R., Turoli, D., De Nisi, G., Tonetto, P., *et al.* 2013. Presence of human milk bank is associated with elevated rate of exclusive breastfeeding in VLBW infants. *Journal of perinatal medicine*, 41(2):129-131.
- Arts, M., Geelhoed, D., De Schacht, C., Prosser, W., Alons, C. & Pedro, A. 2010. Knowledge, beliefs, and practices regarding exclusive breastfeeding of infants younger than 6 months in Mozambique: a qualitative study. *Journal of human lactation*, doi: 0890334410390039.
- Aslinia, F., Mazza, J.J. & Yale, S.H. 2006. Megaloblastic anemia and other causes of macrocytosis. *Clinical medicine & research*, 4(3):236-241.
- Bahl, R., Frost, C., Kirkwood, B.R., Edmond, K., Martines, J., Bhandari, N., *et al.* 2005. Infant feeding patterns and risks of death and hospitalization in the first half of infancy: multicentre cohort study. *Bulletin of the World Health Organization*, 83(6):418-426.
- Baker, R.D. & Greer, F.R. 2010. Diagnosis and prevention of iron deficiency and iron-deficiency anemia in infants and young children (0–3 years of age). *Pediatrics*, 126(5):1040-1050.
- Ballard, O. & Morrow, A.L. 2013. Human milk composition: nutrients and bioactive factors. *Pediatric clinics of North America*, 60(1):49-74.
- Bath, S.C., Steer, C.D., Golding, J., Emmett, P. & Rayman, M.P. 2013. Effect of inadequate iodine status in UK pregnant women on cognitive outcomes in their children: results from the Avon Longitudinal Study of Parents and Children (ALSPAC). *The lancet*, 382(9889):331-337.
- Beard, J.L. 2001. Iron biology in immune function, muscle metabolism and neuronal functioning. *The journal of nutrition*, 131(2):568S-580S.
- Bhatnagar, S. & Taneja, S. 2001. Zinc and cognitive development. *British journal of nutrition*, 85(2):S139-S145.
- Benoist, B.D., McLean, E., Egll, I. & Cogswell, M. 2008. Worldwide prevalence of anaemia 1993-2005: WHO global database on anaemia. Geneva, Switzerland: World Health Organization.

- Bentley, M.E., Johnson, S.L., Wasser, H., Creed-Kanashiro, H., Shroff, M., Fernandez Rao, S., *et al.* 2014. Formative research methods for designing culturally appropriate, integrated child nutrition and development interventions: an overview. *Annals of the New York Academy of Sciences*, 1308(1):54-67.
- Berti, C., Faber, M. & Smuts, C.M. 2014. Prevention and control of micronutrient deficiencies in developing countries: current perspectives. *Nutrition & dietary supplements*, 6:41-57.
- Bhutta, Z.A., Ahmed, T. Black, R.E., Cousens, S., Dewey, K., Giugliani, E., *et al.* 2008. What works? Interventions for maternal and child undernutrition and survival. *The lancet*, 371(9610):417-440.
- Bhutta, Z.A., Das, J.K., Rizvi, A., Gaffey, M.F., Walker, N., Horton, S., *et al.* 2013. Evidence-based interventions for improvement of maternal and child nutrition: what can be done and at what cost? *The lancet*, 382(9890):452-477.
- Bhutta, Z.A., Das, J.K., Walker, N., Rizvi, A., Campbell, H., Rudan, I., *et al.* 2013. Interventions to address deaths from childhood pneumonia and diarrhoea equitably: what works and at what cost? *The lancet*, 381(9875):1417-1429.
- Biasini, F.J., De Jong, D., Ryan, S., Thorsten, V., Bann, C., Bellad, R., *et al.* 2015. Development of a 12 month screener based on items from the Bayley II Scales of Infant Development for use in Low Middle Income countries. *Early human development*, 91(4):253-258.
- Black, M.M. 2003. Micronutrient deficiencies and cognitive functioning. *The journal of nutrition*, 133(11):3927S-3931S.
- Black, M.M. 2008. Effects of vitamin B12 and folate deficiency on brain development in children. *Food and nutrition bulletin*, 29(2):S126.
- Black, M.M. & Aboud, F.E. 2011. Responsive feeding is embedded in a theoretical framework of responsive parenting. *The journal of nutrition*, 141(3):490-494.
- Black, M.M. & Dewey, K.G. 2014. Promoting equity through integrated early child development and nutrition interventions. *Annals of the New York Academy of Sciences*, 1308(1):1-10.
- Black, M.M., Baqui, A.H., Zaman, K., Persson, L.A., El Arifeen, S., Le, K., *et al.* 2004. Iron and zinc supplementation promote motor development and exploratory behavior among Bangladeshi infants. *The American journal of clinical nutrition*, 80(4):903-910.

- Black, M.M., Walker, S.P., Wachs, T.D., Ulkuer, N., Gardner, J.M., Grantham-McGregor, S., *et al.* 2008. Policies to reduce undernutrition include child development. *The lancet*, 371(9611):454-455.
- Black, R.E., Allen, L.H., Bhutta, Z.A., Caulfield, L.E., De Onis, M., Ezzati, M., *et al.* 2008. Maternal and child undernutrition: global and regional exposures and health consequences. *The lancet*, 371(9608):243-260.
- Black, R.E., Alderman, H., Bhutta, Z.A., Gillespie, S., Haddad, L., Horton, S., *et al.* 2013. Maternal and child nutrition: building momentum for impact. *The lancet*, 382(9890):372-375.
- Black, R.E., Victora, C.G., Walker, S.P., Bhutta, Z.A., Christian, P., De Onis, M., *et al.* 2013. Maternal and child undernutrition and overweight in low-income and middle-income countries. *The lancet*, 382(9890):427-451.
- Bland, R., Rollins, N.C., Coutsooudis, A. & Coovadia, H.M. 2002. Breastfeeding practices in an area of high HIV prevalence in rural South Africa. *Acta paediatrica*, 91(6):704-711.
- Bloem, M. 2013. Preventing, stunting: why it matters, what it takes. (*In* Eggersdorfer, M., *ed.* The road to good nutrition. Basel, Switzerland: Karger. p. 13-24).
- Bokor, S., Koletzco, B. & Desci, T. 2007. Systematic review for fatty acid composition of human milk from mothers of preterm compared to full-term infants. *Annals of nutrition and metabolism*, 51:550-556.
- Boulet, S.L., Yang, Q., Mai, C., Kirby, R.S., Collins, J.S., Robbins, J.M., *et al.* 2008. Trends in the postfortification prevalence of spina bifida and anencephaly in the United States. *Birth defects research part A: clinical and molecular teratology*, 82(7):527-532.
- Brenna, J.T., Varamini, B., Jensen, R.G., Diersen-Schade, D.A., Boettcher, J.A. & Arterburn, L.M. 2007. Docosahexaenoic and arachidonic acid concentrations in human breastmilk worldwide. *The American journal of clinical nutrition*, 85(6):1457-1464.
- Brown, K.H., Wuehler, S.E. & Peerson, J.M. 2001. The importance of zinc in human nutrition and estimation of the global prevalence of zinc deficiency. *Food and nutrition bulletin*, 22(2):113-125.
- Cadegan, M., English, R., Pillay, Y. & Barron, P. 2012. Strategic plan for maternal, new-born, child and women's health (MNCWH) and nutrition in South Africa: 2012-2016. Health System Trust: Durban.

- Campoy, C., Escolano-Margarit, M., Anjos, T., Szajewska, H. & Uauy, R. 2012. Omega 3 fatty acids on child growth, visual acuity and neurodevelopment. *British journal of nutrition*, 107(S2):S85-S106.
- Caroli, M., Mele, R., Tomaselli, M., Cammisa, M., Longo, F. & Attolini, E. 2012. Complementary feeding patterns in Europe with a special focus on Italy. *Nutrition, metabolism and cardiovascular diseases*, 22(10):813-818.
- Carruth, B.R., Ziegler, P.J., Gordon, A. & Hendricks, K. 2004. Developmental milestones and self-feeding behaviors in infants and toddlers. *Journal of the American Dietetic Association*, 104:51-56.
- Chan, M. 2014. Investing in early child development: an imperative for sustainable development. *Annals of the New York Academy of Sciences*, 1308(1):vii-viii.
- Chaparro, C.M., Neufeld, L.M., Tena Alavez, G., Eguia-Líz Cedillo, R. & Dewey, K.G. 2006. Effect of timing of umbilical cord clamping on iron status in Mexican infants: a randomised controlled trial. *The lancet*, 367(9527):1997-2004.
- Chen, L.T. & Rivera, M.A. 2004. The Costa Rican experience: reduction of neural tube defects following food fortification programmes. *Nutrition reviews*, 62(suppl 1):S40-S43.
- Cherop, E.C., Etyyang, G.A. & Mbagaya, G.M. 2010. Mixed feeding among infants aged 0-6 months in an urban setting of Eldoret, Kenya. *Journal of food, agriculture & environment*, 8(1):59.
- Colombo, J., Carlson, S.E., Cheatham, C.L., Shaddy, D.J., Kerling, E.H., Thodosoff, J.M., et al. 2013. Long-term effects of LCPUFA supplementation on childhood cognitive outcomes. *The American journal of clinical nutrition*, 98(2):403-412.
- Colombo, J., Zavaleta, N., Kannass, K.N., Lazarte, F., Albornoz, C., Kapa, L.L., et al. 2014. Zinc supplementation sustained normative neurodevelopment in a randomized, controlled trial of Peruvian infants aged 6–18 months. *The journal of nutrition*, jn. 113.189365.
- Connor, W.E. 2000. Importance of n-3 fatty acids in health and disease. *The American journal of clinical nutrition*, 71(1):171S-175S.
- Coovadia, H.M., Rollins, N.C., Bland, R.M., Little, K., Coutsoydis, A., Bennish, M.L., et al. 2007. Mother-to-child transmission of HIV-1 infection during exclusive breastfeeding in the first 6 months of life: an intervention cohort study. *The lancet*, 369(9567):1107-1116.

- Cranmer, L.M., Kanyugo, M., Jonnalagadda, S.R., Lohman-Payne, B., Sorensen, B., Obimbo, E.M., *et al.* 2014. High prevalence of tuberculosis infection in HIV-1 exposed Kenyan infants. *The pediatric infectious disease journal*, 33(4):401.
- Cunnane, S.C., Francescutti, V., Brenna, J.T. & Crawford, M. 2000. Breast-fed infants achieve a higher rate of brain and whole body docosahexaenoate accumulation than formula-fed infants not consuming dietary docosahexaenoate. *Lipids*, 35:105-111.
- Dangour, A.D., Watson, L., Cumming, O., Boisson, S., Che, Y., Velleman, Y., *et al.* 2013. Interventions to improve water quality and supply, sanitation and hygiene practices, and their effects on the nutritional status of children. *Cochrane Database Systematic Reviews*, 8:CD009382.
- De Benoist, B., Darnton-Hill, I., Davidsson, L., Fontaine, O. & Hotz, E. 2007. Conclusions of the joint WHO/UNICEF/IAEA/IZiNCG interagency meeting on zinc status indicators. *Food and nutrition bulletin*, 28(3):S480-S486.
- De Groot R., Palermo T., Handa S., Ragno L.P., Peterman A. 2015. Cash transfers and child nutrition: what we know and what we need to know. Innocenti working paper, No.2015-07. UNICEF Office of Research, Florence.
- De Jong, C., Kikkert, H.K., Fidler, V. & Hadders-Algra, M. 2012. Effects of long-chain polyunsaturated fatty acid supplementation of infant formula on cognition and behaviour at 9 years of age. *Developmental medicine & child neurology*, 54(12):1102-1108.
- De Lourdes Samaniego-Vaesken, M., Alonso-Aperte, E. & Varela-Moreiras, G. 2012. Vitamin food fortification today. *Food & nutrition research*: 56.
- De Onis, M. 2006. Relationship between physical growth and motor development in the WHO Child Growth Standards. *Acta paediatrica*, 95(S450):96-101.
- De Onis, M., Dewey, K.G., Borghi, E., Onyango, A.W., Blössner, M., Daelmans, B., *et al.* 2013. The World Health Organization's global target for reducing childhood stunting by 2025: rationale and proposed actions. *Maternal and child nutrition*, 9(S2):6-26.
- De Wals, P., Tairou, F., Van Allen, M.I., Uh, S.-H., Lowry, R.B., Sibbald, B., *et al.* 2007. Reduction in neural-tube defects after folic acid fortification in Canada. *New England journal of medicine*, 357(2):135-142.

Debes, A.K., Kohli, A., Walker, N., Edmond, K. & Mullany, L.C. 2013. Time to initiation of breastfeeding and neonatal mortality and morbidity: a systematic review. *BMC public health*, 13(3):S19.

Department of Social Development see South Africa

Del Prado, M., Villalpando, S., Elizando, A., Rodriguez, M., Demmelmair, H., Koletzko, B. 2001. Contribution of dietary and newly formed arachidonic acid to human milk lipids in women eating a low fat diet. *American journal of clinical nutrition*, 74:242-247.

De-Regil, L.M., Suchdev, P.S., Vist, G.E., Walleser, S. & Peña-Rosas, J.P. 2013. Home fortification of foods with multiple micronutrient powders for health and nutrition in children under two years of age (review). *Evidence-based child health: a Cochrane review journal*, 8(1):112-201.

Dewey, K.G. 2013. The challenge of meeting nutrient needs of infants and young children during the period of complementary feeding: an evolutionary perspective. *The journal of nutrition*, 143(12):2050-2054.

Dewey, K.G. & Brown, K.H. 2003. Update on technical issues concerning complementary feeding of young children in developing countries and implications for intervention programmes. *Food and nutrition bulletin*, 24(1):5-28.

Dewey, K.G. & Chaparro, C.M. 2007. Session 4: mineral metabolism and body composition iron status of breast-fed infants. *Proceedings of the Nutrition Society*, 66(03):412-422.

Dewey, K.G., Cohen, R.J., Brown, K.H. & Rivera, L.L. 2001. Effects of exclusive breastfeeding for four versus six months on maternal nutritional status and infant motor development: results of two randomized trials in Honduras. *The journal of nutrition*, 131(2):262-267.

Dewey, K.G., Yang, Z. & Boy, E. 2009. Systematic review and meta-analysis of home fortification of complementary foods. *Maternal and child nutrition*, 5(4):283-321.

DOH (Department of Health) see South Africa.

DPME (Department of Performance, Monitoring and Evaluation) see South Africa.

DSD (Department of Social Development) see South Africa

Du Plessis, L.M. 2013. Commitment and capacity for the support of breastfeeding in South Africa: a paediatric food-based dietary guideline. *South African journal of clinical nutrition*, 26(3):S120-S128.

- Du Plessis, L.M., Kruger, H. & Sweet, L. 2013. Complementary feeding: a critical window of opportunity from six months onwards. *South African journal of clinical nutrition*, 26(3):S129-S140.
- Eichler, K., Wieser, S., Rütthemann, I. & Brügger, U. 2012. Effects of micronutrient fortified milk and cereal food for infants and children: a systematic review. *BMC public health*, 12(1):506-518.
- Engelbrechtsen, I.M., Wamani, H., Karamagi, C., Semiyaga, N., Tumwine, J. & Tylleskär, T. 2007. Low adherence to exclusive breastfeeding in Eastern Uganda: a community-based cross-sectional study comparing dietary recall since birth with 24-hour recall. *BMC pediatrics*, 7(1):10.
- Engle-Stone, R., Haskell, M.J., Nankap, M., Ndjebayi, A.O. & Brown, K.H. 2014. Breastmilk retinol and plasma retinol-binding protein concentrations provide similar estimates of vitamin A deficiency prevalence and identify similar risk groups among women in Cameroon but breastmilk retinol underestimates the prevalence of deficiency among young children. *The journal of nutrition*, 144(2):209-217.
- Ertem, I.O., Dogan, D.G., Gok, C.G., Kizilates, S.U., Caliskan, A., Atay, G., *et al.* 2008. A guide for monitoring child development in low-and middle-income countries. *Pediatrics*, 121(3):e581-e589.
- Eussen, S., Alles, M., Uijterschout, L., Brus, F. & van der Horst-Graat, J. 2015. Iron intake and status of children aged 6–36 month in Europe: A systematic review. *Annals of nutrition and metabolism*, doi: 10.1159/000371357.
- Faber, M., Kvalsvig, J.D., Lombard, C.J. & Benadé, A.S. 2005. Effect of a fortified maize-meal porridge on anemia, micronutrient status, and motor development of infants. *The American journal of clinical nutrition*, 82(5):1032-1039.
- Faber, M., Laubscher, R. & Berti, C. 2014. Poor dietary diversity and low nutrient density of the complementary diet for 6-to 24-month-old children in urban and rural KwaZulu-Natal, South Africa. *Maternal and child nutrition*, doi:10.1111/mcn.12146. Date of access: 2014 Aug 19.
- FAO (Food and Agriculture Organization of the United Nations). 2010. Fats and fatty acids in human nutrition: Report on an expert consultation. Geneva, Switzerland.

FAO, IFAD, WFP. 2015. The state of food insecurity in the world. Meeting the 2015 international hunger targets: taking stock of uneven progress. Rome: FAO.

Farquharson, J., Jamieson, E.C., Abbasi, K.A., Patrick, W.J., Logan, R.W. & Cockburn, F. 1995. Effect of diet on fatty acid composition of the major phospholipids of infant cerebral cortex. *Archives of disease in childhood*, 72:198-203.

Fernald, L.C., Kariger, P., Engle, P. & Raikes, A. 2009. Examining early child development in low-income countries. Washington, DC: The World Bank.

Fernandes, M., Stein, A., Newton, C.R., Cheikh-Ismaïl, L., Kihara, M., Wulff, K., *et al.* 2014. The INTERGROWTH-21 st Project Neurodevelopment Package: A novel method for the multi-dimensional assessment of neurodevelopment in pre-school age children. *PLoS ONE*, 9(11): e113360. doi:10.1371/journal.pone.0113360.

Fernandez-Rao, S., Hurley, K.M., Nair, K.M., Balakrishna, N., Radhakrishna, K.V., Ravinder, P., *et al.* 2014. Integrating nutrition and early child-development interventions among infants and preschoolers in rural India. *Annals of the New York Academy of Sciences*, 1308(1):218-231.

Forouzanfar, M.H., Alexander, L., Anderson, H.R., Bachman, V.F., Biryukov, S., Brauer, M., *et al.* 2015. Global, regional, and national comparative risk assessment of 79 behavioural, environmental and occupational, and metabolic risks or clusters of risks in 188 countries, 1990-2013: a systematic analysis of the Global Burden of Disease Study 2013. *The lancet*, [http://dx.doi.org/10.1016/S0140-6736\(15\)00128-2](http://dx.doi.org/10.1016/S0140-6736(15)00128-2).

Galler, J.R., Bryce, C.P., Waber, D.P., Zichlin, M.L., Fitzmaurice, G.M. & Eaglesfield, D. 2012. Socioeconomic outcomes in adults malnourished in the first year of life: a 40-year study. *Pediatrics*, 130(1):e1-e7.

Galler, J.R., Bryce, C.P., Zichlin, M.L., Fitzmaurice, G., Eaglesfield, G.D. & Waber, D.P. 2012. Infant malnutrition is associated with persisting attention deficits in middle adulthood. *The journal of nutrition*, 142(4):788-794.

Gera, T., Sachdev, H.S. & Boy, E. 2012. Effect of iron-fortified foods on hematologic and biological outcomes: systematic review of randomized controlled trials. *The American journal of clinical nutrition*, 96(2):309-324.

- German, J.B. & Dillard, C.J. 2006. Composition, structure and absorption of milk lipids: a source of energy, fat-soluble nutrients and bioactive molecules. *Critical reviews in food science and nutrition*, 46(1):57-92.
- Gibson, R. 2011. Strategies for preventing multi-micronutrient deficiencies: a review of experiences with food-based approaches in developing countries. (In Thompson B, Amoroso, L. eds. Combating micronutrient deficiencies: food-based approach. CABI and FAO: Oxfordshire, UK, pp 1-92).
- Gillespie, S., Haddad, L., Mannar, V., Menon, P., Nisbett, N., Maternal, *et al.* 2013. The politics of reducing malnutrition: building commitment and accelerating progress. *The lancet*, 382(9891):552-569.
- Gidrewics, D.A. & Fenton, T.R. 2014. A systematic review and meta-analysis of the nutrient content of preterm and term breast milk. *BMC Pediatrics*, 2014(14):216.
- Gladstone, M., Lancaster, G.A., Umar, E., Nyirenda, M., Kayira, E., van den Broek, N.R., *et al.* 2010. The Malawi Developmental Assessment Tool (MDAT): the creation, validation, and reliability of a tool to assess child development in rural African settings. *PLoS Med*, 7(5):e1000273.
- Goosen, C., McLachlan, M. & Schübl, C. 2014. Infant feeding practices during the first 6 months of life in a low-income area of the Western Cape Province. *South African journal of child health*, 8(2):50-54.
- Gorstein, J., Sullivan, K.M., Parvanta, I., & Begin, F. 2007. Indicators and methods for cross-sectional surveys of vitamin and mineral status of populations. The Micronutrient Initiative/ Atlanta: Centers For Disease Control And Prevention.
- Graham, S.M., Arvela, O.M. & Wise, G.A. 1992. Long-term neurologic consequences of nutritional vitamin B 12 deficiency in infants. *The Journal of pediatrics*, 121(5):710-714.
- Grantham-McGregor, S. & Ani, C. 2001. A review of studies on the effect of iron deficiency on cognitive development in children. *The journal of nutrition*, 131(2):649S-668S.
- Grantham-McGregor, S. & Baker-Henningham, H. 2005. Review of the evidence linking protein and energy to mental development. *Public health nutrition*, 8(7a):1191-1201.
- Hackman, D.A. & Farah, M.J. 2009. Socioeconomic status and the developing brain. *Trends in cognitive sciences*, 13(2):65-73.

- Haider, B.A. & Bhutta, Z.A. 2011. Neonatal vitamin A supplementation for the prevention of mortality and morbidity in term neonates in developing countries. *Cochrane Database Systematic Reviews*, 10: CD006980.
- Haroon, S., Das, J.K., Salam, R.A., Imdad, A. & Bhutta, Z.A. 2013. Breastfeeding promotion interventions and breastfeeding practices: a systematic review. *BMC public health*, 13(3):S20.
- Heaton, A.E., Meldrum, S.J., Foster, J.K., Simmer, K. 2013. Does docosahexaenoic acid supplementation in term infants enhance neurocognitive functioning? *Frontiers in human neuroscience*, 7(774):45-56.
- Hermoso, M., Vollhardt, C., Bergmann, K. & Koletzko, B. 2011. Critical micronutrients in pregnancy, lactation, and infancy: considerations on vitamin D, folic acid, and iron, and priorities for future research. *Annals of nutrition and metabolism*, 59(1):5-9.
- Hertrampf, E. & Cortés, F. 2004. Folic acid fortification of wheat flour: Chile. *Nutrition reviews*, 62(suppl 1):S44-S48.
- Hess, S.Y., Bado, L., Aaron, G.J., Ouédraogo, J.B., Zeilani, M. & Brown, K.H. 2011. Acceptability of zinc-fortified, lipid-based nutrient supplements (LNS) prepared for young children in Burkina Faso. *Maternal and child nutrition*, 7(4):357-367.
- Innis, S.M. 1991. Essential fatty acids in growth and development. *Progress in lipid research*, 30:39-130.
- Innis, S.M. 2003. Perinatal biochemistry and physiology of long-chain polyunsaturated fatty acids. *Journal of pediatrics*, 143:1-8.
- Honein, M.A., Paulozzi, L.J., Mathews, T., Erickson, J.D. & Wong, L.-Y.C. 2001. Impact of folic acid fortification of the US food supply on the occurrence of neural tube defects. *Journal of the American Medical Association*, 285(23):2981-2986.
- Horta, B.L. & Victora, C.G. 2013. Long-term effects of breastfeeding. Geneva, Switzerland: World Health Organization.
- Hotz, C. & Brown, K.H. 2004. International zinc nutrition consultative group (IZiNCG) Technical Document #1. Assessment of the risk of zinc deficiency in populations and options for its control. *Food and nutrition bulletin*, 25(1):S94-S200.
- Hotz, C., Loechl, C., Lubowa, A., Tumwine, J.K., Ndeezi, G., Masawi, A.N., et al. 2012. Introduction of β -carotene-rich orange sweet potato in rural Uganda resulted in increased

vitamin A intakes among children and women and improved vitamin A status among children. *The journal of nutrition*, 142(10):1871-1880.

HSRC. Human Sciences Research Council. 2013 Nov. The South African National Health and Nutrition Examination Survey (SANHANES-1). Data analysis on infant feeding practices, and anthropometry in children under five years of age: South Africa 2012. Report for UNICEF. Cape Town.

Hurrell, R. & Egli, I. 2010. Iron bioavailability and dietary reference values. *The American journal of clinical nutrition*, 91(5):1461S-1467S.

Hutton, E.K. & Hassan, E.S. 2007. Late vs early clamping of the umbilical cord in full-term neonates: systematic review and meta-analysis of controlled trials. *Journal of the American Medical Association*, 297(11):1241-1252.

Huybregts, L., Houngré, F., Salpéteur, C., Brown, R., Roberfroid, D., Ait-Aissa, M., *et al.* 2012. The effect of adding ready-to-use supplementary food to a general food distribution on child nutritional status and morbidity: a cluster-randomized controlled trial. *PLoS Medicine*, 9(9):e1001313.

(IFPRI) (International Food Policy Research Institute). 2014. Global nutrition report 2014: actions and accountability to accelerate the world's progress on nutrition. Washington, DC.

Iannotti, L.L., Dulience, S.J.L., Green, J., Joseph, S., François, J., Anténor, M.-L., *et al.* 2014. Linear growth increased in young children in an urban slum of Haiti: a randomized controlled trial of a lipid-based nutrient supplement. *The American journal of clinical nutrition*, 99(1):198-208.

Ilcol, Y.O., Ozbek, R., Hamurtekin, E. & Ulus, I.H. 2005. Choline status in new-borns, infants, children, breast-feeding women, breast-fed infants and human breastmilk. *The journal of nutritional biochemistry*, 16(8):489-499.

Iliff, P.J., Piwoz, E.G., Tavengwa, N.V., Zunguza, C.D., Marinda, E.T., Nathoo, K.J., *et al.* 2005. Early exclusive breastfeeding reduces the risk of postnatal HIV-1 transmission and increases HIV-free survival. *Aids*, 19(7):699-708.

Imdad, A. & Bhutta, Z.A. 2011. Effect of preventive zinc supplementation on linear growth in children under 5 years of age in developing countries: a meta-analysis of studies for input to the lives saved tool. *BMC public health*, 11(Suppl 3):S22.

Innocenti. 2005, 22 Nov. Declaration on infant and young child feeding. Florence, Italy.

- Isaacs, E. & Oates, J. 2008. Nutrition and cognition: assessing cognitive abilities in children and young people. *European journal of nutrition*, 47(3):4-24.
- Jones, G., Steketee, R.W., Black, R.E., Bhutta, Z.A., Morris, S.S. & Group, B.C.S.S. 2003. How many child deaths can we prevent this year? *The lancet*, 362(9377):65-71.
- Jurado, M.B. & Rosselli, M. 2007. The elusive nature of executive functions: a review of our current understanding. *Neuropsychology review*, 17(3):213-233.
- Kalanda, B., Verhoeff, F. & Brabin, B. 2006. Breast and complementary feeding practices in relation to morbidity and growth in Malawian infants. *European journal of clinical nutrition*, 60(3):401-407.
- Kent, J.C., Arthur, P.G., Mitoulas, L.R. & Hartmann, P.E. 2009. Why calcium in breastmilk is independent of maternal dietary calcium and vitamin D. *Breastfeeding review*, 17(2):5.
- Khan, N.Z., Muslima, H., Begum, D., Shilpi, A.B., Akhter, S., Bilkis, K., *et al.* 2010. Validation of rapid neurodevelopmental assessment instrument for under-two-year-old children in Bangladesh. *Pediatrics*, 125(4):e755-e762.
- Koletzko, B., Boey, C.C., Campoy, C., Carlson, S.E., Chang, N., Guillermo-Tuazon, M., *et al.* 2014. Current information and Asian perspectives on long-chain polyunsaturated fatty acids in pregnancy, lactation, and infancy: systematic review and practice recommendations from an early nutrition academy workshop. *Annals of nutrition and metabolism*, 65(1):49-80.
- Korenromp, E. 2015. Nutrition targets and indicators for the post-2015 sustainable development goals: Accountability for the measurement of results in nutrition, a technical note, February 2015.
- Kovács, A., Funke, S., Marosvölgyi, T., Burus, I., Desci, T. 2005. Fatty acids in early human milk after preterm and full-term delivery. *Journal of pediatric gastroenterology and nutrition*, 41:454-459.
- Kramer, M.S. & Kakuma, R. 2012. Optimal duration of exclusive breastfeeding. *Cochrane Database Systematic Review*, 8:1-131.
- Labadarios, D, Swart, R., Maunder, E.M.W., Kruger, H.S., Gericke, G.J., Kuzwayo, P.M.N., *et.al.* 2008. The National Food Consumption Survey-Fortification Baseline (NFCS-FB-I): South Africa, 2005. *South African journal of clinical nutrition*, 21(3):245-300.

- Lamberti, L.M., Walker, C.L.F., Noiman, A., Victora, C. & Black, R.E. 2011. Breastfeeding and the risk for diarrhea morbidity and mortality. *BMC public health*, 11(Suppl 3):S15.
- Lamichhane D.K., Leem J.H., Kim H.C., Park M.S., Lee J.Y., Moon S.H. & Ko J.K. 2015. Association of infant and young child feeding practices with undernutrition in children: evidence from the Nepal Demographic and Health Survey. *Paediatrics and international child health*, doi: <http://dx.doi.org/10.1179/2046905515Y.0000000049>.
- Lanigan, J., Bishop, J., Kimber, A. & Morgan, J. 2001. Systematic review concerning the age of introduction of complementary foods to the healthy full-term infant. *European journal of clinical nutrition*, 55(5):309-320.
- Larque, E., Demmelmair, H., Koletzko, B. 2002. Perinatal supply and metabolism of long-chain polyunsaturated fatty acids: importance for the early development of the nervous system. *Annals of the New York Academy of Sciences*, 967:299-310.
- Lassi, Z.S., Das, J.K., Zahid, G., Imdad, A. & Bhutta, Z.A. 2013. Impact of education and provision of complementary feeding on growth and morbidity in children less than 2 years of age in developing countries: a systematic review. *BMC public health*, 13(3):S13.
- Lingam, R., Gupta, P., Zafar, S., Hill, Z., Yousafzai, A., Iyengar, S., *et al.* 2014. Understanding care and feeding practices: building blocks for a sustainable intervention in India and Pakistan. *Annals of the New York Academy of Sciences*, 1308(1):204-217.
- Longini I.M. jr., Nizam, A., Ali, M., Yunus, M., Shenvi, N. & Clemens, J.D. 2007. Controlling endemic cholera with oral vaccines. *PLoS Med*, 4(11):e336.
- Lönnerdal, B. 2003. Nutritional and physiologic significance of human milk proteins. *The American journal of clinical nutrition*, 77(6):1537S-1543S.
- López-Camelo, J.S., Castilla, E.E. & Orioli, I.M. 2010. Folic acid flour fortification: impact on the frequencies of 52 congenital anomaly types in three South American countries. *American journal of medical genetics part A*, 152(10):2444-2458.
- Lowe, N.M., Fekete, K. & Decsi, T. 2009. Methods of assessment of zinc status in humans: a systematic review. *The American journal of clinical nutrition*, 89:2040S-2051S.
- MacDonald, R.S. 2000. The role of zinc in growth and cell proliferation. *The journal of nutrition*, 130(5):1500S-1508S.

- Marriott, B.P., White, A., Hadden, L., Davies, J.C. & Wallingford, J.C. 2012. World Health Organization (WHO) infant and young child feeding indicators: associations with growth measures in 14 low-income countries. *Maternal and child nutrition*, 8(3):354-370.
- Mathews, T. 2007. Trends in spina bifida and anencephalus in the United States, 1991-2005. Hyattsville, MD: National Center for Health Statistics.
- Matias, S., Chaparro, C., Perez-Exposito, A., Peerson J. & Dewey K. 2011. Acceptability of lipid-based nutrient supplement among Guatemalan infants and young children. Washington, DC.: FHI 360/FANTA-2. [homepage on the Internet] 2011:c2013.
- McNamara, R.K. & Carlson, S.E. 2006. Role of omega-3 fatty acids in brain development and function: potential implications for the pathogenesis and prevention of psychopathology. *Prostaglandins leukotrienes & essential fatty acids*, 75:329-349.
- Modi, N. 2006. Donor breastmilk banking. *British medical journal*, 333(7579):1133.
- Moore, E.R., Anderson, G.C., Bergman, N. & Dowswell, T. 2012. Early skin-to-skin contact for mothers and their healthy newborn infants. *Cochrane Database Systematic Review*, 5: CD003519.
- Moynahan, E. 1974. Acrodermatitis enteropathica: a lethal inherited human zinc-deficiency disorder. *The lancet*, 304(7877):399-400.
- Munos, M.K., Walker, C.L. & Black, R.E. 2010. The effect of oral rehydration solution and recommended home fluids on diarrhoea mortality. *International journal of epidemiology*, 39(1):i75–i87.
- Nasreddine, L., Zeidan, M., Naja, F. & Hwalla, N. 2012. Complementary feeding in the MENA region: practices and challenges. *Nutrition, metabolism and cardiovascular diseases*, 22(10):793-798.
- NDOH. (National Department of Health) see South Africa.
- Nel, E., Kruger, H.S., Baumgartner, J., Faber, M. & Smuts, C.M. 2015. Differential ferritin interpretation methods that adjust for inflammation yield discrepant iron deficiency prevalence. *Maternal and child nutrition*, doi: 10.1111/mcn.12175. Date of access: 2015 Feb 26.
- Neufeld, L.M. & Ramakrishnan, U. 2011. Multiple micronutrient interventions during early childhood: moving towards evidence-based policy and program planning. *The journal of nutrition*, 141(11):2064-2065.

- Nor, B., Ahlberg, B.M., Doherty, T., Zembe, Y., Jackson, D. & Ekström, E.C. 2012. Mother's perceptions and experiences of infant feeding within a community-based peer counselling intervention in South Africa. *Maternal and child nutrition*, 8(4):448-458.
- Nores, M. & Barnett, W.S. 2010. Benefits of early childhood interventions across the world: (under)investing in the very young. *Economics of education review*, 29(2):271-282.
- Nyaradi, A., Li, J., Hickling, S., Foster J. & Oddy W.H. 2013. The role of nutrition in children's neurocognitive development, from pregnancy through childhood. *Frontiers in human neuroscience*, 7(97):1-16.
- Pelto, G.H., Armar-Klimesu, M., Siekmann, J. & Schofield, D. 2013. The focused ethnographic study 'assessing the behavioral and local market environment for improving the diets of infants and young children 6 to 23 months old' and its use in three countries. *Maternal and child nutrition*, 9(S1):35-46.
- Pena-Rosas, J.P., De-Regil, L.M., Rogers, L.M., Bopardikar, A. & Panisset, U. 2012. Translating research into action: WHO evidence-informed guidelines for safe and effective micronutrient interventions. *The journal of nutrition*, 142(1):197S-204S.
- Perry, H.B., Zulliger, R. & Rogers, M.M. 2014. Community health workers in low-, middle-, and high-income countries: an overview of their history, recent evolution, and current effectiveness. *Annual review of public health*, 35:399-421.
- Persad, V.L., Van den Hof, M.C., Dubé, J.M. & Zimmer, P. 2002. Incidence of open neural tube defects in Nova Scotia after folic acid fortification. *Canadian Medical Association journal*, 167(3):241-245.
- Phuka, J., Ashorn, U., Ashorn, P., Zeilani, M., Cheung, Y.B., Dewey, K.G., *et al.* 2011. Acceptability of three novel lipid-based nutrient supplements among Malawian infants and their caregivers. *Maternal and child nutrition*, 7(4):368-377.
- Prado, E.L. & Dewey, K.G. 2014. Nutrition and brain development in early life. *Nutrition reviews*, 72(4):267-284.
- Prentice, A.M. & van der Merwe, L. 2011. Impact of fatty acid status on immune function of children in low-income countries. *Maternal and child nutrition*, 7(s2):89-98.
- Pulakka, A., Ashorn, U., Cheung, Y., Dewey, K., Maleta, K., Vosti, S., *et al.* 2014. Effect of 12-month intervention with lipid-based nutrient supplements on physical activity of 18-month-old

Malawian children: a randomised, controlled trial. *European journal of clinical nutrition*, 69:173-178.

Qian, M., Wang, D., Watkins, W.E., Gebiski, V., Yan, Y.Q., Li, M., *et al.* 2005. The effects of iodine on intelligence in children: a meta-analysis of studies conducted in China. *Asia Pacific journal of clinical nutrition*, 14(1):32-42.

Ramakrishnan, U., Nguyen, P. & Martorell, R. 2009. Effects of micronutrients on growth of children under 5y of age: meta-analyses of single and multiple nutrient interventions. *The American journal of clinical nutrition*, 89(1):191-203.

Ray, J.G., Meier, C., Vermeulen, M.J., Boss, S., Wyatt, P.R. & Cole, D.E. 2002. Association of neural tube defects and folic acid food fortification in Canada. *The lancet*, 360(9350):2047-2048.

Rivera, J.A., Hotz, C., González-Cossío, T., Neufeld, L. & García-Guerra, A. 2003. The effect of micronutrient deficiencies on child growth: a review of results from community-based supplementation trials. *The journal of nutrition*, 133(11):4010S-4020S.

Rohner, F., Zimmermann, M., Jooste, P., Pandav, C., Caldwell, K., Raghavan, R., *et al.* 2014. Biomarkers of nutrition for development—iodine review. *The journal of nutrition*, 144:1322S-1342S.

Ruel, M.T. & Alderman, H. 2013. Nutrition-sensitive interventions and programmes: how can they help to accelerate progress in improving maternal and child nutrition? *The lancet*, 382(9891):536-551.

Sabanathan, S., Wills, B. & Gladstone, M. 2015. Child development assessment tools in low-income and middle-income countries: how can we use them more appropriately? *Archives of disease in childhood*, 100:482-488.

Sandstead, H.H., Frederickson, C.J. & Penland, J.G. 2000. History of zinc as related to brain function. *The journal of nutrition*, 130(2):496S-502S.

Sayed, A.R., Bourne, D., Pattinson, R., Nixon, J. & Henderson, B. 2008. Decline in the prevalence of neural tube defects following folic acid fortification and its cost-benefit in South Africa. *Birth defects research part A: clinical and molecular teratology*, 82(4):211-216.

