

**FUNCTIONAL AND NUTRITIONAL CHARACTERISTICS OF BAMBARA
GROUNDNUT MILK POWDER AS AN INGREDIENT IN YOGHURT**

by

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ABSTRACT

The aim of this study was to evaluate Bambara groundnut (BGN) milk subjected to spray drying with a view to establish functional, nutritional and physical properties as an ingredient in BGN yoghurt production. BGN milk powder (BGNMP) was successfully produced employing the spray drying technology. Maltodextrin was used as the drying carrier to elevate total solids of BGNM prior to spray drying. There were three levels of maltodextrin (5, 10 and 15%) employed and 10% was ideal. The optimum spray drying parameters were estimated to be the following; inlet temperature (150°C), outlet temperature (74°C), air pressure (3 bars), flow rate (10% or 16mL/min), and air flow (42.9 m³/h). The functional properties evaluated revealed high water solubility capabilities, making BGNMP readily soluble in water, which is one of the most crucial aspects of milk powders. The water solubility index of BGNMP at all maltodextrin levels ranged from 85.15 to 90.25%. There was a significant ($p < 0.05$) difference amongst BGNMP (5, 10, and 15%) in colour parameters (lightness, yellowness, redness, chroma and hue angle). BGNMP indicated to have a red and yellow colour, but yellow was more dominant. The particle size and particle size distribution of BGNMP ranged from 86.13 to 162.35 μm and 84.04 to 157.0 μm , respectively and did not differ significantly ($p > 0.05$). The water activity (a_w) of BGNMP (5, 10 and 15%) ranged from 0.45 to 0.57 and differed significantly ($p < 0.05$). All a_w levels were below 0.6, which is optimum a_w that allows rapid microbial spoilage. BGNMP had a relatively high protein content of 7.6%, considering the facts that it was not fortified with any protein rich food powder. BGNMP had a low fat content of 1.6% and total dietary fibre content of 3.4%, making it a source of fibre. Furthermore, Bambara groundnut cultured drink (BGNCD) was analysed for physicochemical, rheological and microbiological properties. BGNCD had a low fat content of 0.3%, making it an alternative source of low fat diet food. BGNCD had total dietary fibre content of 3.6% and protein content of 1.2%. There no detection of pathogenic bacteria (*Staphylococcus aureus* and *Escherichia coli*) in BGNCD. Viscoelastic properties of BGNCD revealed it to be stable, with elasticity positioned above the viscosity ($G' > G''$), which implies that the structure of the system is stable and confirms presence of gel like structure. Additionally a 2² factorial design consisting of methylcellulose and gum arabic at varied concentrations was used to estimate optimal parameters for the production of foam-mat dried BGN yoghurt. Optimal formulation parameters to produce most water soluble BGN powdered yoghurt (BGNPY) were estimated to be 50°C/24 h using mixture of gum arabic (6%) and methylcellulose (0.5%), with desirability of 0.956. BGNPY was further evaluated for functional, physical and thermal properties. BGNPY was produced from fresh BGNM and reconstituted BGNMP. The water solubility index (WSI) and water absorption index (WAI) ranged from 71.22 to 73.30% and 1.27 to 1.31 g/g, respectively. There was no significant ($p > 0.05$) difference in WSI and WAI of BGNPY produced from fresh BGNM and BGNPY from reconstituted BGNMP. Sensory evaluation of BGN yoghurt produced from

fresh BGNM and reconstituted BGNMP was investigated. The two BGN yoghurts differed significantly ($p < 0.05$) in appearance, texture and overall consumer acceptability, but did not differ significantly ($p > 0.05$) in colour, taste and aroma.

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DEDICATION

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APPENDICES

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Language and style used in the thesis are in accordance with the requirements of the International Journal of Food Science and Technology. The thesis represents a compilation of manuscript where each chapter is an individual entity and some repetitions between chapters have, therefore, been unavoidable.

GLOSSARY

Terms/Acronyms/Abbreviations	Definition/Explanation
ANOVA	Analysis of variance
BD	Bulk density
BGN	Bambara groundnut
BGNCD	Bambara groundnut cultured drink
BGNM	Bambara groundnut milk
BGNM-R	Bambara groundnut milk-from reconstituted BGN milk
BGNM-NR	Bambara groundnut milk-from fresh BGN milk
BGNMP	Bambara groundnut milk powder
BGNPY-RM	Bambara groundnut powdered yoghurt-reconstituted milk
BGNPY-NRM	Bambara groundnut powdered yoghurt-non reconstituted milk
BNF	British Nutrition Foundation
CAS	Codex Alimentarius Standards
CCI	Centre for Clinical Interventions
DIAP	Dairy Ingredients Application Programme
EAS	Eat African Standards
FA	Fat absorption
FAO	Food and Agriculture Organisation
FC	Foaming capacity
FS	Foaming stability
KEBS	Kenya Bureau of Standards
MANOVA	Multivariate analysis of variance
SEM	Scanning electron microscope
SMP	Skim milk powder
TDF	Total dietary fibre
TTA	Titrateable acidity
WAI	Water absorption index
WMP	Whole milk powder
WSI	Water solubility index

CHAPTER ONE

MOTIVATION AND DESIGN OF THE STUDY

1.1 Introduction

Bambara groundnut (BGN) is a legume extensively grown in Africa and has various names, each language and dialect has its own variation. However in literature the name Bambara groundnut is commonly used (Swanevelder, 1998). The legume consists of two botanical varieties namely, *Vigna subterranea* var. *spontanea* which includes the wild varieties and *V. subterranea* var. *subterranean* which includes the cultivated varieties. BGN belongs to the family *Fabaceae* and is rated the third most important legume in semi-arid Africa (Oyeleke *et al.*, 2012). The legume is widely cultivated in West and Central Africa, but due to the migration of indigenous or native peoples, it can now be found as far south as KwaZulu-Natal, Northern Province of South Africa and Swaziland (Mazahib *et al.*, 2013; Swanevelder *et al.*, 1998). The legume requires little demand on the soil, as it is drought tolerant, adaptable to very hot temperatures, but yet also tolerant to rainfall (Brough *et al.*, 1993).

The bean is highly nutritious, consisting of 15.5 – 22.46% protein, 55.0 – 61.3% carbohydrate, 5.8 – 7.9% fat, 2.5 – 4.5% fibre, 3.1 – 4.2% ash, 0.097% calcium, 0.007% iron, 1.25 potassium and 0.003% sodium (Sirivongpaisal, 2007; Yusuf *et al.*, 2008; Oyeleke *et al.*, 2012)., when the seed is dehulled and raw. BGN is essentially grown for human consumption and makes a complete food, as it contains sufficient quantities of protein, carbohydrates and fat. A study of the flavour and composition of BGN milk (BGNM) in comparison to other plant based milk prepared from other legumes such as cowpea, pigeon pea and soybean has been previously investigated. The acceptability of BGNM was ranked first compared to other plant milks; also the lighter colour of BGNM was preferred (Heller & Mushonga, 1997). Milk prepared from BGN gave a preferred flavour to that of milks from cowpea, pigeon pea and soybean (Atiku *et al.*, 2004). Hence BGN can be an excellent plant milk source

In most African countries staple foods consumed include grains, roots, vegetables and very little milk, which is insufficient for good nutrition (Ikemefuna *et al.*, 1991). Milk has been recognized from the earliest times as the most essential food for infants and growing children (Ikemefuna *et al.*, 1991). Milk contains proteins, fats, carbohydrates as well as vitamins and trace minerals essential for daily nutrition. It is mainly consumed in fresh form and milk for human consumption is mainly sourced from cows (Walstra *et al.*, 2005). Therefore, developing imitation milk extracted from legumes is an alternative way of producing an acceptable nutritious food which is plant based. Such milk can be used to replace animal milk. Considering the richness of milk from legumes it can be used either for babies not given milk either for ethical reasons, as with vegans or for medical reasons such as milk allergies (Ikemefuna *et al.*, 1991)

Yoghurt is a nutritious product, which is obtained by fermenting animal milk using yoghurt micro flora (*Streptococcus thermophilus* and *Lactobacillus bulgaricus*) (Dave & Shah, 1998). The product is highly acceptable to consumers because of its aroma and its flavour attributed to acetaldehyde, which is a volatile organic aroma compound produced by *S. thermophilus* and *L. bulgaricus* (Beshkova *et al.*, 1998). Although an abundant list of volatile compounds has been identified in yoghurt, only a few of them prove to have crucial influence on the resulting aroma. Good quality yoghurt should be thick and smooth showing no signs of syneresis or broken texture. Hence, appearance and physical characteristics are important parameters of yoghurt). Yoghurt shelf life, which is the period of time the product may be stored without becoming unsuitable for use or consumption, is highly dependent on the use of correct processing parameters. Plant based yoghurt can be produced; Murevanhema & Jideani (2012) reported the possibility of producing a Bambara groundnut probiotic beverage with 28 days shelf life. However, its shelf life can further be extended by spray drying.

Spray drying is a commonly used technique in the food industry, more precisely in the dairy sector. It is a process of decreasing the water content, which provides advantages by minimizing microbial deterioration of the spray dried product. It reduces lipid deterioration in fat containing products, and ideally serves as a preservation method (Gaiani *et al.*, 2010). It ensures reduction of storage and transport cost and an easier handling of the material. Spray drying of milk concentrates involves a fast removal of water leading to the formation of a dry matrix containing proteins, lactose, lipids and minerals (Kim *et al.*, 2009). Another drying processing technology which can be employed and is relatively cheap is the foam-mat drying method.

Foam-mat drying is a simple process of drying liquid or solid foods by being mixed with stabilising agent and or foaming agent to produce stable foam, which undergoes air drying temperatures ranging from 50-80°C (Kandasamy *et al.*, 2012; Febrianto *et al.*, 2012). The drying process can be used to dry juice, milk, fruits, beverages, jams (Widyastuti & Srianta, 2011). The foam dried product is then further ground to produce a powdered product. Foam-mat drying is the simplest forms of drying compared to other methods such as freeze drying, spray drying, as it is less expensive, less complicated and is less time consuming (Febrianto *et al.*, 2012).

