MATERNAL BODY COMPOSITION AND INFANT BIRTH WEIGHT IN URBAN INFORMAL SETTLEMENTS IN NAIROBI CITY COUNTY, KENYA

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DECLARATION

This thesis is my original work and has not been presented for award of a degree in the University or any other University.

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DEDICATION

I dedicate this thesis to my parents, brother, friends and colleagues at the African Population and Health Research Center (APHRC) for their constant encouragement and support in the course of my MSc studies.

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ABBREVIATIONS AND ACRONYMS

AMREF	African Medical Research Foundation
ANC	Antenatal Clinic
APHRC	African Population and Health Research Centre
BMI	Body Mass Index
D_2O	Deuterium Oxide
ERC	Ethical Review Committee
FFM	Fat Free Mass
FM	Fat Mass
FTIR	Fourier Transform Infrared Spectroscopy
IAEA	International Atomic Energy Agency
KEBS	Kenya Bureau of Standard
KEMRI	Kenya Medical Research Institute
Kg	Kilograms
KNBS	Kenya National Bureau of Statistics
KUERC	Kenyatta University Ethical Review Committee
LBW	Low Birth Weight
MIYCN	Maternal Infant and Young Child Nutrition
ml	Millilitres
MUAC	Middle Upper Arm Circumference
NACOSTI	National Commission for Science, Technology and Innovation
NUHDSS	Nairobi Urban Health and Demographic Surveillance System
TBW	Total Body Water
UNFPA	United Nations Population Fund
UNICEF	United Nations Children's Fund
VD	Dilution Space
WHO	World Health Organization

DEFINITION OF TERMS

Birth weight	Body weight at birth (World Health Organization, 2010)
Body composition	Body components that comprise an individual's weight, including total body water, fat free mass and fat mass (International Atomic Energy Agency, 2010)
Deuterium	The stable isotope of hydrogen with the symbol 2H, also abbreviated deuterium concentration in saliva samples from studies of body (International Atomic Energy Agency, 2010)
Deuterium oxide (deuterium water)	Water in which 99.8 or 99.9% of the hydrogen atoms are in the form of deuterium (2H2O or D2O) (International Atomic Energy Agency, 2010)
Deuterium dilution method	A well-established method of measuring body composition from which body composition is estimated using a two compartment model, assuming the body is composed of fat and fat free mass (FFM). FFM is 73.2% water in healthy adults. TBW (kg) / 0.732 = FFM (kg). Fat mass is calculated as the difference between FFM and body weight (International Atomic Energy Agency, 2010)
Deuterium enrichment	The concentration of the deuterium isotope in saliva above the normal concentration (International Atomic Energy Agency, 2010)
Fat free mass	Refers to the part of the body that is not fat (International Atomic Energy Agency, 2010)
Fat mass	Refers to the total fat in the body, calculated by subtracting fat free mass from total weight
Fourier Transform Infrared Spectroscopy (FTIR)	A technique used to measure deuterium concentration in saliva samples from studies of body composition and human milk intake (International Atomic Energy Agency, 2010)
Gravidity	The number of times that a woman has been pregnant (World Health Organization, 2010)
Hydration Factor	Water that is contained exclusively within the fat-free mass, which is approximately 73% of fat free mass in adults, but may vary in

	different physiological status e.g. in infant and during pregnancy (International Atomic Energy Agency, 2010)
Low birth weight	Birth weight less than 2500g regardless of the gestation age which the baby is born (World Health Organization, 2010)
Normal birth weight	Birth weight between 2500g and 4000g regardless of the gestation age when the baby is born (World Health Organization, 2010)
High birth weight	Birth weight of more than 4000g regardless of the gestation age when the baby is born (World Health Organization, 2010)
Parity	Number of deliveries that occurs after viability of the pregnancy, usually after 28 weeks (World Health Organization, 2010)
Stable Isotope	Non-radioactive isotopes, for example 2H (Deuterium–Hydrogen isotope), 13C (carbon-13), 15N (nitrogen-15), 18O (oxygen-18). (International Atomic Energy Agency, 2010)
Total body water	Total water content of the body. This includes both intracellular and extracellular fluid (International Atomic Energy Agency, 2010)

OPERATIONAL DEFINITION OF TERMS

Informal Settlements	Areas where groups of housing units have been constructed on land that occupants have no legal claim to, or occupy illegally and hence have limited access to basic infrastructure and amenities including health, education, sanitation etc.
T_0 saliva sample	Saliva sample collected before deuterium ingestion (Baseline Saliva sample), also known as pre-dose sample.
T_1 Saliva sample	Saliva sample collected three hours post deuterium ingestion
T_2–Saliva sample	Saliva sample collected four hours post deuterium ingestion.
Deuterium Enrichment	Concentration of deuterium in the body water above the normal level (0.015 atom %) as a result of the deuterium dosage consumed

ABSTRACT

Nutrition depletion before and during pregnancy compromises foetal programming, and is among the causes of adverse birth outcomes, including low birth weight. Mothers with lower body water and fat free mass are more likely to have children with lower birth weights. Direct maternal body composition assessments during pregnancy are better predictors of birth outcomes than traditional nutritional assessment methods. However, there is paucity of evidence on maternal body composition and the relationship between maternal body composition and birth weight in urban informal settlements in Kenya. The purpose of this study was to assess maternal body composition using the deuterium dilution technique, and establish the relationship between various maternal body components (total body water, fat mass and fat free mass) and birth weight among pregnant mothers in Korogocho and Viwandani informal settlements in Nairobi City. This study was nested in the Maternal Infant and Young Child Nutrition study implemented by the African Population and Health Research Center in Korogocho and Viwandani, in which a cohort of 1000 women were recruited during pregnancy and followed until one year after delivery. For this study, sub sample of 129 participants before their third trimester were recruited to participate in the body composition assessment, using the deuterium dilution technique, and the birth weight of their children was established after delivery. Information on maternal, socio-economic, demographic and health characteristics was also collected. Data collected was analysed using STATA statistical package. Descriptive analysis were used to present participants' body composition characteristics while linear regression analysis was used to determine the relationship between maternal body composition and birth weight at 95% confidence interval (CI). There was a significant relationship between maternal age and gestation stage, with total body water (TBW), fat free mass (FFM) and fat mass (FM) (linear regression, p=0.05). TBW was significantly higher among older mothers, that is 20 to 24 (33.23L, p=0.044), 25 to 30 (34.28L, p=0.009) and 30 years and above (34.31L, p=0.001), compared to those who were less than 20 years (31.11L), and higher among mothers in the second trimester (33.77L, p=0.01) compared to those in the first trimester (31.06L). FFM was significantly higher among participants aged 20 to 24 years (45.56kg, p=0.031), 25 to 29 years (46.97kg, p=0.007), and 30 years and above (47.0kg, p=0.008) compared to those less than 20 years (42.66kg) and higher among participants in the second trimester (46.26kg, p=0.07) compared to those in the first trimester (42.59kg). FM increased significantly for participants aged 20 to 24 (17.47kg, p=0.001), 25 to 29 (16.75kg, p=0.002) and 30 to 35 (21.3kg, p=0.000) years compared to those in the younger than 20 years (13.81kg). The mean birth weight was 3.3kg $\pm$ 0.42kg. Infant's birth weight was positively related to maternal total body water and fat free mass (linear regression, p=0.005, p=0.003 respectively) but not fat mass. These findings suggest that non – fat components of the body, including total body water and fat free mass, positively influence the birth weight of children. In conclusion, it is crucial for governments and other relevant stakeholders to design interventions that improve optimal maternal feeding practices that would subsequently enhance gains in fat free mass and total body water and hence overall birth weight of infants especially among teenage mothers.

CHAPTER ONE: INTRODUCTION

1.1 Background to the study

Birth weight has been demonstrated as an important predictor of child health and nutrition and survival. Normal birth weight is considered as infant weight of between 2.5kg and 3.9kg within the first hours of birth (World Health Organization, 2010), low birth weight (LBW) that is, weight of less than 2.5 kg within the first hours of birth is associated with increased risk of neonatal death (Lawn, Cousens, & Zupan, 2005) and child under-nutrition (Christian et al., 2013). Child under-nutrition is in turn associated with adverse health and developmental consequences from childhood through to adulthood (Victora, 2008). In the concept of foetal origins of adult disease (FOAD), Barker et al hypothesize that growth deprivation in early life have a profound impact on one's risk for development of future adult disease. Low birth weight is highlighted in the FOAD concept as an indication of poor foetal growth and nutrition and is linked to coronary artery disease, hypertension, obesity, and insulin resistance (Barker, 2004; Barker, Osmond, Kajantie, & Eriksson, 2009). Similarly, high birth weight, which is infant's weight of more than 4.0 kg is a major risk factor for childhood obesity, and consequent adulthood obesity, and other chronic diseases (Reilly et al., 2003; Weng, Redsell, Swift, Yang, & Glazebrook, 2012).

Maternal body composition during pregnancy is an indicator of maternal nutrition and an important predictor of birth outcomes, including birth weight (McCarthy, Strauss, Walker, & Permezel, 2004). Body composition is described as the body components that comprise an individual's weight. The two compartment model of body composition assessment subdivides the human total bodyweight into fat mass (FM) and fat free mass (FFM). Fat mass refers to the total amount of fat in the body, while fat free mass refers to the part that is non-fat, including proteins, bone mass, non-bone mass and total body water (TBW) (Widen & Gallagher, 2014). Total body water refers to the total water content of the body, including intracellular and extracellular body water. In normal adults, TBW is approximately 73% of the FFM (International Atomic Energy Agency, 2010; Lof & Forsum, 2004).

Maternal body composition and nutrient stores during pregnancy influence the availability of nutrients for foetal growth and development with significant effects on foetal genetic development (Wu, Bazer, Cudd, Meininger, & Spencer, 2004), immune function (Phillips, 2006) and cardiovascular function (Poston, 2011) and birth weight (Forsum, Lof, Olausson, & Olhager, 2006; Gernand et al., 2012; Kent et al., 2013). As a result, various body composition changes occur during pregnancy to cater for foetal growth and development, including accretion of fat mass (Kopp-Hoolihan, van Loan, Wong, & King, 1999), increase in fat free mass (Contreras et al., 2012) and increase in total body water (Widen & Gallagher, 2014).

Mounting evidence on the role of maternal body composition on foetal growth and development underscores the need to understand the relationship between the various maternal body composition during pregnancy, and birth weight. It is important to note that direct body composition assessment methods such as the Deuterium dilution technique are more objective in body composition assessment than traditional methods like anthropometric and skinfold measurements (McCarthy et al., 2004; Zhao, Li, & Li, 2014b).

Deuterium dilution technique is a gold standard of direct body composition assessment, it is based on the assumption that the water content of FFM is relatively constant at approximately 73 % in normal adults and varies between 0.723% and 0.747% during pregnancy depending on the gestation period, as pregnancy progresses (Lof & Forsum, 2004). In this technique, deuterium water is orally administered to the participants and allowed to mix with the body water for an equilibration time of between 3 to 5 hours, after which the deuterium concentration of either plasma, urine or saliva is measured and used to calculate TBW, FFM and FM (International Atomic Energy Agency, 2010). Deuterium dilution technique has been used in various studies to assess maternal body composition during pregnancy (International Atomic Energy Agency, 2010; Mardones-Santander, Salazar, Rosso, & Villarroel, 1998). It is relatively affordable and easily applicable in the field compared to other direct body composition assessment methods such as the Dual-energy X-ray Absorptiometry (DEXA) technique (Bert 2004; Ellis, 2001).

Urban informal settings are characterized by poor maternal and child health indicators, mainly due to high levels of poverty and poor access to health services (United Nations Human Settlements Programme, 2003). In Kenya, urban slums have specifically been demonstrated to have poorer child health and nutrition indicators compared to their rural counterparts (Kimani-Murage, Fotso, et al., 2014). No study, to the best of knowledge have been conducted to establish maternal nutrition status using the body composition assessment and its association with birth weight in such settings despite the documented poor infant and child nutrition and health indicators. In addition, of the literature available on the use of stable isotope technique to assess body composition is mainly from developed countries.

This study therefore aimed to establish the body composition status of pregnant women using the deuterium dilution technique and investigate the relationship between maternal body composition and birth weight among women residing in two urban informal settlements in Nairobi, Kenya.

1.2 Problem statement

Kenya has made substantial progress in child survival over the past decade, but 52 children out of 1000 live births still die before attaining the age of five years (Kenya National Bureau of Statistics and ICF Macro, 2015). Majority (over 40%) of these deaths happen within the first month of life with a neonatal death rate of 22 deaths per 1,000 live births (Kenya National Bureau of Statistics and ICF Macro, 2015), translating to 40,000 annual deaths in the first month of life (Organisation, 2017). In the recent national demographic and health survey, Nairobi County recorded the highest infant and neonatal deaths in the country (55 and 39 deaths in 1000 live births respectively), neonatal mortality was also 24 percent higher in urban areas than it is in rural areas (26 deaths versus 21 deaths per 1,000 live births) (Kenya National Bureau of Statistics and ICF Macro, 2015). In addition, urban informal settlements, where over 70% of urban dwellers in the country reside (United Nations Human Settlement Programme, 2003) has higher infant mortality rates (57 in 1000 live births) and child mortality rates (24 in 1000 live births) compared to other sub-populations in Kenya (Kimani-Murage, Fotso, et al., 2014).

Low birth weight has globally been found to be an not only an important cause of neonatal and infant deaths (Lawn, Cousens, Zupan, & Lancet Neonatal Survival Steering, 2005), but also an important risk factor for chronic disease in later life, as highlighted in the Barkers hypothesis (Barker et al., 2009). Approximately 8% of infants in Kenya have a low birth weight leading to an estimated 123,000 babies born too small (LBW) and Nairobi county is among the counties with the highest prevalence of low birth at 8.9% (Kenya National Bureau of Statistics and ICF Macro, 2015). Studies in urban informal settings have documented low birth weight as a significant contributor to premature child deaths and that majority of the children born with low birth weight in these settings are stunted before the age of 5 years (Abuya, Ciera, & Kimani-Murage, 2012; Kyobutungi, Ziraba, Ezeh, & Ye, 2008).

Maternal nutrition deprivation during pregnancy is a major cause of low birth weight (Christian et al., 2013; Gernand et al., 2012; Lawn et al., 2005). Maternal nutrition as measured by direct body composition assessment is a better predictors of birth outcomes compared to traditional methods of nutrition assessment (McCarthy et al., 2004; Zhao, Li, & Li, 2014a). In addition, body composition as a measure of nutrition status enables the assessment of individual body component (TBW, FFM, FM), which the traditional methods such as anthropometry do not. The deuterium dilution technique is a gold standard method of direct body composition assessment However, it has not been used to establish the body composition of pregnant women in the urban informal settings. Further, despite the fact that low birth weight in Nairobi's informal settlements contributes largely to child mortality and poor nutrition, there is paucity of evidence on the relationship between maternal body composition in pregnant women and its relationship with birth outcomes (birth weight) in the same setting.

1.3 Purpose of the study

The purpose of this study was to establish the body composition of pregnant women in the first and second trimester using the deuterium dilution technique and its relationship with birth weight in two informal settlements (Korogocho and Viwandani) in Nairobi County.

1.4 Objectives of the study

1. To determine the total body water of pregnant women in Korogocho and Viwandani informal settlements

2. To determine the fat free mass of pregnant women in Korogocho and Viwandani informal settlements

3. To determine the fat mass of pregnant women in Korogocho and Viwandani informal settlements

4. To establish the birth weight of infants in Korogocho and Viwandani informal settlements

5. To establish the relationship between maternal body composition (total body water, fat free mass and fat mass) and infant birth weight in Korogocho and Viwandani informal settlements

1.5 Hypotheses

$H0_1$: There is no significant relationship between maternal total body water and birth weight of infants in Korogocho and Viwandani informal settlements.

$H0_2$: There is no significant relationship between maternal fat free mass and birth weight in Nairobi informal settlements.

$H0_3$: There is no significant relationship between maternal body fat and birth weight in Nairobi informal settlements.

1.6 Significance of the study

Evidence on the body composition of pregnant women in urban informal settlements and the relationship between maternal body composition and birth weight generated from this study is useful in informing the designing of interventions and programs aimed at improving maternal and child health and reducing the number of children born with low birth weight in urban informal settlements. The findings are significant to government agencies such as the Ministry of Health (Unit of Nutrition), the County government of Nairobi (department of health and nutrition), and other stakeholders focusing on maternal and child health.

