EFFECTIVENESS OF BAOBAB-PEARL MILLET BLEND ON BONE MINERAL DENSITY OF WOMEN AGED 40-65 YEARS IN KITUI COUNTY, KENYA

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A PROPOSAL SUBMITTED IN FULFILLMENT OF THE REQUIREMENT FOR THE AWARD OF THE DEGREE OF DOCTOR OF PHYLOSOPY (NUTRITIONAL SCIENCES) IN THE SCHOOL OF HEALTH AND BIOMEDICAL SCIENCES, TECHNICAL UNIVERSITY OF KENYA

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DECLARATION

This proposal is my original work and has not been presented for a degree or any other award in any other university

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ABBREVIATIONS AND ACRONYMS

BDFP- Baobab Dry Fruit Pulp
BMD- Bone Mineral Density
BMI- Body Mass Index
DXA- Dual X-ray Absorptiometry
Gok- Government of Kenya
GPAQ- Global Physical Activity Questionnaire
HRT- Hormonal Replacement Therapy
HH- Household
IDB- International Data Base
IOF- International Osteoporosis Foundation
KOPECS- Kenya Osteoporosis Prevention and Elderly Care Services
MoH- Ministry of Health
NOF- National Osteoporosis Foundation
PAOC- Pan Asia Osteoporosis Congress
PTH- Parathyroid Hormone
SSPS- Statistical Package for Social Sciences
WHO- World Health Organization

DEFINATION OF TERMS

Bone mineral density- The density of the bone can be determined from this ability to absorb radiation and stated as a value on a scale that is compared with a standard or normal data. A single beam of radiation or two beams of radiation coming from two independent sources can be changed by the bone along a specified length of bone in this measurement. By doing this, osteoporosis is detected if it is present.

Dietary Intake- refers to the kinds and quantity of food a person eats.

Food allergy- Is an immunological response that happens quickly after consuming a specific food.

Fracture risk- Depending on previous fracture history and bone mineral density, the anticipated risk of suffering a fracture.

Hormone replacement therapy- a phrase used to describe adding any natural hormone to food. It refers to the replacement of oestrogen or progestogen hormones when given for osteoporosis.

Peak bone mass- An individual's peak bone mass was reached between their mid-20s and early-30s, at the latest. It affects men more than it does women.

OPERATIONAL DEFINATION OF TERMS

Clinical signs- Women with osteoporosis will likely exhibit physical symptoms including limping and swelling ankles and knees.

Nutritional status- health of senior women as a function of food intake, both in terms of amount and quality. In accordance with the WHO health status cutoff criteria, body mass index (kg/m2) will be used to measure nutritional status.

Physical activity- Energy is expended when the body moves in any way for a prolonged amount of time. It will be determined by the physical activity's types, levels, duration, and intensity as determined by the questionnaire the researcher distributed.

ABSTRACT

Osteoporosis is a condition marked by low bone mass and bone tissue loss, which makes a person weak and frail. Osteoporosis, one of the most incapacitating diseases of older people, increases the risk of bone fractures, notably in the hip, spine, and wrist. Women over the age of 45 occupy a third of the hospital beds, on average. 24.3% of Kenyans are estimated to have osteoporosis. The main objective of the study will be to establish the nutrient content of baobab fortified pearl millet flour and assess its impact on bone health among women aged 40-65 years and with osteoporosis at Kitui East sub county, Kitui County. The study will adopt an experimental interventional trial study design. The sample size will be 93 female osteoporotic patients. Bone Mineral Density (BMD) data will be obtained by performing DEXA scan Absorptiometry tests on the women to get their Standard Deviation (SD) levels whereby a SD of 2.5 and above will be considered normal. Baobab dry fruit pulp, pearl millet and baobab fortified pearl millet flour will be analyzed for the nutrient content of selected nutrients of importance to bone health. Calcium, potassium, magnesium, iron, copper, vitamin C and zinc will be analyzed. The interventional trials on women with osteoporosis will make use of two formulations of pearl millet flour supplemented with baobab. Data on the study sample's nutritional status, physical activity level, and health-related characteristics will be gathered using a standardized questionnaire. With the exception of the nutritional status data, which will be entered and analyzed using the Nutri-survey computer package, data from the structured questionnaire will be entered and analyzed using SPSS version 19 software. Bone mineral density, the nutritional value of baobab-fortified pearl millet flour, and respondents' nutritional status will all be interpreted using World Health Organization (WHO) cutoff values. Chi-square (x^2) will be used to establish the associations between categorical data on BMD and Body mass Index (BMI). Persons Correlation (r) will be done to establish the association between continuous data. Data will be generated into means, frequencies and percentages and grouped into tables, charts and graphs. A p value of ($p< 0.05$) will be considered significant. Since many scientific researches has shown that majority of women don’t consume adequate bone health nutrients such as calcium in their diets, there is need of developing a nutrition intervention product which can provide the required bone health nutrients to prevent and manage osteoporosis.

CHAPTER ONE: INTRODUCTION

1.1 Background to the Study

Due to increased bone loss and alteration of the bone's natural design, osteoporosis is a condition that results in porous, weak, and fragile bones. This bone porosity increases the risk of hip, wrist, and spine fractures (World Health Organisation [WHO], 2017). The World Health Organisation (WHO) defines osteoporosis as having a bone mineral density (BMD) that is 2.5 standard deviations (SD) or more below the average value for young, healthy women (WHO, 2017). Osteoporosis is a documented, well-defined disease that affects more than 8.9 million people annually, with more than 4.5 million of those fractures happening in poor countries, according to the International Osteoporosis Foundation (2017). In comparison to older men who are at moderate risk, women have a higher risk of getting osteoporotic fractures (WHO, 2016). According to the Pan Arab Osteoporosis Congress (2015), the two continents where osteoporotic fractures are expected to rise the most in the next ten years are Asia and Africa.

About 2 million people in Nigeria, 1 million in Chad, 5 million in South Africa, and 3 million in Kenya are impacted by osteoporosis in Africa. According to WHO (2015), there are an additional 2 million undiagnosed instances of osteoporosis in Kenya. This is mostly attributable to the Dual X-ray Absorptiometry (DXA) technology's high price and limited availability, particularly in the majority of developing nations (Clark, Carlos, & Martinez, 2016).

Kenyan women are more likely to develop osteoporosis because of the demanding work they do on the farms, including carrying heavy loads of firewood and doing unending housework. According to studies (Kimiywe et al., 2015), they do not consume enough calcium in their diets. According to a study by Chadore et al.

(2018), the baobab fruit pulp is especially high in calcium, which contributes to an increase in bone mineral density. Adequate diet is necessary, especially in calcium, vitamin D, and other minerals like phosphorus, magnesium, zinc, and vitamin C that are crucial for bone health (Brown, 2017). Calcium is an essential component for bone growth and modelling. Additionally, studies have revealed that the body's ability to absorb calcium declines as we age, placing an elevated risk of osteoporosis in the elderly (Kanis, 2017). According to Kenya Osteoporosis Prevention and Elderly Care Services (KOPECS, 2015), osteoporosis not only ranks highly among diseases that render people immobile and have serious repercussions that could be fatal in elderly people, but it is also a primary cause of fractures in Kenya. In order to maintain health, quality of life, and independence in the senior population, osteoporosis and its associated fractures are thought to need to be prevented (Johnell et al., 2017). This is because osteoporosis impacts mobility.

By incorporating the fruit pulp from the Adansonia digitata into most of their meals, African women can increase their calcium intake and levels in their bodies. The naturally sweet and sour pulp is abundant in numerous minerals vital to bone health. Since pulp contains seven times as much vitamin C as oranges, it enhances the body's capacity to absorb and distribute calcium and iron. In comparison to milk, baobab pulp contains calcium that is three times greater (Vila-Real et al., 2017). In comparison to oranges and blueberries, it has ten times more antioxidants. It has seven times as much potassium as a banana and three times as much iron as red meat. Magnesium is 2 times more bioavailable than in spinach. Adansonia digitata also has higher concentrations of the important fatty acids 3, 6, and 9 than sea fish (Kinuthia et al., 2018; Oyoo et al., 2015).

This nutritional profile of baobab pulp suits it as a viable aid in the prevention and effective in treatment of osteoporosis. The minerals and vitamins are beneficial to people suffering from osteoporosis as they contain anti-inflammatory, antipyretic and analgesic components which improve the mobility of the patients (Parvean et al., 2018).