Schoeman, S., Faber, M., Adams, V., Smuts, C., Ford-Ngomane, M., Laubscher, J., *et al.* 2010. Adverse social, nutrition and health conditions in rural districts of the KwaZulu-Natal and

Eastern Cape provinces, South Africa. *South African journal of clinical nutrition*, 23(3):140-147.

Scholl, T.O. 2011. Maternal iron status: Relation to fetal growth, length of gestation and the neonate's iron endowment. *Nutrition Reviews*, 69(1):S23-S29.

Senarath, U., Agho, K.E., Akram, D., Godakandage, S.P., Hazir, T., Jayawickrama, H., *et al.* 2012. Comparisons of complementary feeding indicators and associated factors in children aged 6-23 months in five South Asian Countries. *Maternal and child nutrition*, 8(1):89-106.

Shafir, T., Angulo-Barroso, R., Jing, Y., Angelilli, M.L., Jacobson, S.W. & Lozoff, B. 2008. Iron deficiency and infant motor development. *Early human development*, 84(7):479-485.

Shisana, O., Labadarios, D., Rehle, T., Simbaji, L., Zuma, K., Dhansay, A., *et al.* 2014. The South African National Health and Nutrition Examination Survey: SANHANES-1. HSRC Press; 2014.

Siega-Riz, A.M., Estrada Del Campo, Y., Kinlaw, A., Reinhart, G.A., Allen, L.H., Shahab-Ferdows, S. *et al.* 2014. Effect of supplementation with a lipid-based nutrient supplement on the micronutrient status of children aged 6–18 months living in the rural region of Intibucá, Honduras. *Paediatric and perinatal epidemiology*, 28(3):245-254.

Sizonenko, S.V., Babiloni, C., de Bruin, E.A., Isaacs, E.B., Jönsson, L.S., Kennedy, D.O., *et al.* 2013. Brain imaging and human nutrition: which measures to use in intervention studies? *British journal of nutrition*, 110:S1-S30.

Smuts, C.M., Dhansay, M.A., Faber, M., van Stuijvenberg, M.E., Swanevelder, S., Gross, R. *et al.* 2005. Efficacy of multiple micronutrient supplementation for improving anemia, micronutrient status, and growth in South African infants. *The journal of nutrition*, 135(3):653S–659S.

Smuts, C.M., Lombard, C.J., Benadé, A.S., Dhansay, M.A., Berger, J., de Romaña, G.L., *et al.* 2005. Efficacy of a foodlet-based multiple micronutrient supplement for preventing growth faltering, anemia, and micronutrient deficiency of infants: the four country IRIS trial pooled data analysis. *The journal of nutrition*, 135(3):631S-638S.

South Africa. Department of Health. 2007. Infant and young child feeding policy. Pretoria.

South Africa. Department of Health. 2011. Tshwane declaration of support for breastfeeding in South Africa, 23-24 Aug. Tshwane, South Africa.

South Africa. Department of Health. 2013a. South African infant and young child feeding policy. Pretoria.

South Africa. Department of Health. 2013b. Roadmap for nutrition in South Africa 2013-2017. Pretoria.

South Africa. Department of Health. 2013c. Extension notice R433 and R434: regulations relating to foodstuffs for infants and young children. Pretoria: Government Gazette: 2013.

South Africa. Department of Health & UNICEF South Africa. 2007. A reflection of the South African maize meal and wheat flour fortification programme. Pretoria: South African Department of Health.

South Africa. Department of Social Development. 2015. Draft early childhood development policy. Government Gazette, 597:38558: 13 March 2015.

South Africa. Department of Health, Department of Social Development, Department of Performance, Monitoring and Evaluation. 2014. Final evaluation report: short diagnostic/implementation evaluation of nutrition interventions for children from conception to age 5. Pretoria: South African Department of Health.

Steenkamp, L. & Lategan, R. 2013. The acceptability and intake of lipid-based pastes as a food supplement in a South African context: short report. *South African journal of clinical nutrition*, 26(3):150-151.

Stoltzfus, R. & Underwood, B.A. 1995. Breast-milk vitamin A as an indicator of the vitamin A status of women and infants. *Bulletin of the World Health Organization*, 73(5):703.

SUN. (Scaling up nutrition). 2014 Nov. An introductory to the SUN movement civil society network. *Scaling up nutrition in outline*, 2:1-12.

Tang, G., Qin, J., Dolnikowski, G.G., Russell, R.M. & Grusak, M.A. 2009. Golden Rice is an effective source of vitamin A. *The American journal of clinical nutrition*, 89(6):1776-1783.

Thurnham, D.I., Northrop-Clewes, C.A. & Knowles, J. 2015. The use of adjustment factors to address the impact of inflammation on vitamin A and iron status in humans. *The journal of nutrition*, 145(5):1137S-1143S.

Tolentino, K. & Friedman, J.F. 2007. An update on anemia in less developed countries. *The American journal of tropical medicine and hygiene*, 77(1):44-51.

- Tylleskär, T., Jackson, D., Meda, N., Engebretsen, I.M.S., Chopra, M., Diallo, A.H., *et al.* 2011. Exclusive breastfeeding promotion by peer counsellors in sub-Saharan Africa (PROMISE-EBF): a cluster-randomised trial. *The lancet*, 378(9789):420-427.
- Uauy, R., Mize, C.E. & Castillo-Duran, C. 2000. Fat intake during childhood: metabolic responses and effects on growth. *The American journal of clinical nutrition*, 72(5):1354s-1360s.
- UNICEF (United Nations Children's Fund). 2004. Malaria: A major cause of child death and poverty in Africa. New York, USA.
- UNICEF (United Nations Children's Fund). 2008. The state of the world's children 2009. New York, USA.
- UNICEF (United Nations Children's Fund). 2012. Pneumonia and diarrhoea. Tackling the deadliest diseases for the world's poorest children. New York, USA.
- UNICEF (United Nations Children's Fund). 2014. Statistical tables: economic and social statistics on the countries and areas of the world, with particular reference to the children's well-being. New York, USA.
- UNICEF (United Nations Children's Fund). 2015. Humanitarian action for children: Kenya. New York, USA.
- UNICEF (United Nations Children's Fund), WHO (World Health Organization) & World Bank Group joint malnutrition estimates. 2015. Levels and trends in child malnutrition: key findings of 2015 edition. New York, USA.
- Unnevehr, L., Pray, C. & Paarlberg, R. 2007. Addressing micronutrient deficiencies: alternative interventions and technologies. *The journal of agrobiotechnology management & economics*, 10(3):124-134.
- Vierhaus, M., Lohaus, A., Kolling, T., Teubert, M., Keller, H., Fassbender, I., *et al.* 2011. The development of 3-to 9-month-old infants in two cultural contexts: Bayley longitudinal results for Cameroonian and German infants. *European journal of developmental psychology*, 8(3):349-366.
- Van Dusseldorp, M., Schneede, J., Refsum, H., Ueland, P.M., Thomas, C.M., de Boer, E., *et al.* 1999. Risk of persistent cobalamin deficiency in adolescents fed a macrobiotic diet in early life. *The American journal of clinical nutrition*, 69(4):664-671.

- Von Schenck, U., Bender-Götze, C. & Koletzko, B. 1997. Persistence of neurological damage induced by dietary vitamin B-12 deficiency in infancy. *Archives of disease in childhood*, 77(2):137-139.
- Wainwright, P.E. 2002. Dietary essential fatty acids and brain function: a developmental perspective on mechanisms. *Proceedings of the Nutrition Society*, 61(01):61-69.
- Walker, S.P., Wachs, T.D., Grantham-McGregor, S., Black, M.M., Nelson, C.A., Huffman, S.L., *et al.* 2011. Inequality in early childhood: risk and protective factors for early child development. *The lancet*, 378(9799):1325-1338.
- Walker, S.P., Wachs, T.D., Meeks Gardner, J., Lozoff, B., Wasserman, G.A., Pollitt, E., *et al.* 2007. Child development: risk factors for adverse outcomes in developing countries. *The lancet*, 369(9556):145-157.
- Walter, T. 2003. Effect of iron-deficiency anemia on cognitive skills and neuromaturation in infancy and childhood. *Food and nutrition bulletin*, 24(4):S104-S110.
- Wang, H., Liddell, C.A., Coates, M.M., Mooney, M.D., Levitz, C.E., Schumacher, A.E., *et al.* 2014. Global, regional, and national levels of neonatal, infant, and under-5 mortality during 1990-2013: a systematic analysis for the Global Burden of Disease Study 2013. *The lancet*, 384(9947):957-979.
- Wang, Y. & Chen, H.J. 2012. Use of percentiles and z-scores in Anthropometry. In: Preedy, V.R (ed). *Handbook of Anthropometry. Physical Measures of human form in health and disease.*, ISBN: 978-1-4419-1787-4.
- Wessells, K.R. & Brown, K.H. 2012. Estimating the global prevalence of zinc deficiency: Results based on zinc availability in national supplies and the prevalence of stunting. *PLOS*, 7(11):1-11.
- WHO (World Health Organization) Multicentre Growth Reference Study Group. 2006b. WHO motor development study: Windows of achievements for six gross motor development milestones. *Acta paediatrica*, 450:86-95.
- WHO (World Health Organization). 2003. Guiding principles for complementary feeding of the breastfed child. Washington: Pan American Health.
- WHO (World Health Organization). 2005. Guiding principles for feeding non-breastfed children 6-24 months of age. Geneva, Switzerland.

WHO (World Health Organization). 2006a. WHO child growth standards: Length/height-for-age, weight-for-age, weight-for-length, weight-for-height and body mass index-for-age, methods and development. Geneva, Switzerland.

WHO (World Health Organization). 2008. Indicators for assessing infant and young child feeding practices: part 1: definitions: conclusions of a consensus meeting held 6-8 November 2007 in Washington DC, USA.

WHO (World Health Organization). 2009a. Global prevalence of vitamin A deficiency in populations at risk 1995-2005: WHO global database on vitamin A deficiency. Geneva, Switzerland.

WHO (World Health Organization). 2009b. Infant and young child feeding: model chapter for textbooks for medical students and allied health professionals. Geneva, Switzerland.

WHO (World Health Organization). 2011a. Haemoglobin concentrations for the diagnosis of anaemia and assessment of severity. Vitamin and Mineral Nutrition Information System. Geneva, Switzerland.

WHO (World Health Organization). 2011b. Serum ferritin concentrations for the assessment of iron status and iron deficiency in populations. Vitamin and Mineral Nutrition Information System. Geneva, Switzerland.

WHO (World Health Organization). 2011c. Serum retinol concentrations for determining the prevalence of vitamin A deficiency in populations. Geneva, Switzerland.

WHO (World Health Organization). 2013a. Essential Nutrition Actions: Improving maternal, newborn, infant and young child health and nutrition. Geneva, Switzerland.

WHO (World Health Organization). 2013b. Roadmap for childhood tuberculosis. Geneva, Switzerland.

WHO (World Health Organization). 2014. Maternal, infant and young child nutrition in East and Southern African countries: moving to national implementation, report of a World Health Organization workshop, Entebbe, Uganda, 26–28 November 2013.

WHO (World Health Organization). 2015a. HIV/AIDS <http://www.who.int/mediacentre/factsheets/fs360/en/>. Date of access: 24 Sep.2015.

WHO (World Health Organization). 2015b. Global database on child growth and malnutrition: <http://www.who.int/nutgrowthdb/about/introduction/en/index5.html> Date of access: 24 Sep. 2015.

WHO (World Health Organization). 2015c. World Health Statistics. Geneva: World Health Organization.

WHO & FAO (World Health Organization & Food and Agriculture Organization of the United Nations). 2006. Guidelines on food fortification with micronutrients. Geneva, Switzerland: World Health Organization.

WHO & UNICEF (World Health Organization & United Nations Children's Fund). 2003. Global strategy for infant and young child feeding. Geneva, Switzerland: World Health Organization.

WHO & UNICEF (World Health Organization & United Nations Children's Fund). 2006. Oral Rehydration salts: Production of the new ORS. Geneva, Switzerland: World Health Organization.

WHO & UNICEF (World Health Organization & United Nations Children's Fund). 2009a. Baby-friendly hospital initiative: Revised, updated and expanded for integrated care. Geneva, Switzerland: World Health Organization.

WHO & UNICEF (World Health Organization & United Nations Children's Fund). 2009b. WHO child growth standards and the identification of severe acute malnutrition in infants and children. Geneva, Switzerland: World Health Organization.

WHO & UNICEF (World Health Organization & United Nations Children's Fund). 2014. Global nutrition targets 2025: breastfeeding policy brief. Geneva, Switzerland: World Health Organization.

WHO/UNICEF/WFP (World Health Organization)/United Nations Children's Fund/World Food Program). 2014. Global nutrition targets 2025: wasting policy brief. Geneva, Switzerland: World Health Organization.

Williams, L.J., Mai, C.T., Edmonds, L.D., Shaw, G.M., Kirby, R.S., Hobbs, C.A., *et al.* 2002. Prevalence of spina bifida and anencephaly during the transition to mandatory folic acid fortification in the United States. *Teratology*, 66(1):33-39.

World Bank. 2006. Repositioning nutrition as central to development: a strategy for large-scale action. Washington DC.

Yakoob, M.Y., Theodoratou, E., Jabeen, A., Imdad, A., Eisele, T.P., Ferguson, J., *et al.* 2011. Preventive zinc supplementation in developing countries: impact on mortality and morbidity due to diarrhea, pneumonia and malaria. *BMC public health*, 11(3):S23.

Zeisel, S.H. 2006a. Choline: critical role during fetal development and dietary requirements in adults. *Annual review of nutrition*, 26:229-250.

Zeisel, S.H. 2006b. Choline and brain development. In: Bowman, B.R., Russell, R.M. (ed). *Present knowledge in Nutrition*. 9th ed. 1. ILSI Press; Washington, DC: 352-360.

Zhou, H., Wang, X.-L., Ye, F., Zeng, X.L. & Wang, Y. 2012. Relationship between child feeding practices and malnutrition in 7 remote and poor counties, PR China. *Asia Pacific journal of clinical nutrition*, 21(2):234-240.

CHAPTER 3

This manuscript is formatted according to guidelines of Maternal and Child Nutrition (see Annexure 2), with exception of the line numbers which will be inserted and the line spacing which will be changed to double spacing before submission.

Feeding practices in relation to nutritional status of 6-month old infants from a peri-urban setting in South Africa.

Marinel Rothman¹, Cornelius M Smuts¹, Tonderayi M Matsungu¹, Namukolo Covic², Marike Cockeran³, Mieke Faber⁴

¹ Centre of Excellence for Nutrition (CEN), Faculty of Health Sciences, North-West University, Potchefstroom Campus, Private Bag x6001, Potchefstroom 2520, South Africa.

² Poverty Health and Nutrition Division, International Food Policy Research Institute, P O Box 5689, Addis Ababa, Ethiopia.

³ Medicine Usage in South Africa (MUSA), North-West University, Potchefstroom Campus, Private Bag x6001, Potchefstroom 2520, South Africa.

⁴ Non-communicable Diseases Research Unit, South African Medical Research Council (SAMRC), PO Box 19070, Tygerberg 7505, South Africa.

Corresponding author: CM Smuts; Private Bag X6001, Potchefstroom 2520, South Africa; Tel +27-18-2992086; Fax +27-18-2992464; Email marius.smuts@nwu.ac.za

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Authors' contributors:

M.R.: One of the study-coordinators of the Tswaka study. Involved in questionnaire development and study design, and provided supervision on field data collection and dietary data quality control. Responsible for analysis and interpretation of results, and drafted the paper.

C.M.S.: Principle investigator of the Tswaka trial. Developed protocol and study design of Tswaka study. Provided guidance on biochemical data collection, quality control and analysis. Provided scientific input on data analysis and –interpretation.

T.M.M.: One of the study coordinators of Tswaka trial. Provided supervision of field data collection and quality control of anthropometric data. Revision of paper

N.C.: Academic input. Revision of paper.

M.C.: Provided guidance on statistical analysis and interpretation

M.F.: Co-principle investigator of the Tswaka study. Study design and protocol development of Tswaka trial. Guidance on the assessment of dietary intake and feeding practices. Scientific input on data analysis and –interpretation.

All authors read and approved the final version of the paper.

Abstract

The first two years of life is a period of rapid growth and development. Iron is a key nutrient needed for child growth and cognitive development. In this cross-sectional study, feeding practices, nutrient intake, and nutrient density of the complementary diet were determined in relation to anaemic and iron status of 6-month old infants ($n = 750$) from a peri-urban community of the North-West province, South Africa. For 48.9% of infants, exclusive breastfeeding was practiced up to age 0–2 months. Semi-solid/solid foods were introduced

mostly at age 3–4 months. The most popular first foods were commercial infant cereals (63.8%) and jarred infant foods (20.3%). At the age of 6 months, 70.1% of infants were still being breastfed; food items mostly consumed were infant cereal (68.1%), formula milk (42.0%) and jarred infant foods (22.7%). The nutrient density of the complementary diet was low for > 80% of infants for iron, zinc and calcium, respectively. In total, 36.4% of the infants were anaemic (haemoglobin < 11 g/l); 16.1% had plasma ferritin < 12 µg/l, 30.1% had transferrin receptors > 8.3 mg; and 29.3% were stunted. Dietary intake of iron, zinc, magnesium and B-vitamins were significantly higher for non-anaemic infants. Regression analysis showed that shorter duration of exclusive breastfeeding and frequent consumption of formula milk were associated with a lower chance of infants being anaemic. The early introduction of fortified formula milk and commercial infant cereal may have a protective effect on anaemia and iron deficiency in this study population.

Keywords: Infant feeding practices, nutrients, anaemia, iron, breastfeeding, South Africa

Introduction

During the gestation period and throughout infancy to the age of 2 years (also referred to as *The first thousand days*), good nutrition allows brain functioning to develop and immune systems to develop more strongly (IFPRI 2014). *The first thousand days* of life is a period of rapid growth and development (Bentley et al. 2014) and nutritional deficits during this period may cause long-term impairment in growth and intellectual performance (Prado & Dewey 2014). Iron is one of the key nutrients needed for early child growth and development (Hermoso et al. 2011; Prado & Dewey 2014). Globally, iron deficiency is the most common nutritional deficiency and the primary cause of anaemia. Almost 50% of children younger than 5 years are anaemic (Benoist et al. 2008). Iron deficiency (with or without anaemia) is associated with adverse long-term neurodevelopmental and behavioural outcomes and irreversible negative effects on brain development (Lozoff et al. 2006; Baker & Greer 2010). In South Africa, the 2005 National Food Consumption Survey (NFCS-FB-1) showed that 38.7% of 1–3 year old children were anaemic (Labadarios et al. 2007). Limited data is available on anaemia and iron status of infants younger than 12 months. A systematic review done on iron status of European children aged 6–36 months reported that 2–25% of infants aged 6–12 months were iron deficient. The prevalence of iron deficiency anaemia (IDA) was below 5% for infants and young children in Northern and Western Europe (Eussen et al. 2015). For South African infants, baseline data of intervention studies conducted more than ten years

ago in rural KwaZulu-Natal on infants aged 6–12 months, showed that approximately 40–45% of the children were anaemic (Smuts et al. 2005; Faber et al. 2005).

The World Health Organization (WHO) recommends exclusive breastfeeding for the first 6 months of life with continued breastfeeding until the age of 2 years or older (WHO 2009). Breast milk has low iron content, but infants retain more iron from human milk than cow's milk or infant formulas, because of the presence of lactoferrin and lactalbumin in human milk (Lönnerdal 2003). Still, a WHO committee expressed concerns that exclusively breastfed infants may become iron deficient (WHO 2001). It has been suggested that exclusively breastfed infants with a low birth weight must be supplemented with iron after 4 months of age (Greer 2015; Finkelstein et al. 2013; Baker & Greer 2010; Dube et al. 2010; Dewey & Chaparro 2007). Iron stores for healthy infants is most likely to be inadequate at age 5–7 months since infants must initially rely on iron reserves obtained in utero (Hermoso et al. 2011). Iron status of infants 0–6 months of age has no relation with iron and lactoferrin concentration in the breast milk of the mother (Finkelstein et al. 2013; Raj et al. 2008). However, maternal iron status during pregnancy is associated with infant iron stores postpartum (Finkelstein et al. 2013). Results of a systematic review that included three randomized controlled trials and one observational study showed that infants in developing countries who were introduced to solid foods at the age of 4 months had higher haemoglobin and serum ferritin concentrations compared to infants who were exclusively breastfed for six months (Qasem et al. 2015). The authors highlighted the small sample sizes of the studies included in the systematic review as a major limitation.

A high percentage of infants are introduced to complementary foods before the age of 6 months in both high- and low-income countries (IFPRI 2015). In African countries, including South Africa, grain-based watery porridges with low nutritional quality are often given (Faber et al. 2014; Cherop et al. 2010; Kalanda et al. 2006).

In South Africa, the prevalence of exclusive breastfeeding in infants younger than 6 months is very low (7.4%) (HSRC 2013) and the estimated prevalence of breastfeeding until the age of 2 years is only 30% (UNICEF 2014). The early cessation of breastfeeding may be due to cultural beliefs that breast milk alone is inadequate and also because of the perception of HIV+ mothers that HIV can be transmitted through breastfeeding (Doherty et al. 2012; Ladzani et al. 2011). Infants often receive complementary foods already from the age of 3 months. The complementary diet is often maize-based and of poor nutritional quality, resulting in low

protein-, vitamin-, mineral- and omega-3 fatty acid intake (Du Plessis et al. 2013; Faber et al. 2014). Of concern is the phytate content in maize, as it inhibits iron absorption (Clark 2008).

Against the background of the poor feeding practices during the first six months (low exclusive breastfeeding, early cessation of breastfeeding, early introduction of complementary food of poor nutrient quality) and limited data available on anaemia and iron status of South African infants the aim of this study was to determine early infant feeding practices in relation to iron status at age 6 months in infants in a peri-urban South African community.

Participants and Methods

Study population and study participants

This study reports on the baseline data of a randomized controlled trial involving 750, approximately 6-month-old infants (5.85–7.01 months) which was collected during the period of September 2013—January 2015. The trial was done in the peri-urban Jouberton area in the greater Matlosana Municipality in Klerksdorp in the North West province of South Africa. Jouberton is a low socio-economic community and is 200 km from the nearest metropolitan area (Johannesburg). The number of infants included in the study is based on the sample size calculations for the randomized controlled trial with growth as the main outcome. Infants were excluded if they 1) have never been breast fed 2) suffered from severe obvious congenital abnormalities, 3) were severely anaemic (haemoglobin < 7.0 g/dl); 4) were malnourished (weight-for-length Z-score < -3.00); 5) suffered from a chronic disease; 6) were allergic/intolerant for instance fish, peanuts, soy, milk and/or lactose; 7) received special nutritional supplements, 8) were not a singleton and 9) if their mothers planned to move out of the study area within the next seven months.

The mother-infant pairs were recruited through five primary health care clinics in the community and house-to-house visits. Trained fieldworkers who were fluent in the local language explained the study to the mothers. If the mother expressed willingness to participate, an eligibility questionnaire was administered to determine whether the infant could be included in the study.

All questionnaires used for data collection were completed at a central study site where the mothers were interviewed in their preferred language by trained field workers. The socio-economic information was collected using a structured questionnaire to describe the context

of the study population. The birth weight of the infant was recorded from the infant's clinic card.

Breastfeeding and complementary feeding practices were assessed retrospectively using a structured questionnaire that was developed based on WHO guidelines for assessing infant and young child feeding practices (WHO 2010). Indicators included were the duration of exclusive breastfeeding; age of introduction of liquids, milk feeds and solid foods and meal frequency per day.

Macro- and micronutrient intake were quantified through a single 24-hour dietary recall. Each fieldworker was provided with a standardised dietary kit that contained samples of empty food containers and wrappers, household utensils and three-dimensional sponge models to assist the mother and fieldworker to visualise, quantify and record the amount of food consumed by the infant the previous day. Dry oats was used to quantify portion sizes of certain food items, like cooked food. The mother used the dry oats to indicate the quantity that resembles the amount of food that the infant consumed, which the fieldworker then quantified using a measuring cup.

A set of unquantified food frequency questions was used to obtain descriptive qualitative information on the usual consumption of foods by the infants over the past seven days. These questions were used in similar studies (Faber et al. 2014; Smuts et al. 2005) and were tested for face validity. The mother had a choice of four options to describe the child's usual intake of listed foods. The four options were (i) every day; (ii) most days (not every day but at least four days per week); (iii) once a week (at least once a week, but less often than four days a week); and (iv) never. For each food item, children were grouped according to the usual consumption into two groups namely those who ate the food at least four days per week (frequent consumption) and those who ate the food less often than four days per week (seldom/never).

Iron status was determined by blood sample analysis. Blood samples (4 ml) were drawn into EDTA-coated trace-element free evacuated tubes (Becton Dickinson) by a professional nurse. If blood could not be obtained successfully via antecubital venipuncture of the arm area or dorsal area, a finger prick blood sample was taken to determine haemoglobin (Hb) status. Therefore, Hb values were collected from all infants ($n = 750$) while 4 ml blood samples were obtained from only 485 infants. Hb values were determined by using the Hemocue on an aliquot of whole blood or finger-prick by using the direct cyanmethaemoglobin method [Ames

Mini-Pak haemoglobin test pack and Ames Minilab, Bio Rad Laboratories (PTY) Ltd]. After the Hb test the remaining blood samples aliquots were prepared at 500 X g for 15 min at room temperature and the plasma and plasma aliquots were saved for later analysis. Samples were transported in cooler boxes containing ice cubes from the study site to the main laboratory at the Potchefstroom campus of the North-West University on a daily basis for storage at -80°C until being assayed. Storage temperature was monitored and logged for the duration of the study.

The samples were shipped for analysis to Vitmin Lab Willstaett, Germany in accordance with the National Department of Health regulations and specifications for shipment procedures. Plasma ferritin (Fer) and soluble transferrin receptor (TfR) concentrations were determined by using a sensitive Sandwich ELISA technique (Erhardt et al. 2004). High-sensitivity C-reactive protein (CRP) and alpha-1 glycoprotein (AGP) were measured with an ELISA kit from Human Diagnostics. These respective acute and chronic inflammation markers were assessed in order to eliminate possible falsely elevated plasma ferritin concentrations (Thurnham et al. 2015).

Anthropometric measurements were measured by two fieldworkers who were trained and standardized according to International Society for Advancement of Kinanthropometry (ISAK) procedures (Marfell-Jones et al. 2012). The protocol for the anthropometric measurements was based on the WHO Training Course on Child Growth Assessment (Version 1, November 2006) for length and weight measurements. Length was measured using a mechanical infantometer (Seca 416, Germany) and weight was taken with a calibrated digital scale (Seca 354, Germany). All measurements were done in duplicate. If the two measurements differed by > 0.05 kg for weight or by > 0.3 cm for length a third measurement was done and the two closest values were recorded. The WHO growth standards (2006) were used to calculate age and gender specific Z-scores for weight and length (WHO 2006).

Processing and statistical analysis of the data

All team members were trained to conduct the study according to good clinical practice (GCP). The source data was identified by date, participant number and signature of the data collector and was then stored in individual participant folders as per GCP requirements. Data on infant feeding practices were entered into an EpiInfo data base. Data obtained from socio-economic background, anthropometric status and blood results, were captured by ClinTech (India) International Pvt Ltd. Quality control was conducted to correct obvious errors in the data set.

Questionnaire data was analysed using SPSS for Windows, version 22 (SPSS Inc., Chicago, Illinois). Descriptive analysis was used to describe socio-economic factors and feeding practices.

For the 24-hour recall data, food intake reported in household measures was converted into weight using the MRC Food Quantities Manual (Langenhoven et al. 1991). For exclusively breastfed infants the estimated amount of breast milk intake was taken as 775 ml, and for partially breastfed infants an amount of 675 ml was taken (WHO 1998). For breastfeeding infants who also received bottle milk feeds, the amount of milk feed was subtracted from 675 ml to obtain the estimated amount of breast milk. The coded food intake data was entered into an Excel sheet. The SAS software package (version 9.4; SAS Institute Inc., Cary, NC) was used to convert food intake to macro and micronutrients, using the SAFOODS database (Wolmarans et al. 2010). The vitamin A values for plant foods, given as $\mu\text{g RE}$ in the SAFOODS database were divided by two to obtain $\mu\text{g RAE}$ values; for animal foods and fortified foods $\mu\text{g RE}$ is equal to $\mu\text{g RAE}$. The nutrient data were exported to SPSS for further data analysis. Because a single 24-hour recall was used, we calculated the median (IQR) for groups, but did not calculate the proportion below the Estimated average requirement (EAR) cut-off (Murphy et al. 2006). Nutrient density (amount of each nutrient per 100 kcal) of the complementary diet (excluding breast milk) was calculated and compared to the average desired densities (Dewey & Brown 2003).

Anaemia was defined as haemoglobin ($\text{Hb} < 11.0 \text{ g/dl}$). Iron deficiency (ID) based on plasma ferritin as $\text{Fer} < 12 \mu\text{g/l}$; and iron deficiency anaemia (IDA) as both $\text{Fer} < 12 \mu\text{g/l}$ and $\text{Hb} < 11.0 \text{ g/dl}$ (WHO 2011a, b). ID and IDA based on TfR, as $\text{TfR} > 8.3 \text{ mg/l}$ for ID and $\text{Hb} < 11.0 \text{ g/dl}$ and $\text{TfR} > 8.3 \text{ mg/l}$ for IDA (test-kit reference value). Inflammation was detected by acute phase proteins, $\text{AGP} > 1 \text{ g/l}$ and $\text{CRP} > 5 \text{ mg/l}$ (Thurnham et al. 2015). Individual's Fer concentrations were adjusted by using correction factors (CFs) specific to each subject's inflammatory status. Individuals were categorised into four groups based on their CRP and/or AGP concentrations: 1) an apparently healthy reference group ($\text{CRP} \leq 5 \text{ mg/l}$ and $\text{AGP} \leq 1 \text{ g/l}$); 2) an incubation group ($\text{CRP} > 5 \text{ mg/l}$ and $\text{AGP} \leq 1 \text{ g/l}$); 3) an early convalescence group ($\text{CRP} > 5 \text{ mg/l}$ and $\text{AGP} > 1 \text{ g/l}$) and 4) a late convalescence group ($\text{CRP} \leq 5 \text{ mg/l}$ and $\text{AGP} > 1 \text{ g/l}$). The Fer of the individual was then adjusted by using the relevant group specific CF as a multiplier and repeating the calculation to determine the prevalence of ID after correction. A CF of 0.77, 0.53 and 0.75 was used for the incubation group, early and late convalescence groups, respectively (Thurnham et al. 2015). Stunting was defined as a length-for-age Z-score

(LAZ) < -2 and overweight was defined a length-for-weight Z-score (WLZ) > 2 (Wang & Chen 2012). Low-birth weight was defined as < 2.50 kg (WHO 2011c).

Data was tested for normality by using Shapiro-Wilk test. Descriptive data are reported by percentages and cross-tabulations. Continuous data are reported in terms of mean and 95% confidence intervals (CI) for normal distributed data whereas data not normally distributed (dietary data) are reported as the median and interquartile range (IQR). Significant differences in mean values between groups were determined using an independent t-test or analysis of variance (ANOVA). For dietary intake data, significant differences between anaemic- and non-anaemic groups were determined using the non-parametric Mann-Whitney U test. Associations between categorical data were determined using the Pearson chi-square test. Associations between continuous data namely age of introduction to milk feeds, liquids and semi-solid foods and haemoglobin concentrations were determined by Pearson correlation analysis. Significant findings were further analysed by using binary logistic regression analysis with Hb (anaemic vs non-anaemic) as the dependent variable. Odds ratios (OR) and 95% CIs for the regression coefficients are reported. The nutrient intake was not included in the multiple logistic regression analysis as a single 24-hour dietary recall fails to indicate an individual's food intake. For all analysis, statistical significance was set at $P < 0.05$.

Ethical considerations

The randomized control trial was registered as a clinical trial at Clinicaltrials.gov registry (NCT01845610). Ethical approval was obtained from the Ethics Committees of North-West University and the South African Medical Research Council (SAMRC). After institutional ethical approval was obtained, the study was reviewed by the provincial Department of Health and Social Development for registration with the Directorate for Policy, Planning and Research. All mothers with eligible infants who agreed to participate were asked to read the information sheet and sign the informed consent form and they were given the opportunity to ask questions. For illiterate mothers, the fieldworker read the information out loud to the mother, and the mothers thumb print was used as "signature" on the informed consent form. Infants who were found to be severely anaemic or malnourished during screening were to be referred to the primary health care clinic and excluded from the study.

Results

According to Table 1, most of the primary caretakers interviewed were the mothers of the infants (91.7%). The mean (95% CI) age of the primary caregivers was 28.4 (27.8, 29.0) years, 80% had at least ten years of formal education (grade 10 or above) and only 10.7% were married. For mothers only ($n = 688$), 9.2% were married. The median (IQR) number of people living in the household were 5 (4, 7) with 1 (0, 1) person earning an income and 2 (1, 3) persons receiving social grants. Almost all households had a flush toilet (95.1%) and electricity (92.3%). All households had access to tap water, mostly through a tap either inside (29.3%) or outside (66.5%) the house.

The gender distribution among the infants was 51.6% males and 48.4% females. For those infants of which birth weight was available ($n = 728$), the mean (95% CI) birth weight was 2.98 (2.94, 3.02) kg. Low birth weight was recorded for 14% of the infants.

Table 2 illustrates feeding practices of the study participants. At the time of the survey, at the age of 6 months, 70.1% of the infants were still being breastfed. Breastfeeding was stopped at age 0–4 months for 25.5% of the infants. Exclusive breastfeeding was not widely practiced and 48.9% of infants were exclusively breastfed up to age 0–2 months; only 14.9% were exclusively breastfed after the age of 4 months. A high percentage (94.8%) of infants were given liquids other than breast milk, 46.4% of infants were introduced to liquids at age 0–2 months, mostly water and formula milk. Milk feeds other than breast milk, were introduced at the mean (95% CI) age of 2.3 (2.1, 2.6) months (not shown in table). In terms of milk feeds at the age of 6 months, 52.7% of infants received breast milk, but no other milk feeds, 14.9% received breast milk and formula milk feeds, and for 27.7% formula milk was the only milk feed given. At the age of 6 months, 94.1% of infants were already given semi-solid/solid foods. The semi-solid/solid foods that were introduced first were mostly commercial infant cereal (63.8%); jarred infant foods (20.3%) and maize meal porridge (8.7%). Semi-solid/solid foods were introduced at the mean (95% CI) age of 3.8 (3.6, 4.0) months. The IQR for meal frequency per day was 2–3 times, and 50.5% of the infants were fed with a bottle. A percentage of 13.4% of infants received dietary supplements.

Results from the set of questions on the frequency of foods consumed during the past seven days showed that food consumed on at least four days were mostly infant cereal (68.1%), formula milk feeds (42.0%) and jarred infant foods (22.7%). Sugar and/or salt were added to the infant's food on at least four days for 27.9% and 8.7% of the infants respectively.

Vegetables, fruits and animal-sourced foods were not frequently given (all reported by 5% or less, except for milk – 7%).

Of the 750 infants, 24-hour dietary recall was available for 715 infants; for the remaining 35 infants the primary caregiver could not provide information for a full 24 hours. Foods reported during the 24-hour recall were grouped into six groups; breast milk, formula milk feeds, infant cereals, jarred infant foods, maize meal porridge and all other foods. For consumers of each of these foods groups, dietary iron obtained from these food groups were calculated and is shown in Table 3. The data showed that, for consumers, the median (IQR) amount of iron from formula milk feeds was 3.42 (1.65, 5.86) mg and from infant cereals 3.90 (2.25, 7.65) mg.

Of the 715 infants for whom a 24-hour dietary recall was available, 36 infants were reported to consume breast milk only (with or without water as well) the day before. Information on the complementary diet was therefore available for 679 infants. The nutrient density of the complementary diet (excluding breast milk) is summarised in Table 4. Over 80% of infants did not meet the desired nutrient density for calcium (84.4%), iron (83.4%), zinc (86.0%), and over 60% of infants did not meet nutrient density for niacin (61.8%) and vitamin B6 (63.6%). The nutrient density of vitamin A was not reached by 9.1% of infants.

Anaemia, iron and anthropometric status are presented in Table 5. The mean (95% CI) Hb concentration was 11.35 (11.25, 11.45) g/dl. The mean (95% CI) concentration for Fer and TfR were 31.27 (29.22, 33.31) µg/l and 7.55 (7.31, 7.80) mg/l respectively. The prevalence of raised CRP (> 5 mg/l) and AGP (> 1 g/l) were 14.8% and 32.2% respectively. The prevalence of anaemia was 36.4% (21.7% mild and 14.7% moderate). The prevalence of ID and IDA, based on Fer were 16.1% and 9.3%, respectively after adjustment for inflammation. Based on TfR, the prevalence of ID and IDA was 30.1% and 13.2%, respectively. Almost a third (29.3%) of infants was stunted and 10.1% were overweight. Table 6 indicates a comparison between anaemic and non-anaemic infants according to total nutrient intake as reported by a single 24-hour recall. Although median energy intake did not differ significantly between the two groups, anaemic infants had significantly lower dietary intakes for iron (4.3 versus 5.9 mg; $P = 0.010$), magnesium (35 versus 42 mg; $P = 0.013$), zinc (2.40 versus 2.93 mg; $P = 0.001$), thiamine (0.41 versus 0.46 mg; $P = 0.017$), riboflavin (0.51 versus 0.59 mg; $P = 0.013$), niacin (4.1 versus 5.0 mg; $P = 0.005$), vitamin B₆ (0.263 versus 0.336 mg; $P = 0.006$), vitamin B₁₂ (0.6 versus 0.7 µg; $P = 0.027$) and vitamin C (57 versus 65 mg; $P = 0.041$). When considering the complementary diet by excluding breast milk, the anaemic group had a significantly lower

median (IQR) energy intake [1183 (680, 2004) versus 1440 (807, 2500) kJ; $P = 0.003$], but there was no significant difference in the nutrient density for any of the micronutrients (data not shown).