There is paucity of evidence on body composition assessment among pregnant women in Kenya and other African countries, using the stable isotope technique. This study fills this gap by providing evidence on the body composition of pregnant women in urban informal settings. The study could also provide evidence to other researchers on the use of the stable isotope – deuterium dilution technique in maternal body composition assessment during pregnancy in resource-limited settings.

1.7 Delimitations of the study

The study focused on pregnant women before the third trimester (first and second trimester) only, since the deuterium dilution technique is more reliable for body composition during this pregnancy period, the variations in body composition changes in the third trimester limits its use during this trimester (International Atomic Energy Agency, 2010). The study also focused on urban informal settlements, hence, generalization of the study findings, can only be done to areas and populations of similar characteristics.

1.8 Limitations

Currently there are no published cut-off points for maternal body composition during pregnancy. This study therefore did not categorise maternal body composition (TBW, FFM and FM), but just described the various body composition levels and their relationship with birth weight. In addition, body composition was assessed at one point during pregnancy before the third trimester and other body composition changes in the third trimester which could have an implication on birth weight were not assessed.

1.9 Assumptions

This study used the assumption that hydration of fat free mass for pregnant women in early gestation (1^{st} and 2^{nd} trimesters) is 73 % as reported in other studies (International Atomic Energy Agency, 2010; Lof & Forsum, 2004).

1.10 Conceptual framework

The study adopted the conceptual framework from Chuku et al., (2008). It illustrates that maternal characteristics (socio-economic and demographic characteristics, health and health seeking behaviour) and infant characteristics (gender) have a correlation with birth weight. A significant relationship has been documented between birth weight and maternal weight, height, age, education, ANC attendance and wealth index (Chuku, 2008); maternal socio economic status, religion and marital status (Gathimba, 2014); parity (Karim & Mascie-Taylor, 1997) and health status (Rao et al., 2018). Cchild's sex has also been found to have a significant association with birth weight (Muchemi, Echoka, & Makokha, 2015). Existing literature also reveals some correlation between maternal body composition (total body water, fat free mass and fat mass) and birth weight with several studies recording a positive association especially between birth weight and body water (Butte, Ellis, Wong, Hopkinson, & Smith, 2003; Farah, Stuart, Donnelly, Kennelly, & Turner, 2011; Forsum et al., 2006; Ghezzi et al., 2001; Zhao et al., 2014b).

This study focused on the relationship between maternal TBW, FFM, and FM among pregnant mothers and the birth weight of their infants, in urban informal settings. However, other factors including maternal socio-economic characteristics, socio-demographic characteristics, heath condition, and health seeking behaviour and infant characteristics that could confound the relationship between maternal body compositions and birth weight as illustrated in figure 1.1, were also included in the analysis of data in this study to control for confounding.

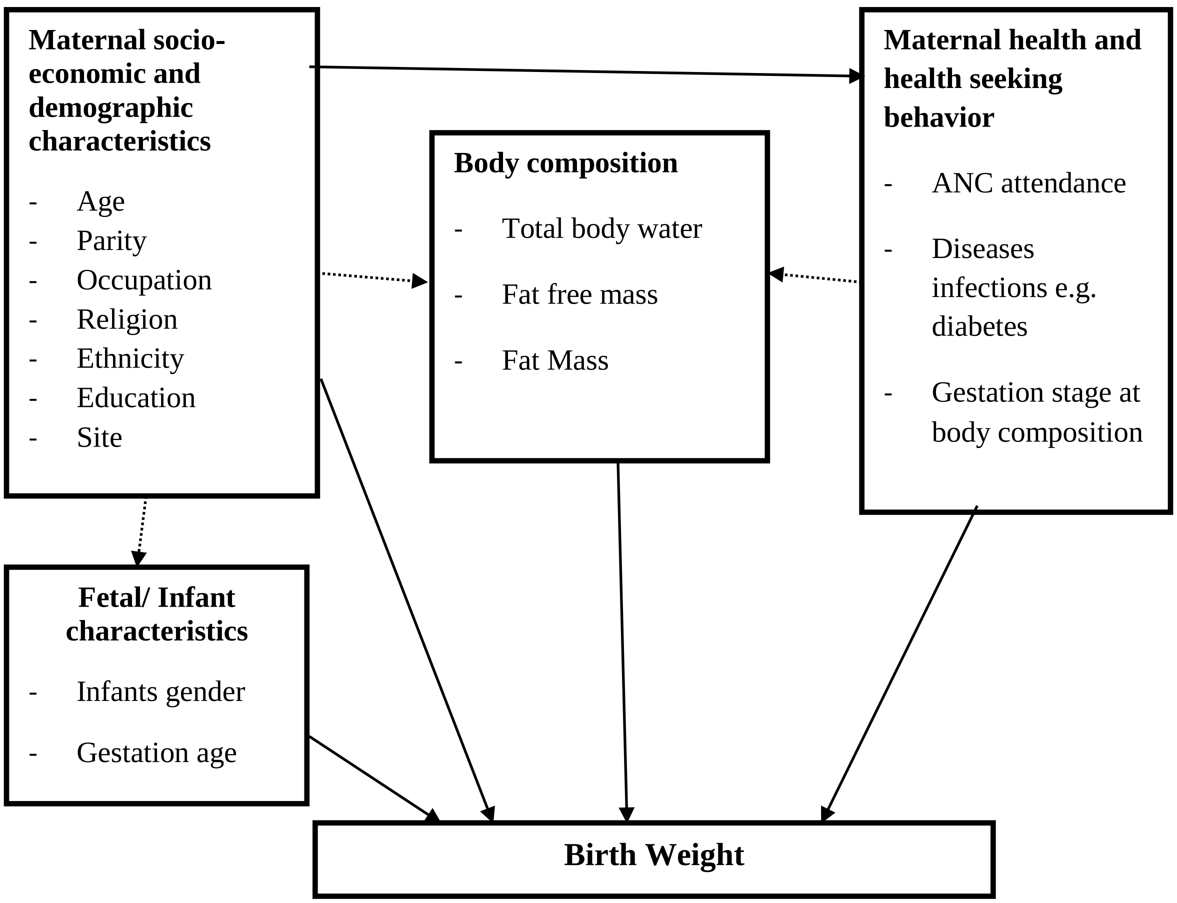

Figure 1.1: Conceptual framework on the relationship between maternal body composition and birth weight.

Source: (Chuku, 2008)

CHAPTER TWO: LITERATURE REVIEW

2.1 Body composition assessment methods

Body composition refers to the various components that comprise an individual's weight, including total body water, fat free mass and fat mass (International Atomic Energy Agency, 2010). It is a measure of an individual's nutrition status and has been shown to be a good predictor of birth weight (McCarthy et al., 2004). The deuterium dilution technique is one of gold standard body composition measurements, that has been used to assess the body composition of pregnant women (International Atomic Energy Agency, 2010; Mardones-Santander et al., 1998). Other methods that have been used for direct body composition include under water weighing also known as hydrodensitiomentry, bioelectric impedance analysis and Dual-energy X-ray Absorptiometry (DEXA) techniques (Duren et al., 2008).The stable isotope is relatively affordable and easily applicable in the field compared to underwater weighing and DEXA (Bert 2004; Ellis, 2001).Anthropometric measurements including weight, height, middle – upper arm circumference and skin fold measurements have also been used for indirect body composition assessment during pregnancy, however, stable isotope technique is more precise and objective compared to anthropometry, and is considered a gold standard method of body composition assessment (McCarthy et al., 2004; Zhao, Li, & Li, 2014b, Duren et al., 2008).

Literature reviewed focused on the use of deuterium dilution technique in body composition assessment during pregnancy, maternal body composition in Kenya and other countries, factors associated with body composition during pregnancy, birth weight at global level and in Kenya and the relationship between maternal body composition during pregnancy and birth weight.

2.1.1 Establishing body composition using the deuterium dilution technique

Deuterium isotope is relatively cheap and non- radioactive compared to previously used tritium isotope, which is very expensive and radioactive, deuterium isotope is also more commonly used for the dilution technique of body composition assessment. (Ellis, 2001). The standardized dose of deuterium isotope for estimating TBW in adults is 30 g for all adult body weights. Deuterium oxide is handled in the body in the same way as water. After mixing with body water, it is eliminated from the body in urine, saliva, sweat and human milk within hours (International Atomic Energy Agency, 2010). The deuterium dilution technique is recognised as a safe body composition assessment method, and has been used to assess body composition in pregnant women and breast milk intake in children in Kenya and other parts of the world with no reports of adverse effects (Ettyang, van Marken Lichtenbelt, Esamai, Saris, & Westerterp, 2005; Kupka et al., 2011; Lof & Forsum, 2004).

2.2 Maternal body composition

There are few published studies on maternal body composition during pregnancy in Kenya. One study conducted mong post-partum women in pastoral communities found that mean body fat percentage measured using the deuterium dilution technique was 26.1%, while the mean FFM was 41.3 kg. Low levels of fat mass in this study were associated with malnutrition and associated indicators. (Ettyang, van Marken Lichtenbelt, Oloo, & Saris, 2003). In a different study done among non-pregnant women in urban and rural setting in Narok County using the deuterium dilution technique, the mean body fat percentage of the women was 30.7% while mean FFM was 38.9kg. Both body fat percentage and fat mass were slightly lower in urban women compared to rural women in this county, although the difference was not significant (Keino, van den Borne, & Plasqui, 2014). Notably, most of the maternal body composition studies conducted in Kenya, have focused on non-pregnant and lactating women, no literature was found on maternal body composition among pregnant women, using the stable isotope technique in Kenya.

2.2.1 Maternal during pregnancy

2.2.1.1Total body water (TBW)

Total body water includes the intracellular and extracellular water in the body. International Atomic Energy Agency (IAEA) estimates that water makes up to approximately 70 to 75% of an individual’s body weight at birth, but decreases to about 40 to 60 % in adulthood (International Atomic Energy Agency, 2010). A systematic review of predictors and outcomes of body composition changes during pregnancy also reveals gains in total body water during pregnancy (Widen & Gallagher, 2014). Other studies have documented an increase in both intracellular and extracellular water content in healthy pregnant women (Ghezzi et al., 2001; Larciprete et al., 2003), but a decrease in body water content in hypertensive women due to decreased plasma volume (Valensise et al., 2000).

The increase in total body water in the course of pregnancy is thought to be among the reasons for weight gain during pregnancy (Butte et al., 2003). Among the components that contribute to increased total body water during pregnancy is the increased blood volume and extracellular fluids (Institute of Medicine (US) Committee on Nutritional Status During Pregnancy and Lactation, 1990). Ghezzi et al, further indicate that total, intracellular and extracellular water significantly increase as pregnancy advances and return to the pre-pregnancy values within 60 days after delivery (Ghezzi et al., 2001).

2.2.1.2 Fat Free mass (FFM)

Maternal FFM, which entails, the protein tissues, bone and none bone minerals have been shown to increase during pregnancy. In USA, fat free mass measured using an infrared spectroscopy method increased during pregnancy (Azizian, Kramer, & Phillips, 2014). In Mexico, Contreras et al., (2012) found an increase in FFM among teenage and mature mothers, using bioelectrical impedance method body composition assessment. However there was a depletion of fat free mass reported among pregnant teenagers in rural Bangladesh (Rah et al., 2008). The World Health

Organisation reports that teenagers may not yet be fully grown, and continue to grow even during pregnancy (World Health Organisation, 2004), it is also documented that nutrition requirements for teenagers is further heightened by pregnancy (Islam, Islam, Bharati, Aik, & Hossain, 2016). This may be the reason for the lower body fat free mass levels among teenagers compared to mature women. The uterine tissue, mammary and soft tissue grow progressively during pregnancy to support foetal development and are thought to contribute to the increased fat free mass during pregnancy (Institute of Medicine (US) Committee on Nutritional Status During Pregnancy and Lactation, 1990).

2.2.1.3 Fat Mass (FM)

Similar to TBW and FFM, FM has been shown to increase during pregnancy. A study in Sweden revealed significant maternal fat accumulation during pregnancy which was directly related to maternal weight gain but had little effect on birth weight (Langhoff-Roos, Lindmark, & Gebre-Medhin, 1987). Kopp- Hoolihan et al, (1999) also found an increase in fat mass during pregnancy, with large inter-individual variability in the course of pregnancy, in the United States of America (USA), It is hypothesized that the fat mass accumulation is also partly responsible for the weight gain during pregnancy (Larciprete et al., 2003). Majority of the fat deposition during pregnancy occurs more around the thighs, subscapular region and in the abdomens subcutaneous and preperitonial layers (Kinoshita & Itoh, 2006). In addition, optimal fat storage during pregnancy is considered essential for the increasing maternal energy requirements during pregnancy, it also promotes optimal foetal and infant development and breast milk production post-partum (Crawford, Hassam, & Stevens, 1981; G. Hornstra, 2000).

2.2.2 Factors associated with maternal body composition

Various studies have documented a corelation between maternal wealth index and maternal age with fat mass. In malawi, wealthier women were found to have a higher arm fat area compared to those with lower wealth index (Ramlal et al., 2013); while adolescent women were found had lower fat mass as compared to mature women in a mexican study (Contreras Campos et al., 2012)

Maternal preconception BMI, parity, age and infant's gender have also been found to affect maternal body composition during pregnancy. In a prospective study by sidebottom et al, women with high preconception BMI had less body fat gains in early pregancy compared to the those with low preconception BMI while primiparous and women carrying males had higher thigh and subscapular body fat gains (Sidebottom, Brown, & Jacobs, 2001) compared to nulliparous women and those carrying female children.

Seasonal variation of body composition has been documented in some studies, for instance, Benise women had significant increase in fat free mass during the post-harvest seasons compared to pre-harvest seasons ((Schultink, Lawrence, van Raaij, Scott, & Hautvast, 1992) while in Malawi exposure to famine was found to cause large losses in arm muscle among HIV infected women (Lederman et al., 1999).

A study on body fat composition in black population from Nigeria, Jamaica and the United States (US) indicates that gender, environmental and ethnicity may influence body composition. Participants from the US had the highest body fat composition while Nigerians had the lowest (Lederman et al., 1999) ethnic influence on body composition is also depicted in a study comparing Singapore, Malaysian and Indian ethnic groups, the results in this study showed significance differences in body composition among these populations with Chinese women having the lowest body fat percentage compared to the Malaysian and Indian women (Deurenberg-Yap, Schmidt, van Staveren, Hautvast, & Deurenberg, 2001).

2.3 Low birth weight in Kenya

The world health organisation classifies birth weight into normal birth weight (2.5kg to 3.9 kg), low birth weight (< 2.5 kg) and high birth weight (≥ 4.0 kg) (World Health Organization, 2010).

Normal birth weight is considered essential for optimal infant and child health, growth and development. Low birth weight (LBW) is associated with increased risks of neonatal deaths (Lawn et al., 2005) and child under-nutrition (Christian et al., 2013), which are further associated with adverse health and developmental consequences in childhood and adulthood (Victora, 2008). (Abu-Saad & Fraser, 2010; Risnes et al., 2011). The barker hypothesis further highlights low birth weight as an important risk factor for chronic diseases in later life including coronary heart diseases, diabetes and hypertension (Barker, 2004).

About 15% of all births in the world are low birth weight (UNICEF, 2004), Majority (95.6%) of which are reported from low and middle income developing countries. In addition about 40 – 80% of all neonatal deaths in the world are attributed to low LBW (Bhutta, Darmstadt, Hasan, & Haws, 2005).

In Kenya, the recent Kenya Demographic and Health Survey reported that 8% of children had low birth weight. Children in urban settings had a slightly higher rates of low birth weight (8.6%) compared to those in rural setting (6.7%). Further, Nairobi county had among the highest prevalence's of low birth weight (8.9%), slightly higher than the national rate (8.9%) (Kenya National Bureau of Statistics and ICF Macro, 2015).

Statistics from the Nairobi cross-sectional survey in 2012 indicated that Nairobi informal settlements had a notable prevalence of LBW (6.3% in some areas) (African Population and Health Research Center, 2014). In addition, urban informal settlements have higher infant mortality rates (75 in 1000 live births) compared to other sub-populations in Kenya (Kimani-Murage, Fotso, et al., 2014), and low birth weight is documented as one of the major contributors to premature child deaths (Kyobutungi et al., 2008) and stunting among children under 5 years (Abuya et al., 2012) in the urban slums.