1.2 Statement of Research Problem

The utilization of BGN as a crop throughout Africa is not extensive. The most common use of BGN throughout African countries is its consumption as roasted or boiled and further used in production of bread, cakes and porridge. Plant milk extracted from BGN has been produced and reported and the physical attributes it poses were reported to be acceptable compared to already existing plant based milks (Brough *et al.*, 1993). Murevanhema &

Jieani (2014) patented a process for obtaining BGNM from BGN flour as well as for a probiotic beverage from BGNM. However nothing has been reported on BGNM powder as an ingredient in BGN yoghurt and powdered yoghurt. Therefore it is necessary to evaluate the efficiency of using BGNM powder as an ingredient in BGN cultured drink (BGNCD).

1.3 Research Objectives

1.3.1 Broad objectives

The aim of this study was to evaluate the functional and nutritional characteristics of Bambara groundnut milk (BGNM) powder as an ingredient in BGN cultured drink (BGNCD).

1.3.2 Specific objectives

The specific objectives included to:

1. Produce spray dried Bambara groundnut milk powder.
2. Establish the functional and nutritional characteristics of BGNM powder.
3. Evaluate the physicochemical, rheological and microbiological characteristics of BGN yoghurt which is referred to as BGN cultured drink (BGNCD).
4. Optimise the production of powdered BGNCD employing the foam-mat drying process.
5. Evaluate the functional, nutritional, thermal and microbiological properties of foam-mat dried BGNCD obtained from BGNMP as an ingredient.

1.4 Hypotheses

The following hypotheses were tested in the study:

1. BGNM powder will be produced and the functional and nutritional characteristics will be suitable for production of BGNCD.
2. The nutritional and functional characteristics of powdered BGN cultured drink will be suitable for consumer acceptability.

1.5 Delineation

1. The seeds were used as purchased and were not sorted into different varieties.

1.6 Significance of the Research

BGN is underutilised, also regarded as a poor man's crop grown by small scale woman farmers. It has been stated that the crop is of little economic importance. However, recently the crop has been proven to be promising in improving food security in the country through research, resulting in product (milk, yoghurt, insoluble and soluble dietary fibre) developments from this crop.

The successful development of powdered BGNM and BGNCD will promote the use of this underutilized legume. This will result in the social development of the country, by creating job opportunities for woman farmers and improve lives of individuals and providing gender empowerment. The success of this study will also provide food nutrition, security and sustainability, as BGN is highly nutritious, widely and readily available throughout Africa regions. In addition dry yoghurt is less susceptible to spoilage, caused by the growth of undesirable bacteria and moulds, mainly stimulated by high moisture content (Kumah & Mishra, 2004). Powdered BGNCD will allow storage at room temperatures with an extended shelf life. This will provide convenience for individuals that have no electricity or refrigerators in the poor rural areas. Furthermore dried yoghurt will be suitable for pilots, submarine workers, independent travellers, astronauts, geologists and forestry workers.

The more the utilization of BGN expands, the greater the production (harvesting) will be. This may encourage exporting of the material improving the country's economy as it will open up new and improved investments opportunities and new markets. Additionally powdered yoghurt requires less packaging, handling and transportation and less storage. This is due to reduction in bulk size compared to the liquid or semi-solid yoghurts, therefore reducing company costs. Successful completion of this study will result in a completed Master's degree, which increases the post graduate output of the Cape Peninsula University of Technology Institute and provides education about this crop and its potential to individuals.

1.7 Expected Outcomes, Results and Contributions of the Research

The following were expected from this study:

- i. Production of Bambara groundnut milk powder.
- ii. New knowledge on the functional and nutritional properties of BGNMP.
- iii. Successfully highlighting the potential of BGN as an important food crop.
- iv. Submission of at least one research article for publication in an accredited journal; thereby contributing new knowledge to the field of food science and technology.
- v. Presenting the research findings at minimum, one local and one international conference.

1.8 Thesis Overview

This thesis was compiled in an article format and consists of six chapters. Chapter one introduces the entire research overview titled motivation and design of the study, which includes research problem, objectives, significance of the research and expected outcomes, results and contributions of the research. Chapter two is the literature review, which elaborated on the background of the research title, including various topics such as background of Bambara groundnut. Bambara groundnut (BGN) is an underutilized legume mainly found in African countries. BGN is known to be a highly nutritious legume, with

notable functional properties, which makes it a good food plant source. Chapter three is the first research chapter focused on the functional and nutritional properties of Bambara groundnut milk powder (BGNMP). In this chapter BGNMP was produced using the spray drying technology from BGN flour rehydrated with water to produce BGN milk (BGNM). The following was tested on BGNMP; functional attributes (wettability, water absorption index, water solubility index, fat absorption, emulsification capacity, foaming capacity, foaming stability), physical attributes (particle size and particle size distribution, colour characteristics, bulk density) and chemical attributes (proximate composition, glass transition temperature). Chapter four focused on the physicochemical, microbiological and rheological attributes of liquid BGN cultured drink (BGNCD), prior to dehydration. Liquid BGNCD was tested for physical attributes (syneresis, colour characteristics and water activity); chemical attributes (pH, titratable acidity, and proximate composition), rheological attributes (time dependent, time independent viscosity and oscillatory viscoelastic properties), microbiological attributes (enumeration of yeast and moulds, total coliforms, total viable counts, detection of *Escherichia coli* and detection of *Staphylococcus aureus*). This chapter further focused on optimising the production of powdered BGNCD using foam-mat drying process.

Liquid BGNCD was dehydrated using gum arabic and methylcellulose as foaming agents to obtain the optimum functional characteristics (water solubility index, water absorption index and wettability) of BGNCD. Chapter five covers the final research chapter which focused on the functional, nutritional, thermal and microbial properties of the optimum powdered BGNCD produced from fresh BGNM and reconstituted BGNMP using the foam-mat drying technology. They are referred to as Bambara groundnut powdered yoghurt-reconstituted (BGNPY-RM) and Bambara groundnut powdered yoghurt non-reconstituted (BGNPY-NRM). Whereby BGNPY-RM implies powdered yoghurt produced from reconstituted BGN milk powder and BGNPY-NRM is powdered yoghurt produced from BGN milk. In this chapter (BGNPY-RM) and (BGNPY-NRM) were analysed for functional rehydration attributes (wettability, water absorption index and water solubility index), physical attributes (bulk density, particle size and particle size distribution and colour characteristics), chemical attributes (proximate composition, thermal properties (glass transition temperature) and microbial properties (enumeration of pathogenic bacteria, spoilage yeast and moulds and lactic acid bacteria). Chapter six is a summary of the entire research focusing on the findings and conclusion of the study.

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CHAPTER TWO

LITERATURE REVIEW

2.1 Definition of Bambara Groundnut, Sources and Types

Bambara groundnut (BGN) is an indigenous African legume. It is a legume that is cultivated throughout Africa, from Senegal across Kenya and from the Sahara to South Africa (Brough *et al.*, 1993). BGN is a plant that belongs to the family Leguminose, with the subfamily papilionadeae. The crop was initially established in the 17th century in literature, where it was referred to as Mandubia Angola. It was then later in 1763 described by the Cinnaeusin Species Plantarum as *Glycine subterranea*. The crop was found in Madagascar in 1806 under vernacular name voanjo, subsequently written as voandzou in French. The name *Voandzeia subterranea* (L) was proposed, which was widely used by researchers over a century.

In 1978 botanical studies were undertaken by Marechal, who established similarities between BGN and plant species of the genus *Vigna*, this confirmed studies done by Vendcourt who took the opportunity to propose the current name *Vigna subterranea* (L).

The crop can be grown on poor soils in hot climates, generally in regions where cultivation of other legumes such as groundnut (*Arachis hypogaea* L.) is too risky because of the threat of drought (Brough *et al.*, 1993). This legume in most regions is the third most important legume after groundnut and cowpea. BGN is a legume that plays an important socio economic role in the semi-arid regions of Africa. It is a reach source of protein and along with the other local source of protein; it could help alleviate the nutritional problems in these areas (Massawe *et al.*, 2005).

There are several types of Bambara groundnut varieties namely; black seed, which matures earlier than other varieties and is usually a small to medium size kernel and mainly one-seeded. Red coloured seed, which matures much late, and consists of large kernels and known for its good yield. Cream/black eye, consists of a large kernel and is also known as a good yielder. Cream/brown eye, consists of a moderate kernel and is also a good yielder. Cream/no eye, consist of very small pods and kernels, produces mainly one seed and its yields are lower. Speckled /flecked/spotted, has a predominant purple colour, which contains small kernels, with mainly one-seeded pods. Brown consists of kernels of medium to large size and continuously varies between light and dark brown (Department of Agriculture, Forestry & Fisheries, 2011). Figure 2.1 clearly depicts the seven types of BGN seeds from different landraces.



Figure 2.1 Seven different colour types of BGN seeds (Kone *et al.*, 2007).

2.2 Importance, Development and Growth of Bambara Groundnut

BGN remains one of the most important legume crops in Africa (Massawe *et al.*, 2005). The crop is grown by subsistence farmers in unfavourable and stressful environments without fertilizers, irrigation or pest and disease control. It is the third most important leguminous crop in the South of Sahara, after cowpeas and groundnut (Ikemefuna *et al.*, 1991; Oyeleke *et al.*, 2012; Aremu *et al.*, 2006). Swanevelder (1998) reported that BGN is the second most important food legume and the third food crop after maize and groundnut. The seed makes a complete food as it contains sufficient quantities of protein, carbohydrates and lipid (Sirivongpaisal, 2008).

Growth and development of BGN is favourable compared to other crops. The seed is suitable for soil where other leguminous crops cannot grow, because it makes little demand on the soil and is drought tolerant (Oyeleke *et al.*, 2012). The crop is grown under traditional low input agricultural systems (Massawe *et al.*, 2005). As this crop is widely considered to be drought tolerant and is reported to outperform other crops in dry environments, there is however limited evidence on how its growth, development and yield are affected by drought (Collinson *et al.*, 1996). Therefore Mwale *et al.* (2007) reported the effect of moisture stress on the yield of BGN crop. Growth and yield of three landraces of BGN seeds under conditions of irrigation and drought has been studied. Their growth and development is adversely influenced by drought as compared to irrigated treatment, even though drought treatment gives reasonable yields. However, different types of landraces of the BGN crop also play a significant role, due to the different growth rate each landrace possesses. Once the BGN seed is fully developed, it is smooth on the surface, covered with a coloured shell, which varies from cream, brown, red or mottled (Figure 2.2).