Table 7 indicates comparison between anaemic and non-anaemic infants when considering gender, birth weight, anthropometric status and feeding practices. Anaemia was more prevalent in male infants than in female infants (39.8% versus 32.8%, $P = 0.046$), and in stunted versus non-stunted infants (41.8% versus 34.2%, $P = 0.047$). Infants still being breastfed at the age of 6 months had a higher prevalence of anaemia when compared to those who were no longer being breastfed (40.8% versus 26.3%, $P = 0.001$). Infants for whom exclusive breastfeeding was stopped at age 0–2 months had a lower prevalence of anaemia versus infants exclusively breastfed until age 5–6 months (31.6% versus 47.3%). A lower prevalence of anaemia was found in infants who were introduced to milkfeeds at age 0–2 months versus infants who were not receiving any milk feeds (27.0% versus 43.6%). A lower prevalence of anaemia was found in infants who were introduced to liquids at age 0–2 months versus infants who were introduced to liquids at age 3–4 months (30.7% versus 45.7%). Anaemia was less prevalent in infants who frequently consumed formula milk (27.6% versus 43.5%, $P = 0.001$) and commercial infant cereal (33.8% versus 41.5%, $P = 0.042$). There was no significant differences in anaemic status observed when considering the frequency of meal times and the use of dietary supplements ($P = 0.842$ and $P = 0.298$), respectively. When comparing ID and non-ID infants, based on Fer, similar results were observed, with the exception stunting and frequent consumption of infant cereals (which showed no differences); and significant differences for mean birth weight ($P = 0.012$), prevalence of overweight ($P = 0.042$) and age of introducing semi-solid/solid food ($P = 0.030$).

For infants who had already been introduced to liquids and/or semi-solid/solid foods, Pearson correlation analysis showed a significant negative correlation between age of introduction to liquids and haemoglobin concentration ($r = -0.128$, $P = 0.001$) and Fer concentration ($r = -0.133$, $P = 0.004$), and a significant positive correlation with TfR concentration ($r = 0.128$, $P = 0.006$) which indicate that infants introduced to liquids at an early age showed better haemoglobin, Fer concentration and TfR concentration. The age of introduction to semi-solid/solid foods was significantly negatively correlated with Fer concentration ($r = -0.106$, $P = 0.024$) and significantly positively correlated with TfR ($r = 0.108$, $P = 0.021$).

According to the results of binary logistic regression analysis gender, exclusive breastfeeding and consumption of formula milk feeds showed associations with anaemia and ID status. For anaemic status: Male infants (OR = 1.388, $P = 0.037$) had a greater chance to be anaemic than female infants, while infants who were exclusively breastfed up to age 0–2 months (OR = 0.620, $P = 0.039$) and infants who frequently consumed formula milk (OR = 0.523, $P < 0.001$) had a lower chance to be anaemic than those infants who were exclusively breastfed up to age 5–6 months and those who seldom/never consumed formula milk, respectively. For ID status based on Fer: Male infants (OR = 2.432, $P = 0.002$) had a greater chance to be ID than female infants, while infants with a higher birth weight (OR = 0.417, $P = 0.005$), infants who were exclusive breastfed up to age 0–2 months (OR = 0.362, $P = 0.022$) and infants who frequently consumed formula milk (OR = 0.219; $P < 0.001$) had a lower chance to be ID, respectively. For ID status based on TfR: Male infants (OR = 2.047, $P = 0.001$) had a greater chance to be ID, and infants with a higher birth weight (OR = 0.498, $P = 0.004$) had a lower chance to be ID.

Discussion

All infants included in the study were initially breastfed as this was an inclusion criterion for enrolment into the randomized controlled trial from which the baseline data were used in this study. Although 70.1% of the infants were still being breastfed at the age of 6 months, exclusive breastfeeding up to the age of 6 months was not widely practiced, mostly because of the early introduction of liquids. Just over half of the infants were exclusively breastfed after the age of 2 months. National data collected in 2012 indicated that 7.4% of South African infants younger than 6 months were exclusively breastfed (HSRC 2013).

Most of the infants had been introduced to semi-solid or solid foods before the age of 6 months, with the most popular first foods being commercial infant cereals and jarred infant foods. National data collected in 2012 also reported early introduction of solid foods; 63.5% of infants were given semi-solid or solid foods before 6 months with an average age of introduction at 3.6 months, and the most common first food being commercial infant cereal (51.2%) (HSRC 2013). In contrast, Faber et al. (2014) reported that maize meal porridge was the most popular first food (69.6% and 59.5% in a rural and urban area in KwaZulu-Natal, respectively), with infant cereals given as first food to < 25% of the infants, indicating differences in feeding practices across diverse settings within South Africa. A systematic review reported that, in developing countries, the introduction of solids to infants at the age of 4 months showed

significant improvements in haemoglobin concentrations versus those introduced to solids at the age of 6 months (Qasem et al. 2015). However, early introduction of complementary feeding might be a risk factor for early exposure to microbial pathogens which increases the risk of infection for diarrheal disease and malabsorption and most digestive enzymes are inadequate for the first 6 months of life (WHO 2003). Also, infants, in particular those who are introduced to solid foods before the age of 4 months, are not yet prepared to digest solid foods and have an increased risk for some chronic diseases such as diabetes and obesity (Clayton et al. 2013).

At the age of 6 months, commercial infant cereals and jarred infant foods were widely consumed, even though a high number of households were recipients of social grants, which points towards the low socio-economic status of the study population. Frequent consumption of commercial baby foods such as infant cereals and jarred infant foods is of concern as several of these products have high sugar content and can thus be seen as a risk factor for future health problems (Walker & Goran 2015).

In South Africa, the legislation on fortifying maize meal with vitamins A, B₁, B₂, B₆, niacin and folic acid and minerals like iron and zinc came into effect in 2003 (DOH & UNICEF, 2007). A small percentage of infants in our study were given maize meal porridge. For consumers, the contribution of maize meal porridge to dietary iron was low (< 1 mg). In contrast, commercial infant cereals and fortified formula feeds contributed significant amounts of dietary iron for consumers of these products (3.90 mg and 3.42 mg, respectively). Yet, when considering the nutrient density of the complementary diet, which comprised mostly of commercial infant cereals, there were more than 80% of infants who did not meet the desired densities for iron, zinc and calcium. These three nutrients have been identified as problem nutrients in the complementary diet of infants in developing countries (Vossenaar & Solomons 2012; Vossenaar et al. 2013), including South Africa (Faber 2005; Faber et al. 2014). The reference values for “desired” nutrient densities should be used cautiously as there are uncertainties regarding the nutrient content of breast milk and there is no clarity on which is the most appropriate reference nutrient intakes to use when developing recommendations for nutrient densities (Dewey & Brown 2003). Also, when using a single 24-hour dietary recall, the nutrient distributions are usually very wide, resulting in an overestimation of inadequate nutrient intake (Murphy et al. 2006).

Although the micronutrient nutrient density of the complementary diet did not differ between the anaemic and non-anaemic infants, when considering total nutrient intake, non-anaemic infants had a higher intake for most of the micronutrients, probably because of the higher number of infants who were given fortified infant formula feeds. For infants who were receiving milk feeds, these milk feeds (mostly fortified formula milk) were introduced mostly at age 0–2 months. A possible explanation for the lower prevalence of anaemia in infants who were exclusively breastfed up to only age 0–2 months might be that infants were introduced to fortified formula milk and infant cereal from an early age. At the age of 6 months, the prevalence of anaemia was significantly lower in infants who were frequently given formula milk. This finding corresponds to studies conducted in Europe, China and South Africa, which reported lower prevalence of anaemia in formula fed infants (Eussen et al. 2015; Luo et al. 2014; Faber 2007). Breast milk contains highly bioavailable iron, but in a low concentration and it is recommended that healthy birth weight infants must be supplied with iron at the age of 6 months (WHO 2009).

Various factors other than feeding practices may affect anaemia and iron status, such as premature birth, parasitic infection and inflammation (Pasricha et al. 2013; Shinoda et al. 2011). The anaemia and iron-status of infants aged 6–8 months should also be interpreted within context of the infant's iron stores at birth. Iron status during the first six months of life is dependent on iron stores at birth and is primarily dependant on birth weight, maternal prenatal iron status and the timing of clamping of the umbilical cord (Dewey & Chaparro 2007). A recent study provided evidence of the potential benefits of iron supplementation after the age of 4 months for infants with depleted iron stores at birth (Greer 2015). We did not observe a significant difference in birth weight between anaemic and non-anaemic infants which concurs with findings from a study conducted in China (Luo et al. 2014). To the contrary, Faber (2007) found that in 6–12 month old infants in a rural community of South Africa low birth weight infants had a higher chance of being anaemic (Faber 2007). Our study did however show that ID infants had a lower birth weight compared to non-ID infants.

Gender was significantly associated with both anaemia and ID, with male infants having a bigger chance to be anaemic or iron deficient. Higher anaemia prevalence in male infants was also found in studies conducted in Ethiopia and Armenia (Woldie et al. 2015; Demirchyan et al. 2015).

Stunted children had a higher prevalence of anaemia compared to non-stunted children. This finding is in agreement with results from infant studies conducted in urban areas of Philippines and rural areas of China where stunting was a strong independent factor associated with anaemia in study infants (Rohner et al. 2013; Yang et al. 2012)

Globally, South Africa has the highest prevalence of HIV in women and the second highest prevalence of HIV in children younger than 5 years (UNICEF 2014). A systematic review done on HIV-associated anaemia in children showed that mild and moderate anaemia was more prevalent with HIV infection (Calis et al. 2008). Although we did not record the HIV status for the infants in our study, there is the possibility that HIV could have contributed to the anaemia prevalence among our study participants, although to a limited extent. The prevalence of HIV in infants may be smaller than expected as evaluation of the national prevention of mother-to-child transmission (MTCT) programme showed that 91.7% of HIV-positive mothers received antiretroviral treatment or prophylaxis and a MTCT rate of 3.5% during pregnancy and intrapartum (Goga et al. 2012).

Regardless of the fact that our study was cross-sectional and observed associations may not provide evidence of causality, information on risk factors and general direction of observed relationships that may contribute to anaemia and ID are indicated. The advantage of this cross-sectional study is that data was collected from a relatively large sample ($n = 750$) within a very narrow age bracket which may reflect relevant associations. Hb results are based on either finger prick or venous samples. Alwan et al. (2013), and Neufeld et al. (2002) found raised Hb from finger prick which may result in excess false negative diagnoses among individuals. However, Simmonds et al. (2011) concluded that in the absence of a venous sample, a capillary sample via finger prick can be used as a reliable alternative in field settings. For the infants in our study, we compared the Hb values obtained from capillary versus venous samples and results showed that there was no significant difference ($P = 0.657$) between the two groups. Although only 485 infants had the full evaluation for ID and IDA, the Hb values and anaemia prevalence derived thereof from this study will remain useful at pragmatic level. Relationship of the inflammatory markers measured to duration of breastfeeding were not determined which could give a bigger perspective to explain the anaemia and ID prevalence.

In conclusion, our results indicated that infant feeding practises in this study population are characterised by frequent consumption of commercial infant foods, of which many are fortified with iron. If the infants were given mostly maize meal porridge or other unfortified foods from

an early age, our results may have looked different. Nutrient density of the complementary diet was low for especially the “problem nutrients” iron, zinc and calcium for most of the infants. Compared to non-anaemic infants, anaemic infants had lower dietary intakes for various nutrients, but nutrient density of the complementary diet did not differ. In this study we observed that frequent consumption of formula milk may have a protective effect on anaemia and ID in 6 month old infants. As adequate iron status is critical for healthy infant development (Hermoso et al. 2011; Prado & Dewey 2014) the prevalence of anemia and iron deficiency within the context of current feeding practices within this age group warrant further investigation.

Key messages

1. Duration of exclusive breastfeeding was short; 48.9% of the infants were exclusively breastfed up to age 0–2 months
2. A high percentage of infants consumed commercial infant cereal and jarred infant food frequently (68.1% and 22.7%, respectively)
3. Nutrient density of the complementary diet indicated was below the desired density for iron, zinc and calcium for more than 80% of infants.
4. Anaemia, ID and IDA prevalence was 36.4%, 16.1% and 9.3% respectively.
5. Early introduction of fortified formula milk and commercial infant cereal seemed to have a protective effect on anaemia and iron deficiency in this study population

References

Alwan AG, Yaqoub YI & Abbas MG (2013) Comparison of the result of some hematological parameters in venous and capillary blood samples. *DJM* 5, 90-94.

Baker R.D. & Greer F.R. (2010) Diagnosis and prevention of iron deficiency and iron-deficiency anemia in infants and young children (0–3 years of age). *Pediatrics* 126(5), 1040–1050.

Benoist B.D., McLean E., Egll I. & Cogswell M. (2008) *Worldwide prevalence of anaemia 1993-2005: WHO global database on anaemia*. World Health Organization: Geneva.

Bentley M.E., Johnson S.L., Wasser H., Creed-Kanashiro H., Shroff M., Fernandez Rao S. et al. (2014) Formative research methods for designing culturally appropriate, integrated

child nutrition and development interventions: an overview. *Annals of the New York Academy of Sciences* 1308(1), 54–67.

Calis J.C, van Hensbroek M.B., de Haan R.J., Moons P., Brabin, B.J., Bates I. (2015) HIV-associated anemia in children: a systematic review from a global perspective. *AIDS* 22(10), 1099–1112.

Cherop E.C., Etyyang G.A. & Mbagaya G.M. (2010) Mixed feeding among infants aged 0-6 months in an urban setting of Eldoret, Kenya. *Journal of Food, Agriculture & Environment* 8 (1), 59–60.

Clark S.F. (2008) Iron deficiency anemia. *Nutrition in Clinical Practice* 23(2), 128–141.

Clayton H.B., Li R., Perrine C.G. & Scanlon K.S. (2013) Prevalence and reasons for introducing infants early to solid foods: Variations by milk feeding type. *Pediatrics* 131, e1108–e1114.

Demirchyan A., Petrosyan V., Sargsyan V. & Hekimian K. (2015) Prevalence and determinants of anaemia among children aged 0–59 months in a rural region of Armenia: a case–control study. *Public Health Nutrition* doi: <http://dx.doi.org/10.1017/S1368980015002451>.

Department of Health (DOH) South Africa & UNICEF South Africa. (2007) *A reflection of the South African maize meal and wheat flour fortification programme*. South African Department of Health: Pretoria.

Dewey K.G. & Brown K.H. (2003) Update on technical issues concerning complementary feeding of young children in developing countries and implications for intervention programs. *Food and Nutrition Bulletin* 24(1), 5–28.

Dewey K.G. & Chaparro C.M. (2007) Session 4: mineral metabolism and body composition iron status of breast-fed infants. *Proceedings of the Nutrition Society* 66 (03), 412–422.

Doherty T., Sanders D., Jackson D., Swanevelder S., Lombard C., Zembe W. *et al.* (2012) Early cessation of breastfeeding amongst women in South Africa: an area needing urgent attention to improve child health. *BMC Pediatrics* 12 (105), 1–10.

Du Plessis L.M., Kruger H. & Sweet L. (2013) Complementary feeding: a critical window of opportunity from six months onwards. *South African Journal of Clinical Nutrition* 26(3), S129–S140.

Dube K., Schwartz J., Mueller M.J., Kalhoff H. & Kersting M. (2010) Iron intake and iron status in breastfed infants during the first year of life. *Clinical Nutrition* 29(6), 773–778.

Erhardt J.G., Estes J.E., Pfeiffer C.M., Biesalski H.K. & Craft N.E. (2004) Combined measurement of ferritin, soluble transferrin receptor, retinol binding protein, and C-reactive protein by an inexpensive, sensitive, and simple sandwich enzyme-linked immunosorbent assay technique. *The Journal of Nutrition* 134(11), 3127–3132.

Eussen S., Alles M., Uijterschout L., Brus F. & van der Horst-Graat J. (2015) Iron intake and status of children aged 6–36 month in Europe: A systematic review. *Annals of Nutrition and Metabolism*, doi: 10.1159/000371357.

Faber M. (2005) Complementary foods consumed by 6-12-month-old rural infants in South Africa are inadequate in micronutrients. *Public Health Nutrition* 8(4), 373–381.

Faber M. (2007) Dietary intake and anthropometric status differ for anaemic and non-anaemic rural South African infants aged 6-12 months. *Journal of Health Population and Nutrition* 3, 285–293.

Faber M., Kvalsvig J.D., Lombard C.J. & Benadé A.S. (2005) Effect of a fortified maize-meal porridge on anemia, micronutrient status, and motor development of infants. *The American Journal of Clinical Nutrition* 82(5), 1032–1039.

Faber M., Laubscher R. & Berti C. (2014) Poor dietary diversity and low nutrient density of the complementary diet for 6-to 24-month-old children in urban and rural KwaZulu-Natal, South Africa. *Maternal & Child Nutrition*. 2014 Aug 19. doi: 10.1111/mcn.12146. [Epub ahead of print]

Finkelstein J.L., O'Brien K.O., Abrams S.A. & Zavaleta N. (2013) Infant iron status affects iron absorption in Peruvian breastfed infants at 2 and 5 mo of age. *The American Journal of Clinical Nutrition* 98(6), 1475–1484.

Goga A.E., Dinh T.H., Jackson D.J. for the SAPMTCTE study group. (2012) *Evaluation of the Effectiveness of the National Prevention of Mother-to-Child Transmission (PMTCT)*

- Programme Measured at Six Weeks Postpartum in South Africa, 2010*. South African Medical Research Council: Cape Town.
- Greer F.R. (2015) How much iron is needed for breastfeeding infants? *Current Pediatric Reviews* 11(4), 298-304.
- Hermoso M., Vollhardt C., Bergmann K. & Koletzko B. (2011) Critical micronutrients in pregnancy, lactation, and infancy: considerations on vitamin D, folic acid, and iron, and priorities for future research. *Annals of Nutrition and Metabolism* 59(1), 5–9.
- Human Sciences Research Council (HSRC). 2013. *The South African National Health and Nutrition Examination Survey (SANHANES-1). Data analysis on infant feeding practices, and anthropometry in children under five years of age: South Africa 2012*. Report for UNICEF, November 2013.
- International Food Policy Research Institute (IFPRI). (2014) *Global Nutrition Report 2014: Actions and Accountability to Accelerate the World's Progress on Nutrition*. Washington, DC.
- International Food Policy Research Institute (IFPRI). (2015) *Global Nutrition Report (2015) Actions and Accountability to Advance Nutrition and Sustainable Development*: Washington DC.
- Kalanda B., Verhoeff F. & Brabin B. (2006) Breast and complementary feeding practices in relation to morbidity and growth in Malawian infants. *European Journal of Clinical Nutrition* 60(3), 401–407.
- Labadarios D. (ed). (2007) *National Food Consumption Survey – Fortification Baseline (NFCS-FB): South Africa, 2005*. Department of Health: Pretoria.
- Ladzani R., Peltzer K., Mlambo M.G. & Phaweni K. (2011) Infant-feeding practices and associated factors of HIV-positive mothers at Gert Sibande, South Africa. *Acta Paediatrica* 100(4), 538–542.
- Langenhoven M., Conradie P., Wolmarans P. & Faber M. (1991) *MRC food quantities manual*. Cape Town: South African Medical Research Council.
- Lönnerdal B. (2003) Nutritional and physiologic significance of human milk proteins. *The American Journal of Clinical Nutrition* 77(6), 1537S–1543S.

Lozoff B., Kaciroti N. & Walter T. (2006). Iron deficiency in infancy: applying a physiologic framework for prediction. *The American Journal of Clinical Nutrition* 84, 1412–1421.

Luo R., Shi Y., Zhou H., Yue A., Zhang L., Sylvia S. *et al.* (2014) Anemia and feeding practices among infants in rural Shaanxi Province in China. *Nutrients* 6(12), 5975–5991.

Marfell-Jones M., Stewart A. & De Ridder H. (2012) *ISAK accreditation handbook*. Upper Hutt, New Zealand: International Society for the Advancement of Kinanthropometry.

Murphy S.P., Guenther P.M. & Kretsch M.J. (2006) Using the Dietary Reference Intakes to assess intakes of groups: pitfalls to avoid. *Journal of the American Dietetic Association* 106, 1550–1553.

Neufeld L, García-Guerra A, Sánchez-Francia D *et al.* (2002) Hemoglobin measured by Hemocue and a reference method in venous and capillary blood: a validation study. *Salud Pública México* 44, 219–227.

Pasricha S.R., Hayes E., Kalumba K. & Biggs B.A. (2013) Effect of daily iron supplementation on health in children aged 4–23 months: a systematic review and meta-analysis of randomised controlled trials. *The Lancet Global Health* 1, e77–e86.

Prado E.L. & Dewey K.G. (2014) Nutrition and brain development in early life. *Nutrition Reviews* 72(4), 267–284.

Qasem W., Fenton T. & Friel J. (2015) Age of introduction of first complementary feeding for infants: a systematic review. *BMC Pediatrics* 15(1), 107.

Raj S., Faridi M., Rusia U. & Singh O. (2008) A prospective study of iron status in exclusively breastfed term infants up to 6 months of age. *International Breastfeeding Journal* 3(3), 1–7.

Rohner F., Bradley A., Woodruff G.J., Aaron E.A, Yakes M.A.O., Lebanan P.R.S. *et al.* (2013) Infant and young child feeding practices in urban Philippines and their associations with stunting, anemia, and deficiencies of iron and vitamin A. *Food and Nutrition Bulletin* 34(2), S17–S34.

Shinoda N., Sullivan K.M., Tripp K., Erhardt J.G., Haynes B.M.H., Temple V.J. *et al.* (2011) Relationship between markers of inflammation and anaemia in children of Papua New Guinea. *Public Health Nutrition*, doi:10.1017/S1368980012001267.

Simmonds MJ, Baskurt OK, Meiselman HJ et al. (2011) A comparison of capillary and venous blood sampling methods for the use in haemorheology studies. *Clin Hemorheol Micro*, 47, 111–119.

Smuts C.M., Dhansay M.A., Faber M., van Stuijvenberg M.E., Swanevelder S., Gross R. *et al.* (2005) Efficacy of multiple micronutrient supplementation for improving anemia, micronutrient status, and growth in South African infants. *The Journal of Nutrition* 135(3), 653S–659S.

Thurnham D.I., Northrop-Clewes C.A. & Knowles J. (2015) The use of adjustment factors to address the impact of inflammation on vitamin A and iron status in humans. *The Journal of Nutrition* 145(5), 1137S–1143S.

UNICEF. (2014) *Statistical tables: Economic and social statistics on the countries and areas of the world, with particular reference to children's well-being*. New York, USA.

Vossenaar M. & Solomons N.W. (2012) The concept of "critical nutrient density" in complementary feeding: the demands on the "family foods" for the nutrient adequacy of young Guatemalan children with continued breastfeeding. *American Journal of Clinical Nutrition* 95(4), 859–866.

Vossenaar M., Hernández L., Campos R., & Solomons N.W. (2013) Several 'problem nutrients' are identified in complementary feeding of Guatemalan infants with continued breastfeeding using the concept of 'critical nutrient density'. *European Journal of Clinical Nutrition* 67(1), 108–114.

Walker R. W. & Goran M. I. (2015) Laboratory determined sugar content and composition of commercial infant formulas, baby foods and common grocery items targeted to children. *Nutrients* 7, 5850-5867.

Wang Y. & Chen H.J. (2012) Use of percentiles and z-scores in Anthropometry. In: Preedy, V.R (ed). *Handbook of Anthropometry. Physical Measures of human form in health and disease*, ISBN: 978-1-4419-1787-4.

WHO (World Health Organization). (1998). *Complementary feeding of young children in developing countries. A review of current scientific knowledge*. WHO/NUT/98.1 World Health Organization: Geneva.

WHO (World Health Organization). (2001) *Iron deficiency anaemia: Assessment, prevention and control. A guide for programme managers*. World Health Organization: Geneva.

WHO (World Health Organization). (2001) *The optimal duration of exclusive breastfeeding: Report of an expert consultation*. World Health Organization: Geneva.

WHO (World Health Organization). (2003) *Guiding principles for complementary feeding of the breastfed child*. Washington: Pan American Health.

WHO (World Health Organization). (2006) *Child growth standards. Length/height-for-age, weight-for age, weight-for-length, weight-for-height and body mass index for age. Methods and development*. World Health Organization: Geneva.

WHO (World Health Organization). (2009) *Infant and young child feeding: model chapter for textbooks for medical students and allied health professionals*. World Health Organization: Geneva.

WHO (World Health Organization). (2010) *Indicators for assessing infant and young child feeding practices. Part 2: Measurement*. World Health Organization: Geneva.

WHO (World Health Organization). (2011a) *Haemoglobin concentrations for the diagnosis of anaemia and assessment of severity*. Vitamin and Mineral Nutrition Information System. Geneva, Switzerland.

WHO (World Health Organization). (2011b) *Serum ferritin concentrations for the assessment of iron status and iron deficiency in populations*. Vitamin and Mineral Nutrition Information System. Geneva, Switzerland.

WHO (World Health Organization). (2011c) *Optimal feeding of low birth weight infants in low and middle income countries*. World Health Organization: Geneva.

Woldie H., Kebede Y. & Tariku A. (2015) Factors associated with anemia among children aged 6–23 months attending growth monitoring at Tsitsika Health Center, Wag-Himra Zone, Northeast Ethiopia. *Journal of Nutrition and Metabolism* 2015, 1–9.

Wolmarans P., Danster N., Dalton A., Rossouw K. & Schönfeldt H. (2010) *Condensed Food Composition Tables for South Africa*. Medical Research Council: Cape Town.

Yang W., Li X., Li Y., Zhang S., Liu L., Wang X. (2012) Anemia, malnutrition and their correlations with socio-demographic characteristics and feeding practices among infants aged 0–18 months in rural areas of Shaanxi province in northwestern China: a cross-sectional study. *BMC Public Health* 12, 11

Table 1. Socio-economic characteristics of the primary caregivers (n=750) and characteristics of the infants

<i>Relationship with child (%)</i>	
Mother	91.7
Grandmother	4.3
Other (father, aunt, not-related)	4.0
Age, years (Mean, 95% CI)	28.4 (27.8, 29.0)
Level of education (%)	
Lower than grade 10/Form III/NTC 1	20.0
grade 10/Form III/NTC 1	20.0
grade 11/Form IV/NTC II	30.1
grade 12/Form V/NTC III	24.8
Higher than grade 12/Form V/NTC III	5.2
Marital status (%)	
Unmarried	55.3
Married	10.7
Living together	28.3
Common-law wife	3.5
Other (divorced, widow)	2.2
<i>Household characteristic (n=750)</i>	
Number of people in household (median, IQR)	5 (4,7)
Number of people in household earning an income (median, IQR)	1 (0,1)
Number of people in household receiving a social grant (median, IQR)	2 (1,3)
Access to a flush toilet (%)	95.1
Access to water (%)	
Tap inside the dwelling	29.3
Tap outside the dwelling	66.5
Public tap	4.1
Electricity in the dwelling (%)	92.3
<i>Infant characteristics (n= 750)</i>	
Age range in months	5.85–7.01
Gender (%)	
male	51.6
female	48.4
Birth weight, kg (Mean, 95%CI)	2.98 (2.94, 3.02)
% Low birth weight (< 2.50 kg)	14

IQR = interquartile range

Table 2. Feeding practices for 6 month old infants (n=750)

	Age intervals when stopped/started (%)			
	%	0–2 mo	3–4 mo	5–6 mo
Still being breastfed	70.1			
No longer being breastfed	29.1	15.2	10.3	3.6
Duration of exclusive breastfeeding		48.9	36.1	14.9 ¹
<i>Type of milk feed</i>				
Breast milk ¹	52.7			
Breast milk and formula milk	14.9			
Breast milk and cow's milk	2.4			
Formula milk	27.7			
Cow's milk	1.1			
No milk	1.2			
Infant is given milk feeds ²	45.6	21.4	15.2	9.0
Infant is given liquids ³	94.8	46.4	33.5	14.9
Infant is given semi-solid/solid ⁴ foods ⁴	94.1	16.6	43.9	33.6
Fed with a bottle/teat	50.5			
Receive dietary supplements	13.4			
Meals per day (median, IQR)	3 (2, 3)			
<i>Food items consumed at least 4 days during the past week</i>				
Breast milk	70.1			
Infant cereal	68.1			
Formula milk feeds	42.0			
Sugar	27.9			
Jarred infant food	22.7			
Rooibos tea	11.5			
<i>Foods consumed at least 4 days per week by 5–10% of the infants</i>				
Maize meal porridge; Instant maize meal porridge; Salt; Milk; cooked porridge, other than maize meal; Potato; Yoghurt; Fruit juice; Margarine				
<i>Foods consumed at least 4 days per week by < 5% of the infants</i>				
Salty snacks e.g. crisps; Vegetables; Fruit; Fish; Cordial drink; Chicken; Egg; Tea; Oil; Peanut butter; Sweets; Fizzy drinks; Rice; Bread; Liver; Meat				

¹ Including the 14 (1.9%) of infants who were still being exclusively breastfed at the time of the survey; ² Type of milk feeds (n= 344): Formula milk 92.4%; Cow's milk 7.6%; ³ Liquids introduced first (n= 713): Water 53.6%; Formula milk 39.1%; Other (rooibos tea, sweetened drink, sugar water, cow's milk) 7.3%; ⁴ Foods introduced first (n= 701): Commercial infant cereal 63.8%; Jarred infant foods 20.3%; Maize meal porridge 8.7%; Other (mabele / oats porridge, vegetables) 7.2%

Table 3. Amount of iron (mg) contribution from main reported food groups, for consumers only

Food group	n	Iron (mg) (median, IQR)
Breastmilk	510	0.20 (0.20, 0.20)
Formula milk	309	3.42 (1.65, 5.86)
Infant cereal	503	3.90 (2.25, 7.65)
Jarred infant foods	196	0.26 (0.16, 0.43)
Maize meal porridge	165	0.76 (0.32, 1.39)
Other	469	0.27 (0.06, 0.81)

IQR = interquartile range

Table 4. Nutrient density (per 100 kcal) of the complementary diet (excluding breast milk) and percentage of infants with an intake below desired density

	Desired density ¹	Nutrient density (Median, IQR) (n=679)	% of infants below the desired density
Energy (kJ)		1354 (754; 2337)	
Total fat (g)		2.50 (1.30; 3.74)	
Carbohydrate (g)		16.41 (13.92; 18.88)	
Protein (g)	1.0	2.23 (1.85, 2.96)	1.26
Calcium (mg)	105	65.19 (43.98; 84.07)	84.4
Iron (mg)	4.5	1.418 (0.993; 2.271)	83.4
Magnesium (mg)	–	8.19 (4.53; 13.31)	
Zinc (mg)	1.1	0.602 (0.347; 0.885)	86.0
Vitamin A (µg RE)	31	92.97 (60.03; 139.44)	9.1
Thiamine (mg)	0.08	0.118 (0.089; 0.156)	17.3
Riboflavin (mg)	0.08	0.123 (0.076; 0.171)	36.8
Niacin (mg)	1.5	1.073 (0.858; 1.566)	61.8
Vitamin B6 (mg)	0.12	0.090 (0.070; 0.121)	63.6
Vitamin B12 (µg)	–	0.141 (0.042; 0.211)	
Vitamin C (mg)	1.5	11.13 (7.95; 15.79)	5.2

¹ WHO (2002) reference values for adequate nutrient density (Dewey and Brown 2003)
n= 679

Table 5. Anaemia-, iron-, CRP- and AGP and anthropometric status of infants

Anaemia status (n= 750)	
Hb concentration (g/dl) (mean, 95%CI)	11.35 (11.25, 11.45)
Anaemic Hb (< 11 g/dl) (%)	36.4
Mild Anaemic (Hb 10.0–10.9 g/dl) (%)	21.7
Moderate Anaemic (Hb 7.0–9.9 g/dl) (%)	14.7
Iron status (n= 485)	
Plasma ferritin (µg/l) (mean, 95% CI) ¹	31.27 (29.22, 33.31)
Transferrin receptor (mg/l) (mean, 95% CI)	7.55 (7.31, 7.80)
CRP > 5 mg/l (%)	14.8
AGP > 1 g/l (%)	32.2
Iron deficiency (%)	
Fer < 12 µg/l (%) ²	16.1
Hb < 11 g/dl+Fer < 12 µg/l (%) ³	9.3
TfR > 8.3 mg/l (%) ²	30.1
Hb < 11 g/l and TfR > 8.3 mg/l (%) ³	13.2
Anthropometry (n=750)	
Stunted ⁴ (%)	29.3
Overweight ⁵ (%)	10.1

¹ after adjustment for inflammation; ² ID; ³ IDA; ⁴ Length-for-age Z-score < -2;

⁵ Weight-for-length Z-score > 2

Table 6. Total nutrient intake for anaemic and non-anaemic infants (median, IQR)

	Total nutrient intake ¹			<i>P</i> ²
	Total group (n=715)	Anaemic (n=265)	Non-Anaemic (n=450)	
Energy (kJ)	2856 (2421; 3365)	2829 (2437; 3392)	2875 (2404; 3358)	0.859
Total fat (g)	32.6 (29.9; 36.2)	33.0 (30.4; 35.5)	32.4 (29.6; 36.4)	0.112
Carbohydrate (g)	85.5 (69.7; 106.1)	83.9 (68.3; 107.9)	86.1 (70.1; 105.8)	0.293
Protein (g)	12.7 (9.9, 16.9)	12.5 (9.2, 16.7)	12.8 (10.1, 16.9)	0.265
Calcium (mg)	361 (274; 468)	361 (269; 461)	360 (280; 475)	0.717
Iron (mg)	5.6 (2.3; 9.5)	4.3 (1.9; 8.5)	5.9 (2.5; 9.7)	0.010
Magnesium (mg)	40 (26; 61)	35 (23; 57)	42 (27; 64)	0.013
Zinc (mg)	2.68 (1.64; 4.37)	2.40 (1.42; 3.70)	2.93 (1.74; 4.74)	0.001
Vitamin A (µg RAE)	634 (504; 821)	609 (496; 792)	647 (508; 829)	0.244
Thiamin (mg)	0.45 (0.27; 0.65)	0.41 (0.26; 0.61)	0.46 (0.30; 0.68)	0.017
Riboflavin (mg)	0.56 (0.38; 0.85)	0.51 (0.36; 0.77)	0.59 (0.39; 0.90)	0.013
Niacin (mg)	4.8 (2.9; 7.2)	4.1 (2.6; 6.9)	5.0 (3.0; 7.4)	0.005
Vitamin B6 (mg)	0.353 (0.201; 0.551)	0.263 (0.146; 0.467)	0.336 (0.149; 0.561)	0.006
Vitamin B12 (µg)	0.7 (0.4; 1.1)	0.6 (0.4; 0.9)	0.7 (0.4; 1.2)	0.027
Vitamin C (mg)	61 (44; 81)	57 (43; 80)	65 (45; 84)	0.041

¹The South African food composition tables (Wolmarans et al. 2010) was used as guideline for the number of decimals to report for each nutrient.

² Mann-Whitney U test

IQR = interquartile range

Table 7. Comparison of anaemic and non-anaemic infants regards birth weight, gender, anthropometry and early feeding practices

	Anaemic (n=273)	Non- Anaemic (n=476)	<i>P</i> ¹	ID ² (n=78)	Non-ID ² (n=407)	<i>P</i> ¹
<i>Gender (%)</i>			0.046			0.012
Male	39.8	60.2		20.1	79.9	
Female	32.8	67.2		11.7	88.3	
<i>Birth weight</i>						
Mean (95% CI)	2.96 (2.89, 3.02)	2.99 (2.95, 3.04)	0.325	2.87 (2.78, 2.97)	3.02 (2.98, 3.07)	0.012
≥ 2.50 kg	35.6	64.4	0.899	16.0	84.0	0.414
< 2.50 kg	36.3	63.7		20.4	79.6	
<i>Anthropometry</i>						
Stunted	41.8	58.2	0.047	20.7	79.3	0.077
Non-stunted	34.2	65.8		14.2	85.8	
Overweight	40.8	59.2	0.401	26.7	73.3	0.042
Non-overweight	35.9	64.1		15.0	85.0	
<i>Breastfeeding at age 6 months</i>			0.001			0.001
Still breastfeeding (%)	40.8	59.2		21.6	78.4	
Not breastfed (%)	26.3	73.7		4.5	95.5	
<i>Age of stopping exclusive breastfeeding</i>			0.007 ³			0.001 ³
0–2 months	31.6	68.4		10.6	89.4	
3–4 months	38.4	61.6		15.6	84.4	
5–6 months	47.3	52.7		37.5	62.5	
<i>Age of introduction of milkfeeds (%)</i>			<0.001 ³			<0.001 ⁴
0–2 months	27.0	73.0		2.0	98.0	
3–4 months	30.1	69.9		2.8	97.2	
5–6 months	29.9	70.1		14.0	86.0	
<i>Age of introduction of liquids (%)</i>			0.011 ³			<0.001 ³
0–2 months	30.7	69.3		10.1	89.9	
3–4 months	38.3	61.7		15.9	84.1	
5–6 months	45.7	54.3		33.3	66.7	
<i>Age of introduction of semi-solid/solid foods (%)</i>			0.181			0.030 ⁵
0–2 months	34.1	65.9		11.4	88.6	
3–4 months	33.1	66.9		12.6	87.4	
5–6 months	41.0	59.0		22.8	77.2	

	Anaemic (n=273)	Non- Anaemic (n=476)	<i>P</i> ¹	ID ² (n=78)	Non-ID ² (n=407)	<i>P</i> ¹
Median feeding times per day (interquartile range)	3 (2.75, 3.00)	3.00 (2.00, 3.00)	0.842	3 (2, 4)	3 (2, 4)	0.423
Received dietary supplements (%)	40	60	0.298	18.5	81.5	0.780
<i>Food items consumed the previous week</i>						
Formula milk						
≥4 days per week	26.7	73.3	0.001	5.5	94.5	0.001
seldom/never	43.5	56.5		24.0	76.0	
Jarred infant foods						
≥4 days per week	36.3	63.7	0.969	15.0	85.0	0.680
seldom/never	36.5	63.5		16.6	83.4	
Infant cereal						
≥4 days per week	33.8	66.2	0.042	15.1	84.9	0.335
seldom/never	41.5	58.5		18.6	81.4	
Rooibos tea						
≥4 days per week	37.6	62.4	0.805	17.0	83.0	0.878
seldom/never	36.3	63.7		16.2	83.8	
Sugar						
≥4 days per week	38.2	61.8	0.543	19.4	80.6	0.260
seldom/never	35.8	64.2		15.1	84.9	

¹ Pearson Chi-Square for categorical data; Analysis of Variance for continuous data

² Based on Fer

³ Bonferroni test indicated differences ($p < 0.05$) between age groups 0–2 months and 3–4 months

⁴ Bonferroni test indicated differences ($p < 0.05$) between age groups 3–4 months and 5–6 months

⁵ Bonferroni test indicated differences ($p < 0.05$) between age groups 0–2 months and 5–6 months

Supplementary Appendix

Table 8. Bivariate relationships between anaemia and various potential associated factors

	B	S.E.	Wald	df	Sig.	Exp(B)	95% C.I. for EXP(B)	
							Lower	Upper
Gender (male) ¹	0.328	0.157	4.348	1	0.037	1.388	1.020	1.888
Exclusive breastfeeding stopped at age 0–2 months ²	–0.478	0.232	4.257	1	0.039	0.620	0.393	0.976
Exclusive breastfeeding stopped at age 3–4 months ²	–0.218	0.235	0.854	1	0.355	0.804	0.507	1.276
Frequent intake of commercial infant cereal ³	–0.164	0.171	0.919	1	0.338	0.849	0.607	1.187
Frequent intake of formula milk ³	–0.630	0.170	13.759	1	<0.001	0.532	0.382	0.743
Constant	–0.071	0.224	0.101	1	0.750	0.931		

Reference categories ¹female; ²5–6 months; ³intake <4 days per week/never

Table 9. Bivariate relationships between ID based on Fer and various potential associated factors

	B	S.E.	Wald	df	Sig.	Exp(B)	95% C.I. for EXP(B)	
							Lower	Upper
Gender(male) ¹	0.889	0.286	9.677	1	0.002	2.432	1.389	4.257
Exclusive breastfeeding stopped at age 0-2 months ²	-1.016	0.442	5.269	1	0.022	0.362	0.152	.862
Exclusive breastfeeding stopped at age 3-4 months ²	-0.559	0.419	1.774	1	0.183	0.572	0.251	1.301
Semi-solid/solid foods introduced at age 0-2 months ³	-0.538	0.589	0.835	1	0.361	0.584	0.184	1.852
Semi-solid/solid foods introduced at age 3-4 months ³	-0.390	0.500	0.608	1	0.435	0.677	0.254	1.805
Semi-solid/solid foods introduced at age 5-6 months ³	-0.653	0.362	3.267	1	0.071	0.520	0.256	1.057
Frequent intake of formula milk ⁴	-1.518	0.356	18.188	1	<0.001	0.219	0.109	0.440
Birth weight (kg)	-0.874	0.313	7.779	1	0.005	0.417	0.226	0.771
Constant	1.898	0.969	3.837	1	0.050	6.671		

Reference categories ¹female; ²Exclusive breastfeeding stopped at age 5–6 months; ³Not being introduced yet; ⁴Intake < 4 days a week/never

Table 10. Bivariate relationships between ID based on TfR and various potential associated factors

	B	S.E.	Wald	df	Sig.	Exp(B)	95% CI for EXP (B)	
							Lower	Upper
Exclusive breastfeeding 0–2 ¹	-.320	.372	.742	1	.389	.726	0.350	1.505
Exclusive breastfeeding 3–4	-.133	.366	.131	1	.717	.876	0.427	1.796
Gender (male) ²	.717	.221	10.552	1	.001	2.047	1.329	3.155
Semi-solid/solid foods introduced at age 0-2 months ³	-.046	.487	.009	1	.924	.955	0.367	2.482
Semi-solid/solid foods introduced at age 3-4 months ³	-.604	.387	2.440	1	.118	.547	0.256	1.166
Semi-solid/solid foods introduced at age 5-6 months ³	-.262	.276	.901	1	.342	.770	0.770	0.448
Birth weight (kg)	-.696	.245	8.092	1	.004	.498	0.308	0.805
Frequent intake of formula milk (\geq 4 days a week)	-.972	.236	16.897	1	<0.001	.378	0.238	0.601
Constant	1.616	.773	4.373	1	.037	5.034		

Reference categories ¹Exclusive breastfeeding stopped at age 5–6 months; ²female; ³Not being introduced yet; ⁴Intake < 4 days a week/never

CHAPTER 4

This manuscript is formatted according to guidelines of Maternal and Child Nutrition (see Annexure 2), with exception of the line numbers which will be inserted and the line spacing which will be changed to double spacing before submission.