2.3.1 Factors associated with low birth weight

Maternal and infant characteristics have been shown to influence birth weight. In the central region of Kenya, child's sex was found to have a significant association with birth weight, with a higher likelihood of low birth weight among girls (Muchemi et al., 2015) while Maternal socio economic status, religion and marital status had a significant association with birth weight in Nairobi county, Kenya (Gathimba, 2014). In the Kenyan coastal region, maternal age was found to have a significant association with birth weight, with higher risks of low birth weight among children born by older mothers (above 25 years) (Fairley et al., 2013). In Lesotho, Mathule et al., (2005) found an association of birth weight with maternal age, parity and birth weight, while in Bangladesh, maternal education and socio economic status had a significant association with birth weight (Karim & Mascie-Taylor, 1997). In addition, Chuku et al., 2008, found a positive correlation between birth weight and maternal weight, height, age, education and wealth index (Chuku, 2008).

2.4 Relationship between maternal body composition and birth weight

2.4.1 Relationship between maternal total body water and birth weight

Evidence from several studies shows a positive correlation between maternal TBW during pregnancy and birth weight. For instance, Ghezzi, in Italy found maternal TBW measured using bioelectric impedance to be positively related with birth weight (Ghezzi et al., 2001). Increase in TBW during pregnancy was also found to have a positive correlation with birth weight in USA, and thought to be a contributor to maternal weight gain during pregnancy (Butte et al., 2003). In Africa, a study done in Tanzania found TBW in the first and second trimester measured using the deuterium dilution technique to have a positive correlation with birth weight among HIV infected mothers (Kupka et al., 2011). In rural Bangladesh, lower TBW in all stages of pregnancy was found to be a risk factor for foetal growth restriction, a major cause for low birth weight (Gernand et al., 2012). However, an evidence gap on maternal body water during pregnancy and its relationship with birth weight among low income settings in Kenya was noted, despite the documented adverse effects of low birth on child health outcome since the same settings (Kyobutungi et al., 2008)

2.4.2 Relationship between maternal fat free mass and birth weight

Maternal FFM, is correlated positively with birth weight and predicts birth weight (Butte et al., 2003; Thame, Osmond, & Trotman, 2013). Longitudinal studies among Caucasian women found that FFM has a strong influence on birth weight (Farah et al., 2011). In Ireland, maternal FFM had a strong correlation with birth weight, pregnant women with high fat free mass were more likely to have bigger babies (Kent et al., 2013). Similarly, in South Chile, maternal fat free mass in late pregnancy had the strongest influence on birth weight (Mardones-Santander et al., 1998). Among Chinese pregnant women, fat free mass was found to be the most important predictor of birth weight, mothers with a fat free mass of more than 40kg were more likely to have babies with high birth weight (>4kg)(Wang et al., 2017). No study was found, documenting the relationship between maternal fat free mass during pregnancy and birth weight in Kenya, including the urban informal setting.

2.4.3 Relationship between maternal fat mass and birth weight

There are varying findings on the relationship between fat mass and birth weight. For instance, Zhao et al., (2014) found maternal fat percentage to be a better predictor of birth outcomes compared to body mass index measurement. In Sweden, maternal body fat composition as measured using the deuterium dilution technique was found to influence foetal growth positively and consequently the birth weight (Forsum et al., 2006). In contrast, some studies in USA, Mexico, Ireland, and Sweden found no significant relationship between fat mass at different gestational ages and birth weight (Butte et al., 2003; Contreras et al., 2012; Kent et al., 2013; Langhoff-Roos et al., 1987; Lederman et al., 1999). There was hence no consensus on the relationship between fat mass and birth weight, among the studies reviewed, further more a dearth of studies on maternal fat free mass during pregnancy and its relationship with birth weight in Kenya and the African context was noted.

2.5 Summary of literature review

Literature reviewed indicates that TBW, FFM and FM increase considerably during pregnancy. It is predicted that the changes in maternal body composition observed during pregnancy cater for foetal growth and development (Bhardwaj, Verma, & Kapoor, 2013; Lof et al., 2005). Maternal FFM and TBW are positively correlated with birth weight. Low TBW was associated with intrauterine growth retardation, a major cause of low birth weight, while high fat free mass was associated with high birth weight babies (Wang et al., 2017). There is no consensus on the relationship between maternal fat mass and birth weight. In Kenya, Urban areas including Nairobi County have higher rates of low birth weight compared to other populations and low birth weight is associated with poor child health and nutrition in urban informal settings, however, there were no studies found, documenting the relationship between maternal body composition and infant's birth weight in these settings. In addition, there is little evidence on the body composition of pregnant women in Kenya and sub-Saharan Africa, particularly using the stable isotope deuterium dilution technique. The relationship between maternal body composition and birth weight is also not extensively studied in the African context. This study seeks to address these gaps by establishing the maternal body composition of pregnant women in urban informal settlements in Kenya using the stable isotope-deuterium dilution technique, and the relationship between maternal body composition during pregnancy and infant's birth weight.

CHAPTER THREE: METHODOLOGY

3.1 Research design

This study adopted a prospective cross-sectional design, using quantitative methods in data collection, analysis and presentation (Mugenda, 2003). A cross-sectional prospective study can be used to test for associations among variables, this design was therefore appropriate for this study. Maternal body composition assessment was conducted during pregnancy, the participants were then followed up after delivery to establish their infants' birth weight.

This study was conducted between December 2012 and June 2014 nested within a broader maternal infant and child nutrition (MIYCN) study, which involved a follow-up of a cohort of over 1000 pregnant women and their respective children after birth, in Korogocho and Viwandani informal settlements. The women in the MIYCN study were followed up from pregnancy until one year after delivery, in order to determine the nutritional status of mothers and children in this cohort (Elizabeth W Kimani-Murage, 2014). The MIYCN study was implemented from 2012 to 2014 and was nested in the Nairobi Urban Demographic Health Surveillance System (NUDHSS), which is run by the African Population and Health Research Center from 2002. The NUHDSS covers approximately 65,000 people living in about 24,000 households in the two informal settlements, and involves a quarterly follow-up of the residents. Key demographic data on pregnancy and pregnancy outcomes, migration, birth, occupation or schooling, health-seeking behavior, morbidity, mortality and causes of death among others are collected every three months. The main objective of the NUDHSS is to provide a platform for investigating linkages between urbanization, health, poverty and other demographic and socioeconomic outcomes among the urban poor (Emina, Beguy, Zulu, Ezeh, Muindi, Elung'ata, et al., 2011).

3.2 Study location

The study was conducted in the Korogocho and Viwandani informal settlements of Nairobi County. The Korogocho informal settlement is located in Kasarani sub-County, about 12 kilometres from the city centre. It is one of the most densely populated informal settlements in Nairobi with about 65,000 inhabitants per square kilometre. The Viwandani Informal settlement is located in Makadara sub-County, about 7 kilometres from the city centre. It is situated in the industrial area of the city and has a population of approximately 53,000 inhabitants per square kilometre.

Since they are not recognized as legal settlements, the two informal settlements are characterized by poor access to basic amenities including portable water, waste disposal, health and education (Abuya et al., 2012; Emina, Beguy, Zulu, Ezeh, Muindi, & Elung'ata, 2011; Kimani-Murage, Fotso, et al., 2014; Kimani-Murage & Ngindu, 2007). They also have poor housing, mainly mud and iron sheets, high levels of food insecurity and are exposed to high levels of unemployment, violence and teenage pregnancy (Beguy, Ndugwa, & Kabiru, 2013; Kabiru, Beguy, Undie, Zulu, & Ezeh, 2010; Kimani-Murage, Schofield, et al., 2014). Poor maternal child health and nutrition indicators are also highly documented in these settings (Kimani-Murage, Fotso, et al., 2014; Ziraba, Madise, Mills, Kyobutungi, & Ezeh, 2009), which justified the selection of the two informal settings as the study areas.

3.3 Measurement of study variables

The dependent variable was infants' birth weight in grams/ kgs as established from the mother-child booklet where birth weight is recorded immediately after delivery. The independent variables were; total body water, fat free mass and fat mass as measured using the deuterium dilution method. The variables controlled for during analysis included maternal health and health seeking behaviour, socioeconomic characteristics and socio-demographic characteristics and infant characteristics (Table 3.1).

Table 3.1 : Study's dependent and independent variables

Dependent Variables	**Measurement**	**Independent Variable**	**Measurement**
Birth Weight	In grams as recorded in the mother – child booklet , after delivery	Maternal total body water	In litres as measured using the stable isotope technique during pregnancy
		Maternal fat free mass	In Kgs as measured using the stable isotope technique during pregnancy
		Maternal fat mass	In Kgs as measured using the stable isotope technique during pregnancy
Birth Weight	In grams as recorded in the mother – child booklet , after delivery	Socio-economic characteristics	Highest level of maternal education and current occupation
		Socio–demographic characteristics	Maternal age in complete years, parity, religion, and ethnicity
		Maternal health characteristics	(Gestation stage (trimester) at body composition assessment, maternal illnesses or complications during pregnancy and antenatal clinic (ANC) attendance).
		Infant characteristics	Infant' sex as recorded from mother – child booklet , after delivery
Maternal total body water	In litres as measured using the stable isotope technique during pregnancy	Socio-economic characteristics	Highest level of maternal education and current occupation
		Socio–demographic characteristics	Maternal age in complete years, parity, religion, and ethnicity
		Maternal health characteristics	(Gestation stage (trimester) at body composition assessment, maternal illnesses or complications during pregnancy and antenatal clinic (ANC) attendance).
		Infant characteristics	Infant' sex as recorded from mother – child booklet , after delivery
Maternal fat free mass	In Kgs as measured using the stable isotope technique during pregnancy	Socio-economic characteristics	Highest level of maternal education and current occupation
		Socio–demographic characteristics	Maternal age in complete years, parity, religion, and ethnicity
		Maternal health characteristics	(Gestation stage (trimester) at body composition assessment, maternal illnesses or complications during pregnancy and antenatal clinic (ANC) attendance).
		Infant characteristics	Infant' sex as recorded from mother – child booklet , after delivery
Maternal fat mass	In Kgs as measured using the stable isotope technique during pregnancy	Socio-economic characteristics	Highest level of maternal education and current occupation
		Socio–demographic characteristics	Maternal age in complete years, parity, religion, and ethnicity
		Maternal health characteristics	(Gestation stage (trimester) at body composition assessment, maternal illnesses or complications during pregnancy and antenatal clinic (ANC) attendance).
		Infant characteristics	Infant' sex as recorded from mother – child booklet , after delivery

3.4 Target population

The target population for this study was pregnant women in their first or second trimester and their respective infants who were participants in the MIYCN study, in Korogocho and Viwandani. Out of the 1000 pregnant women who were enrolled into the MIYCN study, 125 of them were selected to participate in this study.

3.4.1 Inclusion criteria

Women who participated in this study were pregnant women in the first and second trimester, living within the NUDHSS enumeration area in Korogocho and Viwandani, who were enrolled in the MIYCN study by APHRC, and who consented to be part of this sub-study.

3.4.2 Exclusion criteria

Women who reported to have severe nausea and vomiting, which could interfere with the stable isotope – deuterium dilution procedures were excluded.

3.5 Sample size

The formula for calculating the sample size for a one sample continuous outcome was used to determine the sample size for this study; (World Health Organisation, 2001).

$$n = \frac{(Z_{\alpha/2} + Z_{\beta})^2 \sigma^2}{E^2}$$

Where, n was the desired sample size, α was the level of significance (95%), $Z_{\alpha/2}$ was 1.96 which is the standard normal deviation which corresponds to 95% confidence level , Z_{β} = value from the standard normal distribution at a selected power (Z_{β} at 80% power is 0.84), σ was the standard deviation of the outcome variable, the average birth weight of 2.65kg for Nairobi County as reported by Gathimba et al., (2014) with a standard deviation of 0.613 was used in this study, and E was the margin of error, taken as 6% in this study. A sample size of 117 was computed as adequate for this study considering a power of 80%;

$$n = \frac{(1.96+0.84)^2 \times 0.613^2}{(0.06 * 2.65)^{2s}} = 117$$

A 10 % loss to follow up cases was expected; and this was factored using the formula;

N (number to enrol) = Desired sample size / percent retained (Sakpal, 2010)

Therefore;

N =117+12 = 129.

129 women were therefore recruited for the study.

3.6 Sampling technique

Systematic sampling was used to select the study participants. The total sample of mothers recruited in the MIYCN study was 1000, while the calculated sample size for this study was 129 mothers, therefore, using the sampling frame of 1000 mothers, a sampling interval of eight (1000/ 129 = 7.8) was used to identify 129 eligible mothers for this study.

3.7 Research instruments and equipment

3.7.1Researcher- administered questionnaire

A researcher-administered questionnaire (Appendix C), was used to collect data on maternal socio-economic (maternal education, occupation) and demographic (age, parity, religion, ethnicity) characteristics, health status and health seeking behaviour (gestation stage at body composition assessment, maternal illnesses or complications during pregnancy and ANC attendance).

Birth weight data was collected from the mother and child health booklet (recorded at birth) within the first month after delivery.

Gestational age was estimated on the basis of the date of the last menstrual period as recorded in the MCH booklet. For those who did not have the MCH booklet, the self-reported date of their last menstrual period, was used.

3.7.2 Research equipment

a) Analytical balance and Electronic weighing scale

An analytical balance (SECA model) with an accuracy of 0.001g was used to weigh the deuterium that was administered to the study participants while an electronic weighing scale (SECA model) with an accuracy of 0.1kg was used to measure maternal weight.

b) Fourier transform infra-red spectroscope (FTIR) Machine

A FTIR machine (FTRI 650 model) was used to measure deuterium enrichment in the saliva samples.

3.8 Pretesting

The pre-testing of the questionnaires was conducted on a 10 % sample (n=13) of the calculated sample size. The pretesting was conducted in Mukuru Kayaba, an informal settlement neighbouring and with similar characteristics as Viwandani informal settlement. Participants in the pre-test were not included in the main study and their data not included in the analyses. Feedback from the pre-test indicated the sensitivity of the questions on participant’s income or earnings, with majority of those participating in the pre-test indicating that they were uncomfortable talking about their financial income. They were however comfortable answering the questions on their occupation. In response to this feedback, the question on participants income and earnings was expunged from the questionnaire, and occupation used as a proxy for estimating their socio economic status

3.9 Validity and reliability

3.9.1 Validity

The questionnaires were reviewed and validated for use in quantitative data collection by the research team in consultation with experts in the field of maternal and infant and young child nutrition, the dilution technique specialists, and university supervisors. In the interviewer administered questionnaire, questions were adopted from validated questionnaires, used in related studies including the Kenya National Bureau of statistics and the International Atomic Energy Agency (IAEA) (International Atomic Energy Agency, 2010). Sample collection for the body composition assessment was done by the researcher and an experienced specialist in the stable isotope technique from the Kenya Medical Research Institute (KEMRI), the specialist had been trained on the stable isotope technique by the International Atomic Energy Agency (IAEA). The Fourier transform infra-red spectroscope **(**FTIR) machine was calibrated using a standard solution (of 1000mg/kg) prepared by weighing deuterium oxide and diluting with normal water as described by IAEA (International Atomic Energy Agency, 2010).

A 100% editing of the questionnaires was done by the researcher to ensure data quality and validity.

3.9.2 Reliability

Pretesting of the questionnaires was conducted to ensure their reliability in addressing the research questions. To ensure the highest quality of research data process and enhance inter-observer reliability, selection and training of two research assistants was done using standardised procedures. The research assistants were extensively by the researcher trained on quantitative data collection by the researcher. The analytical weighing machine was calibrated fortnightly while in use at the Kenya Bureau of Standards (KEBS), weights and calibration department.

3.10 Recruitment and training of research assistants

Two research assistants (RAS) assisted with the quantitative interviews. The research assistants had a bachelor's degree in Nutrition, with prior experience in conducting quantitative surveys. The RAS were trained by the researcher for five days on the purpose and objectives of the study, interview techniques, research ethics, as well as the deuterium dilution technique protocol. They were also involved in the pretesting and refining of the data collection tools and guides.

3.11 Data collection procedures and techniques

Eligible mothers were contacted by the researcher either by phone calls through the contacts provided during recruitment into the main MIYCN study or by household visits through the household identification numbers obtained from the NUHDSS, for those who had not provided any contacts.

They were informed about the study and invited to a central place within the community (Korogocho and Lungalunga health facilities in Korogocho and Viwandani respectively) for the body composition assessment exercise.

Upon informed consent, face to face interviews were first conducted with individual mothers to elicit information on socio- economic, demographic, health and health seeking behaviour and pregnancy characteristics. The body weight measurements, deuterium dosage and saliva sample collection were then done to establish their body composition. Since the entire process of data collection lasted approximately 6 hours for each individual mother, the participants were requested to come to the venue in the morning, to allow for adequate time for all the data and saliva sample collection activities.