2.3 Uses of Bambara Groundnut

Legumes have previously been part of inexpensive meals throughout the world, as they contribute a major role in the struggle against malnutrition. Therefore it is necessary that their levels of consumption which is already low be increased (Iqbal *et al.*, 2006). Plant proteins provide nearly 65% of the world supply of proteins for humans, 45-50% cereals and 10 -15% legumes. Legumes serve as sources of non-processed proteins for rural and urban dwellers of the population especially in the poor countries of the world (Bamshaiye *et al.*, 2011). Consequently, BGN seed can be utilized to modify and improve the lack of plant nutrients in the poorer areas, as it originates from a nutritious food plant.

BGN is mainly used for human consumption. Table 2.1 is an illustration of some of the food uses of BGN in Africa. The seeds are consumed either immature or full ripe and dry.



Figure 2.2 Illustration of BGN seeds smooth surface in its hard and fully matured state (Alercia, 2013).

Table 2.1

Food uses of Bambara groundnut in Africa

Country	Area	Name of BGN	Preparation of the food	Type of food	
Botswana	Ditloo		Boiled, salted and consumed as a snack	Snack	
			Pounded into flour to make cake.	Cake	
			Pounded into flour and mixed with cereals to make porridge.	Porridge	
Ghana	Azi nogui		Boiled, salted and consumed as a snack	Snack	
			Boiled, crushed into balls and fried into stew.		
			Soaked overnight, boiled until soft into porridge.		
			Canned in tomato sauce with pieces of meat in brine.		
Kenya		'Njugumawe' (Swahili)	Soaked overnight, cooked with maize, mashed and fried into stew.	Pudding	
Nigeria		Okpa	Boiled and eaten as a snack.	Snack	
Zambia		'Ntoyo', 'katoy', '[mbwiila'	Pounded into flour to make bread.	Bread	
Zimbabwe		'Nyimo' (Shona)	Boiled and eaten as a soft porridge.	Porridge	
South Africa		'Njugo beans' (Sesotho)	Boiled with peanut or maize millet to make a stiff dough.	Pap	
			'Ditloo' (Setswana)	Boiled and stirred to make a thin porridge.	Soup/Porridge
			'Tshipupu' (Venda)		

In Botswana immature seeds are boiled, salted and consumed, either on its own or mixed with maize. However at maturity, the seeds are hard and therefore require boiling prior to any specific preparation to be carried out on it (Omoikhoje, 2008).

Dried ripe BGN seeds can be pounded into flour and be used to make cakes or mixed with cereals to prepare porridge (Oyeleke *et al.*, 2012). In Kenya the seed is cooked with maize prior to soaking overnight, then mashed or fried and made into a stew (Heller *et al.*, 1997). The seed is also soaked overnight, and then boiled until soft to produce porridge in Ghana. Other parts of the country such as Zambia BGN flour is used to make bread (Linnemaan, 1990; Brough *et al.*, 1993) and in Nigeria it is used in preparing soup, porridge and various fried or steamed foods such as 'moi-moi' and 'okpa' (Linnemaan, 1988).

According to previous literature, protein functionality test on the ground seed shows that BGN can compete with or replace other conventional flours in a range of processed products (Massawe *et al.*, 2005). BGN can in addition be used to produce vegetable milk that is comparable with soy milk (Massawe *et al.*, 2005; Murevanhema & Jideani, 2012). BGN has also been used as animal feed in Northern Nigeria, the haulm is used for livestock and have been successfully used to feed chicks. These seeds are suitable for animal grazing because they are rich in nitrogen and phosphorus (Bamshaiye *et al.*, 2011).

2.4 Functional Benefits of Bambara Groundnut

The functional properties of raw BGN flour have been reported. Yusuf *et al.* (2008) reported BGN to have a high water absorption capacity (WAC), of 174% compared to benniseed flour (100%), which may be due to the higher polar amino acid residues, having an affinity for water molecules. The oil absorption capacity (OAC) of BGN flour (150%) was also reported to be higher than that of other compared flour, which suggests that BGN flour may have more hydrophobic proteins than benniseed flour. The foaming capacity (FC) is reported to be 72% which is lower than the other compared flours; benniseed flour (82%), sunflower (230%) and soy (170%). However, BGN foam is reported to be more stable (Yusuf *et al.*, 2008).

Roasting is a common local processing method in Africa and has been reported to affect the properties of legume seeds, including decreasing FC and foaming stability (FS) (Yusuf *et al.*, 2008). Yusuf *et al.* (2008) reported that drying wetted cowpea at a temperature of 80 -120°C, decreased the flour functional properties, such as solubility, foam formation and stability. However, it has been reported that in order to remove the beany taste associated with the legume milk; it was necessary and beneficial to heat treat the beans (Brough *et al.*, 1993). Therefore, to avoid maximal decrease in the functional properties of the BGNM powder and BGN powdered yoghurt, excessive heat treatment should be avoided or closely monitored.

The foaming capacity and foaming stability makes the seed a useful foam enhancer in food systems and can be used as an aerating agent in food material that requires the production of stable foam (Oyeleke *et al.*, 2012). Lawal & Adebowale. (2004) further reported the improvement of BGN functional properties using acylation and succination process.

The authors added acetic anhydride (acylation) and succinic anhydride to a BGN protein concentrate. Both processes improved the emulsifying activity and stability of the BGN native protein concentration. Thus BGN has the potential to provide favourable powdered milk as well as powdered yoghurt with the desirable functional properties. Alakali *et al.* (2010) reported the quality of composite BGN flour beef patties. The authors reported meat extended with BGN flour improved the cooking characteristics of the beef patties such as a higher cooking yield, fat and moisture retention and a lower shrinkage of the patties during cooking, which is an indication of BGN potential as functional ingredient.

2.5 Nutritional Benefits of Bambara Groundnut

Legumes have been an important source of protein (Schuster-Gajzago, 2004). They enhance the protein content of cereal based diets and may improve the nutritional status of cereal based diets (Iqbal *et al.*, 2006). Cereal proteins are deficient in certain essential amino acids, particularly lysine (Shad *et al.*, 2009). However, legumes have been reported to contain enough amounts of lysine, but deficient in S-containing amino acids (methionine, cysteine and cysteine) (Farzana & Khalil, 1999). BGN, however has been reported to contain high amounts of lysine and methionine, compared to any other grain legumes (Bamishaiye *et al.*, 2011).

BGN is reported to contain various amounts of nutrients (Table 2.2); 16. 4-22.5% protein, 53.1-61.3% carbohydrate, 5.8-7.9% fat, 3.3-4.5% fibre, 3.1-4.4% ash, 0.097% calcium, 0.007% iron, 1.25% potassium and 0.003% sodium (Sirivongpaisal, 2008; Yusuf *et al.*, 2008; Oyeleke *et al.*, 2012). The protein of BGN is high in lysine and methionine (Okpuzor *et al.*, 2010). Nevertheless Mazahib *et al.* (2013) also reported in BGN, lysine and leucine are the predominant essential amino acids.

BGN is reported to be highly nutritious. The nutritional and functional properties of BGN have been studied and Oyeleke *et al.* (2012) stated that the protein content in the seed can be utilized in the body for repairing and building of new tissue. Aremu *et al.* (2006) studied the chemical composition of two BGN varieties collected from Nigeria and reported the crude protein to be low but nearly the same; cream coated seed and the dark red seed is reported to have 11.6% and 11.1% respectively, however they were not comparable with the 24% protein reported by Poulter (1981) and the 17.5-21.1% reported by Onimawo *et al.* (1998). The difference is due to the genotype and environmental conditions under which

Table 2.2 The chemical composition of Bambara groundnut

Nutrient	Quantity
Proximate (g/100 g)	
Carbohydrates	53.1-61.3
Crude protein	15.5-22.5
Crude fat	5.8-7.9
Crude fibre	3.3-4.5
Ash	3.10-6.3
Energy	362.04
Minerals (mg/100 g)	
Calcium	63.8-256.56
Sodium	10.6-135.3
Iron	5.9-18.51
Potassium	42.7-1702.10
Magnesium	58.0–347.15
Vitamins (mg/100 g)	
Thiamin	0.28
Riboflavin	0.12
Niacin	2.1
Ascorbic acid	1.0

Adapted from (Sirivongpaisal, 2007; Yusu *et al.*, 2008; Oyeleke *et al.*, 2012; Aremu *et al.*, 2006; Salunkhe & Kadam, 2000)

they were grown, thus indicating the nutritive composition of BGN variety may be dependent on the area of harvest.

The fat content of legumes (except for soy and lupin) is about 1 to 2%, including; cowpea (1.3%) and pigeon pea (1.5%) (Schuster-Gajzago, 2004; de Jager, 2013). The oil is composed mainly of polyunsaturated fatty acids (Schuster-Gajzago, 2004). The crude fat content of BGN is approximately 5.8% therefore, the seed can be classified as a source of good fats which can be used in the formulation of low fat diet food (Oyeleke *et al.*, 2012). Okpuzor *et al.* (2010) further reported the fatty acid of BGN is predominantly linoleic, palmitic and linolenic acids. Carbohydrates constitute a major portion of a food legume. These carbohydrates include starch, sugar, crude fibre and other minor carbohydrates such as pectin substances, arabinogalactas and oxyloglucans (Salunkhe & Kadam, 2000). Carbohydrates found in BGN are oligosaccharides namely raffinose (0.35 g/mg) and stachyose (1.57 g/mg), which are responsible for flatulence (Burkill, 1995; Oyeleke *et al.* (2012) and reducing sugars (3.61 g/mg). The chemical composition of starch in legumes is characterized by high amylopectin content (Schuster-Gajzago, 2004). Oyeleke *et al.* (2012) reported starch to be the highest component (48.12 g/mg), which is an indicator of high energy content of the BGN seed. The dietary fibre content of the seeds is a very important factor from a nutritional point of view. Dietary fibre consists of indigestible polymers which are made up of cellulose, hemicelluloses, pectin and lignin. They provide mass in natural food and are resistant to hydrolysis by enzymes in the alimentary tract (Fennema, 1996). The dietary fibre content in BGN (dehulled) is reported to be slightly lower (4.5%), when compared to other legume seeds such as whole sesame seeds. However Oyeleke *et al.* (2012) reported that the crude fibre is sufficient to aid in normal digestion of food, as high values of fibre is associated with some form of disorder in animal, such as colon cancer. Ubom (2007) further elaborates that dietary fibre aids in lowering blood pressure, serum cholesterol and protects against cardiovascular diseases, diabetes and obesity.