A study done by Xing-Nuo Li et.al, (2017) showed that baobab fruit pulp has high nutritional and medical values. Despite the numerous reports on the pharmacological potential, little has been done on developing interventional products to manage osteoporosis. In this study, development of interventional product of baobab-pearl millet blend will be done for consumption by respondents to assess on its efficacy on bone mineral density among the women.

1.2 Problem Statement

Osteoporosis was listed as a major health concern by WHO in 1994, yet it is still a growing issue that affects both developed and developing nations (WHO, 2015). Recent years have seen advancements in our understanding of the illness and how to treat it in women, particularly in the industrialised world. Although osteoporosis takes a severe toll on the health of the elderly, especially African women who are more susceptible to the condition than males, there is still a lack of scientific evidence in most African nations to support interventions and patient care (KOPECS, 2015). An indication of persistent osteoporosis can be seen in many African villages where elderly ladies and men support frail bodies with walking sticks. Restricted access to diagnosis and treatment before the first fracture causes underdiagnosis and undertreatment of the condition in the majority of underdeveloped nations.

There is a dearth of osteoporosis research data in Kenya. Children and women who are close to becoming pregnant have been the subjects of the majority of dietary and health research and treatments. Due of this, elder women have been overlooked while planning health initiatives. Despite food consumption surveys showing that the majority of women do not consume the recommended micronutrients, such as calcium-rich foods, and are not taking any form of micronutrient supplementation, the Kenyan national food fortification plan has excluded calcium, one of the nutrients most important for bone health, from its fortification program. Furthermore, the government of Kenya is yet to provide an accessible comprehensive medical cover for the elderly, majority of whom are left to suffer and often die because they cannot afford medical care. Food insecurity and undernutrition prevalence also remains high among the elderly which compound to the health challenges of osteoporotic patients (Parvean et al.,2018). Planners must prioritize bone health initiatives and care for osteoporotic women based on adequate data.

The majority of hospitals in the nation lack the equipment and medications required for the diagnosis and treatment of issues relating to bone health. This may have led to an incorrect diagnosis of osteoporosis, which resulted in inadequate management and treatment (Adebiyi et al., 2016). The Kenya National Clinical Nutrition and Dietetics Reference Manual (GoK/MoH, 2015)'s exclusion of information on osteoporosis management is another proof that osteoporosis has not received priority attention in Kenya's healthcare systems. According to a clinical study carried out in Kenya by Odulla in 2003, the prevalence of osteoporosis is 24.3% nationwide. Considering the possibility of many unreported cases, this calls for increased efforts to help control the pandemic in the country. This study is set out to conduct an experimental intervention among the women suffering from osteoporosis in order to establish

whether baobab fortified millet flour can boost the body's calcium supply leading to mobilization of calcium in the bones and consequently improving bone health.

In the last 10 years, Kitui Level Five Hospital, the county referral hospital in Kitui, Kenya, has seen an increase in osteoporosis cases from the county's residents (MoH, Kitui Level Five Hospital, 2019 annual hospital report). There is therefore a need to conduct an interventional study among the women with osteoporosis with the aim of improving their nutritional status through an intervention designed to boost dietary calcium through a fortified baobab-pearl millet flour. If successful, this intervention could be scaled up to other parts of the country.

1.3 Purpose of Study

The purpose of the study will be to in cooperate baobab dry fruit pulp into pearl millet and assess its effectiveness on bone mineral density among women with osteoporosis aged 40-65 years in Kitui County, Kenya.

1.4 Objectives of the Study

1.4.1 General objective

1. To determine the impact of baobab-pearl millet blend on the bone mineral density among women with osteoporosis aged 40-65 years in Kitui County, Kenya.

1.4.2 Specific objectives

To achieve the purpose of the study, the specific objectives will be;

1. Determine the nutrient value of baobab dry fruit pulp, pearl millet and baobab-pearl millet blend flour with respect to proximate composition, calcium, potassium, magnesium and zinc.

2. To formulate baobab-pearl millet blend flour that is acceptable and meets the nutrient requirements for bone health.
3. Assess the sensory characteristics and acceptability of baobab-pearl millet blend by women with osteoporosis aged 40-65 years in Kitui County.
4. Assess the nutritional status, morbidity patterns and dietary intake at baseline and after intervention among women suffering from osteoporosis aged 40 to 65 years in Kitui County.
5. Assess baseline and post intervention bone mineral density levels of women with osteoporosis aged 40 to 65 years in Kitui County.
6. Determine the contribution of consumption of baobab-pearl millet porridge on bone mineral density, nutritional status and morbidity patterns of women suffering from osteoporosis aged 40-65 years in Kitui County.

1.5 Hypotheses

H_{o1}: There is no significant association between the bone mineral density and consumption of baobab-pearl millet flour blend among women with osteoporosis aged 40-65 years in Kitui County.

H_{o2}: There is no significant relationship between nutritional status and consumption of baobab-pearl millet flour blend among women with osteoporosis aged 40-65 years in Kitui County.

H_{o3}: There is no significant improvement on morbidity patterns and consumption of baobab-pearl millet flour blend among women with osteoporosis aged 40-65 years in Kitui County.

1.6 Significance of the Study

By introducing a new therapeutic technique in the management of the osteoporosis pandemic, the study's findings will advance our understanding of osteoporosis. It will also avail a new product of baobab fortified millet flour accessible to the elderly who suffer from osteoporosis and the general population which will help improve bone health management. The health care professionals will have research evidence to advise their patients on the consumption of baobab fruit pulp. Policy makers in the Ministry of Health will use the evidence generated by the research to incorporate calcium in the national fortification plan of staple foods with micronutrients. Findings of this study will also be useful to health care partners such as World Health Organization regional office and other stakeholder agencies in the health sector in informing various initiatives on micronutrient fortification of staples

1.7 Assumption of the Study

The study will assume that the women with osteoporosis will be available to collect the flour and consume the porridge for the six months of implementation phase of study.

1.8 Study Limitations

The sample population will be followed up from Sombe primary school in Kitui East Sub County, Kitui County. It might be difficult to follow all of them at home to check on how they are measuring and preparing the porridge. Therefore, demonstrations on how to measure and prepare the porridge will be continuously done at every monthly contact meeting with the respondents at the primary school.

1.9 Study Delimitations

The study population will include women with osteoporosis aged 40-65 years in Kitui County. The nutrient content of baobab dry fruit pulp, pearl millet and baobab-pearl millet flour blend will be analyzed at baseline for important nutrients in bone health including calcium, potassium, magnesium and zinc levels.

1.10 Conceptual Framework

The Conceptual Framework (Figure 1.1) describes the contribution of micronutrients in millet and baobab dry fruit pulp in bone health.

Pearl millet which is common porridge flour in most Kenyan families is high in bone health micronutrients such as calcium, magnesium, manganese, phosphorus and antioxidants. Pearl millet is also an alkaline food which helps in reduction of inflammation and pain in osteoporosis. Many children and adults are at a risk of developing micronutrient deficiencies despite the frequent consumption of millet due to the low bioavailability of nutrients because of the high phytic acid content in the millet. Proper preparation of the millet seeds by soaking in water before milling of the flour is important to reduce the antinutritive elements and enzymes.