3.11.1 The deuterium dosage and saliva sample collection procedure

The deuterium dosage and saliva sample collection was a stepwise procedure involving;

a) Consent process

The participants were first taken through the consent process, before embarking on the subsequent procedures. In this process, the details of the procedure were provided to the participants, including the objectives of the study, a description of the deuterium oxide that they were required to ingest as well as the anticipated benefits and risks in participating in the study, further the participants were informed about the measures that would be taken by the researcher to ensure confidentiality of their information and measurements. Upon provision of all the study details (Appendix A, part 1), the participants were allowed to ask any question and requested for their consent to participate in the study, by signing a consent document (Appendix A, part 2) for those who agreed to participate in the study.

Each mother who agreed to participate in the study was then assigned a study code (*001,002,003…)* that was used to identify their data and measurement in the course of the study for anonymity and confidentiality.

b) Weight measurements

The weight measurements were taken by the researcher, just before the deuterium dosage. For accuracy, the participants were requested to be in the lightest clothing possible while taking their measurements. Weight was taken using the Seca scale to the nearest 0.1kg, with the participants standing upright in light clothing and without shoes. For accuracy and reliability, three weight measurements were taken and the average recorded. In cases where the three measurements differed by more than 0.1kg, they were disregarded and the procedure repeated for that participant.

c) Interviewer administered interview

An interview was then conducted with the participant, collecting their characteristics including social economic, demographic, health and health seeking behaviour and health characteristics.

Their gestation period (number of months pregnant) was confirmed through the date of their last menstrual period (LMP), during the interview in order to ensure their eligibility to participate in the study (first or second trimester). For those with irregular LMP and those who could not remember their LMP, their estimated gestation stage was used. The responses were recorded by the researcher on an electronic data capture equipment (tablet) as they were obtained from the participant.

d) Collection of pre dosage saliva samples (T0)

Pre-dosage saliva sample was collected just before the ingestion of the deuterium isotope, as recommended by the International Atomic Energy Agency (IAEA) protocol (Appendix B) (International Atomic Energy Agency, 2010). For ease of the saliva collection, the participants were provided with a piece of cotton wool and requested to roll it inside their mouths for about 2 minutes, until it was soaked with saliva. The saliva in the cotton wool was then squeezed out into a vial using a 20ml syringe. At least 2ml of saliva was considered adequate for the exercise. The vials containing the saliva were then capped tightly, labelled clearly using white labels indicating the participants code, the type of saliva sample collected (pre dosage sample) and the date and time of the sample collection. These details were also recorded in the study's record book, which was used to document all the samples taken. The vial was then put in a Ziploc bag for transportation to the laboratory for analysis. Collection of the saliva samples was done by the researcher, assisted by the research assistants.

e) Administration of Deuterium (immediately after T0 sample collection)

A standard dose of 30g is recommended by IAEA for adult subject's body composition analysis (International Atomic Energy Agency, 2010). The 30g deuterium oxide in this study was preweighed at KEMRI nutrition laboratory using the SECA analytical balance, and transported in iceboxes to the field in 300ml transparent and tightly capped bottles, which were clearly labelled with the volume of the deuterium contained and the date of preparation. Immediately after the pre-dose saliva collection, the participants were requested to ingest the deuterium, using a straw to avoid spillage. To further ensure that all the deuterium was consumed, the bottle was rinsed with water (half full) twice and the participant requested to drink. The time and volume of the deuterium ingested by each participant was then recorded in the information sheet. Administration of deuterium was done by the researcher and the deuterium dilution technique laboratory expert.

f) Collection of post dosage saliva samples (T1)

It is assumed that the deuterium ingested is evenly distributed into the body compartments (equilibration) and in three to four hours and can easily be detected in body fluids e.g. saliva, urine, plasma, sweat, human milk (International Atomic Energy Agency, 2010). The first post dosage saliva sample was therefore collected by the researcher three hours after ingesting deuterium, using the same procedure described for pre dosage saliva sample.

g) Collection of 4 hour post dosage saliva samples (t2)

The second post dosage saliva sample was collected by the researcher four hours after deuterium ingestion, using the same procedure described for pre- dosage saliva sample.

h) Transportation of the saliva samples from the field to the laboratory and storage.

Each batch of saliva samples (T0, T1 and T2) was packed in a separate zip lock bag and transported to the lab for analysis using separate ice boxes to avoid contamination and evaporation which could occur at room temperature.

In the laboratory, the samples were stored in a deep freezer at –20ºC, to minimise bacterial growth and evaporation while awaiting analysis (International Atomic Energy Agency, 2010).

3.11.2 Face to face interviews (after delivery)

After delivery, each participant was followed up at their homes, to establish their infant's birth weight and gender. For the follow-up exercise, the participants were contacted occasionally during their expected month of delivery to enquire if they had delivered. The participants were visited upon confirmation that they had delivered, and their infants details; birth weight and child sex recorded from the mother and child health booklet where it had been recorded at birth.

3.11.3 Saliva sample analysis

The analysis of the saliva samples to establish the concentration of deuterium oxide (deuterium enrichment) in the pre dosage saliva samples and post dosage saliva samples was done using the FTIR machine at the KEMRI nutrition labs by a deuterium dilution technique laboratory expert. After the deuterium enrichment in the different saliva samples were established for each participant, their TBW, FFM and FM were calculated, taking into account the following assumptions (International Atomic Energy Agency, 2010);

i. Total body weight is composed of fat mass and fat free mass.
ii. Total body water is about 73 % of the body's fat free mass in adults
iii. The deuterium oxide is equally distributed in all body water compartments (e.g. saliva, urine, plasma, sweat, human milk) and the total volume of distribution of deuterium in the body (VD) is 1.041 of total body water due to the exchange of hydrogen atom with the non- aqueous hydrogen in the body.
iv. The rate of equilibration of deuterium oxide is rapid 3 to 4 hours for healthy adults
v. Neither deuterium oxide nor body water is lost during the equilibration time, participants were requested to empty their bladder before the exercise and to minimize water intake and physical activity in order to minimize fluid loss during the activity.

a) Analysis and calculation of total body water, fat mass and fat free mass

Analysis of deuterium enrichment in the saliva samples was done using the Fourier Transform Infrared (FTIR) Spectrometry (International Atomic Energy Agency, 2010). Deuterium enrichment is described as the concentration of deuterium in the body water above the normal body concentration (0.015 atom %) as result of the dosage consumed, it is provided as an output in the FTIR machine in units of mg/kg. After deuterium enrichment is determined (an output in the FTIR machine), total volume of deuterium dilution (VD) will be calculated as;

VD *=Consumed dose of* D_2O *(mg)/deuterium enrichment in saliva (mg/kg)*

With the assumption that VD is 1.041of TBW therefore; total body water was calculated as;

TBW (kg) *= VD /1.041.*

Further, with the assumption that body water (both extracellular and intracellular) is found exclusively within the FFM, and is approximately 73 % of FFM, FFM was calculated as;

FFM (kg) = TBW (kg) / 0.732.

With the assumption that total body weight comprises of fat free mass (all non-fat components in the body including proteins and bones) and fat mass (total body fat), fat mass was calculates as; FM ***(kg)*** *= body weight (kg) – FFM (kg).*

3.12 Data analysis

Electronic data capture system using netbooks, was used for the data collection. After each interviews, the data was upload into the project’s central server, from where it was extracted into STATA 13 software for cleaning and statistical analysis. Descriptive summary statistics such as frequencies and percentages were used for the analysis of socio-economic and demographic characteristics, and body composition measurements. Univariate and multiple linear regression were used to determine the relationship between maternal body composition (TBW, FFM and FM) and the various maternal social demographic, economic and health factors, as well as maternal body composition (TBW, FFM and FM) and birth weight while controlling for potential confounders. P values less than 0.05 were considered statistically significant. A birth weight of less than 2500g was categorised as low birth weight, 2500g to 4000g as normal birth weight, and birth weight higher than 4000g as high birth weight (large babies) according to World Health Organization birth weight classification (World Health Organization, 2010). For the BW, FFM and FM, the analysis was conducted as continuous variables without categorisation (Table 3.2).

Table 3.2: Analysis Matrix

Objective	Variables	Analysis
To determine maternal socio demographic, socio economic and health characteristics	**SES** - Occupation, education level	Frequencies, Percentages
	Socio demographic characteristics - Age - Parity - Occupation - Religion - Ethnicity - Education - Site	Frequencies, Percentages Mean & SD
	Health and health seeking behavior- - Health conditions during pregnancy - Maternal illness during pregnancy - ANC attendance	Frequencies, Percentages
To determine maternal body composition during pregnancy	**Maternal BC** - Total Body Water - Fat Free Mass - Fat Mass	Percentages, Mean & SD
To determine Infant's birth weight.	**Birth weight** - Birth weight (as continuous variable) - Birth weight (categorical) . LBW (< 2500g) . NBW (2500 – 4000g) . HBW (>4000g)	Mean & SD Percentages
To determine the relationship between Maternal body composition and birth weight.	- Total Body Water and Birth weight - Fat Free Mass and Birth weight - Fat Mass and Birth weight	Linear regression analysis

3.12 Logistical and ethical considerations

Approval to conduct this study was sought from the Kenyatta University graduate school, while ethical clearance was sought from Kenyatta University ethical review committee (KU-ERC) (PKU/424/1393), and a research permit obtained from National Commission of Science, Technology and Innovation (NACOSTI) (NACOSTI/P/16/60789/11178), Appendices H, I and J respectively. Further, the MIYCN project within which this study was nested was approved by the KEMRI Ethical Review Board (ERB) (Non SSC protocol No. 327). Voluntary informed consent (Appendix A) was sought from each study participant prior to the interviews and sample collection. For confidentiality, passwords known only to the research team were used to keep track of the data collected. The saliva samples were also assigned unique codes identifiable only by the research team. Snacks (500ml milk) was provided to the participants during the study procedures. Ill or malnourished participants were referred to health facilities for management.

3.13 Risks and benefits to the participants

The study posed no risk to the study participants. The standard dose of 30g deuterium has not been associated with any harmful effects on human adults, in fact, deuterium is naturally found in body water in a concentration of 0.015% (International Atomic Energy Agency, 2010). Deuterium oxide is handled in the body in the same way as water, and is dispersed through the body water within hours. (International Atomic Energy Agency, 2010; Jones & Leatherdale, 1991; Klein & Klein, 1986). The deuterium dilution technique has been used to assess body composition in pregnant women and breast milk intake in children in other studies in Kenya and other parts of the world with no reports of adverse effects (Ettyang et al., 2005; Kupka et al., 2011; Lof & Forsum, 2004). There were no direct benefits to the participants, however, they were reimbursed the transport costs associated with transport to the data collection point. They were also provided with refreshments during the sample collection exercise. No cases of adverse effects were reported in this study.

CHAPTER FOUR: RESULTS

4.1 Maternal characteristics

A total of 125 participants from a calculated sample size of 129 participated in the body composition assessment. Of the 129 recruited, 4 did not complete the steps necessary for body composition analysis per required criteria of 3 saliva samples (International Atomic Energy Agency, 2010), therefore, body composition assessment was successfully completed with 125 mothers, 73 in Korogocho and 52 in Viwandani. All (125) the mothers who participated in the body composition assessment were followed up after delivery in order to establish their infant's birth weight and sex. However, 27 of them were lost to follow-up, for the reasons indicated in Table 4.1.To avoid overburdening the mothers with sensitive questions on the loss of their children, no follow-up interviews were conducted with the bereaved mothers, to establish the causes of miscarriage, still birth or death of their children.

Table 4.1 Reasons for loss to follow-up (missing birth weight)

Reason for lost to follow- up	**n**
Movement out of the study area	18
Miscarriage	2
Still birth	3
Baby not weighed at delivery	2
Refusal	2
Total	**27**

The response rate for the body composition assessment was therefore 96.8% while for birth weight was 75.9 %. A response rate of over 60% is considered adequate for analysis (Mugenda, 2003).

4.1.1 Maternal socio-demographic characteristics

The mean age of the participants was 23.8 (± 5.2) years, with the majority (62%) being in their twenties and 21% being less than 20 years. Majority (76%) of the participants were married or and with at least one other child (65%). Table 4.2 highlights the participant's demographic characteristics.

Table 4.2: Participants socio-demographic characteristics

Characteristics		**N =125| n (%)**	**Mean ± SD**
Informal settlement	Korogocho	73 (58.4)	
	Viwandani	52 (41.6)	
	Total	125 (100)	
Age in complete years	<20	26 (25.9)	
	20-24	42 (33.6)	23.8 ± 5.2
	25-29	36 (30.8)	
	≥ 30	13 (11.1)	
	Don't know/ Can't remember	8 (6.4)	
	Total	125 (100)	
Parity	0	47(37.6)	
	1-2	60 (48)	
	3-4	13(10.4)	
	5-7	5 (4)	
	Total	125 (100)	
Marital status	Single	26 (20.8)	
	Married/ Living with partner	95 (76)	
	Others	4 (3.2)	
	Total	125	
Ethnic group	Somali/ Borana/Garre	7 (5.6)	
	Kikuyu& Embu	38 (30.4)	
	Kamba	18 (14.4)	
	Kisii	4 (3.2)	
	Luhya	26 (20.8)	
	Luo	23 (18.4)	
	Taita	2 (1.6)	
	Others	8 (6.4)	
	Total	125	
Religion	Christian	111 (88.8)	
	Muslim	9 (7.2)	
	Traditional	1 (0.8)	
	Others	4 (3.2)	
	Total	125	

4.1.2 Maternal socio-economic characteristics

Most of the participants had primary level of education (64%). Only 14% had attained secondary and higher level of education. A large proportion (70%) of the participants were unemployed, while 22% of those who were employed worked in the informal sector, mainly as casual labourers, domestic workers or petty traders (green groceries, food vendors, hawkers). The maternal socio-economic characteristics are presented in Table 4.3 below.

Table 4.3: Maternal socio-economic characteristics

Characteristic	**N = 125** **N (%)**
Highest level of education	
None/Less than Primary	21 (16.8)
Primary School	80 (64.0)
Secondary School	16 (12.8)
College/University	2 (1.6)
Missing	6 (4.8)
Total	125 (100)
Occupation	
Unemployed	88 (70.4)
Employed (Informal)	27 (21.6)
Employed (Formal)	6 (4.8)
Missing	4 (3.2)
Total	125 (100)

4.1.3 Maternal pregnancy characteristics

The findings from the participants shows that majority of the participants (84%) recruited to the study were in the second trimester, 16% of the participants had not attended any ante natal clinic (ANC). The number of ANC visits was based on participants' self-report. Incidences of participant's illnesses or complications were also established in this study, through self-reports. The participants were asked on any illnesses that they had experienced since conception to the time the study was conducted. About 70% of the women reported to have experienced at least one pregnancy related complication or illness, with 14% of them reporting to have experienced three or more complications during the pregnancy (table 4.4)

The main complications and / or illnesses mentioned included severe nausea and vomiting (39%), oedema (swollen legs) (17.7%), Malaria (9.2%), fever (22.7%), anaemia (low haemoglobin) (2.5%), bleeding (2.1%), depression (2.1%), fainting (2.10%), varicose veins (0.7%).

Table 4.4: Participant's pregnancy characteristics

Characteristic	n (%)
Pregnancy stage at body composition assessment	
First trimester	20 (16.0)
Second trimester	105 (84.0)
Total	125 (100)
No. of ANC Visits (at the time of BC)	
0	20 (16.0)
1 to 3	50 (40.0)
more than 3	55 (44.0)
Total	125 (100)
No. of complications/ illnesses experienced during pregnancy	
0	38 (30.4)
1	39 (31.2)
2	31 (24.8)
3 to 5	17 (13.6)
Total	125 (100)

4.2 Maternal body composition characteristics

Maternal weight was established through direct weight measurements of the participants', while total body water, fat mass and fat free mass were established through the deuterium dilution technique as described in Chapter three.

4.2.1 Maternal body weight

Maternal weight ranged between 43 and 99 kilograms (kg) with a median of 59.86 kg and a mean 62.7 ($\pm$10.93) kg, in all the pregnancy stages (Table 4.5).