Minerals and vitamins constitute a minor part of a food legume. However, legumes such as soybean, winged bean and BGN are an excellent source of minerals and vitamins. The most common vitamins in these legumes are thiamine, riboflavin, ascorbic acid and niacin. The minerals include; calcium (Ca), phosphorus (P), iron (Fe), magnesium (Mg), manganese (Mn) and zinc (Zn) (Oyeleke *et al.*, 2012). Potassium (K) (1702.10 mg/100 g) is reported to be the highest mineral in BGN followed by phosphorus (738.04 mg/100 g), then magnesium (347.15 mg/100 g), calcium (256.56 mg/100 g), sodium (135.30 mg/100 g), iron (18.51 mg/100 g) and manganese (10.46 mg/100 g) (Oyeleke *et al.*, 2012). However Aremu *et al.* (2006) reported calcium (63.8 mg/100 g) and magnesium (58.0 mg/10 g) as the most abundant minerals in BGN, followed by potassium (42.7 mg/100 g), sodium (10.6 mg/100 g), iron (5.9 mg/100 g), zinc (5.3 mg/100 g) and manganese (2.3 mg/100 g). Calcium is known to be important for blood clotting muscle contraction and in certain enzymes for metabolic

process. Magnesium is known to be the activator of many enzyme systems. Water soluble vitamins such as thiamin, riboflavin, and niacin are significant for the metabolism of major nutrients like carbohydrates, fat and protein (Oyeleke *et al.*, 2012). Hence BGN can be a potential functional food and its powdered milk and yoghurt can provide good nutrition.

2.6 Anti-nutritional Factors and Processing Methods on Bambara Groundnut

Anti-nutritional factors commonly found in food legumes are polyphenols, phytic acid and trypsin inhibitors. Protease inhibitor includes trypsin and chymotrypsin inhibitors. Trypsin inhibitors are protein with molecular weights ranging from 8000-21 500 g/mol (Salunkhe & Kadam, 2000). It is reported that heat treatment reduces trypsin inhibitor activity and increases the digestibility and protein utilization of the legumes. Unheated BGN does not support growth adequately because the trypsin inhibitor contained in the seeds prevents proteolysis in the intestines and a large part of the ingested protein is excreted unused (Obasi, 1998). Soaking of BGN seeds in distilled water allows the seeds to absorb water thereby, decreases and eliminates anti-nutritional factors in legumes. However, soaking too for long has been found to reduce nutritional quality of legumes through the leaching of nutrients into the soak water (Subuola *et al.*, 2012). Soaking and cooking increases the reduction of tannin, phytic acid and polyphenols found in BGN (Mazahib *et al.*, 2013). Obasi. (1998) further reported that raw BGN seeds have high polyphenols content and is reduced dramatically by soaking, boiling and sprouting of the seeds. The loss of polyphenols during sprouting is due to the presence of polyphenols oxidase and enzymatic hydrolysis. Germination enhances desired qualities such as improved digestibility, reduced anti-nutrients like trypsin inhibitors. It improves nutritional quality of the proteins by hydrolyzing them into absorbable polypeptides and essential amino acids. Sprouting improves the availability of vitamin B and C (Subuola *et al.*, 2012).

Tannins are naturally occurring plant polyphenols that can have a large influence on the nutritive value of legumes. Tannins have a positive and negative effect on nutritive value. High concentration of tannins reduces level of consumption, protein and carbohydrate digestibility. Tannins reduce the consumption of legumes by decreasing palatability due to astringency or by negatively affecting digestion. Astringency is caused by the formation of complexes between tannins and salivatory glycoproteins, this astringency increases salivation in the mouth (Lesschaeve & Noble, 2005). Khoddami *et al.* (2013) elaborates on the positive side of phenolics by stating that they are molecules that can act as antioxidants which can prevent heart disease, reduce inflammation and lower incidences of cancer. Therefore, minimal levels of polyphenols in BGN may be acceptable.

Phytic acid is one of the typical anti nutrients in legumes. It forms complexes in plant foods with proteins (protein phytate complex) and chelates essential dietary minerals such as zinc, calcium, magnesium and iron thus decreasing their utilization, which makes them

biologically unavailable for absorption (Igbedioh *et al.*, 1994; Carnovale *et al.*, 1988). However, literature also reports that phytic acid in legumes can be removed or reduced by simple processing methods such as soaking, boiling and roasting. The study reported a decrease in the phytic acid of Bambara groundnut (*Vigna subterranea*) soaked in water and the decrease was progressive with an increase in the soaking period. The reduction of phytic acid is due to the leaching out of phytate ions into the soak water (Obasi, 1998; Igbedioh *et al.*, 1994). It is further reported that the nutritional quality of the Bambara groundnut (*Vigna subterranea*) is affected. The protein and moisture content increases, while the ash and carbohydrate content decreases (Igbedioh *et al.*, 1994). Therefore, anti-nutritional factors should be monitored as milk provides essential human nutrition.

2.7 Milk in Human Nutrition

Milk is the white liquid, which is a lacteal secretion that is obtained by the complete milking of mammalian animals. Milk is an important source of many of the nutrients essential for the proper development and maintenance of the human body (Walstra *et al.*, 2006). Milk produced by animals is mainly intended as nutrition for growing offspring as well as for adults. It is for the growth and development of muscles, bone and tissue, which is dependent on the nutritional composition of the animal milk, namely; protein, vitamin, minerals, as well as fat and milk sugar (lactose), which can be categorized as carbohydrates (Miller *et al.*, 2000). However, there is a growing demand on the shift to plant milk as an alternative to animal milk. This is due to various reasons such as individuals having allergic reactions to animal milk because of lactose intolerance and individuals that choose different life styles such as vegans. Also saturated fats in whole dairy milk has been reported to elevate cholesterol levels (Miller *et al.*, (2000) triggering the shift to plant based milk due to the healthier diet choice of humans.

Plant based “milk” is milk extracts from plant sources, obtained by grinding or mashing plant products (seeds/nuts) with water and then further filtering out the liquid. It is the alternative animal milk. The resulting liquid would then be fortified by adding vitamins or minerals along with the sugar to make the milk extract more palatable (Debiyne, 2005). With plant based milk being nutritious and capable of providing a balanced diet. Resulting in an increased need of processing methods to obtaining plant based milk.

2.8 Common Processing of Legumes to Obtaining Plant Milk

Plant milk is obtained by undergoing long processing methods that require careful monitoring. Various steps are followed to get to the final desired product. The initial step is manually sorting and cleaning of legumes to remove dirt, stones, chaff, spoilt seeds and other foreign materials. The next step is soaking of the seeds in water at different time intervals, which is also known as sprouting; soaking of the seeds may come before or after

the mild heat treatments (Odo, 2003). The seeds would then undergo a mild pre heat treatment of either blanching (soaking seeds in hot or boiling water for a few minutes) or steaming. This is followed by drying of seeds either by roasting the seeds in the oven for a few minutes or using a dryer, legumes are dried on an open frying pan. Once dried, the seeds would be milled; seeds may be wet-milled or dry-milled. A common process flow chart to obtaining plant based milk is shown in Figure 2.3.

Milling is a size reduction process of seeds into smaller particle forms. Wet milling of seeds produces a paste while dry milling results in dry flour production. When dry-milling is completed, re-hydration of the milled flour seeds will be required in order to produce slurry for the plant based milk (Odo, 2003). The resultant slurry would be sieved to obtain liquid milk. Sieving can be done using cheese-cloth or muslin, which removes the residues (Sanni *et al.*, 1999; Amakoromo *et al.*, 2012). The final step is the pasteurization process, which is the most crucial step that ensures hygiene of the final plant milk (Czukur, 2001). Similar processing may be followed.

2.8.1 Previous research on plant milk production

Production of plant milk involves the extraction of the liquid from the seeds using various methods. This section of the study will focus on the different methods to obtain plant milk used by researchers. Table 2.3 shows a short summary over view of different methods that have been employed by different authors to obtain plant based milk. Peanut milk has been prepared by a method reported by Isanga & Zhang (2009). Sorted peanuts were roasted at 130°C for 20 min in an oven. The seeds were then de-skinned and weighed before being soaked in 0.5 g/100 mL NaHCO₃ for at least 12 hours. The de-skinned peanut kernels were then washed with clean water, then mixed with water in a ratio of 1:5 w/v (peanuts: water) and transferred to a blender where they were blended for at least 5 min. The slurry formed was filtered using a double layered cheese cloth to yield the peanut milk. Aidoo *et al.* (2010) described the extraction method of peanut-cowpea milk. The peanut-cowpea ratio of 2:1 was used and a 1:2 (grains: water) ratio was used.

The mixture was wet milled. The slurry was further milled using colloid mill to obtain smooth, fine homogenized milk. The obtained slurry was sieved to obtain the milk extract. Cowpea milk was prepared by a method reported by Sanni *et al.* (1999). Cowpea seeds were cleaned to remove dry particles. The seeds were then soaked in tap water for 12 h. The steeping water was decanted and the beans were washed with tap water. Boiling water sterilized at 121°C for 15 min was used to grind the beans in a warring blender in a ratio of 5:7 w/v (beans:water). Filtration was done by making use of a double layered cheese cloth.