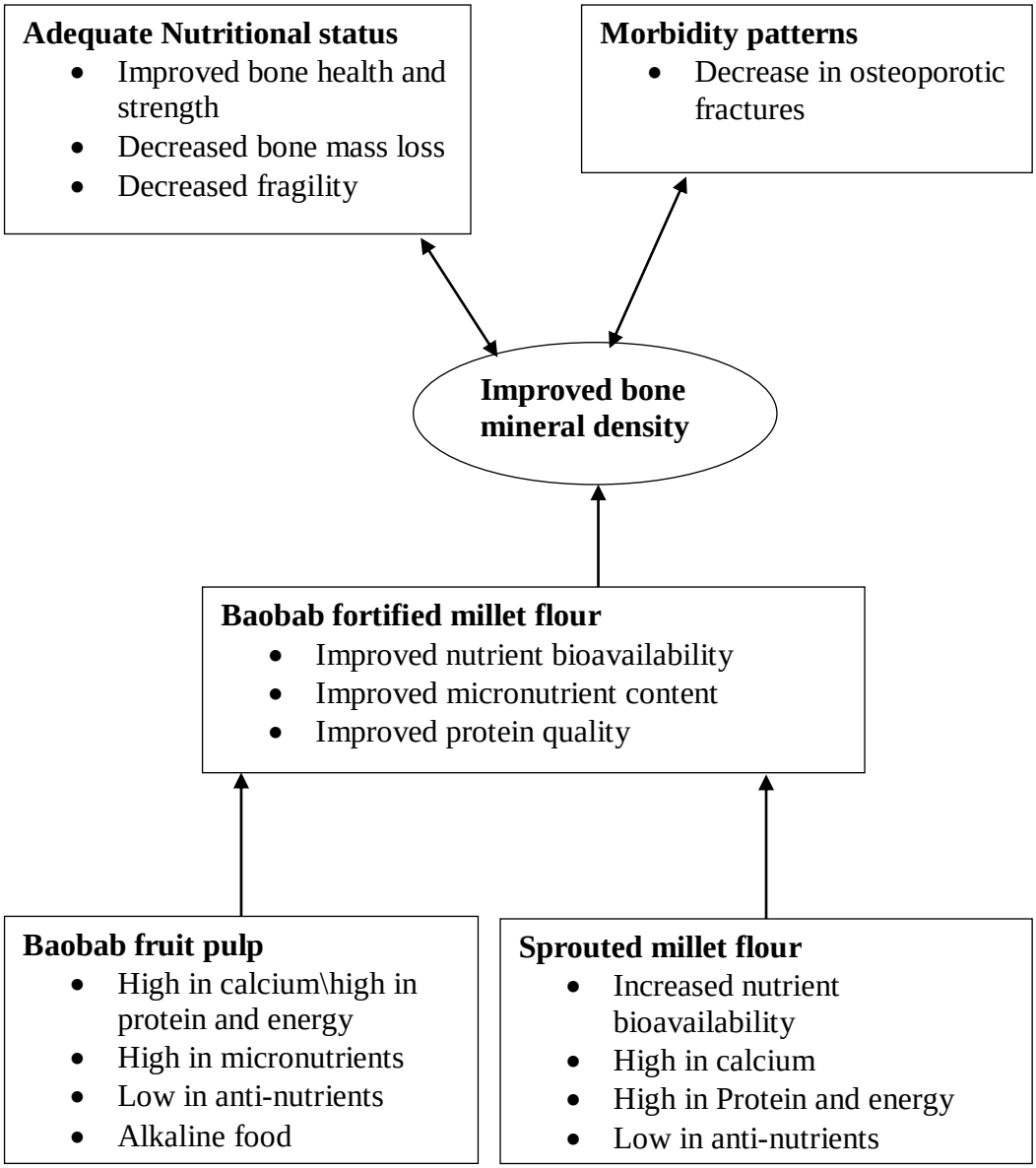

Figure 1 .1: Conceptual Framework on Micronutrient Contribution to Bone Health

Source: Adopted from Brown et al., 2017

Baobab fruit pulp is naturally dry and purely organic food which is rich in fibre, calcium, potassium, magnesium, iron and zinc. Enrichment of pearl millet flour with baobab fruit pulp will result in enhanced micronutrient profile of the composite flour whose consumption is capable of enhancing the dietary intakes of the said

micronutrients Baobab fruits which are sold at low cost are available across many regions in Africa hence their consumption should be encouraged.

CHAPTER TWO: LITERATURE REVIEW

2.1 Overview of Osteoporosis

Depending on the homeostatic mechanism in play, osteoporosis can progress quickly or slowly. Osteoporosis type 1 or postmenopausal osteoporosis is a phase of bone loss that accelerates when oestrogen or testosterone levels are low. The loss of bone mass is larger in women who are more prone to bone loss due to oestrogen decline in the 3 to 5 years following menopause (Brown et al., 2017). Men get osteoporosis later than women do. A woman's ability to eat well can assist reduce the rate of bone loss. After menopause, maintaining bone density depends in large part on consuming enough calcium, vitamin D, and other nutrients, such as protein (National Osteoporosis Foundation, 2017). The slowing down of calcium absorption causes senile osteoporosis, also known as Type II osteoporosis. Due to the effects of gradual bone loss over time, osteoporosis is a condition that is more prevalent as people age (Pfleger, 2015).

Osteoporosis affects 80% of women versus 20% of men, which is a four-fold increase in prevalence. Men have more peak and total bone mass than women because of their larger bodies and denser bones. Due to their sturdier bones than White people, Black people experience less osteoporosis. Men and women experience osteoporosis differently. Wrist fractures are less frequent in women, who also frequently break their hips and backbones. Although fractures at or near the wrist occur more frequently in men with osteoporosis than in women, hip fractures are nonetheless common in this population. One in two women and one in eight men over 50 may have an osteoporosis-related fracture during their lifetimes (Rizzoli et al., 2015).

2.2 Relationship between Bone Mineral Density and Pathogenesis of Osteoporosis

The first three decades of life are when bone develops and mineralizes, during which time bone mass is predominantly gained. For another ten to fifteen years following the adolescent stage, bones continue to widen and accumulate minerals (Asomaring et al., 2016). Between the ages of thirty and forty, bones attain their peak density, or their highest degree of mineral concentration. After that, the mineral content of bone stops rising. Osteoporosis is less likely to occur the higher the peak bone mass. To put it simply, those who have higher peak bone mass have more calcium to lose before their bones begin to degrade and shatter easily. Additionally, because they have greater bone mass to decrease, men are more likely than women to develop osteoporosis. Between the ages of forty and fifty for women and around sixty for males, bone size and density frequently remain quite steady. Low bone density and a higher risk of fractures result from insufficient peak bone mass augmentation and considerable bone loss (Mbora et al., 2015).

2.2.1 Inadequate Bone Mass

Despite the fact that osteoporosis is most frequently noticed in the elderly, the danger for acquiring it in later life starts in childhood and adolescence. When a person develops larger, denser bones during childhood and adolescence, osteoporosis is postponed. A Yugoslavian epidemiological study found that higher calcium consumption throughout adolescence increased peak bone mass regardless of activity or other factors (Johnell et al., 2015). Before the age of 30, calcium intakes of approximately 1200 milligrammes per day along with adequate vitamin D status increased bone mass, according to studies that examined the relationship between

dietary calcium intake, vitamin D status, and bone density. Bone density is likely to be preserved in people over 50 who ingest 1200–1500 mg of calcium and 400–800 mg of vitamin D daily (Brown, 2017).

By transitioning from a low to an adequate diet of calcium from foods like baobab fruit pulp and improving vitamin D status, poor bone mineral density can be somewhat increased. The improvement in bone mineral density is more obvious the earlier in life this occurs. High levels of bone density can be achieved, but they do not last forever; instead, bone needs to be continually regenerated by getting enough calcium and vitamin D. Inactivity and insufficient exercise are the main causes of osteoporosis. Bones are weakened by hospitalization, extended bed rest, and a sedentary lifestyle. Because bone grows in reaction to pressure on the bone tissue, weight-bearing or resistance activities are required to develop bone mass. The body will respond by depositing minerals such as calcium, phosphorus, fluoride, and boron into the bone matrix as you push on the bone more frequently and forcefully, provided that you do not break it.

2.2.2 Increased Bone Loss

The skeleton serves as the body's structural foundation and calcium storage system. Jaw bones and teeth are examples of bone tissue. About 99% of the calcium in an adult is found in the bones and teeth. In blood, soft tissue, and extracellular fluids, the remaining 1% of calcium is found bound to protein. This reserve is necessary for the function of the enzyme systems that control blood coagulation, muscle contraction, and nerve transmission.

Calcium is tightly regulated by hormone systems in order to be regularly accessible to carry out various activities. The body responds to a decrease in calcium levels by producing more PTH through the parathyroid glands. PTH releases calcium from bone, raising blood calcium levels as a result. The thyroid glands are stimulated to secrete calcitonin when there is too much calcium in the blood. Release of calcium from storage is slowed by the hormone calcitonin. To keep enough calcium levels for those calcium messenger activities, bone mineral reserves are continuously broken down and regenerated. Regular dietary intake of the bone-building minerals should be combined with routine weight-bearing activity. Calcium, magnesium, phosphorus, fluoride, boron, and vitamins D and K, which also give calcium when required, keep the skeleton strong. The body's top priority is to maintain blood calcium levels for neuron, muscle, and enzyme activities when a part of this build-dissolve-rebuild cycle is damaged. Insufficient calcium levels, malabsorption, or excessive calcium excretion cause bone loss (Brown, 2017).

Another cause of osteoporosis is a deficiency in the mineral phosphate required for bone mineralization. The bone-building cells can use both elements when there is a balanced ratio of calcium to phosphorus, which is what a diverse diet provides. Phosphorus deficiency encourages the release of calcium from the skeleton. Despite the fact that phosphorus is a plentiful dietary component, some antacids bind to it and prevent the body from using it. When phosphorus is lacking, bone mineralization is postponed until additional phosphate is available. Vitamin D deficiency also hinders the synthesis of minerals. Additionally, as people age, their skin produces less vitamin D from the sun (Klibanskki & Adams-Campell, 2015).