Table 4.5: Maternal body weight during pregnancy

Weight (kg)	N = 125 n (%)	Mean $\pm$ SD
<50	11 (8.8)	
50.0-54.9	21 (16.8)	
55.0-59.9	31 (24.8)	62.7 $\pm$10.93
60.0-64.9	20 (16.0)	
65.0-69.9	13 (10.4)	
>=70	29 (23.1)	
Total 125		

4.2.2 Maternal total body water (TBW)

TBW is the total water content of the body, including the intracellular and extracellular water (International Atomic Energy Agency, 2010). In this study, the participants TBW ranged between 23.3L and 44.8L, the mean TBW was 33.3L (±4.7), which was 53.1% of the mean body weight. When univariate and multiple linear regression analysis was applied, adjusting for socio-demographic (age, marital status, ethnicity, religion, parity, study area), socio–economic (education, occupation) and health related (ANC attendance, pregnancy complications and weight) characteristics, TBW had a significant association with maternal age and the gestation stage (trimester) when the body composition assessment was done. TBW was significantly lower among younger mothers who were aged less than 20 years (31.11L) compared to older mothers who were 20 – 24 years (33.23L, p=0.044), 25-30 years (34.28L, p=0.009) and 30 years and above (34.31L,p=0.010). In addition, participants in the second trimester had a significantly higher TBW (33.77L, p=0.010) compared to those in the first trimester (31.06L).Table 4.6 below outlines the relationship between maternal total body water and maternal characteristics.

Table 4.6: Relationship between maternal characteristics and maternal total body water

	Univariate regression analysis				Multiple linear regression analysis			
Total Body Water	**Coef.**	**P**	**confidence interval**		**Coef.**	**p**	**Confidence interval**	
Site (Korogocho)								
Viwandani	1.30	0.126	-2.99	0.37	-0.91	0.376	-2.97	1.136
Maternal Age (<20 yrs.)								
20-24	2.14	0.063	-0.11	4.41	2.74	0.044*	0.07	5.40
25-29	3.17	0.008*	0.84	5.50	4.13	0.009*	1.04	7.23
≥ 30	3.20	0.041*	-0.39	6.62	5.10	0.010*	1.23	8.97
Marital status (Single)								
Married/living together	1.54	0.131	-0.46	3.55	-0.05	0.963	-2.51	2.39
Ethnicity (Somali)								
Kikuyu/Embu	-2.43	0.189	-6.06	1.20	-8.32	0.052	-16.70	0.06
Kamba	-3.09	0.123	-7.02	0.85	-8.95	0.141	-17.51	-0.39
Kisii	-1.22	0.661	-6.76	4.31	-5.27	0.272	-14.75	4.20
Luhya	0.18	0.926	-3.58	3.94	-4.83	0.248	-13.09	3.43
Luo	0.91	0.636	-2.90	4.72	-4.24	0.304	-12.39	3.91
Taita	-3.87	0.282	-10.95	3.21	-8.13	0.132	-18.78	2.51
Religion (Christian)								
Muslim	0.13	0.937	-3.02	3.27	-4.15	0.233	-11.03	2.72
Traditional	-6.32	0.173	-15.46	2.81	-5.39	0.275	-15.17	4.38
Parity (0)								
1 -2	0.86	0.347	-0.94	2.66	-0.3	0.762	-2.75	2.02
3-4	1.82	0.212	-1.05	4.69	-1.35	0.462	-4.99	2.28
5-7	3.46	0.114	-0.84	7.76	0.12	0.965	-5.45	5.70
Occupation (None)								
Informal	0.99	0.330	-1.01	2.99	-1.72	0.179	-4.25	0.80
Formal	-0.91	0.642	-4.75	2.94	-0.39	0.861	-4.88	4.09
Education (None)								
Primary School	-0.92	0.422	-3.16	1.33	0.19	0.874	-2.27	2.67
Secondary School	-2.35	0.129	-5.39	0.69	1.41	0.462	-2.38	5.20
College/University	-1.31	0.704	-8.09	5.48	-2.11	0.584	-9.76	5.53
Trimester (First)								
Second trimester	2.74	0.024*	0.47	4.96	3.569	0.010*	0.87	6.25
Number of ANC visits								
1 - 3	-0.77	0.542	-3.24	1.71	-1.574	0.273	-4.41	1.26
More than 3	-1.78	0.152	-4.21	0.66	-2.287	0.095	-4.98	0.40
Pregnancy complications (0)								
1	0.10	0.924	-2.03	2.23	-0.222	0.850	-2.54	2.10
2	-0.64	0.575	-2.89	1.61	0.192	0.885	-2.44	2.82
3 -5	-0.20	0.886	-2.89	2.50	-1.279	0.398	-4.27	1.71

* Significant at p<0.05

4.2.3 Fat free mass (FFM)

FFM refers to the part that is non-fat, including proteins, bone mass, non-bone mass (International Atomic Energy Agency, 2010). Participants' FFM ranged between 31.90kg and 61.39kg, with a mean of 45.7kg (± 6.5). The FFM percentage of total body weight ranged between 57.7% and 89% with a mean of 73.1%. Both univariate and multiple linear regression; adjusting for socio-demographic (age, marital status, ethnicity, religion, parity, study area), socio–economic (education, occupation) and health related (ANC attendance, pregnancy complications and weight) characteristics were done to establish the relationship between fat free mass and maternal characteristics. Age and gestations stage were found to have a significant relationship with FFM. FFM increased with age, whereby, mothers aged less than 20 years had significantly lower fat free mass (42.66kg) compared those aged 20 to 24 years (45.56kg, p=0.031), 25 to 29 years (46.97kg, p=0.007) and 30 years and above (47.0kg p= 0.008). In addition, FFM was significantly higher among mothers in the second trimester (46.26kg, p=0.007), compared to those in the first trimester (42.59kg). There was no significant association between FFM and marital status, religion, parity, occupation, education, ANC visits and pregnancy illness/complications during pregnancy; at both univariate and multiple regression analysis (Table 4.7).

Table 4.7: Relationship between maternal characteristics and fat free mass

	Univariate regression analysis				Multiple linear regression			
Fat Free mass (kg)	**Coef.**	**P**	**Confidence interval**		**Coef.**	**P**	**Confidence interval**	
Site (Korogocho)								
Viwandani	-1.73	0.141	-4.06	0.60	-1.40	0.331	-4.25	1.44
Maternal age(< 20)								
20-24	2.90	0.071	-0.25	6.05	4.24	0.031*	.39	8.09
25-29	4.31	0.012*	1.06	7.56	6.18	0.007*	1.74	10.62
≥ 30	4.23	0.087	-0.63	9.08	7.70	0.008*	2.11	13.28
Marital status (Single)								
Married/Living Together	2.159	0.133	-0.60	4.92	-0.33	0.852	-3.76	3.10
Ethnic group (Somali)								
Kikuyu/Embu	-3.32	0.189	-8.30	1.66	-11.90	0.074	-23.47	-0.32
Kamba	-4.23	0.123	-9.62	1.17	-12.72	0.075	-24.53	-.091
Kisii	-1.68	0.661	-9.27	5.91	-7.14	0.271	-20.15	5.88
Luhya	0.47	0.858	-4.7	5.65	-7.21	0.212	-18.65	4.23
Luo	1.25	0.636	-3.98	6.48	-6.13	0.285	-17.35	5.09
Taita	-5.30	0.282	-15.01	4.41	-11.46	0.122	-26.10	3.18
Religion (Christian)								
Muslim	0.13	0.951	-4.20	4.47	-6.09	0.213	-15.60	3.40
Traditional	-8.70	0.172	-21.25	3.85	-6.83	0.325	-20.33	6.68
Parity (0)								
1 -2	1.25	0.324	-1.24	3.74	-.65	0.701	-3.94	2.67
3-4	2.490	1.255	0.21	6.44	-2.02	0.432	-7.03	2.99
5-7	4.74	0.123	-1.17	10.64	0.07	0.971	-7.60	7.74
Occupation (None)								
Informal	1.55	0.271	-1.24	4.35	-2.84	0.132	-6.51	0.83
Formal	-1.24	0.642	-6.52	4.039	-.66	0.831	-6.83	5.50
Education (None)								
Primary School	-1.20	0.443	-4.29	1.89	0.22	0.903	-3.18	3.62
Secondary School	-3.22	0.135	-7.40	0.96	2.05	0.441	-3.17	7.26
College/University	-1.79	0.713	-11.11	7.54	-3.33	0.532	-13.89	7.23
Gestation stage (First trimester)								
Second trimester	3.67	0.021*	0.53	6.87	5.36	0.007*	1.49	9.23
Number of ANC visits (None)								
1- 3	-0.95	0.581	-4.37	2.46	-2.36	0.242	-6.30	1.57
More than 3	-2.43	0.152	-5.78	0.92	-3.20	0.091	-6.91	0.50
Pregnancy complication (None)								
1	0.03	0.993	-2.92	2.98	-0.06	0.971	-3.31	3.19
2	-0.99	0.533	-4.11	2.13	0.54	0.771	-3.14	4.22
3 to 5	-0.38	0.842	-4.10	3.34	-1.68	0.422	-5.78	2.45

* Significant at p<0.05

4.2.4 Fat mass and body fat percentage

Fat mass refers to the total amount of fat in the body while total body fat percentage describes the fat percentage of an individual's weight (International Atomic Energy Agency, 2010). The mean fat mass and body fat percentage in this study were 17.01 kg (± 7.4) and 26.8% (± 8.1) respectively.

At both univariate and multiple regression analysis, there was a significant relationship between FM and maternal age. FM was significantly lower for mothers aged less than 20 years (13.81kg) compared to older mothers who were 20 to 24 years (17.47kg, 0.01), 25 to 29 years (16.75kg, p=0.002) and 30 to 35 years (21.30kg, p=0.000). There was no significant relationship between fat mass and study site, marital status, occupation, education, ANC visits pregnancy complication and gestation stage at body composition. (Table 4.8).

Table 4.8: Relationship between maternal characteristics and maternal fat mass

	Univariate analysis				Multiple regression analysis			
Fat Mass (kg)	**Coef.**	**P**	**Confidence interval**		**Coef.**	**p**	**Confidence**	**Interval**
Site (Korogocho)								
Viwandani	-0.37	0.784	-3.02	2.29	-2.66	0.101	-5.83	0.51
Maternal age (<20 years)								
20-24	3.67	0.03*	0.34	6.99	7.28	0.001*	2.97	
25-29	2.95	0.091	-0.49	6.37	7.89	0.002*	2.94	12.85
≥ 30	7.49	0.02	2.89	12.10	13.29	0.000*	7.05	19.52
Marital status (Single)								
Married/Living Together	-1.69	0.291	-4.83	1.45	-2.06	0.292	-5.89	1.76
Ethnic group (Somali)								
Kikuyu/Embu	0.08	0.981	-5.78	5.93	-14.98	0.023	-27.9	-2.06
Kamba	-1.59	0.622	-7.93	4.76	-16.97	0.062	-30.15	-3.78
Kisii	1.47	0.752	-7.45	10.39	-8.94	0.231	-23.46	5.59
Luhya	-1.73	0.581	-7.82	4.36	-17.31	0.972	-30.13	-4.59
Luo	-2.08	0.512	-8.22	4.07	-15.96	0.321	-28.49	-3.43
Taita	8.53	0.144	-2.88	19.95	-9.61	0.251	-25.95	6.74
Religion (Christian)								
Muslim	-0.83	0.742	-5.76	4.11	-12.82	0.462	-23.42	-2.22
Traditional	-10.32	0.163	-24.61	3.98	-5.82	0.443	-20.89	9.25
parity (None)								
1 - 2	-0.83	0.562	-3.68	2.06	-3.08	0.101	-6.76	0.61
3 - 4	-1.06	0.644	-5.57	3.46	-4.12	0.152	-9.72	1.48
5 - 7	-.98	0.78	-7.74	5.78	-4.98	0.251	-13.54	3.57
Occupation (None)								
Informal	1.84	0.251	-1.32	4.99	-2.92	0.161	-7.02	1.17
Formal	5.08	0.094	-0.88	11.04	4.47	0.202	-2.41	11.36
Education (None)								
Primary School	0.55	0.759	-2.99	4.09	0.77	0.694	-3.03	4.56
Secondary School	1.92	0.428	-2.86	6.71	2.56	0.384	-3.23	8.39
College/University	3.32	0.539	-7.35	13.98	-4.31	0.472	-16.09	7.48
Gestation stage (first trimester)								
Second trimester	2.12	0.251	-1.54	5.77	1.76	0.423	-2.56	6.09
Number of ANC visits (None)								
1-3	-1.37	0.488	-5.24	2.51	-2.14	0.345	-6.52	2.25
More than 3	-1.50	0.436	-5.30	2.30	-2.81	0.184	-6.94	1.33
Pregnancy complications (0)								
1	0.5	0.764	-2.82	3.83	2.19	0.231	-1.44	5.82
2	1.18	0.508	-2.33	4.69	1.31	0.533	-2.79	5.42
3-5	-1.29	0.543	-5.47	2.90	-2.66	0.253	-7.26	1.93

* Significant at $p<0.05$

4.3 Infants birth weight

Infant's birth weight as recorded from the mother child booklet ranged between 2.2kg and 4.2 kg, with a mean birth weight of 3.3kg ± 0.42. Birth weight was further categorised into normal birth weight (2.5kg to 4.0kg), low birth weight (< 2.5kg) and high birth weight (>4.0kg) (World Health Organization, 2010). Majority of the children (94%) had normal birth weight, 3% of the children had low birth weight and the other 3% had high birth weight.

Linear regression analysis were done to establish the relationship between birth weight and maternal characteristics; there was no significant association between birth weight and child's sex, maternal socio-demographic (age, marital status, ethnicity, religion, parity, study area), socio–economic (education, education) and pregnancy characteristics (ANC attendance, pregnancy complications) (table 4.10).

Table 4.9: Relationship between maternal and child characteristics and birth weight

	Linear regression analysis				
Birth weight	**Coef.**	**T**	**P**	**Confidence**	**Interval**
Site (Korogocho)					
Viwandani	-0.04	-0.45	0.650	-0.21	0.13
Child's sex (Female)					
Male	0.14	0.17	0.872	-1.56	0.18
Maternal Age (< 20years)					
20-24	0.14	0.81	0.054	0.00	0.27
25-29	0.11	0.86	0.132	-0.03	0.24
≥ 30	0.11	0.98	0.221	-0.06	0.27
Marital status (Single)					
Married/Living Together	0.09	0.89	0.385	-0.11	0.28
Ethnic group (Somali)					
Kikuyu/Embu	0.20	1.00	0.326	-0.19	0.58
Kamba	0.20	0.90	0.370	-0.24	0.63
Kisii	0.49	1.64	0.115	-0.10	1.08
Luhya	0.16	0.76	0.457	-0.25	0.56
Luo	0.31	1.51	0.141	-0.10	0.71
Taita	-0.58	-1.30	0.201	-1.47	0.31
Religion (Christian)					
Muslim	-0.14	-0.89	0.371	-0.46	0.18
Parity (0)					
1 -2	0.09	1.00	0.342	-.09	0.27
3 – 4	0.05	0.32	0.756	-.25	0.34
5 and above	0.30	1.39	0.173	-.13	0.73
Occupation (None)					
Informal	0.00	0.02	0.982	-0.20	0.21
Formal	0.03	0.16	0.878	-0.35	0.41
Education (None)					
Primary School	0.04	0.34	0.740	-0.18	0.26
Secondary School	0.03	0.18	0.862	-0.29	0.34
College/University	-0.02	-0.07	0.947	-0.63	0.59
Number of ANC visits (None)					
1- 3	0.17	1.07	0.291	-0.15	0.49
More than 3	0.08	0.50	0.623	-0.23	0.39
Pregnancy complications					
1	-0.10	-0.91	0.371	-0.32	0.12
2	-0.06	-0.49	0.636	-0.28	0.17
3-5	0.09	0.71	0.487	-0.17	0.35

* Significant at $p<0.05$

4.4 Relationship between maternal body composition and birth weight

Linear regression was used to establish the relationship between maternal body composition and infant's birth weight. There was a significant relationship between birth weight and maternal weight (p=0.016), TBW (p=0.005) and FFM (p=0.003). There was no relationship between birth weight and maternal fat mass (p=0.28). Table 4.10 below presents the relationship between birth weight and maternal total body water, fat mass and free mass.

Table 4.10: Relationship between maternal total body water, fat free mass and fat mass; and birth weight

Linear regression				
Birth weight	**Coef**	**P**	**Confidence Interval**	
Maternal Weight	6.48	0.016*	1.25	11.72
TBW (L)	0.02	0.005*	0.01	0.04
FFM (kg)	0.02	0.003*	0.01	0.03
FM (kg)	0.01	0.283	0.00	0.02

*Significant at p<0.05

CHAPTER FIVE: DISCUSSION

5.1 Participant's characteristics

Informal settings are associated with high levels of poverty and poor access to basic amenities including health and education (United Nations Human Settlements Programme, 2003). This may explain the low levels of maternal education, unemployment, high teenage pregnancies and poor health seeking behaviour as evidenced by the percentage of mothers who had not attended ANC by the second trimester.