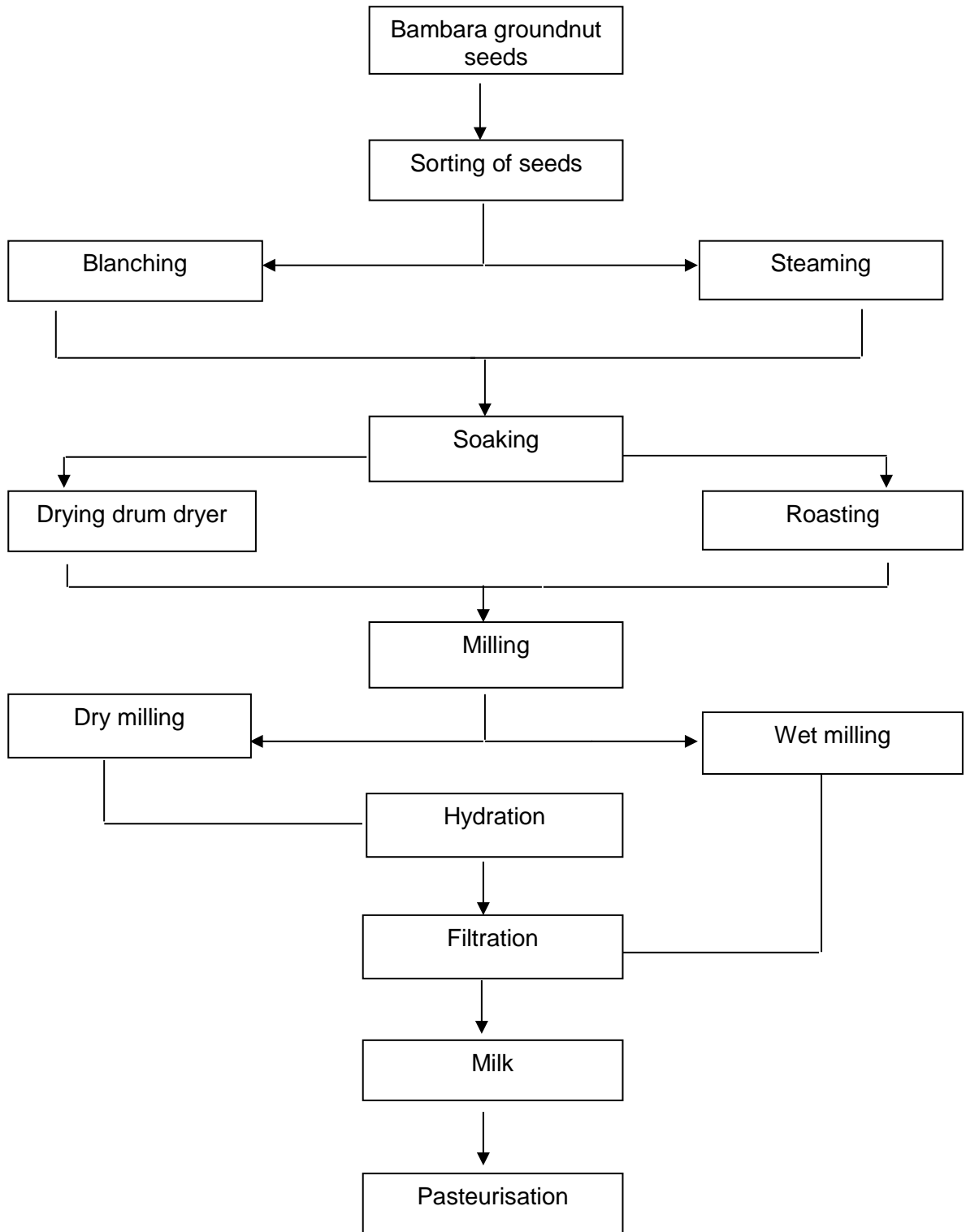


Figure 2.3 Common process flow chart to obtaining plant milk.

Table 2.3 The different methods reported for plant-based milk

Milk	Method	Authors
Peanut milk	Wet milling	Isanga & Zhang (2009)
Peanut-cowpea milk	Wet milling	Aidoo <i>et al.</i> (2010)
Cowpea milk	Wet milling	Sanni <i>et al.</i> (1999)
Soya milk	Wet milling	Salunkhe & Kadam (2000)
Bambara groundnut milk	Wet milling	Brough <i>et al.</i> (1993)
Bambara groundnut milk	Dry milling	Murevanhema & Jideani (2014)

The milk obtained was sterilized in a water bath at 70°C for 20 min. Salunkhe & Kadam (2000) describes the preparation of soya milk from non-defatted soy flour. The beans were initially and soaked in excess water for 12-24 h depending upon the temperature of the water. The water was drained and the beans were washed with tap water. The beans were preheated for 1 min at 94°C. The beans were wet milled with the addition of enough water at 94°C. The ratio of beans to water on a weight basis was approximately 1:10. The slurry was heated at near its boiling point (94°C) for 15 to 20 min to sterilize the product with continuous stirring. The slurry was then sieved to obtain the liquid milk.

BGN seeds weighing 500 g were soaked for 24 h. The soaked sample (wet weighed 830 g) was divided into two, half being used to prepare raw milk directly and the other half roasted to prepare pre-treated milk. Raw milk was prepared by homogenizing soaked beans with hot water (1:2 w/v) initially in a food processor to reduce the size, followed by high speed mixing to ensure a fine suspension of beans and water. The pre-treated milk was produced by roasting the beans prior to homogenization for 5 min or until the colour became slightly transparent. After homogenization, the milk was strained through a muslin cloth to obtain milk sample (Brough *et al.*, 1993). The patented process reported by Murevanhema & Jideani (2014) extracted BGN milk from BGN flour. A dry milling process differing from the common wet milling process was used. Dry milling does not generate waste water unlike wet milling, while wet milling also consumes water increasing the lack of water problems. In addition wet milling requires more energy, compared to dry milling therefore, making dry milling a greener choice (Liaotrakoon *et al.*, 2014).

Germinated milk from cowpea was reported by Abudu & Akinyele (1990). Cowpeas are known to contain various antinutritional factors, these factors are associated with the presence of oligosaccharides and trypsin inhibitors. Antinutritional factors may contribute to the development of flatulence and possibility of diarrhea in children consuming cowpea based product such as milk. Germination is a process known to reduce the amount of oligosaccharides in legumes. It was also reported that the trypsin inhibitor reduces the protein quality found in cowpeas (Abudu & Akinyele, 1990). Germination is reported to reduce the oligosaccharides content.

However, the process of germination also reduces the total solids, energy content, protein content as well as the mineral content of the milk product. Therefore, the process of germination prior to milk extraction from the cowpea was not followed. The advantage of the reduction of the oligosaccharides content was not more important than the reduction in the nutrient content in this study, since the concern is to meet the nutrient requirement so as to prevent malnutrition by promoting legume based foods (Abudu & Akinyele, 1990)

Milk derived from BGN has also been reported by Brough *et al.* (1993). Dry heating of BGN was found to remove the beany taste and flavour from the BGNM. The boiling of the

BGN seeds increased the gelatinization of starch, which greatly hampered the process of straining through the muslin cloth and consequently reduced the total volume of milk. However, roasting the bean was proved effective. BGNM is a promising vegetable milk, which can be produced from BGN seeds as this process has the potential to make possible production of low cost weaning foods. However caution would have to be taken in choosing type of seeds and processing methods, as there is variation in the seed colour which would have an impact on properties such as colour and preference of the consumer. Milk extracted from soya seeds is the most common process that is employed in the industries. Milk extracted from soya seeds has been reported to be easily digestible and a good source of protein. Soya vegetable milk is now an acceptable food product consumed daily by lactose intolerant individuals or those eating vegan diets. However soya products in the market are expensive, due to the fact that soya seeds are not widely grown in Africa as compared to other seeds such as cowpea, pigeon and BGN. Therefore, promoting vegetable milk from native species, especially one that is advantageous in terms of soil growth tolerance, such as BGN, seems more rational (Brough *et al.*, 1993).

2.9 Spray Drying Technology

Spray drying is a process of transforming into powdered form by spraying the feed into a hot drying medium; usually air (Xin & Mujumdar, 2009). The gas generally used is air or more rarely an inlet gas as nitrogen. Spray drying can be used in a wide range of applications where the production of a free flowing powder is required. It is used and successful in various products areas such as pharmaceuticals, beverages, flavours, colouring and plant extraction, milk and egg products. The initial feed material to be converted to powder may be in solution or suspension or paste (Early, 1998). Spray drying involves the formation of droplets from the volume of liquid followed by the removal of moisture from the liquid droplets. The spray drying process employs phases that lead to a dry final product (Filkova *et al.*, 2014)

2.9.1 Heat treatment of milk prior to spray drying

Milk to be spray dried should at least undergo minimum temperature (60-63°C) pasteurization before evaporation and further processing. However, higher temperature heat treatments are used to influence the product characteristics. Heat treatment of milk may affect the biological, physical and chemical changes of milk. This may have an effect on the nutritional and organoleptic properties (Early, 1998). Several heat treatments exist and are applied to milk processing accordingly.

The High Temperature Short Time (HTST) pasteurization has been effectively used for decades. For an effective HTST pasteurization, every particle of milk must be heated in properly operated equipment to 71.7°C (161°F) for 15 sec. HTST pasteurization of milk is

normally carried out by indirect heating systems, such as plate and tubular heat exchangers. In these processes, the milk and the heating medium (superheated steam or hot water) are separated by heat-conducting material, and heat is transferred to the product by conduction and convection. The plate heat exchangers (PHEs) are broadly used for heating and cooling applications because they offer high degrees of intensity and efficiency (Walstra *et al.*, 2006). Heat treatments are often applied in combination with the process of evaporation during spray drying and the reasons for heating milk is to achieve the required microbial specification as milk has the potential to contain some spoilage organisms as well as pathogenic bacteria. Without due diligence in the production of milk powders, the potential to produce a product containing food poisoning is significant. The predominant organisms which can be present in BGN are *Staphylococcus spp*, *Streptococcus spp* and *Enterococcus spp* (Subuola *et al.*, 2012). Hence the heat treatment of the BGN drink is critical in producing an acceptable safe product.