2.3 Nutritional value of baobab dry fruit pulp

Since the native population views the baobab tree as a protector and a symbol of their continent, it is also referred to as the Tree of Life in Africa. Adansonia Digitata is its scientific name. African people and animals depend heavily on the baobab because it gives them food and medicine. The baobab is used in African medicine to cure inflammation, fever, smallpox, diarrhoea, dysentery, and dysentery. It is a deciduous tree that may grow to a height of 30 metres (90 feet) and a circumference of 15 metres (45 feet). During the dry season, the bloated, short, bottle-shaped trunk can hold up to 30,000 gallons of water, providing water storage for the settlements. The spongy wood of the baobab tree, which has a lifespan of up to 5000 years and is fire resistant. The only fruit in the entire world that naturally dries on the branch is this one. The baobab's green, spiky, velvet-like covering turns into a smooth, brown, coconut-like shell after baking in the sun for six months. The fruit's dry, white-ish pulp, which is enclosed in a hard shell, is devoid of any moisture (Kinuthia et al., 2018).

Bone health is known to be impacted by diets with low calcium and vitamin D intake (Bowman & Russell, 2016). In order to maintain adult bone mass, calcium and vitamin D are crucial (Naughton & Greene, 2016). According to a study on food consumption habits by Waudo et al. (2015), calcium intakes among Kenyan women were noticeably low. In a study conducted at Mbagathi Hospital by Kieti (2015), the majority of the Kenyan women interviewed reported poor calcium intakes since they typically drank milk, the main source of calcium, in tea. Calcium, a key mineral in the development and maintenance of strong bones, is provided through the

consumption of calcium-rich foods, such as native vegetables and fruits, as demonstrated by (Mbora, Jamnadass & Lilleso, 2016).

Table 2.1: Nutrient content of baobab dry fruit pulp

Nutrient	**Amount per 100g**
Carbohydrates	80 g
Protein	3.7 g
Fibre	45 g
Ca	344 mg
K	2189 mg
Mg	196 mg
zc	0.92 mg
Vitamin C	173.2 mg

Baobab dry fruit pulp includes vitamin C that is six times as potent as an orange and other vital nutrients needed for bone development. The protein bone matrix collagen, which is necessary for the creation of strong bones, is generated in large part by vitamin C. Additionally, it contains vitamin A, phosphorus, potassium, iron, sodium, zinc, and magnesium, all of which promote calcium absorption and bone production as well as the active transport of calcium in the small intestine and colon. Vitamin B complex is another component found in baobab dry fruit pulp and is a cofactor for collagen metabolism. The interaction between osteoblasts, which create new bone, and osteoclasts, which break down existing bone, may reduce the rate of bone loss (Apalset et al., 2017). Since protein is linked to proper levels of bone growth factors like insulin-like growth factor, protein is also present in baobab as necessary amino

acids, which are crucial nutrients in the prevention of osteoporosis (Budek et al., 2017).

2.4 Baobab-pearl millet flour blend

The availability and accessibility of calcium rich flour which can be palatable, affordable and acceptable to majority of Kenyans is key in prevention and management of osteoporosis. This study will carry out experimental interventional trials by providing baobab fruit pulp in fortified millet flour to women suffering from osteoporosis to establish if it can impact more calcium on their bones hence increasing the bone mineral density. Pearl millet is a super food which contains high amount of calcium and other nutrients needed for bone health such as vitamin D, magnesium, phosphorus, manganese and antioxidants. It's also an alkaline food which helps to reduce the inflammation in osteoporosis hence decreasing pain.

Table 2.2: Nutrient content of pearl millet

Nutrient	Amount per 100g
Carbohydrates	146g
Protein	22g
Fibre	13.3g
Ca	16mg
Zn	3.4mg
P	370mg
K	390mg
Mn	228mg

Hence the fortification of pearl millet flour with baobab will provide super flour with increased calcium content and other nutrients for bone health which can improve the bone mineral density of the women with osteoporosis.

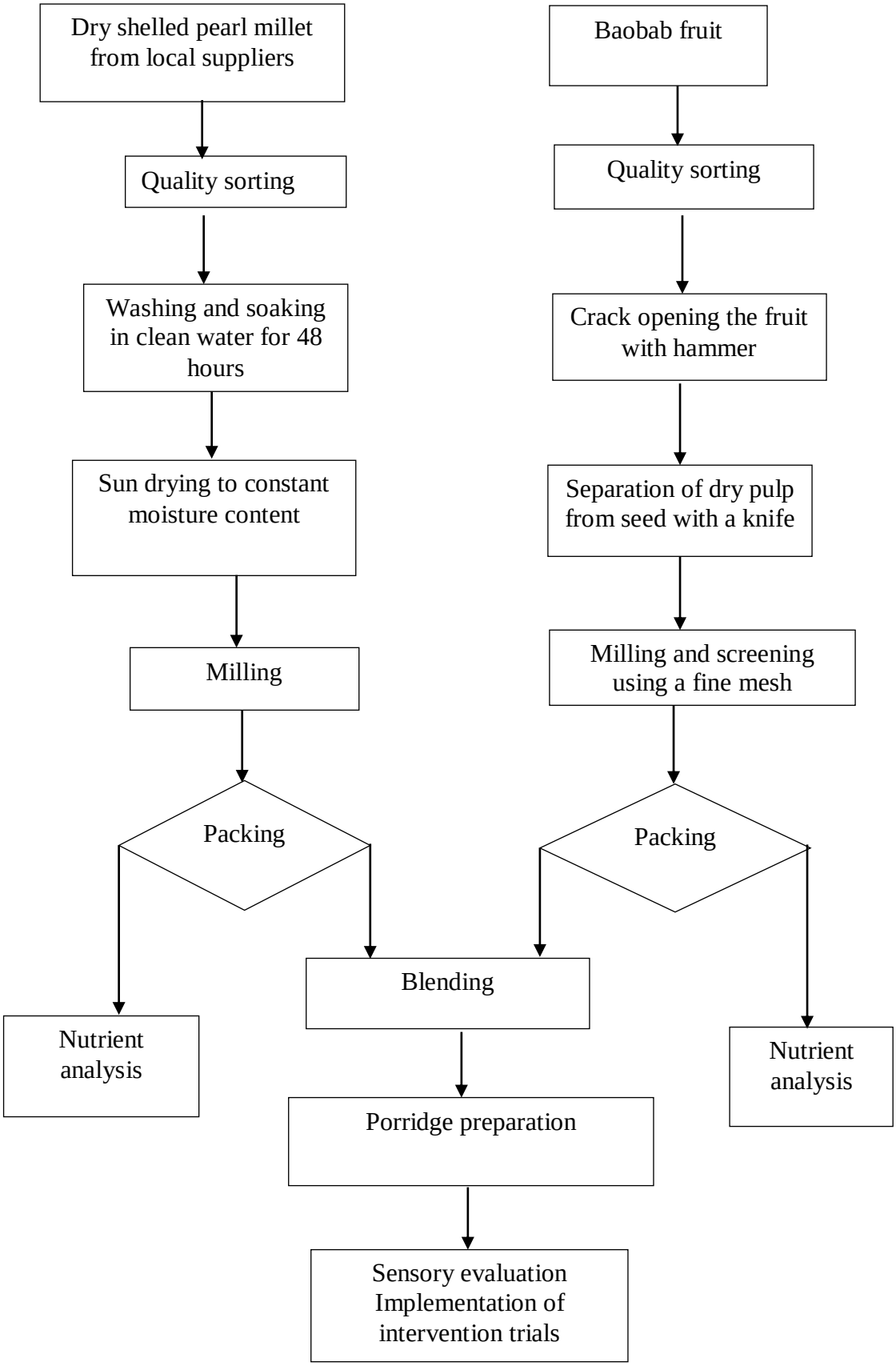

Figure 2.2: Flow Chart of Baobab-pearl Millet Flour Formulation

2.5 Nutritional status and osteoporosis

Low BMI is brought on by inadequate nutritional status, which increases the risk of osteoporosis in early postmenopausal women due to low bone mass and accelerated bone loss as ageing progresses (Ravn et al., 2015). The importance of nutrition in establishing and sustaining bone strength has long been recognized. A risk factor for osteoporosis has been found as a low BMI of less than 19 kg/m2. persons with eating disorders, elderly persons who are frail, and some athletes could have low BMIs and need to be checked for osteoporosis. Low BMI women are more likely to develop osteoporosis. Patients should be urged to maintain a normal weight to assist lower the risk of osteoporosis.