The association of psychomotor development with feeding practices and nutritional status of 6-month-old infants in a peri-urban community of South Africa.

Marinel Rothman¹, Mieke Faber², Namukolo Covic³, Tonderayi M Matsungo¹, Marike Cockeran⁴, Jane D Kvalsvig⁵, Cornelius M Smuts¹.

¹ Centre of Excellence for Nutrition (CEN), Faculty of Health Sciences, North-West University, Potchefstroom Campus, Private Bag x6001, Potchefstroom 2520, South Africa.

² Non-communicable Diseases Research Unit, South African Medical Research Council (SAMRC), PO Box 19070, Tygerberg 7505, South Africa.

³ Poverty Health and Nutrition Division, International Food Policy Research Institute, P O Box 5689, Addis Ababa, Ethiopia.

⁴ Medicine Usage in South Africa (MUSA), North-West University, Potchefstroom Campus, Private Bag x6001, Potchefstroom 2520, South Africa.

⁵ Department of Public Health Medicine, University of KwaZulu-Natal, Durban 4041, South Africa

Corresponding author: CM Smuts; Private Bag X6001, Potchefstroom 2520, South Africa; Tel +27-18-2992086; Fax +27-18-2992464; Email marius.smuts@nwu.ac.za

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Authors' contributors:

MR: One of the study-coordinators of the Tswaka study, supervising field data collection and data quality control of feeding practices and psychomotor development assessments; data analysis and interpretation of results; drafted the paper.

MF: Co-principle investigator of the Tswaka study; training, guidance on data collection, quality control and analysis of feeding practices; academic input and revision of paper.

NC: Provided training, guidance, quality control and direction on data analysis on psychomotor development assessment; revision of paper

TMM: One of the study coordinators of Tswaka trial, supervising field data collection and data quality control of anthropometric measurements; revision of paper

MC: Guidance on statistical analysis and interpretation

JDK: Guidance on psychomotor development assessment; revision of paper

CMS: Principle investigator of the Tswaka study; study design and responsible for overall data collection and quality control; guidance on collection and analysis of biochemical data; academic input and revision of paper.

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Abstract

Inadequate nutrition and development in the first thousand days have long-term effects on cognitive development and school achievement. This study assessed psychomotor development in relation to feeding practices and nutritional status of 6-month-old infants (n= 750) from a peri-urban community in the North West province of South Africa. Psychomotor development was assessed by Kilifi Developmental Inventory and a South African parent rating scale. Infants who were exclusively breastfed for a shorter duration, and those who frequently consumed commercial infant cereal and jarred infant foods reported significant higher parent rating scores [$p= 0.003$ ($d= 0.167$); $p=0.001$ ($d= 0.430$); $p= 0.014$ ($d= 0.212$)], respectively. Anaemic infants had significantly lower scores for both eye-hand coordination and locomotor activities [$p= 0.001$ ($d= 0.497$)] and $p= 0.001$ ($d= 0.463$), respectively, while infants suffering from IDA based on plasma ferritin and soluble transferrin receptor (TfR) had significantly lower scores for eye-hand coordination activities compared to non-IDA infants [$p= 0.007$ ($d= 0.411$)]. Stunted infants had significant lower scores for total combined psychomotor scores and parent rating scores ($p < 0.001$ ($d= 0.312$) and $p= 0.001$ ($d= 0.231$)), respectively. Multivariate regression analysis showed a significant positive correlation between haemoglobin concentration and eye-hand coordination, locomotor and combined psychomotor scores. This study indicated that healthy haemoglobin concentrations are associated with higher psychomotor developmental scores and stressed the importance of optimal infant feeding practices and nutrient status in relation to psychomotor development as a public health priority.

Keywords: Infants, psychomotor development, feeding practices, nutritional status

Introduction

Children attain their optimal potential through a combination of genetic capacity, psychosocial stimulation, adequate nutrition, and a safe, clean physical environment (Bentley et al. 2014). The impact of undernutrition on development in the first thousand days has substantial public health implications. Undernutrition causes a third of the total under-five mortality cases and it is reported that the post-MDG (Millennium Development Goals) era must focus on the reduction of child morbidity to ensure healthy development (Bentley et al. 2014; Liu et al. 2015). Globally, 24.5% of children younger than 5 years are stunted and 23.9% of South African children younger than five years are stunted (WHO 2015). Early childhood stunting is associated with poor cognitive performance (Abubakar et al. 2008; Grantham-McGregor et al. 2007) and after 2 years of age, the effects of malnutrition and some of the functional deficits caused by stunting may be irreversible (Dewey & Adu-afarwuah 2008; Eichler et al. 2012). Nutrition has an influence on brain development, therefore optimal feeding practices are important to ensure optimal psycho-motor development.

In support of healthy development, the WHO recommends exclusive breastfeeding for the first 6 months of life and continued breastfeeding until 2 years of age (WHO 2009). However, globally, a high percentage of infants are introduced to complementary foods before the age of 6 months, replacing breast milk intake (UNICEF 2014). China, with the largest population in the world and with more than 70% of its inhabitants living in rural areas, reported an exclusive breastfeeding rate of only 24.36% for the first 6 months of life (Zhou et al. 2012). The majority of European, American, and Indian infants are introduced to solid foods before 6 months (Caroli et al. 2012; Kuriyan & Kurpad 2012; Lutter 2012). Many African countries have shown increasing rates of exclusive breastfeeding in the last 10 years (IFPRI, 2015) but complementary foods still consist mainly of watery cereal porridges with low energy and nutrient density. Moreover, these complementary foods are often prepared and served under poor hygienic conditions (Black et al. 2008). Exclusive breastfeeding is not widely practiced in South Africa. A national survey that was done in 2012 showed that only 7.4% of infants aged < 6 months were being exclusively breastfed, and that infants were introduced to semi-solid/solid foods, mainly commercial infant cereal, before the age of 4 months (HSRC 2013).

Evidence on the relationship between feeding practices, nutritional status and psychomotor development during infancy is limited, particularly against the global background of early cessation of exclusive breastfeeding. Angulo-Barroso and co-workers (2011) found that cultural background, feeding practices, and iron status were associated with fine and gross motor development of 9-month-old infants in China, Ghana, and the USA (Angulo-Barroso et al. 2011). A Brazilian study of infants of age 6–12 months found that male infants, birth weight

< 1500 g, age-adjusted Z-scores < -1 for length, weight and head circumference, and exclusive breastfeeding for ≤ 2 months were negatively associated with psychomotor development (Eickmann et al. 2012). Apart from feeding practices and nutritional status, the nature of the home environment was also linked to the psychomotor development of infants aged 0–18 months (Sharma & Nagar 2009).

In terms of gross motor development, a longitudinal study in rural Guatemala followed children from birth through 3 years and showed that growth and diet quality – especially the intake of animal protein – were associated with the attainment of walking (Kuklina et al. 2004). These results correspond with findings from a study conducted on children aged 4–17 months in Nepal. The study in Nepal added anaemia as a significant contributor to delayed motor milestone development (Siegel et al. 2005).

While the first 6 months of life is critical in terms of development, and since the effects of nutritional deficits may be irreversible (Prado & Dewey 2014), it is imperative to identify nutritional deficits like stunting and anaemia at an early age, as both are linked to delayed cognitive development (Frongillo et al. 2014). There is a need for more recent literature on the impact of early feeding practices and nutritional status on the development of infants and children (Bhutta et al. 2013), especially under different contextual backgrounds. According to our knowledge, there is no recent literature that has assessed feeding practices and nutritional status in relation to development during infancy in South Africa. This study therefore assessed the association of psychomotor development scores of 6-month-old infants with early feeding practices and nutritional status (by focusing on anaemia, iron status and anthropometric indicators).

Methods

Study population and participants

This study reports on the baseline data of a randomized controlled trial involving 750, approximately 6-month-old infants (5.85–7.01 months) which was collected during the period of September 2013—January 2015. The trial was done in the peri-urban Jouberton area in the greater Matlosana Municipality in Klerksdorp in the North West province of South Africa. Jouberton is a low socio-economic community and is 200 km from the nearest metropolitan area (Johannesburg). The number of infants included and inclusion criteria are based on the sample calculations for the randomized controlled trial. Infants were excluded if they have never been breastfed; suffered from severe obvious congenital abnormalities or a chronic disease; were severely anaemic (Hb < 7.0 g/dl) or severely wasted (weight-for-length Z-score < -3.00); were allergic/intolerant to peanuts, soy, milk and/or lactose; received special

nutritional supplements; were not a singleton, or if their primary caregivers planned to move out of the study area within the next seven months. Caregivers with infants younger than 6 months were recruited for participation in the study through five primary health care clinics and house-to-house visits in order for infants to be six months of age when enrolled into the study.

Socio-economic characteristics, feeding practices, and nutritional status of the study population have been published elsewhere (*paper submitted for publication*).

Data collection

All questionnaires used for data collection were completed at a central study site where the caregivers were interviewed in their preferred language by trained field workers. The methodology used to collect data for socio-economic background, infant feeding practices, and iron and anthropometric status is described in detail elsewhere [*paper submitted for publication*]. These methods are described only briefly hereafter for completeness. Information on breastfeeding and complementary feeding practices were collected using a structured questionnaire that was developed based on WHO guidelines (WHO 2010). A set of unquantified food frequency questions was used to obtain descriptive information on the usual consumption of foods by the 6-month-old infants during the preceding 7 days. For each food item, frequency of consumption was described as “at least 4 days per week” (frequent consumption) or “less often than 4 days per week” (seldom/never).

Iron status was determined by blood sample analysis. If blood could not be obtained successfully via antecubital venepuncture of the arm area or dorsal area, a finger-prick blood sample was taken to determine haemoglobin (Hb) status. Therefore, Hb concentrations were collected from all infants (n= 750) while 4 ml blood samples were obtained from only 485 infants. Hb concentrations were determined by using the Hemocue on an aliquot of whole blood or fingerprick by using the direct cyanmethaemoglobin method (Ames Mini-Pak haemoglobin test pack and Ames Minilab, Bio-Rad Laboratories (PTY) Ltd). For infants from whom blood could be obtained successfully via antecubital venepuncture of the arm area or dorsal area, an Hb test were done and the remaining blood samples aliquots were centrifuged at 500 X g for 15 min at room temperature and the plasma aliquots were stored at -80°C. Plasma ferritin (Fer) and soluble transferrin receptors (TfR) concentrations were determined by using a sensitive Sandwich ELISA technique (Erhardt et al. 2004). High-sensitivity C-reactive protein (CRP) and alpha-1 glycoprotein (AGP) were measured with an ELISA kit from Human Diagnostics. The Fer concentrations were adjusted by using correction factors (CFs) according to Thurnham et al. (2015). Anaemia was defined as haemoglobin (Hb < 11.0 g/dl). Iron deficiency (ID) based on Fer as Fer < 12 µg/l; and iron deficiency anaemia (IDA) as both Fer < 12 µg/l and Hb < 11.0 g/dl (WHO 2011a, b). ID and IDA based on TfR, as TfR > 8.3 mg/l

for ID and Hb < 11.0 g/dl and TfR > 8.3 mg/l for IDA IDA (test-kit reference value).

Anthropometric measurements were taken by two fieldworkers who were trained according to International Society for Advancement of Kinanthropometry (ISAK) procedures (Marfell-Jones et al. 2012). The WHO growth standards were used to calculate age and gender specific Z-scores (WHO 2006). Stunting was defined as a length-for-age Z-score (LAZ) < -2 and overweight was defined as a weight-for-length Z-score (WLZ) > 2 (Wang & Chen 2012). Birth weight was recorded from the infant's clinic card and a low birth weight was defined as < 2.5 kg (WHO 2011c).

The Kilifi Developmental Inventory (KDI) and a South African parent rating scale were used for the assessment of psychomotor development. The KDI was developed for use with children in Africa, incorporating materials readily available in the setting, and requiring limited training to administer. Its psychometric properties have been previously established, and the validation process has included the development of age appropriate cut offs, and normal age ranges (Abubakar et al. 2008, 2010; Kitsao Wekulo 2016). The KDI was translated into the local language by a team member who was fluent in the language. The translation was verified during a pre-testing with volunteers that were not part of the study and corrected where needed. The KDI was used to calculate scores for locomotor skills, eye-hand coordination, as well as combined psychomotor development (which included both locomotor skills and eye-hand coordination). Locomotor skills assessed included movement in space, static and dynamic balance, and motor coordination. Eye-hand coordination was assessed based on the infant's ability to manipulate objects and engage in activities requiring fine motor coordination. Activities were scored on a scale which ranked from 0: Infant not able to perform the task; 1: Infant can perform the task, but not fluently/partially successfully; 2: Infant can perform the task fluently/successfully (Abubakar et al., 2007). The Parent Rating Scale was commissioned by the South African Department of National Education through a process of consultation and age validation to construct developmental standards to guide policy, and practice. The caregiver responds to a series of questions (Kvalsvig et al. 2009). Activities were scored on a scale which ranked from 1: Yes, infant was able; 2: Yes, but caregiver not able to tell when started; 3: Infant is learning; 4: Infant was not able. Both the KDI and parent rating assessments are based on the assessment of psychomotor activities of children aged 6–35 months and in this study only activities that were relevant to 6-month-old infants were utilised. Two field workers were trained and interrater reliability score sheet was used to ensure the comparability of the ranges of scores for each of the assessors. The field workers carry out the assessments in separate rooms to avoid distracting the infants. The caregiver was asked to assist where needed such as to get the attention of the infant.

Processing and statistical analysis of the data

Data quality and integrity were assured by a clinical research associate who visited and assessed the study site on a bi-weekly basis. Team members were trained to conduct the study according to good clinical practice. Data on infant feeding practices was entered manually into an EpiInfo data base. Data on socio-economic background, anthropometric status, blood results, KDI and parent rating scores were captured by ClinTech (India) International Pvt Ltd. Quality control was conducted by the clinical research associate by ensuring that data was correctly translated into the data set from source files in order to correct obvious errors. Questionnaire data, anthropometric and blood values were analysed using SPSS for Windows, version 21 (SPSS Inc., Chicago, Illinois).

KDI scores were calculated by adding up the scores recorded by the fieldworkers (0: unable to perform task; 1: partially able to perform task; 2: able to perform task) (Abubakar *et al.*, 2007). Parent rating scores as recorded by the fieldworkers were recoded and grouped accordingly: 1: infant was able; 0: infant was not able. Data was tested for normality by using the Kolmogorov-Smirnov test which is used for large sample sizes and normality test indicated that all data was normally distributed with $p > 0.05$. Descriptive statistics such as the frequencies, means and 95% confidence intervals (CI) for the mean were calculated. Analysis of variance was used to determine differences in mean psychomotor and parent rating scores between groups, with psychomotor and parent scores as dependent variables, and caregiver characteristic, gender, feeding practises and nutritional status as independent variables. Where analysis of variance showed significant differences for more than two groups, post-hoc analyses was done with a Bonferroni correction. Statistical significance was set at $p < 0.05$. Effect sizes were determined by using Cohen's d value to indicate the standardized difference between two means. Cohen's d was defined as the difference between two means divided by a standard deviation for the data. Correlation between parent rating and psycho-motor scores were determined by using Pearson correlation. Multivariate linear regression analysis was used to investigate associations between developmental scores, feeding practices and nutritional status. For the multivariate linear regression analysis, diagnostics were performed on the residuals of the final model to ensure that statistical assumptions were met.

Ethical considerations

The randomized control trial was registered as a clinical trial at the registry Clinicaltrials.gov (NCT01845610). Ethical approval was obtained from the Ethics Committee of the North-West University and the South African Medical Research Council (SAMRC). After institutional ethical approval was obtained, the study was reviewed by the Provincial Department of Health and Social Development for registration with the Directorate for Policy, Planning and

Research. If the caregiver agreed to participate and her infant was eligible, she was asked to read an information sheet and sign a consent form. Illiterate caregivers were assisted by field workers who read the information sheet to them and explained the content to make sure they understood what was expected of them, with the caregiver's thumbprint subsequently used as a "signature" on the consent form. Infants who were found to be severely anaemic or severely wasted during screening were to be referred to a primary health care clinic and were excluded from the study.

Results

Of the 750 primary caretakers, 91.7% were the mothers of the infants. The mean (95% CI) age of the primary caregivers was 28.4 (27.8, 29.0) years; 80% had attained at least 10 years of formal education (grade 10 or above), and only 10.7% were married. In terms of socio-economic characteristics, the majority of the households had a flush toilet (95.1%) and electricity (92.3%). All households had access to potable water, mostly through a tap either inside (29.3%) or outside (66.5%) the house. The median number of people living in the household was five and on average two people per household were recipients of a social grant.

Table 1 summarizes the main characteristics of the infants as well as the findings on psychomotor development and parent rating scores. In total, 29.3% of the infants were stunted, 10.1% were overweight, and 36.4% were anaemic. The prevalence of ID and IDA based on Fer, after adjustment for inflammation, was 16.1% and 9.3% respectively. ID and IDA prevalence based on TfR were 30.1% and 13.2%, respectively. For the infants in our study, we compared the Hb values obtained from capillary versus venous samples and results showed that there was no significant difference ($P= 0.657$) between the two groups.

At the time of the survey, 70.1% of the infants were still being breastfed. Food items reported to be consumed at least 4 days per week were mostly infant cereal (68.1%), formula milk feeds (41.7%) and jarred infant foods (22.7%). The mean (95% CI; [maximum possible score]) score obtained for combined psychomotor activities was 36.8 (36.3, 37.2 [53]), for eye-hand coordination 20.4 (20.1, 20.7 [27]), for locomotor activities 16.4 (16.1, 16.6 [26]), and for parent rating scores 20.2 (19.9, 20.3 [33]).

Table 2 compares the mean (95% CI) psychomotor development and parent rating scores in terms of duration of exclusive breastfeeding, consumption of frequently consumed food, birth weight, gender, caregiver's education, anaemia, and growth indicators. Parent rating scores were higher for girls compared to boys [$p=0.002$ ($d= 0.217$)], for infants who were cared for by caregivers with a higher education level [$p= 0.007$ ($d= 0.227$)], and for infants who were

exclusively breastfed up to age of 0–2 months compared to infants who were exclusively breastfed up to age 5–6 months [$p= 0.003$ ($d= 0.167$)]. When compared to infants who seldom/never consumed infant cereal, infants who frequently consumed commercial infant cereal reported higher scores for loco-motor activities [$p= 0.011$ ($d= 0.199$)], combined psychomotor development [$p= 0.026$ ($d= 0.167$)] and parent rating [$p= 0.001$ ($d= 0.430$)].

When compared to non-anaemic infants, anaemic infants had significantly lower scores for both eye-hand coordination [$p < 0.001$ ($d= 0.497$)] and locomotor activities [$p < 0.001$ ($d= 0.463$)], and as a result combined psychomotor development [$p < 0.001$ ($d= 0.556$)]. Iron deficient infants based on both Fer and TfR ($n=78$ and $n=146$), respectively, scored lower in combined psychomotor development scores [$p= 0.002$ ($d= 0.202$) and [$p= 0.028$ ($d= 0.204$)], respectively. Infants who suffered from IDA ($n=45$) based on Fer had significantly lower scores for combined psychomotor and eye-hand coordination activities compared to non-IDA infants ($n=440$) [$p < 0.001$ ($d= 0.407$); $p= 0.007$ ($d= 0.411$)], respectively. Infants who suffered from IDA based on TfR ($n=64$) showed lower scores for both eye-hand coordination [$p= < 0.001$ ($d= 0.532$)] and loco-motor activities [$p= 0.001$ ($d= 0.442$)], and therefore also combined psychomotor scores [$p= 0.001$ ($d= 0.532$)].

Lower parent rating scores were recorded for stunted versus non-stunted infants [$p= 0.001$ ($d= 0.231$)], overweight versus non-overweight infants [$p= 0.027$ ($d= 0.267$)], and low-birthweight versus normal birthweight infants [$p= 0.013$ ($d= 0.265$)].

Table 3 reports multivariate linear regression analysis associations of psychomotor development according to education level of the caregiver; gender of the infant; duration of exclusive breastfeeding; frequent intake of formula milk, commercial infant cereal and jarred infant food; haemoglobin, and TfR and Fer concentrations; LAZ-score; WLZ score and birth weight adjusted for cofactors. Table 4 reports multivariate linear regression analysis associations of parent rating scores for the same variables.

Haemoglobin concentration was positively related to eye-hand, locomotor and therefore also combined psychomotor activities [$\beta= 0.677$ (0.343, 1.011), $p= < 0.001$]; [$\beta= 0.439$ (0.164, 0.714), $p= 0.002$]; [$\beta= 1.116$ (0.586, 1.645), $p= < 0.001$], respectively.

Frequent consumption of infant cereal was positively related to locomotor development [$\beta= 0.709$ (0.043, 1.376), $p= 0.037$] and parent rating scores [$\beta= 1.506$ (0.912, 2.100); $p= < 0.001$]. Shorter duration of exclusive breastfeeding (up to the age of 0–2 months) was positively related to parent rating scores [$\beta= 1.544$ (0.673, 2.414), $p= 0.001$] as compared to exclusive breastfeeding up to the age of 5–6 months.

Male versus female gender was negatively related to parent rating scores [$\beta = -0.673$ ($-1.256, -0.089$), $p = 0.024$].

Discussion

It is important to investigate psychomotor development in the context wherein it occurs, especially when protective and risk factors are simultaneously present (Eickmann et al. 2012). Our study showed that differences in developmental scores were based on the level of education of the caregiver, gender, early feeding practices, and nutritional status. These differences varied with regard to the different developmental domains that were tested.

This study supports the relation between iron nutrition and young child development by indicating that haemoglobin concentration was positively associated with both eye-hand coordination and locomotor scores, and therefore combined psychomotor development scores. Consequently, anaemic infants had lower scores for both eye-hand coordination and locomotor activities as well as combined psychomotor activities. All of this is further supported in that infants with ID and IDA based on Fer and TfR, showed lower combined psychomotor development scores. Although Fer probably explains less than 50% of anaemia in this population, the findings emphasise the importance of iron nutrition during early development and the possible impact that the type of complementary food introduced may have on psychomotor development. These findings are in agreement with the results of a study conducted in China, which found that low haemoglobin levels in an older infancy age group (6–12 months) were associated with cognitive and psychomotor delays (Luo et al. 2015). Further studies conducted in China, Ghana and the USA showed that infants (aged 9–10 months) who were suffering from IDA were less successful in achieving eye-hand coordination activities (Shafir et al. 2009; Angulo-Barroso et al. 2011).

Iron is an important component of the haemoglobin molecule and a key nutrient needed for brain development (Prado & Dewey 2014) and iron deficiency is known to be linked to poor cognitive development (Beard 2008). Results of our study showed that fortified commercial infant cereals and fortified formula feeds provided significant amounts of dietary iron, and that infants who were fed formula milk for a longer duration had lower levels of anaemia and iron deficiency (*paper submitted for publication*). Our study further showed that infants who frequently consumed fortified commercial infant cereal reported better loco-motor, combined psycho-motor development and parent rating scores. The associations found between feeding practices and psychomotor development may be explained by the iron status of the infants as a consequence of feeding practices, taking into account that many of the infants are likely to have been born with low iron stores. In your study it was not possible to take iron stores at birth into account.

We did not observe any difference in psychomotor development or parent rating scores between infants who were still being breastfed at the age of six months and infants who were no longer being breastfed at the age of 6 months. Although the benefits of breastfeeding have been well documented it may be difficult to demonstrate without taking into account micronutrient status (including iron status) at birth. The findings may also show that some beneficial effects may only become apparent at a later stage. It may well be that the beneficial effect of breastfeeding at this specific age may only become apparent at a later stage. A large cohort study conducted in Ireland showed that breastfeeding during early infancy was associated with better neurodevelopment in infants at the age of 9-months (McCorry & Murray 2013). A study conducted in urban and rural areas in Spain showed that breastfeeding for longer than 4 months contributed to better mental development at 18 months of age (Gómez-Sanchiz et al. 2003).

Development during infancy is seen as a dynamic interplay between biological and environmental factors (Fernald *et al.*, 2009). Similar to a number of studies (Eickmann et al. 2012; Bruggink et al. 2009; Lima et al. 2004), we found that female infants scored higher in developmental assessments. Infants with a low birth weight and stunted infants scored lower parent rating scores. Intrauterine growth restriction, which is mainly caused by infections and poor maternal nutrition, causes difficulties in fetal nutrition during a crucial period of brain development and studies have reported that low-birth-weight infants showed lower developmental levels later in life (Walker et al. 2007). Stunting during the first thousand days may lead to poorer cognitive and educational outcomes later in life (Black *et al.*, 2013). Undernutrition, together with poor stimulation at home, is seen as key risk factors for poor infant development (Grantham-McGregor et al. 2014) and it is proven that undernourished children could benefit more from stimulation with an improved diet (Black et al. 2013; Grantham-McGregor et al. 2014). We however did not assess the level of stimulation of infants in this study. For future studies, it is therefore recommended to measure stimulation at home-level in combination with feeding practices and nutritional status.

The first six months of life is seen as a period of maximal growth velocity in healthy infants as well as a period of critical long-term neurodevelopment (Pongcharoen et al. 2012). It is therefore imperative to identify undernutrition in infants in this age group (Kerac et al. 2011) while it stresses the need for intervention at an earlier age (Prendergast & Humphrey 2014).

Early intake of long-chain polyunsaturated omega-3 fatty acids (n-3 LC-PUFAs) through fortified foods enhanced child development and visual acuity (Campoy et al., 2012) and supplementation with long-chain poly-unsaturated fatty acids contributes to better cognitive development later in life (De Jong et al., 2012; Colombo et al., 2013; Koletzko et al., 2014).

Only a small number of infants in our study population were given food items rich in essential fatty acids like fish, peanut butter, oil and butter/margarine (*paper submitted for publication*). Home fortification of complementary foods by using products such as nutritabs, micronutrient powders and lipid-based nutritional supplements (LNSs) has been proposed for children younger than 2 years (Gibson, 2011). LNSs rich in energy, protein and EFA are produced in a food base and the ration size for infants is designed to avoid displacement of breastmilk and also allow dietary diversity (Arimond et al., 2013). Small-quantity lipid-based nutrient supplements (SQ-LNS) were found to be acceptable for infants aged 6–12 months and their caregivers in this study population (Rothman et al. 2015) and the effect of thereof on linear growth and psychomotor development has been evaluated (*unpublished data*).

Some limitations of this study might be accuracy of the instruments used to evaluate psychomotor development. The main instrument, “Kilifi Developmental Inventory” (KDI) was not measured in relation to its concurrent validity against any other instrument considered gold standard as the Griffiths Mental Development Scales, Bayley Scales of Infant and Toddler Development or Alberta Infant Motor Scale. As in most parts of Africa, we worked in an area where psychological assessment tools are relatively new and standardized tools for the assessment of psychomotor development in field studies used by uneducated field workers are not in place yet. KDI tasks and materials included in the inventory of our study were based on findings from Kitsao-Wekulo et al. (2016) where the application of the KDI instrument were found to be able to monitor group differences in a research setting and aiding in the identification of developmental delays.

In conclusion, against the background of 1) a short duration of exclusive breastfeeding, 2) the introduction of fortified infant formula and commercial infant foods already between 0–2 months of age and 3) the prevalence of stunting, anaemia and iron deficiency of this study population, our multivariate regression analysis revealed small predictor values in association with changes in developmental scores. However, significant predictor values are useful to identify potential risk factors (low-haemoglobin concentration) and contributors (fortified infant formula and cereals) to psycho-motor development in 6-month-old infants. Finally, the associations found between feeding practices and psychomotor development may be explained by the iron status of the infants’ as a consequence of feeding practices and emphasize the need to know more about the nutritional status of infants from birth the age 6 months. Furthermore, the results should be interpreted with caution as the findings should not take away the long term beneficial effects of breast feeding on infant development and growth.

Key messages

1. Infants suffered from ID and IDA scored lower combined psychomotor scores

2. Non-Anaemic infants had significant higher scores for both eye-hand and locomotor activities as well as combined psychomotor scores
3. Infants who consumed fortified commercial infant cereal at a frequency of ≥ 4 days per week scored higher locomotor development and combined psychomotor development scores as well as higher parent rating scores.
4. Multivariate linear regression analysis showed that haemoglobin concentration was positively associated with eye-hand, locomotor and combined psychomotor development.
5. We need to know more about the nutritional status of infants from birth to the age of 6 months.

References

Abubakar A., Van de Vijver F.J.R., Mithwani S., Obiero N.L., Lewa N., Kenga S. *et al.* (2007) Assessing developmental outcomes in children from Kilifi, Kenya, following prophylaxis for seizures in cerebral malaria. *Journal of Health Psychology* 12 (3), 417-430.

Abubakar A., Holding P., Van Baar A., Newton C. & van de Vijver F.J. (2008) Monitoring psychomotor development in a resource limited setting: an evaluation of the Kilifi Developmental Inventory. *Annals of Tropical Paediatrics: International Child Health* 28 (3), 217–226.

Abubakar A., Holding P., Van de Vijver F., Bomu G. & Van Baar A. (2010) Developmental monitoring using caregiver reports in a resource-limited setting: the case of Kilifi, Kenya. *Acta Paediatrica* 99 (2), 291–297.

Angulo-Barroso R.M., Schapiro L., Liang W., Rodrigues O., Shafir T., Kaciroti N. *et al.* (2011) Motor development in 9-month-old infants in relation to cultural differences and iron status. *Developmental Psychobiology* 53 (2), 196–210.

Arimond M., Zeilani M., Jungjohann S., Brown K.H., Ashorn P., Allen L.H. *et al.* (2013) Considerations in developing lipid-based nutrient supplements for prevention of undernutrition: experience from the International Lipid-Based Nutrient Supplements (iLiNS) Project. *Maternal and Child Nutrition* doi: 10.1111/mcn.12049.

Beard J.L. (2008) Why iron deficiency is important in infant development. *The Journal of Nutrition* 138 (12), 2534–2536.

Bentley M.E., Johnson S.L., Wasser H., Creed-Kanashiro H., Shroff M., Fernandez Rao S. *et al.* (2014) Formative research methods for designing culturally appropriate, integrated child

nutrition and development interventions: an overview. *Annals of the New York Academy of Sciences* 1308 (1), 54–67.

Bhutta Z.A., Das J.K., Rizvi A., Gaffey M.F., Walker N., Horton S. *et al.* (2013) Evidence-based interventions for improvement of maternal and child nutrition: what can be done and at what cost? *The Lancet* 382 (9890), 452–477.

Black R.E., Allen L.H., Bhutta Z.A., Caulfield L.E., De Onis M., Ezzati M. *et al.* (2008) Maternal and child undernutrition: global and regional exposures and health consequences. *The Lancet* 371 (9608), 243–260.

Black R.E., Victora C.G., Walker S.P., Bhutta Z.A., Christian P., De Onis M. *et al.* (2013) Maternal and child undernutrition and overweight in low-income and middle-income countries. *The Lancet* 382 (9890), 427–451.

Bruggink J.L., Einspieler C., Butcher P.R., Stremmelaar E.F., Prechtl H.F. & Bos A.F. (2009) Quantitative aspects of the early motor repertoire in preterm infants: do they predict minor neurological dysfunction at school age? *Early Human Development* 85 (1), 25–36.

Campoy C., Escolano-Margarit M., Anjos T., Szajewska H. & Uauy R. (2012) Omega 3 fatty acids on child growth, visual acuity and neurodevelopment. *British Journal of Nutrition* 107(S2), S85-S106.

Caroli M., Mele R., Tomaselli M., Cammisa M., Longo F. & Attolini E. (2012) Complementary feeding patterns in Europe with a special focus on Italy. *Nutrition, Metabolism and Cardiovascular Diseases* 22 (10), 813-818.

Colombo J., Carlson S.E., Cheatham C.L., Shaddy D.J., Kerling E.H., Thodosoff J.M. *et al.* (2013) Long-term effects of LCPUFA supplementation on childhood cognitive outcomes. *The American Journal of Clinical Nutrition* 98 (2), 403–412.

De Jong C., Kikkert H.K., Fidler V. & Hadders-algra M. (2012) Effects of long-chain polyunsaturated fatty acid supplementation of infant formula on cognition and behaviour at 9 years of age. *Developmental Medicine & Child Neurology* 54 (12), 1102–1108.

Dewey K.G. & Adu-Afarwuah S. (2008) Systematic review of the efficacy and effectiveness of complementary feeding interventions in developing countries. *Maternal & Child Nutrition* 4 (s1), 24–85.

Eichler K., Wieser S., Rütthemann I. & Brügger U. (2012) Effects of micronutrient fortified milk and cereal food for infants and children: a systematic review. *BMC Public Health* 12 (1), 506–518.

Eickmann S.H., Malkes N.F.d.A. & Lima M.D.C. (2012) Psychomotor development of preterm infants aged 6 to 12 months. *Sao Paulo Medical Journal* 130 (5), 299–306.

Erhardt J.G., Estes J.E., Pfeiffer C.M., Biesalski H.K. & Craft N.E. (2004) Combined measurement of ferritin, soluble transferrin receptor, retinol binding protein, and C-reactive protein by an inexpensive, sensitive, and simple sandwich enzyme-linked immunosorbent assay technique. *The Journal of Nutrition* 134 (11), 3127–3132.

Faber M., Kvalsvig J.D., Lombard C.J. & Benadé A.S. (2005) Effect of a fortified maize-meal porridge on anemia, micronutrient status, and motor development of infants. *The American Journal of Clinical Nutrition* 82(5), 1032-1039.

Frongillo E.A., Tofail F., Hamadani J.D., Warren A.M. & Mehrin S.F. (2014) Measures and indicators for assessing impact of interventions integrating nutrition, health, and early childhood development. *Annals of the New York Academy of Sciences* 1308 (1), 68–88.

Gibson R. (2011) Strategies for preventing multi-micronutrient deficiencies: a review of experiences with food-based approaches in developing countries. In: Thompson B, Amoroso L (eds). *Combating micronutrient deficiencies: food-based approach*. CABI and FAO: Oxfordshire, UK, 1–92.

Grantham-McGregor S., Cheung Y.B., Cueto S., Glewwe P., Richter L. & Strupp B. (2007) Developmental potential in the first 5 years for children in developing countries. *The Lancet* 369 (9555), 60–70.

Grantham-McGregor S.M., Fernald L.C., Kagawa R. & Walker S. (2014) Effects of integrated child development and nutrition interventions on child development and nutritional status. *Annals of the New York Academy of Sciences* 1308 (1), 11–32.

Gómez-Sanchiz M., Cañete R., Rodero I., Baeza J.E., Ávila O. (2003) Influence of breastfeeding on mental and psychomotor development. *Clinical Pediatrics* 42, 35-42.

HSRC (Human Sciences Research Council). (2013) The South African National Health and Nutrition Examination Survey (SANHANES-1). Data analysis on infant feeding practices, and anthropometry in children under five years of age: South Africa 2012. Report for UNICEF, November 2013.

IFPRI (International Food Policy Research Institute). (2015). Global Nutrition Report 2015: Actions and Accountability to Advance Nutrition and Sustainable Development. Washington, DC.