2.9.2 Removal of water prior to spray drying

The removal of water by spray drying is an energy requiring process; however spray drying is not the only method employed for the removal of water in milk powder production. Spray drying from low solids results in unacceptable powder properties, significantly a low bulk density and it also increases powder losses (Early, 1998). A method of removing as much water as practical prior to spray drying is required and most instances vacuum evaporation is used. In the case of skimmed milk, vacuum drying can be used to take the total solids content from about 8.7 to 50% creating a milk concentrate which can allow the production of a powder with satisfactory physical properties (Early, 1998). In the production of such a concentrate, about 90% of the water in the original milk is removed through evaporation leaving only around 9.5% to be removed by spray drying, with some 0.5% remaining in the product as bound water. Milk should be evaporated to total solid of 40-50% (Early, 1998). When concentrating milk for spray drying, the highest practical solid level is usually aimed for, though the degree to which the milk can be concentrated is limited by the viscosity of the concentrate and consequently the ability of the spray drying plant to pump and atomize it (Early, 1998). However, Murevanhema & Jideani (2012) reported the total solid of BGNM to be 2.6%, which is a slightly low total solid content as compared to other legume milks such as tigernut milk, which had total solid content of 20.2-23.2% (Ukwuru & Oghodo (2011)). Consequently, spray drying BGNM with such low solids will yield poor quality BGNM powder.

2.10 Component Phases of Spray Drying Technology

The process of spray drying involves multiple stages including atomisation of particle droplets through atomiser into hot drying air stream (drying chamber) and further separated

in the cyclone before going into final product bags. Figure 2.4 depicts a typical spray dryer system equipped with a centrifugal atomizer.

2.10.1 Atomisation phase

Atomization is the most critical step in this process. Liquid atomization in small droplets can be carried out by pressure or centrifugal energy. Numerous atomizer techniques can be employed, which includes pressure nozzle atomization, two-fluid nozzle atomization and centrifugal atomization (Kim *et al.*, 2009a). In a pressure nozzle atomization, spray is created by forcing the fluid through an orifice. It is an energy sufficient method which offers the narrowest particle size distribution. In a two-fluid nozzle atomization, spray is created by mixing the feed with compressed gas. It is useful for making extremely fine particles. In a centrifugal atomization, spray is created by passing the feed through or across a rotator disk (Kim *et al.*, 2009a).

The aim of this stage is to create a maximum heat transferring surface between the dry air and the liquid in order to produce an optimum heat and mass transfer. The atomization process produces spherical droplets. Dried particles may have the same shape and size. The distribution size and shape of droplets is affected by nature of the feed, its solid concentration and viscosity as well as the type of atomizer. Higher energy provided results in finer formed droplets. At constant amount of energy provided, particle size only increases with increasing feed rate (Gharsallaoui *et al.*, 2007) However, Kim *et al.* (2009b) reported that particle size increases when both viscosity and surface tension of the initial liquid is high.

2.10.2 Drying chamber

Within the drying chamber of a spray dryer system, droplet hot air contact occurs. This contact takes place during atomization and initiates the drying stage. In this stage there is co-current drying and counter-current drying. In the co-current process, the feed liquid is sprayed in the same direction as the flow of hot air through the system (Fleming, 1921; Filkova *et al.*, 2014). The hot air inlet temperature typically ranges from 150-220°C and evaporation occurs instantaneously, with an outlet temperature range 50-80°C. Whereas in counter-current drying, the liquid feed is sprayed in the opposite direction of the flow of hot air. This exposes the dry product to high temperatures, thus limiting the application of this process for thermo-sensitive products. However, co-current process is reported to be advantageous, because it is considered as a more economic process in terms of consumed energy (Fleming, 1921). The following paragraph will describe the drying principle of particle droplets of the liquid feed incorporating inlet, outlet temperature and air/gas.

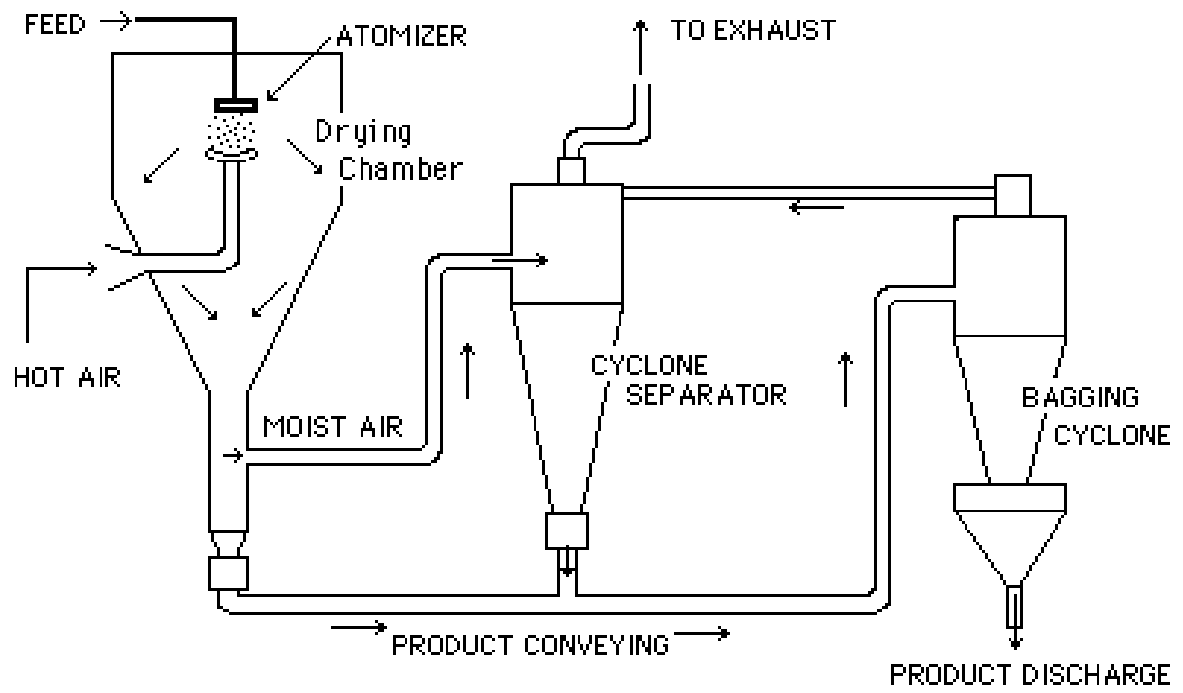


Figure 2.4 Typical spray dryer diagram (GEA, 1981)

At the time of droplets coming into contact with the hot air, balances of temperature and vapour partial pressure is established, between the liquid and gas phases. Thus heat transfer occurs from air towards the product as a result of temperature difference, whereas water transfer occurs in the opposite direction because of the vapour pressure difference (Gharsallaoui *et al.*, 2007). After the hot air comes into contact with the liquid feed product, the hot gas causes an increase of the droplet temperature, which promotes liquid evaporation from the droplet surface and promotes shrinkage of droplets. Heat transfer principally causes the increase of droplet temperature up to constant value, which explains the high inlet temperature of 150-220°C. This value is defined as the air drying humid thermometer temperature. Thereafter, the evaporation of droplet water occurs at a constant temperature and water vapour partial pressure. Finally when the product droplet water content reaches a critical value, a dry crust is formed at the droplet surface and the drying rate decreases rapidly, which explains the lower outlet temperature of 50-80°C (Adhikari *et al.*, 2005)

2.10.3 Separation

In this stage separation of powder from moist gas occurs. This should be carried out in an economic and pollutant free manner, by recycling the drying medium. Separation of fine particles is often occurs in the cyclone placed outside the dryer, to reduce product lose in the atmosphere (Filkova *et al.*, 2014). Larger particles are recovered at the base of the drying chamber, while the finest ones pass through cyclone to be separated from the humid air (Chen & Mujumdar, 2008). Spray drying is the most common and cheapest technique to produce a good quality powdered food material. Equipment is readily available and production cost is lower compared to other drying process such as freeze drying. Hence spray drying will be the best advantageous process to be employed for the drying of BGNM.

2.11 Technical and Economic Purposes of Employing Spray Drying Technology

Spray dried products improves storage stability, minimizes packaging requirements and reduces shipping weights, hence it ensures a reduction of storage, transport cost and easier handling of the material (Gaiani *et al.*, 2010; Sliwinski *et al.*, 2003). Cost of spray drying is directly linked to the use of its energy. There are various reasons the process of spray drying is mostly and commonly used in the food industry. Spray drying enables particle size control. The dry particles are easily controlled by atomization of the liquid feed and the design of the hot gas inlet. The correct spray dryer design and atomization technique enables an increase in the yields of products that require classification. Spray dryer can typically produce between 30 to 500 micron average particle sizes (Xin & Mujumdar, 2009). The shape of most spray dried particles is spherical, which provides for fluid like flow

properties. This enables downstream operations such as packaging, pressing, filtering and handling easier and less costly during production of a product in bulks (Filkova *et al.*, 2014).

Spray drying promotes production of homogenous solid mixtures and produces the most homogenous product for multi component solution and slurries. Each particle produced in the final stage will be of the composition as the initial mixed feed. Spray drying process enables desired evaporative cooling of the final product. The heat and mass transfer during drying occurs in the air and in vapour films surrounding the droplets. This vapour maintains particles undergoing drying at a saturation temperature, which is a state where the particles still contains sufficient moist and prevents it from being too dry. The temperature of the solids will not reach the dryer inlet temperature, at which it is dried at. Hence, many heat sensitive products can be sprayed dried easily at relatively high inlet temperatures (Filkova *et al.*, 2014).

Another advantage of spray drying is the short residence time required to dry a product. The surface area produced by atomization of a liquid feed enables a short gas residence time, ranging from 3-40 s, which permits spray drying without any thermal degradation experienced. Spray drying also reduces the potential of material produced by corrosion e.g. rust. This is because spray drying is a gas suspended process; the dryer chamber remains dry by design. Therefore, many corrosive materials can be processed with carbon steel a primary material of construction of the spray dryer chamber, which reduces capital cost (Filkova *et al.*, 2014). Water removal by spray drying is a common engineering practice. By decreasing water content and water activity, spray drying is generally used in food industries as a food preservation method to ensure a microbiological stable product, avoid risk of chemical or biological degradation, reduce the storage and transport cost and most importantly obtain a dry desired product (Karel & Lund, 2003; Okos *et al.*, 2006). Therefore, the spray drying technology proposed for drying of BGNM should produce the desired BGNM powder.

