AN EVALUATION OF COMMON HEALTH AND NUTRITIONAL RISK FACTORS FOR ANAEMIA IN RURAL WOMEN BETWEEN 25 AND 49 YEARS

UNIVERSITY OF THE
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HEALTH SCIENCES
GESONDHEIDSWETENSKAPPE
SCHOOL FOR ALLIED HEALTH PROFESSIONS
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Elizabeth Margaretha Jordaan

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Elizabeth Margaretha Jordaan

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Supervisor: Prof CM Walsh

Co-supervisor: Dr VL van den Berg

Co-supervisor: Mr FC van Rooyen

I, Elizabeth Margaretha Jordaan declare that the master's research dissertation or publishable interrelated articles that I herewith submit at the University of the Free State, is my independent work and that I have not previously submitted it for a qualification at another institution of higher education.

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LIST OF ABBREVIATIONS

Assuring Health for All in the Free State	AHA-FS
Body mass index	BMI
Baccalaureas Scientiae	BSc
Centimetre	cm
Cluster of differentiation	CD4
Decilitre	dL
Deoxyribonucleic acid	DNA
Department of Health	DoH
Department of Health South Africa	DoHSA
Department of Justice and Constitutional Development	DoJ & CD
Dietary diversity	DD
Dietary diversity score	DDS
Fluid	fl
Food and Agriculture Association	FAO
Free State Rural Development Partnership Programme	FSRDPP
Glycated haemoglobin	HbA1c
Grams	g
Haematocrit	Hct
Haemoglobin	Hb
Holotranscobalamin II	holo TCII
Human immunodeficiency virus	HIV
International Business Machines	IBM
Institute of Medicine	IOM
Intrinsic factor	IF
Kilogram	kg
Litre	L
Mangaung University Community Partnership Programme	MUCPP
Mean corpuscular haemoglobin	MCH
Mean corpuscular haemoglobin concentration	MCHC

Mean corpuscular volume	MCV
Meat-fish-poultry	MFP
Medical Research Council	MRC
Metre	m
Methylene tetrahydrofolate reductase gene	MTHFR
Micromoles	µmol
Millennium Development Goal	MDG
Millilitres	ml
Nanogram	ng
Nanomoles	nmol
National Health Laboratory Service	NHLS
Percentage	%
Picogram	pg
Predictive Analytics SoftWare	PASW
Ribonucleic acid	RNA
South African Health and Nutrition Examination Survey	SANHANES
Statistical Package for the Social Sciences	SPSS
Standard Committee on Nutrition	SCN
Tetrahydrofolic acid	THFA
Total-iron binding capacity	TIBC
Transcobalamin I	TCI
Transcobalamin III	TCIII
United Nations	UN
United Nations Development Programme	UNDP
United Nations Population Fund	UNFPA
World Bank	WB
World Health Organisation	WHO

CHAPTER 1

ORIENTATION TO THE STUDY

1.1 BACKGROUND AND MOTIVATION

"Improve maternal health" is the fifth Millennium Development Goal (MDG) to be achieved by the year 2015. In order to reach this goal and thus ensure a woman's safe passage to motherhood, quality reproductive health services, accompanied by a series of well-timed interventions should be implemented (UN, 2010:30). Worldwide 56 million pregnant and 468 million non-pregnant women were affected by anaemia in the period from 1993 to 2005. The prevalence of anaemia in pregnant and non-pregnant women in Africa was 65.8% and 61.4% respectively. In the 1993 to 2005 period, it was estimated that 21.8% of pregnant women in South Africa had haemoglobin levels below 11g/dL; and 26.4% of non-pregnant women in South Africa had haemoglobin levels below 12g/dL which, according to the World Health Organisation (WHO) (2008:Online), is below the haemoglobin threshold used to define anaemia for these two population groups. Anaemia is a global public health problem that not only affects women in developing countries, but those in developed countries as well. Anaemia holds major consequences for both human health, as well as economic development (WHO, 2008:Online).

Anaemia can be defined as a significant reduction in the mass of circulating red blood cells, resulting in a diminished oxygen binding capacity of blood. Patients suffering from anaemia experience a decrease in the concentration of red blood cells or haemoglobin in peripheral blood (Bunn, 2011:1031). Nutritional anaemias are anaemias that result from the insufficient bioavailability of haemopoietic nutrients (iron, vitamin B12 and folic acid) that are essential for meeting the demands for the synthesis of haemoglobin and red blood cells. A marked decrease in bioavailable haemopoietic nutrients over time resulted from the shift from a hunter-gatherer diet to a more cereal-based diet (Balarajan et al., 2011:2127).

Iron deficiency is the most common worldwide cause of anaemia and mostly affects children and women of childbearing age (Gallagher, 2012:110; Ginder, 2011:1039). Iron deficiency is reported to affect approximately four to five billion people worldwide (SCN, 2004:Online). In South Africa, nationally representative data on iron deficiency anaemia is scarce. A study conducted in 2000 in South Africa estimated that 9-12% of pregnant women suffered from iron deficiency anaemia and that 4.9% of maternal deaths were attributed to iron deficiency anaemia in the year 2000 (Nojilana et al., 2007:744). According to more recent findings of the South African National Health and Nutrition Examination Survey (SANHANES) in 2012, the prevalence of anaemia amongst women of children bearing age was 24.2% in females 15-24 years of age, 24.7% in females 24-34 years of age, 23.1% in females 35-44 years of age and 23.7% in females 45-54 years of age (Shisana et al., 2013:163).

The metabolism of iron is complex as it is involved in many aspects of life, such as red blood cell functioning and myoglobin activity, and influences the roles of various haem and non-haem enzymes. Iron is also involved in immune functioning and cognitive performance (Gallagher, 2012:108). Several biological processes within the body are reliant on the presence of iron (Balarajan et al., 2011:2128). Iron plays a significant role in the blood and respiratory transport of oxygen and carbon dioxide and is an integral function of the haemoglobin molecule (Gallagher, 2012:108; Balarajan et al., 2011:2128).

A shortage of stored iron is the first stage of iron deficiency (prelatent iron deficiency), which is reflected in a reduced plasma ferritin concentration. When the iron stores are depleted, a transport iron deficiency develops (latent iron deficiency). At this stage, haemoglobin synthesis is still adequate. This condition may progress, with additional stress or loss of iron, and manifest an iron deficiency with hypochromic microcytic anaemia (Wick, Pinggera & Lehmann, 2011:26). Clinical and population studies have shown that the haemoglobin value itself is a poor indicator of iron deficiency anaemia, as a drop in haemoglobin concentration is frequently the last measurement to become abnormal in iron deficiency and only signifies advanced iron deficiency (Skikne & Hershko, 2012:252).

Causes of iron deficiency typically include injury, haemorrhage or illness and an iron deficiency may be aggravated by an unbalanced diet containing insufficient iron, protein,

folate and vitamin C (Gallagher, 2012:109). Periods of rapid growth, such as pregnancy, results in substantial demands for iron. The expansion of red-cell mass along with the development and maintenance of the maternal-placental-foetal unit results in a considerable increase in iron requirements during pregnancy. Iron losses in women of childbearing age in the form of menstrual losses can account for approximately 0.48mg of iron loss per day (Balarajan et al., 2011:2128). These losses may accumulate over time and may impact on iron status when taking into consideration that the daily iron requirement of menstruating women is 18mg/day (IOM, 2006:328). Patients with mild iron deficiency anaemia may be asymptomatic due to compensatory physiologic mechanisms. The manifestations of iron deficiency anaemia are non-specific and include general symptoms such as weakness, pallor, dizziness, decreased exercise tolerance, and irritability. Iron deficiency affects various organ systems in the human body (Ginder, 2011:1041). Poor iron status in the mother has been found to increase the vulnerability of infants to iron deficiency anaemia as infants who are born to iron deficient mothers have reduced iron stores at birth (Balarajan et al., 2011:2127).

The term megaloblastic anaemia refers to a group of disorders that are characterised by a distinct morphologic pattern in hematopoietic cells (Antony, 2008:491). Megaloblastic anaemia is a condition that is characterised by large red blood cells with malformed nuclei (Balarajan et al., 2011:2128). A state of unbalanced cell growth and impaired cell division as a result of a defect in deoxyribonucleic acid (DNA) synthesis is a common biochemical feature of megaloblastic anaemia (Antony, 2008:491). According to Elghetany and Banki (2011:562), megaloblastic anaemia is almost always due to vitamin B12 or folic acid deficiency.

According to the Standard Committee on Nutrition's (SCN) Fifth Report on the World Nutrition Situation, folic acid deficiency may be indirectly associated with an increased risk for maternal death and illness (SCN, 2004:Online). Folic acid is an essential nutrient for the synthesis and maturation of red blood cells. Low concentrations of serum and erythrocyte folate may result in changes in cell morphology, intramedullary death of red blood cells and reduced erythrocyte lifespan (Balarajan et al., 2011:2128). A deficiency of folate results in impaired synthesis of DNA and ribonucleic acid (RNA), which in turn results in a reduction in

cell division. Cells in which this reduction is most apparent include red blood cells, leukocytes and epithelial cells of the gastrointestinal tract, all rapidly multiplying cells (Gallagher, 2012:84). Folate is widely available in nature and is synthesised by both microorganisms and plants. Although a balanced Western diet contains sufficient amounts of folate, the net dietary intake in many developing countries still remains insufficient (Antony, 2008:495).

Naturally occurring folate in food is primarily present in the reduced, more unstable polyglutamated form, whereas fortified foods and supplements contain folic acid, the non-natural, synthetic, and fully oxidised monoglutamate form of folate. Folate occurring in food has a lower bioavailability, defined as the proportion of folate that is absorbed and available for metabolic reactions and storage, in relation to folic acid (Caudill, 2010:1455S). Since folate is highly susceptible to breakdown during cooking, cultural and ethnic cooking practices such as boiling of lentils or beans in large volumes of water or frying of foods in an open pan can result in folate losses of between 50 to 95% (Antony, 2008:495).

Folate demands increase during pregnancy and women who enter pregnancy with a poor folate status often develop megaloblastic anaemia (Balarajan et al., 2011:2128). Folate helps with the prevention of malformations that have an effect on the brain and spinal cord of the infant (SCN, 2004:Online). An association exists between low red blood cell folate levels in the mother and the susceptibility of offspring to neural tube defects (Warner, 2007:1451). Cellular folate deficiency during pregnancy can adversely affect cells of the neural tube that are responsible for the closure of the neural tube during embryogenesis. Neural tube closure occurs by the 28th day after conception, thus neural tube defects originate within the first month of pregnancy when most women do not yet know that they are pregnant. It is thus crucial that women of childbearing age ensure that their folate status is adequate prior to conception in order to ensure the availability of folate to the foetus (Antony, 2008:511; Brown, 2010:10). Introducing additional folate after this critical period cannot reverse previous damage due to a lack of this nutrient (Brown; 2010:10).

Folate deficiency may also be associated with an increased risk for the development of ischemic heart disease and stroke as a folate deficiency may lead to raised levels of

homocysteine (Koppel, 2011:2384). Homocysteine is an amino acid derived from the demethylation process of methionine (Gallagher, 2012:201; Ridker & Libby, 2011:927). Severe hyperhomocysteinemia (plasma homocysteine levels greater than 100mmol/L) can develop in persons with rare inherited defects in methionine metabolism and thus have a markedly elevated risk of premature atherothrombosis and venous thromboembolism. A common polymorphism in the methylene tetrahydrofolate reductase gene (MTHFR) has been linked to elevated homocysteine levels and thus increased vascular risk, as increased homocysteine levels enhance blood clot formation and arterial wall damage, particularly in individuals homozygous for the variant (Ridker & Libby, 2011:927). Folate and vitamin B12 play an integral role in the demethylation of dietary methionine, an amino acid (Gallagher, 2012:201; Ridker & Libby, 2011:927). According to Ridker and Libby (2011: 927), familial association studies have reported higher homocysteine levels in offspring of parents with premature coronary artery disease, however, the clinical importance of the MTHFR appears to be modest and little evidence of elevated homocysteine levels, even in those with low folate intake, has been reported (Ridker & Libby, 2011: 927).

It is imperative that women consume a diet sufficient in all the right nutrients before and during pregnancy in order to enhance fertility, to support the development of pregnancy and the growing foetus, and to promote long-term health. It is not uncommon to find women who consume diets low in a variety of nutrients which creates concern as a considerable proportion of pregnancies are unplanned (Derbyshire, 2011:26). Gallagher (2012:349) stated that the effect of poor nutritional status follows both the mother and the infant for decades. According to Brown (2010:3), several aspects of maternal health and lifestyle prior to pregnancy have been shown to have an effect on the mother's subsequent pregnancies with potential to impact the health of her children. The best start to pregnancy, to benefit both mother and child, can be achieved by ensuring the mother's nutrient stores are optimal prior to conception (Derbyshire, 2011:26). Improving maternal health, the fifth MDG, will have a direct effect on reducing death among new-borns and young children, thus influencing the fourth MDG (reduce child mortality) as well (WHO, 2010:Online).

1.2 PROBLEM STATEMENT

Iron deficiency anaemia is associated with approximately 111 000 maternal deaths among pregnant women each year (SCN, 2004:Online). According to Nojilana et al. (2007:742), nationally representative data on the prevalence of iron deficiency in South Africa is limited. The SCN (2004:Online) stated that there is a great need for more and better data to describe this serious nutritional deficiency. Iron deficiency remains one of the leading risk factors and contributors to the global burden of disease (SCN, 2004:Online).

Most of the evidence regarding the health consequences of anaemia relates specifically to iron deficiency anaemia (Balarajan et al., 2011:2130). Folate deficiency, however, also contributes to the development of anaemia and may therefore indirectly be associated with increased risk of maternal death and illness (SCN, 2004:Online). According to Balarajan et al. (2011:2128), little global data exist on the contribution of folate deficiency to the development of anaemia. According to the SCN (2004:Online), numerous studies provide strong support for public health policies and programmes to increase folic acid intake prior to becoming pregnant (Warner, 2007: 1451; SCN, 2004:Online).

Evidence indicates that the mother's nutrition and health status prior to pregnancy should receive attention in ensuring that MDG number five and consequently MDG number four is addressed. Since anaemia is one of the major concerns, not only for maternal health but for child health as well, and may have detrimental effects on the growth and development of the foetus before the mother even knows she is pregnant, it is imperative that the mother's nutrient status before pregnancy receives attention. Evidence regarding the occurrence of deficiencies in iron and folate as well as anaemia due to these deficiencies, need to be determined, as current evidence, especially in women of childbearing age in South Africa, is lacking. Data on anaemia is currently lacking in the Free State as well, with the limited available data mainly focusing on iron status and not on folate. Thus, it was decided to focus on the rural areas in the Free State in the current study.

1.3 AIM AND OBJECTIVES

The main aim of this study was to evaluate common health and nutritional risk factors of anaemia in women between 25 and 49 years of age living in rural areas in the Southern Free State.

1.3.1 Objectives

In order to achieve the main aim, the following were determined:

- dietary intake in order to determine dietary diversity;
- anthropometry (weight, height, skinfold measurements and waist circumference);
- reported health;
- socio-demographics;
- contraceptive use;
- biochemical indices of anaemia (full blood count, serum ferritin, transferrin, homocysteine and red cell folic acid); and,
- relevant associations between anaemia and the above mentioned variables.

1.4 OUTLINE OF THE DISSERATION

Chapter 1 serves are orientation to the study and consists of an overview. Chapter 2 provides a literature review of relevant information and variables researched in the study. Chapter 3 explains the methodology followed in the study. Chapters 4 to 7 are structured as a series of articles compiled according to the aims and objectives of the research study. Chapter 8 summarises the conclusions and recommendations for future interventions, based on the research findings.

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CHAPTER 2

ANAEMIA

2.1 INTRODUCTION

Anaemia is defined as a deficiency in the size or number of erythrocytes or the amount of haemoglobin they contain (Janz & Hamilton, 2014:1586). When anaemia is present, the blood's oxygen binding capacity is reduced and the exchange of oxygen and carbon dioxide between the blood and the tissues is limited (Janz & Hamilton, 2014:1586; Bunn, 2011:1031). Anaemia can develop as decreased erythrocyte production (ineffective erythropoiesis) as a result of impaired proliferation of erythrocyte precursors or ineffective maturation of erythrocytes; or increased loss of erythrocytes through increased destruction (haemolysis) or blood loss; or both. These processes are broadly influenced by nutrition, infectious disease and genetics (Balarajan et al., 2011:2126).

Anaemias are classified based on the size of the erythrocytes, namely macrocytic (larger than normal), normocytic (normal) and microcytic (small) and the haemoglobin content, namely hypochromic (pale) and normochromic (normal) (Janz & Hamilton, 2014:1590).

A lack in sufficient bioavailable haemopoietic nutrients may result in nutritional anaemias due to the inability to meet the demands for haemoglobin and erythrocyte synthesis (Balarajan et al., 2011:2127). Nutritional anaemias can develop due to inadequate intakes of iron, protein, certain vitamins, copper and other heavy metals (Stopler & Weiner, 2012:726; Fishman, Christian & West, 2000:125).

Iron deficiency anaemia is characterised by the production of small (microcytic) erythrocytes with a diminished level of circulating haemoglobin (hypochromic) (Janz & Hamilton, 2014:1590; Elghetany & Banki, 2011:559). Microcytic hypochromic anaemia is the last stage of iron deficiency, and it represents the end point of a long period of iron deprivation (Stopler & Weiner, 2012:727; Elghetany & Banki, 2011:559).

Megaloblastic anaemia reflects a disturbed synthesis of DNA, which results in morphologic and functional changes in erythrocytes, white blood cells, platelets, and their precursors in the blood and bone marrow (Elghetany & Banki, 2011:561). The defective DNA synthesis is the result of a lack of the coenzyme forms of vitamin B12 and folic acid and is characterized haematopoietically by ineffective erythropoiesis and pancytopenia. Macrocytic anaemia presents with larger-than-normal erythrocytes (Janz & Hamilton, 2014:1592).

The prevalence, aetiology, clinical manifestations and prevention and management of iron deficiency anaemia, as well as megaloblastic anaemias due to a vitamin B12 and folic acid deficiency, will be reviewed in this chapter. Brief overviews of the metabolism of these nutrients are also included.

2.2 IRON DEFICIENCY ANAEMIA

2.2.1 Iron metabolism

2.2.1.1 Absorption

Iron balance is mostly maintained through absorption i.e. more iron is absorbed when stores are empty and vice versa. Absorption of iron partly depends on the dietary source (Hallberg, 1981:124). Dietary iron exists in two forms, namely haem iron, which is found in haemoglobin, myoglobin as well as some enzymes (Nojilana, 2007:741; Hallberg, 1981:128); and non-haem iron which is mostly found in plant foods and to a lesser extent in some animal foods as non-haem enzymes and ferritin (Nojilana, 2007:741; Hallberg, 1981:126; Monsen et al., 1978:135).

Iron absorption in the human body is possible only through protein binding of the Fe^{2+} ion (Wick, Pinggera & Lehman, 2011:3). Absorption of iron occurs in the duodenum and upper jejunum (Wick, Pinggera & Lehman, 2011:4). The absorption of iron can be described in four phases. In the first phase, also known as the luminal phase due to its site of occurrence, the haem group is removed from the haemoglobin, myoglobin and other haem containing

protein molecules of food, through the digestion process. Prior to its absorption, non-haem iron needs to be removed from plant foods though digestion (ASSAf, 2013:123).

The second phase of iron absorption is known as the mucosal phase and occurs at the brush border of duodenal mucosal cells responsible for absorption as indicated in figure 2.1 (ASSAf, 2013:123). The translocation of haem across the brush-border membrane occurs with the aid of the apical haem carrier (HCP-1) after which the haem molecule is degraded by haem oxygenase (HO) to release the iron (Muckenthaler & Lill, 2012:31). Entry of non-haem iron at the brush border membrane differs from that of haem iron. At the brush border, duodenal cytochrome B converts Fe^{3+} (ferrous iron) to Fe^{2+} (ferric iron) and the divalent metal transporter (DMT1) transports non-haem iron, via facilitated diffusion down a concentration gradient, into the cells (ASSAf, 2013:123; Muckenthaler & Lill, 2012:31). The iron present in food needs to be reduced as it occurs predominantly in the Fe^{3+} form (Wick, Pinggera & Lehman, 2011:4).

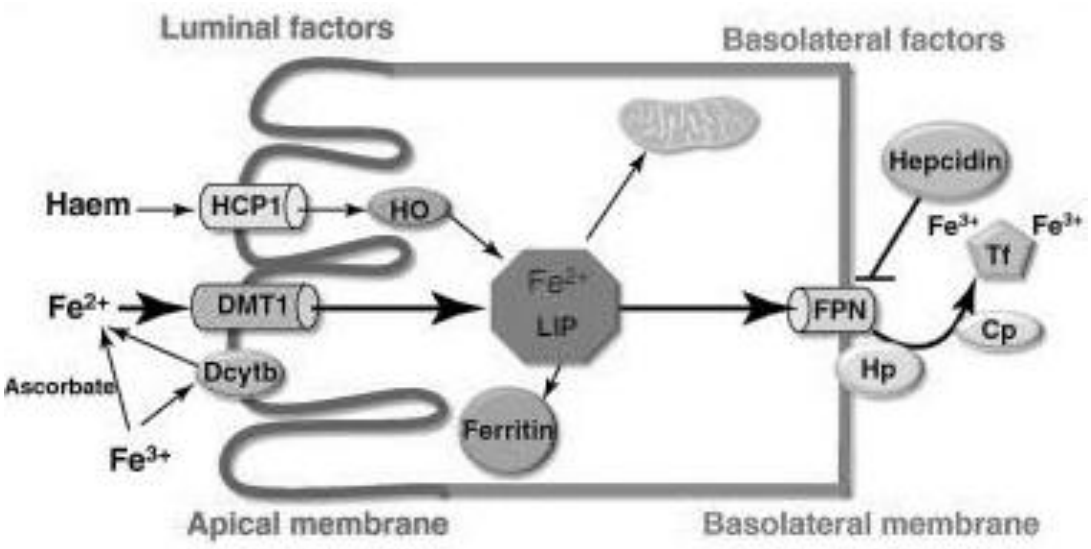

Figure 2.1: Absorption of iron at the brush border of duodenal mucosal cells (Murgia et al., 2011:50).

During the intracellular phase, the third phase of iron absorption, both iron from haem and non-haem sources, either combines with apoferritin to form the storage complex ferritin or it is passed on to the basolateral membrane to follow the exit step of absorption (ASSAf, 2013:123). During the release phase, the fourth phase, the iron is released into the portal

circulation via an active transport mechanism (ASSAf, 2013:123; Muñoz, Villar & García-Erce 2009:4617).

Tight control of the release of iron into the portal circulation occurs through the hormone hepcidin. Production of hepcidin occurs in the liver according to the body's iron needs (ASSAf, 2013:123). Inhibition of the release of iron from mucosal cells is considered to be the major action of hepcidin and its production is thus down-regulated when the need for iron is high (ASSAf, 2013:123; Nemeth & Ganz, 2006:727).

The bioavailability of haem iron is high and is not influenced by dietary factors (Hallberg, 1981:128). The intestinal absorption of non-haem iron can however by affected by various factors, including dietary factors (Hallberg, 1981:126).

a) Conditions in the gastrointestinal tract that affect iron absorption

The solubility and bioavailability of non-haem iron is affected by the degree of gastric acidity; the more acidic the environment, the more bioavailable the iron (Gallagher, 2012:108; Jacobs & Miles, 1969:228). Although iron is usually readily absorbed, primarily in the duodenum, pathologic states that can impair the process include generalized intestinal malabsorption, atrophic gastritis with achlorhydria, and extensive gastric surgery (Ginder, 2011:1041). The use of alkaline substances such as antacids may also interfere with the absorption of non-haem iron. All of these conditions act by prohibiting the solubilisation of iron in gastric and duodenal fluids (Gallagher, 2012:108).

b) Factors that enhance non-haem iron absorption

The absorption of non-haem iron may be enhanced by several dietary factors (Hallberg & Hulthén, 2000:1147). To some extent, the efficiency at which iron absorption occurs may be influenced by the food source from which the iron is derived or with which the iron source is consumed (Gallagher, 2012:108; Fairweather-Tait, 2004:522).

Ascorbic acid is one of the most potent enhancers of iron absorption as it reduces ferric iron to ferrous iron. It also forms a chelate with iron that remains soluble at the alkaline pH of the lower small intestine (Gallagher, 2012:108; Ginder, 2011:1040; Fairweather-Tait, 2004:523). It is important to consume the vitamin C and iron in the same meal (Whitney & Rolfes, 2013:407; Fairweather-Tait, 2004:523). Sugars and sulphur-containing amino acids may also enhance non-haem iron absorption through the formation of chelates with ionic iron (Gallagher, 2012:108; Fairweather-Tait, 2004:523).

Meat, poultry and fish contains haem iron, as well as a peptide called meat-fish-poultry (MFP) factor, of which the specific nature is unknown, that enhances non-haem iron absorption when consumed in the same meal (Whitney & Rolfes, 2013:407; Gallagher, 2012:108; Fairweather-Tait, 2004:523).

c) Factors that inhibit non-haem iron absorption

Various dietary factors bind with non-haem iron thus inhibiting its absorption. Dietary factors that inhibit the absorption of non-haem iron include phytates in legumes, whole grains and rice, the vegetable proteins in soybeans, other legumes and nuts, the calcium in milk, and the polyphenols in tea (tannins), coffee, grain products, oregano and red wine (Whitney & Rolfes, 2013:407; Gallagher, 2012:108; Fairweather-Tait, 2004:523).

2.2.1.2 Transport, cellular uptake, storage and excretion

Following the release of iron into the portal circulation, Fe^{2+} is bound to transferrin which is the main transport protein for iron in circulation (ASSAf, 2013:123). Transferrin is responsible for the transport of iron to the required sites and storage (Srai & Sharp, 2012:11; Ginder, 2011:1040; Newsholme & Leech, 2010:347). Serum transferrin saturation can be used as a measure of iron status (ASSAf, 2013:123).

Transferrin collects iron absorbed from the intestine as well as iron released from the macrophages. The iron is transported to the cells that require it, particularly the cells in the bone marrow and other proliferating cells, where the transferrin binds to receptors on the

plasma membrane in order for the complex to enter the cells via endocytosis (Newsholme & Leech, 2010:348). Ferritin is the major protein associated with the storage of intracellular iron in both the cytoplasm and mitochondria (Srai & Sharp, 2012:11; Ginder, 2011:1040; Newsholme & Leech, 2010:347). Transferrin receptors are expressed on the cells of those tissues that need iron. These receptors bind to transferrin in order for the iron to be absorbed into the cells via a receptor mediated absorption process (ASSAf, 2013:123).

Iron is stored as ferritin in the liver, which serves as the main storage organ for excess iron. Macrophages in the liver and spleen may also serve as sites for iron storage (ASSAf, 2013:123). Since ferritin is present in the circulation, it may also be measured in serum and used as an indicator of iron stores (ASSAf, 2013:123; Gallagher, 2012:108; WHO, 2011:Online). Small amounts of iron are also stored as another storage protein called haemosiderin in the liver (ASSAf, 2013:123). Haemosiderin is produced from ferritin and formation occurs when iron concentrations become abnormally high. Due to its ability to catalyse the formation of free radicals and in itself act as a free radical which can result in cellular damage, free iron is extremely toxic (Ginder, 2011:1040; Valko, Morris & Cronin, 2005:1161). Free iron that is unstable and not incorporated into porphyrin rings is thus associated with proteins (Ginder, 2011:1040).

Most iron is lost from the body through bleeding (Ginder, 2011:1040). Only very small amounts of iron are lost through defecation, perspiration and the normal shedding of hair and skin (Whitney & Rolfes, 2013:408; Gallagher, 2012:108). Iron lost in faeces is mostly from food that could not be absorbed in the digestive tract. The shedding of the cells of the gastrointestinal epithelium and iron present in bile may also contribute to daily iron losses. Urine contains almost no iron (Fuqua, Vulpe & Anderson, 2012:116; Gallagher, 2012: 108).

Wick, Pinggera & Lehman (2011:12) stated that menstruating women lose on average between 30 to 60 mL of blood each month, which is equivalent to an iron loss of approximately 15 to 30 mg of iron. Newholme and Leech (2010:348), however, indicated that blood loss during menstruation can, on average, be between 100 to 200 mL per month, resulting in a loss of approximately 200 mg of iron.

2.2.2 Iron recycling in the body

The maintenance of iron homeostasis in the body is carefully regulated through the absorption, transport, storage, recycling and losses of iron. The hormone hepcidin is produced in the liver and is central to regulating the balance of iron in the body. Hepcidin plays a role in maintaining blood iron within normal range by limiting iron absorption from the small intestine, and controlling the release of iron from the liver, spleen and bone marrow (ASSAf, 2013:124; Whitney & Rolfes, 2013:408).

Approximately 20 to 25 mg of iron is required each day in order for haemoglobin synthesis to take place in the bone marrow (Newsholme & Leech, 2010:347). Iron turnover as a result of senescent erythrocyte degradation is also approximately 20 to 25 mg per day and exceeds the daily iron intake and excretion (Wick, Pinggera & Lehman, 2011:12). Iron is obtained either from the diet or from recycled iron through senescent erythrocyte breakdown (Newsholme & Leech, 2010:347).

Extremely economical recycling of available iron reserves is necessary to meet the requirements for haemoglobin, myoglobin and enzyme synthesis (Wick, Pinggera & Lehman, 2011:12). The reticuloendothelial macrophage system responsible for iron recycling is a potent conservation mechanism through which an average of approximately 1 to 2 mg of iron is lost per day (Ginder, 2011:1040).

In order to ensure homeostasis is maintained, only about 1 to 2 mg of absorbed iron is required daily due to the low rate of iron loss under normal circumstances (Ginder, 2011:1040). According to Wick, Pinggera & Lehman (2011:12), 5 mg of iron is required per day under normal circumstances. The daily iron requirement of menstruating women is 18mg per day (IOM, 2006:328).

2.2.3 Functions

Iron is involved in many aspects of life (Ginder, 2011:1040). Iron is a critical constituent of haemoglobin and myoglobin porphyrin rings, that transport oxygen, cytochromes and other

vital enzymes, due to its' ability to accept or donate electrons through the conversion between the ferrous (Fe^{2+}) and ferric (Fe^{3+}) forms (Ginder, 2011:1040). As a component of haemoglobin and cytochromes, iron is one of the most significant biocatalysts within the body (Wick, Pinggera & Lehmann, 2011:3).

Due to its oxidation-reduction properties, iron plays a role in the blood and respiratory transport of oxygen and carbon dioxide, as well as in the process of cellular respiration and energy generation (Gallagher, 2012:108; Ginder, 2011:1040). Erythrocytes are responsible for the transport of oxygen from the lungs to the tissues and carbon dioxide from the tissues to the lungs. The amount of haemoglobin, its oxygen affinity and blood flow influences the transport of oxygen (Janz & Hamilton, 2014:1586). The haemoglobin content of erythrocytes is determined by the coordinated production of globin protein, the haem porphyrin ring, and the availability of iron (Ginder, 2011:1039).

Another function of iron within the body is the involvement of iron in the iron-containing cytochrome P-450 system (Gallagher, 2012:108; Meunier, de Visser & Shaik, 2004:3947). This system is responsible for the transformation of a variety of water-insoluble and endogenous organic molecules to allow for the secretion of these molecules in bile for elimination (Gallagher, 2012:108; Haseman et al., 1995:41).

Normal immune functioning requires the precise regulation of iron in the body due to iron's ability to compromise cellular functioning in the presence of both deficient and excessive levels. Dietary sufficiency in terms of iron intake is thus required for normal functioning of the immune system (Gallagher, 2012:109; Cherayil, 2011:1).

Since the cells of the brain also need iron for normal functioning (Piňero & Connor, 2000:435), cognitive development and performance will be affected by iron status (ASSAf, 2013:121). The production and functioning of neurotransmitters, and possibly myelin, may also be influenced by iron levels in the body (Piňero & Connor, 2000:435).

2.2.4 Iron deficiency

2.2.4.1 Aetiology

Significant differences in the causes of iron deficiency during different life stages, genders and socioeconomic circumstances exist. Infants, growing children, adolescents during the growth spurt, and menstruating and pregnant women are at risk for developing iron deficiency due to increased physiological requirements (Skikne & Hershko, 2012:251). Several studies however affirm that the use of oral contraceptives is associated with decreased iron deficiency risk (Casabellata et al., 2007:201; Harvey et al., 2005:563; Milman, Kirchhoff & Jorgensen, 1992:101). Iron deficiency can arise from insufficient (total or bioavailable) intake of iron to meet the iron needs or to make up for increased losses (Balarajan et al., 2011:2127).

Other causes of iron deficiency anaemia can include inadequate absorption as a result of diarrhoea, achlorhydria, intestinal disease, or drug interference; inappropriate use secondary to chronic gastrointestinal problems; increased iron needs for increased blood volume or excessive menstrual blood loss, or haemorrhage from injury; faulty release of iron into the plasma from the stores; or ineffective iron use as a result of chronic inflammation or other chronic disorders (Stopler & Weiner, 2012:727; Ginder, 2011:1040). Occult gastrointestinal blood loss as a result of non-steroidal anti-inflammatory drugs, cancer of the stomach or colon, benign gastric ulceration or angiodysplasia; and malabsorption due to coeliac disease, gastrectomy or Helicobacter Pylori infection may also lead to deficiency (Goddard et al., 2011:1310).

According to Skikne & Hershko (2012:251), iron deficiency may develop as a result of a single disorder; however, in most cases an interplay of multiple causative factors leads to the development of iron deficiency. Fanou-Fogny (2010:574) stated that significant correlations between serum iron, fat mass and BMI have been reported in various studies which may suggest that adiposity may possibly have a negative effect on iron status. A study that investigated the association between anaemia and BMI and waist circumference

among Chinese adult women found that overweight/obesity as well as central obesity was inversely associated with anaemia (Qin et al., 2013:2).

According to Ginder (2011:1040), the average Western diet contains approximately 20 mg of iron per day. The efficiency of iron absorption in the duodenum is also sufficient to maintain the amount of iron required to maintain homeostasis (Ginder, 2011:1040). Lower intakes of bioavailable haemopoietic nutrients over time, as well as absorption enhancers such as vitamin C have been experienced as a result of a shift to more cereal-based diets with more heat exposure during food preparation. The accompanied increase in the intake of other dietary factors such as polyphenols, phytates, and calcium, all of which reduce the bioavailability of iron, further complicates the situation. Physiological and pathophysiological factors can further influence the absorption of nutrients that promote haemopoiesis (Balarajan et al., 2011:2127). When the diet contains few inhibitors of iron absorption, approximately 15% of the iron is absorbed which is equivalent to approximately 1.8mg of iron. However, if the diet contains inhibitors of iron absorption, absorption of iron decline by approximately 66% resulting in less than 1 mg of iron being absorbed (Skikne & Hershko, 2012:259). Nutritional anaemias can be worsened in vulnerable groups where access to micronutrient-rich diets is limited (Balarajan et al., 2011:2127).

2.2.4.2 Clinical manifestations

Persons suffering from mild iron deficiency anaemia may be asymptomatic as a result of compensatory physiologic mechanisms (Bunn, 2011:1033; Ginder, 2011:1041). Initially some patients may report symptoms such as fatigue, dyspnoea and palpitations (Bunn, 2011:1033). According to Stopler & Weiner (2012:727), reduced immunocompetence, particularly due to defects in cell-mediated immunity and the parasitic activity of neutrophils, may possibly be an early sign of iron deficiency.