Concurring with the finding that majority of the mothers in this study were unemployed, Emina et al ., 2011 documented that about half of the female residents in the area were economically inactive, and that most of those in employment were either in casual employment or in unestablished businesses. Low education could explain the fact that majority of them were unemployed (Moindi, 2012). In addition, maternal low education has been associated with adverse pregnancy outcomes including low birth weight (Muula, Siziya, & Rudatsikira, 2011).

It is worth noting that a considerable proportion (20%) of the participants in this study were teenage mothers (< 20 years of age). A recent report on adolescent's reproductive and sexual health highlighted the growing number of teenage motherhood in Kenya, which is even higher among low income settings, where one in every four girls aged 14 to 19 years are already mothers. Beguy et al in 2013 estimated the population of teenage mothers living in the study area as 15% (Beguy, Ndugwa, & Kabiru, 2013). Teenage motherhood, has been highlighted by the World health Organisation as among the main health risks for teenagers and a major cause for education disruption for women and girls, and hence their employment prospects (World Health Organisation, 2004).

Since majority of the pregnant mothers were in the second trimester of their pregnancy, it is expected that they would have at least one anti natal clinic (ANC) visit by this pregnancy period, as recommended (World Health Organisation, 2017), however, 16% of the participants had not attended any ANC. Poor ANC attendance is associated with poor pregnancy outcomes including low birth weight (Chuku, 2008).

5.2 Maternal total body water

The International Atomic Energy Agency (IAEA) estimates that water makes up to approximately 70 to 75% of an individual's body weight at birth, but decreases to about 40 to 60 % in adulthood. Total body water in this study accounted for 53% of the total weight, which was within the IAEA estimated ranges. In addition, the IAEA estimates that optimal TBW among women (non pregnant) ranges between 26 to 33L (International Atomic Energy Agency, 2010). The mean TBW in this study was 33.3, slightly higher than the IAEA estimates among non-pregnant women, this could be due to the pregnancy, since studies have shown an increase in TBW levels in the course of pregnancy, Butte et al., (2003).

The mean total body water of pregnant women in this study was slightly higher (33.3L) than that of non-pregnant women in studies conducted in Pokot, Nandi and Narok counties, (24.0L, 31.8L and 29.3L respectively) (Ettyang et al., 2005; Keino et al., 2014). Gains in total body water during pregnancy as documented by Widen and Gallagher., (2014) could be the reason for the slightly higher TBW levels among pregnant women in this study compared to other Kenyan studies conducted among non-pregnant women. In fact Lof et al., (2004) documents higher total body water levels among pregnant women compared to non-pregnant women in the USA. Similar to this study, Larciprete et al., (2003) found the mean total body water of pregnant women in the first and second trimester to be between 38.68L and 40.61L respectively in Italy (Larciprete et al., 2003).

The finding of higher TBW levels among those assessed in the second trimester compared to those assessed in first trimester in this study shows similar trends with a study by Lof et al., (2004) in Sweden, which found women in the third trimester (32 weeks) to have higher total body water levels compared to those in the second trimester (14 weeks gestation) and at preconception stage. Butte et al., (2003) hypothesized that the increase in total body water in the course of pregnancy is among the reasons for weight gain during pregnancy. Ghezzi et al., (2001), further indicate that total intracellular and extracellular water significantly increase as pregnancy advances and return to the pre-pregnancy values within 60 days after delivery. Among the components that contribute to increased total body water during pregnancy is the increased blood volume and extracellular fluids, which are natural changes that occur during pregnancy to support foetal development (Contreras et al., 2012; Institute of Medicine (US) Committee on Nutritional Status During Pregnancy and Lactation, 1990).

Similar to this study, younger mothers (20 years) in Mexico had slightly lower total body water compared to older mothers in both second and third trimester (Contreras et al., 2012). WHO reports that pregnant teenagers have not achieved full growth and development and continue to grow even during the pregnancy (World Health Organisation, 2004), this could explain the lower TBW levels among the young mothers compared to the older and mature mothers. Low total body water measurement during pregnancy has been identified as a risk factor for intrauterine growth retardation (Gernand et al., 2012) although there are no clear cut-off points for normal or low total body water levels during pregnancy (International Atomic Energy Agency, 2010).

5.3 Maternal fat free mass

The mean fat free mass of pregnant women in this study (45.7kg ± 6.5) was slightly higher than those reported in a study conducted among Pokot and Nandi post-partum women who had 32.8kg and 44.8kg respectively, and non-pregnant women in Narok county whose mean fat free mass was 38.9kg (Ettyang et al., 2005; Keino et al., 2014). The difference could be due to the fact that fat free mass increases during pregnancy and is hence higher among pregnant than non-pregnant mothers as documented in several studies (Contreras et al., 2012; Lof & Forsum, 2004).

The lower fat free mass among younger mothers in this study compares with those of Contreras et al., (2012). In their study, older women had higher FFM compared to younger mothers. Rah et al., (2008) highlights that teenage pregnancy may deplete lean body mass among teenage mothers, this coupled by the fact that teenage mothers have not achieved optimal growth and development by the time they get pregnant (World Health Organisation, 2004) could be the probable cause of the lower fat free mass among younger mothers compared to older mothers as recorded in this study. In addition, the growth spurts among adolescents increases their nutrient requirements which are increased further by pregnancy .(Islam et al., 2016), This may also have contributed to the lower body fat free mass levels among the younger mothers compared to the older mothers, who are otherwise fully grown.

The higher level of fat free mass in the second trimester compared to first trimester of pregnancy in this study has also been documented in Sweden and Mexico, where mothers in the first trimester had lower fat free mass compared to those in the second and third trimesters (Contreras et al., 2012; Lof & Forsum, 2004). The progressing growth of the uterine tissue, mammary and soft tissue during pregnancy is thought to contribute to the increase in fat free mass as pregnancy progresses (Institute of Medicine (US) Committee on Nutritional Status During Pregnancy and Lactation, 1990).

5.4 Maternal fat mass

The Kenya healthy diets and physical activity guidelines highlight that optimal body fat percentage for non pregnant women ranges between 12% – 25% (Ministry of Health., 2017). The mean body fat percentage for women in this study was 26.8%, slightly higher than the optimal ranges as stated in the guidelines, which could be as a result of the pregnancy. Maternal fat accretion during pregnancy occurs as a natural process to cater for foetal growth and development (Kopp-Hoolihan et al., 1999). It is further hypothesized that the fat mass accumulation is partly responsible for the weight gain during pregnancy (Larciprete et al., 2003) and that fat deposition occurs more around the thighs, subscapular region and in the abdomens subcutaneous and preperitonial layers (Kinoshita & Itoh, 2006) (Crawford et al., 1981; G. Hornstra, 2000; Kinoshita & Itoh, 2006).

Pregnant women in this study had a slightly higher fat mass (17.01kg) compared to non-pregnant women in Pokot and Nandi counties in Kenya, who had a mean fat mass as of 8.7 kg and 16.7kg respectively (Ettyang et al., 2005; Keino et al., 2014). Studies have documented possible increase in fat mass accumulation during pregnancy compared to non – pregnant states (Langhoff-Roos et al., 1987). This could explain the higher fat mass levels among women in this study compared to non-pregnant women in the other studies.

The increase in fat mass with increasing age agrees with the study by Contreras et al., (2012) in Mexico. Older mothers in this study had a higher fat mass compared to younger mothers. The lower fat mass values among younger women compared to older women in this study could be due to the fact that teenage pregnancy hinders their growth and depletes maternal body fat and lean body mass as evidenced by the study conducted in Bangladesh (Rah et al., 2008). It is worth noting that lower fat mass levels are associated with maternal malnutrition in Kenya (Ettyang et al., 2003).

This study found no relationship between fat mass and maternal gestation stage, however, in USA, fat mass was found to have a correlation with gestation stage (Kopp-Hoolihan et al., 1999), Villar also documents some relationship between fat mass and pregnancy stage, indicating that fat mass increases mainly in the second and third trimester compared to the non- pregnant state (Villar et al., 1992).

Although no relationship was found between body composition and maternal socioeconomic status and parity in this study, Ramal et al. (2013) in Malawi found a correlation between fat mass and mother’s social economic status. That is, wealthier women had higher fat mass accumulation compared to women of lower socio-economic status. In a different study, parity was positively correlated with fat mass, with prim parous mothers having higher fat mass compared to multiparous women in USA (Sidebottom et al., 2001). Both studies used anthropometric measurements of fat mass rather than the deuterium hydration methodology used in this study.

5.5 Infant's birth weight

The proportion of children with normal birth weight in this study (94%) is consistent with that of the national level in Kenya (>90%) (Kenya National Bureau of Statistics and ICF Macro, 2015). However, the percentage of children with low birth weight in this study was slightly lower than that recorded by Mutua (6.4%) in the same study area and at National level and in Nairobi county (8% and 9% respectively) (Kenya National Bureau of Statistics and ICF Macro, 2015; Mutua et al., 2015).

Birth weight is a predictor of both child and adult health, since low birth weight increases the risks of neonatal and infant mortality as well as stunting while higher birth weight is related to adolescent and adulthood obesity and non-communicable diseases (Lawn et al., 2005; Weng et al., 2012). Three percent of children in this study had low birth weight, meaning that they are at a higher risk of adverse health and nutrition outcomes including neonatal mortality and stunting (Abuya et al., 2012; Lawn et al., 2005).

None of the maternal and child characteristics that were assessed in this study had any association with birth weight. Consistent with this, the study by Gathimba et al. (2014) in Nairobi County found no correlation between birth weight and maternal education and ANC attendance. In Mombasa county, Kenya, maternal socio economic status including education also had no association with birth weight (Fairley et al., 2013). In contrast however, some studies in various parts of Kenya have found significant relationships between birth weight and various child and maternal characteristics. For instance, maternal socio economic status, religion and marital status had a significant association with birth weight in Nairobi county, Kenya (Gathimba, 2014) In the central region of Kenya, child's sex was found to have a significant association with birth weight, with a higher likelihood of low birth weight among girls (Muchemi et al., 2015). Maternal age in the coastal region was found to have a significant association with birth weight, with higher risks of low birth weight among children born by older mothers (above 25 years) (Fairley et al., 2013). In Lesotho, Mathule et al., (2005) found an association of birth weight with maternal age, parity and birth weight, while in Bangladesh, maternal education and socio economic status had a significant association with birth weight (Karim & Mascie-Taylor, 1997).

5.6 Relationship between birth weight and maternal body composition

5.6.1 Relationship between birth weight and maternal weight

Maternal weight has been documented to be a good predictor of birth weight (Karim & Mascie-Taylor, 1997; Mohanty et al., 2006; Ververs, Antierens, Sackl, Staderini, & Captier, 2013). While the mean weight in this study was 62.7kg, a systematic review in African and Asian contexts indicates that weight less than 45 kg at any gestational age could be a predictor of low birth weight. Further, studies by mohatny et al. (2006) and Bisai et al. (2007) also indicate that a maternal weight of ≤ 45 kg could be a risk factor for low birth weight (Bisai, Mahalanabis, Sen, Bose, & Datta, 2007; Mohanty et al., 2006; Ververs et al., 2013).

5.6.2 Relationship between birth weight and total body water

The finding in this study that TBW had a positive relationship with birth weight agrees with a study by Butte et al., (2003) who found a positive correlation between total body water and birth weight in the USA using the deuterium dilution method. In a different study, Kupka et al., (2011) using the deuterium dilution methodology found a positive relationship between TBW and birth weight among HIV infected women in Tanzania. Although there are no established TBW cut-off points for normal birth weight, it is important to note that lower TBW has been associated with intrauterine growth retardation, one of the risk factors for low birth weight (Gernand et al., 2012).

5.6.3 Relationship between birth weight and fat free mass

The positive relationship between FFM and birth weight in this study compares with studies among Jamaican women in their first trimester and among Caucasian pregnant women in their second and third trimester (Butte et al., 2003; Farah et al., 2011). In these two studies, FFM was found to have a positive relationship with birth weight and to be a strong predictor of birth weight (Butte et al., 2003; Farah et al., 2011). Higher maternal FFM was linked with higher birth weights compared with lower FFM (Farah et al., 2011). There are limited published maternal FFM cut-off points for normal birth weight, but a fat free mass higher than 40.76kg has been associated with about a 3-fold risk for high birth weight babies (>4kg) among Chinese women (Wang et al., 2017), and hence increased risks of obesity in childhood and adult hood, and associated chronic diseases (Weng et al., 2012).

5.6.4 Relationship between birth weight and fat mass

Similar to this study Butte et al., (2003) found no correlation between FM as measured using the deuterium dilution technique and birth weight. Similarly, Kent et al. (2013) found no relationship between fat mass and birth weight among mothers in the first trimester, using the bioelectric impedance method. In contrast, there was a positive correlation between birth weight and fat mass of women in the third trimester in a Swedish study, conducted using anthropometric assessment (Forsum et al., 2006). It is hypothesized that the fat mass accumulation is also partly responsible for the weight gain during pregnancy (Larciprete et al., 2003). Although no relationship was found between fat mass and birth weight maternal stores of certain types of fats such as the poly unsaturated fatty acids (PUFA) have been shown to promote optimal foetal and infant development and breast milk production post-partum (Crawford et al., 1981; Gerard Hornstra, 2000{Innis, 2008 #221)}.

This study indicates that both maternal total body water and fat free mass during pregnancy are positively associated with infant’s birth weight, while maternal fat mass has no association with infant's birth weight. Butte et al., (2003) found a positive association between maternal total body water and birth weight, while Farah et al., (2011) found a strong correlation between maternal fat free mass and birth weight. In addition Kent et al., (2013) found no relationship between fat mass and birth weight. This signifies the important role played by total body water which includes the intracellular and extracellular body fluids and fat free mass which includes proteins and body tissue, in the foetal growth and development, and hence their influence on the infants general birth weight.

The use of the deuterium dilution technique assessment a gold standard method of body composition assessment, to determine body composition is especially a strength of the study as this technique is considered as a more objective method of nutrition assessment compared to other methods such as anthropometry which have been used to establish maternal nutrition status in this setting.

CHAPTER SIX: SUMMARY, CONCLUSIONS AND RECOMENDATIONS

6.1 Summary

The purpose of this study was to assess maternal body composition including total body water, fat free mass and fat mass during pregnancy and establish the relationship between the body composition and infants' birth weight.

One fifth of the participants were younger mothers aged younger than 20 years. Most of the mothers (84%) were in their second trimester and about 20% had not started attending ante natal clinics. A majority of the mothers had less then secondary level education (85 %) and most were unemployed (70%). The participants' mean weight was 62.7kg ± 10.9kg.

TBW ranged between 23.3 L and 44.8 L and the mean total body water was 33.3 L (+ 4.7L). Total body water was significantly related to participant's age and gestation stage; young mothers (<20 years) had significantly lower TBW values compared to older mothers while mothers in the second trimester had significantly higher TBW values compared to those in the first trimester.

FFM ranged between 31.9 kg and 61.3 kg, the mean FFM was 45.7kg (± 6.5kg). FFM was significantly associated with maternal age and gestation stage, with younger mothers and those in the first trimester having lower FFM values compared to older mothers and those in the second trimester respectively.

The mean fat mass was 17.01 kg (± 7.42). FM was significantly associated with maternal age, with lower fat mass levels among younger mothers (less than 20 years) compared to older mothers who were 20 years and above.

The mean birth weight was 3.3kg ± 0.42kg. A majority (94%) of the children had normal birth weight (2.5kg to 4.0kg), only 3% had low birth weight (<2.5kg). No relationship was found between infant weight and any of the maternal characteristics including socio demographic (age, marital status, ethnicity, religion, parity, study area), socioeconomic (education, education) and health related (ANC attendance, pregnancy complications and weight) characteristics.

Infant's birth weight was significantly related with maternal weight, maternal TBW and FFM. However, there was no significant relationship between birth weight and maternal fat mass.

6.2 Conclusion

Overall, the maternal body composition findings among pregnant women indicate that total body water, fat free mass and fat mass are significantly lower among younger mothers (less than 20 years) and increase as pregnancy progresses.