2.12 Foam-Mat Drying Technology

Foam-mat drying is a simple process of drying liquid-solid foods by mixing with stabilising agent and or foaming agent to produce stable foam, which undergoes air drying temperatures ranging from 50-80°C (Kandasamy *et al.*, 2012; Febrianto *et al.*, 2012). The foam-mat drying process can be used to dry juice, milk, fruits, beverages, jams (Widyastuti & Srianta, 2011). The foam dried product is then further ground to produce a powdered product. Foam-mat drying is the simplest form of drying compared to other methods such as freeze drying and spray drying, as it is less expensive, less complicated and is less time consuming (Febrianto *et al.*, 2012).

2.12.1 Foam-mat drying processing technique and foaming mechanism

Foam mat drying is a process where liquid foods are whipped into stable foams and then air dried. During the heated air drying it is required that the foams remain stable and retain typical open structure to simplify rapid drying and detraining. Should foams collapse during drying, this unwanted feature increases drying rate, reduces the quality of the product and prevents detraining (Sankat & Castaigne, 2004). Degree of drying in the foam-mat drying process is reasonably very high because of the massive increase in the liquid-gas interface, even though the heat transfer is hindered by a large volume of gas present in the foamed mass. Drying occurs in more than one constant rate periods because of the periodic bursting of successive layers of foam bubbles, thus exposing new surfaces to heat and mass transfer as the drying progresses (Kandsamy *et al.*, 2012).

However, it is recognised that pure liquids do not foam and the foaming solutions depict an obvious relationship with the surface activity of the solute. Foaming is not distinct in mixtures of liquids with the same chemical type, it is also not pronounced in aqueous solutions of highly hydrophilic solutes, glycerol or sucrose. Transient foams are caused by solutes that lower the surface tension moderately for example the short chain aliphatic alcohols and acids and persistent foams arise only with solutes which lower the surface tension in dilute solutions, for example proteins. For foaming to take place a “foreign compound” must be added to produce a stable gas-liquid dispersion. This type of compound is normally an existing or added active agent that reduces the surface tension of liquid and therefore encourages a surface layer of a composition that differs from the rest of the liquid. This layer performance as a buffer that hinders the natural coalescence of gas bubbles dispersed in a liquid (Tayeb, 1994). Figure 2.5 depicts general structure of foam system.

Drying foamed material enables a high speed of drying because of the open structure of foam. Foam-mat dried materials have desirable properties such as favourable rehydration and the retention of volatiles Jakubczyk *et al.* (2011). Krasaekoopt & Bhatia (2012) further reported that the collaboration of air bubbles into foam is important and affects the drying speed. Drying rate of the foam-mat drying process is relatively high because of the huge surface exposed to the drying air ensuring fast moisture removal. The foam stability during drying is critical, if the foam collapses, breaking of cellular structure will occur which can cause serious damage of the drying process.

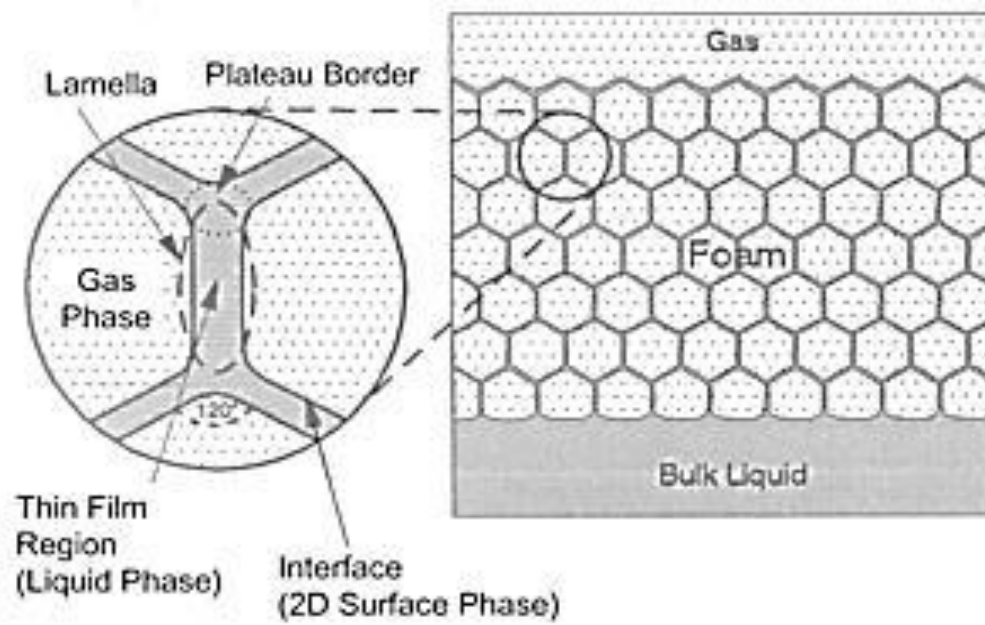


Figure 2.5 General foam system structure (Anon, 2012).

The foams become stable when high viscosity and low surface tension occurs at the air/aqueous interface. However there are a lot of factors that affect the foam characteristics and foam properties such as food composition, type and concentration of the foaming agent as well as the mixing time. Small bubbles in the foaming mass are exposed to an enormous surface for removal of moisture. The fast drying is due to the movement of moisture by capillarity in the liquid films separating foam bubbles. The foaming makes the mass undergoing drying extremely porous and submissive or compliant to drying its inner layers (Rajkumar *et al.*, 2007).

2.12.2 Economic and technical purposes for employing foam-mat drying

Foam-mat drying has the ability to process hard to dry materials to produce products of the desired and required properties, retaining volatiles that otherwise would be lost during drying of non-foamed materials (Wilson *et al.*, 2012). Foam-mat drying process produces end product with favourable rehydration, controlled density and retain volatiles that would be lost when using other forms of drying (Kudra & Ratti, 2006). Foam-mat drying process provides the advantage of lower temperature and shorter drying times compared to non-foamed material in the same type of dryer and conditions (Widyastutu & Srianta, 2011).

The foam-mat drying process is suitable for heat sensitive, viscous, sticky and high sugar content food products (Rajkumar *et al.*, 2007), which cannot be dried using other forms of drying methods such as spray drying. Foam-mat dried products contain improved reconstitution characteristics due to their open structure. More interest has developed for foam-mat drying because of its simplicity, cost effectiveness, high speed drying and improved product quality. Foam-mat drying does not comprise a large capital outlay, product is reduced into light and porous form, which when packaged allows great stability (Kandasamy *et al.*, 2012).

Foam-mat drying offers the advantage of air drying, affordability and accessibility. Foamed structures offer advantages in spreading, in drying, in surface removal, in crinkling and rehydration of the product sample. Foaming assists in the difficulty of thickness control of a product (Tayeb, 1994). The foam-mat drying technique has the advantage of using lower temperatures and also the foaming system incorporated speeds up the evaporation of water, it is low cost and easy to do. With foam-mat drying method, expected drying time can be faster, which makes it cheaper and easier when compared to other usual methods of drying (Febrianto *et al.*, 2012).

2.12.3 Additives in foam-mat drying methods

In production of papaya pulp powder, methyl cellulose was incorporated as a foaming agent at different concentrations (0.25%, 0.50%, 0.75% and 1%). The maximum stable foam formed was 83% for the 0.75% concentration (Kandasamy *et al.*, 2012). In the development of functional drink of foam-mat dried papaya, egg white was used as the foaming agent at different concentrations (10%, 15% and 20%). Egg white contains good foaming properties, which is because of the proteins found in egg white, which has the ability to encapsulate and retain air. Increasing concentration of egg white, results in rapid drying, due to the surface area that is high. The finest concentration of egg white depends on the type of fruit being dried (Widyastuti & Srianta, 2011). Sankat & Castaigne (2004) reported that during the drying of ripe banana puree, soy protein was employed as the foam inducer. However, other foaming agents were employed such as glyceryl monostearate, which did not induce any foam. Dream whip and gelatine produced foam, but the foams produced were not suitable for successive drying. Soy protein foaming agent reduced the density of the initial ripe banana product from 0.93 g/mL to 0.50 g/mL after being whipped for 12 min by adding 10 g/100 g of the soy protein (Sankat & Castaigne, 2004). Some foods naturally contain soluble proteins and monoglycerides and produce foams when whipped; however the foams produced are not adequate enough for drying process. Therefore the addition of foaming agents and stabilisers are necessary to induce foaming and give proper stability during drying. Glyceryl monostearate (GMS) and solubilised soy protein (SP) are the most often reported foaming agents. Mango pulp was foam-mat dried using egg white at varying concentrations of 0, 3, 5, 7 and 9% as a foaming agent (Wilson *et al.*, 2012).

The bulk density and reconstitution rate of citrus powder was studied using the method of foam-mat drying. For the experiment, GMS foaming agent and soya albumin foam stabiliser with Methocel (10 cps) also known as MSA/MC (Wagner *et al.*, 1964). Foam-mat drying of mango pulp using egg albumin (10%) as a foaming agent combined with 0.5% of methyl cellulose as a stabiliser gave the optimum results (Rajkumar *et al.*, 2006). The two combined together retained significantly high amounts of biochemical content. Apple puree which contained no additives was foam-mat dried using different foam-mat drying methods to study the physical properties and the aroma of the apple puree powder. Egg albumin (Fluka) with the concentration of 2% was employed as a foaming agent and 0.5% of methylcellulose (Methocel, 65HG, and Fluka) (Jakubczyk *et al.*, 2011). Febrianto *et al.* (2012) reported the drying of milk powder using foam-mat drying. In this particular method a filler material was used combined with an emulsifier. Tween 80 was used as an emulsifier at a concentration of 1% combined with the two filler materials gum arabic (2, 4, and 6%) and maltodextrin (5, 10, and 15%) (Febrianto *et al.*, 2012).