Since anaemia is the final manifestation of chronic iron deficiency, malfunction of various systems within the human body can be experienced with iron deficiency anaemia (Stopler & Weiner, 2012:727; Ginder, 2011:1040). As is the case with most types of anaemia, the symptoms of iron deficiency anaemia are nonspecific including symptoms of weakness,

pallor, dizziness, reduced exercise tolerance or irritability (Ginder, 2011:1041). Decreased work performance and exercise intolerance may be reflective of inadequate muscle function. Fatigue, anorexia, and “pica”, the craving of non-food substances, are behavioural changes that may be indicative of neurological involvement (Stopler & Weiner, 2012:727; Ginder, 2011:1041).

The structure and function of epithelial tissues are greatly affected as iron deficiency anaemia becomes more severe. Epithelial tissues commonly affected include tissues of the tongue, nails, mouth, and stomach. Pale skin and light pink appearance of the lower eyelids may be signs of iron deficiency anaemia (Stopler & Weiner, 2012:728; Ginder, 2011:1041). Changes in and around the mouth may include atrophy of the epithelium of the tongue with burning or soreness, and redness; in severe cases, a completely smooth, waxy and glistening tongue; and angular stomatitis (Stopler & Weiner, 2012:728; Elghetany & Banki, 2011:559).

Gastrointestinal symptoms may occur as a result of the shunting of blood away from the splanchnic bed (Bunn, 2011:1033). Chronic gastritis may occur frequently and can lead to decreased gastric secretions (achlorhydria) (Stopler & Weiner, 2012:728; Elghetany & Banki, 2011:559).

Females with iron deficiency anaemia may develop problems with menstruation, where either amenorrhoea or increased bleeding occurs (Bunn, 2011:1033). Other symptoms may include restless legs syndrome with accompanying leg pain or discomfort, and thin and flat fingernails that eventually appear concave or spoon-shaped (Koilonychia) (Stopler & Weiner, 2012:728; Ginder, 2011:1041).

Where haemoglobin levels fall below 7.5 g/dL, the resting cardiac output may become elevated with an accompanying increase in the stroke volume and heart rate may occur, resulting in patients complaining of a rapid, pounding sensation in the precordium (Bunn, 2011:1033).

2.2.4.3 Management

a) Medical management

The underlying cause of iron deficiency anaemia needs to be identified and corrected (Stopler & Weiner, 2012:730; Bunn, 2011: 1039; Elghetany & Banki, 2011:561). Treatment aims should include the restoration of haemoglobin concentrations and red cell indices to normal levels as well as the replenishment of iron stores (Goddard et al., 2011:1312). According to Stopler & Weiner (2012:730), oral supplementation in the form of ferrous iron is the first line treatment and should be prescribed according to the severity of the anaemia and the patients' tolerance. Recommendations on the length of iron therapy differ slightly with some recommending iron supplementation for at least two months after the normalising of haemoglobin (Elghetany & Banki, 2011:561), and others recommending at least 3 months following the normalisation of haemoglobin levels (Goddard, McIntyre & Scott, 2000:iv3).

The correction of iron deficiency anaemia through oral supplementation may fail in patients who do not take the prescribed medication as indicated; who experience bleeding that continues at a rate that is much faster than the rate of replacement of erythrocytes; or in the case where the supplemental iron is not being absorbed as a result of malabsorption secondary to other conditions such as celiac disease (Ginder, 2011:1042). Patients who do not tolerate or respond to oral iron supplementation may receive parenteral iron therapy (Stopler & Weiner, 2012:730; Goddard et al., 2011:1312). The replenishment of iron stores via the parenteral route is faster, but it is not as safe as administration via the oral route and is more expensive and painful (Goddard, McIntyre & Scott, 2000:iv3).

Transfusion of patients suffering from anaemia may be challenging, however severe anaemia accompanied by myocardial or cerebral ischemia or congestive heart failure, may be an indication for the administration of packed red cells (Bunn, 2011:1039). Transfusions should be followed by oral iron supplementation in order to replenish stores (Goddard et al., 2011:1312).

b) Medical nutrition therapy

The amount of absorbable iron in the diet should be addressed in order to compliment iron supplementation (WHO, 2011:Online). The absorption of iron is best on an empty stomach, but the risk for gastric irritation is increased. Gastrointestinal side effects that may occur when taking iron supplements on an empty stomach include nausea, epigastric discomfort and distension, heartburn, diarrhoea or constipation. Patients who experience these side effects are advised to take the iron supplements with meals (Ginder, 2011:1041).

The bioavailability of dietary iron is important in correcting and preventing iron deficiency, thus factors that improve and inhibit iron absorption should be considered (WHO, 2011:Online). Good food sources of iron include liver, dried beans, egg yolks, kidney, lean beef, dark meat of chicken, salmon, tuna and dried fruits among others (Nojilana et al., 2007:741). Haem iron is well absorbed, even though it accounts for approximately 10% of the average daily iron intake and is found in beef, pork and lamb. The remaining 90% is accounted for by the less well absorbed non-haem iron (Hurrell & Egli, 2010:1461S).

2.3 MEGALOBLASTIC ANAEMIA

Megaloblastic anaemia refers to a group of disorders with a distinct morphologic pattern in in hematopoietic cells (Antony, 2008:491). Characteristics of megaloblastic anaemias include the presence of large, immature abnormal precursors in the bone marrow with decreased capacity for oxygen transfer (Stopler & Weiner, 2012:732; Aslinia, Mazza & Yale, 2006:236). Defective DNA synthesis with lesser alterations in ribonucleic acid (RNA) and protein synthesis that result in a state of unbalanced cell growth and impaired cell division is a common biochemical feature of megaloblastic anaemia (Antony, 2008: 491).

According to Elghetany and Banki (2011:562), megaloblastic anaemia is almost always due to a vitamin B12 or folic acid deficiency. Clinically, the deficiency is observed in those tissues with rapid cell turnover such as hematopoietic cells and cells of mucosal surfaces (Janz & Hamilton, 2014:1592). Vitamin B12 and folic acid are important for nucleoprotein synthesis. Similar clinical results are seen in vitamin B12 and folic acid deficiencies, even though the

developmental histories differ. Folic acid deficiency usually appears before a vitamin B12 deficiency (Janz & Hamilton, 2014:1592; Stopler & Weiner, 2012:732).

Folate levels within the body are normally depleted within 2 to 4 months where individuals consume a folate deficient diet. Vitamin B12 stores, however, are only depleted after following a vitamin B12 deficient diet for several years. The haematological manifestations of a vitamin B12 deficiency can be masked by folic acid supplementation, while irreversible neuropsychiatric damage may occur and can only be corrected with timely vitamin B12 supplementation (Stopler & Weiner, 2012:732; Elghetany & Banki, 2011:564). The diagnosis of macrocytic anaemia thus requires the determination of appropriate folic acid as well as vitamin B12 biochemical values (Wick, Pinggera & Lehman, 2010:42).

The development of a functional folate deficiency may be the result of a vitamin B12 deficiency (Hamilton & Blackmore, 2012:203). The methylfolate trap theory is proposed for this interrelationship between vitamin B12 deficiency and folate deficiency and involves the entrapment of folate in the metabolically useless form of 5-methyltetrahydrofolate. Methionine synthase requires vitamin B12 and plays an essential role in the metabolism of folate as seen in figure 2.2 (Scott & Molloy, 2012:244). This resultant unavailability of folate negatively impacts on pyrimidine and purine synthesis (Hamilton & Blackmore, 2012:203).
The discovery that methionine synthase required vitamin B 12, but also used folate and
was essential for folate metabolism within the cell.

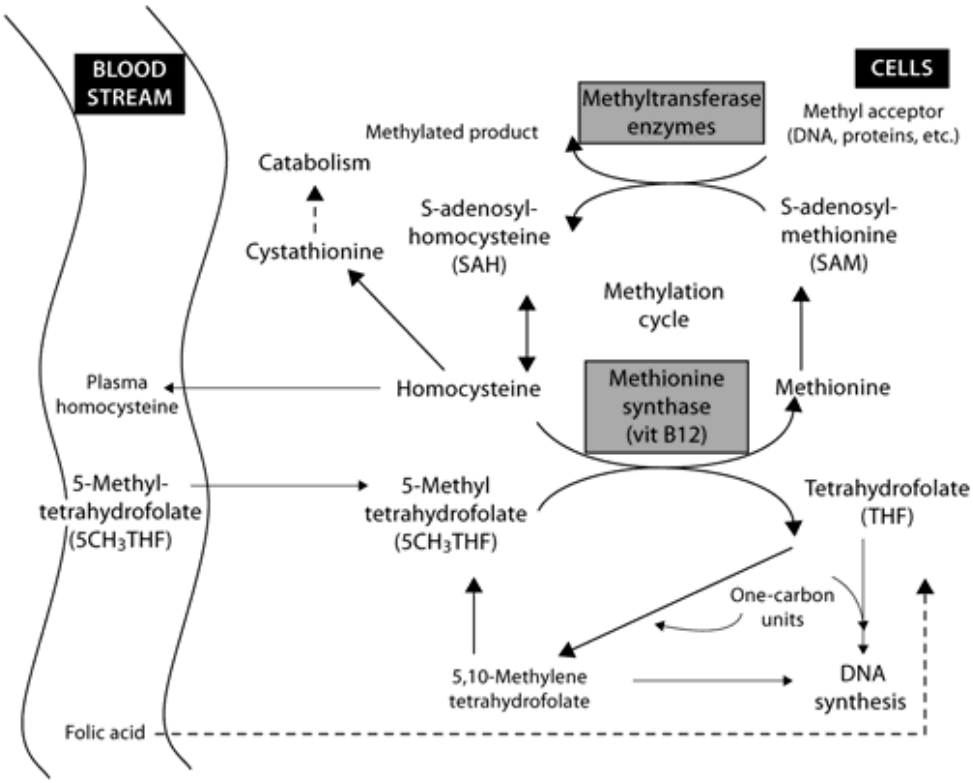

Figure 2.2: Folate-vitamin B12 interactions showing methionine synthase as a central enzyme in the uptake and retention of folate cofactors (Scott & Molloy, 2012:244).

2.3.1 Folate deficiency

2.3.1.1 Metabolism

a) Absorption and digestion

Folate refers to a family of water soluble B vitamins that occurs naturally in food and in biologic organisms (Crider et al., 2012:21). Folic acid refers to the synthetic form of the vitamin used in food fortification and supplements (Stover, 2014:359; Allen, 2008:S28). The efficacy of the absorption of dietary folate in the polyglutamate form is not as high as dietary folate in the monoglutamate form (Hamilton & Blackmore, 2012:203; Antony, 2008:495). Folate present in food occurs predominantly in the polyglutamate form and should thus be converted to the monoglutamate form, by folate hydrolase, in order for effective absorption to take place (Gallagher, 2012:82; Hamilton & Blackmore, 2012:203; Antony, 2008:495).

Folate hydrolase functions best at a pH of 5.5 and can be found in the brush border of the proximal jejunum (Elghetany & Banki, 2011:564). In the jejunum, folate is mainly absorbed via active transport; however, absorption via passive diffusion can also take place in the case of ingestion of large quantities of folate (Gallagher, 2012:82; Antony, 2011:1078).

b) Transport, cellular uptake, storage and excretion

Cells are capable of only taking up monoglutamate derivatives occurring in plasma. Folate uptake from the plasma into tissues occurs at a rapid rate by means of two physiologic transport processes (Antony, 2011:1078). Cellular uptake of these derivatives occurs via an energy-dependent process with specific folate-binding proteins or via a carrier-mediated process. Following uptake by the intestinal mucosal cell, folate is reduced to tetrahydrofolic acid (FH_4). FH_4 is methylated to 5-methyl-FH_4, within the cell, where it is stored at an intracellular level through the binding to intracellular molecules. Low concentrations of 5-methyl-FH_4 occur in tissues with high rates of cell division, where tissues with low rates of cell division have higher levels of 5-methyl-FH_4 (Gallagher, 2012:82). The liver is the most important storage organ of folate (Gallagher, 2012:82; Elghetany & Banki, 2011:564).

The metabolism of folate can occur in the kidney and liver via an enzyme reductase responsible for reduction of the pterin ring; through the activity of the enzyme polyglutamate synthase on the polyglutamyl side chain; or by the acceptance of single-carbon moieties at specific positions on the pterin ring (Gallagher, 2012:82; Antony, 2008:495). The metabolic activation of folate requires the conversion to one of several derivatives and involves the covalent bonding of single-carbon units at the N-5 or N-10 (or both) positions on the pterin ring (Stover, 2014:359; Gallagher, 2012:83; Antony, 2011:495). Folate is degraded to a variety of water-soluble side-chain metabolites that can be excreted in urine and bile (Gallagher, 2012:83; Antony, 2008:497).

2.3.1.2 Functions

Folate is involved in many synthesis reactions, as an enzyme co-substrate, particularly in the metabolism of amino acids and nucleotides by either donating or accepting single carbon

units (Gallagher, 2012:83; Antony, 2008:498). Folate plays an essential role in the remethylation of methionine from homocysteine (Forges et al., 2007:225) and in this process plays a role in the activation of vitamin B12 (Antony, 2008:499; Forges et al., 2007:225).

The biosynthesis of ribonucleotides and deoxyribonucleotide precursors for DNA synthesis also requires the presence of folate (Crider, 2012:21). Due to its role in normal cell division, folate is particularly important during embryogenesis, and may influence the occurrence of serious birth defects. Folate also plays an indispensable role the formation and maturation of red and white blood cells in the bone marrow (Gallagher, 2012:83; Antony, 2008:511).

2.3.1.3 Aetiology

The most common food sources of folate include green vegetables, fortified cereals and fruit (Janz & Hamilton, 2014:1592). The most common causes of folate deficiency include prolonged inadequate diets, faulty absorption and increased requirements during rapid phases of growth (Elghetany & Banki, 2011:32). Causes of folate deficiency, in developing countries especially, comprise reduced availability of folate-containing foods due to seasonal changes; cooking techniques responsible for the destruction of folate; and anorexia that is associated with certain chronic diseases (Antony, 2011:1078). Gluten sensitive enteropathy, idiopathic steatorrhoea, nontropical sprue, and certain medications may be among other causes of folate deficiency (Stopler & Weiner, 2012:733; Antony, 2008:501).

Folate may be completely destroyed during cooking, particularly during periods lasting longer than 15 minutes (Janz & Hamilton, 2014:1592; Elghetany & Banki, 2011:564; Antony, 2008:495). Cooking practices of certain cultural and ethnic groups that involve prolonged boiling in spices and large volumes of water or frying of foods in an open pan may result in folate losses of up to 50 to 95%. The bioavailability of folate in foods may be reduced as a result of oxidation by means of nitrites that are used in the curing of meats (Antony, 2008:495).

Allen (2008:S30), stated that folate deficiency, evidenced by low serum or erythrocyte folate, is commonly experienced in chronic alcoholics. Alcohol not only impairs the nutritional status when used excessively, it may also block the metabolism of folate and thus have a direct effect in suppressing haematopoiesis. The absorption and enetrohepatic circulation of folate can also be affected by alcohol which can result in increased folate losses in the urine (Antony, 2008:495).

Certain medications are known to influence folate status and include anticonvulsants, such as methotrexate which acts as a folate antagonist; sulfasalazine, which is used for the treatment of ulcerative colitis; and pyrimethamine, which is used for the treatment of malaria (Allen, 2008:S30). Proton pump inhibitors may inhibit the human proton-coupled folate transporter and so reduce folate absorption. Oral contraceptives may increase the catabolism of folate (Antony, 2011:1078). Many non-steroidal anti-inflammatory drugs (Baggott et al., 1992:197) and antacids (Russel et al., 1988:458) may also interfere with folate levels within the body as aspirin inhibits the action of folate-requiring enzymes and antacids inhibit folate absorption.

A functional folate deficiency may develop in the presence of a vitamin B12 deficiency as the entrapment of the metabolically inactive form of folate, 5-methyl FH4, will occur (Rolfes, Pinna & Whitney, 2012:310; Stopler & Weiner, 2012:734; Scott & Weir, 1981:337). Vitamin B12 is necessary to remove the 5-methyl unit to form FH4 (Elghetany & Banki, 2011:564). In the presence of either folate of vitamin B12 deficiency, homocysteine levels may rise and spill into the circulation as the conversion of homocysteine to methionine is also dependent on folate and vitamin B12 levels (Litchford, 2012:201; Scott & Weir, 1981:337).

Folate deficiency can occur in four stages. During stage one an early negative folate balance is evident, followed by stage 2 where a negative folate balance as well as a reduction in erythrocyte folate levels develops. In stage three, damaged folate metabolism associated with folate deficient erythrocyte production and slowed DNA synthesis occurs, followed by the final stage, in which a clinical folate-deficient anaemia with an elevated MCV is evident (Stopler & Weiner, 2012:735).

2.3.1.4 Clinical manifestations

The morphologic features of folate deficiency may be indistinguishable from that of vitamin B12 deficiency; however, these features develop much faster in folate deficiency (Heimburger, 2014:761). Folate deficiency may manifest as hematologic, cardiopulmonary, gastrointestinal, dermatologic, genital, infertility, and psychiatric abnormalities (Antony, 2008:510).

Common sings of folate deficiency are fatigue, dyspnoea, sore tongue, diarrhoea, irritability, anorexia, glossitis and weight loss (Stopler & Weiner, 2012:735; Antony, 2008:510). Approximately 20% of patients with folate deficiency may present with peripheral neuropathy (Heimburger, 2014:761). In the absence of folate, DNA damage results in the destruction of erythrocytes as they attempt to divide and mature. Fewer and larger erythrocytes, which are incapable of carrying oxygen or of travelling through the capillaries as efficiently as normal erythrocytes, are the result (Bailey & Gregory, 1999:780).

The growth of tissues may be impaired as a deficiency of folate impairs cell division and protein synthesis. The regeneration of erythrocytes and cells of the gastrointestinal tract is impaired, resulting in anaemia and gastrointestinal tract deterioration respectively (Rolfes, Pinna & Whitney, 2012:312; Antony, 2008:510). The effects on the gastrointestinal tract causes further folate malabsorption which results in a vicious cycle of folate deficiency in the short term and vitamin B12 deficiency in the long term (Antony, 2008:510).

Folate deficiency can have profound adverse effects on cells of the neural tube and neural crests as these cells are responsible for midline closure during embryogenesis and critical periods of proliferation occurs in embryonic neural tube and neural crest cells (Antony, 2008:511). The neural tube develops into the spinal column and brain during the first 28 days after conception (Derbyshire, 2011:135; Antony, 2008:511). Folic acid supplementation in the mother appears to be the most effective intervention in preventing neural tube defects (Stover, 2014:365; Czeizel & Dudas, 1992:1832). Neural tube defects thus develop before many women know they are pregnant. In order to ensure availability of folate for the

embryo, it is thus vitally important that women of childbearing age pay attention to folate intake prior to conception (Antony, 2008:511).

Associations between impairments in folate metabolism and cardiovascular disease, cancers (Blom & Smulders, 2011:80) and cognitive decline, as shown primarily by an elevation in plasma homocysteine or low circulating folate concentrations, have been found (Wald, Kasturirtne & Simmonds, 2010:522).

2.3.2 Vitamin B12 deficiency

2.3.2.1 Vitamin B12 metabolism

a) Absorption and digestion

Vitamin B12 comprises of a family of cobalamin compounds that all contain the porphyrin-like cobalt-centered corrin nucleus (Antony, 2008:492). Several steps exist in the absorption of vitamin B12 (Newsholme & Leech, 2010:334). Dietary vitamin B12 is bound to protein and thus requires release through pepsin digestion in the stomach. Following its release in the stomach, vitamin B12 combines with R protein after which it moves to the small intestine. Hydrolysis of the R proteins is followed by the binding of intrinsic factor (IF) with vitamin B12, once inside the small intestine (Gallagher, 2012:85; Elghetany & Banki, 2011:562; Antony, 2008:492). IF is produced in the stomach and serves as a specific binding protein for vitamin B12. The majority of vitamin B12 is absorbed via active transport, a process for which IF is critical, as only approximately 1% can be absorbed via simple diffusion, even when high amounts of vitamin B12 are present (Gallagher, 2012:85; Antony, 2008:492).

b) Transport, cellular uptake, storage and excretion

Intrinsic factor forms an IF-vitamin B12 complex to enable uptake into the enterocyte through binding with a specific membrane receptor on the ileal brush border (Gallagher, 2012:85; Newsholme & Leech, 2010:334). Vitamin B12 binds to the plasma R proteins, known as transcobalamins (TCs), TCI, TCII and TCIII (Gallagher, 2012:85; Elghetany & Banki,

2011:562). These R proteins also serve to protect the vitamin (Newsholme & Leech, 2012:334). The newly absorbed vitamin B12 is mainly transported in the peripheral tissues by TCII (Gallagher, 2012:85; Elghetany & Banki, 2011:562; Antony, 2008:493). TCII rapidly delivers vitamin B12 to the liver, haematopoietic cells as well as other dividing cells (Elghetany & Banki, 2011:562). Mediation via a specific TC receptor that internalises the TC-vitamin B12 complex controls the uptake of vitamin B12 at cellular level. Lysosomal degradation is needed to free the vitamin B12 in order for it to bind to vitamin B12 dependent enzymes as further intracellular metabolic processes (Gallagher, 2012:85; Antony, 2008:494). The liver is the main site for storage of vitamin B12 in the human body (Elghetany & Banki, 2011:562).

Vitamin B12 is only metabolically active as derivatives that contain either a 5”-deoxyadenosine or a methyl group that is attached covalently to the coring ring cobalt atom. The vitamin is excreted intact via renal and biliary routes as little or no metabolism of the corrinoid ring system takes place. Only cobalamins that occur freely in plasma are available for excretion (Gallagher, 2012:85; Antony, 2008:495).

Methylmalonyl-coenzyme A (CoA) mutase and methionine synthase bind more than 95% of the intracellular vitamin B12. Inside the mitochondria, vitamin B12 plays a role in the metabolism of propionate. Vitamin B12, along with folate, plays an important role in methionine synthesis from homocysteine. Elevated levels of methylmalonic acid and homocysteine in the blood may be an indication of vitamin B12 deficiency (Antony, 2011:1075).

The development of vitamin B12 deficiency can be observed as a series of stages. During stage one an early negative vitamin B12 balance as well as depletion of the primary delivery protein, TCII, can be observed. Stage two is characterised by low vitamin B12 on TCII and a lowering of vitamin B12 in the storage protein, haptocorrin. Damaged metabolism and vitamin B12-deficient erythropoiesis is evident in stage three, whereas clinical damage and vitamin B12 deficiency anaemia follows in stage four (Stopler & Weiner, 2012:737).

2.3.2.2 Functions

Vitamin B12 functions in two coenzyme forms namely adenosylcobalamin and methylcobalamin that are involved in the metabolism of propionate, amino acids, and single carbons (Gallagher, 2012:85; Antony, 2008:491). Vitamin B12, in conjunction with folate, plays a role in various processes within the human body including DNA synthesis, erythrocyte formation and myelinisation of nerves (Singh & Sachan, 2011:858). Vitamin B12 is thus vital for the growth of all cells including the cells in the GI tract, bone marrow and nervous tissue (Gallagher, 2012:85; Antony, 2008:499).

2.3.2.3 Aetiology

Certain microorganisms synthesise vitamin B12 which is ingested by animals. Dietary sources of vitamin B12 thus include meat and meat products (including shellfish, fish and poultry) and to a lesser degree milk and milk products (Newsholme & Leech, 2010:334). Vitamin B12 is thus entirely dependent of the amount of dietary animal sources, except in cases where fortified foods are available (Allen, 2008:S21). Even though vitamin B12 producing bacteria are present in the large bowel of humans, they are located too distal, below the ileum, for physiologic absorption (Antony, 2008:491; Allen, 2008:S21).

Vitamin B12 deficiency rarely develops as a result of inadequate dietary intake (Koppel, 2011:2384). Development of deficiency in strict vegetarians who do not consume any vitamin B12, except for traces found in plants due to contamination by vitamin B12 synthesising microorganisms, is rare (Stopler & Weiner, 2012:736; Elghetany & Banki, 2011:562). Low maternal vitamin B12 status is associated with adverse pregnancy outcomes in these patient groups (Antony, 2011:1075). The majority of the reported cases of clinical and biochemical vitamin B12 deficiency symptoms are seen in infants who were born to, and breastfed by mothers following vegan diets (Allen, 2008:S21). In general, it however takes decades for individuals who are strict vegetarians, who do not receive any vitamin B12 supplementation to develop a vitamin B12 deficiency, due to normal enterohepatic circulation (Stopler & Weiner, 2012:736; Antony, 2008:491).

Malabsorption results in a much more rapid depletion of vitamin B12 than low dietary intakes. Malabsorption can occur as a result of a lack of intrinsic factor (IF), intestinal abnormalities or gastric atrophy resulting in poor release of the vitamin from food (Allen, 2008:S23). Gastric parietal cell atrophy is associated with insufficient hydrochloric acid secretion of which the causes can include total or partial gastrectomy, autoimmune destruction, or destruction of the gastric mucosa by ingestion of corrosive substances (Antony, 2011:1076). Peptic ulcer and chronic gastritis are conditions that are associated with hypochlorhydria, lowered IF production, malabsorption of vitamin B12 and pernicious anaemia. Both of these conditions can be caused by Helicobacter pylori (Stopler & Weiner, 2012:737; Elghetany & Banki, 2011:562). Helicobacter pylori infection may possibly be the main cause of food-bound vitamin B12 malabsorption and gastric atrophy in the elderly as it eventually results in achlorhydria and gastric atrophy (Allen; 2008:S26).

Vitamin B12 deficiency may develop as a result of an autoimmune disorder in which antibodies destroy parietal cells in the stomach (Allen, 2008:S23). This abnormality results in the failure of the gastric mucosa to secrete IF due to gastric parietal cell atrophy (Stabler, 2013:150; Antony, 2011:1076; Elghetany & Banki, 2011:652). Vitamin B12 is consequently malabsorbed and leads to vitamin B12 deficiency, known as pernicious anaemia (Allen, 2008:S23). Pernicious anaemia usually only manifests later in life, with an average age of onset of approximately 60 years, and less than 10% occurs in persons younger than 40 years of age (Antony, 2011:1076; Elghetany & Banki, 2011:562).

Overgrowth of bacteria within the small bowel that develops from stasis or impaired motility, resulting in colonisation, can accumulate free vitamin B12 before it can bind to IF. A short course of antibiotics can solve this problem (Antony, 2011:1077; Allen, 2008:S26). Parasitic infection due to *Diphyllobothrium latum*, the fish tapeworm, may also lead to vitamin B12 deficiency. Consumption of raw or undercooked fish from cold-water lakes and rivers can lead to infection with this parasite. Competition for vitamin B12 develops between the tapeworm and the human host (Allen, 2008:S26).

According to Allen (2008:S27), drugs that impair gastric acid and pepsin secretion (e.g. H2-receptor antagonists and proton pump inhibitors), subsequently impairing the release of

vitamin B12 from food proteins, is the most commonly reported adverse effect of medications on vitamin B12. Drugs, such as the biguanides, can decrease IF and gastric acid secretion and impair the transepithelial transport of vitamin B12 (Antony, 2011:1077; Stopler & Weiner, 2012:736).

Other causes of vitamin B12 may include antibodies to IF in saliva or gastric juice; disorders of the small intestine such as celiac disease, idiopathic steatorrhoea, tropical sprue, cancers affecting the small intestine, and long term alcohol ingestion or use of calcium-chelating agents (Stopler & Weiner, 2012:736; Antony, 2008:504). Patients with HIV may present with low plasma vitamin B12 concentrations. Malabsorption of vitamin B12 may occur in persons with gastrointestinal symptoms and in more advanced stages of the disease (Allen, 2008:S27).

2.3.2.4 Clinical manifestations

The blood, gastrointestinal tract and central as well as peripheral nervous system are affected by a vitamin B12 deficiency (Stopler & Weiner, 2012:2384; Elghetany & Banki, 2011:562). Peripheral neuropathy together with numbness and a tingling sensation in the hands and feet is commonly seen in vitamin B12 deficient individuals. Other common symptoms develop as a result of inadequate myelinisation of nerves include memory loss, changes in personality, occasional psychosis, reduction in the sense of vibration and position, poor muscle coordination and hallucinations (Stopler & Weiner, 2012:737; Koppel, 2011:2394; Elghetany & Banki, 2011:562). Damage to the nervous system may be irreversible with prolonged vitamin B12 deficiency even with vitamin B12 treatment (Stopler & Weiner, 2012:737; Antony, 2008:520).

Physical examinations of patients with vitamin B12 deficiency may show pallor, oedema, jaundice, and changes in in the pigment of the skin (Stabler, 2013:151). Skin pallor in combination with jaundice may give patients a lemon-yellow appearance to the skin. Patients may also suffer from a sore, smooth and pale tongue (atrophic glossitis) or a red

and raw tongue (acute glossitis). Gastrointestinal symptoms may also be experienced and include episodic abdominal pain, constipation and diarrhoea (Elghetany & Banki, 2011:562).

Vitamin B12 deficiency is an important modifiable risk factor for osteoporosis and adults have a greater risk for developing osteoporosis as the average bone mineral density is lower in adults (Stopler & Weiner, 2012:737; Tucker et al., 2005:152). Swart, van Schoor and Lips (2013:217), found that epidemiological cohort studies indicate strong associations between low vitamin B12 and high serum homocysteine levels and the incidence of fractures, however, the relationship with low bone mineral density is unclear.

An association between low levels of vitamin B12 and increased levels of homocysteine exist, but a relationship to vascular disease or vascular-related dementia has not yet been established (Koppel, 2011:2384). However, reactions between vitamin B12, folate and homocysteine aggravate heart disease (Stopler & Weiner, 2012:737; HOPE 2, 2006:1567).

2.3.3 Management of megaloblastic anaemia

2.3.3.1 Medical management

It is imperative to diagnose the cause of the megaloblastic anaemia correctly prior to initiation of treatment since the administration of folate will correct the anaemia from either vitamin B12 or folate deficiency. The neurologic damage of vitamin B12 deficiency may be masked, allowing nerve damage to progress to the point where it is irreversible (Stopler & Weiner, 2012:736; Elghetany & Banki, 2011:565). Even though severely anaemic patients may need to be treated with both folate and vitamin B12, it is usually possible to determine the cause of deficiency and treat it specifically (Elghetany and Banki, 2011:565).

Replenishment of stores may be achieved with an oral dosage of 1mg of folate taken daily for 2 to 3 weeks. In order to maintain stores after repletion, an absolute minimum oral folate intake of 50 to 100µg is required. Treatment should continue until complete recovery is evident (Antony, 2008:520). Increased alertness, cooperation and appetite may be observed within 24 to 48 hours of treatment, however, improvement in haematological

values is a gradual process that takes approximately eight weeks (Aslinia, Mazza & Yale, 2006:240).

Treatment of vitamin B12 deficiency anaemia usually consists of a once a week intramuscular or subcutaneous injection of 1000µg for one month, followed by a reduced frequency of administration of monthly 1000µg injections until remission can be achieved (Aslinia, Mazza & Yale, 2006:240). Pernicious anaemia is treated with a daily dose of 1000µg of vitamin B12 during the first week, twice weekly injections for the second week, followed by once a week for the following four weeks, and the once a month for the patient's lifetime (Elghetany & Banki, 2011:565).

2.3.3.2 Medical nutrition therapy

Following the correction of the anaemia, patients should be advised to consume at least one fresh, uncooked fruit or vegetable or to drink a glass of fruit juice per day (Stopler & Weiner, 2012:736). Fruits and vegetables consumed in the fresh, uncooked state are food sources of folate as the heating process and the oxidation that takes place during storage destroys folate (McKillop et al., 2002:681).

It is recommended that women consume 400µg of folate per day (IOM, 2006:244). Foods rich in folate include dark green leafy vegetables, such as spinach and broccoli; legumes, such as black beans, kidney beans and black-eyed peas; liver and some fruits, especially citrus fruits and juices. Consumption of fortified grain products also contribute to folate intake (Simpson et al., 2009:1330). It is recommended that women of childbearing age, who may fall pregnant, consume 600µg of synthetic folic acid from fortified foods and supplements in addition to folate from dietary sources (IOM, 2006:244).

The recommended daily allowance of vitamin B12 is set at 2.4µg per day (IOM, 2006:244). Meat, particularly beef and pork, eggs, milk and milk products are good sources of vitamin B12 (Stopler & Weiner, 2012:736; Scott, 1999:444).

2.4 PREVENTION STRATEGIES

The South African Department of Health (SADoH) has committed itself to prioritise activities towards virtually eliminating vitamin A, iodine and iron deficiencies (SADoH, 2007:6). Legislation requiring any person who is responsible for manufacturing, importing, or selling bread wheat flour and maize meal to fortify these products came into effect on 7 October 2003. As stipulated under the Foodstuffs, Cosmetics and Disinfectants Act (Act No. 54) of 1972, maize meal and wheat flour should be fortified with vitamin A, thiamine, riboflavin, niacin, folic acid, pyridoxine, iron and zinc (Labadarios et al., 2008:253). Fortification of everyday staple products focuses on providing significant levels of essential vitamins and minerals through an affordable diet to those who are most deprived (SADoH, 2007:7).

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CHAPTER 3

METHODOLOGY

3.1 INTRODUCTION

This study formed part of the Assuring Health for All in the Free State (AHA-FS) study. The study sample, measurements, techniques, procedures, the role of the researcher, statistical analysis and ethical considerations of this study will be discussed in this chapter.

3.2 STUDY DESIGN

The AHA-FS study is an epidemiological study undertaken in 2007 in the rural areas and in 2009 in the urban areas with the main aim of determining how living in rural and in urban areas influences the lifestyles of populations that predispose them to both chronic diseases (such as obesity, diabetes mellitus and cardiovascular disease), as well as undernutrition and HIV/AIDS. For the purpose of this study, data from the baseline rural phase was used and applied as a cross-sectional study.

3.2.1 Population

The AHA-FS study was planned to coincide with the service learning functions of the University of the Free State where two key community service delivery sites, the Mangaung University Community Partnership Programme (MUCPP) and the Free State Rural Development Partnership Programme (FSRDPP) were included. The urban baseline phase included the areas serviced by the MUCPP, whereas the rural baseline phase included the areas serviced by the FSRDPP. As part of the larger study protocol of the AHA-FS study, human immunodeficiency virus (HIV) status of the participants is known.

3.2.2 Sample

For the purpose of this study, all households in the rural towns of Trompsburg, Philippolis and Springfontein were eligible to participate. Low socio-economic areas in these towns were targeted to make up the population (excluding farms), as these communities are in the process of experiencing transitions in lifestyle that have already been experienced by the other populations. The households were estimated, with the help of the local municipalities, as follows:

Trompsburg:	3000 Black, 400 Coloured
Philippolis:	2500 Black, 800 Coloured
Springfontein:	2500 Black, 600 Coloured

The rural AHA-FS study included all adult participants, 25-64 years of age, who provided informed consent. From this sample, women between the age of 25 and 49 years who were HIV-uninfected who gave informed consent and who were not pregnant at the time of data collection were selected for the current study. A total number of 359 women were included in the rural AHA-FS data set of which 191 were between the ages of 25-49 years. A further 57 women had to be excluded from the study due to HIV infection, pregnancy or incomplete data sets for anaysis. The final sample of this study thus consisted of 134 women.

3.3 INFORMATION COLLECTED DURING THE BASELINE SURVEY

The households were visited by fieldworkers to explain the study to adult members of the households, using an information letter (Appendix A), and to invite those between the ages of 25 and 64 years of age to participate in the study. Those households who were willing to participate were issued with an information document, which included more information on the project, and an informed consent document (Appendix B) was completed. A participation letter (Appendix C) stating and explaining all the relevant arrangements regarding the study was issued to each adult participant who gave written, informed consent.