2.6 Morbidity factors associated with Osteoporosis

For older persons, preventing fractures is crucial since they increase the risk of dying young. In a study carried out in Europe by (Johnell et al., 2017), 13% of older women who shattered a hip died within six months of the fracture. Death is caused by complications from the break rather than the fracture itself. Impaired mobility is one of these consequences, which makes it difficult to complete daily tasks like eating and exercising. The wrist, hip, and spine are where the majority of these fractures happen (Johnnell & Kanis, 2017). Most spinal fractures don't cause any symptoms, unlike hip fractures. Due to the height loss brought on by osteoporosis, women with compression in the spinal column experience a condition known as shrinking height, which can result in the dowager's hump or kyphosis. The process of losing height is typically gradual and painless; for instance, a lady who was 5 feet 6 inches tall at age 30 would only be 5 feet tall at age 83 (Brown, 2017).

As these conditions prevent mineral deposition in the bones, people with conditions including diabetes, rheumatoid arthritis, and rickets are more likely to develop osteoporosis later in life (Guardiola et al., 2016). Women should take nutritional supplements after menopause, notably calcium and vitamin D, as their absorption declines with ageing (Burckhardt, 2016). These supplements offer pain alleviation for the body's loss of minerals due to the body's decreased oestrogen production during menopause. Many older women lack the financial resources necessary to buy nutritional supplements for bone health. This investigation will identify the respondents' patterns of morbidity. To stop the onset of osteoporosis, it's crucial to adopt a healthy lifestyle early in life. Compared to non-smokers, smokers incur higher bone deterioration and fractures. Abuse of alcohol can cause osteoporosis and an increase in bone fractures (Johnell & Kanis, 2017).

2.7 Physical activity levels contribution in osteoporosis

Both low and high levels of physical activity as well as body weight have a major impact on bone health. As a result of the pressure that muscles apply to the bones during contraction, the bones become denser and develop more trabeculae (Harsh, Stefarick, & Stafford, 2016). As a result, active bones are denser than sedentary ones. Weight gain and heavier body weights both put equivalent strain on the bones and increase their density. Obesity, however, as well as strenuous housework like carrying large loads of firewood or water, or digging for extended periods of time, can result in decalcification of the bones and osteoporosis.

2.8 Summary of Literature Review

Despite the fact that osteoporosis is most frequently noticed in the elderly, the danger of getting it in later life starts in childhood and adolescence. By raising knowledge of the value of healthy food, exercise, and lifestyle changes to delay the start of the disease, the high prevalence of osteoporosis in our nation can be reduced. Between the ages of 30 and 40, bones attain their peak density, or their highest degree of mineral concentration. Following that, no longer increases in bone mineral content. Osteoporosis is less likely to occur the higher the peak bone mass. Simply said, those with higher peak bone mass have more calcium to lose before their bones start to deteriorate and break readily.

CHAPTER THREE: METHODOLOGY

3.1 Introduction

Presented in this chapter is the methodology of the study. Research design, study site, sample size determination and sampling procedures are among the subtitles discussed.

3.2 Research Design

Experimental interventional trial study design will be adopted. The study design will enable the collection of data to assess the respondents on the impact of baobab- pearl millet flour blend before and after the intervention. Food nutrient analysis will be done to determine the nutrient value of baobab dry fruit pulp, pearl millet and baobab-pearl millet flour blend. Dual x-ray Absorptiometry tests will be done on the respondents to determine their bone mineral density with an aim of formulating a new intervention product for management of osteoporosis. In the experimental phase, the respondents will be shown how to prepare and consume the baobab millet flour blend in form of porridge.

3.3 Study Site

The study will be carried out at five villages, in Kitui East sub County of Kitui County. The villages are served by an all-weather road which makes it easily accessible. The main livelihood activity in the villages is subsistence farming whereby growing of indigenous crops such as pearl millet is being promoted by Food and Agricultural Organization of the United Nations together with other stakeholders. Kitui east Sub County is densely populated by the African baobab trees which makes the baobab pulp to be used for the intervention easily available. Baobab fruits are

consumed by the indigenous people hence there will be no risk of toxicity when being used for the intervention.

3.4 Study Population

The study targets women aged 40-65 years with osteoporosis living in Kitui East sub County of Kitui County. This is because degeneration of bones starts after age 40 (Brown et al., 2017).

3.5 Inclusion and Exclusion criteria

The study will include women aged 40-65 years who will been diagnosed to have osteoporosis by Dual X-ray Absorptiometry test of less than 2.5 SD. Women with osteoporotic fractures and diseases related to osteoporosis will also be included in the study. The study will exclude women aged 40-65 years with diseases not related to osteoporosis. Those with motor accident fractures and other non-osteoporotic fractures will also be excluded.

3.6 Dependent and Independent Variables

The women's bone mineral density will be the main dependent variable in this study. The target group's nutritional health, consumption of a baobab-pearl millet flour blend, the respondents' patterns of morbidity, and the amount of physical activity within the study population will all be independent factors.

3.7 Sample Size Determination

A baseline study conducted at Kitui East sub county on women with osteoporosis aged 40-65 years in 2019 (MoH, Kitui county referral hospital 2019) come up with a sample size of 154 respondents. Therefore 60% of the 154 which is 93 respondents will be used to conduct the interventional study at the same village. This sample will be deemed sufficient since, according to Gall and Borg (1997), comparative studies

should include at least 30 people per group. The respondents will be assigned numbers from one to one fifty-four. Simple random sampling procedure will be used to pick the first 93 numbers which will be randomly computer generated from the 154 numbers.

3.8 Sampling Procedure

The five villages in Kitui East sub County of Kitui County will be purposively chosen. This is because there are ongoing agricultural support initiatives promoting farming of indigenous crops such as millet, sorghum, figure millet and green grams. Women aged 40- 65 years who will have painful limbs will be tested for bone mineral density and those found to have a SD of less than 2.5 will be recruited for the experimental interventional trials. A total of 93 respondents will be recruited.

Numbers in the calculated sample size of 93 respondents will be randomized into two groups which will be one experimental group and one control group by the researcher assisted by a Biostatician. The numbers 1 to 93 will be printed on a piece of paper then each number will be cut in the same size and folded in the same pattern as all the numbers. Then the papers will be put in an opaque container covered with a lid which will be thoroughly shaken before a piece of paper is picked by the respondents. Simple systemic random sampling will be done where by those respondents who will pick uneven numbers will belong to the experimental group of 47 respondents and those who will pick even numbers will belong to the control group of 46 respondents.

3.9 Data Collection Instruments

A dual x- ray Absorptiometry machine will be used to obtain the bone mineral density of the respondents. Adansonia digitata dry fruit pulp and millet flour will be used to make the different blends of baobab fortified millet flour. Nutri-survey computer software will be used to determine which baobab fortified millet flour blend has the highest nutrient content of calcium and other minerals for bone health to be used for the intervention. Flame flatometer, UV-vis spectoflatometer, HPLC and AAS will be used to analyze the nutritional value of baobab dry fruit pulp. Information on nutritional status, health status, and physical activity levels of the study population will be collected using a researcher administered questionnaire (Appendix 1).

3.10 Pre-testing Research Instruments

The pretesting size should be between 1% and 10% of the sample size, according to Mugenda & Mugenda (2003). The research questionnaire will be pretested on 10 women with osteoporosis at the nearby Kawea village. In this instance, 10 osteoporotic women will make up 10% of the total population of 93. The women will be invited to remark on how well-written the structured questionnaire items are before changes are made to increase validity and reliability. To produce trustworthy and precise nutritional parameters, the tool validation of the laboratory equipment that will be utilized for nutrient analysis will be calibrated using established standards. Validity is the precision with which a tool evaluates the characteristics of the data that will be collected, whereas reliability is a gauge of how consistently a research tool produces results over time.

To test for sensory acceptability of the baobab millet flour blend the two blends analyzed to have high nutrient content for bone health will be used. Porridge will be prepared by the researcher and served to the patients in 50mls portions in the morning hours at Kawea primary school in Kawea village. The palatability will be assessed in terms of the aroma, taste, the facial expressed as they take the porridge, amount taken and the duration taken to consume the porridge. The sensory evaluation will be carried out using the 9-point hedonic scale where by 1=dislike extremely and 9=like extremely (Appendix 2). The respondents will be provided with water to rinse their mouths after each sampling and take 5 to 10 minutes before taking the next blend of porridge.