Kerac M., Blencowe H., Grijalva-Eternod C., McGrath M., Shoham J., Cole T.J. *et al.* (2011) Prevalence of wasting among under 6-month-old infants in developing countries and implications of new case definitions using WHO growth standards: a secondary data analysis. *Archives of Disease in Childhood* 96 (11), 1008–1013.

Kitsao-Wekulo P., Holding P., Abubakar A., Kvalsvig J., Taylor G.H. & King C.L. (2016). Describing normal development in an African setting: The utility of the Kilifi Developmental Inventory among young children at the Kenyan coast. *Learning and Individual Differences*, doi: 10.1016/j.lindif.2015.11.011.

Koletzko B., Boey C.C., Campoy C., Carlson S.E., Chang N., Guillermo-Tuazon M. *et al.* (2014) Current information and Asian perspectives on long-chain polyunsaturated fatty acids in pregnancy, lactation, and infancy: systematic review and practice recommendations from an early nutrition academy workshop. *Annals of Nutrition and Metabolism* 65 (1), 49–80.

Kuklina E.V., Ramakrishnan U., Stein A.D., Barnhart H.H. & Martorell R. (2004) Growth and diet quality are associated with the attainment of walking in rural Guatemalan infants. *The Journal of Nutrition* 134 (12), 3296–3300.

Kuriyan R. & Kurpad A. (2012) Complementary feeding patterns in India. *Nutrition, Metabolism and Cardiovascular Diseases* 22 (10), 799–805.

Kvalsvig J.D, Govender K, and Taylor M (2009) Research on the age validation of NELDS related to the cognitive development of children between 0 and 4 years of ages. Report to UNICEF and the Department of Education.

Lima M., Eickmann S., Lima A., Guerra M., Lira P., Huttly S. *et al.* (2004) Determinants of mental and motor development at 12 months in a low income population: a cohort study in northeast Brazil. *Acta Paediatrica* 93 (7), 969–975.

Liu L., Oza S., Hogan D., Perin J., Rudan I., Lawn J.E., *et al.* (2015) Global, regional, and national causes of child mortality in 2000–13, with projections to inform post-2015 priorities: an updated systematic analysis. *The Lancet* 385 (9966), 430-440.

Luo R., Shi Y., Zhou H., Yue A., Zhang L., Sylvia S. *et al.* (2015) Micronutrient deficiencies and developmental delays among infants: evidence from a cross-sectional survey in rural China. *BMJ Open* 5 (10), e008400.

Lutter C. (2012) Growth and complementary feeding in the Americas. *Nutrition, Metabolism and Cardiovascular Diseases* 22 (10), 806–812.

Marfell-Jones M., Stewart A. & De Ridder H. (2012) ISAK accreditation handbook. Upper Hutt, New Zealand: International Society for the Advancement of Kinanthropometry.

McCrory C. & Murray A. (2013) The effect of breastfeeding on neuro-development in infancy. *Maternal and Child Health Journal* 17 (9), 1680–1688.

Pongcharoen T., Ramakrishnan U., DiGirolamo A.M., Winichagoon P., Flores R., Singkhornard J. *et al.* (2012) Influence of prenatal and postnatal growth on intellectual functioning in school-aged children. *Archives of Pediatrics & Adolescent Medicine* 166 (5), 411–416.

Prado E.L. & Dewey K.G. (2014) Nutrition and brain development in early life. *Nutrition Reviews* 72 (4), 267–284.

Prendergast A.J. & Humphrey J.H. (2014) The stunting syndrome in developing countries. *Paediatrics and International Child Health* 34 (4), 250–265.

Rothman M., Berti, C., Smuts C.M, Faber M., Covic N.M. (2015) Acceptability of novel small-quantity lipid-based nutrient supplements for complementary feeding in a peri-urban South Africa community. *Food and Nutrition Bulletin* 36: 455-466.

Shafir T., Angulo-Barroso R., Su J., Jacobson S.W. & Lozoff B. (2009) Iron deficiency anemia in infancy and reach and grasp development. *Infant Behavior and Development* 32 (4), 366–375.

Sharma S. & Nagar S. (2009) Influence of home environment on psychomotor development of infants in Kangra district of Himachal Pradesh. *Journal of Social Sciences* 21 (3), 225–229.

Siegel E.H., Stoltzfus R.J., Kariger P.K., Katz J., Khatry S.K., LeClerq S.C. *et al.* 2005. Growth indices, anemia, and diet independently predict motor milestone acquisition of infants in south central Nepal. *The Journal of Nutrition* 135 (12), 2840–2844.

Thurnham D.I., Northrop-Clews C.A. & Knowles J. (2015) The use of adjustment factors to address the impact of inflammation on vitamin A and iron status in humans. *The Journal of Nutrition* 145 (5), 1137S–1143S.

UNICEF. (2014) Statistical tables: Economic and social statistics on the countries and areas of the world, with particular reference to children's well-being. New York, USA.

Walker S.P., Wachs T.D., Meeks Gardner J., Lozoff B., Wasserman G.A., Pollitt E. *et al.* (2007) Child development: risk factors for adverse outcomes in developing countries. *The Lancet* 369 (9556), 145–157.

Wang Y. & Chen H.J. (2012) Use of percentiles and z-scores in Anthropometry. In: Preedy, V.R (ed). *Handbook of Anthropometry. Physical Measures of human form in health and disease*, ISBN: 978-1-4419-1787-4.

WHO (World Health Organization) (2006). *Child growth standards. Length/height-for-age, weight-for age, weight-for-length, weight-for-height and body mass index for age. Methods and development*. World Health Organization: Geneva.

WHO (World Health Organization). (2009) *Infant and young child feeding: model chapter for textbooks for medical students and allied health professionals*. Geneva, Switzerland: World Health Organization, ISBN-13: 978-92-4-159749-4.

WHO (World Health Organization). (2010) *Indicators for assessing infant and young child feeding practices. Part 2: Measurement*. World Health Organization: Geneva.

WHO (World Health Organization). (2011a) Haemoglobin concentrations for the diagnosis of anaemia and assessment of severity. Vitamin and Mineral Nutrition Information System. Geneva, Switzerland: World Health Organization.

WHO (World Health Organization). (2011b) Serum ferritin concentrations for the assessment of iron status and iron deficiency in populations. Vitamin and Mineral Nutrition Information System. Geneva, Switzerland: World Health Organization.

WHO (World Health Organization). (2015) *World Health Statistics*. Geneva: World Health Organization.

WHO (World Health Organization) Multicentre Growth Reference Study Group. (2006) WHO motor development study: Windows of achievements for six gross motor development milestones. *Acta Paediatrica*, 450, 86–95.

Zhou H., Wang X.-L., Ye F., Zeng X.L. & Wang Y. (2012) Relationship between child feeding practices and malnutrition in 7 remote and poor counties, PR China. *Asia Pacific Journal of Clinical Nutrition* 21 (2), 234

Table 1. Gender, nutritional status, duration of exclusive breastfeeding of and scores (mean, 95% CI) obtained in psychomotor development- and parent rating activities of 6-month-old infants

<i>Infant characteristics (n=750)</i>		
Male / Female (%)		51.6 / 48.4
Low birth weight ¹ (%)		14
Stunted ² (%)		29.3
Overweight ³ (%)		10.1
Anaemic ⁴ (%)		36.4
Iron deficient based on Fer ^{5,6} (%)		16.1
Iron deficiency anaemia based on Fer ^{5,7} (%)		9.3
Iron deficient based on TfR ^{5,8} (%)		30.1
Iron deficiency anaemia based on TfR ^{5,9} (%)		13.2
Still being breastfed at age 6 months (%)		70.1
<i>Duration of exclusive breastfeeding (%)</i>		
Age 0–2 months		48.9
Age 3–4 months		36.1
Age 5–6 months		14.9
<i>Psychomotor development scores (n=750)</i>	<i>Max possible score</i>	<i>Mean (95% CI)</i>
Combined psychomotor score	53	36.8 (36.3, 37.2)
<i>Eye-hand coordination sub-scale</i>	27	20.4 (20.1, 20.7)
Locomotor skills sub-scale	26	16.3 (16.1, 16.6)
Parent Rating	33	20.2 (19.9, 20.4)

¹ < 2.5 kg; ² Length-for-age Z-score < 2; ³ Weight-for-length Z-score > 2; ⁴ Hb < 11 g/dl; ⁵ n= 485; ⁶ Fer < 12 µg/l, after adjustment for inflammation; ⁷ Hb < 11 g/l and Fer < 12 µg/l, after adjustment for inflammation; ⁸ TfR > 8.3 mg/l; ⁹ Hb < 11 g/l and TfR > 8.3 mg/l

Table 2. Comparison between development scores and caregiver's level of education, gender, early feeding practices, frequently consumed food items, anaemic- and iron status, growth status and birth weight of 6-month-old infants (mean, CI)

	Eye-hand			Locomotor			Combined psychomotor			Parent rating			
	n	(max score 27)	<i>P</i> ¹	<i>d</i>	(max score 26)	<i>P</i> ¹	<i>d</i>	(max.score 53)	<i>P</i> ¹	<i>d</i>	(max score 33)	<i>P</i> ¹	<i>d</i>
Caregiver education													
≥ Grade 10	601	20.3 (20.0, 20.7)	0.462	0.069	16.4 (16.1, 16.6)	0.812	0.020	36.7 (36.2, 37.2)	0.879	0.031	20.3 (20.1, 20.6)	0.007	0.227
< Grade 10	138	20.6 (19.9, 21.3)			16.3 (15.7, 16.9)			36.9 (35.9, 38.0)			19.5 (18.9, 20.1)		
Gender													
Male	387	20.4 (20.0, 20.8)	0.988	0.001	16.4 (16.1, 16.7)	0.618	0.036	36.8 (36.2, 37.4)	0.662	0.017	19.8 (19.5, 20.1)	0.002	0.217
Female	363	20.4 (19.9, 20.8)			16.3 (15.9, 16.6)			36.7 (36.0, 37.4)			20.5 (20.2, 20.8)		
Duration of EBF													
Age 0–2 months	367	20.5 (20.1, 20.9)	0.559	0.079	16.4 (16.1, 16.7)	0.103	0.018	36.9 (36.3, 37.6)	0.864	0.038	20.5 (20.2, 20.8)	0.003 ²	0.167
Age 3–4 months	271	20.2 (19.7, 20.7)			16.5 (16.1, 16.9)			36.7 (35.9, 37.5)			19.9 (19.6, 20.3)		
Age 5–6 months	112	20.5 (19.9, 21.1)			15.8 (15.2, 16.3)			36.3 (35.3, 37.3)			19.4 (18.8, 20.1)		
Breastfed at age 6 months													
Yes	525	20.5 (20.2, 20.8)	0.383	0.064	16.3 (16.1, 16.6)	0.662	0.034	36.8 (36.3, 37.4)	0.206	0.026	20.18 (19.9, 20.5)	0.745	0.026
No	224	20.2 (19.6, 20.8)			16.4 (16.0, 16.8)			36.7 (35.8, 37.5)			20.1 (19.7, 20.5)		
Frequently consumed foods													
Formula milk													
Frequent ³	311	20.6 (20.0, 21.0)	0.366	0.063	16.7 (16.3, 17.1)	0.021	0.169	37.3 (36.5, 37.9)	0.274	0.127	20.4 (20.1, 20.8)	0.067	0.132
Seldom/never	430	20.3 (19.9, 20.6)			16.1 (15.8, 16.5)			36.4 (35.8, 37.0)			20.0 (19.7, 20.3)		
Commercial infant cereal													
Frequent ³	503	20.5 (20.2, 20.9)	0.177	0.103	16.6 (16.3, 16.9)	0.011	0.199	37.1 (36.6, 37.7)	0.026	0.167	20.6 (20.4, 20.9)	0.001	0.430
Seldom/never	236	20.1 (19.6, 20.6)			15.9 (15.5, 16.4)			36.1 (35.3, 36.8)			19.21 (18.8, 19.6)		
Jarred infant foods													
Frequent ³	168	20.7 (20.2, 21.2)	0.263	0.096	16.6 (16.1, 17.0)	0.092	0.069	37.3 (36.4, 38.2)	0.842	0.096	20.7 (20.3, 21.1)	0.014	0.212
Seldom/never	573	20.3 (19.9, 20.6)			16.3 (16.1, 16.6)			36.6 (36.1, 37.2)			20.0 (19.8, 20.3)		

	n	Eye-hand			Locomotor			Combined psychomotor			Parent Rating		
		(max score 27)	<i>P</i> ¹	<i>d</i>	(max score 26)	<i>P</i> ¹	<i>d</i>	(max.score 53)	<i>P</i> ¹	<i>d</i>	(max score 33)	<i>P</i> ¹	<i>d</i>
Anemic status													
Anemic ⁴	273	19.0 (18.7, 19.4)	<0.001	0.497	15.3 (15.0, 15.6)	<0.001	0.463	34.3 (33.8, 34.9)	<0.001	0.556	20.1 (19.7, 20.5)	0.530	0.046
Non-Anemic	477	21.2 (20.8, 21.6)			16.9 (16.6, 17.3)			38.1 (37.5, 38.8)			20.2 (19.9, 20.5)		
ID status													
ID ⁵	78	19.9 (19.0, 20.7)	0.190	0.161	15.9 (15.2, 16.6)	0.121	0.282	35.8 (34.5, 37.1)	0.002	0.202	20.0 (19.4, 20.7)	0.690	0.049
Non-ID	407	20.5 (20.1, 20.9)			16.5 (16.2, 16.9)			37.1 (36.5, 37.7)			20.2 (19.9, 20.5)		
ID ⁶	146	19.9 (19.3, 20.7)	0.115	0.205	15.9 (15.4, 16.5)	0.022	0.218	35.9 (34.8, 37.0)	0.028	0.204	19.9 (19.5, 20.5)	0.492	0.066
Non-ID	339	20.6 (20.2, 21.0)			16.7 (16.3, 17.0)			37.3 (36.6, 37.9)			20.2 (19.9, 20.6)		
IDA status													
IDA ⁷	45	18.9 (18.1, 19.8)	0.007	0.411	15.6 (14.9, 16.3)	0.065	0.282	34.5 (33.2, 35.9)	<0.001	0.407	20.1 (19.2, 21.0)	0.981	0.004
Non-IDA	440	20.6 (20.2, 20.9)			16.5 (16.2, 16.9)			37.1 (36.5, 37.7)			20.2 (19.9, 20.4)		
IDA ⁸	64	18.7 (18.1, 19.4)	<0.001	0.532	15.1 (14.6, 15.7)	0.001	0.442	33.9 (32.9, 34.9)	0.001	0.532	20.4 (19.6, 21.1)	0.533	0.083
Non-IDA	421	20.7 (20.3, 21.1)			16.6 (16.3, 16.9)			37.3 (36.7, 37.9)			20.1 (19.8, 20.4)		
LAZ													
Stunted ⁹	220	20.3 (19.8, 20.8)	0.657	0.035	16.4 (15.9, 16.7)	0.982	0.002	36.6 (35.8, 37.5)	0.930	0.024	19.6 (19.1, 20.0)	0.001	0.231
Non-stunted	530	20.4 (20.1, 20.8)			16.4 (16.1, 16.6)			36.8 (36.3, 37.3)			20.4 (20.2, 20.7)		
WLZ													
Overweight ¹⁰	76	20.3 (19.4, 21.2)	0.869	0.019	16.0 (15.3, 16.8)	0.328	0.116	36.3 (34.8, 37.8)	0.684	0.071	19.4 (18.7, 20.1)	0.027	0.267
Not-overweight	674	20.4 (20.1, 20.7)			16.4 (16.2, 16.6)			36.8 (36.3, 37.3)			20.2 (20.0, 20.5)		
Birth weight													
< 2.50 kg	102	19.8 (19.1, 20.6)	0.156	0.150	16.4 (15.8, 16.9)	0.947	0.007	36.3 (35.1, 37.5)	0.813	0.092	19.5 (18.8, 20.1)	0.013	0.265
≥ 2.50 kg	626	20.5 (20.2, 20.8)			16.4 (16.1, 16.6)			36.9 (36.4, 37.4)			20.3 (20.1, 20.5)		

¹ Analysis of variance

² Bonferroni post-hoc test indicated significant difference between 0-2 months and 4-6 age groups ($p = 0.005$)

³ ≥ 4 days a week; ⁴ Hb < 11g/l; ⁵ Fer < 12µg/l; ⁶ TfR > 8.3 mg/l; ⁷ Hb < 11g/l and Fer < 12µg/l; ⁸ Hb < 11g/l and TfR > 8.3 mg/l; ⁹ Length-for age Z-score < -2; ¹⁰ Weight-for-length Z-score: > 2

d (Cohen's *d*-value): 0 no practical significance; 0.2 small practical significance; 0.5 medium practical significance; 0.8 large practical significance

EBF: Exclusive breastfeeding

Table 3. Multivariate linear regression analysis for psychomotor development

Variable	Eye-hand coordination			Locomotor			Combined psychomotor		
	β^1 (95% CI)	β^2	<i>P</i>	β^1 (95% CI)	β^2	<i>P</i>	β^1 (95% CI)	β^2	<i>P</i>
Caregiver education									
\geq Grade 10 ³	0.198 (-.742, 1.139)	-0.19	0.679	0.153 (-0.622, 0.928)	-0.018	0.699	0.351 (-1.139, 1.841)	-0.069	0.644
Gender									
Male ⁴	-0.141 (-0.936, 0.654)	0.017	0.727	0.397 (-0.258, 1.052)	-0.060	0.234	0.256 (-1.003, 1.516)	0.109	0.690
Duration of exclusive breastfeeding									
Age 0-2 months ⁵	-0.758 (-1.943, 0.428)	0.005	0.210	-0.237 (-1.214, 0.740)	0.066	0.634	-0.994 (-2.873, 0.885)	-0.129	0.299
Age 3-4 months ⁵	-0.712 (-1.905, 0.481)	0.064	0.242	0.216 (-0.767, 1.199)	0.024	0.666	-0.496 (-2.387, 1.395)	-0.171	0.607
Frequent intake of formula milk ⁶	-0.395 (-1.207, 0.417)	0.048	0.340	0.146 (-0.523, 0.815)	-0.022	0.668	-0.294 (-1.536, 1.038)	-0.032	0.704
Frequent intake of jarred infant food ⁶	-0.021 (-0.943, 0.901)	0.002	0.964	0.167 (-0.593, 0.927)	-0.021	0.666	0.146 (-1.315, 1.608)	-0.052	0.844
Frequent intake of infant cereal ⁶	0.504 (-0.305, 1.313)	-0.058	0.221	0.709 (0.043, 1.376)	-0.100	0.037	1.214 (-0.068, 2.495)	-0.228	0.063
Plasma ferritin ($\mu\text{g/l}$)	-0.011 (-0.030, 0.007)	-0.064	0.236	0.013 (-0.002, 0.028)	0.092	0.090	0.002 (-0.027, 0.031)	-0.072	0.894
Haemoglobin (g/l)	0.677 (0.343, 1.011)	0.196	<0.001	0.439 (0.164, 0.714)	0.155	0.002	1.116 (0.586, 1.645)	-0.089	<0.001
Transferrin receptor (mg/l)	-0.087 (-0.234, 0.061)	-0.059	0.249	-0.008 (-0.129, 0.113)	-0.007	0.896	-0.095 (-0.328, 0.139)	0.014	0.426
LAZ score	0.161 (-0.275, 0.597)	0.041	0.468	0.256 (-0.103, 0.615)	0.079	0.162	0.417 (-0.274, 1.108)	0.075	0.236
WLZ score	-0.162 (-0.509, 0.184)	-0.045	0.358	-0.060 (-0.346, 0.226)	-0.020	0.679	-0.223 (-0.772, 0.327)	-0.083	0.427
Birth weight	0.884 (-0.149, 1.916)	0.099	0.093	-0.684 (-1.535, 0.167)	-0.093	0.115	0.200 (-1.436, 1.836)	0.073	0.810
	Adjusted R ²	0.04		Adjusted R ²	0.03		Adjusted R ²	0.037	

¹ Unstandardized; ² Standardized

LAZ: Length-for-age Z-score; WLZ: Weight-for-length Z-score

Reference categories: ³< Grade 10; ⁴ Female; ⁵ Age 5–6 months; ⁶ seldom /never

Table 4. Multivariate linear regression analysis for parent rating score

Variable	Parent Rating score		
	β ¹ (95% CI)	β^2	<i>P</i>
Caregiver education			
\geq Grade 10 ³	0.535 (-0.155, 1.226)	-0.069	0.128
Gender			
Male ⁴	-0.673 (-1.256, -0.089)	0.109	0.024
Duration of exclusive breastfeeding			
Age 0-2 months ⁵	1.544 (0.673, 2.414)	-0.129	0.001
Age 3-4 months ⁵	0.722 (0.446, 1.619)	-0.171	0.106
Frequent intake of formula milk ⁶	0.198 (-0.399, 0.794)	-0.032	0.510
Frequent intake of jarred infant food ⁶	0.388 (-0.289, 1.065)	-0.052	0.261
Frequent intake of infant cereal ⁴	1.506 (0.912, 2.100)	-0.228	<0.001
Plasma ferritin ($\mu\text{g/l}$)	-0.010 (-0.023, 0.004)	-0.072	0.165
Haemoglobin (g/l)	-0.233 (-0.478, 0.012)	-0.089	0.063
Transferrin receptor (mg/l)	0.016 (-0.092, 0.124)	0.014	0.773
LAZ score	0.226 (-0.094, 0.546)	0.075	0.166
WLZ score	-0.228 (-0.483, 0.027)	-0.083	0.079
Birth weight	0.500 (-0.258, 1.259)	0.073	0.195
	Adjusted R ²	0.106	

¹ Unstandardized; ² Standardized

LAZ: Length-for-age Z-score; WLZ: Weight-for-length Z-score

Reference categories:³< Grade 10; ⁴Female; ⁵ Age 5–6 months; ⁶ seldom /never

CHAPTER 5

This manuscript is formatted according to guidelines of Food and Nutrition Bulletin (see Annexure 3), with exception of the line numbers and line spaces. This manuscript is accepted for publication (doi: 10.1177/0379572115616057) (Annexure 4).

Acceptability of novel small-quantity lipid-based nutrient supplements for complementary feeding in a peri-urban South African community

Marinel Rothman, Cristiana Berti, Cornelius M Smuts, Mieke Faber, Namukolo Covic

Marinel Rothman, Cristiana Berti, Marius Smuts and Namukolo Covic are affiliated with North-West University, Potchefstroom; South Africa. Mieke Faber is affiliated with the Non-Communicable Disease Research Unit of the South African Medical Research Council, Cape Town, South Africa.

Please address all inquiries to the corresponding author: Marinel Rothman, Centre of Excellence for Nutrition, North-West University, South Africa; Potchefstroom Campus; Private Bag X6001; Potchefstroom; 2520. E-mail: marinel.rothman@nwu.ac.za

ABSTRACT

Background: Small quantity lipid-based nutritional supplements may potentially be used for home-fortification in poor settings where low nutrient dense complementary foods are commonly used for infant feeding. However, they need to be acceptable to succeed.

Objective: This study assessed the acceptability of two novel, small-quantity lipid-based nutritional supplements (A & B) for supplementing complementary foods among infants 6–12 months old in a peri-urban South African community.

Methods: Both supplements were soy-based pastes and contained micronutrients and essential fatty acids. In addition, supplement B contained docosahexaenoic acid, arachidonic acid, phytase and L-lysine. Mother/infant pairs were enrolled in a two-part trial. Part 1 ($n = 16$) was a test-feeding trial with a cross-over randomized design, and a five-point hedonic scale was used for sensory evaluation (disagree = 1, agree = 5). Part 2 ($n = 38$) was a two-week home-use trial followed by focus group discussions.

Results: In Part 1, more than 70% of mothers reported a score ≥ 4 on sensory attributes for both small-quantity lipid-based nutritional supplements indicating that both supplements were well perceived. In Part 2, the mean reported consumption over the two week period was $65.3 \pm 34.2\%$ and $62.0 \pm 31.3\%$ of the 20 g daily portion for supplement A and B, respectively. Focus group discussions confirmed a positive attitude towards the supplements in the study population.

Conclusions: This study showed acceptance of both small-quantity lipid-based nutritional supplements in terms of sensory characteristics as well as in terms of practicality for home-use.

Keywords: lipid-based supplements, infants, acceptability, complementary feeding

Introduction

Worldwide, undernutrition consisting of stunting, wasting, vitamin A and zinc deficiency, as well as suboptimum breastfeeding causes nearly 3.1 million (45%) of deaths of children younger than 5 years¹. One of the six global targets for the improvement of infant and young child nutrition by 2025 is a 40% reduction in children younger than five years who are stunted. In 2012, 36% of all stunted children, 29% of all underweight children and an estimated 28% of all severely wasted children were living in Africa². South Africa is among the 34 countries with the highest burden of stunting on a global level³. The highest percentage of stunting falls in the age category 0-3 years at 26.9% and 25.9% for South African boys and girls, respectively⁴.

South Africa has the lowest level (8%) of exclusive breastfeeding in Sub-Saharan Africa⁵. It has been reported that in some areas 35% to 50% of mothers stop all breastfeeding before 3 months postpartum, that complementary foods are introduced as early as 6 weeks of age^{6,7}, and that the nutrient density of the complementary diet is poor^{8,9}. These suboptimal feeding practices are of concern, particularly against the background that the prevalence of stunting in the 1-3 years age group increased by 3% from 2005 to 2012⁴. Strategies to improve dietary quality of complementary foods are therefore urgently needed.

Small-quantity lipid-based nutrient supplements (SQ-LNS), which contribute to energy, protein and essential fatty acids (EFA) intake, are easy to use and require minimum change in dietary behaviours^{10,11}. Presently, many organisations and companies have shown interest in the development of SQ-LNS and their potential use in a variety of cultural and geographical settings¹⁰. Recently, a study conducted in the Honduras indicated that SQ-LNS improved micronutrient status in non-malnourished children aged 6-18 months¹¹. Iannotti et al.¹² showed that daily consumption of SQ-LNS resulted in a significant increase in linear growth in healthy infants aged 6–11 months in Haiti. To ensure large scale use of SQ-LNS, these products should be acceptable within a given socio-cultural context. Although acceptability trials on SQ-LNS were conducted among children younger than 5 years in some countries like Malawi, Burkina Faso, Ghana and Guatemala¹³⁻¹⁶, these did not contain added docosahexaenoic acid (DHA), arachidonic acid (ARA) and phytase.

Two new SQ-LNS were developed for potential use in the South African setting. Both SQ-LNS (A and B) are made from soy, which is less expensive than peanuts; and contain essential fatty acids and micronutrients needed for infant development^{17,18}. In addition SQ-LNS B contains ARA and DHA, which may provide additional benefits in terms of vision,

growth and psychomotor development¹⁹⁻²¹, phytase to improve iron and zinc bioavailability^{17,22}, and L-lysine, which is an essential amino acid that is lacking in maize. In South Africa, as in many African countries, maize meal porridge is often used for complementary feeding in infants^{8,23}. However, the acceptance of home use of SQ-LNS for infant feeding has not yet been established in South Africa.

The aim of this study was, to determine the acceptability of two newly developed SQ-LNS pastes in a peri-urban South African community. The specific objectives of the study presented in this paper were: 1) to determine the acceptability in terms of sensory characteristics to infants and their primary caregivers, as rated by the primary caregiver on a 5-point hedonic scale; and 2) to obtain information on how the caregivers experienced using the products by conducting a two-week home-use trial followed by focus group discussions (FGDs).

Materials and Methods

Overview and Design

The two SQ-LNS were produced by two European companies, namely DSM and UNILEVER, with a presence and capacity to produce in South Africa. The two SQ-LNS were delivered to the nutrition laboratory at the North-West University, Potchefstroom. Both SQ-LNS were packed in single-dose 20 g plastic tubs with lids. The nutrient composition of the SQ-LNS A and B is presented in Table 1. The study was single blinded and for the purpose of the study the two products, utensils, as well as documentation used were colour coded. The products were only referred to in colour codes at all times to the field staff facilitators and participants.

Study setting

The trial was conducted in February-April 2013 in a low socio-economic peri-urban community in the North West Province in South-Africa. This trial preceded a randomized controlled trial that assessed the effect of SQ-LNS on infant's growth from 6-12 months, therefore this trial recruited infants in the same age category.

Ninety-four percent of the caregivers who participated in the study were the mothers of the infants. For this reason, the term "mother" is used throughout the rest of the article.

Recruitment process

The mother-infant pairs were recruited at five clinics in the community. These clinics allocate specific days for mother-infant pairs and the recruitment was done on these specific days. Trained fieldworkers explained the study to the mothers by using a structured information sheet explaining the meaning and importance of the trial as well as some background of the SQ-LNS; the mothers were also given an information sheet in their local language. If the mother expressed willingness to participate, an eligibility questionnaire was administered to determine whether the infant could be included in the study. To be eligible to participate in the study, the infant had to be 6 – 12 months of age, have consumed some breast milk previously, be apparently healthy with no fever and not suffering from an acute illnesses such as acute respiratory tract infection and diarrhoea, not be severe malnourished (mid-upper arm circumference [MUAC] \leq 115 mm), and with no known allergy to peanuts, soy, milk/lactose, and fish. Also, the mother/infant pairs should plan to remain in the study area for at least the following four weeks to ensure no adverse reactions occurred. Before

enrolment, a consent form was signed by the mother. Different infants were recruited for Part 1 and 2 of the acceptability trial to avoid a carry-over effect. A different information- and consent form were used for Part 2 which included information regarding the focus group discussions. Eligibility criteria was the same for Part 2.

MUAC was taken in order to determine if the infant was severe malnourished by using a standard MUAC tape following standard procedures²⁴. Infants excluded from participation for having failed the MUAC screening, were to be referred to the nearest clinic.

Research approach used

The methodology used in this study was a combination of methodological elements from the acceptability trials that were conducted in Malawi, Ghana and Guatemala^{13,15,16}, that were adapted for our specific setting. This was a single-blind study as only the researchers were able to distinguish between the SQ-LNS A and –B. This study was carried out in two parts. Part 1 was a sensory evaluation in which the mothers were asked to taste each SQ-LNS product mixed with maize porridge and then score the SQ-LNS-mixture on different sensory attributes. The mothers were also asked to feed the SQ-LNS-mixture to their infants and then rate their perception of the infant's liking of the products. The maize meal porridge used was prepared by a fieldworker from the community. Part 2 was a two-week home-use trial to obtain information on how mothers experienced the use of the SQ-LNS at household level. At the end of Part 2, FGDs were conducted to get insight on how the mothers experienced using the SQ-LNS during the home use trial. The number of mother-infant pairs enrolled in Part 1 and Part 2 of the study were based on similar acceptability trials conducted in Malawi¹³, Burkina Faso¹⁴ and Guatemala¹⁶. For FGDs, at least 8 participants per group are needed, therefore the 40 mother-infants pairs enrolled in Part 2 were deemed sufficient to be able to have at least 3 focus groups for data saturation²⁵.

Part 1: Sensory Evaluation

The sensory evaluation was done within a few days after recruitment by using a cross-over randomized design in February 2013. The design used was adapted from the method of Matias *et al.*¹⁶. The envelope method was used for randomization²⁶ which was done by researchers (MR and CB.). Figure 1 is a representation of the research approach used for Part 1.

Half of the mother-infant pairs started with SQ-LNS A whereas the other half started with SQ-LNS B. The mothers were invited to come to the field station at 08:00 am on each

sensory evaluation day to ensure that the infants were fed within the timeframe in which they would normally consume their first complementary meal of the day.

On day 1, recruited infants were first screened for eligibility. All infants were found eligible and were given 5 g of either SQ-LNS A or B and were observed by a study nurse for at least 20 min to ensure that no adverse reaction occurred.

On day 2, the sensory evaluation was done only if no adverse reaction was observed by the study nurse. The study nurse measured the infant's temperature by using an electronic ear thermometer and children with aural body temperature $> 38^{\circ}\text{C}$ were excluded. Breastfeeding mothers were first asked to breastfeed their child. No other liquids or food were allowed for the next 60 min during which a fieldworker interviewed the mother to complete a socio-economic questionnaire. After the 60 min, the sensory evaluation was started. Under the guidance of a fieldworker, the mother mixed 20 g SQ-LNS with 20 g maize porridge and then tasted a spoonful of the SQ-LNS-mixture. The rest of the SQ-LNS-mixture, together with the standard bowl and spoon used, was weighed before starting to feed the infant. A stopwatch was set at 15 min (maximum time given to consume the paste and porridge mixture) and the countdown started when the mother started feeding her infant the given portion of the SQ-LNS-mixture. The mother could stop if the infant refused to eat further and, if so, this was noted by the fieldworker. The mother was kindly asked to stop feeding her infant when 15 min was over. The remaining SQ-LNS-mixture, together with the bowl and spoon was weighed again; and the amount of SQ-LNS-mixture consumed by the infant was calculated. If there were any left-overs after 15 min, the mother could then continue feeding the infant after the weighing had been done, if the infant still wanted to eat. On day 3 and 4 the second SQ-LNS were given, following the same process. On day 5, all infants were provided with 40 g of the control-porridge (plain maize porridge) without any added SQ-LNS, following the same sensory evaluation procedure described above.

To assess the general liking and acceptance of the SQ-LNS-mixes and control porridge, an adapted 5-point hedonic scale was used on which the points were indicated by a drawing of a facial expression depicting 1 = Disagree (Dislike extremely), 2 = Tend to disagree (Dislike), 3 = Undecided, 4 = Tend to agree (Like) and 5 = Agree (Like extremely)¹⁶. The mother used the 5-point hedonic scale to rate the taste, smell, appearance, colour, texture, mouth-feel and her overall liking, as well as her perception of the infant's liking of the SQ-LNS-mixtures and control porridge.

Part 2: Two-week home-use trial

The home-use trial commenced in April 2013. To avoid a carryover effect from the sensory evaluation, a different group of 40 mother-infant pairs were recruited from the same clinics for the home-use trial. The same informed consent process as for the sensory evaluation was followed. The mother-infant pairs were randomly assigned to one of the two SQ-LNS for home use by researchers (MR. and CB.) by using the envelope method²⁶. Figure 2 is a representation of the research approach used for Part 2.

On the first day, recruited infants were first screened for eligibility at the central study site. All infants were found to be eligible. Eligible infants were given 5 g of the SQ-LNS. The home use was continued only if the study nurse did not observe any negative reaction within 20 min. The mother was interviewed by a fieldworker to complete a socio-economic questionnaire, and was given a 13 day supply (14 minus 1, as 1 tub was used for day 1 on site) of 20 g tubs of either SQ-LNS A or B according to the randomization. The fieldworker demonstrated to the mother how to mix the SQ-LNS with whatever complementary food the mother would be giving for the next 13 days.

Instructions for use were: For the infant's first meal of the day, the mother was asked to dish-out 2–3 tablespoons of the complementary food she was going to give to her infant into a standard serving bowl provided and mix into it one complete 20 g tub of SQ-LNS. The mother was required to keep the empty SQ-LNS tubs in a plastic zip-lock bag provided for the adherence monitoring. She was trained to record on a pictorial "Daily adherence form" the estimated amount the infant had consumed on the day and the mother was also asked to record the type of complementary food used to mix with the SQ-LNS.

A fieldworker visited each mother three times (days 2, 7 and 14) to make sure that the mother understood the use of the SQ-LNS as well as the completion of the daily adherence form. At each visit the fieldworker counted and recorded the used and remaining SQ-LNS tubs and also completed a weekly adherence questionnaire which also included information on any illnesses that may have occurred and the types of food the mothers used with the SQ-LNS.

At the end of the two week period, a fieldworker conducted an exit interview with the mother to get information on the mother's perception of the infant's acceptance of SQ-LNS, the ease of using the SQ-LNS, the possible intra-household distribution of the SQ-LNS as well as the complementary foods used with the SQ-LNS.

Two days following the two weeks home-use trial, FGDs were conducted at the central field station to obtain additional information on the mothers' perceptions on the ease of using the

two SQ-LNS. The FGDs were facilitated by trained facilitators using a structured field guide adapted from Rabiee²⁵. The kind of questions asked that were asked included: “What was your overall feeling about the product?”; “What was your experience of using the product?” and “Would you recommended the product for the feeding of the baby to other mothers/caregivers?”. The participants were allocated into three different discussion groups. Group A: Mothers who used SQ-LNS A; Group B: Mothers who used SQ-LNS B; Group C: Mixed group. The facilitator was aware of the colour codes (for SQ-LNS A or B) but was not aware what the differences were. The same was the case for the participants. Two research assistants took notes, and the discussions were audio taped. Before starting the focus group, mothers were asked to rate the general acceptance of the SQ-LNS-mixture on a scale from 1 to 10. The facilitator then elicited discussion on the practicality and experience of using the SQ-LNS for feeding the infant, as well as general characteristics of the SQ-LNS. The discussions were conducted in the local language, Setswana and lasted 30–45 min depending on information saturation, meaning that no new ideas emerged on the specific topics during a specific FGD²⁵.

Ethics

Ethical approval was obtained from the Ethics Committee of North-West University (NWU-00011-11-A1) and the Ethics Committee of the South African Medical Research Council (EC011-03/2012). Approval and permission to do the study was also given by both the Provincial and District Department of Health, as well as at community health care facility level where recruitment of mother infant pairs was done. The study has been registered with the North West Provincial Department of Health Directorate for Planning, Research, Monitoring and Evaluation. The actual transport costs were reimbursed for each mother participating in the trial.

Data Analysis

Statistical analyses were done using IBM SPSS Statistics 22 package. Socio-economic characteristics of participants were analysed by using descriptive statistics. For Part 1, the median and interquartile range are reported for consumption time and percentages for the hedonic scales. For Part 2, the mean (SD) proportion of the SQ-LNS consumed was calculated. The information on sensory acceptance from the hedonic scales is reported as frequencies.

In Part 2, for the calculation of percentage intake of both SQ-LNS, the standard amount of 5 g given on the first day of phase 2 was excluded. Thus, percentage intake over the two weeks was calculated only from Day 2 as the first determined day.

A reflective notebook was kept by a research assistant to note all verbal communication expressed by the focus group participants. A quality check of the FGD transcripts was done by a research assistant fluent in both English and Setswana and who was present during the FGDs to ensure that the transcripts were translated without the original meaning being lost in translation. The researcher also compared the transcribed work to the notes taken during the discussions. Direct quotes from participants were included in the description of the findings, in order to give more meaning to the context²⁵. The audio tapes were transcribed verbatim and translated to English by Globosonic DFH, a professional company specialized in transcription services before content analysis. Transcripts were analysed according to guidelines given by Rabiee²⁵; a thematic framework was compiled by identifying themes (key words); tabulating was done by managing, cleaning and simplifying the data; and interpretation was done by identifying main expressions.