Data was obtained from the participants at the research venue, which was at the community halls in the rural areas. In order to make sure that the participants met the criteria for the age, each of their ID documents were screened. Participants were required to rotate between different research stations in the venue. The venues where data was collected included stations for the collection of blood and urine samples; a food station; medical examination; as well as an anthropometric station. Blood and urine samples were transported back to the laboratory directly after the days' samples had been drawn. The stations were indicated on each participant's checklist and each station was signed or checked off after completion. During the interviews or examinations, the following data were included:

3.3.1 Individual questionnaires

An adapted 24-hour recall (Appendix D) as well as a short food frequency questionnaire was used to determine individual dietary intake and eating habits. Each 24-hour recall was also used to determine the dietary diversity score (DDS) of each household (Appendix E).

Physical activity was calculated by using a 24-hour recall of physical activity which was used to calculate energy expended in order to categorize activity levels as sedentary, low active, active and very active.

Anthropometric measurements were taken on all participants and included height, weight, waist, hip and mid-arm circumferences as well as skinfold measurements. Anthropometric information was recorded on an anthropometry form (Appendix F).

A health questionnaire (Appendix G) that included information on social support (group membership, network of friends, family structure), tobacco and alcohol consumption patterns, medical history and medications (including contraception use), family medical history, alternative medical practices, levels of stress and behaviours related to stress control was completed for all adult participants in each household. One adult in each household also completed a questionnaire on knowledge, attitudes and practices about nutrition.

Medical examinations involved of the assessment of blood pressure, heart rate, visible signs and symptoms, cardiovascular abnormalities, respiratory abnormalities, abdominal pathology, nervous system abnormalities and skin pathology, as well as HIV pre-test counselling. All medical examinations were performed by an experienced medical practitioner and recorded on a medical examination form. A referral letter was issued to participants who needed further medical support.

A full blood count; glycated haemoglobin (HbA1c), glucose and fibrinogen levels; as well as HIV testing and cluster of differentiation (CD4) counts were taken from all adult members in each household. A spot urine sample was also collected from each participating adult.

For the purpose of the current study, information on the socio demographics, dietary intake, reported health, anthropometry and biochemical values related to anaemia were used. Dietary intake information from the 24-hour recall questionnaire was be used to determine individual dietary diversity.

3.3.2 Household questionnaires

A socio-demographic questionnaire (Appendix H) was completed for each household. This questionnaire included basic demographics of household members; structure of the house; household income; house amenities; access to water and sanitation; employment status and cooking facilities. Questions pertaining to language, race, gender, age, education, employment status and income, type of dwelling and household density were also included. Information regarding water, sanitation, source of energy and food storage facilities was obtained in terms of household information.

A household food security questionnaire was completed for each household and included questions regarding agricultural practices, crop and livestock ownership as well as a hunger scale.

3.4 MEASUREMENTS

3.4.1 Variables and operational definitions

Certain variables measured on the adult participants from the rural areas in the AHA-FS study were used for the purpose of this study. Measurements included individual dietary intake (dietary diversity), anthropometry (body mass index and body fat percentage) as well as fasting blood samples.

3.4.1.1 Individual dietary intake

A 24-hour recall was used to obtain individual dietary intake. The 24-hour recall method of data collection requires the participant to remember the specific foods and amounts of foods consumed within the previous 24 hours (Hammond, 2012:140). Nutrient adequacy was evaluated according to the DDS. The DDS is determined by summarising the number of food groups obtained from the 24-hour recall into 9 standardised food groups as developed in the standardised tool for individual dietary diversity (DD) of the Food and Agriculture Organization (FAO) (FAO, 2011:8). DD was scored as indicated in Table 3.1.

Table 3.1 Dietary diversity scores (FAO, 2011:8)

Number of food groups consumed	**Score**
≤ 3	Low
4-5	Medium
≥ 6	High

3.4.1.2 Anthropometry

For the purpose of this study, anthropometric measurements included height, weight, triceps, biceps, subscapular and supra-ileac skinfolds as well as waist circumference. These measurements were used to calculate body mass index (BMI) and fat percentage.

a) Body mass index

The BMI of the participants was calculated to determine whether the participant was underweight, normal weight, overweight or obese. BMI is thus used to measure undernutrition or overnutrition (Lee & Nieman, 2013:14). BMI was used to interpret the anthropometric measurements of the participants and was classified as suggested by the World Health Organization (WHO) (Table 3.2).

Table 3.2 International classification of adult underweight, overweight and obesity according to BMI (WHO, 2013:Online)

BMI (kg/m^2)	**Classification**
< 18.50	Underweight
18.50 – 24.99	Normal weight
25.00 – 29.99	Overweight
30.00 – 34.99	Obesity class I
35.00 – 39.99	Obesity class II
≥ 40.00	Obesity class III

b) Waist circumference

The presence of excess body fat around the abdominal area that is out of proportion to the total body fat is a risk factor for chronic diseases associated with obesity and metabolic syndrome (Hammond & Litchford, 2012:169). Waist circumference measurement is an approach for assessing body fat distribution (Lee & Nieman, 2013:187). Waist circumference was classified as normal (below 80 centimetres), at risk (greater and equal to 80 centimetres), or high risk (greater and equal to 88 centimetres) (Lee & Nieman, 2013:187; WHO, 2008a:15).

c) Body fat percentage

Body fat percentage was calculated using the sum of four skinfolds, namely, triceps, biceps, subscapular and suprailiac skinfolds. Body fat percentage was determined by adding the four skinfolds together and reading the answer off the "Percentage of body fat based on four skinfold measurements" chart which gives the actual fat percentage (Durnin & Woemersley, 1974:77-97). Body fat ranges for persons older than 18 years of age used to interpret the body fat percentage of the participants are indicated in Table 3.3 (Lee & Nieman, 2013:199).

Table 3.3 Body fat ranges for persons 18 years of age and older (Lee & Nieman, 2013:199)

Classification	**Males**	**Females**
Unhealthy range (too low)	≤ 5%	≤ 8%
Acceptable range (lower end)	6 – 15 %	9 – 23%
Acceptable range (upper end)	16 – 24%	24 – 31%
Unhealthy (too high)	≥ 25%	≥ 32%

3.4.1.3 Fasting blood samples

For the purpose of this study, fasting blood samples were used to measure full blood count, serum ferritin and transferrin levels, homocysteine and red cell folic acid levels in all participants. The National Health Laboratory Sercive (NHLS) and Ampath reference values were used for the interpretation of each biochemical parameter. The reference values of the WHO were used for the interpretation of iron deficiency and iron deficiency anaemia.

a) Full blood count, serum ferritin and transferrin

Haematocrit (Hct) and haemoglobin (Hb) form part of a routine full blood count and are used in conjunction with each other to evaluate iron status. Hct measures the percentage of red blood cells in the total blood volume. The Hb concentration measures the total amount

of Hb in the peripheral blood. Hb and Hct are both below normal in iron deficiency anaemia and megaloblastic anaemia (Litchford, 2012:200). Hb levels below 12.1g/dL and Hct levels below 0.371L/L are considered low (NHLS Reference values). Cut-off values differ between sea level and the Free State. At sea level, the reference range for Hb is 12.0-15.0g/dL and the reference range for Hct is 36.0-46.0g/dL in females (Ampath, 2010:87).

Mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH) and mean corpuscular haemoglobin concentration (MCHC) also forms part of a routine full blood count and can also be used to further classify the anaemia (Litchford, 2012:195). The reference range for MCV is 79.1 to 98.9fl. A raised MCV is indicative of macrocytic cells and is associated with megaloblastic anaemia (folate or vitamin B12 deficiency). A lowered MCV indicates microcytic red cells and is associated with iron deficiency anaemia (NHLS Reference values; Ampath, 2010:88).

The reference range for MCH is 27.0 to 32.0pg per cell and values below the reference are indicative of hypochromic (pale) red cells as seen with iron deficiency anaemia. MCH values above the reference range indicate hyperchromic red cells as seen with a megaloblastic anaemia. The reference value for MCHC is 32.0 to 36.0g/dL. A raised MCHC is associated with megaloblastic anaemia where a decreased value of MCHC is associated with iron deficiency anaemia (NHLS Reference values; Ampath, 2010:88).

Ferritin is the storage protein that isolates the iron normally collected in the liver, spleen and marrow. Serum ferritin is the best indicator of iron stores in the human body as it reflects tissue stores in a direct manner (Litchford, 2012:200). Normal concentrations of ferritin are between 6 to 120ng/ml (NHLS Reference values). Transferrin saturation is a direct measure of available proteins responsible for the binding of mobile iron. Transferrin saturation is dependent on the number of free binding sites on the plasma iron-transport protein transferrin (Litchford, 2012:200). Transferrin is normally between 15% and 50% saturated with iron and saturation below 15% indicates the possible presence of iron deficiency (Ampath, 2010:50). Iron deficiency anaemia develops in three stages. Firstly, depletion of iron stores occurs and is confirmed by serum ferritin levels below the reference value. Deficient erythropoiesis with normal haemoglobin but increased red cell

protoporphyrin follows as stage two. During this stage, the iron concentration of serum ferritin and serum iron falls, the synthesis of transferrin is increased and the percentage of transferrin saturation becomes decreased. During the third and final stage, iron deficiency anaemia in which both iron and haemoglobin are low, a microcytic, hypochromic anaemia is visible on blood morphology (Gaw et al., 2008:112).

According to the WHO (2008b:4), iron deficiency is diagnosed when ferritin levels are below 15.0ng/mL and haemoglobin levels are still greater than 12.0g/dL; and iron deficiency anaemia is diagnosed when both ferritin and haemoglobin levels are below 15.0ng/mL and 12.0g/dL respectively. These cut-off values will also be used to define iron deficiency and iron deficiency anaemia.

b) Homocysteine and red cell folate levels

Folate and vitamin B12 are required for the synthesis of methionine-S-adenosylmethionine, the biochemical precursor involved in the production of homocysteine. In return, homocysteine is involved in the production of the amino acids cysteine and methionine. The conversion of homocysteine to methionine is dependent on folate and vitamin B12 levels as indicated in figure 3.1. Homocysteine levels rise and spill into the circulation when either folate or vitamin B12 is lacking. Thus, elevated levels of homocysteine may be an indication of a folate or vitamin B12 deficiency (Litchford, 2012:201). The reference range for homocysteine is 2.10 to 15.70 µmol/L (NHLS Reference values).

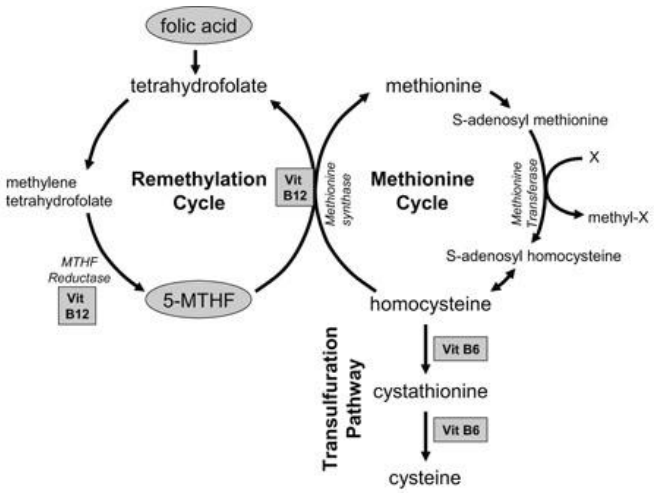

Figure 3.1: Homocysteine metabolic pathways (Cianciolo et al., 2008: 942)

Red blood cell folate concentration is a better indicator of folate status than serum folate, as folate is much more concentrated in red blood cells than in serum and red blood cell folate offers a closer indication of tissue stores. Red blood cell folate is thus considered the most reliable indicator of folate status (Litchford, 2012:201). A red blood cell folate concentration below 372nmol/L is indicative of a folate deficiency (NHLS Reference values).

3.5 TECHNIQUES

Techniques used in this study to determine socio demographics and dietary diversity; and to measure anthropometry and fasting blood samples are described in the following section.

3.5.1 Questionnaires

Questionnaires were used to obtain information from household members in a structured interview conducted by final year students from the Department of Nutrition and Dietetics. Separate stations were assigned for the completion of each questionnaire. Participants rotated between the different stations in order to complete all the relevant questionnaires. A trained Sotho-speaking interviewer was used in cases where participants were unable to understand English or Afrikaans.

Questionnaires used for the purpose of this study include the following:

a) Individual dietary intake information and dietary diversity score – A 24-hour recall of usual intake was completed during individual interviews with all adults that participated in the study (Appendix D) and was used to determine DDS (Appendix E). In order to determine dietary diversity, each 24-hour recall was summarized in the nine different food groups defined by the FAO (FAO, 2011:8) for the calculation of individual dietary diversity namely starchy staples; dark green leafy vegetables; other vitamin A rich fruits and vegetables; other fruits and vegetables; organ meat; meat and fish; eggs; legumes, nuts and seeds; and milk and milk products.

b) Anthropometry – Measurements were taken on each participant by a final year dietetics student and recorded immediately after the measurement was taken (Appendix F).

c) Reported health information was obtained from each individual via structured interviews conducted by final year dietetics students (Appendix G).

d) Socio demographic and household information obtained from one member of the household by structured interview by final year dietetics students (Appendix H).

3.5.2 Anthropometry

Trained fourth year BSc. Dietetics students under supervision of lecturers from the Department of Nutrition and Dietetics at the University of the Free State were responsible for taking the anthropometric measurements.

3.5.2.1 Body weight

Body weight is one of the most important measurements in nutritional assessment (Lee & Nieman, 2013:170). Weight can be used to provide a rough estimate of overall fat and muscle stores. Weight relevant to height can be interpreted by calculating the BMI (Hammond & Litchford, 2012:166).

Weight was measured by using a digital electronic foot scale and recorded to the nearest 0.1kg. Scales were placed on a flat, hard surface so as to ensure the scale is secure and does not rock or tip. Participants were advised to wear minimal clothing and no shoes, after an overnight fast. Participants were instructed to stand still and straight in the middle of the scale, without any support, with the body weight distributed evenly between both feet. The scale was checked and adjusted to zero prior to each measurement (Lee & Nieman, 2013:170; Gibson, 2005:253).

3.5.2.2 Height

Height measurements may be valuable when used in combination with other measurements (Hammond & Litchford, 2012:165).

A stadiometer was used to measure height. Participants were advised to wear minimal clothing when measured in order to see their posture clearly. Shoes, socks and hats were also removed before measuring. Subjects were required to stand with their heels together, arms to their sides, legs straight, shoulders relaxed, and their head facing straight ahead in the Frankfurt horizontal plane (the top of the external ear canal and the top of the lower bone of the eye socket should be in a horizontal plane parallel to the floor). The measurer made sure that the participant's heels, buttocks, scapulae, and back of the head was against the vertical surface of the stadiometer if possible. The participant was advised to inhale deeply, to hold their breath, and to maintain an erect posture while the headboard is lowered on the highest point of the head with sufficient pressure to compress the hair. The measurement was read to the closest 0.1cm at eye level with the headboard in order to prevent any errors caused by parallax (Lee & Nieman, 2013:168; Gibson, 2005:246).

3.5.2.3 Waist circumference

A non-stretchable tape measure was used to obtain the measurement. In measuring waist circumference, two accepted procedures exist, i.e. measurement at the natural waist or measurement at the umbilicus level. For the purpose of this study, waist circumference was measured at the level of the natural waist. Participants were instructed to wear minimal clothing in order to allow the tape to be positioned correctly. The measurer ensured that the participant was standing upright, facing straight forward, with the abdomen relaxed, arms at the sides, feet together, and weight distributed evenly between the two feet (Gibson, 2005:281). The measurer stood facing the participant and placed the non-stretchable tape around the participant, in a horizontal plane at the narrowest part of the torso (Firsancho, 2011:12). The tape measure was wrapped firmly, but not too tight around the marked area. The participant was asked to breathe normally and to breathe out gently at the time of the measurement to prevent the contraction of muscle or the participant

from holding their breath when the measurement was taken. The measurement was taken to the nearest 1cm (Gibson, 2005:281).

3.5.2.4 Skinfold measurements

Skinfold measurements were obtained by using precision thickness callipers. In order to determine the percentage body fat of the participants, triceps, biceps, subscapular and suprailiac skinfold measurements were obtained.

a) Triceps skinfold

The triceps is the most commonly measured skinfold site (Lee & Nieman, 2013:190). The participant's arm was bare with the top of the shoulder accessible to the measurer. The triceps skinfold was measured at the midpoint of the upper right arm, between the acromion process at the shoulder and the tip of the olecranon at the elbow. While the participant's arm was bent at a 90 degree angle at the elbow, the midpoint was determined and marked by using a non-stretchable tape and a soft pen or indelible pencil. Following the marking of the midpoint, the right arm was extended to hang loosely at the side with the palm of the hand facing anteriorly in order to properly determine the posterior midline. The skinfold site was marked along the posterior midline of the upper arm at the same level as the midpoint marked previously. In order to take the skinfold measurements, the measurer stood behind the subject and then grasped the skinfold with the thumb and index finger of the left hand approximately 1cm proximal to the skinfold site after which the measurer inserted the calliper approximately 1cm from the thumb and fore finger. The measurer ensured that the calliper jaws were placed at right angles, exactly at the marked point and that the skinfold remained held between the fingers while the measurement was taken (Lee & Nieman, 2013:192; Gibson, 2005:276).

b) Biceps skinfold

Biceps skinfold was measured as the thickness of a vertical fold on the front of the upper arm. The measurements were taken directly above the center of the cubital fossa, i.e. the

midpoint between the acromion process at the shoulder and the olecranon process at the elbow on the front of the upper arm, at the same level as the triceps skinfold (Hammond & Litchford, 2012:168; Gibson, 2005:275).

c) Subscapular skinfold

The subscapular skinfold was measured below and lateral to the angle of the shoulder blade, with the participant's shoulder and arm relaxed. The participant's arm was bare with their back accessible to the measurer. The measurer could place the participant's arm behind their back to assist with the identification and marking of the site. The long-axis of the skinfold is on a 45 degree angle directed down and to the side, in the same direction as the inner border of the scapula. The skinfold was grasped 1cm above the site (Lee & Nieman, 2013:192; Gibson, 2005:274).

d) Suprailiac skinfold

The suprailiac skinfold was measured in the mid-axillary line, just above the iliac crest. The measurer ensured the participant stood upright, faced straight ahead, with feet together and arms to the sides. The measurer grasped the skinfold approximately 1cm posterior to the midaxillary line and parallel to the cleavage lines of the skin (Lee & Nieman, 2013:193; Gibson, 2005:275).

3.5.3 Fasting blood samples

Blood samples were collected by chemical pathologists and analysed according to standard techniques. Respondents were required to fast overnight with blood samples being collected the next morning. Requirements for the purpose of this study included 60ml of blood from adults collected in purple plug EDTA-containing tubes.

The Sysmex XT-2000*i* Automated Hematology Analyzer was used to measure the full blood count. This system uses fluorescent flow cytometry and hydrodynamic focusing technologies in order to provide a sensitive measure of cell types in whole blood (Sysmex,

2014:Online). Iron levels were determined by using the Beckman Coulter Synchron LX20 which is a microprocessor-controlled, random-access clinical analyser that is capable of processing a wide variety of operator-selected chemistries in a single run (Mikolaenko et al., 2000:387). Transferrin and homocysteine was determined by using the BN Prospec System nephelometric technology. Serum folate levels were determined by using the Bayer Advia Centaur System (Hadler et al., 2008: S262).

3.6 VALIDITY AND RELIABILITY

Validity can be defined as the extent to which a measuring instrument measures what it is intended to measure. Reliability is defined as the consistency with which a measuring instrument delivers a certain, consistent result when the unit being measured has not changed (Leedy & Ormrod, 2013:91).

3.6.1 Questionnaires

Validity

All issues addressed by the questionnaires pertained directly to the aim and objectives of the study. The 24 hour recall questionnaire is a quick list recall by the respondent of the previous 24 hour's intake with a detailed interview elaborating the list followed by a thorough review of the detailed interview, a method that may ensure validity (Bates et al., 2005:581). The information obtained in the 24-hour recall was used to determine dietary diversity, for which a standardised tool was developed by the FAO (FAO, 2011:8).

Reliability

Dietary assessment methods are considered reliable where the method delivers similar results with repeated use in the same circumstances. Replication of observations in dietary assessment is not possible; it is thus difficult to establish reproducibility (Gibson, 2005:129). Reliability of the socio-demographic questionnaire was assessed by interviewing 10% of the study population again. In the case where answers deviated with more than 20%, the

question was considered unreliable and consequently the results of those questions were not reported.

3.6.2 Anthropometry

Validity

In ensuring validity, it is important to ensure the tools used for data collection are calibrated and that measurements are taken using standardised, established techniques as described in the literature (Lee & Nieman, 2013; Frisancho, 2011; Gibson, 2005). In order to further ensure validity, each measurement was taken three times to ensure the accuracy of the average value. The scale was calibrated after every 20th participant measured.

Reliability

In order to ensure reliability of the anthropometric measures, the researcher used consistent, standardised techniques as described in the literature (Lee & Nieman, 2013:166-220). Researchers responsible for taking the anthropometric measurements received training in anthropometric measurements prior to data collection.

3.6.3 Fasting blood samples

Validity

To ensure validity, standardised measuring techniques were used and measured by experienced medical technologists. Blood samples were collected early in the morning after overnight fasting. Samples were immediately stored in ice filled containers and transported to the laboratory. Analysis of the samples was done by using standardised laboratory techniques.

Reliability

A well-planned environment using laboratory controls in which to perform the procedure improves the efficacy and accuracy and thus the reliability of the blood samples. Blood samples were collected by medical technologists according to standardised methods.

3.7 PROCEDURES FOR THE CURRENT STUDY

The procedures followed during the larger AHA study are described in section 3.7. Figure 3.2 outlines the procedures that were followed for the current study after the data collection period:

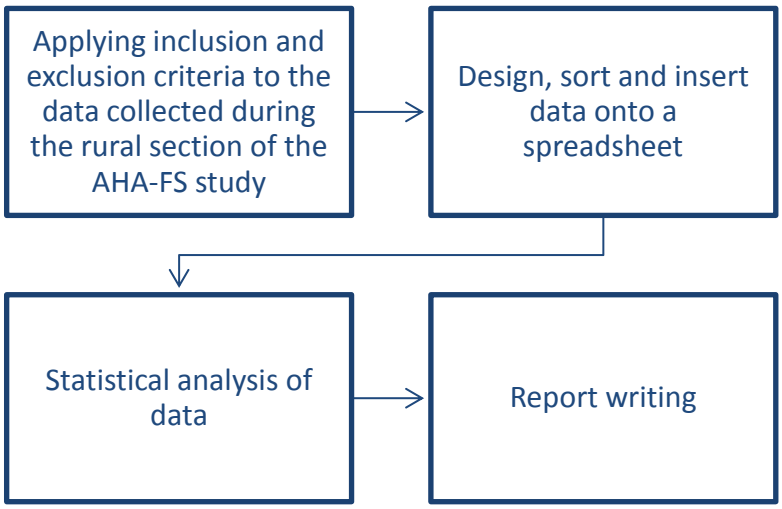

Figure 3.2: Procedures for the current study

3.8 THE ROLE OF THE RESEARCHER

The researcher was involved in rendering nutrition counselling/ education services after the data collection period. For the current study, the researcher compiled a new data collection sheet/ format for gathering data for this study from the existing data and performed all the relevant statistical analyses and interpretation of the data.

3.9 STATISTICAL ANALYSIS

The researcher performed the analysis by using the Predictive Analytics SoftWare (PASW) Statistics Student version 22.0 software by Statistical Package for the Social Sciences (SPSS): *An IBM Company* (IBM, 2013). Descriptive statistics, namely frequencies and percentages for categorical data and means and standard deviations or medians and percentiles for continuous data were determined. Due to the non-parametric nature of the data in the current study, the Kruskal-Wallis test was used to determine associations between continuous variables and categorical variables and the Chi-square test was used to determine associations between categorical variables.

3.10 ETHICAL CONSIDERATIONS

This study forms part of a larger study for which approval has been obtained from the Ethics Committee of the Faculty of Health Sciences at the University of the Free State (Etovs number 21/07).

An informed consent form (Appendix B) along with an information document (Appendix A) was provided to all participants in the language of their choice.

Strict confidentially of the information has been maintained by ensuring that no names are made known, or written in questionnaires. In order to further ensure confidentiality, codes have been used in data analysis and results.

Participation was voluntary and participants were free to withdraw from the study at any time. Participants received a light snack after the fasting blood samples were drawn. The results of the blood analyses were provided to the participants. Participants were also referred to relevant local or provincial medical services if necessary.

3.11 SUMMARY

A sample of 134 female participants between the ages of 25 and 49 years of age provided informed consent and could be included in this study. A wide variety of data was collected at three central locations as part of the rural section of the AHA-FS study. For the purpose of this study, dietary diversity, anthropometry and fasting blood samples were determined using valid and reliable techniques and measurements.

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CHAPTER 4

ANAEMIA PREVALENCE AND DIETARY DIVERSITY AMONG WOMEN IN THE RURAL FREE STATE, SOUTH AFRICA

ABSTRACT

Background: Anaemia is a global public health problem, particularly in women, and holds major consequences for human health. Optimal nutrition is important to prevent anaemia and to ensure optimal health of women, especially of childbearing age.

Objectives: Determining the dietary diversity, prevalence of anaemia and contraception use in rural women between 25–49 years in the Free State province, South Africa.

Method: A cross-sectional descriptive study design was applied in a sample of 134 women. A 24-hour recall was completed in a structured interview to determine dietary diversity, which was categorised as low (≤3 food groups), medium (4–5 food groups) and high (≥6 food groups). Blood samples were collected and analysed according to standard techniques. Full blood counts, transferrin saturation, ferritin, homocysteine and red cell folate levels were determined. Questions regarding contraceptive use were included in a questionnaire on reported health.

Results: The median age of the women in the sample was 41 years. Half (51.5%) of the women in the sample had medium dietary diversity and 44.7% had low dietary diversity. Overall, 76.9% of the women consumed flesh meats and fish, which are good sources of the bioavailable haem iron. Only a quarter (25.4%) of the women ate dark green leafy vegetables which are sources of non-haem iron, as well as folate. All the women consumed starchy foods, some of which are sources of folate and iron due to mandatory fortification. The prevalence of anaemia in the sample was 4.6% (6/134). Iron deficiency anaemia (haemoglobin < 12.0g/dL; ferritin < 15.0ng/mL) was present in only 0.7% (1/134) of the sample and 1.5% (3/134) had an iron deficiency (haemoglobin > 12.0g/dL; ferritin < 15.0ng/mL). Overall, 7.5% of the sample presented with elevated homocysteine levels, however, only 3.8% presented with low levels of red cell folate indicative of folate deficiency. Only 54.1% of the women reported that they regularly menstruate and 71.6%

currently used, or had previously used, injectable contraceptives. Significant associations between median MCV and MCH levels and dietary diversity score possibly indicates that the mandatory food fortification programme has a positive impact on the nutritional status of these women.

Conclusion: A diet with moderate variety was consumed. The prevalence of iron deficiency, iron deficiency anaemia and folate deficiency was low. The low prevalence of these conditions could be related to the older median age of the sample and the fact that approximately half of the women did not menstruate regularly. Attention should, however, still be given to the women's diets, as foods rich in haemopoietic nutrients were not consumed by all.

Keywords: Anaemia; dietary diversity; folate; iron; iron deficiency

4.1 INTRODUCTION

The global public health problem of anaemia affects women living in both developed and developing countries and holds major consequences for human health as well as economic development (WHO, 2008:Online). According to findings of the South African National Health and Nutrition Examination Survey (SANHANES-1) published in 2013, the prevalence of anaemia among women of children-bearing age (15–54 years) was in the range 23.1%–24.7% (Shisana et al., 2013:163). Research directed at nutritional anaemias largely focuses on iron-deficiency anaemia, which is associated with approximately 111 000 deaths among pregnant women globally each year (SCN, 2004:Online). In contrast, however, little global data exist on the contribution of folate deficiency towards the development of anaemia (Balarajan et al., 2011:2128).

Anaemia can be defined as a significant reduction in the mass of circulating red blood cells, resulting in a diminished oxygen binding capacity of blood (Bunn, 2011:1031). Nutritional anaemias result from the insufficient bioavailability of haemopoietic nutrients (iron, vitamin B12 and folic acid) (Biesalski & Erhardt, 2007:38). A study conducted by Abrahams, Mchiza and Steyn (2011:811) concluded that a large proportion of the South African population is

currently in a stage of nutrition transition where changes in dietary patterns are affecting health outcomes. This transition could also impact on anaemia.

"Improve maternal health" is the fifth Millennium Development Goal to be achieved by the year 2015. In order to obtain this goal and thus ensure a woman's safe passage into motherhood, quality reproductive health services, accompanied by a series of well-timed interventions, should be implemented (UN, 2010:30). It is a given that women need to consume a diet sufficient in all the required nutrients before and during pregnancy in order to enhance fertility, to support the development of pregnancy and the growing foetus, and to promote long-term health (Derbyshire, 2011:26), as the effect of poor nutritional status may follow both the mother and the infant for decades (Gallagher, 2012:349). Individual dietary diversity can be used to determine the nutrient adequacy of women's diets (FAO, 2011:5).

Contraception could possibly play a role in lowering the risk for iron deficiency anaemia as it aids in reducing the number of pregnancies and the time interval between consecutive pregnancies (UNDP/UNFPA/WHO/WB, 1998:261). The effects of contraceptive use on menstrual blood loss may also potentially influence iron status by reducing the amount of blood lost during this time (Yeasmin et al., 2010:25; UNDP/UNFPA/WHO/WB, 1998:262).

According to the World Health Organization (WHO) (2008:Online), estimates of the prevalence of anaemia are only useful when associated with the prevailing causal factors in specified settings. It is essential to collect accurate information concerning these factors, as they are multiple and complex and can serve as the basis for the development of appropriate interventions (WHO, 2008:Online).

Various studies related to anaemia have been conducted in South Africa; however, limited data are available regarding the prevalence of anaemia among Southern African women in general, and in the Free State province in particular. This study aimed to determine the prevalence of anaemia, the dietary factors that may contribute to the development of nutritional anaemias, as well as contraceptive use in women aged 25–49 years in three rural towns in the Free State.

4.2 RESEARCH METHOD AND DESIGN

4.2.1 Research approach

The work reported here formed part of the 'Assuring Health for All in the Free State' (AHA-FS) research programme, which is an epidemiological study aimed at determining how living in rural and urban areas affects lifestyle and indicators of health. The Free State Rural Development Partnership Programme (FSRDPP) includes the Trompsburg, Philippolis and Springfontein municipalities, where students from the department of Nutrition and Dietetics at the University of the Free State do their rural internship. These three towns formed part of the rural phase of the AHA-FS study. A multidisciplinary research team investigated the socio-demographic status, household food security, dietary intake, levels of physical activity, as well as the knowledge, attitudes and practices related to nutrition, and reported health status of the study population by using standardised questionnaires. In addition to a medical examination, anthropometric measurements and blood specimens were also obtained for various investigations. For the purpose of this study, data from the rural phase of the original study were used and applied in a cross-sectional investigation.

Haematological abnormalities of both HIV-infected and HIV-uninfected persons in the AHA study population have been published by Groenewald et al. (2011) and van Zyl et al. (2010), but data on the risk factors associated with the development of anaemia within these communities have not previously been investigated. The aim of this particular component of the study was thus to evaluate dietary diversity and anaemia in the women between 25–49 years in these areas and to determine associations between biochemical indices and dietary diversity.

4.2.2 Population and sampling

All households in the African and coloured settlements (excluding farms) in the rural towns of Trompsburg, Philippolis and Springfontein were eligible to participate in the original study. An increase in the level of poverty in low socio-economic townships in rural areas, where health care coverage is an additional challenge, is commonly observed in these areas.

Low socio-economic communities in these towns are in the process of undergoing transitions in lifestyle, particularly changes in their nutrition and physical activity, which have already been experienced by the populations living in more affluent areas in these towns.

From the database of the original study, women aged 25–49 years who were HIV-uninfected, who gave informed consent and who were not pregnant at the time of data collection, were selected for the current study, resulting in a final sample of 134 women.

Confidentiality of the data was ensured by allocating a number to each respondent. Ethical approval was obtained from the Ethics Committee of the Faculty of Health Sciences of the University of the Free State (ETOVS number 21/07), the Department of Health and local municipalities. Participation was voluntary and could be terminated at any time.

4.2.3 Measuring instruments and procedures

Information was obtained via structured interviews conducted by fourth-year students from the Department of Nutrition and Dietetics at the University of the Free State. A 24-hour recall was used to determine individual dietary diversity. The dietary diversity score was determined by using the tool developed by the Food and Agriculture Organization (FAO). This tool summarizes the number of food groups obtained from the 24-hour recall into 9 standardised food groups, namely starchy staples; dark green leafy vegetables; other vitamin A-rich fruit and vegetables; other fruits and vegetables; organ meat; flesh meat and fish; eggs; legumes; nuts and seeds and milk and milk products (FAO, 2011:8). According to this tool, a low dietary diversity score was allocated if three food groups or less were consumed, a medium score where four to five food groups were consumed, and a high score where six or more food groups were consumed.

Respondents were required to fast overnight with blood samples being collected by chemical pathologists the next morning. These samples were stored immediately in ice-filled containers and transported to the laboratory. The blood samples were analysed according to standard techniques at the National Health Laboratory Service Laboratory at

the University of the Free State. Full blood counts were determined using the Roche Sysmex XT 2000i® analyser (Sysmex, 2014:Online), serum ferritin using the Beckman Coulter Synchron LX20 (Mikolaenko et al., 2000:387), and transferrin and homocysteine levels using the BN Prospec System nephelometric technology. Serum folate levels were recorded using the Bayer Advia Centaur System (Hadler et al., 2008: S262).

Anaemia was diagnosed when haemoglobin levels were below 12.1g/dL (NHLS reference values). According to the WHO (2008:Online), iron deficiency is diagnosed when ferritin levels are below 15.0 ng/mL and haemoglobin levels are still greater than 12.0 g/dL; and iron deficiency anaemia is diagnosed when both ferritin and haemoglobin levels are below 15.0ng/mL and 12.0 g/dL, respectively. These cut-off values were also used in the current study to define iron deficiency and iron deficiency anaemia.

Questions relating to contraceptive use were included in the health questionnaire.

4.2.4 Statistical analysis

The researcher conducted statistical analysis of the data, using the Predictive Analytics SoftWare (PASW) Statistics Student version 22.0 by Statistical Package for the Social Sciences (SPSS) (IBM, 2013). Frequencies and percentages for categorical data and medians and percentiles for continuous data were recorded. Only six out of the 134 women in the sample suffered from anaemia (haemoglobin <12.1g/dL) which made determining associations, especially associations where those women suffering from anaemia were compared to those who did not, difficult. It was thus decided to determine associations by using the Kruskal-Wallis Test to compare medians within the entire population across different categorical variables.

4.3 RESULTS

Of the complete data set available as part of the “Assuring Health for All in the Free State” study, 38.5% related to women between 25–49 years of age. After applying the exclusion

criteria, 23.3% of the original data set could be included in the current study, resulting in a sample size of 134 women, the median age of whom was 41 ± 6.7 years.

Table 4.1 ranks the relative intake of the nine different food groups, and shows that the women's diet was predominantly based on starchy staples. Flesh meat and fish, which are rich sources of iron, were consumed by more than three quarters of the women, whereas sources of folate (dark green leafy vegetables and organ meats) were consumed by only approximately a quarter of the sample.

Table 4.1 Consumption per food group (*n* = 134)

Food group	Percentage of women consuming the food item (%)
Starchy staples	100
Dark green leafy vegetables	25.4
Other vitamin A-rich fruit and vegetables	11.2
Other fruits and vegetables	58.2
Organ meat	1.5
Flesh meat and fish	76.9
Eggs	10.4
Legumes, nuts and seeds	8.2
Milk and milk products	81.3

Overall, 44.7% of the sample had a low dietary diversity score, while almost half (51.5%) had a medium dietary diversity score. Only a relatively small percentage (3.7%, 5/134) of the sample had a high dietary diversity score (Table 4.2).