3.11 Data Collection Procedures

The lead investigator and four research assistants will administer the questionnaire during the scheduled interviews to gather data on the study population's dietary status, morbidity patterns, and physical activity. Information on BMD, nutritional value of Adansonia digitata pulp and millet and formulation of baobab millet flour blend will be collected using the following procedures:

3.11.1 Bone Mineral Density Assessment-

On a Hologic QDR-2000 dual-energy x-ray absorptiometer, we will employ a fan beam and an array of detectors to measure the anteroposterior and lateral lumbar-spine and hip bone mineral densities (Hologic, Waltham, Massachusetts). Radial bone density will be evaluated using pencil-beam imaging. The women will be lying supine throughout the anteroposterior and lateral lumbar-spine scans. The femoral neck, the anteroposterior lumbar spine (vertebrae L1 to L4), and the lateral lumbar-

spine vertebral bodies (vertebrae L2 to L4) will all have their area density evaluated in grammes per square centimeter. By multiplying the lateral lumbar spine measures by the average vertebral body width determined from the anteroposterior scan, grammes per cubic centimeter are obtained. Both the absolute bone mineral density and the normalized standard deviation will be used to express each value. These lumbar-spine tissue scans will be performed both before and after the six-month intervention. The scans will be examined by an orthopedic surgeon to ensure that no iliac crests, ribs, or regions of vascular calcification or degenerative arthritis are crossed or included in the measurements.

3.11.2 Nutrient Content Analysis of pearl millet and baobab dry fruit pulp

The baobab dry fruit pulp and millet will be analyzed at the baseline of the study for selected macronutrients and micronutrients of importance to bone health. The samples will be analyzed in triplicate and average taken. The Nutrient content analysis will be done at the Food, Nutrition and Dietetics Laboratories at Kenyatta University and KEMRI.

3.11.2.1 Determination of Moisture Content

The hot air-drying method, as specified by AOAC methodologies (AOAC, 2005), will be used to measure moisture. Two grammes of samples will be weighed into dry, pre-weighed crucibles and cooked for five hours at 105 oC in a hot air oven (Gellenkamp, UK). After cooling in desiccators, the dry sample will be weighed with an electronic balance (NBY323/64: Avery East Africa). The difference in weight will then be obtained and moisture content calculated as:

MC = (Wt. of crucible + fresh sample) – (wt. of crucible + dry sample)

3.11.2.2 Crude protein

The Kjeldahl technique will be used to calculate the crude protein concentration based on the total nitrogen content (AOAC, 2005). One Kjeldahl tablet will be used to digest one gramme of the material, which will then be distilled in 40% sodium hydroxide. 0.1N hydrochloric acid will be added to the resultant solution to titrate it using a mixed indicator of methyl red and bromocresol green. The percentage nitrogen (N) will be calculated using the following equation of % nitrogen= (S−B) ×N×0.014×D×100/W×V, where D = Dilution factor, T= titer value = (S-B), W =weight of sample, 0.014 = constant value. Crude protein will be obtained by multiplying the corresponding total nitrogen (N) content by a conventional factor of 6.25. Therefore, the crude protein (CP) content will be calculated as the product of % (CP) =N (%) x 6.25).

3.11.2.3 Crude fiber

The finely grounded sample will be digested for 30 minutes with a mixture of nitric and trichloroacetic acids in 70 percent of acetic acid. Starch, protein, lignin, and the majority of the pentosanes will be brought into solution, but cellulose and fat won't be affected. Ethyl ether will be used for fat extraction. The unprocessed fiber will be removed, dissolved in H2SO4, and then exposed to potassium dichromate for oxidation. After titrating the excess dichromate with regular sodium thiosulfate, the amount of crude fiber is estimated using the formula: crude fiber %= {(w1-w2)/w} x 100.

Where w1= weight of acid and alkaline digested sample

w2= weight of incinerator sample after acid and alkaline digestion and

w= weight of food sample

3.11.2.4 Determination of Crude Fat

The soxhlet extraction method will be used to determine crude fat in accordance with the guidelines provided by AOAC (2005). The dried samples' fat content will be extracted at 60ºC into an organic solvent (petroleum ether) and then refluxed for six hours. The percentage of fat content will subsequently be determined by multiplying the weight of the fat extracted by the sample's weight by 100.

3.11.2.5 Determination of Total Ash

The amount of total ash will be calculated in accordance with AOAC (2005). Samples weighing five grammes will be placed in dry crucibles, carbonized on a hot plate, and heated for eight hours at 600ºC in a muffle furnace (Nerberthem model L9/11/C6, Germany). Ash content will be calculated using the weight difference following desiccation of the samples to room temperature.

3.11 .2.6 Determination of Total Carbohydrates

The difference approach will be used to estimate the carbohydrate content, which will be determined using the OCQC method (2005). It will be calculated using the equation below; Carbohydrate (%) =100 − (moisture +fat +protein + ash) %.

3.11.2.4 Vitamin C

High Performance Liquid Chromatography (HPLC) technology will be used to identify vitamin C. Reversed-phase liquid chromatography will be used to analyze vitamin C, together with an isocratic elution process and photodiode assay detection at 254 nm. The separations will be done on a 250mm x 4.6mm Carbon 18 (C18) column from LiChro CART in Darmstadt, Germany. The stationary phase will use a mixture of methanol - water (5.95, v/v). The flow rate of the mobile phase will be 1.0 ml min-1 and 20 µL injection volume of samples and standard will be used in

quantitative analysis. The analytical column will be maintained at a constant 25ºC laboratory temperature. The external standard for vitamin C will contain a natural vitamin C derivative at 1 mg/ml.

3.11.2.5 Mineral Content

Calcium, Potassium and Phosphorus will be analyzed using Flame emission photometer while Magnesium, Manganese, Zinc, Iron and copper using flame atomic absorption spectrophotometer (AA-6200 series). Weighing and digestion with strong nitric acid and hydrogen peroxide of two grammes of each sample will be done. The determination of iron (Fe) and zinc (Zn) at wavelengths of 259.9 nm and 213.9 nm, respectively, will be done using an air-acetylene flame. To measure the concentration of iron, ferrous ammonium sulphate (Fe (NH4)2(SO4)2) will be used as a reference solution.

3.11.3 Baobab-pearl millet flour bend

3.11.3.1 Processing of Baobab dry fruit pulp

The baobab dry fruit pulp will be obtained from the Baobab fruit which is first harvested from the wild by the local women of Kibwezi in Makueni County. The study team will hand separate the seeds and fibrous material from the fruit juice using a knife after using a hammer to break open the fruit's tough shell. After that, the dry fruit pulp will be ground and screened through a fine mesh. Last but not least, the milled dry fruit pulp will be kept in sanitary food-grade packaging containers and marked with the date of packing. Dry baobab fruit pulp has a shelf life of 24 months from the time of harvest.

3.11.3.2 Processing of Pearl Millet Flour

Every month for a period of six months two sacks of 90 kilograms each of quality sorted dry shelled pearl millet grains will be purchased from the local market. The millet will be washed and soaked for three days to reduce on the phytate inhibitors before it is sun dried to 12% moisture content and then whole milling will be done to produce the millet flour. The flour will be packed into one-kilogram portions and stored away from any dirt, direct sunlight and moisture.

3.11.3.3 Preparation of Baobab-Pearl Millet Flour blend

Four formulations of baobab fortified millet flour will be constituted using different proportions of millet and baobab dry fruit pulp. Nutri-survey computer software will be used to analyze and design the flour formulations by applying linear programming. The two formulations out of the four which will have higher nutrient content will be used for the sensory testing before one formulation which will be highly accepted by the respondents is chosen to be used for the intervention.

3.11.3.4 Preparation of Baobab-Pearl Millet blend porridge

The researcher will demonstrate on how to cook the porridge at the primary school to make sure the respondents know how to prepare, consume and store it. 60 grams of the fortified flour will be added to 60 milliliters of water and stirred to make a smooth paste. The paste will be left to ferment in a warm location in the kitchen near the fire place for 2 days while being stirred at least once a day. The fermented paste will be added to 1 liter of boiling water and stirred continuously for 5-10 minutes. Once the porridge is well cooked, sugar will be added to taste as desired. The patients will be given a 300 milliliters cup to use to take the porridge in the morning

and evening for the six months of the interventional trials. A 500 milliliters jar will also be provided to the respondents for storing the portion to be consumed in the evening.