Results

Participant characteristics

The characteristics of the participants are shown in Table 2. The mean age of the infants was 9.0 ± 2.0 months for Part 1 and 9.0 ± 1.8 months for Part 2. The mothers' mean age was 25.0 ± 5.2 years for part 1 and 26.0 ± 8.1 years for part 2. Fifty percent of the mothers (for both Parts) reported attending school for ≤ 10 years, and $> 60\%$ were single. Electricity was available to $> 90\%$ of households. All households had access to tap water, either from their own tap outside the house (57%) or from a tap inside their house (43%).

Part 1: Sensory Evaluation. Twenty-five mother-infant pairs were recruited, but only 20 mothers came for screening. All of them were found eligible and enrolled in the study. There were 4 dropouts: One infant developed a rash (not related to the product), 2 infants refused to eat the SQ-LNS-mixture, and 1 was withdrawn by the mother for personal reasons. Thus, 16 infants completed the sensory evaluation.

Part 2: Home-use trial. Fifty infant-mother pairs were recruited of whom 40 showed up for the start of the two-week home-use trial. Two mothers in the SQ-LNS B group withdrew their infants from the trial for personal reasons. Thirty-eight infants completed the home-use trial. Twenty-six mothers participated in three FGDs.

Part 1: Sensory Evaluation

All the infants ate the entire given portion of SQ-LNS-mixture (20 g SQ-LNS plus 20 g maize porridge) within the 15 min provided for (Figure 3). The median (interquartile range) time recorded for consumption for SQ-LNS A, -B and the control porridge were 3.2 (1.8, 5.3), 4.3 (2.7, 6.6) and 2.4 (1.8, 2.8) respectively. The frequencies of different time intervals for all three groups are shown in Figure 3

Figure 4 shows the mother's and her child's liking of the SQ-LNS-mixtures and the control porridge. Most (> 70%) of the mothers reported liking the sensory attributes, taste, smell, appearance, colour, texture and mouth-feel. Based on the results from the hedonic scale, categories 4 and 5 of 'tend to agree' and 'agree', respectively were combined because most mothers (> 60%) reported a score of 5 (agree) with a small percentage (< 20%) reporting 4 (tend to agree) for all sensory attributes when considered both mother-and infant hedonic scale. Scores 1 (disagree), 2 (tend to disagree) and 3 (undecided) were reported by < 10% when considering all sensory attributes for both scales. The overall liking for both pastes was 77.8% and 83.3% for SQ-LNS-A and SQ-LNS-B mixture, respectively. Most of the mothers (83.3%) reported not experiencing any problems when feeding both mixtures to their infants.

Table 3 shows the mother's score of her practical experience of using the porridge SQ-LNS-mixtures. Most of the mothers reported that both SQ-LNS were easy to mix with porridge (88.9% and 83.3% for SQ-LNS A- and SQ-LNS B-mixture, respectively). Mothers also reported that they were positive to give both porridge SQ-LNS-mixtures to their infants (83.3% and 72.2% for SQ-LNS A- and B-mixture, respectively). Without providing any specific possible pricing information, most mothers (83.3%) reported that they were willing to buy both SQ-LNS if available on the market.

Part 2: Home-use trial

The mothers reported mixing the given SQ-LNS with a variety of foods like commercial infant cereal (18.4%), bottled pureed infant food (15.8%) and breakfast cereals (13.2%) and some food items like soft maize porridge, mabele porridge and mashed potatoes were reported by less than 10% of the mothers respectively. Over the two week period, the mean percentage of reported consumption was 65.3 ± 34.2 and 62.0 ± 31.3 of the recommended amount for SQ-LNS A and B, respectively (Table 4). The mother's own acceptance and perception of her infant's acceptance is reported in Table 4. At the end of the home use trial,

more than 80% of the mothers reported that both SQ-LNS were acceptable to them and their infants.

Three FGDs were conducted including 8 participants each for the SQ-LNS A, and B, and 10 participants for the mixed group. FGDs reflected that the mothers were happy with their infants liking and eating of the SQ-LNS-mixture, as reflected in the comment “*Baby enjoys it*”. Some mothers mentioned that their infants preferred the SQ-LNS-mixture over the complementary foods they normally fed their infants (“*Baby does not want to eat something else*”). Some mothers also reported perceiving their infants showing more positive active behaviour after eating the SQ-LNS-mixture (“*baby has more energy*”). Some mothers felt that the SQ-LNS increased their infants’ appetite with quotes like: “*I like this product because it gave my baby an appetite, she never ate a lot*” and “*After giving her this paste she eats more than she used to and I like that*”. The overall experience for both SQ-LNS was favourable in terms of ease of mixing and convenience of adding to the different types of complementary foods used. Some mothers spontaneously reported that they were willing to buy the SQ-LNS (A/B) and would encourage other mothers to give the pastes to their infants. No pricing information formed part of the discussion guide.

Discussion

This study indicated that, based on sensory characteristics, both soy-based SQ-LNS, when mixed with maize meal porridge, were acceptable in terms of taste, appearance, colour, texture, mouth-feel and overall-liking. The aim of the study was not to determine which SQ-LNS was liked most, but rather to assess whether both SQ-LNS were acceptable. The control porridge was included to ensure that the porridge that was used for mixing the SQ-LNS was acceptable and did not negatively affect acceptance of the SQ-LNS mixture. Both SQ-LNS mixed in maize porridge were perceived to have a better taste, texture, colour and appearance than the control maize porridge by the mothers (Fig. 4). When considering the sensory acceptance, Menella et al.²⁷ assumed that variance in responses to tastes may be affected by the infant’s feeding history in terms of breast milk and/or formula milk consumption. According to Adu-Afarwuah et al.¹⁵ either the proportion of offered food consumed or the duration taken to consume the amount offered without force-feeding can be used as a proxy indicator for acceptability. Soy-based products have not previously been reported for complementary feeding in the South African setting⁹ and the infants would not therefore have been expected to be familiar with the sensory characteristics of the two given SQ-LNS-mixtures. Similar products tested in other countries did not contain added DHA and

ARA¹³⁻¹⁶. Therefore, based on the short time taken to consume both SQ-LNS-mixtures and despite lack of familiarity with such SQ-LNS-mixtures the products tested can be considered to have been acceptable to the infants.

The home-use trial supported the findings from the sensory evaluation in terms of the perceptions towards the novel SQ-LNS. More than 80% of the mothers and their infants (based on mothers' perceptions) reported both SQ-LNS to be acceptable at home-use level. As was the case in the study done by Adu-Afarwuah et al.¹⁵, this study presented a challenge in terms of determining the infants' perceived liking of the SQ-LNS which relied entirely on the mother's perception. However, the mother's level of acceptance and perceptions are essential, as she is the person primarily responsible for deciding what the child eats²⁸.

On average, 62.0 - 65.3% of the recommended amount of both SQ-LNS was consumed over the 2 weeks home use period. This lower than recommended amount consumed may have an influence on the intake of the beneficial constituents like DHA, ARA, phytase and L-lysine. The randomized controlled trial that followed after this trial will indicate the impact of the SQ-LNS on the growth and development of infants in this age category in relation to the actual intake of the SQ-LNS over a period of six months. In the home-use trial, a variety as foods, e.g. pureed jarred infant foods, commercial infant cereals, soft maize porridge and vegetables were used to mix the SQ-LNS into. Most mothers (> 80%) reported ease of mixing with the SQ-LNS despite the variety of foods used and this may have contributed towards the mothers reported acceptance. In practice this information is of interest as it implies that the sensory characteristics of both SQ-LNS A and B are likely applicable to a variety of food items used in the home.

During the FGDs following the home use period, some mothers referred to being pleased with perceived improvements in their infant's appetite and activity level after consuming the SQ-LNS-mixtures. Peltó et al.²⁹ conducted a focused ethnographic study in urban and rural communities in South Africa and argued that the strongest association South African mothers had with a healthy child was an active child with a healthy appetite and steady weight gain. The mothers' perceptions in this study that their children's appetite and activity level improved are also in line with mother's comments reported in acceptability trials done in Ghana, Malawi, Guatemala and Burkina Faso¹³⁻¹⁶. Although pricing did not form part of the FGD guide, mothers' expressed willingness to buy the products. This further confirmed a positive attitude of the mothers towards the two SQ-LNS.

Although the SQ-LNS were shown to be acceptable in our study, for large scale implementation, good communication and promotion (i.e., development of educational messages accompanying the SQ-LNS distribution) would be needed to ensure that sufficient amounts are eaten by the infant, so that the infant can get the full benefit of the SQ-LNS²⁹. It is well recognised that communication and nutrition education form an integral part of any sustainable strategy aimed at improving the quality of complementary feeding³.

A strength of the study is that all participating infants were tested for any allergic reactions which might occur from these novel SQ-LNS. A study nurse was appointed for the duration of the study specifically for this purpose. The SQ-LNS tested are relevant to all areas where under nutrition is prevalent and the methodology used may provide a template that could be used in a different study population. The fact that, in part 1, the control porridge was always last, may be seen as a limitation as this may have resulted in bias in the reporting of the sensory evaluation. It should however be noted that the aim of the sensory evaluation of the control porridge was to verify that the porridge used for mixing the SQ-LNS with was also accepted and did not negatively influence the sensory evaluation of the SQ-LNS mixture.

In conclusion, we found that the sensory characteristics of these two novel SQ-LNS, when mixed into maize porridge and or other complementary foods, were found acceptable to the mother-infant pairs and that the mothers experienced the use of these products positively. The results suggest that these newly developed SQ-LNS may have potential for successful use in food-based interventions aimed at improving the nutritional quality of commonly used complementary foods in resource poor settings.

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Authors' contributions

Marinel Rothman: Study design; data collection, -analysis and –interpretation; wrote the first draft of the paper.

Cristiana Berti: Study design; data collection, -analysis and –interpretation; revision of paper.

Cornelius M Smuts: Scientific input in data analysis and –interpretation; revision of paper.

Mieke Faber: Scientific input in data analysis and –interpretation; revision of paper.

Namukolo Covic: Study design; scientific input in data interpretation; revision of paper.

All authors read and approved the final version of the paper.

Conflict of interest

This manuscript is an original work, and has not been published elsewhere. None of the authors have any conflict of interest.

References

1. Black RE, Victora CG, Walker SP, Bhutta ZA, Christian P, De Onis M, Ezzati M, Grantham-McGregor S, Katz J, Martorell R. Maternal and child undernutrition and overweight in low-income and middle-income countries. *Lancet* 2013;382:427-51.
2. WHO HQ/AFRO. Maternal, infant and young child nutrition in East and Southern African countries: moving to national implementation, report of a World Health Organization workshop, Entebbe, Uganda, 26–28 November 2013. 2014.
3. Bhutta ZA, Das JK, Rizvi A, Gaffey MF, Walker N, Horton S, Webb P, Lartey A, Black RE. Evidence-based interventions for improvement of maternal and child nutrition: what can be done and at what cost? *Lancet* 2013;382:452-77.
4. Shisana O, Labadarios D, Rehle T, Simbaji L, Zuma K, Dhansay A, Reddy P, Parker W, Hoosain E, Naidoo P, Hongoro C, Mchiza Z, Steyn NP, Dwane N, Makoae M, Maluleke T, Ramlagan S, Zungu N, Evans MG, Jacobs L, Faber M, & the SANHANES-1 Team.. The South African National Health and Nutrition Examination Survey:SANHANES-1. HSRC Press; 2014.
5. Tylleskär T, Jackson D, Meda N, Engebretsen IMS, Chopra M, Diallo AH, Doherty T, Ekström E-C, Fadnes LT, Goga A. Exclusive breastfeeding promotion by peer counsellors in sub-Saharan Africa (PROMISE-EBF): a cluster-randomised trial. *Lancet* 2011;378:420-7.
6. MacIntyre U, De Villiers F, Baloyi P. Early infant feeding practices of mothers attending a postnatal clinic in Ga-Rankuwa. *S Afr J Clin Nutr* 2005;18:70-5.
7. Kruger R, Gericke G. Breast feeding practices of mothers with children (aged 0-36 months) in a rural area of South Africa: A qualitative approach. *JFECS* 2001;29:60-71.
8. Faber M. Complementary foods consumed by 6–12-month-old rural infants in South Africa are inadequate in micronutrients. *Public Health Nutr* 2005;8:373-81.
9. Faber M, Laubscher R, Berti C. Poor dietary diversity and low nutrient density of the complementary diet for 6- to 24-month-old children in urban and rural KwaZulu-Natal, South Africa. *Matern Child Nutr* 2014. doi:10.1111/mcn.12146.

10. Arimond M, Zeilani M, Jungjohann S, Brown KH, Ashorn P, Allen LH, Dewey KG. Considerations in developing lipid-based nutrient supplements for prevention of undernutrition: experience from the International Lipid-Based Nutrient Supplements (iLiNS) Project. *Matern Child Nutr* 2013. doi: 10.1111/mcn.12049.
11. Siega-Riz AM, Estrada Del Campo Y, Kinlaw A, Reinhart GA, Allen LH, Shahab-Ferdows S, Heck J, Suchindran CM, Bentley ME. Effect of supplementation with a lipid-based nutrient supplement on the micronutrient status of children aged 6–18 months living in the rural region of Intibucá, Honduras. *Paediatr Perinat Epidemiol* 2014;28:245-54.
12. Iannotti LL, Dulience SJL, Green J, Joseph S, François J, Anténor M-L, Lesorogol C, Mounce J, Nickerson NM. Linear growth increased in young children in an urban slum of Haiti: a randomized controlled trial of a lipid-based nutrient supplement. *Am J Clin Nutr* 2014;99:198-208.
13. Phuka J, Ashorn U, Ashorn P, Zeilani M, Cheung YB, Dewey KG, Manary M, Maleta K. Acceptability of three novel lipid-based nutrient supplements among Malawian infants and their caregivers. *Matern Child Nutr* 2011;7:368-77.
14. Hess SY, Bado L, Aaron GJ, Ouédraogo JB, Zeilani M, Brown KH. Acceptability of zinc-fortified, lipid-based nutrient supplements (LNS) prepared for young children in Burkina Faso. *Matern Child Nutr* 2011;7:357-67.
15. Adu-Afarwuah S, Lartey A, Zeilani M, Dewey KG. Acceptability of lipid-based nutrient supplements (LNS) among Ghanaian infants and pregnant or lactating women. *Matern Child Nutr* 2011;7:344-56.
16. Matias S, Chaparro C, Perez-Exposito A, Peerson J, Dewey K. Acceptability of lipid-based nutrient supplement among Guatemalan infants and young children. Technical report. Washington, D.C.:FHI 360/FANTA-2 2011:1-33.
17. Gibson RS, Bailey KB, Gibbs M, Ferguson EL. A review of phytate, iron, zinc, and calcium concentrations in plant-based complementary foods used in low-income countries and implications for bioavailability. *Food Nutr Bull* 2010;31:134-46.
18. Georgieff MK. Nutrition and the developing brain: nutrient priorities and measurement. *Am J Clin Nutr* 2007;85:614S-20S.

19. Hoffman DR, Boettcher JA, Diersen-Schade DA. Toward optimizing vision and cognition in term infants by dietary docosahexaenoic and arachidonic acid supplementation: a review of randomized controlled trials. *Prostaglandins Leukot Essent Fatty Acids* 2009;81:151-8.
20. Hoffman DR, Theuer RC, Castañeda YS, Wheaton DH, Bosworth RG, O'Connor AR, Morale SE, Wiedemann LE, Birch EE. Maturation of visual acuity is accelerated in breast-fed term infants fed baby food containing DHA-enriched egg yolk. *J Nutr* 2004;134:2307-13.
21. Clandinin MT, Van Aerde JE, Merkel KL, Harris CL, Springer MA, Hansen JW, Diersen-Schade DA. Growth and development of preterm infants fed infant formulas containing docosahexaenoic acid and arachidonic acid. *J Pediatr* 2005;146:461-8.
22. Cercamondi CI, Egli IM, Mitchikpe E, Tossou F, Hessou J, Zeder C, Hounhouigan JD, Hurrell RF. Iron bioavailability from a lipid-based complementary food fortificant mixed with millet porridge can be optimized by adding phytase and ascorbic acid but not by using a mixture of ferrous sulfate and sodium iron EDTA. *J Nutr* 2013;143:1233-9.
23. Du Plessis LM, Kruger H, Sweet L. Complementary feeding: a critical window of opportunity from six months onwards. *S Afr J Clin Nutr* 2013;26:S129-S40.
24. Marfell-Jones M, Stewart A, De Ridder H. ISAK accreditation handbook. Upper Hutt, New Zealand: International Society for the Advancement of Kinanthropometry 2012.
25. Rabiee F. Focus-group interview and data analysis. *Proc Nutr Soc* 2004;63:655-60.
26. Schulz KF, Grimes DA. Allocation concealment in randomised trials: defending against deciphering. *Lancet* 2002;359:614-8.
27. Mennella JA, Forestell CA, Morgan LK, Beauchamp GK. Early milk feeding influences taste acceptance and liking during infancy. *Am J Clin Nutr* 2009;90:780S-8S.
28. Ruel MT, Alderman H. Nutrition-sensitive interventions and programmes: how can they help to accelerate progress in improving maternal and child nutrition? *Lancet* 2013;382:536-51.

29. Pelto GH, Armar-Klemesu M, Siekmann J, Schofield D. The focused ethnographic study 'assessing the behavioral and local market environment for improving the diets of infants and young children 6 to 23 months old' and its use in three countries. *Matern Child Nutr* 2013;9:35-46.

Table 1. Nutrient content of the two SQ-LNS pastes

	SQ-LNS A	SQ-LNS B
Amount (g) (1 portion)	20	20
Energy (kcal)	114	113
Energy density (kcal/g)	5.7	5.7
Protein (g)	3.0	3.7
% calories from protein	10	13
Fat (g)	8.0	8.8
% calories from fat	63	70
Linoleic acid (g)	1.5	1.8
α -linolenic acid) (mg)	265	348
n-6/n-3 ratio	5.7	5.0
Docosahexaenoic acid (mg)	-	75
Arachidonic acid (mg)	-	75
Vitamin A (μ g RE)	200	200
Vitamin D (μ g)	2.5	2.5
Vitamin E (mg)	2.5	2.5
Vitamin K (μ g)	7.5	7.5
Thiamin (mg)	0.25	0.25
Riboflavin (mg)	0.25	0.25
Niacin (mg)	3.0	3.0
Pantothenate (mg)	1.0	1.0
Vitamin B6 (mg)	0.25	0.25
Biotin (mg)	4.0	4.0
Folic acid (μ g)	80	80
Vitamin B12 (μ g)	0.45	0.45
Vitamin C (mg)	23.3	23.3
Calcium (mg)	250	396
Iodine (μ g)	45	45
Iron (mg)	5.8	5.8
Zinc (mg)	6.2	6.2

	SQ-LNS A	SQ-LNS B
Copper (mg)	0.28	0.28
Selenium (µg)	8.5	8.5
Magnesium (mg)	-	30
Manganese (mg)	-	0.6
Phosphorus (mg)	-	230
Potassium (mg)	-	257
L-Lysine (mg)	-	160
Phytase (FTU) ¹	-	200

¹ FTU: unit of phytase activity

Table 2. Characteristics of the participants for Part 1 and Part 2 of the study

Participant characteristics	Part 1	Part 2
Infant-mother pairs (n)	16	38
Infants' mean (\pm SD) age, months	9 \pm 2.0	9 \pm 1.8
Mothers		
Mean (\pm SD) age, years	25 \pm 5.2	26 \pm 8.1
Unmarried n (%)	14 (88)	26 (68)
School attendance n (%)	16 (100)	34 (89)
10 years and less	8 (50)	17 (50)
11 and 12 years	8 (50)	16 (47)
Source of drinking water n (%)		
Own tap outside house	9 (56)	22 (58)
Own tap inside house	7 (44)	16 (42)
Only cold running water available n (%)	9 (56)	36 (95)
Electricity availability inside household n (%)	15 (94)	36 (95)

Table 3. Hedonic scores reported by the mother on her practical experience of using the two porridge-SQ-LNS-mixtures in Part 1 of the study

Practical attribute	SQ-LNS-A mixture (n = 18)	SQ-LNS-B mixture (n = 18)	Control porridge (n = 18)
Easy to mix n (%)			NA
▪Agree	16 (88.9)	15 (83.3)	
#Disagree	0	1(5.6)	
Liked giving n (%)			NA
▪Agree	15 (83.3)	13 (72.2)	
#Disagree	1 (5.6)	3 (16.7)	
Willingness to buy if available n (%)			NA
▪Agree	15 (83.3)	15 (83.3)	
▪Agree	1 (5.6)	1 (5.6)	
#Disagree			

▪ - The term 'Agree' refers to the combination of 'agree' and 'tend to agree'; #The term 'Disagree' is the combination of 'disagree' and 'tend to disagree'.

* - $p < 0.05$

NA – Not Applicable

Table 4. Frequencies of the mother's own acceptance and perception of infant acceptance of the SQ-LNS-pastes after two weeks of home-use (Part 2 of the study) expressed as percentage.

		SQ-LNS A (<i>n</i> = 20)	SQ-LNS B (<i>n</i> = 18)
Percentage of SQ-LNS consumed (Mean ± SD)		65.3 ± 34.2	62.0 ± 31.3
Percentage (95%CI) SQ-LNS acceptable to mother	Yes	90 (75, 100)	89 (72, 100)
	No	5 (0, 15)	6 (0, 17)
	Not sure	0	6 (0, 17)
	No answer	5 (0, 15)	0
Percentage (95%CI) SQ-LNS acceptable to infant as perceived by the mother	Yes	80 (60, 95)	89 (72, 100)
	No	15 (0, 35)	0
	Not sure	0	11 (0, 28)
	No answer	5 (0, 5)	0

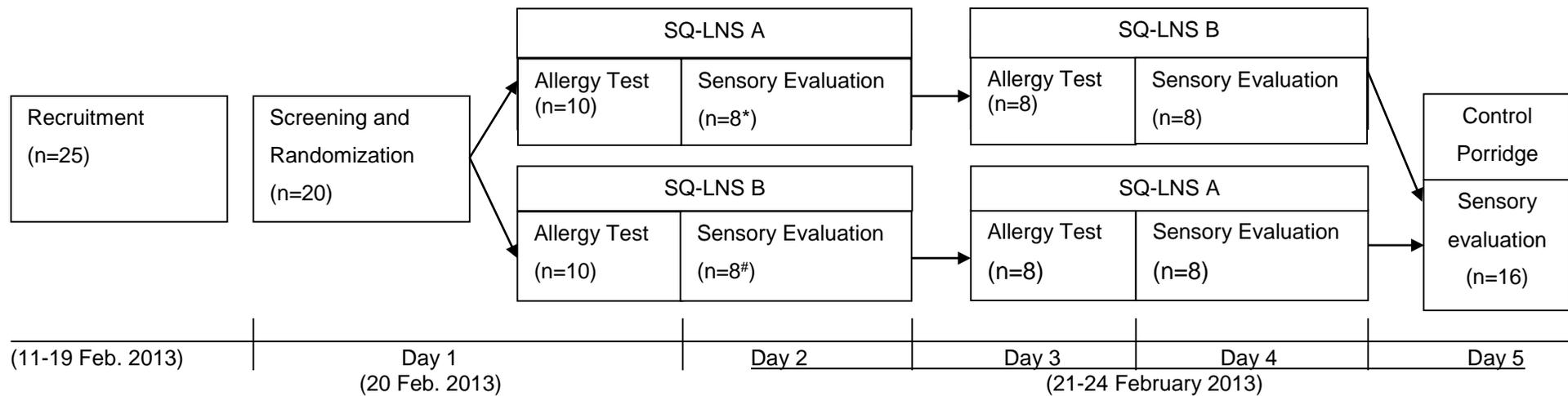


Figure 1. Representation of the research approach used for Part 1.

* 2 Drop-outs from day 1: 1 infant developed a rash (not related to product) and 1 mother withdrawn from the study for personal reasons

2 Drop-outs from day 1: infants refused to eat the product

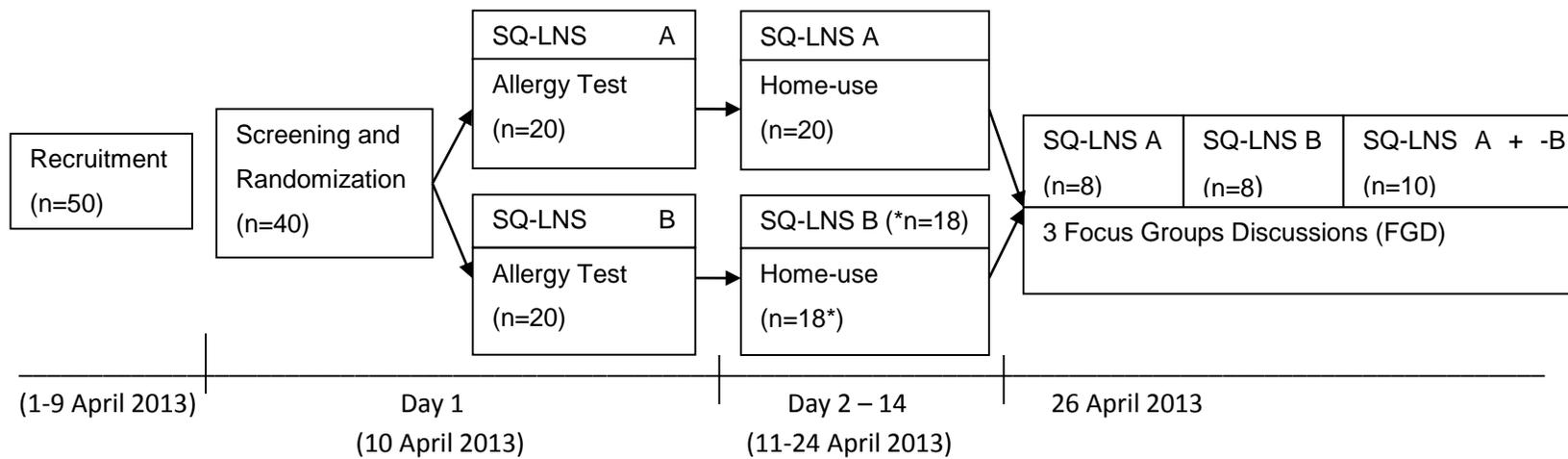


Figure 2. Representation of the research approach used for Part 2.

* 2 Mothers in SQ-LNS B group withdrew for personal reason

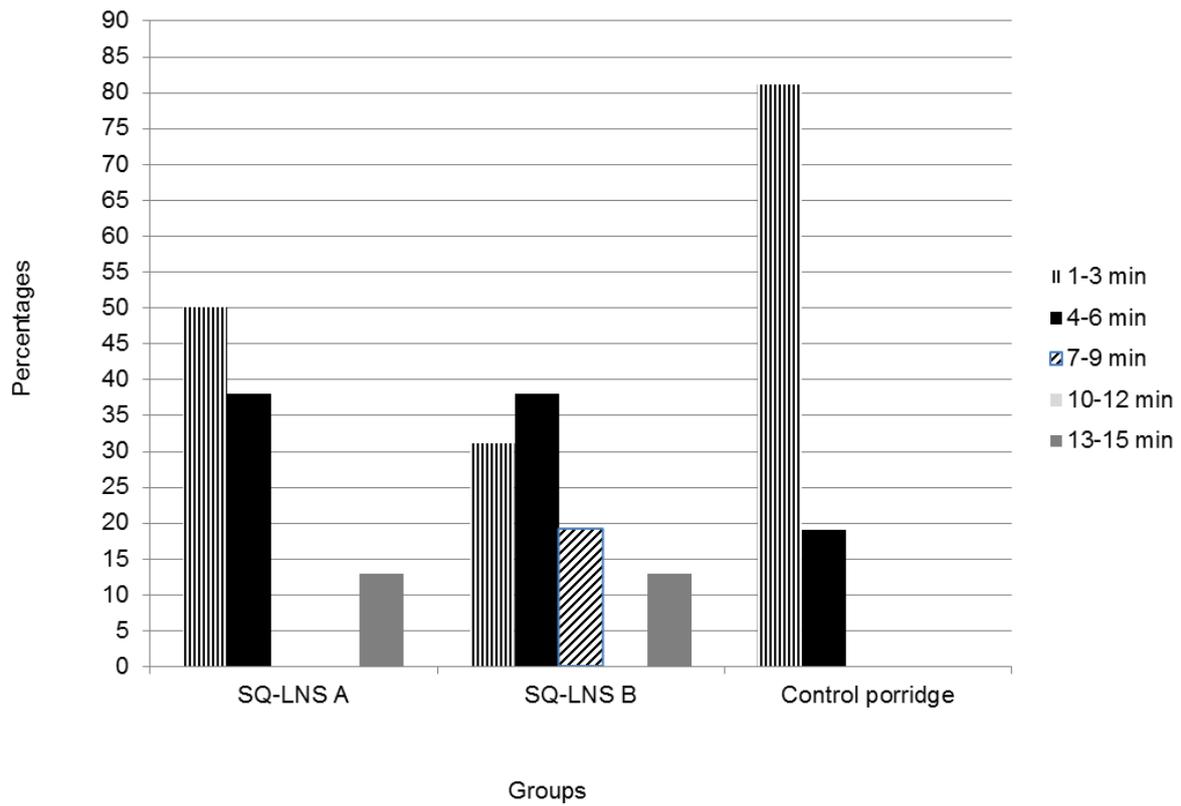


Figure 3. Time used to consume SQ-LNS A and –B and control porridge respectively

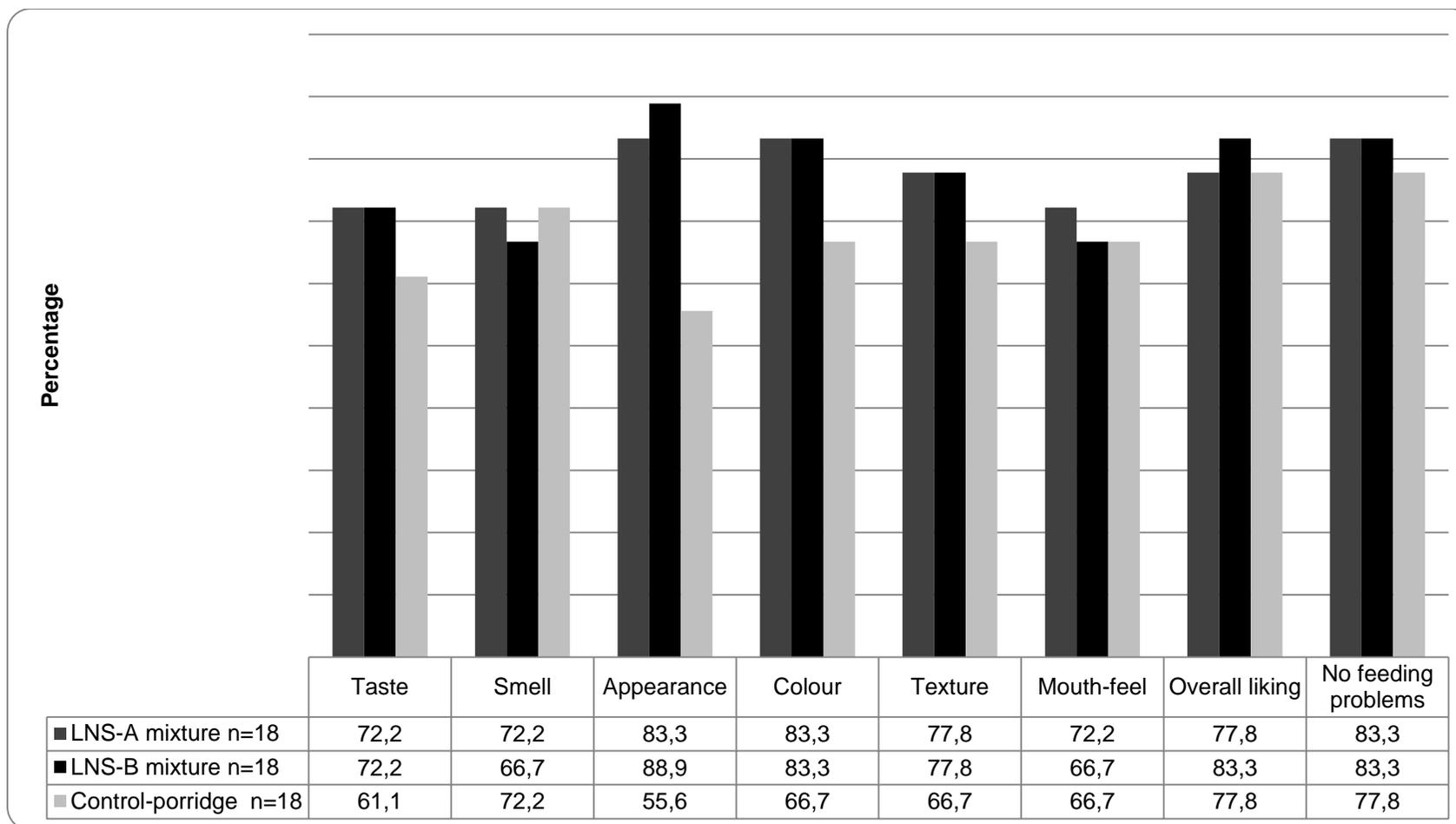


Figure 4. Percentage of mothers who reported 'Agree' on both her own liking and her perception of the infant's liking of the two porridge-SQ-LNS-mixtures and the control porridge in Part 1 of the study; *The term 'Agree' refers to the combination of 'agree' and 'tend to agree'

CHAPTER 6

Conclusion

Infancy is a critical time of development where rapid physical and mental development occurs. Infant feeding practices have an effect on the growth and development (Sudfeld *et al.*, 2015; Nyaradi *et al.*, 2013; Saha *et al.*, 2008).

Using baseline data of a randomized controlled trial, this study investigated feeding practices, nutritional status, and psychomotor development of 6-month-old infants (n=750) residing in a peri-urban area of poor socio-economic status.

Feeding practices

Introduction of liquids and semi-solid/solid foods

All children were initially breastfed, as never having received breast milk was an exclusion criteria for enrolment into the randomized controlled trial. Liquids, mostly water and formula milk, were introduced at age 0–2 months for 46.4% of the infants. Because of the early introduction of liquids, exclusive breastfeeding was not widely practiced; 48.9% of infants were exclusively breastfed only up to age of 0–2 months.

More than half (60.5%) of the infants had been introduced to semi-solid/solid at age 4 months or younger; the most popular first foods being commercial infant cereals and jarred infant foods. Our results reflect the effect of urbanisation on infant feeding practices in this study population, as characterised by frequent consumption of commercial infant foods regardless of the fact that a high number of households were recipients of social grants which is indicative of a low socio-economic environment. Our findings are in line with national data which reported that 63.5% of infants were given semi-solid foods at an average age of 3.6 months, with commercial infant cereal being the most common semi-solid initial food (HSRC, 2013). Early introduction of complementary feeding may be a risk factor for early exposure to microbial pathogens, which increases the infant's risk of infection with diarrheal disease and malabsorption, and additionally, most digestive enzymes are inadequate for the first 6 months of life (WHO, 2003).

Milkfeeds and dietary intake at age 6 months

Most (70.1%) of the infants in our study population were still being breastfed at the age of 6 months. For 52.7% of all infants breast milk was the only milk feed given, 14.9% received breast milk and formula milk, and for 27.7% formula milk was the only milk feed given. Complementary foods frequently consumed were mostly infant cereal (68.1%) and jarred

infant foods (22.7%). Sugar and/or salt were added to the infant's food at least 4 days per week for 27.9% and 8.7% of the infants respectively. It has been reported that many commercial baby foods such as infant cereals and jarred baby foods contain a high sugar content, which can be seen as a potential risk factor for future health problems (Cogswell et al., 2015; Walker & Goran, 2015). A small number of infants were given vegetables and fruits (both round 3%) frequently. Animal-sourced foods and food items rich in essential fatty acids like eggs, oil and peanut butter were also not frequently given. For consumers, commercial infant cereals and fortified formula feeds provided significant amounts of dietary iron (3.90 and 3.42 mg, respectively). In contrast, for consumers of maize meal porridge, maize meal provided <1 mg iron. Although a high percentage of infants in our study population consumed fortified formula milk and infant cereals on a frequent basis, the desired nutrient density of the complementary diet (excluding breast milk) was not met – especially for the “problem nutrients” iron, zinc, and calcium.

Nutritional status

Prevalence of stunting, anaemia and iron deficiency

At this young age, almost 30% of infants were stunted, a percentage that is already higher than the global (25%) and national prevalence (22%) for children younger than 5 years (UNICEF, 2014; HSRC, 2013). Almost 40% of the infants were anaemic, which resembles the national prevalence (38.7%) reported for children aged 1–3 years in 2005 (Labadarios et al., 2008). Based on Fer, 16.1% of the infants were ID and 6% had IDA. Transferrin receptor concentrations indicated an ID and IDA prevalence of 30.1% and 13.2%, respectively.

Association of feeding practices, iron status and anaemia.

A shorter duration of exclusive breastfeeding and frequent consumption of formula milk showed to have a protective effect on anaemia and ID. A possible explanation for the lower prevalence of anaemia in infants who were exclusively breastfed up to only age 0–2 months might be that infants were introduced to fortified formula milk and infant cereal from an early age. The introduction of solids to infants in developing countries – at the age of 4 months versus 6 months of age – showed significant improvements in haemoglobin, an outcome that resembles our findings (Qasem et al., 2015). Although the micronutrient nutrient density of the complementary diet did not differ between the anaemic and non-anaemic infants, when considering total nutrient intake, non-anaemic infants had a higher intake for most of the micronutrients (including iron), probably because of the higher number of infants who were given fortified infant formula feeds.

Various factors other than feeding practices and dietary intake may affect the prevalence of anaemia, iron deficiency and IDA in infants. The anaemia and iron-status of infants aged 6–8 months should also be interpreted within context of the infant's iron stores at birth. Iron status during the first 6 months of life is dependent on iron stores at birth and is primarily dependant on birth weight, maternal prenatal iron status and the timing of clamping of the umbilical cord (Dewey & Chaparro 2007). Adequate iron status is critical for healthy cognitive development in infants (Hermoso et al. 2011; Prado & Dewey 2014).

The investigation of early feeding practices, anaemia and iron status in relation to psychomotor development and growth will provide a more conclusive representation of infant development

Feeding practices, nutritional status and psychomotor development

Infants who were exclusively breastfed up to age 0-2 months showed significantly higher parent rating scores versus those who were exclusively breastfed up to age 5–6 months. This finding may be supported by the evidence that infants who were fed formula milk for a longer duration had a smaller chance of being either anaemic and/or iron deficient. It is well known that iron deficiency is linked with poor cognitive development (Beard, 2008). Anaemic infants had lower scores for both eye-hand and locomotor activities, and as a result combined psychomotor development scores whereas infants suffering from ID and IDA had significantly lower combined psychomotor development scores. Therefore, the associations found between feeding practices and psychomotor development may be explained by the iron status of the infants as a consequence of feeding practices.