Table 4.2 Dietary diversity scores among the women (*n*=134)

Dietary diversity score	Percentage of the women (%)
Low (≤3 food groups consumed per day)	44.7
Medium (4–5 food groups consumed per day)	51.5
High (≥6 food groups consumed per day)	3.7

Table 4.3 compares the percentage of the sample that presented with low dietary diversity scores (≤3 food groups consumed per day) with results from other studies related to dietary diversity undertaken in South Africa.

Table 4.3 Low dietary diversity scores compared with other South African studies (%)

Study	% of sample
Current study	
Women aged 25–49 years	44.7
SANHANES-1 (Shisana et al., 2013)	
Men and women in the Free State (>15 years)	45.1
Oldewage-Theron & Kruger (2011)	
Women in a peri-urban informal settlement in South Africa	80.9
Labadarios, Steyn & Nel (2011)	
Adult men and women in the Free State	26.6

Table 4.4 provides a description of the study population in terms of haemoglobin, haematocrit, MCV, MCH, transferrin saturation, ferritin, homocysteine and red cell folate levels. The medians for all the blood values were within normal limits.

Table 4.4 Description of the study population in terms of haemoglobin, haematocrit, MCV, MCH, transferrin saturation, ferritin, homocysteine and red cell folate levels

Blood samples	Normal reference value/ range	N	Median	Low (%)	Normal (%)	High (%)
Haemoglobin	> 12.1 g/dL	130	13.8	4.6	95.4	-
Haematocrit	> 0.371 L/L	131	0.429	3.1	96.9	-
MCV	79.1–98.9 fl	131	93.8	3.1	77.1	19.8
MCH	27.0–32.0 pg/cell	131	30.4	7.6	67.2	25.2
Transferrin saturation	> 15%	74	26.1	12.2	87.8	-
Ferritin	6–120 ng/mL	74	94.0	1.4	58.1	40.5
Homocysteine	2.10–15.70 µmol/L	134	9.6	-	92.5	7.5
Red cell folate	> 372 nmol/L	131	575.3	3.8	96.2	-

Ampath. 2010. Ampath desk reference: guide to laboratory tests. Centurion: Ampath.

National Health Laboratory Services Reference values. 2014.

Low haemoglobin levels were observed in 4.6% (6/134) of the sample with only a small percentage presenting with low haematocrit (3.1%, 4/134), mean corpuscular volume (MCV) (3.1%, 4/134) and mean corpuscular haemoglobin (MCH) (7.6%, 10/134) levels. Elevated MCH levels were present in 25.2% (33/134) of the study population.

Transferrin saturation levels below normal were found in 12.2% (9/134) of the sample, with 1.4% (1/134) of the women displaying low ferritin levels. Table 4.5 compares the prevalence of anaemia, iron deficiency and iron deficiency anaemia in the current study with other related investigations. Even though only 4.6% of the sample suffered from anaemia, those with low MCH levels, but normal haemoglobin levels (5.2%, 7/134), were at risk for developing anaemia.

Both the medians for homocysteine and red cell folate were also within normal limits. Elevated homocysteine levels occurred in 7.5% (10/134) of the sample and 3.8% (5/134) had low red cell folate levels.

TABLE 4.5 Prevalence of anaemia and iron deficiency anaemia in comparison to other studies

	Anaemia (%)	Iron deficiency (%)	Iron deficiency anaemia (%)
Current study:			
Women aged 25–49 years	4.6	1.5	0.7
SANHANES (Shisana et al., 2013):			
Women	24.7 (25–34 years) 23.1 (35–44 years)	5.9 (16–35 years)	9.7 (16–35 years)

When assessing the prevalence of iron deficiency, it is important to consider menstruation patterns and contraceptive use (Table 4.6). Just over half (54.1%) of the women regularly menstruated and almost three quarters (71.6%) of them currently or previously made use of injectable contraceptives.

Table 4.6 Menstruation patterns and contraceptive use

Questions	N	Yes (%)	No (%)
Still menstruated regularly	133	54.1	45.9
Currently or previously used injectable contraceptives	134	71.6	28.4

As expected, a significant association was found between median haemoglobin and whether the women still menstruate or not (p=0.008) (Table 4.7). These median haemoglobin levels were within the normal range for both women who menstruated (13.7g/dL) and women who did not (14.2g/dL). No significant association was found between haemoglobin levels and those women who currently or previously used injectable contraceptives and those that did not (p=0.847). No significant associations were found between the dietary diversity score and any of the blood parameters (p>0.05), except for the median MCV levels (p=0.029) and median MCH levels (p=0.019). These median MCV levels were within the normal range for both the group with low dietary diversity scores (94.9fl) and those with medium to high dietary diversity scores (92.9fl). The median MCH levels were also within

the normal range for those who had a low dietary diversity score (31.0pg/cell), as well as those who had a medium to high score (30.0pg/cell). No significant associations were found between haemoglobin and the intake of flesh meats and fish food group (p>0.05) and the organ meats group (p>0.05).

Table 4.7 Associations between blood parameters and other variables

Variables	n	Median blood value	p-value for median differences
Median haemoglobin levels across categories of menstruation:			0.008*
Menstruated regularly	70	13.7g/dL	
Did not menstruate regularly	59	14.2g/dL	
Median haemoglobin levels across categories of contraceptive use:			0.847
Made use of injectable contraceptives	94	13.8g/dL	
Did not use injectable contraceptives	36	13.9g/dL	
Median haemoglobin levels across categories of dietary diversity score:			0.481
Low dietary diversity	58	13.7g/dL	
Medium to high dietary diversity	72	13.8g/dL	
Median haematocrit levels across categories of dietary diversity score:			0.319
Low dietary diversity	58	0.428L/L	
Medium to high dietary diversity	73	0.430L/L	
Median MCV levels across categories of dietary diversity score:			0.029*
Low dietary diversity	58	94.9fl	
Medium to high dietary diversity	73	92.9fl	
Median MCH levels across categories of dietary diversity score:			0.019*
Low dietary diversity	58	31.0pg/cell	
Medium to high dietary diversity	73	30.0pg/cell	
Median transferrin saturation levels across categories of dietary diversity score:			0.129
Low dietary diversity	30	29.5%	
Medium to high dietary diversity	44	24.8%	
Median ferritin levels across categories of dietary diversity:			0.750
Low dietary diversity	30	88.5ng/mL	
Medium to high dietary diversity	44	98.0ng/mL	

Median homocysteine levels across categories of dietary diversity score:			0.594
Low dietary diversity	60	9.6µmol/L	
Medium to high dietary diversity	74	9.5µmol/L	
Median red cell folate levels across categories of dietary diversity score:			0.320
Low dietary diversity	58	570.5nmol/L	
Medium to high dietary diversity	73	576.1nmol/L	
Median haemoglobin levels across categories of flesh meat and fish consumption:			0.511
Consumed flesh meat and fish	101	13.8g/dL	
Did not consume flesh meat and fish	29	13.6g/dL	
Median haemoglobin levels across categories of organ meat consumption:			0.281
Consumed organ meat	2	13.0g/dL	
Did not consume organ meat	128	13.8g/dL	

* Statistically significant association

4.4 DISCUSSION

With this study we aimed to assess the dietary diversity, presence of anaemia, and related factors (menstruation and contraceptives use), in a group of women aged 25–49 years living in low socio-economic townships in the rural Free State towns of Trompsburg, Philippolis and Springfontein. As a result of the far-reaching effects of anaemia on human health (WHO, 2008:Online), it was considered desirable to determine the need for intervention studies within this population.

The median dietary diversity score of the women in this study agrees with the findings of a study conducted by Labadarios, Steyn and Nel (2011:35), on adult men and women in South Africa. It was, however, slightly higher than that of the SANHANES-1 study, where the median score was low. Even though the nutrient adequacy in terms of the median dietary diversity score of these women was medium, almost half of the women had a low score indicating that their diet needs attention in terms of the nutrient adequacy. The prevalence

of low dietary diversity in this study was similar to that of the SANHANES-1 study (Table 4.3) (Shisana et al., 2013:169).

Even though the median dietary adequacy of the women fell within the range of 4–5 groups, the food groups included in their diet, in terms of iron and folate, were inadequate. Almost a quarter of the women did not consume flesh meat and fish, which are excellent sources of the more bioavailable haem iron (Nojilana et al., 2007:741). The diets of the women seemed to be largely starch-based, with low intakes of other iron-rich foods including organ meats, eggs and dark green leafy vegetables (Nojilana et al., 2007:741). Dark green leafy vegetables are excellent sources of folate and the less bioavailable form of non-haem iron (Nojilana et al., 2007:741); only a quarter of the women were observed to consume them and an even smaller proportion ate legumes, which are also a rich source of folate. The iron present in these vegetables is largely in the form of less bioavailable non-haem iron, and its absorption can be impaired by the phytates present in some grains. The low intake of iron and folate-rich sources was similar to findings in a study conducted on non-pregnant women older than 19 years living in rural areas in KwaZulu-Natal (Kolahdooz, Spearing & Sharma, 2013:2).

In this study, only a small proportion of the women had low haemoglobin levels indicative of anaemia which is much lower than those reported in the SANHANES-1 study, where almost a quarter of the women between 25–34 years and 35–44 years in South Africa and 17.6% of women between the ages of 16–35 years in the Free State, suffered from anaemia (Shisana et al., 2013:162). The prevalence of anaemia in the current study is also much lower than the results from a systematic review by Stevens et al. (2013:e19), that looked at population representative data, where 28% of non-pregnant women aged 15–49 years in 2011 in South Africa were reported to be anaemic. According to the National Food Consumption Survey – Fortification Baseline (NFCS-FB) survey conducted in 2005 among women of reproductive age (15–35 years), 23.2% of women in the Free State province suffered from anaemia (Labadarios et al., 2007:458). These studies however, did not report on menstruation and contraceptive use, which may account for some of the differences.

It was evident in our study population that almost half of the women were not menstruating, and a large proportion of the sample made use of injectable contraceptives, both of which may have decreased their risk for developing iron deficiency and thus possibly influencing the low prevalence of iron deficiency in the current sample. Women who still menstruated regularly were more likely to have lower haemoglobin levels as the median haemoglobin levels were statistically significantly lower in women who still menstruated. Results from the current study show that a large number of women currently or previously made use of injectable contraceptives. A study conducted by Arias et al. (2006:237), concluded that the reduction of iron deficiency is one of the non-contraceptive benefits of using injectable contraceptives, specifically depot medroxyprogesterone acetate. A study conducted by Yeasmin et al. (2010:28) among women aged 20–40 years from a low socio-economic background in Dhaka, Bangladesh, found that women who used oral contraceptives had significantly higher haemoglobin levels than those who did not. The SANHANES-1 study did not report on information on contraceptive use and the effects thereof on anaemia.

Iron deficiency anaemia develops in three stages. The first stage is characterised by the depletion of iron stores which is confirmed by low serum ferritin levels. During the following stage, serum ferritin decreases further with transferrin saturation decreasing as well. These decreases are followed by the development of microcytosis and hypochromia after which signs and symptoms of anaemia appear (Gaw et al., 2008:112).

Low MCV levels, indicating microcytic anaemia, and low MCH levels, indicating hypochromic anaemia, in combination with haemoglobin levels, indicate iron deficiency anaemia. A quarter (25.2%) of the women had increased levels of MCH, which is seen in macrocytic anaemia. Considering these factors in combination with ferritin levels, it is evident that a very small percentage (1.4%, 1/134) had low iron stores. Serum ferritin levels in the current study were much higher than results from the SANHANES-1 where 13.5% of women aged 16–35 years living in informal rural areas had low serum ferritin levels (Shisana et al., 2013:162). A study conducted on non-pregnant women, aged 18–40 years, in Bangladesh, Chile, China, the Dominican Republic, Pakistan, Thailand and Tunisia in the period 1988–1992 concluded that contraceptive use has a beneficial on ferritin levels

(UNDP/UNFPA/WHO/WB, 1998:261) which could serve as a possible explanation for the above mentioned differences. It is important to consider that serum ferritin levels may be falsely elevated in the prevalence of inflammation and infection (Gaw et al., 2008:112), thus the prevalence of iron deficiency and consequently iron deficiency anaemia could have been underestimated.

When comparing haemoglobin and serum ferritin levels to the WHO (2007:Online) cut off values for diagnosing iron status, 0.7% of the sample suffered from iron deficiency anaemia and 1.5% of the sample had an iron deficiency. Iron deficiency anaemia was found to be present in 9.7% of women of reproductive age in South Africa in the SANHANES-1 study. Differences in the prevalence of anaemia and iron deficiency anaemia could also be attributed to the older median age of the sample. When comparing the number of women in each age range to that of the SANHANES-1 study, where approximately similar numbers of women were included in the 25–34 years and 35–44 years (Shisana et al., 2013:162), the current study had more older women (35–49 years) than younger women (25–34 years) which could also be a possible reason for the differences as women of younger fertile age have a greater risk for developing anaemia, particularly due to iron deficiency, as a result of losses occurring during menstruation (Asres, Yemane & Gedefaw, 2014:Online).

Homocysteine levels rise and spill into the circulation when either folate or vitamin B12 is lacking, thus possibly indicating folate or vitamin B12 deficiency (Litchford, 2012:201). Overall, 3.8% of the sample had red cell folate levels below the reference value, indicating low folate status, which is much lower than the estimated prevalence of folate deficiency in 25–72% of women of reproductive age (Milman, 2011:370). This difference could be due to the small sample size in the current study, but also because of the intake of starchy staples, fortified with folic acid, by the entire population. Optimal folate status in women of childbearing age is of utmost importance as neural tube closure occurs at 28 weeks of pregnancy, a time when most pregnant women do not yet know that they are pregnant (Derbyshire, 2011:135).

Median MCV and MCH levels decreased significantly as dietary diversity scores increased, however, all of these medians were still within the normal range. The unexpected direction

of these associations may reflect more on the effects of the mandatory fortification of food in South Africa.

Although no significant association could be found between any of the blood parameters, except for MCH, and dietary diversity score, as well as any of the blood parameters and the flesh meats and fish food group and the organ meats group, there is a predominance of literature suggestive of the fundamental importance of iron and folate in maternal health (Derbyshire, 2011:26; WHO, 2006:Online). When considering what is known now regarding the low prevalence of anaemia and the adequacy of the women's diets in this study, it may possibly be an indication of the effect of the mandatory food fortification of maize meal and wheat flour with folic acid and iron, amongst others, that came into effect in South Africa in October 2003 (DoHSA, 2007:4). Food fortification may be a solution to addressing micronutrient malnutrition, particularly in terms of iron and folate, however, implementing other solutions as well as further studies may add value.

4.5 LIMITATIONS OF THE STUDY

The small sample size proved to be the greatest limitation of the current study, which may cast doubts on its representativity. The older median age may also have impacted on the representativity as majority of the women may be presenting with menopause which may have impacted on the low prevalence of anaemia. The prevalence of iron deficiency and iron deficiency anaemia also have been underestimated since no indicator of infection was measured to determine the prevalence of inflammation or infection. However, we believe that this study is still of value as it provides an indication of the dietary adequacy and the prevalence of anaemia in these women, which would otherwise not be known.

Although it was planned to include vitamin B12 levels in the current study, this parameter was not measured in the AHA data set. Even though the possibility exists that the remainder of the sample with elevated homocysteine levels in the absence of elevated red cell folate levels may possibly suffer from a vitamin B12 deficiency, this is unlikely since the women did not follow a vegetarian diet and were not at risk for age-related achlorhydria.

The dietary assessment reflects only one day of the diet of the women, which is not necessarily an accurate representation of the overall dietary adequacy. When interpreting dietary diversity scores, it is important to keep in mind that the quantity of food consumed is not measured; the diet can vary across seasons where some foods are only available in large quantities and at low affordable prices for short periods of time; and that the variety available in rural areas may differ from that of urban areas (FAO, 2011:27).

4.6 CONCLUSION AND RECOMMENDATIONS

The women in the current study consumed a diet with moderate variety which was based on starchy staple foods. Even though these women consumed little foods from the iron and folate containing food groups, the prevalence of iron deficiency, iron deficiency anaemia and folate deficiency was low. The low prevalence could be attributed to the fact that almost half of the women did not menstruate and the median age was older than that in other studies conducted on women of childbearing age. Women with a low dietary diversity score may be more prone to develop a megaloblastic anaemia than women who have a medium to high dietary diversity due to a greater variety of foods in their diet.

The results of this study will be used to plan sustainable community-based intervention strategies to improve the dietary adequacy of the women living in the Free State, where the dietetics students from the Department of Nutrition and Dietetics of the University of the Free State provide key nutrition services. These strategies should be appropriate and culturally acceptable and should involve the diversification of the diets of women living in the Free State, specifically focusing on improving the intake of iron and folate-rich food sources, as well as reducing those factors that may inhibit the absorption. We believe that projects encouraging the growing of vegetable gardens that are currently being implemented in the low socio-economic communities in Trompsburg, Philippolis and Springfontein should receive more attention, along with the promotion of raising chickens.

These interventions are important, even though the prevalence of anaemia in our study population is low, as the consequences of the nutrition transition may still impact on the health of these women and their offspring. We believe that the effectiveness of already

established nutrition intervention strategies, as well as the strategies mentioned earlier, would benefit from further research on this population. Reducing the burden of disease in developing communities in South Africa can be expected to contribute to sustainable livelihoods and improved health outcomes.

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CHAPTER 5

ANAEMIA AND ASSOCIATED ANTHROPOMETRIC VARIABLES AMONG WOMEN IN THE RURAL FREE STATE, SOUTH AFRICA

ABSTRACT

Background: Obesity and anaemia, particularly due to iron deficiency, both remain global public health problems which hold major consequences for human health. Optimal nutritional status, including optimal body composition, plays an important role in ensuring optimal health of women, particularly women of childbearing age.

Objectives: Determining the body composition, prevalence of anaemia, contraception use and the association between anaemia and body composition as well as contraception use in rural women between 25–49 years in the Free State province, South Africa.

Method: A cross-sectional descriptive study design was applied in a sample of 134 women. Weight, height, waist circumference, triceps, biceps, subscapular, and suprailiac skinfold measurements were measured according to standard techniques. Weight and height were used to calculate body mass index (BMI) which was categorised as underweight (<18.50kg/m^2), normal weight (18.50–24.99kg/m^2), overweight (25.00–29.99kg/m^2), obesity class I (30.00–34.99kg/m^2), obesity class II (35.00–39.99kg/m^2) and obesity class III (≥40.00kg/m^2). Waist circumference was categorised as normal (<80cm), at risk (≥80cm) and high risk (≥88cm). Body fat percentage was determined by means of the sum of the four skinfolds and categorised as too low (≤8%), acceptable lower end (9–23%), acceptable upper end (24–31%) and too high (≥32%). Blood samples were collected and analysed according to standard techniques. Full blood counts, transferrin saturation, ferritin, homocysteine and red cell folate levels were determined. Questions regarding contraceptive use were included in a questionnaire on reported health.

Results: Medians for BMI (28.7kg/m^2), waist circumference (90.8cm) and body fat percentage (38.8%) were all in the unhealthy ranges. With regard to body mass index, 25.4% of the women were classified as overweight and 45.4% were classified as obese. When considering waist circumference, 21.5% were classified as at risk and 57.7% as at high risk

for chronic diseases. Overall, 86.2% of the women presented with body fat percentages that were too high and unhealthy. Iron deficiency anaemia (haemoglobin < 12.0g/dL; ferritin < 15.0ng/mL) was present in only 0.7% (1/134) of the sample and 1.5% (3/134) had iron deficiency (haemoglobin > 12.0g/dL; ferritin < 15.0ng/mL). Overall, 7.5% of the sample presented with elevated homocysteine levels, however, only 3.8% presented with low levels of red cell folate indicative of folate deficiency. Only 54.1% of the women reported that they regularly menstruate and 71.6% currently or had previously used injectable contraceptives. Significant associations were found between median MCV, MCH and transferrin saturation levels across categories of BMI, waist circumference, as well as body fat percentage. The medians for these blood parameters decreased with increasing BMI, waist circumference and body fat percentage. An unexpected inverse association was also found between median homocysteine levels across BMI categories.

Conclusion: A predominant pattern of malnutrition, characterised by overweight and obesity, high rates of abdominal obesity and unhealthy body fat percentages were prevalent. Significant associations between BMI, waist circumference and body fat percentage categories with MCV, MCH and transferrin saturation levels indicate that risk for iron deficiency is associated with obesity. Attention should be given to improving the nutritional status of these women in order to reduce their risk for chronic diseases and anaemia.

Keywords: Anaemia; body composition; folate; iron; iron deficiency

5.1 INTRODUCTION

Populations undergoing a nutrition transition present with a high prevalence of chronic diseases associated with obesity on the one hand, and high rates of micronutrient deficiencies, including iron deficiency, on the other (Mujica-Coopman et al., 2014:Online). Obesity and iron deficiency still remain major independent contributing factors to the global burden of disease (WHO, 2008a:Online; WHO, 2000:Online). At present, obesity is considered to be a global pandemic (Popkin, Adiar & Nq, 2011:4), whereas worldwide, iron deficiency remains the most prevalent single micronutrient deficiency (WHO, 2008a:Online).

Many South African women from African decent are unwilling to lose weight as obesity is more culturally and aesthetically acceptable and even sought after, in these communities (Kruger et al., 2002:422). Elevated body mass index (BMI) is however associated with increased risk for chronic disease, particularly coronary heart disease, diabetes, stroke and some cancers in women (Kruger et al., 2002:422; Puoane et al., 2002:1038).

Contraception may potentially lower the risk for iron deficiency anaemia by helping to reduce the number of pregnancies and the time interval between consecutive pregnancies (UNDP/UNFPA/WHO/WB, 1998:261). The amount of blood lost during menstruation may also be reduced with contraception use (Yeasmin et al., 2010:25; UNDP/UNFPA/WHO/WB, 1998:262). Many women and clinicians believe that an association exists between hormonal contraceptive use and weight gain (Gallo et al., 2014:2).

Anaemia is defined as a significant reduction in the mass of circulating red blood cells leading to a diminished oxygen binding capacity of blood (Bunn, 2011:1031). Nutritional anaemias result from the insufficient intake and bioavailability of haemopoietic nutrients (iron, vitamin B12 and folic acid) (Biesalski & Erhardt, 2007:38).

Research on nutritional anaemias mainly focuses on iron-deficiency anaemia. Globally, approximately 111 000 deaths among pregnant women are attributed to iron-deficiency anaemia each year (SCN, 2004:Online). Unfortunately, little global data exists on the contribution of folate deficiency towards the development of anaemia (Balarajan et al., 2011:2128). It is important to not just focus on the prevalence of anaemia, but to determine the exact causal mechanisms in order to develop appropriate interventions (WHO, 2008a:Online).

Associations between obesity and iron deficiency in children and adults have been described in the literature (Cheng et al., 2011; Zekanowska et al., 2011; del Giudice et al., 2009). No studies have however investigated the relationship in women, particularly women of child bearing age, in South Africa. Limited data is also available on the prevalence of anaemia in general and in the Free State province in particular. This study thus aimed to determine the prevalence of anaemia, body composition, and association between anaemia and body

composition as well as contraceptive use in women aged 25–49 years in three rural towns in the Southern Free State.

5.2 RESEARCH METHOD AND DESIGN

5.2.1 Research approach

The work reported on in this article formed part of the 'Assuring Health for All in the Free State' (AHA-FS) research programme, an epidemiological study aimed at determining how living in rural and urban areas affects lifestyle and indicators of health. Trompsburg, Philippolis and Springfontein municipalities, form part of the Free State Rural Development Partnership Programme (FSRDPP), and were included in the rural phase of the AHA-FS study. As part of the data collection process, a multidisciplinary research team made use of standardised questionnaires to investigate socio-demographic status, household food security, dietary intake, levels of physical activity, knowledge, attitudes and practices relating to nutrition, and reported health of the study population. In addition to a medical examination, anthropometric measurements and blood specimens were also obtained for various investigations. For the purpose of this study, data from the rural phase of the original study were applied in a cross-sectional investigation.

Since very few studies, to our knowledge, have been conducted to determine the relationship between biochemical markers for anaemia and anthropometric status, the aim of this particular component of the study was to evaluate anthropometric variables, contraceptive use and anaemia in women between 25–49 years in these areas.

5.2.2 Population and sampling

All households in the low socio-economic townships (excluding farms) in the rural towns of Trompsburg, Philippolis and Springfontein in the Free State were eligible to participate in the original study. Low-socio economic communities in the townships of these towns are in the process of undergoing transitions in lifestyle, particularly changes in their nutrition and

physical activity, which have already been experienced by the populations living in formal settlements, who were thus excluded.

From the database of the original study, women aged 25–49 years who were HIV-uninfected, who gave informed consent and who were not pregnant at the time of data collection, were selected for the current study, resulting in a final sample of 134 women.

5.2.3 Methodology

Anthropometric measurements used in the current study included height, weight, triceps, biceps, subscapular and supra-ileac skinfolds as well as waist circumference. Trained fourth year dietetics students obtained all the measurements using standardised measuring techniques as described in the literature (Lee & Nieman, 2013:170; Gibson, 2005:253). Weight and height measurements were used to calculate BMI and the sum of the four skinfolds was used to calculate body fat percentage.

The BMI of the participants were used to determine whether the participants were underweight, normal weight, overweight or obese and was classified as suggested by the World Health Organization (WHO) (Table 5.1).

Table 5.1 International classification of adult underweight, overweight and obesity according to BMI (WHO, 2013:Online)

BMI (kg/m^2)	Classification
< 18.5	Underweight
18.5 – 24.99	Normal weight
25.0 – 29.99	Overweight
30.0 – 34.99	Obesity class I
35.00 – 39.99	Obesity class II
≥ 40.00	Obesity class III

Waist circumference was used to assess body fat distribution (Lee & Nieman, 2013:187) and was classified as indicated in table 5.2.

Table 5.2 Waist circumference cut-off values (Lee & Nieman, 2013:187; WHO, 2008b:Online)

Waist circumference measurement	Classification
< 80cm	Normal
80–87cm	At risk
≥88cm	High risk

The sum of four skinfolds, namely, triceps, biceps, subscapular and suprailiac skinfolds were used to calculate body fat percentage. The four skinfolds were added up and the corresponding actual fat percentage was read off the "Percentage of body fat based on four skinfold measurements" chart (Durnin & Woemersley, 1974:77-97). Body fat ranges for persons older than 18 years of age used to interpret the body fat percentage of the participants, are indicated in table 5.3 (Lee & Nieman, 2013:199).

Table 5.3 Body fat ranges for persons 18 years of age and older (Lee & Nieman, 2013:199)

Classification	Females
Unhealthy range (too low)	≤ 8%
Acceptable range (lower end)	9 – 23%
Acceptable range (upper end)	24 – 31%
Unhealthy (too high)	≥ 32%

For the purpose of this study, fasting blood samples were used to measure full blood count, serum ferritin levels and transferrin saturation, homocysteine levels, and red cell folic acid levels in all participants.

Respondents were required to fast overnight with blood samples being collected by chemical pathologists the next morning. These samples were stored immediately in ice-filled containers and transported to the laboratory. The blood samples were analysed

according to standard techniques at the National Health Laboratory Service Laboratory at the University of the Free State. Full blood counts were determined using the Roche Sysmex XT 2000i® analyser (Sysmex, 2014:Online), iron levels using the Beckman Coulter Synchron LX20 (Mikolaenko et al., 2000:387), and transferrin saturation and homocysteine levels using the BN Prospec System nephelometric technology. Serum folate levels were recorded using the Bayer Advia Centaur System (Hadler et al., 2008: S262).

Haemoglobin levels below 12.1g/dL were diagnosed as anaemia (NHLS reference values). According to the WHO (2008a:Online), iron deficiency is diagnosed when ferritin levels are below 15.0ng/mL and haemoglobin levels are greater than 12.0g/dL; and iron deficiency anaemia is diagnosed when ferritin and haemoglobin levels are below 15.0ng/mL and 12.0g/dL respectively. These cut-off values were used to define iron deficiency and iron deficiency anaemia in this sample.

As part of the reported health questionnaire, participants had to answer questions relating to menstruation and contraceptive use in a structured interview with trained students.

5.2.4 Study procedures

Fieldworkers visited all the households in the three areas to explain the study to the adult members and to invite those between the ages of 25-64 years to participate. Data was obtained from the participants at the community halls in each of the rural areas. In order to make sure that the participants met the criteria for age, ID documents were screened. Participants were required to rotate between different research stations in the venue.

In order to ensure confidentiality of the data, each respondent was assigned a specific number. Ethical approval was obtained from the Ethics Committee of the Faculty of Health Sciences of the University of the Free State (ETOVS number 21/07), the Department of Health and local municipalities. Participation was voluntary and could be ended at any time.

5.2.5 Statistical analysis

Statistical analysis was conducted using the Predictive Analytics SoftWare (PASW) Statistics Student version 22.0 by Statistical Package for the Social Sciences (SPSS) (IBM, 2013). Frequencies and percentages were determined for categorical data and medians and percentiles for continuous data were recorded. The Kruskal-Wallis test was used to determine p-values in order to compare medians between continuous and categorical variables to describe associations between variables within the entire sample.

5.3 RESULTS

After applying the exclusion criteria, data from 134 females could be included in the analysis, however, missing values had an effect on the n-value of most variables. The median age, BMI, waist circumference and body fat percentage of the sample is indicated in table 5.4. All median values for the measurements were in the unhealthy categories.

Table 5.4 Median age, BMI, waist circumference and body fat percentage

Variable	N	Median	Minimum	Maximum
Age (years)	134	41.0	25.0	49.0
BMI (kg/m^2)	130	28.7	11.9	52.2
Waist circumference (cm)	130	90.8	56.0	135.0
Body fat percentage (%)	123	38.3	21.5	48.8

Overall, the majority (70.8%) of the women presented with BMI values above normal, and only a small percentage (6.9%) were classified as underweight (Table 5.5).

Table 5.5 BMI categories (%)

BMI categories	n	%
Underweight (< 18.5kg/m^2)	9	6.9
Normal weight (18.5 – 24.99kg/m^2)	29	22.3
Overweight (25.0 – 29.99kg/m^2)	33	25.4
Obesity class I (30.0 – 34.99kg/m^2)	27	20.8
Obesity class II (35.0 – 39.99kg/m^2)	13	10.0
Obesity class III (≥40kg/m^2)	19	14.6

Almost 60% of the women presented with waist circumference measurements in the high risk category (Table 5.6).

Table 5.6 Waist circumference categories (%)

Waist circumference categories	n	%
Normal (<80cm)	27	20.8
At risk (80cm-87cm)	28	21.5
High risk (≥88cm)	75	57.7

None of the women in the sample presented with body fat percentages that were too low. It is however, concerning to note that almost 90% presented with body fat percentages in the too high, unhealthy range (Table 5.7).

Table 5.7 Body fat percentage categories (%)

Fat percentage categories	n	%
Unhealthy, too low (≤8%)	0	0
Acceptable range, lower end (9-23%)	2	1.6
Acceptable range, upper end (24-31%)	15	12.2
Unhealthy, too high (≥32%)	106	86.2

Only 4.6% (6/134) of the women presented with low haemoglobin levels (Table 5.8), with even fewer women presenting with low haematocrit (3.1%, 4/134), mean corpuscular

volume (MCV) (3.1%, 4/134) and mean corpuscular haemoglobin (MCH) (7.6%, 10/134) levels. Elevated MCH levels were present in 25.2% (33/134) of the study population.

Table 5.8 Haemoglobin, haematocrit, MCV, MCH, transferrin saturation, ferritin, homocysteine and red cell folate levels

Blood samples	Normal reference value	N	Median	Low (%)	Normal (%)	High (%)
Haemoglobin	> 12.1g/dL	130	13.8	4.6	95.4	-
Haematocrit	> 0.371L/L	131	0.429	3.1	96.9	-
MCV	79.1–98.9fl	131	93.8	3.1	77.1	19.8
MCH	27.0–32.0pg/cell	131	30.4	7.6	67.2	25.2
Transferrin saturation	> 15%	74	26.1	12.2	87.8	-
Ferritin	6–120ng/mL	74	94.0	1.4	58.1	40.5
Homocysteine	2.10–15.70µmol/L	134	9.6	-	92.5	7.5
Red cell folate	> 372nmol/L	131	575.3	3.8	96.2	-

Ampath. 2010. Ampath desk reference: guide to laboratory tests. Centurion: Ampath.

National Health Laboratory Services Reference values. 2014.

Transferrin saturation below normal (<15 %) were present in 12.5% (9/134) of the sample, with 1.4% (1/134) of the women displaying low ferritin levels (<6 ng/mL). Overall, only 4.6% of the women presented with anaemia, 2.3% with iron deficiency and 0.7% with iron deficiency anaemia. Elevated homocysteine levels (>15.70 µmol/L) occurred in 7.1% (10/134) of the sample and 4.0% (5/134) had low red cell folate levels (<372 nmol/L). None of the participants however presented with anaemia due to folate deficiency, however, those with low levels of red cell folate levels were categorised as at risk of developing folate deficiency.

When assessing the prevalence of anaemia, particularly iron deficiency anaemia, it is important to note that 45.9% of the women did not menstruate regularly, and at the time of data collection, 71.6% were currently using, or had previously used, injectable contraceptives (Table 5.9).

Table 5.9 Information pertaining to menstruation patterns and contraceptive use

Questions	N	Yes (%)	No (%)
Still menstruated regularly	133	54.1	45.9
Currently or previously used injectable contraceptives	134	71.6	28.4

Statistically significant inverse associations were found between BMI categories and median MCV (p=0.0001), median MCH (p=0.0001), median transferrin saturation (p=0.011) and median homocysteine (p=0.006) levels (Table 5.10).

Table 5.10 Associations between blood parameters and BMI categories

Variables	n	Median blood values	p-value for median differences
Median haemoglobin levels across BMI categories:			0.129
Underweight	9	13.5g/dL	
Normal weight	27	14.4g/dL	
Overweight	32	14.1g/dL	
Obese class 1	27	13.6g/dL	
Obese class 2	12	13.9g/dL	
Obese class 3	19	13.8g/dL	
Median haematocrit levels across BMI categories:			0.600
Underweight	9	0.416L/L	
Normal weight	28	0.438L/L	
Overweight	32	0.430L/L	
Obese class 1	27	0.424L/L	
Obese class 2	12	0.428L/L	
Obese class 3	19	0.434L/L	
Median MCV levels across BMI categories:			0.0001*
Underweight	9	98.6fl	
Normal weight	28	97.4fl	
Overweight	32	92.9fl	
Obese class 1	27	90.9fl	
Obese class 2	12	93.8fl	
Obese class 3	19	90.5fl	

Median MCH levels across BMI categories:			0.0001*
Underweight	9	31.5pg/cell	
Normal weight	28	31.7pg/cell	
Overweight	32	30.6pg/cell	
Obese class 1	27	29.4pg/cell	
Obese class 2	12	30.3pg/cell	
Obese class 3	19	29.2pg/cell	
Median transferrin saturation across BMI categories:			0.011*
Underweight	3	42.0%	
Normal weight	14	31.9%	
Overweight	15	28.9%	
Obese class 1	16	19.8%	
Obese class 2	6	28.5%	
Obese class 3	16	21.5%	
Median ferritin levels across BMI categories:			0.137
Underweight	3	127.0ng/mL	
Normal weight	14	179.5ng/mL	
Overweight	15	98.0ng/mL	
Obese class 1	16	76.5ng/mL	
Obese class 2	6	89.0ng/mL	
Obese class 3	16	49.0ng/mL	
Median homocysteine levels across BMI categories:			0.006*
Underweight	9	12.2µmol/L	
Normal weight	29	11.3µmol/L	
Overweight	33	10.4µmol/L	
Obese class 1	27	9.1µmol/L	
Obese class 2	13	8.8µmol/L	
Obese class 3	19	9.2µmol/L	
Median red cell folate levels across BMI categories:			0.809
Underweight	9	560.0nmol/L	
Normal weight	28	603.8nmol/L	
Overweight	32	557.1nmol/L	
Obese class 1	27	541.6nmol/L	
Obese class 2	12	556.6nmol/L	
Obese class 3	19	575.3nmol/L	

* Statistically significant association

Median MCV levels decreased significantly between underweight and overweight and obesity and between normal weight and overweight, obesity class 1 and class 3. Similar trends were observed for median MCV levels and transferrin saturation. A similar,

unexpected association was found with median homocysteine levels decreasing as BMI increased.