3.11.3.5 Description of the intervention

The experimental intervention trials will provide one formulation of baobab-pearl millet flour blend to the women aged 40-65 years with osteoporosis. The study will use one experimental group and one control. The experimental group will consume porridge from the blend tested to have the highest nutrient content of bone health nutrients and most liked by the respondents. The flours will be distributed biweekly to the respondents at the primary school. Each respondent will be provided with 60 grams of the flour per day to prepare porridge of medium consistency which they will consume in 300mls twice a day in the morning and evening for six months. The porridge will be cooked by adding 60mls of water into 60 grams of the flour then ferment it for 2 days and the fermented paste poured into 1 liter of boiling water. A standardized cooking time of 10 minutes will be used to prepare the porridge hence ensuring uniformity in preparation method. The cooking method and how to take the porridge will be demonstrated before the start of the intervention trials and once monthly for the six months period of the intervention trials to emphasize on uniformity of preparing and consumption the porridge. The research assistants will keep record of distribution of the flours and ensure that all the respondents collected their biweekly refills. The principal researcher together with the research assistants will conduct sporadic checks to check on how the respondents will be preparing and consuming the porridge at home. This will be part of the continuous monitoring and evaluation of the interventional trials to check on performance and gaps which could

be addressed during the six months of implementation of the study. Morbidity patterns and physical activity levels will be assessed biweekly when the respondents come to collect the flour while nutritional status will be assessed monthly. The Bone Mineral Density will be analyzed at baseline and after six months of the interventional trials. The baobab dry fruit pulp and pearl millet will be analyzed once at baseline of the study before they are used to formulate the baobab-pearl millet flour blend.

3.11.4 Anthropometric Assessment

We measured the subjects' weight and arm span anthropometrically. A three-time average of the subjects' weight and arm span will be determined. Because of kyphosis and the distortion of the limbs brought on by osteoporosis, arm spread is chosen above height. The respondents will weigh as trained research assistants take their measurements while wearing no shoes and only the barest minimum of apparel (Kamal, 2009). Using an electronic scale that has been calibrated to zero and placed on a flat surface, the weight will be reduced to the nearest 0.1 kg. Using a tape measure, arm span measures will be obtained to the nearest 0.1 cm, and the values will then be converted into height measurements, per Mohanty et al. (2011).

3.11.5 Morbidity Patterns

By asking the respondents to recount any instances of fractures and illnesses connected to osteoporosis in their lives, morbidity patterns will be identified. The number of times, location and type of fractures and illnesses will be established.

3.11.6 Physical Activity

The structured questionnaire's physical activity assessment portion will be used to gauge participants' levels of physical activity. The African Physical Activity Questionnaire (APAQ), a WHO instrument for monitoring physical activity in impoverished countries, was followed for crafting the questionnaire's questions (Armstrong & Bull, 2010). The type, length, frequency, and intensity of the physical activity will all be covered in the questions.

3.11.7 Training Research Team

The research will have a total of 10 team members. I will lead and coordinate all the research work while being assisted by a junior researcher holding a master’s degree in food science and nutrition. The lead researcher will provide five days of training on the proposed research to four research assistants who have bachelor's degrees in food, nutrition, and dietetics. The goal, objectives, and monitoring of the research will all be highlighted. Additionally, they will receive instruction on obtaining and recording anthropometric measurements, primarily through role playing and additional practise during the pretest. There will also be training in effective questioning methods and data entry into the questionnaires. One Kenya Registered community health nurse will be trained on how to demonstrate on how to issue biweekly flour refills to the respondents and how to capture the information in the commodity book with support from the research assistants. One orthopedic surgeon will be trained on the way of prescribing the dual x-ray absorptiometry test request since we will take the BMD of one particular area of the bone. She/he will help to ascertain the accuracy of the BMD so that it is taken at the same area for all the respondents. One data clerk will be recruited and trained on data entry, coding and will be supported by one biostatician in data analysis.

The principal researcher will train the whole team on porridge preparation method and how to consume it so that each member will emphasis on how to us it at any time of contact with a respondent.

3.12 Data Analysis and Presentation

Every day, all completed questions will be collected, double-checked for accuracy, and coded. The Statistical Package for Social Sciences (SPSS) computer programme, version 17, will be used to enter and analyze all data. The Body Mass Index will be determined using the 2005 Nutri-survey version since it is accurate and takes into consideration variables including age, sex, physical activity, rest, and sleep. The Body Mass Index (BMI), controlled for age, sex, and level of activity, will be used to interpret the nutritional status of the study population. A BMI of less than 18.5 kg/m2 indicates undernutrition, of 18.5 kg/m2 - 24.9 kg/m2 indicates normal health status, and of greater than 25 kg/m2 indicates overnutrition. Using SPSS, data on physical activity and morbidity will be analyzed, and results will be categorized by frequency, duration, intensity, kind, and location. Quantitative data that will be organized into tables, charts, and graphs will be described using descriptive analysis, which includes means, standard deviations, frequencies, and percentages.

To determine the relationships between categorical data, such as bone mineral density values and BMI, Analysis of Variance (ANOVA) and Chi-square will be used. Using Pearson's Product Moment Correlation Coefficient, the direction and strength of the association between the variables will be assessed. A p-value of less than 0.05 is the threshold for statistical significance.

3.13 Logistical and Ethical Considerations

Technical University Graduate School approval will be requested before beginning the research. The National Council of Science, Technology, and Innovations (NACOSTI) will also issue a research permit. The Technical University Ethical Review Committee will also sort the ethical clearance certificate. A further research authorization will be requested from the Kitui County Department of Health. On the study site, permission will be obtained from the area chief and sub chief who will inform and introduce the research team to the village managers. The village managers will support the study team in identifying the households with women aged 40-65 years. The respondents will be informed and requested to sign their informed consent for administering the questionnaire and taking the scans. The respondents will be informed about confidentiality and privacy of data collected.

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APPENDICES
APPENDIX I
LETTER OF INTRODUCTION

Effectiveness of baobab-pearl millet blend on bone mineral density of women with osteoporosis aged 40-65 years in Kitui County, Kenya

Dear respondent,
My name is Immaculate Kieti, a PhD student from The Technical University of Kenya. I am undertaking a study on the "Impact of baobab fortified millet flour on bone mineral density of women suffering from osteoporosis". I am seeking your consent to participate in this study whose findings will be beneficial to you.

Please read the information relating to the study and your participation in the consent form before accepting to participate in this study. You may forward any concerns or complaints relating to the study through the contacts below.

Your participation will be highly appreciated.

Immaculate Kieti
Principal Investigator
Cell phone: 0725540181
Email: kieti8382@gmail.com

The Technical University of Kenya
Technical University of Kenya Ethics Review Committee
PO Box 43844 – 00100, Nairobi
Tel: 87120901
Email: secretary. tukerc@.tuk.ac.ke

Thank you.

APPENDIX II: INFORMED CONSENT

Effectiveness of baobab-pearl millet blend on bone mineral density of women with osteoporosis aged 40-65 years in Kitui County, Kenya

Dear respondent,

Before agreeing to participate in this research, it is important that you read and understand the information below about the study. Then you can indicate your willingness to participate in the research. Please feel free to ask any questions that you may have.

Principal Investigator: Immaculate Kieti

Purpose of the Research Study: To establish the nutritional value of baobab fruit pulp and assess its impact on bone health among women suffering from osteoporosis in Kitui County Kenya.

Procedures Description:
1. You will be given measured portions of two porridge samples prepared by the researcher and asked to rate your opinions of specific sensory attributes indicated in a form that will be provided for the purpose.
2. You will also be given a baobab fortified millet flour to prepare porridge after being shown how to prepare it which you will take every morning and evening for the six months of the study.
3. Your Bone mineral density will be measured by scan at the beginning and the end of the study.

Benefits of participation: You will benefit from the study by knowing your bone mineral density which may be improved by consuming the porridge from baobab fortified millet flour which you will be given in this study.

Risks or Discomforts of this study: There are no anticipated risks if you decide to take part in this study
.
Complaints: Concerns or complaints arising from this study during its implementation or afterwards may be forwarded to the contacts given in the letter of introduction
.
Payment: Kindly remember that there will be no payment for participating in this study.
Your responses will be taken with a lot of confidentiality and will only be used for the purpose of this study
.
Your participation is entirely voluntary, and you have the right to withdraw from this study at any time.
Thank you

Immaculate Kieti

Principal Investigator

Respondent
I have understood the purpose of this study and hereby willingly give my consent to participate.