Anaemia, iron-status, and psychomotor development of infants aged 6–8 months should be interpreted within context, especially when protective and risk factors are simultaneously present. Development during infancy is seen as a dynamic interplay between biological and environmental factors (Fernald *et al.*, 2009). Still, interrelated factors like iron and iodine deficiency, chronic undernutrition (stunting), and poor stimulation at home are key risk factors for delayed cognitive development (Grantham-McGregor *et al.*, 2014). Our study showed that iron deficiency, anaemia, and chronic undernutrition (stunting) were associated with lower psychomotor scores.

Stunted infants had significantly lower scores for combined psychomotor skills and parent rating scores. Infants with a low birth weight (< 2.50 kg) also scored lower in combined psychomotor and parent rating scores. Stunting during the first thousand days may indicate poorer cognitive and educational outcomes later in life (Black *et al.*, 2013). The first 6 months of life is seen as a period of maximal growth velocity in healthy infants and is also seen as a period of critical long-term neurodevelopment (Pongcharoen *et al.*, 2012). It is, therefore,

imperative to identify undernutrition in infants in this very young age group (Kerac *et al.*, 2011) while stressing the need for intervention at an earlier age (Prendergast & Humphrey, 2014) – a process that can already be started by advising women of childbearing age. Intermittent iron supplementation can reduce the risk of anaemia by 49% and iron deficiency by 76% in children younger than 24 months (Bhutta *et al.*, 2013).

Our study population showed that, regardless of the evidence that infants consumed fortified commercial infant cereal before 6 months of age, the nutrient density of the complementary diet was still less than the required density for key nutrients like iron, zinc and calcium. Intervention in the form of home fortification must therefore be explored.

Home fortification of complementary foods has been proposed as an innovative intervention for improving micronutrient intake in children younger than 2 years. Products that can be used for home fortification include nutritabs, micronutrient powders and lipid-based nutritional supplements (LNSs) (Gibson, 2011). LNSs rich in energy, protein and EFA are produced in a food base and the ration size for infants is designed to avoid displacement of breastmilk and also allow dietary diversity (Arimond *et al.*, 2013)

Acceptance of 2 novel SQ-LNS for complementary feeding by infants and their primary caregivers.

Our acceptability trial indicated that, based on sensory characteristics, novel soy-based SQ-LNS, when mixed with maize meal porridge, were acceptable in terms of taste, appearance, colour, texture, mouth-feel and overall-liking (Rothman *et al.*, 2015). The acceptability trial presented a challenge in terms of determining the infants' perceived liking of the SQ-LNS which relied entirely on the mother's perception. However, the mother's level of acceptance and perceptions are essential, as she is the person primarily responsible for deciding what the infant eats (Ruel *et al.*, 2013). The home-use trial found that more than 80% of the mothers and their infants (based on mothers' perceptions) reported SQ-LNS to be acceptable at home-use level. In support, the FGDs revealed that some mothers referred to being pleased with perceived improvements in their infant's appetite and activity level after consuming the SQ-LNS-mixtures which are also in line with mother's comments reported in acceptability trials done in Ghana, Malawi, Guatemala and Burkina Faso on SQ-LNS (Adu-Afarwuah *et al.*, 2011; Hess *et al.*, 2011; Matias *et al.*, 2011; Phuka *et al.*, 2011).

Limitations and strengths

This was a cross-sectional study and therefore all conclusions based on the results should be interpreted carefully as no causation can be concluded. The advantage of this cross-sectional study is that data was collected from a relatively large sample size (n=750) within a very

narrow age bracket, which may reflect relevant associations. This study is the first to provide evidence on associations between feeding practices, nutritional status and psychomotor development of 6-month-old infants in South Africa.

In terms of dietary intake, one limitation of this study is the use of a single 24-hour recall as this provides information on nutrient intake on group level, but not at the individual level. Early feeding practices were assessed retrospectively and this information therefore relied on the respondent's memory.

Implications

National policy of infant and young child feeding aims to define strategies and actions which support and protect optimal infant and young child feeding practices within the context of HIV. In order to provide continuity support at community level, a public health and primary health care approach is imperative to implement these strategies and actions. These strategies and activities are driven by best available evidence (DOH, 2013).

In 2011, the National Department of Health phased out the distribution of free infant formula as part of prevention of mother-to-child transmission. Free infant formula may only be provided for infants with specific medical conditions. Our study indicated that more than 40% of the infants received formula milk at the age of 6 months. Primary health care services (e.g. local clinics) and community health workers should therefore inform mothers/caregivers about the importance of breast milk. However, the decision to breastfeed is a personal matter and depends on various factors like cultural practices and the home-environment (Nor *et al.*, 2012). Non-breastfed mothers should be informed about age specific types of infant formula and how to prepare formula milk feeds in a hygienically safe manner (DOH, 2013). Irrespective of the outcome of this cross-sectional study the promotion of good breast feeding practices is imperative as the advantages of breast feeding are far beyond the scope and implications of the associations indicated by this study. The study though provide some evidence that feeding practices at a very young age, including breast feeding practices, can have implications on nutritional status and/or iron status with consequences on infant development. It further emphasises the importance of iron nutrition and iron status based on its association with psychomotor development. The study provides further evidence that infants are at risk at 6 months of age for anaemia and ID and that complementary food provided at this critical stage of development can contribute to improve their iron status and nutrient intake important for more optimal development. The intervention that follows these baseline observations will in fact address these issues by providing the infants with acceptable SQ-LNS (Rothman *et al.* 2015) that contains minerals and vitamins as well as essential fatty acids.

Recommendations

- Our study evidenced that the early introduction of fortified formula milk and commercial infant cereal have a protective effect against anaemia and iron deficiency, which in turn contribute to improved psycho-motor development. We recommend that the haemoglobin concentration of infants be tested already before 6 months of age in order to detect and address anaemia already at an early age.
- We found that SQ-LNS was acceptable in this study community. The acceptance of these products must also be determined in different communities with different cultural backgrounds and feeding practices.
- The drivers for frequent consumption of commercial infant foods within the context of low socio-economic environments should be explored
- In spite of mandatory fortification of maize meal in South Africa, maize meal porridge did not significantly contribute to dietary iron (for those who consumed maize meal porridge). The National food fortification programme should therefore consider fortifying a maize meal product targeting infants who consume a small amount of food, but have high nutrient requirements because of rapid growth and development.
- Products for home fortification addressing iron deficiency in infants should be developed and the impact thereof should be evaluated randomized controlled trials.
- In our study we looked at early feeding practices, nutritional status and psychomotor development. For future studies, it is recommended that, in addition, stimulation of the child is also assessed.

References

Adu-Afarwuah, S., Lartey, A., Zeilani, M., Dewey, K.G. 2011. Acceptability of lipid-based nutrient supplements (LNS) among Ghanaian infants and pregnant or lactating women. *Maternal and child nutrition*, 7:344-56

Arimond, M., Zeilani, M., Jungjohann, S., Brown, K.H., Ashorn, P., Allen, L.H., *et al.* 2013. Considerations in developing lipid-based nutrient supplements for prevention of undernutrition: experience from the International Lipid-Based Nutrient Supplements (iLiNS) Project. *Maternal and child nutrition*, doi: 10.1111/mcn.12049.

Beard, J.L. 2008. Why iron deficiency is important in infant development. *The Journal of nutrition*, 138(12):2534–2536.

Bhutta, Z.A., Ahmed, T. Black, R.E., Cousens, S., Dewey, K., Giugliani, E., *et al.* 2008. What works? Interventions for maternal and child undernutrition and survival. *The lancet*, 371(9610):417-440.

Black, R.E., Alderman, H., Bhutta, Z.A., Gillespie, S., Haddad, L., Horton, S., *et al.* 2013. Maternal and child nutrition: building momentum for impact. *The lancet*, 382(9890):372-375.

Cogswell, M. E., Gunn, J. P., Yuan, K., Park, S. & Merritt, R. 2015. Sodium and sugar in complementary infant and toddler foods sold in the United States. *Pediatrics*, 2014–3251.

Dewey, K.G. & Chaparro, C.M. 2007. Session 4: mineral metabolism and body composition iron status of breast-fed infants. *Proceedings of the Nutrition Society*, 66(03):412-422.

Fernald, L.C., Kariger, P., Engle, P. & Raikes, A. 2009. Examining early child development in low-income countries. Washington, DC: The World Bank.

Gibson, R. 2011. Strategies for preventing multi-micronutrient deficiencies: a review of experiences with food-based approaches in developing countries. (*In* Thompson B, Amoroso, L. eds. Combating micronutrient deficiencies: food-based approach. CABI and FAO: Oxfordshire, UK, pp 1-92).

Grantham-McGregor, S.M., Fernald, L.C., Kagawa, R. & Walker, S. 2014. Effects of integrated child development and nutrition interventions on child development and nutritional status. *Annals of the New York Academy of Sciences*, 1308(1):11–32.

Hermoso, M., Vollhardt, C., Bergmann, K. & Koletzko, B. 2011. Critical micronutrients in pregnancy, lactation, and infancy: considerations on vitamin D, folic acid, and iron, and priorities for future research. *Annals of nutrition and metabolism*, 59(1):5-9.

- Hess, S.Y., Bado, L., Aaron, G.J., Ouédraogo, J.B., Zeilani, M., Brown, K.H. 2011. Acceptability of zinc-fortified, lipid-based nutrient supplements (LNS) prepared for young children in Burkina Faso. *Maternal and child nutrition*, 7:357-67.
- Human Sciences Research Council (HSRC). 2013. The South African National Health and Nutrition Examination Survey (SANHANES-1). Data analysis on infant feeding practices, and anthropometry in children under five years of age: South Africa 2012. Report for UNICEF, November 2013.
- Labadarios, D, Swart, R., Maunder, E.M.W., Kruger, H.S., Gericke, G.J., Kuzwayo, P.M.N., *et al.* 2008. The National Food Consumption Survey-Fortification Baseline (NFCS-FB-I): South Africa, 2005. *South African journal of clinical nutrition*, 21(3):245-300.
- Matias, S., Chaparro, C., Perez-Exposito, A., Peerson, J., Dewey, K. 2011. Acceptability of lipid-based nutrient supplement among Guatemalan infants and young children. Technical report. Washington, D.C.:FHI 360/FANTA-2 2011:1-33.
- Nor, B., Ahlberg, B.M., Doherty, T., Zembe, Y., Jackson, D. & Ekström, E.C. 2012. Mother's perceptions and experiences of infant feeding within a community-based peer counselling intervention in South Africa. *Maternal and child nutrition*, 8(4):448-458.
- Nyaradi, A., Li, J., Hickling, S., Foster, J. & Oddy, W.H. 2013. The role of nutrition in children's neurocognitive development, from pregnancy through childhood. *Frontiers in human neuroscience*, 7(97):1–16.
- Phuka, J., Ashorn, U., Ashorn, P., Zeilani, M., Cheung, Y.B., Dewey, K.G., Manary, M., Maleta, K. 2011. Acceptability of three novel lipid-based nutrient supplements among Malawian infants and their caregivers. *Maternal and child nutrition*, 7:368-77.
- Pongcharoen, T., Ramakrishnan, U., DiGirolamo, A.M., Winichagoon, P., Flores, R., Singkhornard, J., *et al.* 2012. Influence of prenatal and postnatal growth on intellectual functioning in school-aged children. *Archives of pediatrics & adolescent medicine*, 166(5):411–416.
- Prado, E.L. & Dewey, K.G. 2014. Nutrition and brain development in early life. *Nutrition reviews*, 72(4):267-284.
- Prendergast, A.J. & Humphrey, J.H. 2014. The stunting syndrome in developing countries. *Paediatrics and international child health*, 34(4):250–265.
- Qasem, W., Fenton, T. & Friel, J. 2015. Age of introduction of first complementary feeding for infants: a systematic review. *BMC Pediatrics*, 15(1):107.

Rothman, M., Berti, C., Smuts, C.M., Faber, M., Covic, N. 2015. Acceptability of novel small-quantity lipid-based nutrient supplements for complementary feeding in a peri-urban South African community. *Food and nutrition bulletin*, doi: 10.1177/0379572115616057.

Ruel, M.T. & Alderman, H. 2013. Nutrition-sensitive interventions and programmes: how can they help to accelerate progress in improving maternal and child nutrition? *The lancet*, 382(9891):536-551.

Saha, K.K., Frongillo, E.A., Alam, D.S., Arifeen, S.E., Persson, L.Å. & Rasmussen, K.M. 2008. Appropriate infant feeding practices result in better growth of infants and young children in rural Bangladesh. *The American journal of clinical nutrition*, 87(6):1852–1859.

South Africa. Department of Health. 2013. South African infant and young child feeding policy. Pretoria.

South Africa. Department of Health South Africa & UNICEF South Africa. 2007. A reflection of the South African maize meal and wheat flour fortification programme. Pretoria: South African Department of Health.

Sudfeld, C.R., McCoy, D.C., Fink, G., Muhihi, A., Bellinger, D.C., Masanja, H., *et al.* 2015. Malnutrition and Its Determinants Are Associated with Suboptimal Cognitive, Communication, and Motor Development in Tanzanian Children. *The Journal of nutrition*, doi: 10.3945/jn.3115.215996

UNICEF (United Nations Children's Fund). 2014. Statistical tables: economic and social statistics on the countries and areas of the world, with particular reference to the children's well-being. New York, USA.

Walker, R. W. & Goran, M. I. 2015. Laboratory determined sugar content and composition of commercial infant formulas, baby foods and common grocery items targeted to children. *Nutrients*, 7:5850-5867.

WHO (World Health Organization). 2003. Guiding principles for complementary feeding of the breastfed child. Washington: Pan American Health.

ANNEXURE 1

Additional policies, principles and indicators

Box 1. The Tshwane Declaration (DOH, 2011).

The Tshwane Declaration of Support for Breastfeeding in South Africa were implemented in 2011 and focus on:

- The support and strengthening of efforts to promote breastfeeding
- The adoption of the 2010 WHO guidelines on HIV and Infant feeding
- Finalisation of the National Regulations on the Code on Marketing of Breast milk substitutes
- Commitment by government and relevant bilaterals, partners and funders and excluding the formula industry as well as to promote, protect and support breastfeeding with included updated guidelines on HIV and Infant feeding. HIV mothers must also be considered and should breastfed for 12 months according to National guidelines.
- Promotion and support of human milk banks
- Implementation of Baby Friendly Health Initiative (BFHI) and Kangaroo Mother Care (KMC)
- Implementation of community based Interventions and support
- Continuation of research, monitoring and evaluation
- Not to provide formula feeds at public health facilities except for nutritional supplements including formula feeds prescribed by a health care professional for mothers, infants and children with approved medical conditions (DOH 2013).

Table 1. Guiding principles for complementary feeding of the breastfed child (Source: WHO 203, WHO, 2005).

Breastfed child	Non breastfed child
Introduce complementary foods at 6 months of age while continuing to breastfeed on-demand until 2 years of age or beyond.	
Start at 6 months of age with small amounts of food and increase the quantity as the child gets older, while maintaining frequent breastfeeding.	Daily dietary intake must meet the energy needs of the infant (approximately 600 kcal per day at age 6-8 months, 700 kcal at age 9-11 months and 900 kcal at age 12-23 months). A variety of nutrient-dense food should be served 4-5 times per day with additional nutrient dense snacks 1-2 times per day according to the infant's energy requirements.
Gradually increase food regularity and variety of nutrient-rich foods as the infant grows older, by adapting to the infant's requirements and abilities.	
Use fortified complementary foods or vitamin-mineral supplements for the infant, as needed.	Fortified food must contain iron (8-10 mg for 6-12 month old infants and 5-7 mg/d for 12-24 month old infants. Zinc, calcium and vitamin B12 should be supplemented to infants who consumed a small amount of animal-source foods.
<ul style="list-style-type: none"> • Practice responsive feeding, applying the principles of psychosocial care. 	
<ul style="list-style-type: none"> • Practice good hygiene and proper food handling. 	
<ul style="list-style-type: none"> • Increase fluid intake during illness, including more frequent breastfeeding, and encourage the child to eat soft, favourite foods. After illness, give more food more often than usual and encourage the child to eat more. 	

Box 2. Infant and Young Child Feeding Indicators (WHO, 2008).

	Indicator	Description
1	Breastfeeding initiation	proportion of children born in the last 24 months who were put to the breast within one hour of birth
2	Exclusive breastfeeding under 6 months	proportion of infants 0-5 months of age who are fed exclusively with breast milk
3	Continued breastfeeding at one year	proportion of children 12-15 months of age who are fed breast milk
4	Introduction of solid, semi-solid or soft foods	proportion of infants 6-8 months of age who receive solid, semi-solid or soft foods
5	Minimum dietary diversity	proportion of children 6-23 months who received foods from 4 or more food groups
6	Minimum meal frequency	proportion breastfed and non-breastfed children 6-23 months of age who receive solid, semi-solid, or soft foods (also included milk feeds for non-breastfed children) the minimum number of times of more
7	Minimum acceptable diet	proportion of children 6-23 months of age who receive a minimum acceptable diet (apart from breast milk)
8	Consumption of iron-rich or iron-fortified foods	proportion of children 6-23 months of age who receive an iron-rich food or iron-fortified food that is specially designed for infants and young children, or fortified at home
9	Child ever breastfed	proportion of children born in the last 24 months who were ever breastfed
10	Continued breastfeeding at 2 years	proportion of children 20-23 months of age who are fed breast milk
11	Age appropriate breastfeeding	proportion of children 0-23 months of age who are appropriately breastfed
12	Predominant breastfeeding under 6 months	proportion of infants 0-5 months of age who are predominantly breastfed
13	Duration of breastfeeding	
14	Bottle feeding	proportion of children 0-23 months of age who are fed with a bottle
15	Milk feeding frequency for non-breastfed children	median duration of breastfeeding among children less than 36 months of age

Box 3. Policy framework of Global Strategy for Infant and Young Child Feeding (WHO, 2003).

- Inappropriate feeding practices and their consequences are major obstacles to sustainable socioeconomic development and poverty reduction.
- Appropriate evidence-based feeding practices are essential for attaining and maintaining proper nutrition and health.
- Mothers and babies form an inseparable biological and social unit; the health and nutrition of one group cannot be separated from the health and nutrition of the other.
- Keeping improved infant and young child feeding high on the public health agenda is crucial to consolidating gains made during the past two decades.
- Twenty years after the adoption of the International Code of Marketing of Breast-milk Substitutes and 10 years into the World Declaration on Plan of action for Nutrition, the Innocenti Declaration and the Baby-friendly Hospital Initiative, it is time for governments, the international community and the other concerned parties to renew their commitment to promoting the health and nutrition of infants and young children and to work together for this purpose.
- An integrated comprehensive approach is needed for the implementation of the strategy.
- Existing health and inter-sectoral structures must be used and reinforced where necessary.
- Political commitment at the highest level must be achieved as well as the assembling of the indispensable human and financial resources
- Additional high-priority for success include definition of suitable goals and objectives, a realistic timeline for their achievement, and measurable process and output indicators that will permit an accurate monitoring and evaluation of action taken and a rapid response to identified needs.

Box 4. Baby-friendly Hospital Initiative (WHO and UNICEF, 2009).

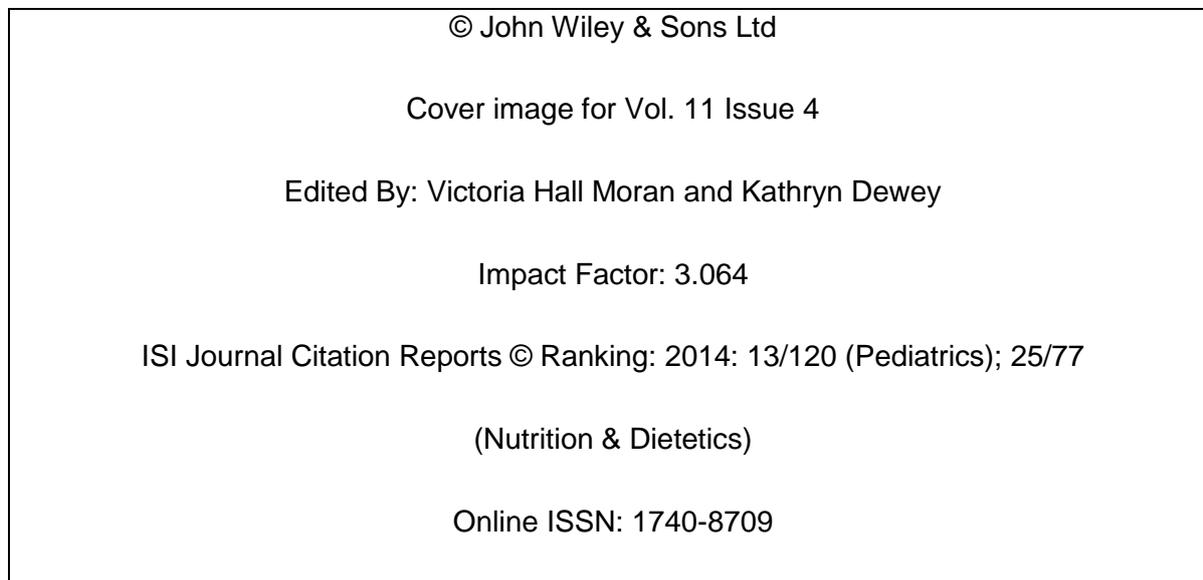
1. Have a written breastfeeding policy that is routinely communicated to all health care staff.
2. Train all health care staff in skills necessary to implement this policy.
3. Inform all pregnant women about the benefits and management of breastfeeding.
4. Help mothers initiate breastfeeding within one half-hour of birth.
5. Show mothers how to breastfeed and maintain lactation, even if they should be separated from their infants
6. Give newborns infants no food or drink other than breast milk, unless medically indicated.
7. Practice rooming in – that is, allow mothers and infants to remain together 24 hours a day.
8. Encourage breastfeeding on demand.
9. Give no artificial teats or pacifiers (also called dummies or soothers) to breastfeeding infants.
10. Foster the establishment of breastfeeding support groups and refer mothers to them on discharge from hospital or clinic.

Box 5. The four operations target of the The Innocenti Declaration (22 November 2005, Florence, Italy).

1. Appointing a national breastfeeding coordinator with appropriate authority, and establishing a multi-sectoral national breastfeeding committee composed of representatives from relevant government departments, nongovernmental organisations and health professional associations
2. Ensuring that every facility providing maternity services fully practices all "Ten steps to successful breastfeeding set out in the WHO/UNICEF statement on breastfeeding and maternity services
3. Giving effect to the principles and aim of the International code of Marketing of Breast-milk Substitutes and subsequent relevant Health Assembly resolutions in their entirety
4. Enacting imaginative legislation protecting the breastfeeding rights of working women and establishing means for it enforcement

ANNEXURE 2

Content and style guidelines for Maternal and Child Nutrition



The scope of Maternal & Child Nutrition includes pre-conceptual nutrition, antenatal and postnatal maternal nutrition, women's nutrition throughout their reproductive years, and fetal, neonatal, infant and child nutrition, up to and including adolescence. The Journal welcomes submission of (1) Original Research Papers, (2) Systematic Reviews that provide original information on maternal and/or child nutrition and (3) Letters to the Editor (Correspondence), usually commenting on a recent publication in the journal. The journal also publishes Special Issues, which bring together collections of papers on a particular theme, usually edited by a Guest Editor, and subject to customary reviewing process.

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University of Central Lancashire School of Health

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Baby Friendly Health Initiative (2012) 10 Steps to Successful Breastfeeding. Available at: <http://www.babyfriendly.org.au/about-bfhi/ten-steps-tosuccessful-breastfeeding/> (Accessed 23 November 2014).

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OnlineOpen is available to authors of primary research articles who wish to make their article available to non-subscribers on publication, or whose funding agency requires grantees to archive the final version of their article. With OnlineOpen, the author, the author's funding agency, or the author's institution pays a fee to ensure that the article is made available to non-subscribers upon publication via Wiley Online Library, as well as deposited in the funding agency's preferred archive. For the full list of terms and conditions, see <http://olabout.wiley.com/WileyCDA/Section/id-406241.html>. Prior to acceptance there is no requirement to inform an Editorial Office that you intend to publish your paper OnlineOpen if you do not wish to. All OnlineOpen articles are treated in the same way as any other article. They go through the journal's standard peer-review process and will be accepted or rejected based on their own merit.

8.6 Author material archive policy

Please note that, unless specifically requested, Wiley-Blackwell will dispose of all hardcopy or electronic material submitted 2 months after publication. If you require the return of any material submitted, please inform the Editorial Office or Production Editor as soon as possible if you have not yet done so.

9. Manuscript referrals to the open access journal Food Science & Nutrition

This journal works together with Wiley's open access journal, Food Science & Nutrition, to enable rapid publication of good quality research that is unable to be accepted for publication by our journal. Authors may be offered the option of having the paper, along with any related peer reviews, automatically transferred for consideration by the Editor of Food Science & Nutrition. Authors will not need to reformat or rewrite their manuscript at this stage, and

publication decisions will be made a short time after the transfer takes place. The Editor of Food Science & Nutrition will accept submissions that report well-conducted research which reaches the standard acceptable for publication. Food Science & Nutrition is a Wiley Open Access Journal and article publication fees apply. For more information, please go to www.foodscience-nutrition.com.details on online production tracking and for a wealth of resources including FAQs and tips on article preparation, submission and more.

ANNEXURE 3

Content and style guidelines for the Food and Nutrition Bulletin

The editors of the Food and Nutrition Bulletin (FNB) welcome contributions relating to nutrition in the developing world (please see Content guidelines below.) Submission of an article does not guarantee publication; acceptance depends on the judgment of the editors and reviewers as to its relevance and quality. Contributors should examine recent issues of the Bulletin for content and style, and should also review the FNB Editorial Policy prior to submitting an article.

The following is an outline of the content and style guidelines and formatting required for all FNB submissions. Please review carefully, and prepare your manuscript according to these guidelines. **Manuscripts that do not conform to these guidelines will be returned to the author for correction.**

Content guidelines:

The Food and Nutrition Bulletin accepts policy analyses, state-of-the-art summaries, and original scientific articles relating to multidisciplinary efforts to alleviate the problems of world hunger and malnutrition in the developing world. All potentially acceptable manuscripts are peerreviewed.

Articles published in the FNB must be broadly applicable; scientific articles of interest only to individuals in a single discipline or within a single country or region will not be accepted. **Other Content:** In addition to the types of articles outlined above, Letters to the Editor are encouraged and will be printed if judged to have an adequate basis and to be of sufficient general interest. Also of interest are "Short Communications," defined as papers that explore a topic with the customary scientific rigor but that do not need extensive analysis and discussion of the data or that have useful information that is not sufficient for a full article. These would be identified as such at the beginning of each article.

Formatting guidelines:

Language. All submissions must be in English.

Format. Manuscripts should be prepared on a computer in MS Word, double-spaced, with ample margins, and submitted electronically via email directly to the Managing Editor (fnb@inffoundation.org). **Hard copy submissions are not accepted.** Please review page 3 for formatting instructions of the cover page and text of your manuscript.

Abstract. An abstract of not more than 250 words should be included at the beginning of the manuscript, in the following format:

- *Background.* The context of the problem that was investigated, with relevant historical information.
- *Objective.* A one- or two-sentence description of the purpose of the study and what the authors expected to find.
- *Methods.* Outline of study design, subject selection, analytical methods, data analysis.
^{3/4}
- *Results.* What was found, based on the data. Give specific data and their statistical significance here, if possible.
- *Conclusions.* Provide a one- or two-sentence description of the conclusions, based on the results.

Emphasize new and important aspects of the study or observations. Do not include any information that is not given in the body of the article. Do not cite references or use abbreviations or acronyms in the abstract.

Key words. Authors should provide a minimum of four key words for the article.

Ethical approval of studies and informed consent. For investigations of human subjects, authors should state in the Methods section the manner in which informed consent was obtained from the study participants (i.e., oral or written), and describe how the study investigators protected the rights of participants as described in the Declaration of Helsinki.

Conflicts of interest and other disclosures. Authors must confirm upon submission that the manuscript is an original work, and has not been submitted for publication elsewhere. Additionally, authors must explicitly state any potential financial or other conflicts of interest in a cover letter accompanying submission of their manuscript. Finally, the corresponding author will need to verify that each author listed has seen and approved the form and content of the submitted article.

Tables and Figures. Tables and figures should be placed on separate pages at the end of the manuscript file. Footnotes should be keyed to the relevant data points by letters or symbols. **The original data files for figures that use bar graphs, scatterplots, or other graphic representations of data must be included as part of the initial submission.** Please double-check your data for accuracy and consistency with the text. Photographs.

Photographs should be submitted electronically in jpg., tiff. or bmp. format. If used, photographs will be published only in black and white.

Units of measure. All measurements should be expressed in metric units. If other units are used, their metric equivalent should be indicated.

Abbreviations. Please spell out all abbreviations used on the first reference.

Text of manuscript:

- Head 1 (eg., Abstract, Keywords, Intro, Methods, Results, Discussion, Acknowledgment, References) should be in 14 point bold.
- Head 2 should be in 12 point bold.
- Head 3 should be in 12 point bold italic.
- Please do not use numbers to designate order in headers or titles.

Footnotes: If possible, please try to keep footnotes concise and to a minimum. Remember that FNB pages are slightly smaller, and long footnotes will extend to subsequent pages.

References in the text should be numbered in consecutive order, and should be **enclosed in square brackets**. (eg, "...better than that of the population of Nepal [1].") Please do NOT use parentheses or superscript.

For more detailed information on references, please see References section below.

Cover page

Please format the cover page and text of your manuscript as follows: Title in 14 point bold

First and last name of all authors – please do NOT use superscript

Description of affiliations - please do NOT use superscript or titles (eg., Dr., Professor, job titles, etc.). Affiliations should include just the name of the organization, city and state and/or country. University departments, etc. should not be included.

Corresponding author information – include name, mailing address, and email.

Running head must also be given.

References

- ***We prefer that authors use the software EndNote® or the Endnote feature in MS Word when compiling references.** If further instruction is needed on how to use this MS Word tool, please contact the Managing Editor. If references are created manually, the author will be responsible for renumbering all references in the text and reference section if changes are required.

- References should be listed at the end of the article, and **must be provided in the format listed below (see examples).**

- Please number references consecutively in the order in which they are first mentioned in the text. Identify references in the text and tables and figure legends by arabic numerals **enclosed in square brackets**. (Please do NOT use parentheses.) References cited only in tables or figure legends should be numbered in accordance with the first mention of the relevant table or figure in the text.

- ***Be sure references are complete and current; the accuracy of references is the responsibility of the author.** Please double-check that reference numbers correspond to the correct numbers in the text.

- Unpublished papers, and papers submitted for publication but not yet accepted, should not be listed as references.

- If the material in the article has been previously presented or is planned to be published elsewhere in the future—in the same or modified form—a note should be included giving the details.

Please review these examples of reference formatting:

—*Standard journal article (list all authors – do not use “et al.”)*

1. Alvarez MI, Mikasic D, Ottenberger A, Salazar ME. Características de familias urbanas con lactante desnutrido: un análisis critico. Arch Latioam Nutr 1979;29:220-30.

—*Corporate author:*

2. Committee on Enzymes of the Scandinavian Society for Clinical Chemistry and Clinical Physiology. Recommended method for the determination of gammaglutamyltransferase in blood. Scand J Clin Lab Invest 1976;36:119-25.

Book or other monograph reference

—personal author(s):

3. Brozek J. Malnutrition and human behavior: experimental, clinical and community studies. New York: Van Nostrand Reinhold, 1985.

—corporate author:

4. American Medical Association, Department of Drugs. AMA drug evaluations, 3rd ed. Littleton, Mass, USA: Publishing Sciences Group, 1977. —editor, compiler, chairman as author: 5. Medioni J, Boesinger E, eds. Mécanismes éthologiques de l'évolution. Paris: Masson, 1977.

—chapter in book:

6. Barnett HG. Compatibility and compartmentalization in cultural change. In: Desai AR, ed. Essays on modernization of underdeveloped societies. Bombay: Thacker, 1971:20-35.

ANNEXURE 4

Published Article

Acceptability of Novel Small-Quantity Lipid-Based Nutrient Supplements for Complementary Feeding in a Peri-Urban South African Community

Marinel Rothman, MSc¹, Cristiana Berti, MD¹, Cornelius M Smuts, PhD¹, Mieke Faber, PhD², and Namukolo Covic, MD¹

Abstract

Background: Small-quantity lipid-based nutritional supplements (SQ-LNS) may potentially be used for home fortification in poor settings, where low nutrient-dense complementary foods are commonly used for infant feeding. However, they need to be acceptable to succeed.

Objective: This study assessed the acceptability of 2 novel, SQ-LNS (A and B) for supplementing complementary foods among infants aged 6 to 12 months in a peri-urban South African community.

Methods: Both supplements were soy-based pastes and contained micronutrients and essential fatty acids. In addition, supplement B contained docosahexaenoic acid, arachidonic acid, phytase and L-lysine. Mother–infant pairs were enrolled in a 2-part trial. Part 1 ($n = 16$) was a test-feeding trial with a crossover randomized design, and a 5-point hedonic scale was used for sensory evaluation (*disagree* = 1, *agree* = 5). Part 2 ($n = 38$) was a 2-week, home-use trial followed by focus group discussions.

Results: In part 1, more than 70% of mothers reported a score ≥ 4 on sensory attributes for both SQ-LNSs indicating that both supplements were well perceived. In part 2, the mean reported consumption over the 2-week period was $65.3\% \pm 34.2\%$ and $62.0\% \pm 31.3\%$ of the 20 g daily portion for supplements A and B, respectively. Focus group discussions confirmed a positive attitude toward the supplements in the study population.

Conclusion: This study showed acceptance of both SQ-LNSs in terms of sensory characteristics as well as in terms of practicality for home use.

Keywords

lipid-based supplements, infants, acceptability, complementary feeding

Introduction

Worldwide, undernutrition consisting of stunting, wasting, vitamin A and zinc deficiency as well as suboptimum breast-feeding causes nearly 3.1 (45%) million deaths of children younger than 5 years.¹ One of the 6 global targets for the improvement in infant and young child nutrition by 2025 is a 40% reduction in children younger than 5 years who are stunted. In 2012, 36% of all

¹ North-West University, Potchefstroom, South Africa

² Non-Communicable Disease Research Unit of the South African Medical Research Council, Cape Town, South Africa

Corresponding Author:

Marinel Rothman, Centre of Excellence for Nutrition, North-West University, Potchefstroom Campus, Private Bag X6001, Potchefstroom 2520, South Africa.

Email: marinel.rothman@nwu.ac.za

children with stunting, 29% of all underweight children, and an estimated 28% of all children with severe wasting were living in Africa.² South Africa is among the 34 countries with the highest burden of stunting on a global level.³ The highest percentage of stunting falls in the age category 0 to 3 years at 26.9% and 25.9% for South African boys and girls, respectively.⁴

South Africa has the lowest level (8%) of exclusive breast-feeding in sub-Saharan Africa.⁵ It has been reported that in some areas 35% to 50% of mothers stop all breast-feeding before 3 months postpartum, that complementary foods are introduced as early as 6 weeks of age,^{6,7} and that the nutrient density of the complementary diet is poor.^{8,9} These suboptimal feeding practices are of concern, particularly against the background that the prevalence of stunting in the 1 to 3 years age-group increased by 3% from 2005 to 2012.⁴ Strategies to improve dietary quality of complementary foods are therefore urgently needed.

Small-quantity lipid-based nutrient supplements (SQ-LNSs), which contribute to energy, protein, and essential fatty acids (EFA) intake, are easy to use and require minimum change in dietary behaviors.^{10,11} Presently, many organizations and companies have shown interest in the development of SQ-LNS and its potential use in a variety of cultural and geographical settings.¹⁰ Recently, a study conducted in the Honduras indicated that SQ-LNS improved micronutrient status in non-malnourished children aged 6 to 18 months.¹¹ Iannotti et al¹² showed that daily consumption of SQ-LNS resulted in a significant increase in linear growth in healthy infants aged 6 to 11 months in Haiti. To ensure large-scale use of SQ-LNS, these products should be acceptable within a given sociocultural context. Although acceptability trials on SQ-LNS were conducted among children younger than 5 years in some countries such as Malawi, Burkina Faso, Ghana and Guatemala,¹³⁻¹⁶ these did not contain added docosahexaenoic acid (DHA), arachidonic acid (ARA), and phytase.

Two new SQ-LNS were developed for potential use in the South African setting. Both SQ-LNS-A and SQ-LNS-B are made from soy, which is less expensive than peanuts and contain essential fatty acids and micronutrients needed for infant development.^{17,18} In addition, SQ-LNS B contains ARA

and DHA, which may provide additional benefits in terms of vision, growth, and psychomotor development¹⁹⁻²¹; phytase to improve iron and zinc bioavailability^{17,22}; and L-lysine, which is an essential amino acid that is lacking in maize. In South Africa, as in many African countries, maize meal porridge is often used for complementary feeding in infants.^{8,23} However, the acceptance of home use of SQ-LNS for infant feeding has not yet been established in South Africa.

The aim of this study was to determine the acceptability of 2 newly developed SQ-LNS pastes in a peri-urban South African community. The specific objectives of the study presented in this article were (1) to determine the acceptability in terms of sensory characteristics to infants and their primary caregivers, as rated by the primary caregiver on a 5-point hedonic scale, and (2) to obtain information on how the caregivers experienced using the products by conducting a 2-week home-use trial followed by focus group discussions (FGDs).

Materials and Methods

Overview and Design

The 2 SQ-LNSs were produced by 2 European companies, namely DSM and UNILEVER, with a presence and capacity to produce in South Africa. The 2 SQ-LNSs were delivered to the nutrition laboratory at the North-West University, Potchefstroom. Both SQ-LNS were packed in single-dose 20 g plastic tubs with lids. The nutrient composition of the SQ-LNS A and B is presented in Table 1. The study was single blinded, and for the purpose of the study, the 2 products, utensils as well as documentation used were color coded. The products were only referred to in color codes at all times to the field staff facilitators and participants.

Study Setting

The trial was conducted in February to April 2013 in a low socioeconomic peri-urban community in the North-West Province in South Africa. This trial preceded a randomized controlled trial that assessed the effect of SQ-LNS on infant's growth from 6 to 12 months, therefore this trial recruited infants in the same age category.

Table 1. Nutrient Content of the 2 SQ-LNS Pastes.