Significant associations were also found between waist circumference categories and median MCV levels (p=0.003), MCH levels (p=0.001) and transferrin saturation (p=0.002), however, no significant associations were found between waist circumference categories and median haemoglobin, homocysteine, red cell folate and ferritin levels (Table 5.11). Median MCV, MCH levels and transferrin saturation all decreased as waist circumference categories increased.

Table 5.11 Associations between blood parameters and waist circumference categories

Variables	n	Median blood values	p-value for median differences
Median haemoglobin levels across waist circumference categories:			0.629
Normal	27	13.6g/dL	
At risk	30	13.7g/dL	
High risk	69	14.0g/dL	
Median haematocrit levels across waist circumference categories:			0.259
Normal	28	0.418L/L	
At risk	30	0.425L/L	
High risk	69	0.434L/L	
Median MCV levels across waist circumference categories:			0.003*
Normal	28	97.3fl	
At risk	30	96.7fl	
High risk	69	91.6fl	
Median MCH levels across waist circumference categories:			0.001*
Normal	28	31.6pg/cell	
At risk	30	31.1pg/cell	
High risk	69	29.6pg/cell	

Median transferrin saturation across waist circumference categories:			0.002*
Normal	12	39.0%	
At risk	14	29.9%	
High risk	45	24.5%	
Median ferritin levels across waist circumference categories:			0.434
Normal	12	129.0ng/mL	
At risk	14	88.0ng/mL	
High risk	45	92.0ng/mL	
Median homocysteine levels across waist circumference categories:			0.178
Normal	28	10.5µmol/L	
At risk	30	11.0µmol/L	
High risk	72	9.4µmol/L	
Median red cell folate levels across waist circumference categories:			0.494
Normal	28	603.0nmol/L	
At risk	30	598.2nmol/L	
High risk	69	576.1nmol/L	

* Statistically significant association

As seen in BMI categories, associations between body fat percentage and median MVC levels (p=0.025), MCH levels (p=0.019) and transferrin saturation (p=0.013) were also identified, with no significant associations found between body fat percentage and medians of other blood parameters (Table 5.12). All median MCV and transferrin saturation again decreased with increasing body fat percentage categories. Median MCH levels decreased significantly between acceptable range, upper end and unhealthy, too high body fat percentages.

Table 5.12 Associations between blood parameters and body fat percentage categories

Variables	n	Median blood values	p-value for median differences
Median haemoglobin levels across body fat percentage categories:			0.808
Unhealthy, too low	0		
Acceptable range, lower end	2	13.5g/dL	

Acceptable range, upper end	15	13.6g/dL	
Unhealthy, too high	103	13.8g/dL	
Median haematocrit levels across body fat percentage categories:			0.891
Unhealthy, too low	0		
Acceptable range, lower end	2	0.425L/L	
Acceptable range, upper end	15	0.421L/L	
Unhealthy, too high	104	0.430L/L	
Median MCV levels across body fat percentage categories:			0.025*
Unhealthy, too low	0		
Acceptable range, lower end	2	99.7fl	
Acceptable range, upper end	15	97.6fl	
Unhealthy, too high	104	93.0fl	
Median MCH levels across body fat percentage categories:			0.019*
Unhealthy, too low	0		
Acceptable range, lower end	2	31.7pg/cell	
Acceptable range, upper end	15	31.8pg/cell	
Unhealthy, too high	104	30.1pg/cell	
Median transferrin saturation across body fat percentage categories:			0.013*
Unhealthy, too low	0		
Acceptable range, lower end	0		
Acceptable range, upper end	7	36.0%	
Unhealthy, too high	60	25.2%	
Median ferritin levels across body fat percentage categories:			0.114
Unhealthy, too low	0		
Acceptable range, lower end	0		
Acceptable range, upper end	7	134.0ng/mL	
Unhealthy, too high	60	88.0ng/mL	
Median homocysteine levels across body fat percentage categories:			0.277
Unhealthy, too low	0		
Acceptable range, lower end	2	14.6µmol/L	
Acceptable range, upper end	15	9.4µmol/L	
Unhealthy, too high	106	9.6µmol/L	

Median red cell folate levels across body fat percentage categories:			0.541
Unhealthy, too low	0		
Acceptable range, lower end	2	681.4nmol/L	
Acceptable range, upper end	15	908.8nmol/L	
Unhealthy, too high	104	576.6nmol/L	

* Statistically significant association

A significant association was found between median haemoglobin levels and whether women menstruated regularly or not (p=0.008), with no significant association between median haemoglobin levels and whether women currently or previously used injectable contraceptives or not (Table 5.13). Median haemoglobin levels were higher in those women who menstruated regularly compared to those who did not.

Table 5.13 Associations between haemoglobin, menstruation and contraceptive use

Variables	n	Median blood value	p-value for median differences
Median haemoglobin levels across categories of menstruation:			0.008*
Menstruated regularly	70	13.7g/dL	
Did not menstruate regularly	59	14.2g/dL	
Median haemoglobin levels across categories of contraceptive use:			0.847
Made use of injectable contraceptives	94	13.8g/dL	
Did not use injectable contraceptives	36	13.9g/dL	

* Statistically significant association

5.4 DISCUSSION

Obesity and anaemia are two major global health concerns (WHO, 2008a:1; WHO, 2000:16). Although obesity and iron deficiency usually represent opposite ends of the spectrum of malnutrition, some studies have shown a link between these two conditions (Cheng et al., 2011; Zekanowska et al., 2011; Cepeda-Lopez, Aeberli & Zimmermann, 2010; del Giudice et al., 2009). Since both obesity and anaemia pose a threat to human health, the current study aimed to determine the need for intervention studies in this population.

The median BMI in the women fell in the overweight category (28.7kg/m^2), with 25.4% of the sample presenting with overweight and 45.4% presenting with obesity. These results are similar to the findings of the SANHANES-1 study where 28.0% and 36.3% of women aged 25–34 years were overweight or obese respectively. Among women aged 35–44 years, 26.4% were overweight and 44.8% were obese (Shisana et al., 2013:140). The South African Health and Demographic Survey conducted in 2003, found that the mean BMI among non-pregnant South African women 25–34 years, 35–44 years and 45–54 years were also in the overweight category at 26.8kg/m^2, 28.6kg/m^2 and 29.4kg/m^2 respectively (DoH, MRC & OrcMacro, 2007:277). Among non-pregnant women in the Transition and Health During Urbanisation of South Africans (THUSA) study of African descent, 26.8% and 24.6% aged 25–34 years, 31.1% and 39.0% aged 35–44 years, and 25.0% and 35.2% aged 45–54 years, were overweight or obese, respectively (Kruger et al., 2002:424).

Almost 60% of the women presented with a waist circumference that fell in the high risk category, thus putting them at increased risk for developing chronic diseases due to obesity. The median waist circumference was 90.8cm, also within the high risk category. These findings were again similar to that of the South African Health and Demographic Survey of 2003 where the mean waist circumference measurements were 82.3cm in women 25–34 years, 85.1cm in women 35–44 years, and 88.4cm in women 45–54 years (DoH, MRC & OrcMacro, 2007:281). These results were also similar to those of the SANHANES-1 study where the mean waist circumference in women aged 25–34 was 87.7cm and in women aged 35–44 was 90.9cm (Shisana et al., 2013:142).

The median body fat percentage of the women in the current study was 38.3% which falls in the unhealthy category. Almost 90% of the women had body fat percentages in the unhealthy range. The mean body fat percentage among African women in a study conducted on South African women from the Cape Town Metropole Area with mean age of 40 years was 34.0%, which is very similar to the results from the current study (Mciza et al., 2005:514).

Only a small percentage of women in the current study suffered from anaemia (Table 5.8), which is much lower than that of women in the SANHANES-1 study where 24.7% of women

aged 25–34 years and 23.1% of women aged 35–44 years were reported to be anaemic (Shisana et al., 2013:142). The prevalence of anaemia in the current study is also much lower than the results from a systematic review by Stevens et al. (2013:e19), that looked at population representative data among women aged 15–49 years, where 28% of non-pregnant women in South Africa were anaemic. Occurrence of anaemia in these two studies was thus much higher than that of the current study. Possible reasons for these difference could be that almost half of the women in the current study did not menstruate regularly and a large proportion of the sample were currently using, or had previously made use of, injectable contraceptives. These results could also be further explained by the fact that a significant association was found between the medians for haemoglobin and whether the women still menstruated regularly, which could indicate that women who still menstruated were more likely to have lower levels of haemoglobin. Other studies, including the SANHANES-1 study, did not report on menstruation and contraceptive use, making it difficult to compare results. A study conducted by Yeasmin et al. (2010:28) among women aged 20–40 years from a low socio-economic background in Dhaka, Bangladesh, found that women who used oral contraceptives had significantly higher haemoglobin levels than those who did not. The current study did not find a significant association between contraceptive use and haemoglobin levels.

Iron deficiency anaemia develops in three stages. Depletion of iron stores occurs during the first stage and is confirmed by low serum ferritin levels. During the following stage, serum ferritin decreases further with transferrin saturation decreasing as well. These decreases are followed by the development of microcytosis and hypochromia after which signs and symptoms of anaemia appear (Gaw et al., 2008:112).

Other parameters often seen in combination with low haemoglobin levels in iron deficiency anaemia include decreased levels of MCV (microcytic anaemia) and MCH (hypochromic anaemia). Only 2/134 (1.5%) women presented with microcytic, hypochromic anaemia and only 2/134 (1.5%) women presented with hypochromic anaemia with normal MCV values. A quarter (25.2%) of the women had increased levels of MCH, which is seen in macrocytic anaemia.

A very low percentage of the women in the current study presented with low ferritin levels, which are indicative of low iron stores. When compared to the SANHANES-1 study where 13.5% of women aged 16–35 years living in informal rural areas had low serum ferritin levels (Shisana et al., 2013:162), the women in the current study had much higher iron stores. The median age of 41 years in the current study could also partly explain differences in the prevalence of anaemia, iron deficiency as well as low iron stores. When comparing the number of women in each age range to that of the SANHANES-1 study where approximately similar number of women were included in the 25–34 years and 35–44 years (Shisana et al., 20113:161), more older women (35–49 years) were included in the current study than younger women (25–34 years). Women of younger fertile age have a greater risk for developing anaemia, particularly due to iron deficiency, as a result of losses occurring during menstruation (Asres, Yemane & Gedefaw, 2014:Online).

According to the WHO (2007:Online), iron deficiency is diagnosed when ferritin levels are below 15ng/mL and haemoglobin levels are still above 12g/dL; and iron deficiency anaemia is diagnosed when both ferritin and haemoglobin levels are below 15ng/mL and 12mg/dL respectively. When using this diagnosis, only one woman (0.7%) in the sample suffered from iron deficiency anaemia and two women (1.5%) in the sample had an iron deficiency. Iron deficiency anaemia was found to be present in 9.7% of women of reproductive age in South Africa in the SANHANES-1 study (Shisana et al., 2013:162), which is a much higher percentage than that in the current study.

Various factors may lead to the development of anaemia, including deficiencies of other micronutrients besides iron. Folate or vitamin B12 deficiency may be present when homocysteine levels are elevated as homocysteine levels rise and spill into the urine when either folate or vitamin B12 is lacking (Litchford, 2012:201). Red cell folate levels below the reference value were found in 3.8% of the women, which is much lower than the estimated prevalence of folate deficiency in 25–72% of women of reproductive age living in developing countries (Milman, 2011:370).

Although no significant associations were found between median haemoglobin levels and categories of BMI, waist circumference and body fat percentage, significant associations

were found between median MCV, MCH levels and transferrin saturation and all three anthropometric measurements (median MCV, MCH levels and transferrin saturation decreased as the anthropometric measurements increased). Unfortunately, very few studies have reported on the relationship between these blood parameters and anthropometric variables with most only referring to haemoglobin. A study conducted among adults in Washington DC did however find that MCV levels and transferrin saturation decreased as BMI increased (Yanoff et al., 2007:1414). A study conducted by Vuong et al. (2014:673), using data of 25–55 year old men and women from the National Health and Nutrition Examination Survey in the United States' dataset, also found that MCV levels, as well as MCH levels, decreased with higher waist circumferences similar to the results of the current study. No studies on the association between body fat percentage and MCV, MCH levels and transferrin saturation could be found.

Some studies have reported a significant association between haemoglobin and overweight and obesity, central obesity (Qin et al., 2013:Online) and body fat percentage (Mohamed & Alhessain, 2014:Online) as well as serum ferritin concentrations and obesity (Mujica-Coopman et al., 2014:Online).

Median homocysteine levels in the current study decreased as BMI increased. This result is unexpected as both obesity (Vayá et al., 2012:49) and elevated homocysteine have been found to be independent risk factors for atherosclerosis and thrombosis (Rosolova et al., 2002:93). Various studies have assessed the association between these two risk factors (Vayá et al., 2012; Rosolova et al., 2002; Sánchez-Margalet et al., 2002). Most studies have found a direct relationship between obesity and homocysteine, with levels of homocysteine increasing as weight increases (Vayá et al., 2012; Sánchez-Margalet et al., 2002). Although these findings differ from those of the current study, a study conducted by Rosolova et al. (2002:96), among healthy Czech men and women also found an indirect relationship between homocysteine and obesity, just as the current study did. A very small percentage of the women in the current study suffered from a folate deficiency and since folate is required for the conversion of homocysteine to methionine, this may serve as a possible reason for the unexpected inverse association.

Significant associations between anthropometric variables and markers of iron status may be due to inflammation, as various studies conducted in obese adults have shown increased levels of pro-inflammatory molecules which may impair iron status by reducing the bioavailability of iron (Cheng et al., 2011; Zekanowska et al., 2011; del Giudice et al., 2009). A recent meta-analysis of all controlled studies on adult obesity and low iron stores concluded that decreased transferrin saturation is consistent with the mechanism of obesity-related inflammation (Cheng et al., 2011:157). The high prevalence of obesity in the current study and significant associations between median transferrin saturation and BMI, waist circumference and body fat percentage categories thus mirrors the findings of other studies.

5.5 LIMITATIONS OF THE STUDY

The small sample size proved to be the greatest limitation of the current study, which may question representativity. The older median age may also have impacted on the representativity. However, we believe that this study is still of value as it gives an idea of the prevalence of anaemia and obesity as well as the potential relationship between these variables in these women, which would otherwise not be known.

Although it was planned to include vitamin B12 levels in the current study, this parameter was not measured in the AHA data set. Those participants that presented with elevated homocysteine, but normal folate levels may have suffered from a vitamin B12 deficiency, however, these women were not at risk for vitamin B12 deficiency as they did not follow a vegetarian diet and were not at risk for achlorhydria that is associated with older age.

5.6 CONCLUSION AND RECOMMENDATIONS

The results of this study suggest that there was a predominant pattern of malnutrition, characterised by overweight and obesity, high rates of abdominal obesity and unhealthy body fat percentages, among the women included in this study. These women were at high risk for developing chronic diseases of lifestyle due to obesity. The prevalence of iron deficiency, iron deficiency anaemia and folate deficiency was low. The low prevalence could

be attributed to the fact that almost half of the women did not menstruate anymore and the median age was older than that in other studies conducted on women of childbearing age. The mandatory food fortification of certain staple foods may also be a possibly reason. Significant associations between BMI, waist circumference and body fat percentage categories and MCV, MCH levels and transferrin saturation indicate that the risk for iron deficiency is associated with obesity.

Haemoglobin levels below normal cut off values on its own does not identify the type of anaemia. It is thus important for future research to include other parameters of iron and folate status.

Culturally sensitive and sustainable community-based interventions need to be planned and implemented in the study area, based on the findings of the current study. It is possible to implement these interventions as final year dietetics students from the Department of Nutrition and Dietetics already provide nutrition services in these areas. These interventions should be focused on changing the perception of these women that obesity is not unhealthy as well as promoting physical activity along with an appropriate diet and lifestyle.

Although the prevalence of anaemia in our study was low, the importance of interventions in reducing the risk of anaemia cannot be disregarded. The strategies mentioned earlier, in addition to already established nutrition intervention strategies in these communities would benefit from further research. Efforts directed towards reducing the burden of disease in developing communities in South Africa have the potential to contribute to sustainable livelihoods and improved health outcomes.

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CHAPTER 6

THE ASSOCIATION OF REPORTED HEALTH STATUS AND LIFESTYLE FACTORS WITH ANAEMIA AMONG WOMEN IN THE RURAL FREE STATE, SOUTH AFRICA

ABSTRACT

Background: Indicators of women's health in Africa are generally poor, and progress towards achieving the Millennium Development Goals has been slow. Priority interventions for addressing non-communicable diseases, include accelerated tobacco control, salt reduction, promotion of healthy diets and physical activity, reduction of harmful alcohol consumption and access to essential drugs and technologies. Investing in safeguarding women's health can greatly impact upon societal growth and advancement.

Objectives: Determining reported health, prevalence of anaemia and contraceptive use as well as association between anaemia and reported health among rural women aged 25–49 years in the Free State province, South Africa.

Method: A cross-sectional descriptive study design was applied in a sample of 134 women. A reported health questionnaire was completed for each woman in a structured interview and included information on social support (group membership, network of friends, family structure), tobacco and alcohol consumption patterns, medical history and medications, contraceptive use, family medical history, alternative medical practices, levels of stress and behaviours related to stress control. Blood samples were collected and analysed according to standard techniques. These included full blood counts, transferrin saturation, ferritin, homocysteine and red cell folate levels.

Results: Almost half of the women (47.0%) smoked and 29.3% used snuff. The majority currently or formerly used alcohol (71.5%), and 38.7% consumed more than five drinks per day, at least once a month. Some reported breathlessness with usual activity (41.0%), loose stools/diarrhoea for at least three days (17.3%), vomiting (18.0%), loss of appetite (41.0%), blood in their urine (7.5%), as well as involuntary weight loss of more than 3kg (42.5%) in the past six months. More than half of the women suffered from hypertension (56.4%). Medication was regularly used by 64.2% and hypertension medication was the most

common (45.1%). Almost one in five of the women (17.9%) had been hospitalised within the past 24 months. More than half (54.1%) of the women in the sample menstruated regularly, 71.6% currently or previously used injectable contraceptives, and 68.9% had given birth to three or fewer children. Only 0.7% of women suffered from iron deficiency anaemia (haemoglobin < 12.0g/dL; ferritin < 15.0ng/mL) and 1.5% had an iron deficiency (haemoglobin > 12.0g/dL; ferritin < 15.0ng/mL). Less than 10% (7.5%) of the sample presented with elevated homocysteine levels, however, only 3.8% presented with low levels of red cell folate indicative of folate deficiency. Median haemoglobin levels were significantly higher among those women who smoked, compared to those who did not, possibly due to the exposure to carbon monoxide that binds haemoglobin, reducing its oxygen carrying capacity. Unexpectedly, median haemoglobin levels were significantly higher among those women who experienced breathlessness with usual activity, probably due to other reasons not investigated in the current study. As expected, the group of women who menstruated regularly had a significantly lower median haemoglobin level, than those who did not.

Conclusion: Overall, the women's risk for anaemia in terms of other health factors assessed in this study, seem to be low. The prevalence of iron deficiency, iron deficiency anaemia, as well as folate deficiency in the sample was low, possibly due to the fact that half of the women were not menstruating. Results from the current study indicate that smoking could potentially mask the presence of anaemia.

Keywords: Anaemia, health, folate, iron, iron deficiency

6.1 INTRODUCTION

Investing in the safeguarding of women's health can greatly impact upon societal growth and advancement (Izugbara & Covan, 2014:697). The majority of maternal deaths can be avoided through interventions aimed at providing appropriate health care services by skilled health care providers with the necessary equipment and supplies (UN, 2010:30). Therefore, "improving maternal health" is one of the priorities of the Millennium Development Goals set to be achieved by 2015 (UN, 2010:30). According to the United Nations (UN) (2010:30) report on the progress of these goals, haemorrhage and hypertension are the leading

causes of maternal deaths in developing regions. In Africa, women's health indicators are however, still poor and progress towards achieving the Millennium Development Goals, particularly those that pertain to women's health and wellbeing, has been slow (Izugbara & Covan, 2014:697).

Anaemia remains a major concern that affects global development in high as well as low and middle income countries (Jatav et al., 2014:9; WHO, 2008:Online). The prevalence of non-communicable diseases, including anaemia, keeps rising and remains a global challenge (Jatav et al., 2014:9; Beaglehole et al., 2011:1438). Anaemia is commonly associated with fatigue, dyspnoea, palpitations (Bunn, 2011:1033), weakness, pallor, dizziness, reduced exercise tolerance or irritability (Ginder, 2011:1041). It is of utmost importance to determine associations between health outcomes and the prevalence of anaemia in order to develop appropriate interventions to address the problem (WHO, 2008:Online). Priority interventions for addressing non-communicable diseases include accelerated tobacco control, salt reduction, promotion of healthy diets and physical activity, reduction of harmful alcohol consumption and access to essential drugs and technologies (Beaglehole et al., 2011:1440).

Contraceptive use has the potential to decrease the risk for anaemia as contraceptives reduce the number of pregnancies as well as the time interval between pregnancies (UNDP/UNFPA/WHO/WB, 1998:261). Menstrual blood loss, which could further contribute to iron deficiency, is also decreased with contraceptive use (Yeasmin et al., 2010:25; UNDP/UNFPA/WHO/WB, 1998:262).

Even though a number of studies that relate to anaemia have been conducted in South Africa, limited data are available regarding the prevalence of anaemia in Southern African women in general, and in the Free State province in particular, including the relationship between anaemia and reported health. This study thus aimed to determine the prevalence of anaemia, reported health, as well as contraceptive use and associations between anaemia and reported health in women aged 25–49 years in three rural towns in the Free State.

6.2 METHOD

6.2.1 Ethical considerations

Ethical approval to conduct the current study was obtained from the Ethics Committee of the Faculty of Health Sciences of the University of the Free State (ETOVS number 21/07). Permission was also obtained from the Department of Health and local municipalities. Participation was voluntary and could be ended at any time. Confidentiality of all participants was ensured by assigning a specific number to each participant.

6.2.2 Reference population and sampling

All households in the low socio-economic townships (excluding farms) in the rural towns of Trompsburg, Philippolis and Springfontein in the Free State were eligible to participate in the original study. low socio-economic communities in the townships of these towns are in the process of undergoing transitions in lifestyle, particularly changes in their nutrition and physical activity, which have already been experienced by the populations living in formal settlements.

Women who did not fall within the age range of 25–49 years, who were HIV infected and who were pregnant at the time of data collection, were excluded from the current study. All participants gave written informed consent. The final sample consisted of 134 women.

6.2.3 Measurement process

The work reported on in the current study formed part of the 'Assuring Health for All in the Free State' (AHA-FS) research programme. The AHA-FS study is an epidemiological study aimed at determining how living in rural and urban areas affects lifestyle and indicators of health. Trompsburg, Philippolis and Springfontein municipalities, form part of the Free State Rural Development Partnership Programme (FSRDPP), and formed part of the rural phase of the AHA-FS study.

Fieldworkers visited all the households in the three areas to explain the study to the adult members and to invite those between the ages of 25-64 years to participate. Data were obtained from the participants at the community halls in the rural areas. In order to ensure that the participants met the criteria for the age, their ID documents were screened. Participants were required to rotate between different research stations in the venue.

A multidisciplinary research team made use of standardised questionnaires to investigate socio-demographic status, household food security, dietary intake, levels of physical activity, knowledge, attitudes and practices relating to nutrition, and reported health of the study population. A medical examination, anthropometric measurements and blood specimens were also obtained for various investigations. Data collected from the rural phase of the original study were used and applied in a cross-sectional investigation in the current study.

Haematological abnormalities of both HIV-infected and HIV-uninfected persons in the AHA study population have been published by Groenewald et al. (2011) and van Zyl et al. (2010), but data on the association between anaemia indices and known health risk behaviours within these communities have not previously been investigated. The aim of this particular component of the study was thus to evaluate reported health, the prevalence of anaemia and contraceptive use in women between 25–49 years in these areas.

6.2.4 Data handling and analysis

A health questionnaire was used to obtain information on reported health from each woman in a structured interview. This questionnaire included information on tobacco and alcohol consumption patterns, medical history and medication use, contraceptive use and family medical history.

Fasting blood samples were used to measure full blood count, serum ferritin and transferrin saturation, homocysteine levels and red cell folic acid levels in all participants.

Respondents were required to fast overnight to allow the collection of blood samples by medical doctors the following morning. These samples were stored immediately in ice-filled

containers and transported to the laboratory. The blood samples were analysed according to standard techniques by chemical pathologists at the National Health Laboratory Service Laboratory at the University of the Free State. Full blood counts were determined using the Roche Sysmex XT 2000i® analyser (Sysmex, 2014:Online), iron levels using the Beckman Coulter Synchron LX20 (Mikolaenko et al., 2000:387), and transferrin saturation and homocysteine levels using the BN Prospec System nephelometric technology. Serum folate levels were recorded using the Bayer Advia Centaur System (Hadler et al., 2008: S262).

Anaemia was diagnosed when haemoglobin levels were below 12.1g/dL (NHLS reference values). Iron deficiency was diagnosed when ferritin levels were below 15.0ng/mL while haemoglobin levels were still greater than 12.0g/dL; and iron deficiency anaemia was diagnosed when both ferritin and haemoglobin levels were below 15.0ng/mL and 12.0g/dL respectively (WHO, 2008:Online).

Statistical analysis was conducted using the Predictive Analytics SoftWare (PASW) Statistics Student version 22.0 by Statistical Package for the Social Sciences (SPSS) (IBM, 2013). Frequencies and percentages were determined for categorical data and medians and percentiles for continuous data were recorded. The Kruskal-Wallis test was used to compare medians of continuous and categorical variables in order to describe the associations between variables. The Chi-square test was also used to determine associations between categorical variables.

6.3 RESULTS

A large percentage of the women (47.0%) reported that they were current smokers at the time of the data collection, or had previously smoked. Almost three quarters (73.5%) of these women, had started smoking before the age of 20 years (Table 6.1). The majority of the women (70.7%) reported never using snuff, and most of those who did use snuff, used it three times or less per day (77.5%). The majority of the women were currently, or had formerly, used alcohol (71.5%), of which more than a third reported consuming more than five drinks per day at least once a month.

A relatively large percentage of women reported having experiencing breathlessness with usual activity (41.0%), loose stools/diarrhoea for at least three days (17.3%), vomiting (18.0%), loss of appetite (41.0%), blood in urine (7.5%) and involuntary weight loss of more than 3kg (42.5%), over the 6 months prior to data-collection. More than half of the women suffered from hypertension (56.4%).

Almost a third of women (64.2%) used medication regularly, with hypertension medication being used most often (45.1%). Almost one in five women (17.9%) had been hospitalised during the past 24 months.

More than half (54.1%) of women in the sample menstruated regularly with almost three quarters of the women (71.6%) reporting that they were currently, or had previously, used injectable contraceptives. None of the women in the current study made use of oral contraceptives and other methods of contraception were not asked. The majority of the women reported giving birth to three or fewer children (68.9%).

Table 6.1 Questions regarding reported health

Variable	n	%
Which best describes your smoking history?		
Never smoked	71	53.0
Currently smoke	39	29.1
Formerly smoked	24	17.9
If currently or formerly smoked, how many cigarettes per day?		
1	3	7.0
2	13	30.2
3	3	7.0
4	2	4.7
5	9	20.9
6	4	9.3
7	2	4.7
9	1	2.3
10	2	4.7
20	4	9.3

If currently or formerly smoked, at what age did you start?		
≤ 20	36	73.5
20–30	11	22.5
33	1	2.0
42	1	2.0
Which best describes your history of snuffing?		
Never used snuff	94	70.7
Currently use snuff	29	21.8
Formerly used stuff	10	7.5
If currently or formerly used snuff, how many times per day do you snuff?		
1	7	22.6
2	7	22.6
3	10	32.3
4	5	16.1
5	1	3.2
8	1	3.2
If currently or formerly used snuff, at what age did you start?		
≤ 20	9	28.1
21–30	3	9.4
31–40	15	46.9
41–48	5	15.6
Which best describe your history of alcohol use?		
Never used alcohol products	38	28.6
Currently use alcohol products	59	44.4
Formerly used alcohol products	36	27.1
At least once a month, do you consume more than 5 drinks per day?		
Yes	24	38.7
No	38	61.3
At what age did you start using alcohol?		
≤ 20	41	48.8
21–30	32	38.1
31–40	9	10.7
41–47	2	2.4
Have you experienced breathlessness in the last 6 months?		
Yes	55	41.0
No	79	59.0

Have you experienced loose stools / diarrhoea for at least 3 days in the last 6 months?		
Yes	23	17.3
No	110	82.7
Have you experienced vomiting in the last 6 months?		
Yes	24	18.0
No	109	82.0
Have you experienced appetite loss in the last 6 months?		
Yes	55	41.0
No	79	59.0
Have you experienced blood in your urine in the last 6 months?		
Yes	10	7.5
No	124	92.5
Have you experienced involuntary weight loss of more than 3kg in the last 6 months?		
Yes	57	42.5
No	77	57.5
Have you ever been diagnosed with diabetes?		
Yes	7	5.2
No	127	94.8
Have you ever been diagnosed with high blood pressure?		
Yes	75	56.4
No	58	43.6
Have you ever been diagnosed with stroke?		
Yes	7	5.2
No	127	94.8
Have you ever been diagnosed with heart disease/ angina/ heart attack?		
Yes	20	14.9
No	114	85.1
Have you ever been diagnosed with heart failure?		
Yes	1	0.7
No	133	99.3
Have you ever been diagnosed with cancer?		
Yes	1	0.7
No	133	99.3

Have you ever been diagnosed with liver disease?		
Yes	2	1.5
No	132	98.5
Have you ever been diagnosed with lung disease?		
Yes	15	11.2
No	119	88.8
Have you ever been diagnosed with tuberculosis?		
Yes	7	5.2
No	127	94.8
Have you ever been diagnosed with epilepsy?		
Yes	4	3.0
No	130	97.0
Have you ever been diagnosed with allergy?		
Yes	23	17.3
No	110	82.7
Are you taking medications regularly (i.e. at least once per week)?		
Yes	86	64.7
No	47	35.3
Types of medication:		
Allergies	3	2.0
Arthritis	10	6.5
Asthma pills	11	7.2
Asthma pump	3	2.0
Diabetes	7	4.6
Hypertension	69	45.1
Heart	3	2.0
Ointment for joints	4	2.6
Pain medication	23	15.0
Other	20	13.6
During the past 24 months, have you been hospitalised?		
Yes	24	17.9
No	110	82.1
If yes, how many times?		
1	18	72.0
2	6	24.0
3	1	4.0

If yes, give details (e.g. for specific operation or treatment):		
Cancer	2	8.0
Orthopaedics	6	24.0
Maternity	3	12.0
Breast problems	2	8.0
Gynaecology	2	8.0
Operation	3	12.0
Other	7	28.0
If yes, for how many days?		
≤ 10	18	72.0
> 10	7	28.0
Do you still have periods?		
Yes	72	54.1
No	61	45.9
Have you ever used an injectable contraceptive?		
Yes	96	71.6
No	38	28.4
How many live children have you given birth to?		
1	20	16.4
2	29	23.8
3	35	28.7
4	23	18.9
5	14	11.5
7	1	0.8

Only 4.6% of the women presented with low haemoglobin levels (Table 6.2). Overall, only 3.1% presented with low haematocrit, 3.1% with low mean corpuscular volume (MCV), and 7.6% low mean corpuscular haemoglobin (MCH) levels. A quarter of the women (25.2%) presented with elevated MCH levels. Overall, 12.5% of the women presented with low transferrin saturation and 1.4% with low ferritin levels. Overall, 4.6% of the women could be classified as having anaemia, 2.3% as being iron deficient and 0.7% as having iron deficiency anaemia.

Elevated homocysteine levels were found in 7.1% of the sample and 4.0% had low red cell folate levels.

Table 6.2 Description of the study population in terms of haemoglobin, haematocrit, MCV, MCH, transferrin saturation, ferritin, homocysteine and red cell folate levels

Blood samples	Normal reference value	N	Median	Low (%)	Normal (%)	High (%)
Haemoglobin	> 12.1 g/dL	130	13.8	4.6	95.4	-
Haematocrit	> 0.371 L/L	131	0.429	3.1	96.9	-
MCV	79.1–98.9 fl	131	93.8	3.1	77.1	19.8
MCH	27.0–32.0 pg/cell	131	30.4	7.6	67.2	25.2
Transferrin saturation	> 15%	74	26.1	12.2	87.8	-
Ferritin	6–120 ng/mL	74	94.0	1.4	58.1	40.5
Homocysteine	2.10–15.70 µmol/L	134	9.6	-	92.5	7.5
Red cell folate	> 372 nmol/L	131	575.3	3.8	96.2	-

Ampath. 2010. Ampath desk reference: guide to laboratory tests. Centurion: Ampath.

National Health Laboratory Services Reference values. 2014.

A significant association was found between median haemoglobin levels and smoking (*p*=0.001). Median haemoglobin levels were higher among those women who were smoking than those who did not. The median haemoglobin levels were lower among those women who did not experience breathlessness with usual activity, than those who did (*p*=0.027). Median haemoglobin levels were significantly lower among those women who menstruated regularly than those who did not (*p*=0.008). No statistically significant differences between the means for haemoglobin were seen of those women who were currently using, or who previously used, injectable contraceptives (Table 6.3).