Maternal total body water was related to maternal age and gestation period. Younger mothers (< 20years) has significantly lower total body water compared to older mothers (> 20 years). In addition, mothers in the first trimester had significantly lower total body water compared to those in the second trimester.

Maternal fat free mass was similarly related to maternal age and gestation period. Younger mothers (< 20years) has significantly lower total body water compared to older mothers (> 20 years) while mothers in the first trimester had significantly lower total body water compared to those in the second trimester.

Maternal fat mass was significantly lower among younger mothers (< 20years) compared to older mothers (> 20 years).

The maternal body components that were specifically found to have a significant relationship with birth weight were total body water and fat free mass, but not fat mass. Therefore,

Ho_1: There is no relationship between maternal total body water and infant's birth weight; was rejected

Ho_2: There is no relationship between fat free mass and infant’s birth weight; was rejected.

Ho_3: There is no relationship between maternal fat mass and infant’s birth weight; was not rejected.

There are no published cut off points for body composition (TBW, FFM and FM) during pregnancy, and hence this study only provides a description of the general body composition, without defining whether the body composition findings are either high, normal or low. Further, Paucity of body composition studies among pregnant women in Kenya and indeed in Africa was noted, this study however fills this gap, by contributing to the body of knowledge on the body composition levels of pregnant women living in low income settings, in Kenya. It fills the gap identified in the literature review, on the paucity of body composition data among pregnant women in Kenya and indeed sub-Saharan Africa, especially using the stable isotope technique.

6.3 Recommendations

6.3.1 Recommendations for policy and practice

a) Recommendations for policy

Given the findings of lower TBW and FFM found among younger mothers (less than 20 years) compared to older mothers, and the positive relationship between these two body components with birth weight, the Ministry of Health should design policies and interventions focusing on improving maternal nutrition during pregnancy that would enhance gains in FFM and TBW, in these and similar settings especially among young mothers. This could be done through integrating nutrition counselling and education within the youth friendly programs that are specifically designed and targeted for youths and adolescents.

Interventions to delay child bearing among teenagers should be promoted, as they were found to have significantly lower total body water and fat free mass which is are risk factors for low birth weight.

b) Recommendations for practice by health workers

Nutritional counselling by health workers, on optimal maternal feeding practices is recommended for pregnant mothers and especially younger mothers in this and other similar settings, in order to improve the nutrition status especially the body components that were found to influence birth weight (TBW, FFM). The National Guidelines for Quality Obstetrics and Perinatal Care provide guidance on optimal energy and protein consumptions for pregnant women, including adolescents (Ministry of Public Health and Sanitation and Ministry of Medical Services., 2012). Breaking down this information into actual locally available and affordable foods that could be used to attain the recommended levels should be a key component in the nutrition education provided to pregnant mothers in this context especially the teenage pregnant mothers who have significantly lower body composition (TBW and FFM) as identified in this study.

6.3.2 Recommendations for further research

The paucity of data on body composition among pregnant women in Kenya was noted, and further research is therefore recommended to better establish the maternal body composition cut-off points among pregnant women. A similar study in rural settings, inclusive of women from a range of socioeconomic status (low to high), and in all three trimesters of pregnancy would best shed some light on the body composition variation in these populations and environments, allowing for targeted strategies to improve mother and child health outcomes among the most vulnerable.

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APPENDICES

Appendix A: CONSENT FORM.

1. **Information sheet.**

Introduction
Hello. My name is **Milka Njeri**; I am from Kenyatta University, I will be conducting research in Korogocho and Viwandani informal settlements to **determine the maternal body composition and birth weight** Your participation in this study will help us understand the matter well which will inform us better on designing effective interventions. We will then share what we have learned from this community with policy-makers and encourage them to make proper decisions that will help the people of these communities.
Explanation of Procedures for Questionnaire and Anthropometric Measurements
Being part of this study will involve participating in an interview that will take about 45 - 60 minutes, we will ask you questions regarding yourself, the pregnancy, antenatal care and your birth history and fill in a questionnaire. We will also visit you again after you deliver to conduct an interview and collect the weight of your child.
Explanation of Procedures for Anthropometric Measurements and Administration of Deuterium Oxide
In addition to the interviews, we will measure your weight, height and your body composition. The procedure used to measure your body composition will involve the administration of Deuterium Oxide (Heavy water) to you. We will obtain an initial saliva sample before administration; we will then collect another sample of saliva three and four hours after administration of the Deuterium

Oxide. The saliva samples will be collected using a sterile cotton wool which you will be given to soak it with saliva for around 5 minutes. This exercise will take only a few minutes (less than half an hour each time it is done). These saliva samples will be taken to KEMRI for analysis. There are no side effects of Deuterium Oxide since it will be administered in small doses and it naturally exists in the human body. In addition, the procedure is not invasive and does not involve any break in the skin or internal body cavity.
Confidentiality and Voluntary Participation
To enter in this study you have been randomly picked as a member of this community. The information that we collect in this research will be treated with. Your name will not appear on any reports or publications from this study. Your participation is **voluntary**, and if you choose not to participate, you will not be treated with prejudice. Please note that if at any point in time you feel you do not want to continue participating, you can withdraw freely. If at any point you feel uncomfortable about any of the questions, you do not have to answer them. You can skip the question or end the interview at any time.
Benefits
You will not get any material gift for participating in this study. However, you will be reimbursed Ksh 300 for transport and time taken to participate in the sample collection exercise, refreshments will also be provided after the sample collection exercise. The data collected from you and your child will be useful in informing policy and interventions that would benefit this community as a whole.
Questions and Your Rights as a Participant
You have the right to ask, any questions you may have about this research. If you have any questions or concerns about this study or the results you can ask me before or after the interview or

you can contact me **+254 020 400 1000**. If you have questions about your rights as a research participant, or concerns or complaints about the research, you may contact the Scientific Steering Committee Member on **+254 020 2722541**

2. Participant's Consent Certificate

Declaration of participant: I declare that the purpose and other details about the study have been given to me. I have also been given an opportunity to ask any questions I may have had and all such questions or inquiries have been answered to my satisfaction. I hereby give my consent to participate in this study. I understand that I will be given a signed copy of this document to keep.

Participant's Name ________________________________

Signature /Thumbprint ________ **Date _______________**

Appendix B: STABLE ISOTOPE – DEUTERIUM DILUTION PROTOCOL

1. Materials and equipment to be used

i) Materials

- Gloves
- Vials
- Syringes
- Cotton wool roll
- Deuterium Oxide (Weighed at KEMRI – 30g for adults)
- Straws
- Weighing machine
- Stadiometer
- Zip lock bags (small and large)
- Labels
- Stationery

ii) Equipment

- Analytical balance (SECA) for weighing deuterium
- Cooler box for transportation of deuterium (from KEMRI lab to the field) and saliva samples (from the field to KEMRI lab)
- Electronic weighing scale (SECA) for maternal weight measurements
- FTIR machine for measuring deuterium enrichment

2. Procedure

i) Consent form – Explain;

a. All the procedures and the duration of the exercise.

b. Deuterium Oxide Deuterium isotope

c. Importance/ reason for the exercise

d. Risks and benefits

e. Confidentiality

f. Allow time for any questions

g. Further contact details in case of complaints or questions.

h. Request the participants to sign the consent form (two copies – one for her and one for documentation)

i. Assign each mother a code e.g. *001,002,003*

ii) Weight measurements

a) Request mothers to empty their bladder/bowel before weight measurements and the procedures

b) Take the weight measurements while the participant is standing on the straight on the weighing scale and record on the information sheet

c) Take two weight measurements to the nearest 0.1kg and calculate the average weight. If the two weight measurements differ by more than 0.1, disregard and repeat the procedure for that participant.

iii) Other data to be collected and recorded on the information sheet

i. Mother's Name

ii. Mother's DOB

iii. Last menstrual period (LMP)

iv) Collection of baseline saliva samples (T0)

a) Cut a reasonable piece of cotton wool, give to the mother and request them to roll inside their mouths for about 2 minutes (until it soaks with saliva)

b) Squeeze the saliva into a vial using the 20 ml syringes and cap the vials tightly (at least 2ml of saliva is required)

c) Cap the vials tightly and record the time of saliva collection in the information sheet.

d) Label the vials clearly, indicating the participants code, time of sample collection, date of collection, and the type of sample (if T0, t1 or t2)

e) Put each vial in a zip lock bag and label the bag clearly (as the label appears on the vial)

f) Put all the small Ziploc bags containing T0 samples in a larger ziplock bag and label indicating the date, type of sample and study site *e.g. 3rd May 2015 T1,Korogocho.*

v) Administration of Deuterium (immediately after T0 sample collection)

a) Give the deuterium to the mothers to drink using the provided straws request them to be careful not to spill (pre weighed in KEMRI)

b) Rinse the bottles with water (half full) and request the mother to drink using the straw

c) Repeat the above (b) procedure 3 times.

d) Record the time of deuterium intake in the information sheet.

e) Record the Deuterium dosage taken by each participant in grams in the information sheet(usually indicated on the deuterium bottle)

vi) Collection of 3 hour post dosage saliva samples (T1)

a) Procedure similar to collection of T0.

b) Record the time of sample collection for each sample in the record book.

c) After collecting the samples, put each vial in a Zip lock bag and label as it appears in the vial.

d) Pack all the small zip lock bags containing the vials in a larger Zip lock bags and label as T1 samples, date and study site

e) The participants should be advised not to eat or drink anything at least 30 minutes before collection of T2 samples.

vii) Collection of 4 hour post dosage saliva samples (t2)

a. Repeat procedure for collecting T0 and T1

3. <u>Assumptions in stable isotope – deuterium dilution technique</u>

vi. Total body weight is composed of fat mass and fat free mass.

vii. Total body water is about 73 % of the body's fat free mass in adults

viii. The deuterium oxide is equally distributed in all body water compartments (e.g. saliva, urine, plasma, sweat, human milk) and the total volume of distribution of deuterium in the body (VD) is 1.041 of total body water due to the exchange of hydrogen atom with the non- aqueous hydrogen in the body.

ix. The rate of equilibration of deuterium oxide is rapid 3 to 4 hours for healthy adults

x. Neither deuterium oxide nor body water is lost during the equilibration time, participants will be requested to empty their balder, minimize water intake and physical activity in order to minimize fluid loss during the activity.

4. <u>Analysis and calculation of total body water, fat mass and fat free mass</u>

Analysis of deuterium enrichment in the saliva samples will be done using the Fourier Transform Infrared (FTIR) Spectrometry(International Atomic Energy Agency, 2010). Deuterium enrichment is described as the concentration of deuterium in the body water above the normal level (0.015 atom %) as result of the dosage consumed, it is provided as an output in the FTIR machine after analysis in units of mg/kg.

After deuterium enrichment is determined (an output in the FTIR machine), total volume of dilution (VD) will be calculated as;

VD *=Consumed dose of* D_2O *(mg)/deuterium enrichment in saliva (mg/kg)*

With the assumption that VD is 1.041of TBW therefore; total body water will be calculated as;

TBW (kg) *= VD /1.041*

With the assumption that body water (both extracellular and intracellular) is found exclusively within the FFM, and is approximately 73 % of FFM, FFM will be calculated as;

FFM (kg) = TBW (kg) / 0.732.

With the assumption that total body weight comprises of fat free mass (all non-fat components in the body including proteins and bones) and fat mass (total body fat), fat mass will be calculates as;

FM (kg) *= body weight (kg) – FFM (kg),*

(International Atomic Energy Agency, 2010)

Appendix C: QUESTIONNAIRE

QUESTIONNAIRE

1	**BACKGROUND**	
1.0	VILLAGE OF RESIDENCE (CODESHEET B5)	
1.1	START TIME (24HRS)	
1.2	FIELD WORKER'S CODE	
1.3	DATE OF INTERVIEW (DD/MM/YYYY)	
1.4	HOUSEHOLD HEAD NAME………………………………………………………………………………………	
1.5	ID OF ROOM WHERE HOUSEHOLD HEAD SLEEPS	
1.6	HOUSEHOLD ID	
1.7	MOTHER'S NAME………………………………………………………………………………………	
1.8	MOTHER'S ID	
1.90	CURRENT MARITAL STATUS OF THE WOMAN **(1=Currently Married ; 2=Living together; 3=Separated; 4=Divorced; 5=Widowed; 6=Never Married)**	
1.10	MOTHERS DATE OF BIRTH (DD/MM/YYYY)	
1.11	MOTHER'S HIGHEST LEVEL OF EDUCATION **(0= Illiterate , 1=Primary, 2= Secondary, 3= Tertiary)**	
1.12	NUMBER OF YEARS AT THE HIGHEST LEVEL	
1.13	RELIGION OF THE MOTHER **(1= Christian , 2= Muslim, 3=Traditional, 4= Hindu, 96=Other)**	
1.14	SPOUSE DATE OF BIRTH (DD/MM/YYYY)	
1.15	SPOUSE HIGHEST LEVEL OF EDUCATION **(0= Illiterate , 1=Primary, 2= Secondary, 3= Tertiary)**	
1.16	SPOUSE NUMBER OF YEARS AT THE HIGHEST LEVEL	

	QUESTIONS AND FILTERS	CODING CATEGORIES	SKIP
2	**OCCUPATION**		
2.0	What is the current occupation of the mother (1=SALARIED; = SELF EMPLOYED; 3= CASUAL; 4 = UNEMPLOYED; 5 = STUDENT)		
2.1	What is the current occupation of the father (1=SALARIED; = SELF EMPLOYED; 3= CASUAL; 4 = UNEMPLOYED; 5 = STUDENT)		

3 PREGNANCY DETAILS

3.1 Are you currently pregnant? (1=YES, 2=NO) (IF NO, SKIP TO 13.0) [ ][ ]

3.2 Date of Last Menstrual Period [ ][ ][ ][ ][ ][ ][ ][ ]

3.3 How many months pregnant are you? [ ][ ]

3.4 Have you experienced any of the following pregnancy related complications with this pregnancy?

	Y	N	DK
HBP	1	2	98
Gestational Diabetes	1	2	98
Anaemia	1	2	98
Depression	1	2	98
Bleeding / Spoting	1	2	98
Severe nuasea and vomit	1	2	98
Malaria	1	2	98
Fainting	1	2	98
Varicose veins	1	2	98
Swollen legs	1	2	98
Other (Specify)__________		96	

4 ANTENATAL CARE

4.0 Have you seen anyone for antenatal care during this pregnancy?

YES..............................	1
NO...............................	2

4.1 Whom did you see?

PROBE (Anyone else?) FOR THE TYPE OF PERSONS AND RECORD ONLY THE PERSON WITH THE HIGHEST QUALIFICATION

Doctor.........................	1
Nurse.............................	2
Midwife/ Auxillary midwife..	3
Traditional birth attendant..	4
Other (Specify)................	6
________________	

4.2 Where did you receive antenatal care for this pregnancy?

(FW: IF HEALTH FACILITY, PROBE AND WRITE ITS NAME, CODE AND LOCATION)

Name of HF ____________________

HF code [ ][ ][ ]

Location ____________________

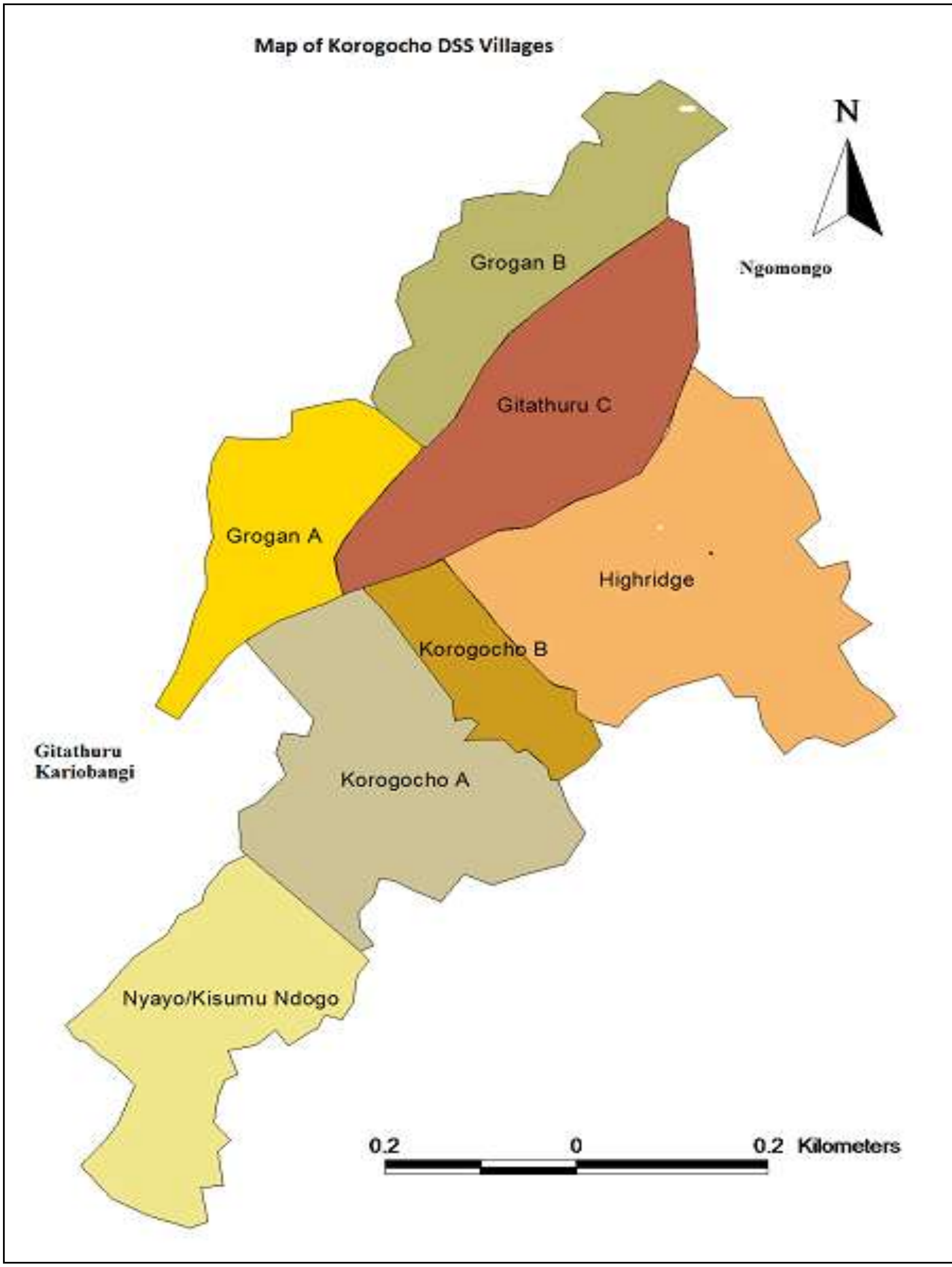
(FW: IF MORE THAN ONE PLACE MENTIONED, RECORD THE MOST RECENT PLACE BEFORE DELIVERY.)