	SQ-LNS A	SQ-LNS B
Amount, g (1 portion)	20	20
Energy, kcal	114	113
Energy density, kcal/g	5.7	5.7
Protein, g	3.0	3.7
% calories from protein	10	13
Fat, g	8.0	8.8
% calories from fat	63	70
Linoleic acid, g	1.5	1.8
α -linolenic acid, mg	265	348
n-6/n-3 ratio	5.7	5.0
Docosahexaenoic acid, mg	–	75
Arachidonic acid, mg	–	75
Vitamin A, μ g RE	200	200
Vitamin D, μ g	2.5	2.5
Vitamin E, mg	2.5	2.5
Vitamin K, μ g	7.5	7.5
Thiamin, mg	0.25	0.25
Riboflavin, mg	0.25	0.25
Niacin, mg	3.0	3.0
Pantothenate, mg	1.0	1.0
Vitamin B6, mg	0.25	0.25
Biotin, mg	4.0	4.0
Folic acid, μ g	80	80
Vitamin B12, μ g	0.45	0.45
Vitamin C, mg	23.3	23.3
Calcium, mg	250	396
Iodine, μ g	45	45
Iron, mg	5.8	5.8
Zinc, mg	6.2	6.2
Copper, mg	0.28	0.28
Selenium, μ g	8.5	8.5
Magnesium, mg	–	30
Manganese, mg	–	0.6
Phosphorus, mg	–	230
Potassium, mg	–	257
L-Lysine, mg	–	160
Phytase FTU	–	200

Abbreviations: SQ-LNS, small-quantity lipid-based nutrient supplements; FTU, unit of phytase activity.

Ninety-four percent of the caregivers who participated in the study were the mothers of the infants. For this reason, the term “mother” is used throughout the rest of the article.

Recruitment Process

The mother–infant pairs were recruited at 5 clinics in the community. These clinics allocate specific days for mother–infant pairs, and the

recruitment was done on these specific days. Trained fieldworkers explained the study to the mothers using a structured information sheet explaining the meaning and importance of the trial as well as some background of the SQ-LNS. The mothers were also given an information sheet in their local language. If the mother expressed willingness to participate, an eligibility questionnaire was administered to determine whether the infant could be included in the study. To be eligible to participate in the study, the infant had to be 6 to 12 months of age; have consumed some breast milk previously; be apparently healthy with no fever and not suffering from an acute illnesses such as acute respiratory tract infection and diarrhoea; not be severe malnourished (mid-upper arm circumference [MUAC] \leq 115 mm); and with no known allergy to peanuts, soy, milk/lactose, and fish. Also, the mother–infant pairs should plan to remain in the study area for at least the following 4 weeks to ensure no adverse reactions occurred. Before enrollment, a consent form was signed by the mother. Different infants were recruited for parts 1 and 2 of the acceptability trial to avoid a carryover effect. A different information- and consent form were used for part 2 which included information regarding the FGDs. Eligibility criteria was the same for part 2.

Mid-upper arm circumference was taken in order to determine whether the infant was severely malnourished using a standard MUAC tape following standard procedures.²⁴ Infants excluded from participation for having failed the MUAC screenin, were to be referred to the nearest clinic.

Research Approach Used

The methodology used in this study was a combination of methodological elements from the acceptability trials that were conducted in Malawi, Ghana, and Guatemala,^{13,15,16} which were adapted for our specific setting. This was a single-blind study, as only the researchers were able to distinguish between the SQ-LNS-A and -B. This study was carried out in 2 parts. Part 1 was a sensory evaluation in which the mothers were asked to taste each SQ-LNS product mixed with maize porridge and then score the SQ-LNS

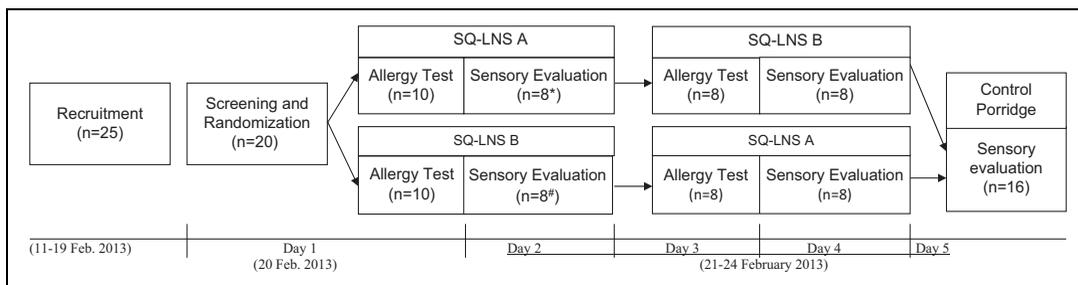


Figure 1. Representation of the research approach used for Part 1. *2 dropouts from day 1: 1 infant developed a rash (not related to product) and 1 mother withdrew from the study for personal reasons. #2 dropouts from day 1: infants refused to eat the product.

mixture on different sensory attributes. The mothers were also asked to feed the SQ-LNS mixture to their infants and then rate their perception of the infant's liking of the products. The maize meal porridge used was prepared by a fieldworker from the community. Part 2 was a 2-week home-use trial to obtain information on how mothers experienced the use of the SQ-LNS at household level. At the end of part 2, FGDs were conducted to get insight on how the mothers experienced using the SQ-LNS during the home use trial. The number of mother–infant pairs enrolled in parts 1 and 2 of the study were based on similar acceptability trials conducted in Malawi,¹³ Burkina Faso,¹⁴ and Guatemala.¹⁶ For FGDs, at least 8 participants per group are needed, therefore the 40 mother–infant pairs enrolled in part 2 were deemed sufficient to be able to have at least 3 focus groups for data saturation.²⁵

Part 1: sensory evaluation. The sensory evaluation was done within a few days after recruitment using a crossover randomized design in February 2013. The design used was adapted from the method of Matias et al.¹⁶ The envelope method was used for randomization²⁶ which was done by researchers (MR and CB.). Figure 1 is a representation of the research approach used for part 1.

Half of the mother–infant pairs started with SQ-LNS-A, whereas the other half started with SQ-LNS-B. The mothers were invited to come to the field station at 08:00 AM on each sensory evaluation day to ensure that the infants were fed within the time frame in which they would normally consume their first complementary meal of the day.

On day 1, recruited infants were first screened for eligibility. All infants were found eligible and were given 5 g of either SQ-LNS-A or SQ-LNS-B and were observed by a study nurse for at least 20 minutes to ensure that no adverse reaction occurred.

On day 2, the sensory evaluation was done only if no adverse reaction was observed by the study nurse. The study nurse measured the infant's temperature using an electronic ear thermometer and children with aural body temperature $>38^{\circ}\text{C}$ were excluded. Breast-feeding mothers were first asked to breast-feed their child. No other liquids or food were allowed for the next 60 minutes during which a fieldworker interviewed the mother to complete a socioeconomic questionnaire. After the 60 minutes, the sensory evaluation was started. Under the guidance of a fieldworker, the mother mixed 20 g SQ-LNS with 20 g maize porridge and then tasted a spoonful of the SQ-LNS mixture. The rest of the SQ-LNS mixture, together with the standard bowl and spoon used, was weighed before starting to feed the infant. A stopwatch was set at 15 min (maximum time given to consume the paste and porridge mixture) and the countdown started when the mother started feeding her infant the given portion of the SQ-LNS mixture. The mother could stop if the infant refused to eat further and, if so, this was noted by the fieldworker. The mother was kindly asked to stop feeding her infant when 15 minutes was over. The remaining SQ-LNS mixture, together with the bowl and spoon, was weighed again, and the amount of SQ-LNS mixture consumed by the infant was calculated. If there were any leftovers after 15 minutes, the mother could then continue feeding the infant after

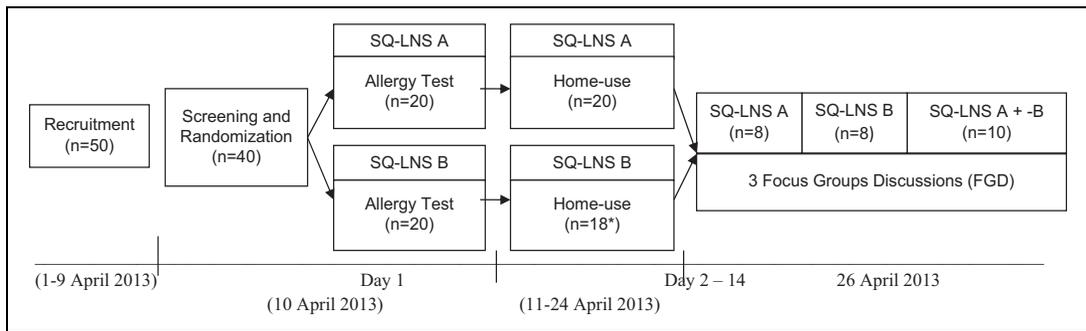


Figure 2. Representation of the research approach used for Part 2. *2 mothers in Small-quantity lipid-based nutrient supplement (SQ-LNS) B group withdrew for personal reasons.

the weighing had been done, if the infant still wanted to eat. On days 3 and 4, the second SQ-LNS was given, following the same process. On day 5, all infants were provided with 40 g of the control porridge (plain maize porridge) without any added SQ-LNS, following the same sensory evaluation procedure described earlier.

To assess the general liking and acceptance of the SQ-LNS mixes and control porridge, an adapted 5-point hedonic scale was used on which the points were indicated by a drawing of a facial expression depicting 1 = *disagree* (dislike extremely), 2 = *tend to disagree* (dislike), 3 = undecided, 4 = *tend to agree* (like) and 5 = *agree* (like extremely).¹⁶ The mother used the 5-point hedonic scale to rate the taste, smell, appearance, color, texture, mouth feel, and her overall liking as well as her perception of the infant's liking of the SQ-LNS mixtures and control porridge.

Part 2: Two-week home-use trial. The home-use trial commenced in April 2013. To avoid a carryover effect from the sensory evaluation, a different group of 40 mother–infant pairs were recruited from the same clinics for the home-use trial. The same informed consent process as for the sensory evaluation was followed. The mother–infant pairs were randomly assigned to 1 of the 2 SQ-LNSs for home use by researchers (MR and CB) using the envelope method.²⁶ Figure 2 is a representation of the research approach used for Part 2.

On the first day, recruited infants were first screened for eligibility at the central study site. All infants were found to be eligible. Eligible infants were given 5 g of the SQ-LNS. The home use was

continued only if the study nurse did not observe any negative reaction within 20 minutes. The mother was interviewed by a fieldworker to complete a socioeconomic questionnaire and was given a 13-day supply (14 minus 1, as 1 tub was used for day 1 on site) of 20-g tubs of either SQ-LNS-A or -B according to the randomization. The fieldworker demonstrated to the mother how to mix the SQ-LNS with whatever complementary food the mother would be giving for the next 13 days.

Instructions for use were as follows: For the infant's first meal of the day, the mother was asked to dishout 2 to 3 tablespoons of the complementary food she was going to give to her infant into a standard serving bowl provided and mix into it one complete 20 g tub of SQ-LNS. The mother was required to keep the empty SQ-LNS tubs in a plastic ziplock bag provided for the adherence monitoring. She was trained to record on a pictorial “Daily adherence form” the estimated amount the infant had consumed on the day, and the mother was also asked to record the type of complementary food used to mix with the SQ-LNS.

A fieldworker visited each mother 3 times (days 2, 7, and 14) to make sure that the mother understood the use of the SQ-LNS as well as the completion of the daily adherence form. At each visit, the fieldworker counted and recorded the used and remaining SQ-LNS tubs and also completed a weekly adherence questionnaire which also included information on any illnesses that may have occurred and the types of food the mothers used with the SQ-LNS.

At the end of the 2-week period, a fieldworker conducted an exit interview with the mother to get information on the mother's perception of the

infant's acceptance of SQ-LNS, the ease of using the SQ-LNS, the possible intrahousehold distribution of the SQ-LNS as well as the complementary foods used with the SQ-LNS.

Two days following the 2-weeks home-use trial, FGDs were conducted at the central field station to obtain additional information on the mothers' perceptions on the ease of using the 2 SQ-LNSs. The FGDs were facilitated by trained facilitators using a structured field guide adapted from Rabiee.²⁵ The kind of questions that were asked included: "What was your overall feeling about the product?"; "What was your experience of using the product?" and "Would you recommend the product for the feeding of the baby to other mothers/caregivers?". The participants were allocated into 3 different discussion groups: group A: mothers who used SQ-LNS A; group B: mothers who used SQ-LNS B; and group C: mixed group. The facilitator was aware of the color codes (for SQ-LNS A or B) but was not aware what the differences were. The same was the case for the participants. Two research assistants took notes, and the discussions were audio-taped. Before starting the focus group, mothers were asked to rate the general acceptance of the SQ-LNS-mixture on a scale from 1 to 10. The facilitator then elicited discussion on the practicality and experience of using the SQ-LNS for feeding the infant as well as general characteristics of the SQ-LNS. The discussions were conducted in the local language, Setswana, and lasted 30 to 45 minutes depending on information saturation, meaning that no new ideas emerged on the specific topics during a specific FGD.²⁵

Ethics

Ethical approval was obtained from the Ethics Committee of North-West University (NWU-00011-11-A1) and the Ethics Committee of the South African Medical Research Council (EC011-03/2012). Approval and permission to do the study was also given by both the Provincial and the District Department of Health, as well as at community health care facility level where recruitment of mother infant pairs was done. The study has been registered with the North West Provincial Department of Health Directorate for

Planning, Research, Monitoring and Evaluation. The actual transport costs were reimbursed for each mother participating in the trial.

Data Analysis

Statistical analyses were done using IBM SPSS Statistics 22 package. Socioeconomic characteristics of participants were analyzed using descriptive statistics. For part 1, the median and interquartile range are reported for consumption time and percentages for the hedonic scales. For part 2, the mean (SD) proportion of the SQ-LNS consumed was calculated. The information on sensory acceptance from the hedonic scales is reported as frequencies.

In part 2, for the calculation of percentage intake of both SQ-LNS, the standard amount of 5 g given on the first day of phase 2 was excluded. Thus, percentage intake over the 2 weeks was calculated only from Day 2 as the first determined day.

A reflective notebook was kept by a research assistant to note all verbal communication expressed by the focus group participants. A quality check of the FGD transcripts was done by a research assistant fluent in both English and Setswana and who was present during the FGDs to ensure that the transcripts were translated without the original meaning being lost in translation. The researcher also compared the transcribed work to the notes taken during the discussions. Direct quotes from participants were included in the description of the findings, in order to give more meaning to the context.²⁵ The audio tapes were transcribed verbatim and translated to English by Globosonic DFH, a professional company specialized in transcription services before content analysis. Transcripts were analyzed according to guidelines given by Rabiee²⁵; a thematic framework was compiled by identifying themes (key words); tabulating was done by managing, cleaning, and simplifying the data; and interpretation was done by identifying main expressions.

Results

Participant Characteristics

The characteristics of the participants are shown in Table 2. The mean age of the infants was 9.0 ±

2.0 months for part 1 and 9.0 ± 1.8 months for part 2. The mothers' mean age was 25.0 ± 5.2 years for part 1 and 26.0 ± 8.1 years for part 2. Fifty percent of the mothers (for both Parts) reported attending school for ≤ 10 years and $>60\%$ were single. Electricity was available to $>90\%$ of households. All households had access to tap water, either from their own tap outside the house (57%) or from a tap inside their house (43%).

Part 1: Sensory Evaluation: Twenty-five mother–infant pairs were recruited, but only 20 mothers came for screening. All of them were found eligible and enrolled in the study. There were 4 dropouts: 1 infant developed a rash (not related to the product), 2 infants refused to eat the SQ-LNS mixture, and 1 was withdrawn by the mother for personal reasons. Thus, 16 infants completed the sensory evaluation.

Part 2: Home-use trial: Fifty infant–mother pairs were recruited of whom 40 showed up for the start of the 2-week home-use trial. Two mothers in the SQ-LNS-B group withdrew their infants from the trial for personal reasons. Thirty-eight infants completed the home-use trial. Twenty-six mothers participated in 3 FGDs.

Part 1: Sensory Evaluation. All the infants ate the entire given portion of SQ-LNS-mixture (20 g SQ-LNS plus 20 g maize porridge) within the 15 minutes provided for (Figure 3). The median (interquartile range) times recorded for consumption for SQ-LNS-A, -B, and the control porridge were 3.2 (1.8–5.3), 4.3 (2.7–6.6), and 2.4 (1.8–2.8), respectively. The frequencies of different time intervals for all 3 groups are shown in Figure 3.

Figure 4 shows the mother's and her child's liking of the SQ-LNS mixtures and the control porridge. Most ($>70\%$) of the mothers reported liking the sensory attributes, taste, smell, appearance, color, texture, and mouth-feel. Based on the results from the hedonic scale, categories 4 and 5 of "tend to agree" and "agree," respectively, were combined because most mothers ($>60\%$) reported a score of 5 (agree) with a small percentage ($<20\%$) reporting 4 (tend to agree) for all sensory

Table 2. Characteristics of the Participants for Part 1 and Part 2 of the Study.

Participant Characteristics	Part 1	Part 2
Infant–mother pairs, n	16	38
Infants' mean (\pm SD) age, months	9 ± 2.0	9 ± 1.8
Mothers		
Mean (\pm SD) age, years	25 ± 5.2	26 ± 8.1
Unmarried, n (%)	14 (88)	26 (68)
School attendance, n (%)	16 (100)	34 (89)
10 years and less	8 (50)	17 (50)
11 and 12 years	8 (50)	16 (47)
Source of drinking water, n (%)		
Own tap outside house	9 (56)	22 (58)
Own tap inside house	7 (44)	16 (42)
Only cold running water available, n (%)	9 (56)	36 (95)
Electricity availability inside household, n (%)	15 (94)	36 (95)

Abbreviation: SD, standard deviation.

attributes when considered both mother and infant hedonic scale. Scores 1 (*disagree*), 2 (*tend to disagree*), and 3 (*undecided*) were reported by $<10\%$ when considering all sensory attributes for both scales. The overall liking for both pastes was 77.8% and 83.3% for SQ-LNS-A and SQ-LNS-B mixture, respectively. Most (83.3%) of the mothers reported not experiencing any problems when feeding both mixtures to their infants.

Table 3 shows the mother's score of her practical experience of using the porridge SQ-LNS mixtures. Most of the mothers reported that both SQ-LNS were easy to mix with porridge (88.9% and 83.3% for SQ-LNS A and SQ-LNS B mixture, respectively). Mothers also reported that they were positive to give both porridge SQ-LNS mixtures to their infants (83.3% and 72.2% for SQ-LNS A and B mixture, respectively). Without providing any specific possible pricing information, most (83.3%) mothers reported that they were willing to buy both SQ-LNS if available on the market.

Part 2: Home-use trial. The mothers reported mixing the given SQ-LNS with a variety of foods such as commercial infant cereal (18.4%), bottled pureed infant food (15.8%), breakfast cereals (13.2%), and some food items such as soft maize porridge, mabele porridge, and mashed potatoes

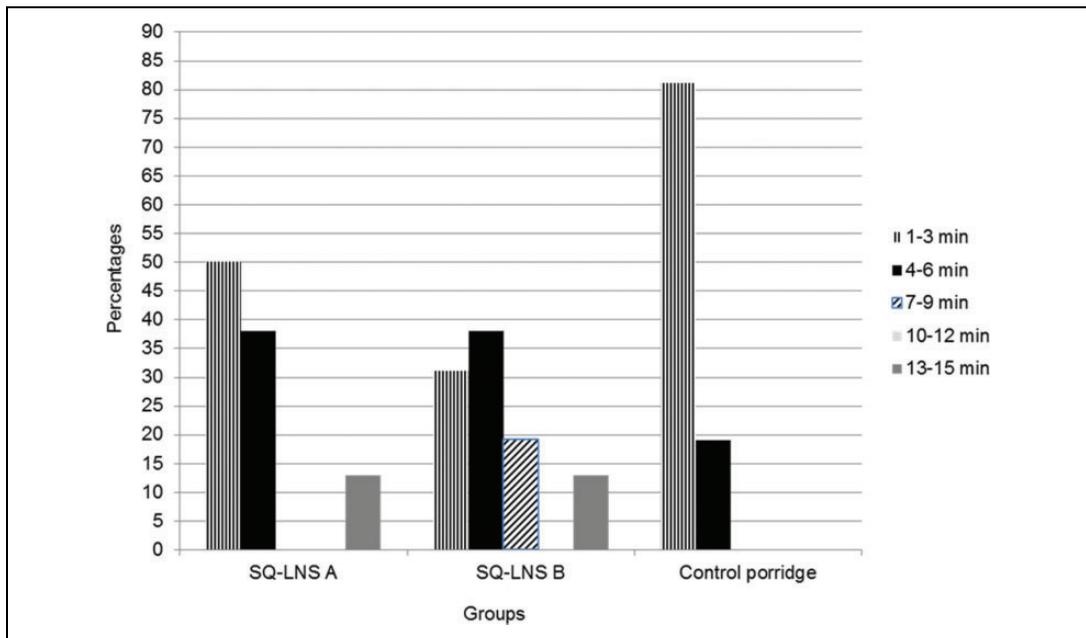


Figure 3. Time used to consume Small-quantity lipid-based nutrient supplements (SQ-LNS) A and B and control porridge, respectively.

were reported by less than 10% of the mothers, respectively. Over the 2-week period, the mean percentage of reported consumption was 65.3 ± 34.2 and 62.0 ± 31.3 of the recommended amount for SQ-LNS-A and -B, respectively (Table 4). The mother's own acceptance and perception of her infant's acceptance is reported in Table 4. At the end of the home-use trial, more than 80% of the mothers reported that both SQ-LNS were acceptable to them and their infants.

Three FGDs were conducted including 8 participants each for the SQ-LNS-A, and -B, and 10 participants for the mixed group. The FGDs reflected that the mothers were happy with their infants liking and eating of the SQ-LNS mixture as reflected in the comment "Baby enjoys it." Some mothers mentioned that their infants preferred the SQ-LNS mixture over the complementary foods they normally fed their infants ("Baby does not want to eat something else"). Some mothers also reported perceiving their infants showing more positive active behavior after eating the SQ-LNS mixture ("baby has more energy"). Some mothers felt that the SQ-LNS increased their infants' appetite with quotes like: "I like this product because it

gave my baby an appetite, she never ate a lot" and "After giving her this paste she eats more than she used to and I like that." The overall experience for both SQ-LNS was favorable in terms of ease of mixing and convenience of adding to the different types of complementary foods used. Some mothers spontaneously reported that they were willing to buy the SQ-LNS (A/B) and would encourage other mothers to give the pastes to their infants. No pricing information formed part of the discussion guide.

Discussion

This study indicated that, based on sensory characteristics, both soy-based SQ-LNS, when mixed with maize meal porridge, were acceptable in terms of taste, appearance, color, texture, mouth-feel, and overall liking. The aim of the study was not to determine which SQ-LNS was liked most but rather to assess whether both SQ-LNS were acceptable. The control porridge was included to ensure that the porridge that was used for mixing the SQ-LNS was acceptable and did not negatively affect acceptance of the SQ-LNS mixture. Both SQ-LNS mixed in maize porridge were perceived to have a better taste,

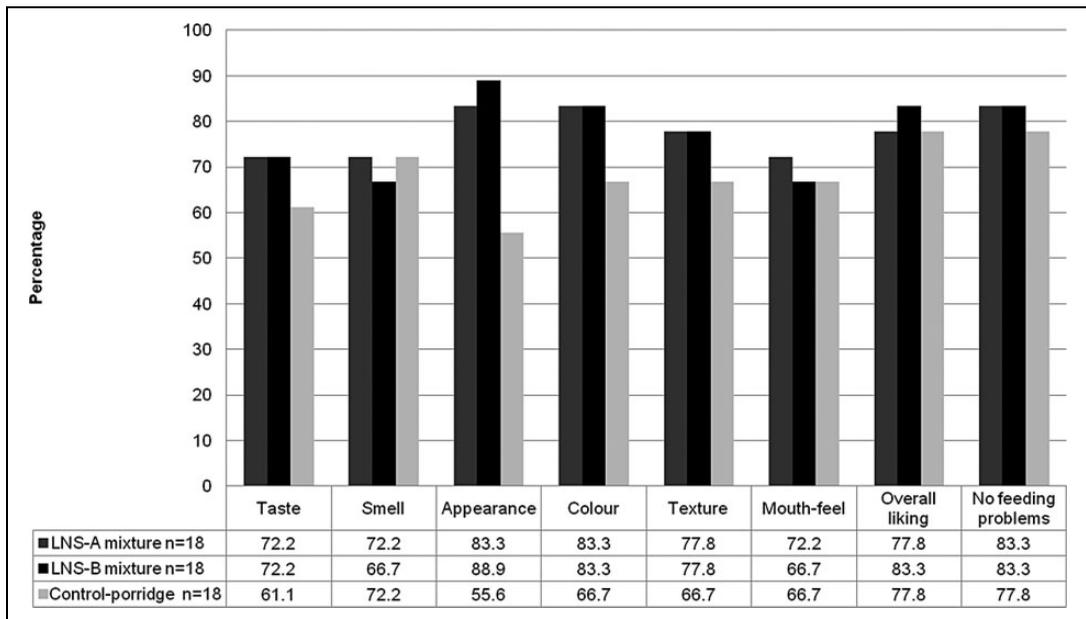


Figure 4. Percentage of mothers who reported “agree” on both her own liking and her perception of the infant’s liking of the 2 porridge Small-quantity lipid-based nutrient supplements (SQ-LNS) mixtures and the control porridge in part I of the study. *The term “agree” refers to the combination of “agree” and “tend to agree.”

Table 3. Hedonic Scores Reported by the Mother on her Practical Experience of Using the 2 Porridge SQ-LNS Mixtures in Part I of the Study.^a

Practical attribute	SQ-LNS-A mixture (n = 18)	SQ-LNS-B mixture (n = 18)	Control porridge (n = 18)
Easy to mix, n (%)			NA
Agree ^b	16 (88.9)	15 (83.3)	
Disagree ^c	0	1 (5.6)	
Liked giving, n (%)			NA
Agree ^b	15 (83.3)	13 (72.2)	
Disagree ^c	1 (5.6)	3 (16.7)	
Willingness to buy if available, n (%)			NA
Agree ^b	15 (83.3)	15 (83.3)	
Disagree ^c	1 (5.6)	1 (5.6)	

Abbreviations: NA, not applicable; SQ-LNS, small-quantity lipid-based nutrient supplements.

^aP < .05.

^bThe term “agree” refers to the combination of “agree” and “tend to agree.”

^cThe term “disagree” is the combination of “disagree” and “tend to disagree.”

texture, color, and appearance than the control maize porridge by the mothers (Fig. 4). When considering the sensory acceptance, Mennella et al²⁷ assumed that variance in responses to tastes may be affected by the infant’s feeding history in terms of breast milk and/or formula milk consumption. According to Adu-Afarwuah et al¹⁵ either the

proportion of offered food consumed or the duration taken to consume the amount offered without force-feeding can be used as a proxy indicator for acceptability. Soy-based products have not previously been reported for complementary feeding in the South African setting,⁹ and the infants would not therefore have been expected to be familiar with the

Table 4. Frequencies of the Mother's Own Acceptance and Perception of Infant Acceptance of the SQ-LNS Pastes after 2 Weeks of Home-Use (Part 2 of the Study) Expressed as Percentage.

		SQ-LNS A (n = 20)	SQ-LNS B (n = 18)
Percentage of SQ-LNS consumed, Mean \pm SD		65.3 \pm 34.2	62.0 \pm 31.3
Percentage (95%CI) SQ-LNS acceptable to mother	Yes	90 (75-100)	89 (72-100)
	No	5 (0-15)	6 (0-17)
	Not sure	0	6 (0-17)
	No answer	5 (0-15)	0
Percentage (95%CI) SQ-LNS acceptable to infant as perceived by the mother	Yes	80 (60-95)	89 (72-100)
	No	15 (0-35)	0
	Not sure	0	11 (0-28)
	No answer	5 (0-5)	0

Abbreviations: CI, confidence interval; SQ-LNS, small-quantity lipid-based nutrient supplements; SD, standard deviation.

sensory characteristics of the 2 given SQ-LNS mixtures. Similar products tested in other countries did not contain added DHA and ARA.¹³⁻¹⁶ Therefore, based on the short time taken to consume both SQ-LNS mixtures, and despite lack of familiarity with such SQ-LNS mixtures, the products tested can be considered to have been acceptable to the infants.

The home-use trial supported the findings from the sensory evaluation in terms of the perceptions toward the novel SQ-LNS. More than 80% of the mothers and their infants (based on mothers' perceptions) reported both SQ-LNSs to be acceptable at home-use level. As was the case in the study done by Adu-Afarwuah et al,¹⁵ this study presented a challenge in terms of determining the infants' perceived liking of the SQ-LNS which relied entirely on the mother's perception. However, the mother's level of acceptance and perceptions are essential, as she is the person primarily responsible for deciding what the child eats.²⁸

On average, 62.0% to 65.3% of the recommended amount of both SQ-LNS was consumed over the 2-week home-use period. This lower than recommended amount consumed may have an influence on the intake of the beneficial constituents such as DHA, ARA, phytase, and L-lysine. The randomized controlled trial that followed after this trial will indicate the impact of the SQ-LNS on the growth and development of infants in this age category in relation to the actual intake of the SQ-LNS over a period of 6 months. In the home-use trial, a variety as foods, for example, pureed jarred infant

foods, commercial infant cereals, soft maize porridge, and vegetables, were used to mix the SQ-LNS. Most mothers (>80%) reported ease of mixing with the SQ-LNS, despite the variety of foods used, and this may have contributed toward the mothers reported acceptance. In practice, this information is of interest, as it implies that the sensory characteristics of both SQ-LNS A and B are likely applicable to a variety of food items used in the home.

During the FGDs following the home-use period, some mothers referred to being pleased with perceived improvements in their infant's appetite and activity level after consuming the SQ-LNS-mixtures. Pelto et al²⁹ conducted a focused ethnographic study in urban and rural communities in South Africa and argued that the strongest association South African mothers had with a healthy child was an active child with a healthy appetite and steady weight gain. The mothers' perceptions in this study that their children's appetite and activity level improved are also in line with mother's comments reported in acceptability trials done in Ghana, Malawi, Guatemala, and Burkina Faso.¹³⁻¹⁶ Although pricing did not form part of the FGD guide, mothers' expressed willingness to buy the products. This further confirmed a positive attitude of the mothers toward the 2 SQ-LNS.

Although the SQ-LNS were shown to be acceptable in our study, for large-scale implementation, good communication and promotion (i.e., development of educational messages accompanying the SQ-LNS distribution) would be needed to

ensure that sufficient amounts are eaten by the infant so that the infant can get the full benefit of the SQ-LNS.²⁹ It is well recognized that communication and nutrition education form an integral part of any sustainable strategy aimed at improving the quality of complementary feeding.³

A strength of the study is that all participating infants were tested for any allergic reactions which might occur from these novel SQ-LNSs. A study nurse was appointed for the duration of the study, specifically for this purpose. The SQ-LNSs tested are relevant to all areas where undernutrition is prevalent, and the methodology used may provide a template that could be used in a different study population. The fact that, in part 1, the control porridge was always last may be seen as a limitation, as this may have resulted in bias in the reporting of the sensory evaluation. It should however be noted that the aim of the sensory evaluation of the control porridge was to verify that the porridge used for mixing the SQ-LNS was also accepted and did not negatively influence the sensory evaluation of the SQ-LNS mixture.

In conclusion, we found that the sensory characteristics of these 2 novel SQ-LNSs, when mixed into maize porridge and or other complementary foods, were acceptable to the mother–infant pairs and that the mothers experienced the use of these products positively. The results suggest that these newly developed SQ-LNSs may have potential for successful use in food-based interventions aimed at improving the nutritional quality of commonly used complementary foods in resource poor settings.

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Authors' Note

Marinel Rothman: study design; data collection, analysis, and interpretation; and wrote the first draft of the paper. Cristiana Berti: study design; data collection, analysis, and interpretation; and revision of paper. Cornelius M Smuts: Scientific input in data analysis and interpretation and revision of paper. Mieke Faber: Scientific input in data analysis and interpretation and revision of paper. Namukolo Covic: Study design; scientific input in data interpretation; and revision of paper.

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References

1. Black RE, Victora CG, Walker SP, et al. Maternal and child undernutrition and overweight in low-income and middle-income countries. *Lancet*. 2013;382(9890):427-451.
2. WHO HQ/AFRO. Maternal, infant and young child nutrition in East and Southern African countries: moving to national implementation, report of a World Health Organization workshop, Entebbe, Uganda, 26–28 November 2013. Geneva: World Health Organization; 2014.
3. Bhutta ZA, Das JK, Rizvi A, et al. Evidence-based interventions for improvement of maternal and child nutrition: what can be done and at what cost? *Lancet*. 2013;382(9890):452-477.
4. Shisana O, Labadarios D, Rehle T, et al. *The South African National Health and Nutrition Examination Survey: SANHANES-1*. Cape Town: HSRC Press; 2014.
5. Tylleskär T, Jackson D, Meda N, et al. Exclusive breastfeeding promotion by peer counsellors in sub-Saharan Africa (PROMISE-EBF): a cluster-randomised trial. *Lancet*. 2011;378(9789):420-427.
6. MacIntyre U, De Villiers F, Baloyi P. Early infant feeding practices of mothers attending a postnatal clinic in Ga-Rankuwa. *S Afr J Clin Nutr*. 2005;18:70-75.
7. Kruger R, Gericke G. Breast feeding practices of mothers with children (aged 0-36 months) in a rural area of South Africa: a qualitative approach. *JFECS*. 2001;29:60-71.
8. Faber M. Complementary foods consumed by 6–12-month-old rural infants in South Africa are inadequate in micronutrients. *Public Health Nutr*. 2005;8(4):373-381.

9. Faber M, Laubscher R, Berti C. Poor dietary diversity and low nutrient density of the complementary diet for 6- to 24-month-old children in urban and rural KwaZulu-Natal, South Africa [published online August 19, 2014]. *Matern Child Nutr.* 2014. doi:10.1111/mcn.12146.
10. Arimond M, Zeilani M, Jungjohann S, et al. Considerations in developing lipid-based nutrient supplements for prevention of undernutrition: experience from the International Lipid-Based Nutrient Supplements (iLiNS) Project [published online May 6, 2013]. *Matern Child Nutr.* 2013. doi:10.1111/mcn.12049.
11. Siega-Riz AM, Estrada Del Campo Y, Kinlaw A, et al. Effect of supplementation with a lipid-based nutrient supplement on the micronutrient status of children aged 6–18 months living in the rural region of Intibucá, Honduras. *Paediatr Perinat Epidemiol.* 2014;28(3):245-254.
12. Iannotti LL, Dulience SJL, Green J, et al. Linear growth increased in young children in an urban slum of Haiti: a randomized controlled trial of a lipid-based nutrient supplement. *Am J Clin Nutr.* 2014;99(1):198-208.
13. Phuka J, Ashorn U, Ashorn P, et al. Acceptability of three novel lipid-based nutrient supplements among Malawian infants and their caregivers. *Matern Child Nutr.* 2011;7(4):368-377.
14. Hess SY, Bado L, Aaron GJ, Ouédraogo JB, Zeilani M, Brown KH. Acceptability of zinc-fortified, lipid-based nutrient supplements (LNS) prepared for young children in Burkina Faso. *Matern Child Nutr.* 2011;7(4):357-367.
15. Adu-Afarwuah S, Lartey A, Zeilani M, Dewey KG. Acceptability of lipid-based nutrient supplements (LNS) among Ghanaian infants and pregnant or lactating women. *Matern Child Nutr.* 2011;7(4):344-356.
16. Matias S, Chaparro C, Perez-Exposito A, Peerson J, Dewey K. *Acceptability of Lipid-Based Nutrient Supplement among Guatemalan Infants and Young Children* (Technical report FHI 360). Washington, DC: FANTA-2; 2011:1-33.
17. Gibson RS, Bailey KB, Gibbs M, Ferguson EL. A review of phytate, iron, zinc, and calcium concentrations in plant-based complementary foods used in low-income countries and implications for bioavailability. *Food Nutr Bull.* 2010;31(2 suppl):134-146.
18. Georgieff MK. Nutrition and the developing brain: nutrient priorities and measurement. *Am J Clin Nutr.* 2007;85(2):614S-620 S.
19. Hoffman DR, Boettcher JA, Diersen-Schade DA. Toward optimizing vision and cognition in term infants by dietary docosahexaenoic and arachidonic acid supplementation: a review of randomized controlled trials. *Prostaglandins Leukot Essent Fatty Acids.* 2009;81(2-3):151-158.
20. Hoffman DR, Theuer RC, Castañeda YS, et al. Maturation of visual acuity is accelerated in breast-fed term infants fed baby food containing DHA-enriched egg yolk. *J Nutr.* 2004;134(9):2307-2313.
21. Clandinin MT, Van Aerde JE, Merkel KL, et al. Growth and development of preterm infants fed infant formulas containing docosahexaenoic acid and arachidonic acid. *J Pediatr.* 2005;146(4):461-468.
22. Cercamondi CI, Egli IM, Mitchikpe E, et al. Iron bioavailability from a lipid-based complementary food fortificant mixed with millet porridge can be optimized by adding phytase and ascorbic acid but not by using a mixture of ferrous sulfate and sodium iron EDTA. *J Nutr.* 2013;143(8):1233-1239.
23. Du Plessis LM, Kruger H, Sweet L. Complementary feeding: a critical window of opportunity from six months onwards. *S Afr J Clin Nutr.* 2013;26(3):S129-S40.
24. Marfell-Jones M, Stewart A, De Ridder H. *ISAK Accreditation Handbook*. Upper Hutt, New Zealand: International Society for the Advancement of Kinanthropometry; 2012.
25. Rabiee F. Focus-group interview and data analysis. *Proc Nutr Soc.* 2004;63(4):655-660.
26. Schulz KF, Grimes DA. Allocation concealment in randomised trials: defending against deciphering. *Lancet.* 2002;359(9306):614-618.
27. Mennella JA, Forestell CA, Morgan LK, Beauchamp GK. Early milk feeding influences taste acceptance and liking during infancy. *Am J Clin Nutr.* 2009;90(3):780S-788 S.
28. Ruel MT, Alderman H. Nutrition-sensitive interventions and programmes: how can they help to accelerate progress in improving maternal and child nutrition? *Lancet.* 2013;382(9891):536-551.
29. Peltó GH, Armar-Klemesu M, Siekmann J, Schofield D. The focused ethnographic study 'assessing the behavioral and local market environment for improving the diets of infants and young children 6 to 23 months old' and its use in three countries. *Matern Child Nutr.* 2013;9(suppl 1):35-46.