Table 6.3 Associations between haemoglobin and other variables

Variables	n	Median blood values	p-value for median differences
Median haemoglobin levels across categories of history of smoking:			0.001*
Yes	63	14.2g/dL	
No	67	13.6g/dL	

Median haemoglobin levels across categories of history of using snuff:			0.226
Yes	37	13.7g/dL	
No	92	14.0g/dL	
Median haemoglobin levels between across categories of history of alcohol use:			0.296
Yes	92	13.9g/dL	
No	37	13.7g/dL	
Median haemoglobin levels across categories of breathlessness with usual activity:			0.027*
Yes	57	14.2g/dL	
No	76	13.7g/dL	
Median haemoglobin levels across categories of had loose stools/ diarrhoea for at least three days:			0.091
Yes	22	13.6g/dL	
No	107	13.9g/dL	
Median haemoglobin levels across categories of vomiting in the last 6 months:			0.106
Yes	24	14.4g/dL	
No	105	13.7g/dL	
Median haemoglobin levels across categories of loss of appetite in the last 6 months:			0.288
Yes	54	13.9g/dL	
No	76	13.8g/dL	
Median haemoglobin levels across categories of blood in urine in the last 6 months:			0.088
Yes	10	13.2g/dL	
No	120	13.9g/dL	
Median haemoglobin levels across categories of involuntary weight loss of more than 3kg:			0.886
Yes	56	13.9g/dL	
No	74	13.8g/dL	
Median haemoglobin levels across categories of diabetes:			0.051
Yes	7	14.6g/dL	
No	123	13.7g/dL	

Median haemoglobin levels across categories of hypertension:			0.081
Yes	74	14.1g/dL	
No	55	13.7g/dL	
Median haemoglobin levels across categories of stroke:			0.536
Yes	7	14.6g/dL	
No	123	13.8g/dL	
Median haemoglobin levels across categories of heart attack/ angina/ heart disease:			0.830
Yes	19	14.1g/dL	
No	111	13.7g/dL	
Median haemoglobin across categories of heart failure:			0.424
Yes	1	14.5g/dL	
No	129	13.8g/dL	
Median haemoglobin levels across categories of cancer:			0.090
Yes	1	9.9g/dL	
No	129	13.8g/dL	
Median haemoglobin levels across categories of liver disease/ hepatitis/ jaundice:			0.791
Yes	2	13.7g/dL	
No	128	13.8g/dL	
Median haemoglobin levels across categories of lung disease:			0.423
Yes	14	14.3g/dL	
No	116	13.8g/dL	
Median haemoglobin levels across categories of tuberculosis:			0.115
Yes	7	14.7g/dL	
No	123	13.8g/dL	
Median haemoglobin levels across categories of epilepsy:			0.599
Yes	4	14.1g/dL	
No	126	13.8g/dL	

Median haemoglobin levels across categories of allergy:			0.861
Yes	22	13.9g/dL	
No	107	13.8g/dL	
Median haemoglobin levels across categories of regular medication use:			0.384
Yes	84	13.9g/dL	
No	54	13.7g/dL	
Median haemoglobin levels across categories of hospitalisation in the past 24 months:			0.368
Yes	22	13.6g/dL	
No	108	13.8g/dL	
Median haemoglobin levels across categories of regular menstruation:			0.008*
Yes	70	13.7g/dL	
No	59	14.2g/dL	
Median haemoglobin levels across injectable contraceptive use categories:			0.847
Yes	94	13.8g/dL	
No	36	13.9g/dL	
Median haemoglobin levels across categories of live children given birth to:			0.384
1	19	14.0g/dL	
2	28	13.9g/dL	
3	34	13.7g/dL	
4	22	13.6g/dL	
5	14	13.6g/dL	
7	1	14.7g/dL	

* Statistically significant association

Only six women presented with haemoglobin levels below 12.1g/dL. No statistically significant difference was however found between symptoms of breathlessness in women with low haemoglobin levels compared to those who had normal haemoglobin levels ($\chi 2=0.676$, df=1, $p>0.01$) (Table 6.4).

Table 6.4 Association between symptoms of breathlessness and haemoglobin levels.

Variable	n	Breathlessness with usual activity	No breathlessness with usual activity
Low haemoglobin (<12.1g/dL)	5	1	4
Normal haemoglobin (≥12.1g/dL)	125	53	72

p=1.000

6.4 DISCUSSION

With this study we aimed to assess the reported health status, presence of anaemia and contraceptive use in a group of women aged 25–49 years living in low socio-economic townships in the rural Free State towns of Trompsburg, Philippolis and Springfontein. Due to the potential impact of improving women's health on societal growth and advancement (Izugbara & Covan, 2014:697), it was considered desirable to determine the need for intervention studies within this population.

A much larger percentage of women in the current study reported smoking than the 22.8% of women between 25–54 years in the South African National Health and Nutrition Examination Survey (SANHANES-1) (Shisana et al., 2013:102). A study conducted by Ayo-Yusuf & Omole (2008:64b) looking at women aged 25–70 years in South Africa found that 14.6% of the women in their study used snuff which is slightly lower than the findings of the current study where 29.3% of women used snuff.

As with smoking, the percentage of women in the current study who consumed alcohol (44.4%) at the time of data collection was higher than that of a South African national population-based survey conducted by Peltzer et al. (2011:33) that reported that 20.5% of women aged 25–34 years, 21.4% of women aged 35–44 years and 20.0% of women aged 45–54 years used alcohol. More women in the current study (38.7%) also reported binge drinking in the past month than compared to the Peltzer et al. (2011:33) study where 6.0% of women aged 25–34 years, 4.2% of women aged 35–44 years and 2.9% of women aged 45–54 years reported to binge drink in the past month.

Breathlessness with usual activity could be a symptom of anaemia (Bunn, 2011:1033) and conditions such as diarrhoea, vomiting, loss of appetite, blood in urine and involuntary weight loss could potentially, if experienced in the long term, contribute to the development of anaemia (Bayraktar & Bayraktar, 2010:2720). A relatively large percentage of women in the current study reported experiencing these symptoms.

Various disease states, like diabetes (Thomas et al., 2003:1164) and heart disease (Qaseem et al., 2013:770), as well as certain medication, have the potential to influence anaemia risk and, in turn, anaemia has the potential to influence the prognosis of various diseases (Qaseem et al., 2013:770; Sico et al., 2013:271; Foy & Labhasetwar, 2011:9155; Thomas et al., 2003:1164,). Relatively few women in the sample suffered from disease, apart from hypertension (56.4%). Of all the medications used, antihypertensive medications were also used most often. Antihypertensive medications have been reported to be associated with decreased haemoglobin levels which could develop due to haemodilution, haemolytic anaemia or the suppression of red blood cell production most often caused by angiotensin-converting enzyme inhibitors and angiotensin receptor blockers (Sica & Mannino, 2007:723). However, no significant associations were found in this study population between haemoglobin levels and any of the types of medications used.

The prevalence of anaemia among the women in the current study was much lower than the reported in the SANHANES-1 study, where the national prevalence of anaemia among women aged 25–34 years and women aged 35–44 years was 24.7% and 23.1%, respectively (Shisana et al., 2013:142). Similarly, a systematic review that determined the global trends in anaemia prevalence from 1995 to 2011 from population representative data, found that the prevalence of anaemia among non-pregnant women in South Africa, aged 15–49 years, decreased slightly from 33% in 1995 to 28% in 2011 (Stevens et al., 2013:e19).

Iron deficiency anaemia develops in three stages. During the first stage, depletion of iron stores occurs and is confirmed by low serum ferritin levels. The following stage is characterised by further decreases in serum ferritin with transferrin saturation decreasing as well. These decreases are followed by the development of microcytosis and hypochromia after which signs and symptoms of anaemia appear (Gaw et al., 2008:112). Only 1.5% of the

women in the current study presented with microcytic, hypochromic anaemia (low MCV and MCH levels) and 1.5% with normocytic, hypochromic anaemia (normal MCV and low MCH levels). A quarter of the women (25.2%) presented with macrocytic anaemia (elevated MCH levels). Also, a small percentage of women (1.4%) presented with low iron stores (low ferritin levels) which is a much lower prevalence than in the SANHANES-1 study where 13.5% of women aged 16–35 years had low ferritin levels (Shisana et al., 2013:162). These differences could be explained by the findings from a study conducted on non-pregnant women, aged 18–40 years, in Bangladesh, Chile, China, the Dominican Republic, Pakistan, Thailand and Tunisia in the period 1988–1992 where it was concluded that contraceptive use could have a beneficial effect on ferritin levels (UNDP/UNFPA/WHO/WB, 1998:261).

Only one woman (0.7%) in the sample presented with iron deficiency anaemia and two women (1.5%) with iron deficiency. Iron deficiency anaemia was found in 9.7% of women of reproductive age in South Africa in the SANHANES-1 study (Shisana et al., 2013:162). Low red cell folate levels were found in 3.8% of the women, which is much lower than the estimated prevalence of folate deficiency in 25–72% of women of reproductive age (Milman, 2011:370).

Differences in the prevalence of anaemia between the current study and others may be due to the large percentage of women in the current sample (45.1%) that did not menstruate regularly, as well as the fact that their median haemoglobin levels were significantly higher than those women who menstruated regularly. Furthermore, the high percentage of injectable contraceptive use can provide another explanation. Furthermore, other studies, including the SANHANES-1 study, did not report on menstruation and contraceptive.

A large percentage of the women in the current study were currently using, or previously made use of, injectable contraceptives (71.6%). A study conducted by Arias et al. (2006:237), concluded that the reduction of iron deficiency is one of the non-contraceptive benefits of using injectable contraceptives, specifically depot medroxyprogesterone acetate. A study among women aged 20–40 years from a low socio-economic backgrounds in Dhaka, Bangladesh found that women who did not use oral contraceptives had significantly lower levels of haemoglobin than those who did (Yeasmin et al., 2010:28). However, no significant

association was found between median haemoglobin levels and contraceptive use in the current study, which may have been influenced by the low percentage of women with anaemia.

Fewer women in the current study were in the younger age category of 25–34 years (20.1%) than in the older age category of 35–49 years (79.9%), which differs from the SANHANES-1 study, where almost equal numbers of women were included in the 25–34 years and 35–44 years groups (Shisana et al., 20113:161). These differences as well as the older median age of the women in the current study may also have resulted in a lower prevalence in anaemia in the current study as women of younger fertile age have a greater risk for developing anaemia, particularly due to iron deficiency, due to losses occurring during menstruation (Asres, Yemane & Gedefaw, 2014:Online).

An association was found between haemoglobin and smoking, with median haemoglobin levels being higher among those who smoked than those who did not. A meta-analysis on anaemia and cigarette smoking found that haemoglobin levels increase in healthy individuals who smoke, which is possibly facilitated by the exposure to carbon monoxide as it binds to haemoglobin, reducing its oxygen carrying capacity. The body compensates for the reduced oxygen delivery by maintaining a higher level of haemoglobin, resulting in a masking effect on the detection of anaemia (Leifert, 2008:177). According to the WHO (2011:Online), an adjustment of -0.3g/L can be made to the measured haemoglobin levels in persons who smoke.

An association between haemoglobin and breathlessness with usual activity was found as median haemoglobin levels were higher among those women who did experience breathlessness. No statistically significant association was however found when breathlessness with activity, were cross tabulated with anaemia (Table 6.4). The small number of women presenting with low haemoglobin levels may also have impacted on the significance. This may possibly indicate that the symptoms of breathlessness experienced by the women in the sample as a whole, could have been due to other reasons not investigated in the current study.

6.5 LIMITATIONS OF THE STUDY

The sample size may influence the representativity of the current study. However, we still believe that this study is important as it explored associations between variables that have not been looked at in South Africa, especially in the Free State.

Vitamin B12 levels were not measured in the AHA-FS data set, and even though we would have liked to include it in the current study, it is unlikely that these women would have suffered from a vitamin B12 deficiency as they did not follow a vegetarian diet and were not at risk for age-related achlorhydria.

6.6 CONCLUSION AND RECOMMENDATIONS

Results from the current study indicate that smoking could potentially mask the presence of anaemia which could worsen as if it goes undetected for a period of time. Overall, the women's risk for anaemia in terms of other health factors assessed in the current study seems to be low. Few women in the current study suffered from iron deficiency, iron deficiency anaemia, as well as folate deficiency. Possible reasons could be the older median age of the women, as well as the fact that almost half of the women in the sample did not menstruate regularly (possibly due to contraceptive use or menopause).

Significant associations were found between median haemoglobin levels in those women who were smoking and those who did not, as well as those women who menstruated regularly and those who did not. Thus indicating that those women who smoke and who menstruate regularly may have an increased risk for developing anaemia. An unexpected inverse association was found between median haemoglobin levels and those women who experienced breathlessness with usual activity, but could be due to other reasons not investigated in the current study.

Interventions directed at improving the health of these women that can form part of these services should thus be planned for this population. These interventions will mostly be in the form of nutrition education that can be provided at health care facilities in these areas,

as well as during home visits and should focus on improving the nutritional status of the women and so improving their health as well. These interventions should be culturally sensitive and adjusted to suit the specific needs of the women in this population. Furthermore, ensuring sustainability is of utmost importance as it impacts on health outcomes and has the potential to reduce the burden of disease in developing countries.

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CHAPTER 7

ANAEMIA PREVALENCE AND CONCOMITANT SOCIO-DEMOGRAPHY AMONG WOMEN IN THE RURAL FREE STATE, SOUTH AFRICA

ABSTRACT

Background: Access to basic services including water, electricity and sanitation are critical indicators of poverty which are often associated with standards of living and health. Poverty has the potential to influence risk for developing many diseases, including anaemia. Anaemia is a global public health problem that affects women living in both developed and developing countries. Determining the causal factors of anaemia can contribute to addressing the problem through appropriate interventions.

Objectives: To determine the socio demographic status, prevalence of anaemia, contraceptive use and associations between anaemia and socio demographic factors among rural women aged 25–49 years in the Free State province, South Africa.

Method: A cross-sectional descriptive study design was applied in a sample of 134 women. A socio-demographic questionnaire which assessed basic demographics of household members; structure of the house; household income; amenities; access to water and sanitation; employment status and cooking facilities was completed for each household. Questions pertaining to language, race, gender, age, employment status and income as well as type of dwelling were also included. Information related to water, sanitation, source of energy and food storage facilities was obtained in terms of household information. Blood samples were collected and analysed according to standard techniques. These included full blood counts, transferrin saturation, ferritin, homocysteine and red cell folate levels. Questions regarding contraceptive use were included in a questionnaire on reported health.

Results: The majority of the women (79.9%) fell in the older age group (35–49 years). Overall, 37.7% of the women were unemployed with only 21.6% of the women having a husband or partner who was a full time wage earner. One (44.0%) or two (39.6%) members contributed to the total household income which mostly ranged between R501–R1000 (33.6%) and R1001–R3000 (38.8%) per month. More than half of the women (52.7%) relied

on social grants as their main source of income. Most women lived in brick or concrete houses (75.4%), had access to electricity (92.5%), had their own tap for water (94.0%) and had a flush toilet (93.3%). Electricity was the fuel source mostly used for cooking (63.4%) and most of the houses had a working refrigerator or freezer (64.2%) and a working stove or hot plate (74.6%). A third of the women (33.5%) made use of cast iron pots for cooking. Only 0.7% of women suffered from iron deficiency anaemia (haemoglobin < 12.0g/dL; ferritin < 15.0ng/mL) and 1.5% had an iron deficiency (haemoglobin > 12.0g/dL; ferritin < 15.0ng/mL). Overall, 7.5% of the sample presented with elevated homocysteine levels, however, only 3.8% presented with low levels of red cell folate indicative of folate deficiency. Only 54.1% of the women reported that they regularly menstruate and 71.6% had currently or previously used injectable contraceptives. A significant association was found between median haemoglobin level and type of toilet, with those that had a flush toilet being more likely to have a higher median haemoglobin.

Conclusion: Even though most women had access to basic infrastructure, low levels of income and dependence on social grants as main source of income show that poverty was prevalent. The percentage of women with iron deficiency, iron deficiency anaemia as well as folate deficiency was low, probably due to the fact that half of the women did not menstruate. In the poorest households (with no flush toilet), women were more likely to have a lower median haemoglobin.

Keywords: Anaemia, socio demographics, folate, iron, iron deficiency

7.1 INTRODUCTION

South Africa is currently experiencing various transformations, the first being the struggle to overcome the burden of race, class and gender-based inequalities (Durrheim & Dixon, 2010:273). In addition, a nutrition transition is being experienced, with a large proportion of the South African population transitioning from a traditional to a more Western type of eating pattern, affecting health outcomes (Abrahams, Mchiza & Steyn, 2011:811).

Access to basic services including water, electricity and sanitation are critical indicators of poverty that impact on standards of living (Kehler, 2001:42). Poverty is often characterised

by sustained or chronic deprivation of resources, capabilities, choices, security and power needed to enjoy an adequate standard of living (UN, 2001:Online). Income is one of the key indicators of poverty (ODI: Poverty Briefings, 1999:2). Socio-economic rights that are mentioned in the Constitution of South Africa, include access to safe water, health care, sufficient food and water as well as social security (DoJ & CD, 2014:Online). Since 1994, the South African government has been providing poor South Africans (with a combined income of R3500 per month or less) with low cost housing, with the main aim of providing clean water, electricity, health care, sanitation and access to education. This low cost housing consists of simple single storey brick structures that are big enough to accommodate a single family (Greyling, 2009:Online). The Social Assistance Act has also been put in place for the realisation of social security in the form of grants for the elderly, children living in poverty, people with disabilities, children in need of foster care, and people in social distress (Mirugi-Mukundi, 2009:18).

At an individual level, poverty is associated with poor health outcomes, including an increased risk for iron deficiency (Ahnquist, Wamala & Lindstrom, 2012:938). It is suggested that a cycle exists between iron deficiency, diminished work capacity, work output and productivity, low income, and the subsequent risk of anaemia (Semba, 2003:S107). Anaemia is a global public health problem that affects women living in both developed and developing countries. It is generally assumed that iron deficiency can be attributed to 50% of anaemia cases worldwide. Estimating the prevalence of anaemia alone is of no value, unless related to action. Associations with the prevailing causal factors of anaemia in specified settings need to be determined in order for relevant interventions to be developed (WHO, 2008:Online).

The risk for anaemia may potentially be lowered by the use of contraceptives as the number of pregnancies and the time interval between pregnancies are reduced (UNDP/UNFPA/WHO/WB, 1998:261). Contraceptives also reduce menstrual blood loss which may further influence iron status (Yeasmin et al., 2010:25; UNDP/UNFPA/WHO/WB, 1998:262).

Various studies have been conducted in South Africa that relate to anaemia and socio demography; however, limited data are available regarding the prevalence of anaemia in Southern African women in general, and in the Free State province in particular, as well as the relationship between anaemia and prevailing socio demographic factors. This study thus aimed to determine the prevalence of anaemia and socio demographic factors that may potentially contribute to anaemia, as well as contraceptive use and associations between anaemia and socio demographic factors in women aged 25–49 years, in three rural towns in the Free State.

7.2 RESEARCH METHOD AND DESIGN

7.2.1 Research approach

The work reported on in this article formed part of the 'Assuring Health for All in the Free State' (AHA-FS) research programme, which is an epidemiological study with the aim of determining how living in rural and urban areas affects lifestyle and indicators of health. Trompsburg, Philippolis and Springfontein are included in the Free State Rural Development Partnership Programme (FSRDPP), and formed part of the rural phase of the AHA-FS study. As part of the data collection process, a multidisciplinary research team made use of standardised questionnaires to investigate socio-demographic status, household food security, dietary intake, levels of physical activity, knowledge, attitudes and practices relating to nutrition, and reported health of the study population. In addition to a medical examination, anthropometric measurements and blood specimens were also obtained for various investigations. For the purpose of this study, data from the rural phase of the original study were used and applied in a cross-sectional investigation.

The aim of this particular component of the study was to evaluate the socio-demography, prevalence of anaemia and contraceptive use in women between 25–49 years in these areas.

7.2.2 Population and sampling

All households in the low socio-economic townships (excluding farms) in the rural towns of Trompsburg, Philippolis and Springfontein in the Free State were eligible to participate, due to transitions in lifestyle that are experienced by low socio-economic communities. The final sample consisted of 134 women aged 25–49 years who were HIV-uninfected, who gave informed consent, and who were not pregnant at the time of data collection.

7.2.3 Measuring techniques and procedures

A socio-demographic and household questionnaire was completed for each household. This questionnaire included basic demographics of household members; structure of the house; household income; amenities; access to water and sanitation; employment status and cooking facilities. Questions pertaining to language, race, gender, age, employment status and income as well as type of dwelling were also included. Information regarding water, sanitation, source of energy and food storage facilities was also obtained.

Information was obtained from one adult household member in a structured interview conducted by final year students from the Department of Nutrition and Dietetics. A trained Sotho-speaking interviewer assisted in cases where participants were unable to understand English or Afrikaans.

For the purpose of this study, fasting blood samples were used to measure full blood count, serum ferritin levels and transferrin saturation, homocysteine and red cell folic acid levels in all participants. Blood samples were collected in the morning by medical doctors after an overnight fast. After collection, these samples were immediately stored in ice-filled containers and transported to the National Health Laboratory Service Laboratory at the University of the Free State where they were analysed according to standard techniques. Full blood counts were determined using the Roche Sysmex XT 2000i® analyser (Sysmex, 2014:Online), iron levels using the Beckman Coulter Synchron LX20 (Mikolaenko et al., 2000:387), and transferrin saturation and homocysteine levels using the BN Prospec System

nephelometric technology. Serum folate levels were recorded using the Bayer Advia Centaur System (Hadler et al., 2008: S262).

Anaemia was diagnosed when haemoglobin levels were less than 12.1g/dL (NHLS reference values). The World Health Organization's (WHO) cut off values were used for the diagnosis of iron deficiency and iron deficiency anaemia: iron deficiency is diagnosed when ferritin levels are below 15.0ng/mL and haemoglobin levels are still greater than 12.0g/dL; and iron deficiency anaemia is diagnosed when both ferritin and haemoglobin levels are below 15.0ng/mL and 12.0g/dL respectively (WHO, 2008:Online).

As part of the reported health questionnaire, participants had to answer questions relating to menstruation and contraceptive use.

Prior to data collection, field workers visited all the households in the three areas to explain the purpose and procedures of the study to adult members and to invite those between the ages of 25-64 years to participate on a given date. Data was obtained from the participants at the community halls in the rural areas. In order ensure that the participants met the inclusion criteria for age, ID documents were screened. Participants were required to rotate between different research stations in the venue until the necessary data had been collected.

Ethical approval was obtained from the Ethics Committee of the Faculty of Health Sciences of the University of the Free State (ETOVS number 21/07), the Department of Health and local municipalities. In order to ensure confidentiality of the data, each respondent was assigned a specific number. Participation was voluntary and participants could withdraw at any time.

7.2.4 Statistical analysis

Statistical analysis was conducted using the Predictive Analytics SoftWare (PASW) Statistics Student version 22.0 by Statistical Package for the Social Sciences (SPSS) (IBM, 2013). Frequencies and percentages were determined for categorical data and medians and

percentiles for continuous data were recorded. Since only five out of the 134 women in the sample suffered from anaemia (haemoglobin<12.0g/dL), it was difficult to determine associations between those who had low haemoglobin levels and those who had normal levels. It was thus decided to use the Kruskal-Wallis test in order to compare medians of continuous and categorical variables.

7.3 RESULTS

After applying the inclusion and exclusion criteria, 134 women could be included in the sample. These women had a median age of 41 years and most (79.9%) were between 35–49 years of age (Table 7.1). The majority of the women (61.2%) were of black ethnicity and 40.3% of the women spoke Afrikaans. Overall, 38.1% of the women were married. Approximately three quarters (75.4%) of the women lived in brick or concrete houses. Most of the women in the sample had access to their own tap (94.0%) for drinking water and had a flush toilet (93.3%) at their house.

The majority of the households had electricity (92.5%) and electricity was also mostly used for cooking (63.4%). Almost two thirds (66.4%) of the women never used cast iron pots for cooking. Overall, 64.2% of the households had a refrigerator or freezer, 74.6% had a stove or hot plate, 47.0% had a primus or paraffin stove, and 18.3% had a microwave; all in working condition.

Overall, 37.3% of the women were unemployed, with only 21.6% having a husband or partner who was a full time wage earner. With regard to monthly income, 91.1% of households had a monthly income of R3000 or less. In most cases (76.1%), this income was the same as during the past six months and mostly came from government grants (52.7%). In most of the households, between one (44.0%) and two (39.6%) members in the household contributed to the total income.

Table 7.1 Socio demographic and household information

Variable	N	%
Race of the family		
Black	82	61.2
Coloured	30	22.4
Mixed	22	16.4
Age		
25–34 years	27	20.1
35–49 years	107	79.9
First language of household		
Sotho	38	28.4
English	1	0.7
Afrikaans	54	40.3
Other	41	30.6
Marital status:		
Never married	28	20.9
Currently married / traditional marriage	51	38.1
Living with partner	22	16.4
Widowed	15	11.2
Separated	14	10.4
Divorced	4	3.0
Employment status		
Housewife by choice	5	3.7
Unemployed	50	37.3
Self-employed	4	3.0
Full time wage earner	13	9.7
Other	62	46.3
Husband/ partner's employment status		
Retired by choice	4	3.0
Unemployed	10	7.5
Full time wage earner	29	21.6
Other	32	23.9
Not applicable	59	44.0
Type of dwelling		
Brick, concrete	101	75.4
Corrugated iron	32	23.9
Other	1	0.7

Where do you get drinking water most of the time?		
Own tap	126	94.0
Communal tap	7	5.2
Other	1	0.7
What type of toilet does this household have?		
Flush	125	93.3
Pit	1	0.7
Bucket, pot	5	3.7
Other	3	2.2
Does the household have electricity?		
Yes	124	92.5
No	10	7.5
What fuel is used for cooking most of the time?		
Electricity	85	63.4
Gas	8	6.0
Paraffin	38	28.4
Wood, coal	3	2.2
Do you use a cast iron pot for cooking?		
Never	89	66.4
Less or equal to once a week	31	23.1
More than once a week	9	6.7
Every day	5	3.7
Does the home have a working refrigerator/freezer?		
Yes	86	64.2
No	48	35.8
Does the home have a working stove/hot plate?		
Yes	100	74.6
No	34	25.4
Does the home have a working primus/paraffin stove?		
Yes	63	47.0
No	71	53.0
Does the home have a working microwave?		
Yes	25	18.7
No	109	81.3

How many people contribute to the total income?		
0	4	3.0
1	59	44.0
2	53	39.6
3 or more	18	13.4
Household income per month		
None	2	1.5
R100-R500	23	17.2
R501-R1000	45	33.6
R1001-R3000	52	38.8
R3001-R5000	8	6.0
Over R5000	1	0.7
Don't know	3	2.2
What is your main source of income?		
Wages and salaries from formal employment	19	14.5
Self-employment	3	2.3
Casual employment	19	14.5
Crop production and livestock sales	0	0
Sale of assets	0	0
Land/ flats/ equipment rental	0	0
Old age pension or state grant	69	52.7
Domestic work	1	0.8
Other	20	15.3
Is this income more or less than the income that you had over the past six months?		
More	9	6.7
Less	23	17.2
The same	102	76.1

Low haemoglobin levels were present in only 4.6% of the women (Table 7.2), with only 3.1% presenting with low haematocrit levels, 3.1% with low mean corpuscular volume (MCV) levels, and 7.6% with low mean corpuscular haemoglobin (MCH) levels. Elevated MCH levels were present in 25.2% of the sample.

Table 7.2 Haemoglobin, haematocrit, MCV, MCH, transferrin saturation, ferritin, homocysteine and red cell folate levels

Blood samples	Normal reference value	N	Median	Low (%)	Normal (%)	High (%)
Haemoglobin	> 12.1 g/dL	130	13.8	4.6	95.4	-
Haematocrit	> 0.371 L/L	131	0.429	3.1	96.9	-
MCV	79.1–98.9 fl	131	93.8	3.1	77.1	19.8
MCH	27.0–32.0 pg/cell	131	30.4	7.6	67.2	25.2
Transferrin saturation	> 15%	74	26.1	12.2	87.8	-
Ferritin	6–120 ng/mL	74	94.0	1.4	58.1	40.5
Homocysteine	2.10–15.70 µmol/L	134	9.6	-	92.5	7.5
Red cell folate	> 372 nmol/L	131	575.3	3.8	96.2	-

Ampath. 2010. Ampath desk reference: guide to laboratory tests. Centurion: Ampath.

National Health Laboratory Services Reference values. 2014.

Low transferrin saturation were identified in 12.5% of the sample, with only 1.4% of the women presenting with low ferritin levels. Only 4.6% of the women presented with anaemia, 2.3% with iron deficiency and 0.7% with iron deficiency anaemia. Elevated homocysteine levels were identified in 7.1% of the sample and 4.0% had low red cell folate levels. Almost half (45.9%) of the women did not menstruate regularly and 71.6% were currently or had previously made use of injectable contraceptives (Table 7.3).

Table 7.3 Menstruation patterns and contraceptive use

Questions	N	Yes (%)	No (%)
Still menstruated regularly	133	54.1	45.9
Currently or previously used injectable contraceptives	134	71.6	28.4

A significant association was found between median haemoglobin levels and whether the women were still menstruating or not, with median haemoglobin levels being lower in those women who menstruated regularly compared to those who did not (Table 7.4).

Table 7.4 Associations between haemoglobin, menstruation and contraceptive use

Variables	n	Median blood value	p-value for median differences
Median haemoglobin levels across categories of menstruation:			0.008*
Menstruated regularly	70	13.7g/dL	
Did not menstruate regularly	59	14.2g/dL	
Median haemoglobin levels across categories of contraceptive use:			0.847
Made use of injectable contraceptives	94	13.8g/dL	
Did not use injectable contraceptives	36	13.9g/dL	

* Statistically significant association

As far as indicators of socio-demography are concerned, a significant association was found between median haemoglobin level and the type of toilet system available, with median haemoglobin levels being higher among those who had flush toilets compared to those who used the bucket system (Table 7.5). None of the other indicators of socio-economic status were significantly associated with haemoglobin levels.

Table 7.5 Associations between median haemoglobin levels and socio-demographic variables

Variables	n	Median blood values	p-value for median differences
Median haemoglobin levels across categories of marital status:			0.279
Never married	27	13.7g/dL	
Currently married/ traditional marriage	50	13.8g/dL	
Living with partner	20	13.6g/dL	
Widowed	15	14.2g/dL	
Separated	14	13.7g/dL	
Divorced	4	13.0g/dL	

Median haemoglobin levels across categories of respondent's employment status:			0.957
Housewife by choice	5	13.5g/dL	
Unemployed	48	13.8g/dL	
Self employed	4	13.6g/dL	
Full time wage earner	12	13.6g/dL	
Other	61	14.0g/dL	
Median haemoglobin levels across categories of husband/partner's employment status:			0.299
Retired by choice	4	14.7g/dL	
Unemployed	9	13.6g/dL	
Full time wage earner	28	14.0g/dL	
Other	31	13.7g/dL	
Not applicable	58	13.8g/dL	
Median haemoglobin levels across categories of type of dwelling:			0.187
Brick, concrete	98	14.0g/dL	
Corrugated iron	31	13.6g/dL	
Other	1	13.1g/dL	
Median haemoglobin levels across categories of sources of drinking water:			0.345
Own tap	122	13.8g/dL	
Communal tap	7	13.7g/dL	
Other	1	12.5g/dL	
Median haemoglobin levels across categories of type of toilet:			0.025*
Flush	121	13.8g/dL	
Pit	1	14.5g/dL	
Bucket, pot	5	12.8g/dL	
Other	3	12.5g/dL	
Median haemoglobin levels between women who had access to electricity and those who did not:			0.738
Yes	121	13.8g/dL	
No	9	13.7g/dL	
Median haemoglobin levels across categories of fuel sources for cooking:			0.327
Electric	82	14.1g/dL	
Gas	8	14.2g/dL	
Paraffin	37	13.6g/dL	
Wood, coal	3	14.2g/dL	

Median haemoglobin levels between women who used cast iron pots for cooking and those who did not:			0.341
Never	86	13.7g/dL	
Less than once a week	31	14.2g/dL	
More than once a week	8	13.3g/dL	
Every day	5	14.2g/dL	
Haemoglobin levels between women who had a working refrigerator/ freezer and those who did not:			0.774
Yes	84	13.9g/dL	
No	46	13.7g/dL	
Median haemoglobin levels between those women who had a working stove/ hot plate and those who did not:			0.468
Yes	97	13.9g/dL	
No	33	13.7g/dL	
Median haemoglobin levels between women who had a working primus/ paraffin stove and those who did not:			0.184
Yes	61	13.7g/dL	
No	69	14.2g/dL	
Median haemoglobin levels across categories of number of people contributing to the total income:			0.248
0	4	13.2g/dL	
1	57	14.0g/dL	
2	51	13.7g/dL	
3	12	14.1g/dL	
4	5	13.7g/dL	
6	1	13.4g/dL	
Median haemoglobin levels across categories of household monthly income per month:			0.085
None	2	13.6g/dL	
R100-R500	21	13.8g/dL	
R501-R1000	44	14.1g/dL	
R1001-R3000	52	13.9g/dL	
R3001-R5000	7	12.9g/dL	
Over R5000	1	14.4g/dL	
Don’t know	3	12.6g/dL	

Median haemoglobin levels across categories of sources of income:			0.638
Wages and salaries from formal employment	21	14.3g/dL	
Self-employment	3	14.7g/dL	
Casual employment	19	13.6g/dL	
Old age pension or state grant	67	13.8g/dL	
Domestic work	1	12.7g/dL	
Other	19	13.7g/dL	
Median haemoglobin levels across categories of changes in monthly income over the past 6 months:			0.434
More	8	13.9g/dL	
Less	23	13.8g/dL	
The same	99	13.8g/dL	

* Statistically significant association

7.4 DISCUSSION

According to the 2011 Census, 77.6% of South Africans lived in formal dwellings and 89.1% of the population in the Free State province had access to tap water inside their dwelling or yard at that time (Statistics South Africa, 2012:Online). These results are similar to findings from the current study where 75.4% of the women lived in brick/concrete houses and 94.0% had access to their own tap for drinking water. About 3 out of 4 women in the current study (74.6%) had a working stove or hot plate which was similar to that of the 2011 Census, where 77.0% of South African households had a working electric or gas stove (Statistics South Africa,2012:Online).

The unemployment rate of women in the current study (37.3%) was the same as that reported in the 2011 Census, (the Free State province had an unemployment rate of 32.6%). Similar findings were observed between the 38.8% of the women in the current study, who had a monthly income between R1001–R3000, and the Free State population as a whole in the SANHANES-1 study, where 40.0% had a monthly income between R801–R3200 (Shisana et al., 2013:68). In the current study, 52.3% of households had a monthly income less than R1000 which is comparable to the results of a study conducted by Oldewage-Theron et al., (2006:798) in an informal settlement in the Vaal Triangle, South Africa where 58.3% of households had a monthly income less than R1000.

A study conducted by Singh (2013:283) that looked at nationally representative data collected between 2005–2006 among women in India aged 15–49 years, found that women who were married had more severe anaemia compared to those who were either divorced, widowed, separated, and not living together. Although these results differ from the current study where no significant association was found, this may be due to the low prevalence of anaemia in the current study.

The majority of the households in the current study had access to electricity (92.5%) and 89.8% made use of electricity for cooking. The results of the 2011 Census reported that a smaller percentage of Free State households made use of electricity for cooking at 63.4% (Statistics South Africa, 2012:Online). Overall, 93.3% of the women in the current study had a flush toilet at their house, which was higher than the 57.0 % of South Africans reported during the 2011 Census (Statistics South Africa, 2012:Online). The second most used fuel source for cooking in the current study, was paraffin (28.4%), which was only used by 8.5% of the South African according to the 2011 Census (Statistics South African, 2012:Online).

According to the Census 2011, the average annual household income in the Free State was R75 312 (Statistics South Africa, 2012:Online), which is higher than the findings of the current study where only 0.7% of households had a monthly income of more than R5000 (i.e. more than R60 000 annually). The SANHANES-1 study reported that 36.4% of households in the Free State province received no monthly income (Shisana et al., 2013:68), compared to only 1.5% in the current study.

The prevalence of general anaemia as defined by low haemoglobin levels among the women in the current study was much lower than that reported in other South African studies. The prevalence of anaemia in the SANHANES-1 study among women aged 25–34 years and women aged 35–44 years was 24.7% and 23.1% respectively (Shisana et al., 2013:142). Stevens et al. (2013:e19) conducted a systematic review on population representative data and found that the prevalence of anaemia among non-pregnant women in South Africa, aged 15–49 years, decreased slightly from 33% in 1995 to 28% in 2011. These differences may be due to the fact that the above mentioned studies did not consider menstruation or

contraceptive use, making it difficult to compare the findings. Almost half of the women in the current study did not menstruate regularly and almost three quarters were currently using, or had previously used injectable contraceptives (Table 4.3). As expected, women who did not menstruate regularly were more likely to have higher haemoglobin levels (Table 7.4) Similar results was reported by Yeasmin et al. (2010:28) among women aged 20–40 years from a low socio-economic-background in Dhaka, Bangladesh, where women who used contraceptives had significantly higher haemoglobin levels than those who did not.