Code----------optional Sign---------- Date----------
-

APPENDIX III

DATA SHEET FOR BONE MINERAL DENSITY LEVELS

Measurement	**1st reading**	**2nd reading**	**3rdreading**	**Mean**
Bone mineral density (SD)				

APPENDIX IV
NUTRIENT ANALYSIS DATA SHEETS

Baobab dry fruit pulp and pearl millet nutrient content analysis

Proximate analysis

Analyte	Content per 100g (dry weight)				
	Unit of analysis	Sample 1	Sample 2	Sample 3	Mean
Moisture content					
Crude protein					
Crude fibre					
Total carbohydrates					
Crude fat					
Crude ash					

Vitamin and mineral analysis

Analyte	Content per 100g (dry weight)				
	Unit of analysis	Sample 1	Sample 2	Sample 3	Mean
Vitamin C					
Calcium					
Phosphorus					
Sodium					
Magnesium					
Iron					
Zinc					
Copper					

APPENDIX V

INTERVIEW SCHEDULE QUESTIONNAIRE

Effectiveness of baobab-pearl millet blend on bone mineral density of women with osteoporosis aged 40-65 years in Kitui County, Kenya

Questionnaire No---------- Questionnaire checked----------

Name of enumerator---------- Date checked----------

Date of interview----------

Section A: Nutritional status

1. Anthropometric assessment

Measurement	**1st reading**	**2nd reading**	**3rd reading**	**average**
Weight (kg)				
Armspan/Height (m)				
BM I (kg/m^2)				

Section B: Health status

2. Have you had any fracture not related to motor vehicle accident in your life?

   1) Yes 2) No

3. If yes, what was the location of the fracture? Confirm from individual clinic files.

   1) Wrist 2) Hip 3) spine 4) Ankle

   5) Others specify_____

4. How many times have you had these fractures? _____

5. Do you suffer from backache? 1) Yes 2) No

6. Have you ever suffered from the following illnesses? 1) Diabetes 2) Arthritis

   3) Rickets

7. Are you taking any supplements related to Osteoporosis? 1) Yes 2) No

8. If yes, which one? _____

Section C: Physical activity

9. How would you rate your level of physical activity when you were below 30 yrs?

1) Very active 2) Moderately active 3) Sedentary lifestyle

10. How would you rate your level of physical activity when you were above 30 yrs?

1) Very active 2) Moderately active 3) Sedentary lifestyle

11. What physical activities were you involved in when you were below 30 yrs?

1) Walking 2) Running 3) Jogging 4) Gymnasium exercise

5) Normal household chores 6) Gardening 7) Others (specify) _____

12. What physical activities were you involved in when you were above 30 yrs?

1) Walking 2) Running 3) Jogging 4) Gymnasium exercise

5) Normal household chores 6) Gardening 7) Others (specify)

13. How many times in a week do you currently take up the physical activity?

1) Daily 2) Three times a week 3) More than three times a week 4) Less than three times a week

14. How long does the physical activity last?

1) 30 mins 2) Between 30 mins-1 hour 3) more than 1 hour 4) less than 30 mins

15. What is the intensity of your physical activity?

1) Vigorous 2) moderate 3) slow

APPENDIX VI

SENSORY CHARACTERISTICS EVALUATION

Effectiveness of baobab-pearl millet blend on bone mineral density of women with osteoporosis aged 40-65 years in Kitui County, Kenya

Date ---------- Time ---------- place ----------

Product code ---------- Code no. of respondent----------

Numerical score	Preference rating	Attributed score			
		Aroma	Taste	Consistency	Overall acceptability
1	Like extremely				
2	Like much				
3	Like moderately				
4	Like slightly				
5	Neither like nor dislike				
6	Dislike slightly				
7	Dislike moderately				
8	Dislike much				
9	Dislike extremely				

NB: kindly tick as honestly as possible for each attribute

APPENDIX VII

RECRUITMENT FORM FOR COLLECTION OF BAOBAB FORTIFIED PEARL MILLET FLOUR

Form No----------

Name of the respondent----------

Height (kg) ----------

Weight (m) ----------

BMI (kg/m2) ----------

APPENDIX VIII

QUESTIONNAIRE FOR INTERVENTION PERIOD

Effectiveness of baobab-pearl millet blend on bone mineral density of women with osteoporosis aged 40-65 years in Kitui County, Kenya

Questionnaire No---------- Questionnaire checked----------

Name of enumerator---------- Date checked----------

Date of interview----------

Section A: Nutritional status

1. Anthropometric assessment

Measurement	**1st reading**	**2nd reading**	**3rd reading**	**average**
Weight (kg)				
Arm span/Height (m)				
BM I (kg/m^2)				

Section B: Health status

2. Have you had any fracture not related to motor vehicle accident after beginning this study?

1) Yes 2) No

3. If yes, what is the location of the fracture?

1) Wrist 2) Hip 3) spine 4) Ankle

5) Others specify_____

4. How many times have you had these fractures? _____

Section C: Physical activity

5. Has your physical activity improved after starting this study? 1) Yes 2) No

6. If yes, how would you rate your level of physical activity improvement after starting this study? 1) Has improved very much 2) Has improved much 3) Has improved slightly

APPENDIX IX

IMPACT EVALUATION TOOL

Effectiveness of baobab pearl millet blend on bone mineral density of women with osteoporosis aged 40-65 years in Kitui County, Kenya

Variable	Indicator	Remarks
Bone mineral density	% with} 2.5 SD	
Acceptability of baobab	% with increased consumption	
Nutritional status	% with}18.5 kg/m^2	
Morbidity patterns	% with decreased no. of fractures	
Physical activity	% with increased mobility	
Overall intervention acceptability	% with sustained baobab consumption and ownership	

APPENDIX X

WORK PLAN

Activity	Description	Duration
Registration		1 month
Concept paper	Concept development and presentation	1 month
Proposal preparation	Proposal writing	2 months
	Proposal defense	1 month
Acquisition of samples	Purchase of millet	1 month
	Purchase of baobab fruit	1 month
Laboratory analysis	Analysis of nutrient content of baobab	1 month
Preparation of intervention flours	Formulation of baobab fortified millet flour	1 month
Research permits	Obtaining relevant permits	2 months
Field testing	Sensory acceptability evaluation	1 month
Interventional trials	Implementation of intervention	6 months
Data analysis	Data entry	1 month
	Data analysis	1 month
Findings presentation	Submission and review of findings	1 month
	Presentation of findings	1 month
Thesis preparation	Thesis writing	1 month
	Thesis notice of submission	1 month
	Submission	1 month
Thesis defense	Thesis defense	1 month
Thesis correction and submission	Revision and submission of corrected thesis	3 months
	Submission of final thesis	1 month

APPENDIX XI: RESEARCH BUDGET

Item	Description	Amount required	Unit cost	Total cost
Bone mineral density test	Dual x-ray absorptiometry	186 pple	7,100	1,320,600
Laboratory analysis	Nutrient content analysis of baobab dry fruit pulp and millet			211,500
Millet	Clean millet grains to be fortified with baobab pulp	540 kgs	100	54,000
Baobab fruit pulp	Dry baobab fruit pulp to fortify the millet flour	240 kgs	100	24,000
Processing of millet	Milling of sprouted millet grains for flour	540 kgs	10	5,400
Processing baobab fruit pulp	Separation of the pulp from the baobab seeds	240 kgs	30	7,200
Packaging materials	Materials for Packing the flour	1200	30	36,000
Sealing machine	Will help in sealing the packed flour	1	5,000	5,000
Demonstration utensils	Porridge cooking and serving utensils	1 set	5,000	5,000
Consumption utensils	Cups and jars	93 sets	300	27, 900
Acceptability testing	Sensory evaluation of porridge	4 pple	2000	8,000
Transport	Fare for collection of baobab pulp			15,000
Research permits	Approvals to conduct research	3	1,000	3,000
Questionnaires	Printing and photocopying	186 sets	30	5,580
Writing materials	Pens, notebooks and rulers	10 sets	100	1,000
Mobile phone credit	For communication during the study	10 pple	3000	30,000
Training	Training of research Team	10 pple	2,000	20,000
Data collection	Hiring research assistants	40 days	8,000	320,000
Data analysis	Data entry and analysis	1	50,000	50,000
Thesis preparation	Typing, printing and binding	1	20,000	20,000
Conferencing	Conference attendance fee	3	30,000	90,000
Publication	Publication of completed work	3	10,000	30,000
Study feedback	One day workshop with respondents and key stakeholders	150pple	500	75,000
Subtotal				2,359,180
Contingencies				20,000
Total				2,379,180