Home............................	1
Traditional birth attendant's	2
Health facility....................	3
Other (Specify)................	6
________________	

4.3	How many months pregnant were you when you first received antenatal care for this pregnancy?	Months [ ][ ] Don't Know.................... 98		
4.4	How many times have you received antenatal care during this pregnancy?	No. of times [ ][ ] Don't Know............. 98		
5	**BIRTH HISTORY DETAILS**			
	Now I would like to ask you questions about all the births you have had in your lifetime.			
5.0	Do you have any children to whom you have given birth who are now living with you?	YES........................... 1 NO............................. 2		→ 4.1
5.0b	How many of your children live with you?	[ ][ ]		
5.1	Do you have any children to whom you have given birth who are alive but do not live with you?	YES........................... 1 NO............................. 2		→ 4.2
5.1b	How many of your children who are alive that DO NOT live with you?	[ ][ ]		
5.2	Have you ever given birth to children who were born alive but later died? **(IF NO, PROBE)**: Any baby who cried or showed signs of life but survived only a few hours or days?	YES........................... 1 NO............................. 2		→ 4.3
5.3	How many children have died? **IF NONE, RECORD 00**	Number [ ][ ]		
5.4	**(FW: SUM ANSWERS 5.0bb, 5.1b, AND 5.3, AND ENTER TOTAL)** IF NONE, RECORD 00 (PARITY)	TOTAL...... [ ][ ]		
5.5	**CHECK 3.7** Just to make sure that I have this right: you have in total _______ births during your life. Is that correct? **IF NO, PROBE AND CORRECT 5.0b - 5.3 AS NECESSARY**	**(FW: TICK THE APPROPRIATE BOX)** YES...... [ ] NO....... [ ]		
5.6	Women sometimes have pregnancies that do not result in a live born child. That is, a pregnancy can end early, in a miscarriage, or the child can be born dead. Have you had any such pregnancy that did not result in a live birth?	YES........................... 1 NO............................. 2		→ 4.9
5.6b	In all how many of the pregnancies did not end in a live born child? IF NONE, RECORD 00	TOTAL [ ][ ]		
5.7	In all, how many of the pregnancies that did not end in a live born child lasted more than 6 months? IF NONE, RECORD 00	TOTAL...... [ ][ ]		
5.8	**SUM ANSWERS 4.4 AND 4.6b AND ENTER TOTAL (GRAVIDA)**	TOTAL...... [ ][ ]		

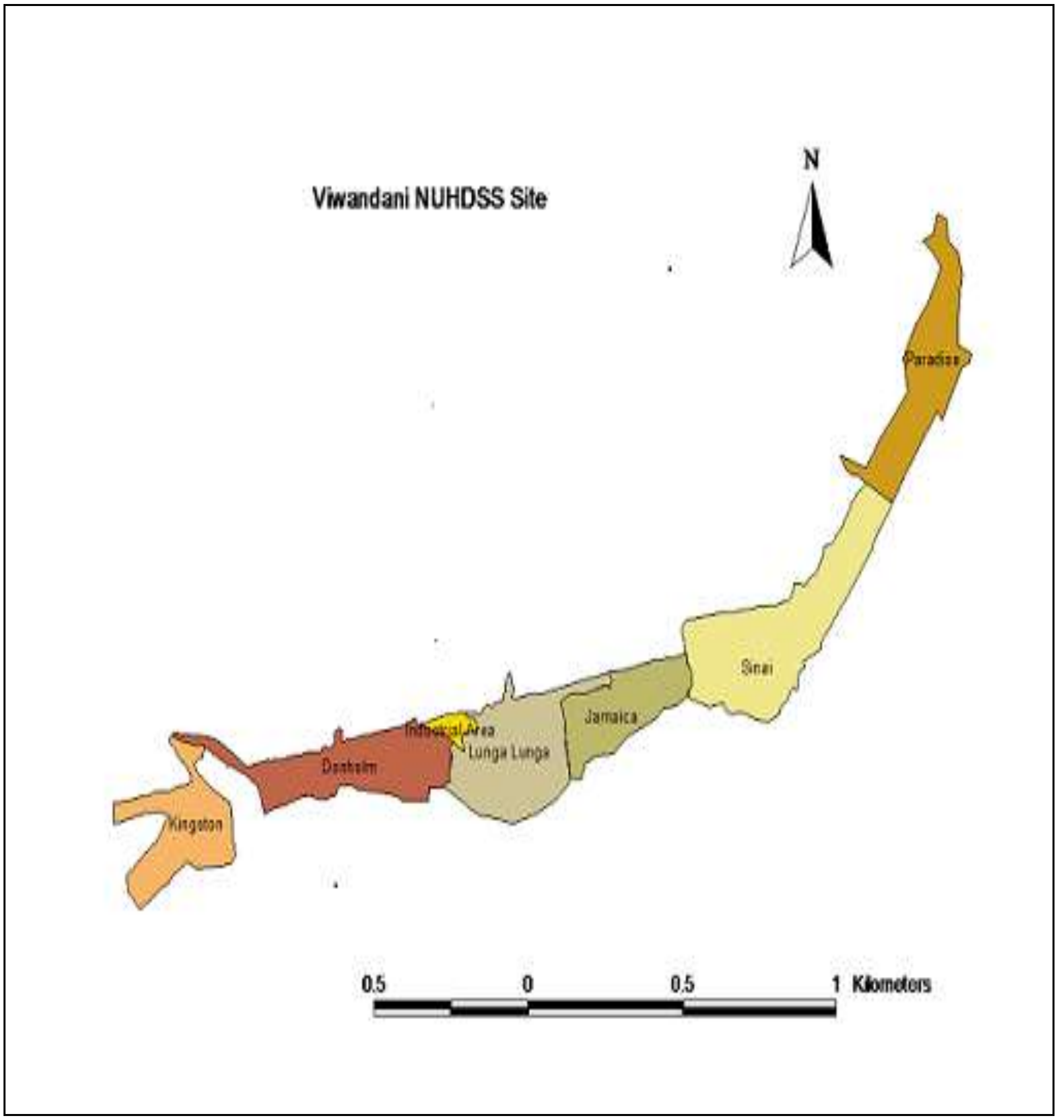
6	**SMOKING HISTORY**		
6.0	Have you ever smoked (and not just testing)	YES .... 1 NO 2	
6.1	Are you still smoking?	YES..................... 1 NO 2	
6.2	How many cigarettes do you smoke per day?	NUMBER OF CIGARETTES	
7	**INFANT DETAILS**		
7.1	CHILD'S NAME..		
7.2	CHILD'S DATE OF BIRTH (DD/MM/YYYY)		
7.2	CHILD'S SEX (1=MALE; 2=FEMALE)		
7.3	When (NAME) was born, was he/she very small, smaller than usual, about usual size, larger than usual, very large or you don't know?	Very small....................... 01 Smaller than usual.......... 02 About usual size............. 03 Larger than usual........... 04 Very large..................... 05 Don't Know/ Remember...... 98	
7.40	Was (NAME) weighed at birth?	YES......................... 1 NO................................ 2 DON'T KNOW................ 8	(2, 8) → 9.0
7.50	How much did (NAME) weigh? RECORD BIRTH WEIGHT IN GRAMS FROM HEALTH CARD IF AVAILABLE IF NO CARD / CAN'T RECALL FILL IN 9999	Weight from card Weight from recall	
8	**ANTHROPOMETRIC MEASUREMENTS FOR MOTHER.**		
8.1	ENTER THE **WEIGHT OF THE MOTHER/CARER** IN KG **1st reading** _ _ _ . _ **2nd Reading** _ _ _ . _ **Avg** _ _ _ . _		
	END TIME (24 HRS)		
9	**RECORD ANY GENERAL COMMENTS**		

Appendix D: MAP OF KOROGOCHO



Ap

Appendix E: MAP OF VIWANDANI



Appendix F: MIYCN STUDY ETHICAL APROVAL

KENYA MEDICAL RESEARCH INSTITUTE

P.O. Box 54840-00200, NAIROBI, Kenya
Tel (254) (020) 2722541, 2713349, 0722-205901, 0733-400003; Fax: (254) (020) 2720030
E-mail: director@kemri.org info@kemri.org Website:www.kemri.org

KEMRI/RES/7/3/1 **11th June, 2015**

TO: **DR. ELIZABETH KIMANI (PRINCIPAL INVESTIGATOR)**
APHRC,
P.O. BOX 10787-00100,
NAIROBI, KENYA

Dear Madam,

RE: **NON-SSC PROTOCOL NO. 327 (*REQUEST FOR ANNUAL RENEWAL AND PROTOCOL DEVIATION)* EFFECTIVENESS OF COMMUNITY BASED NUTRITIONAL COUNSELING ON INFANT FEEDING PRACTICES.**

Thank you for the continuing review report for the period **18th March 2014** to **17th March 2015** and a protocol deviation on late submission of the application.

This is to inform that during the 240th A meeting of the KEMRI Scientific and Ethics Review Committee held on 9th June 2015, the Committee **conducted the annual review and approved** the above referenced application for another year.

The Committee also noted that a protocol deviation form has been submitted, as the request for annual renewal was done after the expiration date of the last approval. The measures taken to prevent this from recurring in the future are satisfactory.

This approval is valid from **June 9, 2015** through to **June 8, 2016**. Please note that authorization to conduct this study will automatically expire on **June 8, 2016**. If you plan to continue with data collection or analysis beyond this date please submit an application for continuing approval to the **SERU** by **27th April 2016.**

You are required to submit any amendments to this protocol and other information pertinent to human participation in this study to the SERU for review prior to initiation.

You may continue with the study.

Yours faithfully,

PROF. ELIZABETH BUKUSI,
ACTING HEAD,
KEMRI/SCIENTIFIC AND ETHICS REVIEW UNIT

In Search of Better Health

Appendix G: KENYATTA UNIVERSITY ETHICAL APPROVAL

KENYATTA UNIVERSITY
ETHICS REVIEW COMMITTEE

Email: chairman.kuerc@ku.ac.ke
secretary.kuerc@ku.ac.ke
kuerc2008@gmail.com
Website: www.ku.ac.ke

P. O. Box 43844 - 00100 Nairobi
Tel: 8710901/12
Fax: 8711242/8711575

Our Ref: KU/R/COMM/51/608

Date: 18th January, 2016

Milka Njeri,
Kenyatta University,
P.O Box 43844,
Nairobi

Dear Njeri,

RE APPLICATION NUMBER PKU/424/I 393 "Maternal body composition and infant birth weight in urban informal settlement in Nairobi County, KENYA".

1. IDENTIFICATION OF PROTOCOL

The application before the committee is with a research topic "Maternal body composition and infant birth weight in urban informal settlement in Nairobi County, Kenya" received on 2nd October, 2015.

2. APPLICANT

Milka Njeri, Department of Food, Nutrition and Dietetics

3. STUDY SITE

Korogocho and Viwandani Informal Settlements, Kenya

4. DECISION

The committee has considered the research protocol in accordance with the Kenyatta University Research Policy (section 7.2.1.3) and the Kenyatta University Ethics Review Committee Guidelines AND APPROVED that the research may proceed for a period of ONE year from 18th January,2016.

5. ADVICE/CONDITIONS

i. Progress reports are submitted to the KU-ERC every six months and a full report is submitted at the end of the study.
ii. Serious and unexpected adverse events related to the conduct of the study are reported to this board immediately they occur.
iii. Notify the Kenyatta University Ethics Committee of any amendments to the protocol.
iv. Submit an electronic copy of the protocol to KUERC.

When replying, kindly quote the application number above.
If you accept the decision reached and advice and conditions given please sign in the space provided below and return to KU-ERC a copy of the letter.

KENYATTA UNIVERSITY OFFICE OF THE CHAIRMAN 18 JAN 2016 ETHICS REVIEW COMMITTEE P.O BOX 43844 - 00100 NAIROBI

DR. TITUS KAHIGA
CHAIRMAN ETHICS REVIEW COMMITTEE

I Milkah Njeri N accept the advice given and will fulfill the conditions therein.

Signature........ M .... Dated this day of........ 8/3/15 .... 2016.

cc. Vice-Chancellor
DVC-Research Innovation and outreach

Appendix H: NACOSTI RESEARCH PERMIT

NATIONAL COMMISSION FOR SCIENCE, TECHNOLOGY AND INNOVATION

Telephone: +254-20-2213471, 2241349,3310571,2219420
Fax: +254-20-318245,318249
Email:dg@nacosti.go.ke
Website: www.nacosti.go.ke
when replying please quote

9th Floor, Utalii House
Uhuru Highway
P.O. Box 30623-00100
NAIROBI-KENYA

Ref. No. **NACOSTI/P/16/60789/11178**

Date:
16th June, 2016

Milka Njeri Wanjohi
Kenyatta University
P.O. Box 43844-00100
NAIROBI.

RE: RESEARCH AUTHORIZATION

Following your application for authority to carry out research on "***Maternal body composition and infant birth-weight in urban informal settlements in Nairobi County Kenya,***" I am pleased to inform you that you have been authorized to undertake research in **Nairobi County** for the period ending **13th June, 2017.**

You are advised to report to **the County Commissioner and the County Director of Education, Nairobi County** before embarking on the research project.

On completion of the research, you are expected to submit **two hard copies and one soft copy in pdf** of the research report/thesis to our office.

BONIFACE WANYAMA
FOR: DIRECTOR-GENERAL/CEO

Copy to:

The County Commissioner
Nairobi County.

The County Director of Education
Nairobi County.

Appendix I: NACOSTI RESEARCH CERTIFICATE

THIS IS TO CERTIFY THAT:
MISS. MILKA NJERI WANJOHI
of KENYATTA UNIVERSITY, 10787-100
Nairobi,has been permitted to conduct
research in *Nairobi County*

on the topic: *MATERNAL BODY COMPOSITION AND INFANT BIRTH-WEIGHT IN URBAN INFORMAL SETTLEMENTS IN NAIROBI COUNTY KENYA*

for the period ending:
15th June,2017

Applicant's
Signature

Permit No : NACOSTI/P/16/60789/11178
Date Of Issue : 16th June,2016
Fee Recieved :ksh 1000

Director General
National Commission for Science,
Technology & Innovation