Another possible reason for the differences in the prevalence of anaemia could be the older median age (41 years) of the women in the current study. Fewer women (20.1%) fell in the younger age group (25–34 years) than in the older age group (79.9%) (35–49 years) when compared to the SANHANES-1 study where almost equal numbers of women were included in the 25–34 years and 35–44 years groups (Shisana et al., 20113:161). Women of younger fertile age have a greater risk for developing anaemia, particularly due to iron deficiency, as a result of losses occurring during menstruation (Asres, Yemane & Gedefaw, 2014:Online).

Iron deficiency anaemia develops in three stages. Depletion of iron stores occurs during the first stage and is confirmed by low serum ferritin levels. During the following stage, serum ferritin decreases further with transferrin saturation decreasing as well. These decreases are followed by the development of microcytosis and hypochromia after which signs and symptoms of anaemia appear (Gaw et al., 2008:112). Only 1.5% of the women in the current study presented with microcytic, hypochromic anaemia (low MCV and MCH levels) and only 1.5% of the women presented with hypochromic anaemia with normal MCV values. Research has shown that the use of iron post for cooking may have the potential to reduce the risk for iron deficiency anaemia (Charles, 2012:121). Although a third of the women in the current study used cast iron pots, a significant association could not be demonstrated between median haemoglobin levels and cast iron pot use. A quarter (25.2%) of the women had increased levels of MCH, which is seen in macrocytic anaemia. Ferritin levels below the reference ranges were only present in 1.4% of the women which is much lower than the results of the SANHANES-1 study, where 13.5% of women aged 16–35 years had low ferritin levels (Shisana et al., 2013:162). A study conducted on non-pregnant women, aged 18–40 years, in Bangladesh, Chile, China, the Dominican Republic, Pakistan,

Thailand and Tunisia in the period 1988–1992 concluded that contraceptive use has a beneficial effect on ferritin levels (UNDP/UNFPA/WHO/WB, 1998:261) and could thus serve as a possible explanation for the above mentioned differences.

Iron deficiency and iron deficiency anaemia was present in only 0.7% and 1.5%, respectively, which was much lower than the 9.7% in women of reproductive age in the SANHANES-1 study that had iron deficiency (Shisana et al., 2013:162). Low red cell folate levels were found in 3.8% of the women, which is also much lower than the estimated prevalence of folate deficiency in 25–72% of women of reproductive age (Milman, 2011:370). The mandatory food fortification programme of staple foods which have been in place since 2003, and included iron and folate may also have contributed to the low prevalence.

Similar to the results of the current study, a study conducted by Haverkate et al. (2013:6), using data of women aged 15–49 years from demographic and health surveys conducted from 2003–2010 from 21 different African countries, found a positive association between haemoglobin levels and the presence of a toilet facility. A study looking at the household environment and the prevalence of anaemia among children under 5 years of age in India also reported poor toilet facilities were associated with iron deficiency anaemia (Baranwal et al., 2014:1). Such households would be considered the poorest of the poor. Poor sanitation may lead to parasitic infections, which could potentially cause anaemia due to chronic blood loss (Ardhapurkar & Nalawade, 2010:212).

Despite relatively good infrastructure and facilities in most households, there were households that did not have access to basic requirements such as flush toilets. In these households the risk of low haemoglobin was higher.

7.5 LIMITATIONS OF THE STUDY

The small sample size in the current study may cast doubts on representativity. The older median age of the women, as well as the fact that almost half of the women in the sample did not menstruate regularly, may have influenced the ability to determine associations.

We believe that this study is however still of value, since it provides an overview of the situation in the rural Southern Free State, that has not been documented before.

Initial planning of the study included the evaluation of vitamin B12 levels, however, this parameter was not measured in the AHA data set. Even though the possibility exists that the remainder of the sample with elevated homocysteine levels in the absence of elevated red cell folate levels may possibly suffer from a vitamin B12 deficiency, this is unlikely since the women do not follow a vegetarian diet and are not at risk for age-related achlorhydria.

7.6 CONCLUSION AND RECOMMENDATIONS

Overall, the results of the current study show that poverty was prevalent in this sample, even though most of the women had access to basic infrastructure. Despite this, occurrence of iron deficiency, iron deficiency anaemia as well as folate deficiency in the sample was low. The significant association that was found between haemoglobin and type of toilet facilities available could indicate that households that could be considered to be the poorest of the poor with improper sanitation have an increased risk for anaemia.

The results of this research provide a better understanding of the underlying factors associated with poverty in this community. The focus of intervention studies within these communities should be to improve employment opportunities in an effort to reduce poverty. Health promotion programmes with the aim of teaching skills and improving knowledge should also be implemented. Education should focus on teaching women to make the best choices for their health with the resources that they have available. The implementation of food gardens that could serve as a source of income generation may also help to supplement the existing income of the households.

“Eradicate extreme poverty and hunger” and “improve maternal health” are two of the Millennium Development Goals to be achieved by the year 2015. The most noticeable gap between rich and poor is seen with maternal health. In order to bridge this gap and achieve these goals, quality reproductive health services, accompanied by a series of well-timed

interventions, should be implemented (UN, 2010:30) in order to improve the health of the women in the current study.

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CHAPTER 8

CONCLUSIONS AND RECOMMENDATIONS

8.1 INTRODUCTION

This research study investigated nutritional, social and health factors that could potentially contribute to the development of anaemia in low socio-economic women in the rural towns of Trompsburg, Philippolis and Springfontein. As stated by the World Health Organization (WHO) (2008:Online), "determining the prevalence of anaemia will only be useful if associations between anaemia and the prevailing causal factors in specified setting are determined". Accurate information regarding these causal factors needs to be collected in order for appropriate interventions to be developed (WHO, 2008:Online), and thus served as the motivation for the current study. The conclusions that stemmed from the current study will be discussed according to prevalence of anaemia in the sample and each of the four main aims, namely dietary diversity and anaemia, anthropometric variables and anaemia, reported health and anaemia and socio-demography and anaemia.

8.2 PREVALENCE OF ANAEMIA IN THE SAMPLE

Nutritional anaemias develop as a result of insufficient bioavailable haemopoietic nutrients such as iron, vitamin B12 and folate (Balarajan et al., 2011:2127). These anaemias manifest as iron deficiency anaemia and megaloblastic anaemia (due to either folate or vitamin B12 deficiency). Even though the concentration of haemoglobin is the most commonly used

indicator of anaemia, it lacks specificity (Balarajan et al., 2011:2126). Additional tests are thus required to determine the type of anaemia.

Overall, 4.6% (6/134) of the women in the sample presented with anaemia. Occurrence of iron deficiency anaemia (0.7%) and iron deficiency (1.5%) among the women was low. Elevated homocysteine levels were present in 7.5% of the sample with only 3.8% presenting with low red cell folate levels indicative of folate deficiency. More than half of the women (54.1%) reported that they menstruated regularly and 71.6% had currently or previously used injectable contraceptives. Women who menstruated regularly had significantly lower median haemoglobin levels than those who did not.

The low prevalence of anaemia could be attributed to the fact that almost half of the women did not menstruate and the median age was older than that in other studies conducted on women of childbearing age. The mandatory fortification of certain staple foods could also have contributed to the low prevalence of anaemia seen in the sample.

8.3 DIETARY DIVERSITY AND ANAEMIA

The women in the current study consumed a diet with moderate variety that consisted of starchy staple foods, with 51.5% having a medium dietary diversity score. Quite a large number of women had a low dietary diversity score (44.7%). More than three quarters of the women (76.9%) consumed flesh meats and fish which are good sources of the more bioavailable haem iron, however, only a quarter (25.4%) of the women ate dark green leafy vegetables which are sources of non-haem iron as well as folate. All the women consumed starchy foods, some of which are sources of folate and iron due to their mandatory fortification. Significant associations between median MCV and MCH levels and dietary diversity score were identified, with median MCV and MCH levels increasing as dietary diversity scores decreased, possibly indicating that the mandatory food fortification programme most probably had a positive impact on the nutritional status of these women.

8.4 ANTHROPOMETRIC VARIABLES AND ANAEMIA

A predominant pattern of malnutrition, characterised by overweight (25.4%) and obesity (45.4%), high rates of abdominal obesity (79.2%) and unhealthy body fat percentages (86.2%), was observed among the women in the current study. Medians for BMI (28.7kg/m^2), waist circumference (90.8cm) and body fat percentage (38.8%) were also within the unhealthy ranges. These women were thus at high risk for developing chronic diseases of lifestyle due to obesity. Significant associations between BMI, waist circumference and body fat percentage categories and MCV, MCH levels and transferrin saturation indicate that obesity is associated with an increased risk for iron deficiency.

8.5 REPORTED HEALTH AND ANAEMIA

Overall, the women's risk for anaemia in terms of health factors assessed in this study, seemed to be low, except menstruation. Almost half of the women (47.0%) in the current study smoked and more than a quarter (29.3%) used snuff. Almost three quarters of the women used alcohol (71.5%), with 38.7% having consumed more than five drinks per day, at least once a month. A relatively large percentage of women reported breathlessness with usual activity (41.0%), loose stools/ diarrhoea for at least three days (17.3%), vomiting (18.0%), loss of appetite (41.0%), blood in their urine (7.5%) and involuntary weight loss of more than 3kg (42.5%) within the past six months.

More than half of the women suffered from hypertension (56.4%). Medication was used by 64.2% on a regularly basis, with hypertension medication being used the most (45.1%). More than half (54.1%) of the women in the sample still menstruated regularly, with 71.6% currently or previously used injectable contraceptives and 68.9% having given birth to three or fewer children.

Results from the current study indicate that smoking could potentially mask the presence of anaemia as a significant association was found between median haemoglobin levels of those women who smoked and those who did not, where median haemoglobin levels where higher among women who smoked compared to those who did not.

A significant and expected association was found between median haemoglobin levels between those women who menstruated regularly and those who did not, indicating that women who menstruate have an increased risk for anaemia. Median haemoglobin levels were significantly higher among those women who experienced breathlessness with usual activity, but did not differ significantly between women who suffered from anaemia and those who did not, which could indicate that the breathlessness was due to other reasons not investigated in the current study.

8.6 SOCIO-DEMOGRAPHY AND ANAEMIA

The results of the current study show that poverty was prevalent among the women included in the study, even though most of the women had access to basic infrastructure, such as living in a brick/ concrete house (75.4%), access to electricity (92.5%) and own tap for drinking water (94.0%). Despite relatively good infrastructure, few women earned a full time wage (9.7%), with only 21.6% of the women having a husband/ partner who earned a full time wage. The total household income was low and mostly ranged between R501–R1000 (33.6%) and R1001–R3000 (38.8%) per month and more than half of the women (52.7%) relied on social grants as their main source of income.

A significant association was found between haemoglobin and type of toilet facility available, where median haemoglobin levels were higher among those who used flush or pit toilets thatn those who used the bucket/ pot or any other system, which could indicate that women from households that could be considered to be the poorest of the poor with improper sanitation had an increased risk for anaemia.

8.7 RECOMMENDATIONS

8.7.1 Recommendations to address anaemia and other health issues

According to Balarajan et al. (2011:2131), strategies for anaemia prevention and control should include the improvement of dietary intake and food diversifications, the fortification of food, micronutrient supplementation, disease control and education. Findings from the current study support these recommendations.

Although the prevalence of anaemia in the current study was relatively low, the underlying causes of inappropriate dietary and lifestyle practices that were identified in the current study need to be addressed. These included high prevalence of smoking, snuff use and alcohol consumption; low variety and intake of iron and folate rich foods in the women's diets as well as high prevalence of overweight and obesity. The results of this study will be used to plan and implement sustainable community-based interventions, mostly though home visits, within these communities. These interventions should take cultural sensitivity and sustainability into consideration and should be adjusted to suit the exact needs of the women in this study.

The focus of interventions within these communities should be on improving employment opportunities, reducing poverty, improving dietary adequacy, changing perceptions regarding overweight and obesity and promoting physical activity in order to improve the nutritional status and in turn the health of women.

One strategy that should form part of these interventions, that has been suggested by Oldewage-Theron, Dicks & Napier (2006:803), is the promotion of household food gardens. The aim of these gardens is to increase the consumption of vegetables in order to promote higher micronutrient intakes that may help to protect against micronutrient deficiencies associated with anaemia. Appropriate nutrition education regarding bioavailability issues of the nutrients in vegetables should also be provided. Vegetables can also be sold to contribute to the household income.

Promotion of nutrition education has been suggested by various researchers in the literature (Balarajan et al., 2011:2131; Upadhyay et al., 2011:34; Oldewage-Theron, Dick & Napier, 2006:80) as a strategy to promote better food choices and healthier lifestyles in order to prevent health problems. The findings from the current study support these recommendations. Nutrition education should be tailored to specifically address the needs of individuals in poor communities.

Balarajan et al. (2011:2131) have recommended that investment in the quality of community health workers and increasing community awareness and social acceptability of these community-based interventions will help to achieve the desired results. Even though the current study did not assess any of these aspects, it is still important to ensure that health workers and members of the community are involved in the planning and implementation of the interventions in these areas in order to ensure success. Health workers and members of the community can play a valuable role in ensuring interventions are relevant and culturally appropriate.

Even though the prevalence of anaemia in the current study was low, the importance of interventions to reduce the risk of anaemia cannot be disregarded. Efforts directed towards reducing the burden of disease in developing communities in South Africa have the potential to contribute to sustainable livelihoods and improved health outcomes.

8.7.2 Recommendations for further research

The strategies mentioned earlier, in addition to already established nutrition intervention strategies in these communities would benefit from further research.

According to Balarajan et al. (2011:2132), information related to the different types of anaemia remains necessary as the contribution of the different causes of anaemia and the prevalence of the different types of anaemia in different settings remains unclear. It is thus recommended that studies consider including a wider range of parameters that will give a better indication of the different types of anaemia, i.e. iron deficiency anaemia, megaloblastic anaemia associated with folate or vitamin B12 deficiency, as haemoglobin on its own does not provide enough information.

A study conducted by Mujica-Coopman et al. (2014:Online), recommended that more robust designs are needed to determine the cause-effect between iron status and obesity (and other health outcomes), particularly in groups that are vulnerable to anaemia. These authors also recommended that various biomarkers of iron, such soluble transferrin

receptor, should be included in further research, a recommendation that is supported by the current study (Mujica-Coopman et al., 2014:Online).

Other recommendations that stem from the limitations of the current study include:

- the inclusion of a larger group of women, with different ages, so as to increase the sample size to improve representativity; and
- the measuring of vitamin B12 levels in order to determine the prevalence of B12 deficiency in combination with folate deficiency.

8.8 RESEARCH SIGNIFICANCE

The incidence and impact of anaemia has been discussed in detail in the literature. The significance of this study was that various contributing factors were investigated and associations with other factors were determined in order to identify factors associated with anaemia in these women. The current study identified associations between poverty and anaemia, smoking and anaemia as well as adiposity and anaemia. The current study also confirmed that women who menstruated regularly are at increased risk for anaemia.

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SUMMARY

Anaemia, a global public health problem that particularly affects women, holds major consequences for human health. For this reason, the factors that play a role in the development of anaemia need to be identified. Determining the causal factors of anaemia can contribute to addressing the problem through appropriate interventions. The aim of this study was to determine the prevalence of anaemia, dietary diversity, anthropometric status, reported health status, socio-demography and associations between these factors among rural women aged 25–49 years.

A cross-sectional descriptive study design was applied in a sample of 134 women living in the rural towns of Trompsburg, Springfontein and Philippolis in the Southern Free State, South Africa. Women who were pregnant at the time of data collection and who were HIV positive were excluded from the current study. This study made use of data collected as part of the Assuring Health for All in the Free State study.

Blood samples were collected and analysed according to standard techniques. These included full blood counts, transferrin saturation, ferritin, homocysteine and red cell folate levels.

A 24-hour recall was completed in a structured interview to determine dietary diversity, categorised as low (≤3 groups), medium (4–5 groups) and high (≥6 groups). A reported health questionnaire was completed for each woman and included information on tobacco and alcohol consumption patterns, medical history and medications as well as menstruation patterns and contraceptive use. A socio-demographic questionnaire was completed for each household which assessed basic demographics of household members; structure of the house; household income; amenities; access to water and sanitation; employment status and cooking facilities. Questions pertaining to language, race, gender, age, employment status and income as well as type of dwelling were also included. Information related to water, sanitation, source of energy and food storage facilities was obtained in

terms of household information. Information for all questionnaires were obtained through structured interviews.

Weight, height, waist circumference, triceps, biceps, subscapular, and suprailiac skinfold measurements were measured according to standard techniques. Weight and height were used to calculate body mass index (BMI) which was categorised as underweight (<18.50kg/m^2), normal weight (18.50–24.99kg/m^2), overweight (25.00–29.99kg/m^2), obesity class I (30.00–34.99kg/m^2), obesity class II (35.00–39.99kg/m^2) and obesity class III (≥40.00kg/m^2). Waist circumference was categorised as normal (<80cm), at risk (≥80cm) and high risk (≥88cm). Body fat percentage was determined by means of the sum of the four skinfolds and categorised as too low (≤8%), acceptable lower end (9–23%), acceptable upper end (24–31%) and too high (≥32%).

The median age of the women in the study was 41 years with most of the women (79.9%) falling in the older age group (35–49 years). Occurrence of anaemia (4.6%), iron deficiency anaemia (0.7%) and iron deficiency (1.5%) among the women was low. However, the prevalence of anaemia of more than 4.9% within a specific population is considered a mild public health problem by the WHO (2008:Online) which is close to the 4.6% of the women in the current study. Elevated homocysteine levels were present in 7.5% of the sample with only 3.8% presenting with low red cell folate levels indicative of folate deficiency. More than half of the women (54.1%) reported that they menstruated regularly and 71.6% had currently or previously used injectable contraceptives. As expected, women who menstruated regularly had significantly lower median haemoglobin levels than those who did not.

With regard to the women's diets, almost half (44.7%) of the women in the sample had a low dietary diversity with flesh meats and fish (good sources of haem iron) consumed by 76.9% of the women. Only a quarter (25.4%) of the women ate dark green leafy vegetables (sources of non-haem iron and folate). All the women consumed starchy foods, some of which are sources of folate and iron due to their mandatory fortification. Significant associations between median MCV and MCH levels and dietary diversity score may indicate

that the mandatory food fortification programme is having a positive impact on the micronutrient intake of these women.

A predominant pattern of malnutrition, characterised by overweight and obesity (70.8%), high rates of abdominal obesity (79.2%) and unhealthy body fat percentages (86.2%) were prevalent. Significant associations between BMI, waist circumference and body fat percentage categories with MCV, MCH levels and transferrin saturation indicate that risk for iron deficiency is associated with obesity.

In terms of the women's reported health, median haemoglobin levels were significantly higher among those women who smoked compared to those who did not. A small percentage of the women (17.9%) had been hospitalised within the past 24 months with some women reporting breathlessness with usual activity (41.0%), loose stools/ diarrhoea for at least three days (17.3%), vomiting (18.0%), loss of appetite (41.0%), blood in their urine (7.5%) and involuntary weight loss of more than 3kg (42.5%) in the past six months. Unexpectedly, median haemoglobin levels were significantly higher among those women who experienced breathlessness with usual activity, but did not differ significantly between women who suffered from anaemia and those who did not, which could indicate that the breathlessness was due to other reasons not investigated in the current study.

Poverty was prevalent in the sample with 37.7% of women being unemployed and only 21.6% having a husband or partner who was a full time wage earner. Even though most women had access to basic infrastructure, low levels of income and dependence on social grants as main source of income (52.7%) show that poverty was prevalent. In the poorest households (with no flush toilet), women were more likely to have a lower median haemoglobin.

Results from the current study thus indicate that regular menstruation, poverty, smoking and obesity are factors that influenced the women's risk for anaemia. Attention should be given to improving the nutritional status and lifestyles of these women in order to improve their overall health and to reduce their risk for chronic diseases and anaemia.

Keywords: Anaemia, haemoglobin, iron, iron deficiency, folate, folate deficiency, dietary diversity, body composition, health, socio-demographics.

OPSOMMING

Anemie, ‘n wêreldwye openbare gesondheidsprobleem wat veral vroue beïnvloed, hou ‘n groot risiko vir menslike gesondheid in. Om hierdie rede is dit nodig om die faktore wat ‘n rol in die ontwikkeling van anemie speel te identifiseer. Die bepaling van faktore as oorsaak van anemie kan bydra tot die aanspreek van dié probleem deur middel van toepaslike intervensie. Die doel van hierdie studie was om die voorkoms van anemie, dieet diversiteit, antropometriese status, gerapporteerde gesondheidstatus, sosio-demografie en verbande tussen hierdie faktore onder vroue in landelike areas tussen 25-49 jaar te bepaal.

'n Dwarssnit, beskrywende studie-ontwerp is toegepas in 'n steekproef van 134 vroue wat in die landelike dorpe van Trompsburg, Springfontein en Philippolis in die Suid-Vrystaat, Suid-Afrika woon. Vroue wat swanger en/of HIV positief was tydens data-opname, was uit die huidige studie uitgesluit. Hierdie studie maak gebruik van data wat as deel van die “Assuring Health for All in the Free State” studie ingesamel is.

Bloedmonsters is versamel en ontleed volgens standaard-tegnieke. Dit sluit in volbloedtelling, transferrien-versadiging, ferritien, homosisteïen en rooiselfolaat-vlakke.

'n 24-uur herroep is in 'n gestruktureerde onderhoud voltooi om dieet-diversiteit wat as laag (≤3 groepe), medium (4-5 groepe) en 'n hoog (≥6 groepe) geklassifiseer is te bepaal. ‘n Gerapporteerde gesondheidvraelys is vir elke vrou voltooi en het inligting oor tabak- en alkoholgebruikspatrone, mediese geskiedenis, medikasie sowel as menstruasiepatrone en die gebruik van voorbehoedmiddels ingesluit. 'n Sosio-demografiese vraelys is vir elke huishouding voltooi en het die basiese demografie van lede van die huishouding; struktuur van die huis; huishoudelike inkomste; geriewe; toegang tot water en sanitasie; indiensnemingstatus en kookgeriewe bepaal. Vrae met betrekking tot taal, ras, geslag, ouderdom, indiensnemingstatus en inkomste sowel as die tipe woning is ook ingesluit. Inligting met betrekking tot water, sanitasie, bron van energie en voedselberging-fasiliteite is verkry in terme van huishoudelike inligting. Inligting vir alle vraelyste is deur middel van gestruktureerde onderhoude verkry.

Massa, lengte, middelomtrek, triseps, biseps, subskapulêre en suprailiaak velvoumates is volgens standaardtegnieke bepaal. Massa en lengte is gebruik om liggaamsmassa-indeks (LMI) te bepaal wat as ondermassa (<18.50kg /m^2), normale massa (18.50-24.99kg /m^2), oormassa (25.00-29.99kg /m^2), vetsugklas I (30.00-34.99kg /m^2), vetsugklas II (35.00-39.99kg /m^2) en vetsugklas III (≥40.00kg /m^2) geklassifiseer is. Middelomtrek was as normaal (<80cm), risiko (≥80cm) en 'n hoë risiko (≥88cm) geklassifiseer. Liggaamsvet-persentasie is bepaal deur middel van die som van die vier velvoue bereken en as té laag (≤8%), aanvaarbare onderste grens (9-23%), aanvaarbare boonste grens (24-31%) en té hoog (≥32 %) gekategoriseer.

Die mediaan-ouderdom van die vroue in die studie was 41 jaar met die meeste van die vroue (79,9%) wat in die ouer ouderdomsgroep (35-49 jaar) val. Die voorkoms van anemie (4.6%), ystertekort-anemie (0,7%) en ystertekort (1.5%) onder die vroue was laag. Die voorkoms van anemie van meer as 4.9% binne ‘n spesifieke populasie word egter as ‘n matige publieke-gesondheidsprobleem deur die WGO beskou, wat baie naby aan die 4.6% van die vroue in die huidige studie is. Verhoogde homosisteïen-vlakke was teenwoordig in 7.5% van die steekproef met slegs 3.8% wat lae rooiselfolaat-vlakke getoon het. Dit is ‘n aanduiding van folaattekort. Meer as die helfte van die vroue (54.1%) het gerapporteer dat hulle gereeld menstrueer en 71.6% het tans of voorheen gebruik gemaak van inspuitbare voorbehoedmiddels. Soos verwag, het die vroue wat gereeld menstrueer, aansienlik laer mediaan-hemoglobienvlakke gehad as diegene wat nie gemenstrueer het nie.

Met betrekking tot die vroue se dieet het byna die helfte (44.7%) van die vroue in die steekproef 'n lae dieet diversiteit gehad met 76.9% van die vroue wat vleis en vis (goeie bronne van heemyster) ingeneem het. Slegs 'n kwart (25.4%) van die vroue het donkergroen blaargroentes (bronne van nie-heem yster en folaat) geëet. Al die vroue neem styselrykevoedsel in, waarvan sommige bronne van folaat en yster as gevolg van hul verpligte verryking. Beduidende verbande is tussen mediaan MCV- en MCH-vlakke asook dieet-diversiteittelling gevind wat daarop kan dui dat die verpligte voedselverrykingprogram 'n positiewe impak op die mikrovoedingstofinname van hierdie vroue het.

‘n Oorheersende patroon van wanvoeding, wat gekenmerk word deur oormassa en vetsug (70.8%), 'n hoë voorkoms van abdominale vetsug (79,2%) en ongesonde liggaamsvetpersentasie (86,2%), was teenwoordig. Beduidende verbande tussen LMI, middelomtrek en liggaamsvetpersentasie-kategorieë met MCV, MCH en transferrienversadiging dui daarop dat die risiko vir ystertekort geassosieer word met vetsug.

In terme van die vroue se gerapporteerde gesondheid, was die mediaan-hemoglobienvlakke aansienlik hoër onder vroue wat gerook het in vergelyking met diegene wat nie gerook het nie. 'n Klein persentasie van die vroue (17,9%) was die afgelope 24 maande in die hospitaal opgeneem en 'n paar vroue het benoudheid met gewone aktiwiteit (41.0%), los stoelgange / diarree vir ten minste drie dae (17,3%), braking (18.0 %), verlies van eetlus (41.0%), bloed in urine (7.5%) en onwillekeurige massaverlies van meer as 3 kg (42.5%), ervaar. Mediaan-hemoglobienvlakke was aansienlik hoër onder vroue wat benoudheid met gewone aktiwiteit ervaar het, maar geen beduidende verskil was tussen vroue wat aan anemie gely het of nie. Dit daarop kan dui dat die benoudheid as gevolg van ander redes, wat nie in die huidige studie ondersoek is nie, was.

Armoede het oor die algemeen onder die vroue voorgekom met 37,7% van vroue wat werkloos was en slegs 21,6% met 'n man of lewensmaat wat 'n voltydse loon verdien. Selfs al het die meeste vroue toegang tot basiese infrastruktuur dui lae vlakke van inkomste en afhanklikheid van maatskaplike toelaes as hoofbron van inkomste (52,7%) dat armoede algemeen was. In die armste huishoudings (met geen spoeltoilet), was vroue meer geneig om 'n laer mediaan-hemoglobien te hê.

Resultate van die huidige studie dui dus aan dat gereelde menstruasie, armoede, rook en vetsug faktore is wat die vroue se risiko vir anemie beïnvloed. Aandag moet aan die verbetering van die voedingstatus en lewenstyl van hierdie vroue gegee word, om so hul algehele gesondheid te verbeter en om hul risiko vir chroniese siektes en anemie te verminder.

APPENDIX A

INFORMATION LETTER TO COMMUNITIES

Assuring Health for All (AHA) in the Free State
INFORMATION DOCUMENT

Study title: Assuring Health for All (AHA) in the Free State

Thank you for being willing to help us in this very important project. We are sure that the project will contribute to improving the health of all the people of the Free State.
We, the University of the Free State, Faculty of Health Sciences, are doing research on determining the factors involved in causing disease and disability in the Southern Free State. Research is just the process to learn the answer to a question. In this study we want to learn what factors need to be addressed in health programmes in the Free State. The study involves research and is not part of routine medical care.

Invitation to participate: We are asking/inviting you to participate in this research study, or/andasking for your permission to include your child in this research study.

What is involved in the study: The aim of the project is to get enough information regarding the development of chronic diseases like diabetes, stroke, high blood pressure and heart disease as well as HIV/AIDS to plan appropriate health and nutrition intervention strategies for the people of the Free State. Trompsburg, Philippolis and Springfontein have been chosen as the rural areas and Mangaung as the urban area.

For this study we need households whom we can follow for 12 years. The baseline survey will be done during March 2009 in Mangaung. You will be asked to visit the MUCPP clinic for one day to take the necessary measurements and to complete the questionnaires. After the baseline survey has been completed, we will implement a nutrition intervention in your community to address the problems identified in the baseline survey. This intervention will form part of the service learning interventions of the University. In addition to the services that we will render in the community, we will visit your community again after three to six years to repeat the measurements.

All the questionnaires will be filled out at MUCPP clinic by students from the University of the Free State. Respondents from the chosen households will be asked to complete the following questionnaires in an interview with the students:

- Socio-demographic and household questionnaire,
- Household food security and food procurement questionnaire,
- Health questionnaire,
- Knowledge, practices and attitudes (KPA) about nutrition questionnaire,
- Diet and physical activity questionnaire.

We will also take some measurements such as weight, height, skinfold thicknesses, blood pressure, blood samples and a urine sample. With your permission we will draw 60ml of blood in adults and this will only be done once. In adults blood and urine samples will be used to determine the following: Full blood count; HbA1c; Glucose; Insulin; Lipogram; Homocysteine; Red cell Folate; Serum Vitamin B12; Fibrinogen; Gamma glutermyl transferases (GGT); Carbohydrate-deficient transferrin (CDT); Ferritin; Uric acid; Creatinin; C-reactive protein; Albumin; Pre-albumin; Transferrin; Retinol-binding protein; TSH; Iodine (urine); Leptin; Tumour Necrosis Factor alpha; Interleuken 6; Melatonin; Brain natriuretic peptide; ACTH; Cortisol; Orexin; Urotensin-11; Endothelin 1; Plasminogen Activation Inhibitor (PAI-1); Adiponectin; Micro-albuminuria (urine); Glucose tolerance (sub-sample); FFA (sub-sample).

A short medical examination will be performed on all participants members to identify any serious health problems.

We would like to retain some of the same blood in storage for possible future research related to the present research question. Blood samples will be stored anonymously for a period of five years at the

Department of Chemical Pathology at the University of the Free State. If you are unhappy to have your blood stored for future research, it will be disposed of at the end of the study, once the sample storage and record-keeping requirements of good research practice have been met.

It is very important that we gather quality data and knowledge. Because HIV/AIDS is a devastating illness and affects almost all aspects of health, it is necessary to know if HIV is absent before we analyse the data. It will be to your benefit as well as the benefit of the research to determine your HIV status. Therefore we will also ask permission to draw blood to determine your HIV status and ask questions about your HIV status which you are allowed not to answer. You will be asked to sign a separate consent form for the HIV test. You will receive pre- and post-testing counselling by a medical practitioner and all results will be kept strictly confidential in accordance with the guidelines of the Health Professions Council of South Africa (HPCSA). You will only be informed of your HIV result if you choose to be. All respondents who choose to be informed of the results will be informed by a medical doctor and referred for relevant management. None of the researchers (other than one doctor) will know the HIV status of any participants.

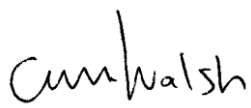
Blood tests will involve an analysis of the genetic composition of red blood cells and are aimed at increasing the understanding of the causes and behaviour of chronic diseases of lifestyle such as obesity, diabetes and heart disease. Genes are what you inherit from your parents. They are found in every part of your body and therefore they will be present in the blood that we draw. The findings may benefit/eventually benefit others in terms of prevention or treatment of diseases. You are free to refuse consent and you do not have to give reasons for doing so. The following arrangements have been made to ensure privacy and confidentiality of your genetic information: All blood samples will be stored anonymously. Your genetic material and information will be used in an identifiable form. The research may reveal information of potential importance to the future health of an identifiable or potentially identifiable participant or the participant's offspring.

Researchers will endeavour to provide information about the outcome of the research. If research generates information about you which may be of relevance to the health of other family members, your consent will be sought before offering to disclose such information to the family members concerned. Your material and information will not be released for other uses without consent, unless required by law.

Risks of being involved in the study: Medical doctors and registered nurses will be responsible for safely drawing blood samples. In the unlikely event that an adverse event results from the procedure, you will be compensated for any expenses.
Benefits of being in the study: By participating in the study you will help us to develop health and nutrition strategies that will benefit the people of the Free State. You will be given pertinent information on the study while involved in the project and after the results are available.

Participation is voluntary, and refusal to participate will involve no penalty or loss of benefits to which you are entitled; you may discontinue participation at any time without penalty or loss of benefits to which you are otherwise entitled.
Confidentiality: Efforts will be made to keep personal information confidential. Absolute confidentiality cannot be guaranteed. Personal information may be disclosed if required by law. Organizations that may inspect and/or copy your research records for quality assurance and data analysis include groups such as the Ethics Committee for Medical Research and the Medicines Control Council. If results are published, this may lead to individual/cohort identification.

Kind regards



PROF CORINNA WALSH
Contact details: 083 297 6030 / 051 4013818(W)

INLIGTINGSDOKUMENT

Studietitel: Assuring Health for All (AHA) in the Free State

Dankie dat u bereid is om ons te help met hierdie baie belangrike projek. Ons is seker dat die projek sal bydra om die gesondheid van alle persone in die Vrystaat te verbeter. Ons, die Universiteit van die Vrystaat, Fakulteit Gesondheidswetenskappe, doen navorsing oor die faktore wat betrokke is by die oorsake van siekte in die Vrystaat. Navorsing is slegs die proses waardeur die antwoord op 'n vraagstuk verkry word. In hierdie studie wil ons leer watter faktore aangespreek moet word in gesondheidsprogramme in die Vrystaat. Die studie behels navorsing en is nie deel van roetine mediese behandeling nie.

Uitnodiging om deel te neem: Ons versoek/nooi u uit om aan 'n navorsingstudie deel te neem of/en vra u toestemming om u kind by die navorsingstudie in te sluit.

Wat behels die studie – Die doelwit van hierdie projek is om genoeg inligting in te samel oor die ontwikkeling van chroniese siektes soos diabetes, beroerte, hoë bloeddruk en hartsiektes sowel as MIV/VIGS om toepaslike gesondheids- en voedingintervensie strategieë te kan beplan vir die mense van die Vrystaat. Trompsburg, Philippolis en Springfontein is as die plattelandse areas gekies en Mangaung as die stedelike area.

Vir die studie benodig ons huishoudings wat ons vir 12 jaar kan opvolg. Die basislyn opname sal in Mangaung gedoen word tydens Maart 2009. U sal gevra word om die MUCPP kliniek vir een dag te besoek waar die nodige metings gedoen sal word en vraelyste voltooi sal word. Nadat die basislynopname voltooi is, sal ons 'n voedingintervensieprogram in u area implementeer om die probleme wat in die basislynopname identifiseer is aan te spreek. Hierdie intervensie vorm deel van die diensleer intervensies van die universiteit. Tesame met die intervensie sal ons ook die gemeenskap elke drie jaar tot ses jaar besoek om die metings te herhaal.

Al die vraelyste sal by MUCPP voltooi word deur studente van die Universiteit van die Vrystaat. Respondente van die gekose huishoudings sal gevra word om die volgende vraelyste te voltooi in 'n onderhoud met die student:

- Sosio-demografiese en huishoudelike vraelys,
- Huisehoudelike voedselsekuriteit en voedselverkrygings vraelys,
- Gesondheids vraelys,
- Kennis, praktyke en houding teenoor voeding vraelys,
- Dieet en fisiese aktiwiteit vraelys.

Ons sal ook sekere metings soos gewig, lengte, velvoudiktes, bloeddruk, bloed monsters en uriene monsters neem. Met u toestemming sal ons in volwassenes 60ml bloed trek en dit sal slegs een keer geskied. In volwassenes sal bloed en uriene monsters gebruik word om die volgende te bepaal: Volbloedtelling; HbA1c; Glukose; Insulien; Lipogram; Homositeïen; Rooisel Folaat; Serum Vitamien B12; Fibrinogeen; Gamma glutermyl transferases (GGT); Carbohydrate-deficient transferrin (CDT); Ferritin; Uriensuur; Kreatinien; C-reactiewe proteïen; Albumien; Pre-albumien; Transferrien; Retinol-binding proteïen; TSH; Jodium (uriene); Leptien; Tumor Nekrosis Faktor alfa; Interleuken 6; Melatonien; Brain natriuretic peptide; ACTH; Kortisol; Orexin; Urotensien-11; Endothelien 1; Plasminogen Activation Inhibitor (PAI-1); Adiponektien; Mikro-albumienuria (uriene); Glukose toleransie (sub-sample); FFA (sub-sample).

'n Kort mediese ondersoek sal ook gedoen word op sekere lede van die huishouding om ernstige gesondheidsprobleme te identifiseer.

Ons wil graag van die bloed bêre vir moontlike toekomstige navorsing wat verband hou met die huidige navorsingsvrae. Bloed monsters sal anoniem gestoor word vir 'n periode van vyf jaar. As u ongelukkig daaroor voel dat u bloed vir toekomstige navorsing geberg word sal daar aan die einde van die studie daarmee weggedoen word sodra die monsterbergings- en aantekeningsvereistes van goeie navorsingspraktyk nagekom is.

Dit is baie belangrik dat ons inligting van 'n hoë kwaliteit versamel. Omdat MIV/VIGS 'n siekte is wat amper alle aspekte van gesondheid beïnvloed, is dit nodig dat ons weet of MIV afwesig is voordat ons die data ontleed. Dit sal tot voordeel van uself en die navorsing strek indien u HIV status bepaal kan word. Dus sal ons toestemming vra om bloed te trek om u MIV status te bepaal en vrae oor HIV vra wat u nie hoef te antwoord indien u nie wil nie. U sal gevra word om 'n aparte teostemmingsvorm te

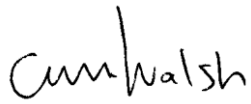
voltooi vir die HIV toets. U sal voor en na die toets berading ontvang deur ‘n mediese dokter en alle uitslae sal streng vertroulik hanteer word volgens die riglyne van die Health Professions Council of South africa (HPCSA). U sal slegs van u MIV uitslae in kennis gestel word indien u kies om dit te ontvang. Alle respondente wat kies om van hulle uitslae in kennis gestel te word sal deur ‘n mediese dokter in kennis gestel word en verwys word vir die relevante hantering. Die navorsers (met uitsluiting van een dokter) sal nie weet wat u uitslae is nie.

Bloedtoetse sal die analise van die genetiese samestelling van rooibloedselle insluit en is gemik daarop om die oorsake en gevolge van chroniese siektes soos vetsug, diabetes en hartsiektes beter te verstaan. Gene is dit wat u van u ouers erf en word in elke deel van u liggaam aangetref. Daarom sal dit in enige weefsel of bloed wat deur ons verwyder word teenwoordig wees. Die bevindings kan tot ander se voordeel strek met betrekking tot voorkoming en behandeling van die toestand. Dit staan u vry om toestemming te weier en u hoef geen redes daarvoor te verstrek nie. Die volgende reëlings is getref om privaatheid en vertroulikheid van u genetiese inligting te verseker: Alle bloedmonsters sal anoniem geberg word. U genetiese materiaal en inligting sal in ‘n identifiseerbarevorm gebruik word. Die navorsing mag inligting openbaar wat van potensiële belang mag wees vir die toekomstige gesondheid van ‘n identifiseerbare of potensieel identifiseerbare deelnemer of die deelnemer se nakomelinge.

Navorsers sal poog om inligting oor die uitkoms van die navorsing te verskaf. As navorsing inligting aan die lig bring wat van belang mag wees vir die gesondheid van u familielede, sal u toestemming verkry word voordat sodanige inligting aan die betrokke familielede bekend gemaak word. U bloed en inligting sal nie sonder toestemming vir ander gebruike beskikbaar gestel word nie tensy vereis deur die wet.

Risikos van deelname aan die studie: Mediese dokters of geregistreerde verpleegkundiges sal verantwoordelik wees vir die veilige neem van bloedmonsters. In die onwaarskynlike geval dat ‘n negatiewe gevolg ontstaan as gevolg van die prosedure sal u vir enige onkostes vergoed word.
Voordele van deelname aan die studie: Deur aan die studie deel te neem sal u ons help om gesondheids- en voedingstrategieë te ontwikkel wat die mense van die Vrystaat sal baat. Die proefpersoon sal pertinente inligting oor die studie ontvang tydens betrokkenheid by die projek en agterna wanneer die resultate beskikbaar is.
Deelname is vrywillig, en weiering om deel te neem sal geen boete of verlies van voordele waarop die deelnemer andersins geregtig is behels nie; die proefpersoon kan te eniger tyd aan deelname onttrek sonder boete of verlies van voordele waarop die proefpersoon andersins geregtig is.
Vertroulikheid: Daar sal gepoog word om persoonlike inligting vertroulik te hou. Volkome vertroulikheid kan nie gewaarborg word nie. Persoonlike inligting kan bekend gemaak word as die wet dit vereis. Organisasies wat u navorsingsrekords mag ondersoek en/of kopieer vir kwaliteitsversekering en data-analise sluit groepe soos die Etiekkomitee vir Mediese Navorsing en die Medisynebeheerraad in. As resultate gepubliseer word kan dit lei tot individuele/groepsidentifikasie.

Vriendelike groete



Prof CORINNA WALSH
Kontakbesonderhede: 083 297 6030 / 051 4013818(W)

Assuring Health for All (AHA) in the Free State
(Tshepiso ya Bophelo ho bohle ba Foreisitata)

LENGOLO LA TLHAHISO LESEDING

Sehloho sa dipatlisiso: Assuring Health for All in the Free State

Re leboha ha o dumetse ho re thusa dipatlisisong tsena tse bohlokwa. Re tshepa ha dipatlisiso di tla re thusa ho ntlafatsa maphelo a batho bohle ba Foreisitata.
Rona, re le Yunivesithi ya Foreisitata, Lefapha la tsa Maphelo, re etsa dipatlisiso ho shebana le dintho tse bakang mafu le boqhwala mona Foreisitata e Borwa.
Dipatlisiso ke mokgwa wa ho ithuta karabo ya potso e itseng. Ka dipatlisiso tsena re batla ho ithuta hore na ke dintho dife tse hlokang hore di amuwe ditsamaisong tsa tsa bophelo mona Foreisitata. Tsena ke dipatlisiso feela, mme ha se karolo ya tshebeletso ya tsa bongaka.

Sememo sa ho nka karolo: Re a o kopa/ re a o mema ho nka karolo dipatlisisong tsena, ebile/ kapa re kopa tumello ya hao ho sebedisa ngwana wa hao dipatlisisong tsena.
Dipatlisiso tsena di kenyeleditse eng: Sepheo sa dipatlisiso tsena ke ho fumana tlhahiso leseding e lekaneng mabapi le tswello pele ya mafu a kang diabetes, stroke, high blood pressure le mafu a pelo hammoho le HIV/AIDS hore ho tle ho thalwe mekgwa ya tsamaiso ya tsa bophelo le phepo bakeng sa batho ba Foreisitata. Trompsburg, Phillippolis le Springfontein di kgethilwe jwalo ka dibaka tsa mahaeng mme Mangaung e kgethilwe e le sebaka sa ditoropong tse tla sebediswang dipatlisisong.
Bakeng sa dipatlisiso tsena re hloka ho sebetsa le malapa ohle motseng wa lona bakeng sa dilemo tse 12. Dipatlisiso tsa pele di tla qala ka Hlakubele (March) 2009. O tla kopuwa ho etela MUCPP letsatsi le le leng hore o tle o tsebe ho tlatsa diforomo le hore o methuwe. Kamora dipatlisiso tsena tsa pele, ho kenywa tshebetsong mekgwa ya tlhokomelo mabapi le tsa phepo sebakeng seo o dulang ho sona hore ho tle ho lokisanwe le mathatha a tla be a hlahelletse dipatlisisong tsa pele. Tlhokomelo ena e tla ba karolo ya ho ithuta ho bile ho fanwa ka ditshebeletso ke baithuti ba Yunivesithi. Hammoho le ditshebetso tseo re tla be re fana ka tsona motseng, re tla etela motse wa hao ka mora dilemo tse tharo hoisa ho tse tseletseng hore ho phethwe ho metha.

Diforomo tsohle di tla tlatswa sebakeng seo liphuputso li tla tswareloa teng ke baithuti ba tswang Yunivesithing ya Foreisitata kapa ke bathusi ba kwetlisitsweng ba tswang motseng wa hao. Batho ba nkang karolo ho tswa malapeng a kgethilweng ba tla kopuwa ho tlatsa diforomo tse latelang ho ya ka dipotso tseo ba tla beng ba di botswa ke moithuti kapa mosebeletsi wa setjhaba:

- Dipotso ka maemo a hao le ka lelapa la hao,
- Theko ya dijo le 'food security',
- Tsebo, tshebetso le mekgwa e amanang le phepo,
- Tsela ya ho ja le boikwetliso.

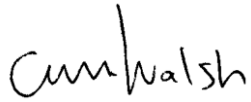
Re tla o metha boima, botelete, botenya ba momeno wa letlalo (skinfold) le kgatello ya madi (blood pressure), o tla nkuwa madi hammoho le metsi. Ho batho ba baholo re tla hula madi a kana ka dimililitara tse 60 hona ho tla etswa hanngwe feela. Ho tla etswa dihlahlobo tsa bophelo ho batho ba itseng ba lelapa ho shebana le mathata a bophelo a tshosetsang. Ka tumello ea hau re tla ntsa madi a ka bang dimelimetara tse mashome a tseletseng (60ml) ho motho e moholo le tse leshome le metso e mehlano, mme hona ho tla etwa hangoe feela. Madi le metsetse e nkuoeng ho batho ba baholo e tla sebediswa ho hlalhoba matsoai le matsooeana a fumanehang ho tsona a kenyeletsang tse latelang: Full blood count; HbA1c; Glucose; Insulin; Lipogram; Homocysteine; Red cell Folic acid; Serum Vitamin B12; Fibrinogen; Gamma glutermyl transferases (GGT); Carbohydrate-deficient transferrin (CDT); Ferritin; Uric acid; Creatinin; C-reactive protein; Albumin; Pre-albumin; Transferrin; Retinol-binding protein; TSH; Iodine (urine); Leptin; Tumour Necrosis Factor alpha; Interleuken 6; Melatonin; Brain natriuretic peptide; ACTH; Cortisol; Orexin; Urotensin-11; Endothelin 1; Plasminogen Activation Inhibitor (PAI-1); Adiponectin; Micro-albuminuria (urine); Glucose tolerance (sub-sample); FFA (sub-sample). Tse ding tsa matsoai le matsoaeana ana di tla hlahlojoa mading le metsetseng ea bana
Re ka rata ho boloka a mang a madi hore a tle a tsebe ho sebediswa ka nako e tlang dipatlisisong tse tshwanang le tsena. Madi ana a tla bolokwa kantle le ho ngolwa mabitso bakeng sa dilemo tse hlano. Ha o sa rate ha madi a hao a bolokwa bakeng sa dipatlisiso tse ding, a tla lahlwa ha ho qetwa ka dipatlisiso le hang feela ha a ile a bolokwa hantle e bile dintho tse hlokahalang bakeng sa dipatlisiso di ile tsa fumanwa ka mokgwa o nepahetseng.

Ho bohlokwa haholo hore tlhahiso leseding eo re e fumanang ke e hlwahlwa. Hobane lefu la HIV e le le hlokofatsang haholo, ebile le ama maphelo a rona, ho a hlokahala hore re tsebe hore kokwana-hloko ena e teng kapa tjhe. Ke molemong oa hau le oa mofuputse ho fumana boemo ha hau ba HIV. Ka hona re tla kopa tumello ea ho ntsa madi ho uena ho ea hlahloba boemo ba hau ba HIV mme re tla u botsa lipotso tse ling tse amanang le boemo ba hau ba HIV tseo u sa qobelloeng ho li araba. U ka li araba ka ho rata ha hau. U tla kopuoa ho saena foromo eo u re fang tumello ea ho hlahloba boemo ba hau ba HIV. Ka hoo re tla hula madi le ho o botsa dipotso mabapi le maemo a hao a HIV. O dumelletswe ho se arabe dipotso tsena. Ho tla buisanwa le wena ho tebesitswe maikutlo pele le ka mora dihlahlobo tse tla etswa ke ngaka, mme diphetho tsohle di tla ba sephiri ho ya ka ditaelo tsa Health Professions Council ya South Africa (HPCSA). O tla tsebiswa ka maemo a hao a HIV feela ha o kgetha hore o tsebiswe. Batho bohle ba batlang ho tsebiswa ka sephetho sa dihlahlobo tsa HIV, ba tla tsebiswa ke ngaka, mme ba tla romellwa ho batho ba tla tseba ho ba fa thuso. Babatlisisi ba bang (ka ntle ho dingaka) ba ke ke ba tseba maemo a ba nka karolo a HIV.

Ho hulwa ha madi ho kenyelleditse ho hlahloba 'genetic composition' e ho di 'red blood cells', mme hona ho tla thusa ho phahamisa kutlwisiso ya dintho tse bakang mafu a kang monono, diabetes le mafu a pelo. Di 'genes', ke dintho tseo o di futsang ho tswa ho batswadi ba hao. Di fumanwa dikarolong tsohle tsa mmele kahoo di a fumaneha le ho madi a tla hulwa. Dintlha tse tla fumanwa di tla thusa batho ba bang ka ho thusa ho thibela, kapa ho phekola mafu a fapaneng. O na le tokelo ya ho hana ho nka karolo ebile ha ho hlokahale hore o fane ka lebaka. Ho netefatsa hore dintlha tse tla fumanwang ho wena di dula e le sephiri, ho entswe dintho tsena tse latelang: Madi ohle a tla bolokwa ntle le ho ngolwa mabitso. 'Genetic material' ya hao e tla sebediswa ka mokgwa o tsebahalang. Dipatlisiso tsena di ka nna tsa fana ka tlhahiso leseding e bohlokwa bakeng sa bokamoso ba monka karolo kapa ba bana ba hae.

Babatlisisi ba tla leka ho fana ka lesedi mabapi le sephetho sa dipatlisiso. Ha dipatlisiso di ka fana ka dinthla tse leng bohlokwa bakeng sa maphelo a ba bang ba lelapa, tumello e tla kopuwa ho wena pele batho ba amehang ba tsebiswa. Dintlha tsohle tsa hao di ke ke tsa sebediswa nqeng tse ding kantle ho tumello ya hao, kantle le ha ho ka hlokeha hore ho etswe jwalo ho ya ka molao.

Kotsi tse ka bang teng dipatlisisong: Dingaka le baoki ba tla ikarabella ho huleng ha madi ka mokgwa o bolokehileng. Ha ho ka etsahala hore ho be le se o etsahallang se sa lokang o tla lefuwa ditshenyehelo tsa hao tsohle.
Ditholwana tsa ho nka karolo: Ha o nka karolo o tla thusa ho tsweletsa pele metjha ya tsa bophelo le phepo ho thusa batho ba Foreisitata. O tla fuwa tlhahiso leseding e bohlokwa tsamaong ya dipatlisiso le ha sephetho sa dipatlisiso se fumaneha.
O nka karolo ka ho ithaopa, mme ha o hana ho nka karolo o ke ke wa lahlehelwa ke letho; o ka tlohella ho nka karolo nako e nngwe le e nngwe ntle le ho lahlehelwa ke letho.
Sephiri: Ho tla etswa maleba-leba a hore dintlha tsa hao di dule di le lekunutu. O tshepiswa lekunutu ka hohle-hohle. Dintlha tsa hao di phatlalatswa feela ha molao o re jwalo. Mekgatlo e tla hlahloba kapa e tla kopisa dintlha tsa hao ho lekola boleng e kenyeleditse e kang Ethics Committee ya Medical Research hammoho le Medicine Control Council.
Ha diphetho di ka phatlalatswa, hona ho ka lebisa ho tsebiswa ha motho kapa sehlopha.



Ka boikokobetso
Prof Corinna Walsh
Mohala: 083 297 6030 / 051 401 3818

APPENDIX B

INFORMED CONSENT FORM

Assuring Health for All (AHA) in the Free State

CONSENT TO PARTICIPATE IN RESEARCH

You have been asked to participate in a research study.

You have been informed about the study by …… .

You may contact Prof Corinna Walsh at 083 297 6030 at any time if you have questions about the research or if you are injured as a result of the research.

You may contact the Secretariat of the Ethics Committee of the Faculty of Health Sciences, UFS at telephone number (051) 4052812 if you have questions about your rights as a research subject.

Your participation in this research is voluntary, and you will not be penalized or lose benefits if you refuse to participate or decide to terminate participation.

If you agree to participate, you will be given a signed copy of this document as well as the participant information sheet, which is a written summary of the research. You are also giving permission that some of the same blood can be retained in storage for possible future research related to the present research question.

The research study, including the above information has been verbally described to me. I understand what my involvement in the study means and I voluntarily agree to participate.

_____ _____

Signature of Participant Date

Assuring Health for All (AHA) in the Free State

TOESTEMMING TOT DEELNAME AAN NAVORSING

U is versoek om aan ‘n navorsingstudie deel te neem.
U is oor die studie ingelig deur …….. .

U kan Prof Corinna Walsh enige tyd kontak by 083 297 6030 indien u vrae oor die navorsing het of as gevolg van die navorsing beseer is.

U kan die Sekretariaat van die Etiekkomitee van die Fakulteit Gesondheidsweteskappe, UV by telefoonnommer (051) 4052812 kontak indien u enige vrae het oor u regte as ‘n proefpersoon.

U deelname aan hierdie navorsing is vrywillig, en u sal nie gepenaliseer word of voordele verbeur as u weier om deel te neem of besluit om deelname te staak nie.

As u instem om deel te neem, sal ‘n ondertekende kopie van hierdie dokument sowel as die deelnemerinligtingsblad, wat ‘n geskrewe opsomming van die navorsing is, aan u gegee word .

Die navorsingstudie, insluitend die bogenoemde inligting is verbaal aan my beskryf. Ek begryp wat my betrokkenheid by die studie beteken en ek stem vrywillig in om deel te neem.

_____ _____

Handtekening van deelnemer Datum

Assuring Health for All (AHA) in the Free State
(Tshepiso ya Bophelo ho bohle ba Foreisitata)

TUMELLO YA HO NKA KAROLO DIPATLISISONG

O kopuwe ho nka karolo dipatlisisong.

O tsebisitswe ka dipatlisiso tsena ke .....

O ka ikopanya le Prof Corinna Walsh ho 083 297 6030 nako e nngwe le e nngwe ha o na le dipotso ka dipatlisiso kapa ha o ka wa lematseha ka lebaka la dipatlisiso.

O ka ikopanya le mongodi wa komiti ya Ethics ho Faculty ya Health Sciences, Yunivesithing ya Foreisitata nomorong ya (051) 4052812 ha o ena le dipotso ka ditokelo tsa hao jwalo ka motho ya nkang karolo dipatlisisong.

O nka ka karolo dipatlisisong ka ho ithaopa, ka hoo o ke ke wa ahlolwa kapa wa lahlehelwa ke di letho ha o ka wa hana ho nka karolo kapa wa tlohella ho nka karolo.

Ha ebe o dumela ho nka karolo, o tla fuwa lengolo le tshwanang le lena le saenuweng hammoho le lengolo le fanang ka tlhahiso leseding, eo e leng tlhaloso e ngotsweng ya dipatlisiso tsena.

Ke hlaloseditswe sepheo sa dipatlisiso, hammoho le tlhahiso leseding ena e ka hodimo ka molomo. Ke utlwisisa ho nka karolo ha ka dipatlisisong mme ke dumela ho nka karolo ka ho ithaopa, ntle le ho qobellwa.

_____ _____

Signature ya ya nkang karolo Letsatsi

APPENDIX C

PARTICIPATION LETTER

Assuring Health for All (AHA) in the Free State

Participation letter

Dear Participant

Thank you for being willing to help us in this very important project. We are sure that the project will contribute to improving the health of all the people of the Free State.

At the time you receive this letter you would have already been visited by a fieldworker and you have already signed consent to give a blood sample. This letter serves to inform you of the date and time the blood sample and other measurements will be taken at the research unit (hall) closest to your household.

IMPORTANT INFORMATION

1. You must be at the research unit (hall) in your community on **……** by 0….h00.
2. You **MUST NOT EAT OR DRINK** anything after ten o'clock of the previous night (10 pm of the night before). This is necessary for the glucose test to be accurate.
3. You **MUST BRING YOUR ID DOCUMENT** with you
4. You will receive something to eat and drink after the blood sample is taken.
5. If you are employed, please show this letter to your employer.

Dear Employer

This serves to ask you to give one day's paid leave to……. in order to allow him/her to attend his appointment with the research team of the Faculty of Health Sciences at the University of the Free State.

Thank you for your cooperation. For any further information please contact Dr Corinna Walsh at 083 297 6030.

C Walsh (project leader)

Assuring Health for All (AHA) in the Free State

Deelname brief

Beste Deelnemer

Dankie dat u bereid is om ons te help met hierdie belangrike projek. Ons is seker dat die projek sal bydra tot die verbetering van gesondheid van al die mense in die Vrystaat.

Teen die tyd wat u hierdie brief ontvang het ‘n veldwerker u al reeds besoek en u het toestemming gegee om deel te neem aan die projek en ‘n bloed monster te gee Met hierdie brief wil ons u graag in kennis stel van die datum en tyd wat die bloed getrek sal word en ander mates geneem sal word by die navorsingseenheid (saal)naaste aan u woning.

BELANGRIKE INLIGTING

1. U moet by die navorsingseenheid (saal) in u gemeenskap wees op **……** teen 0….h00.
2. U **MOET NIKS EET OF DRINK** na tien uur die vorige aand (10 pm van die aand voor die toets). Dit is nodig vir die glukose toets om betroubaar te wees.
3. U **MOET U ID DOKUMENT** saam met u kliniek toe bring
4. U sal ‘n ligte ete en iets om te drink ontvang nadat die bloedtoets voltooi is.
5. Indien u werk, moet u asseblief hierdie brief aan u werkgewer wys.

Geagte Werkgewer

Met hierdie brief vra ons dat u een dag betaalde verlof toestaan aan……. om dit vir haar/hom moontlik te maak om hierdie afspraak met die navorsingsspan van die Fakulteit Gesondheidswetenskappe by die Universiteit van die Vrystaat by te woon.

Dankie vir u samewerking. Vir verdere inligting kontak asseblief vir Dr Corinna Walsh by 083 297 6030.

C Walsh (projekleier)

APPENDIX D

INDIVIDUAL DIETARY INTAKE QUESTIONNAIRE

Assuring Health for All (AHA) in the Free State

Dietary intake questionnaire

24-hour recall

Town: _____ | 1

Household number: | 2-5

Member number (as on socio-demo form): | 6-7

Interviewer:_____ | 8-9

Food and fluid intake (expressed in exchanges):

Food/Drinks and amounts	**Milk and milk products**	**Meat and meat alternatives**	**Legumes**	**Fruit ß-carotene**	**Vegetables ß-carotene**	**Fruit Vit C**	**Vegetables Vit C**	**Fruit other**	**Vegetables other**	**Bread and cereals**	**Fats and oils**	**Sweets/Sugar**	**Alcohol**
Breakfast and mid-morning													
Lunch and mid afternoon													
Supper and late night													
Total:													

APPENDIX E

DIETARY DIVERSITY QUESTIONNAIRE

Dietary Diversity Questionnaire

Town/ Area: _____ 1

Household number 2-4

Member number (as on socio-demo form) 5-6

Number	Food group	Consumed: Yes (1) / No (2)	
1	Starchy staples		7
2	Dark green leafy vegetables		8
3	Other vitamin A rich fruits and vegetables		9
4	Other fruits and vegetables		10
5	Organ meat		11
6	Meat and fish		12
7	Eggs		13
8	Legumes, nuts and seeds		14
9	Milk and milk products		15

APPENDX F

ANTHROPOMETRY FORM

Assuring Health for All (AHA) in the Free State
Anthropometry

Area in Mangaung:_____ [ ] 1

Household number: [ ][ ][ ] 2-4

Member number (as on socio-demographic form): [ ][ ] 5-6

Interview Date: **D D M M Y Y Y Y** [ ][ ][ ][ ][ ][ ][ ][ ] 7-14

Measurer (interviewer):_____ [ ][ ] 15-16

Weight (kg):_____ [ ][ ][ ].[ ] 17-21

Height (cm): _____ [ ][ ][ ].[ ] 22-26

If height cannot be measured:

Knee height (cm):_____ [ ][ ][ ].[ ] 27-31

Demispan (cm):_____ [ ][ ][ ].[ ] 32-36

Circumferences (cm):

Upper-arm (adults and children):_____ [ ][ ].[ ] 37-40

Waist (adults): _____ [ ][ ][ ].[ ] 41-45

Hip (adults): _____ [ ][ ][ ].[ ] 46-50

Wrist (adults):_____ [ ][ ].[ ] 51-54

Head circumference (children):_____ [ ][ ].[ ] 55-58

Bio-impedance fat percentage (adults):_____ [ ][ ].[ ] 59-62

Skinfold thicknesses (mm):

Triceps (adults and children):_____ _____ _____ [ ][ ] 63-64

Biceps (adults): _____ _____ _____ [ ][ ] 65-66

Supra-ileac (adults): _____ _____ _____ [ ][ ] 67-68

Subscapular (adults): _____ _____ _____ [ ][ ] 69-70

Thigh (adults): _____ _____ _____ [ ][ ] 71-72

Calf (adults): _____ _____ _____ [ ][ ] 73-74

APPENDIX G

HEALTH QUESTIONNAIRE

Assuring Health for All (AHA) in the Free State

HEALTH QUESTIONNAIRE

(All information in this questionnaire is confidential).

Area in Mangaung:_____ | 1

Household number: | 2-5

Member number (as on socio-demographic form): | 6-7

Interviewer: _____ | 8-9

Interview Date: D D M M Y Y Y Y | 10-17

Marital status: | 18

1. Child
2. Never married
3. Currently married/ Traditional marriage
4. Living with partner
5. Widowed
6. Separated
7. Divorced
8. Other, specify_____

Which best describes your history of smoking? | 19

1. Never smoked
2. Currently smoke
3. Formerly smoked

If yes, how many cigarettes per day?_____ | 20-21

If yes, at what age did you start? _____ | 22-23

Which best describes your history of snuffing? | 24

1. Never used snuff
2. Currently use snuff
3. Formerly used snuff

If yes, how many times per day do you snuff_____ | 25-26

If yes, at what age did you start? _____ | 27-28

Which best describes your history of alcohol use? | 29

1. Never used alcohol products
2. Currently use alcohol products

3. Formerly used alcohol products

If currently, what form of alcohol do you use regularly (at least once a week)? 1=yes 2=no

1. Spirits (rum, whisky, gin, vodka etc.)		30
2. Wine		31
3. Beer		32
4. Homemade beer		33
At least once a month, do you consume >5 alcoholic drinks per day? 1=yes 2=no		34
At what age did you start using alcohol? _____		35-36
On weekends, how many alcohol-containing drinks do you consume?_____		37-38
Do you feel tired on Monday after heavy alcohol consumption (more than 5 drinks per day) during the weekend? 1=yes 2=no		39

Usual sleeping habits:

What time do you usually go to bed at night?_____	:	40-44
What time do you usually wake up in the morning?_____	:	45-49
Do you usually take naps during the day? 1=yes 2=no		50

Current disability: 1=yes 2=no

1. Do you have any trouble walking about?		51
2. Do you have trouble seeing someone across the room (with glasses worn)?		52
3. Do you have trouble reading or seeing individual grains of rice/corn on your plate (with glasses)?		53
4. Do you have trouble speaking and being understood?		54
5. Do you have trouble hearing?		55

Have you experienced any of the following in the last six months? 1=yes 2=no

1. Chest pain or tightness with usual activity		56
2. Breathlessness with usual activity		57
3. Cough for at least 2 weeks		58
4. Wheezing or whistling in the chest		59
5. Loose stools/ diarrhoea for at least 3 days		60
6. Vomiting		61
7. Loss of appetite		62
8. Swelling of feet		63
9. Blood in urine		64
10. Involuntary weight loss of > 3 kg		65
11. Skin rash		66
12. Joint pain		67
13. Sexually transmitted diseases		68

Have YOU ever been diagnosed with the following? 1=yes 2=no

1. Diabetes		69
2. High blood pressure		70
3. Stroke		71
4. Heart disease/ Angina/ Heart attack		72
5. Heart failure		73
6. Cancer		74
7. Liver disease/ Hepatitis/ Jaundice		75
8. Lung disease e.g. emphysema or asthma		76
9. Tuberculosis		77
10. HIV/AIDS		78
11. Epilepsy		79
12. Allergy		80

Has a family member (parents, siblings, children) ever been diagnosed with the following? 1=yes 2=no

1. Diabetes		1
2. High blood pressure		2
3. Stroke		3
4. Heart disease/ Angina/ Heart attack		4
5. Heart failure		5
6. Cancer		6
7. Liver disease/ Hepatitis/ Jaundice		7
8. Lung disease e.g. emphysema or asthma		8
9. Tuberculosis		9
10. HIV/AIDS		10
11. Epilepsy		11
12. Allergy		12

Medication

Are you taking medication regularly (ie. at least once per week) 1=yes 2=no | | 13

If yes, list the medication that you are currently using (including traditional medicine).

_____		14-15
_____		16-17
_____		18-19
_____		20-21
_____		22-23
_____		24-25
_____		26-27

_____ | | | 28-29

During the past 12 months, have you been hospitalised? 1=yes 2=no | | 30

If yes, how many times? _____ | |

If yes, give details (e.g. for specific operation or treatment)_____ | | | 23-32

If yes, for how many days?_____ | | | 33-34

For women only: 1=yes 2=no

1. Are you currently pregnant? | | 35
2. Do you still have periods? | | 36
3. Have you ever used an injectable contraceptive? | | 37
4. How many live children have you given birth to?_____ | | 38
5. Did you breastfeed any of your children? | | 39
6. If yes, at what age (months) did you add anything other than breast milk to the diet?_____ | | | 40-41

Social situation and stress:

Are you a member of a church?_____ 1=Yes 2=No | | 42

Do you attend services at least 2x/month? 1=Yes 2=No | | 43

Stress is defined as feeling irritable or filled with anxiety, or as having sleeping difficulties as a result of conditions at work or at home. How often have you felt stress in the last 2 months? | | 44

1. Never
2. A few periods of stress
3. Several periods of stress
4. Permanent stress

Have you experienced any of the following during the past 12 months? 1=yes 2=no

1. Loss of job | | 45
2. Retirement | | 46
3. Loss of crop/ business failure | | 47
4. Household break in | | 48
5. Marital separation/ divorce | | 49
6. Other major intra-family conflict? If yes, specify _____ | | 50
7. Major personal injury or illness | | 51
8. Violence | | 52
9. Death of a spouse | | 53
10 Death or major illness of another family member | | 54
11. Wedding of family member | | 55
12. New job | | 56

13. Birth in the family		57
14. Separation from family		58
15. Unavailablity of food/ Food insecurity		59
16 Other major stress? If yes, specify _____		60
During the past 12 months, was there ever a time when you felt sad, blue or depressed for two weeks or more in a row? 1=yes 2=no		61
Are you willing to answer questions related to HIV/AIDS? 1=yes 2=no		62
If yes, do you know people who have HIV/AIDS? 1=yes 2=no		63
If yes, which of these people: 1=yes 2=no		
1. Your children		64
2. Your grandchildren		65
3. Your spouse		66
4. Your family members		67
5. Your friends		68
6. People in the community		69
Do you care for orphans in your household? 1=yes 2=no		70

APPENDIX H

SOCIO-DEMOGRAPHIC QUESTIONNAIRE

Assuring Health for All (AHA) in the Free State

SOCIO-DEMOGRAPHIC AND HOUSEHOLD QUESTIONNAIRE

(All information in this questionnaire is confidential).

Area in Mangaung:_____ 1

Household number: 2-5

Member number: 6-7

8-9

Interviewer: _____

Interview Date: D D M M Y Y Y Y 10-17

For each individual living in the household (5-7 days per week), provide the following:

Household Member	Name	Relation to head of household	Sex	Education	Birth date D D M M Y Y Y Y	
01	_____					18-31
02	_____					32-45
03	_____					46-59
04	_____					60-73
05	_____					1-14
06	_____					15-28
07	_____					29-42
08	_____					43-56
09	_____					57-70
10	_____					1-14
11	_____					15-28
12	_____					29-42

For each individual who was living in the house and died within the past 2 years, provide the following:

Name	Relation	Sex	Education	Birth date D D M M Y Y Y Y	
_____					43-54
_____					55-66
_____					67-78
_____					1-12

Household member number of person with whom interview is being conducted: [ ][ ] 13-14

How many years have you been living in an urban area _____ [ ][ ] 15-16

Encircle the appropriate answer:

First language of household: [ ] 17

1. Sotho
2. Tswana
3. English
4. Afrikaans
5. Other, specify_____

Employment status of respondent: [ ] 18

1. Housewife by choice
2. Unemployed
3. Self Employed
4. Full time wage earner (receive a salary)
5. Other, specify (part-time, piece job etc.)_____
6. Don't Know

Husband/ partner's employment status: [ ] 19

1. Retired by choice
2. Unemployed
3. Self Employed
4. Full time wage earner (receive a salary)
5. Other, specify (part-time, piece job etc.)_____
6. Not Applicable e.g. dead

Type of dwelling: [ ] 20

1. Brick, Concrete
2. Traditional mud
3. Tin
4. Plank, wood
5. Other, specify_____

Total number of rooms in house: _____ [ ][ ] 21-22

Number of bedrooms: _____ [ ] 23

Do you have a bathroom in the house? 1=Yes 2=No [ ] 24

Do you have a bathroom outside? 1=Yes 2=No [ ] 25

Do you have a kitchen or cooking area inside the house? 1=Yes 2=No [ ] 26

Does the household have electricity? 1=Yes 2=No [ ] 27

Where do you get drinking water most of the time? [ ] 28

1. Own tap
2. Communal tap
3. River, dam
4. Borehole, well
5. Other, specify _____

What type of toilet does this household have? [ ] 29

1. Flush

2. Pit
3. Bucket, pot
4. VIP
5. Other, specify _____

What fuel is used for cooking most of the time? ☐ 30

1. Electric
2. Gas
3. Parrafin
4. Wood, Coal
5. Sun
6. Open fire

Do you use a cast iron pot for cooking? ☐ 31

1. Never
2. ≤ Once a week
3. > Once a week
4. Every day

Does the home have a working:

Refrigerator and/or freezer ☐ 32
1.Yes
2.No

Stove (Gas, Coal or electric) or Hot Plate ☐ 33
1. Yes
2. No

Primus or Paraffin Stove ☐ 34
1. Yes
2. No

Microwave ☐ 35
1. Yes
2. No

Radio ☐ 36
1. Yes
2. No

Television ☐ 37
1. Yes
2. No

How many people contribute to the total income? _____ ☐☐ 38-39

Household income per month (including wages, rent, sales of vegs, etc. State grants). ☐ 40

1. None
2. R100-R500
3. R501- R1000
4. R1001-R3000
5. R3001-R5000
6. Over R5000
7. Don't know

Is this more or less the income that you had over the past six months? ☐ 41

1. More
2. Less
3. The same

Race of the family: ☐ 42

1. Black
2. Coloured
3. White
4. Mixed