Foaming agents commonly used are enzymatically hydrolysed soya protein (0.1-5% by weight of the dry solids), glycerides or sucrose fatty acid ester, lecithin and poly glyceryl

stearate other foaming agents are Hyfoama (prepared by hydrolysis of casein), powdered egg white, Vega foam D (soy protein concentrate) and milk protein concentrate. Foaming stabilisers that are hydrophilic colloids are soluble starches, sodium carboxyl methyl cellulose, agar, methyl cellulose, gum arabic, sodium alginate, pectin, dextrin, sodium carboxyl methyl starch, sodium carboxyl methyl amylose, pentosans, albumin, gelatin and dried egg white. Monostearate is a surface active agent acting as a foam stabiliser. Mono glycerides such as glycerol mono palmitate, glycerol mono myristate, glycerol mono laurate, glycerol mono stearate and glycerol mono oleate are well known foam stabilisers in aqueous systems. There are also some well-known emulsifiers, thickeners and consistency developers. Albumin acts as an emulsifier for syrups, while guar gum is added as a thickening agent and sodium alginate is used to raise viscosity of syrups. Consistency may also be elevated by adding hydrophilic colloids such as methyl cellulose in juice, gelatin in meat puree, pectin in berry juice and guar gum in lemonade (Tayeb, 1994).

2.12.4 Drying of liquid and solid foods using foam-mat drying methods

Commercial yoghurt was foam-mat dried using two types of foaming agents; methyl cellulose; (0.5, 1.0, 1.5 and 2%) and egg albumin (1, 2, 3 and 4%). Yoghurt foam was produced by mixing the foaming agents with the yoghurt and blended for 12 min. The yoghurt foam was then poured into Teflon trays and dried in an oven at 50, 60 and 70°C for 3 h. It was observed that the appropriate conditions for producing the optimum yoghurt powder is by using 3% egg albumin and drying at 60°C for 3 h (Krasaekoopts & Bhatia, 2012). Alphonso mango pulp was foam-mat dried using egg albumin (5%, 10% and 15%) with methyl cellulose of 0.5% as foaming agents. The foamed pulps were dried at 60, 65 and 75°C (Rajkumar *et al.*, 2007). The optimum results were obtained using 10% egg albumin foaming agent with 0.5% methyl cellulose stabilising agent, dried at 60°C. Febrianto *et al.* (2012) reported drying of liquid milk using foam-mat drying process. Fresh milk was measured to a volume of 200 mL and cooled to 0°C optimum temperature for treatment.

The fresh milk was then mixed with an emulsifier Tween 80, 1% v/v of fresh milk (2 mL) in plastic and glass containers and further homogenised using a mixer speed of 1 and increased to 2 and 3, respectively. The fresh milk with the emulsifier was then added to filler materials used at different concentrations; maltodextrin (5, 10 and 15%) and gum arabic (2, 4 and 6%). The mixtures were stirred using a mixer at speed 1 and then increased to 2 and 3 alternately for 5 min. The foamed mixture was poured into stainless steel pans which were coated with plastic film. The pans containing foamed mixture were placed in the vacuum dryer and dried at temperature of 70°C for 7 h. Best milk results were obtained using 1% emulsifier with 15% maltodextrin.

2.13 Yoghurt Production and Ingredients

Yoghurt is a food material that is produced by adding two starter cultures, *Lactobacillus delbreuckii subsp. Bulgaricus* and *Streptococcus thermophilus* (Marttila-Sandholm & Saarela, 2011) into liquid milk. Milk is the main ingredient of yoghurt, while other ingredients are allowed in yoghurt to adjust the composition, such as cream to adjust the fat content and non-fat dry milk to adjust the solid content. The solid content of yoghurt is often adjusted above the 8.25% minimum to provide better body and texture to the finished yoghurt. Stabilizers are part of yoghurt ingredients, which are added to improve the body and texture by increasing firmness due to their thickening effect, preventing separation (syneresis) and helping to keep uniform mixing within the yoghurt (Mckinley, 2005). Common examples of stabilizers used in yoghurts are alginate (carrageenan), gelatins, gums (locust bean, guar), pectin and starch. Other ingredients added during the production of yoghurt are sweeteners and flavourants, which are used to provide variety to the consumer (Marttila-Sandholm & Saarela, 2001).

Lactobacillus bulgaricus and *Streptococcus thermophilus* cultures perform specific functions during the production of yoghurt. The function of these beneficial bacteria is to ferment lactose (milk sugar) to produce lactic acid bacteria. The increase in the lactic acid decreases the pH and causes the milk to form clots or form the soft gel that shows yoghurt characteristics. The fermentation of lactose also produces the flavour compounds known as acetaldehyde produced by *S. thermophilus* and *L. bulgaricus*. Other cultures such as *Lactobacillus acidophilus*, *lactobacillus subsp.* and *Bifidobacteria* may be added to yoghurt as probiotic cultures. Probiotic cultures benefit human health by improving lactose digestion, gastrointestinal function and stimulating the immune system (Tamime, 2002).

2.14 Yoghurt General Processing Procedure and Parameters

Yoghurt making can be achieved by using the following general manufacturing procedure. The composition of milk is adjusted to achieve the desired fat and solid content. It is then followed by pasteurizing the milk at temperatures of 85°C for 15 to 30 min or at 95°C for 10 min. This treatment kills vegetative bacteria and expels most of the oxygen and produces reducing substance, which help to initiate and maintain anaerobic conditions in milk suitable for growth of the inoculum (Lutchmedial *et al.*, 2004). Homogenization is the following step. The mixture is homogenized to mix all ingredients thoroughly to improve the consistency of the yoghurt. After the homogenization, the mixture is cooled to 42°C to bring the yoghurt to the ideal growth temperature for the starter culture. Next is to add the starter cultures into the cooled milk. The milk is then held at 42°C until the pH of 4.5 is reached, the holding process allows the fermentation to progress to form a soft gel with yoghurt characteristics.

The yoghurt is then cooled to 7°C by being stored under refrigeration to stop the fermentation process (Tamime & Robinson, 1999).

2.15 Plant Based Yoghurt as an Alternative

Yoghurt produced from cow's milk is consumed in both developing and industrialized countries. However there is a growing demand for alternatives to cow's milk due to the problems such as allergenicity caused in individuals who are lactose intolerant. Lactose intolerance is caused by a carbohydrate found in cow's milk known as lactose disaccharide. Some individuals suffer from lactose mal-digestion, which is a difficulty in digesting lactose. So once the carbohydrate enters the large intestines in a human body, it is fermented by colonic microflora causing adverse gastrointestinal symptoms in some individuals, such as flatulence, diarrhea and abdominal pain (British Nutrition Foundation, 2013).

Demand is also due to the desire for vegetarian as well as the reduction in the economy cost, by using plant based milk from legumes. Therefore, soy-based yoghurt is one of the cow's milk alternatives yoghurt (Marttila-Sandholm & Saarela, 2011). Soya beverage and cow's milk were produced with *S. thermophilus* and *Lactobacillus delbreuckii subsp bulgaricus* to analyse the differences in production and outcome of the final product. The yoghurts were produced with the yoghurt starter in combination with either probiotic bacteria such as *Lactobacillus johnsonii*, *Lactobacillus rhamnosus* or human derived *Bifidobacteria*. During the production of soya based yoghurt in comparison to normal milk based yoghurt, it is reported that the drop in pH was faster in the soy beverage than in milk. This suggests a greater rate of production of organic acids in the soy beverage than in cow's milk. It is also reported that probiotic bacteria and *Bifidobacteria* uses different sugars to support their growth (Marttila-Sandholm & Saarela, 2011). Since BGNM makes an acceptable plant-based product study previously reported by Murevanhema & Jideani (2014). It will be beneficial seeing that it is free of allergenicity properties. It can be consumed by all different groups such as vegetarians and vegans and thus BGNM powdered yoghurt will be an acceptable plant based yoghurt.

2.16 Plant Yoghurt Processes and Ingredients

Other legume milk based yoghurt has been successfully produced such as from peanut milk. In the previously mentioned study (Section 2.8.1) physicochemical parameters of the peanut milk yoghurt was evaluated. Peanut milk was produced by soaking raw peanuts with water to get slurry subject to filtration. Alternatively, it may also be produced by grinding unsoaked roasted peanuts/raw full fat or partially defatted peanuts to form flour, to which water may later be added to the flour to make an emulsion. Milk-like product was then homogenized and pasteurized in much the same way as fresh milk and also supplemented with vitamins, minerals and sometimes flavourants. The peanut milk for yoghurt production is described;

the peanut milk was however fortified with skimmed milk powder. The peanut milk of 12 g/100 g was mixed with 4 g/100 g of skimmed milk powder. Mixture was stirred and warmed at 43°C for 30 min (Isanga & Zhang, 2009). Sucrose is added to the mixture to impart flavour and sweetness. The blend was homogenized at 25 MPa. Homogenized milk was further pasteurized at 85°C for 30 min cooled to 43°C in a water bath, then inoculated with the starter cultures *L. bulgaricus* and *S. thermophilus* and incubated at 43°C for 4-5 hours until a pH of about 4.6 was reached. The peanut milk yoghurt was then cooled then cooled and transferred to the refrigerator. The study revealed that the peanut milk yoghurt has higher protein content, fat, caloric value and apparent viscosity. However peanut milk yoghurt has a relatively lower lactose level. Therefore peanut milk yoghurt is reported to be suitable for lactose intolerant individuals. It is also reported that yoghurt from peanut milk has a higher proportion of unsaturated fatty acids (mainly oleic and linoleic), therefore it can be considered to be more health promoting. In terms of flavour, texture and appearance, the peanut based yoghurt is reported to be appreciated by the panelist (Isanga & Zhang, 2009).

Legume based yoghurt has been produced using cowpea seeds. Cowpea yoghurt was developed reported by (Sanni *et al.*, 1999). Initially cowpea milk was prepared. Seeds were sprouted. Sprouting is the practice of germinating seeds to be eaten raw or cooked. Once the sprouting process completed, the water was decanted and the seeds are mixed with sterilized water and ground. The blended slurry was filtered by making use of a double layered cheese cloth. Milk obtained was sterilized in a water bath at 70°C for 20 min. Pineapple flavour was used and added to enhance a pineapple flavor to the milk. The pineapple flavour was obtained by initially removing the fruit peels followed by washing, grating and mixing the pineapple fruit pulp with distilled water in the ratio 4:1. Artificial flavor of 1% was also added. The flavourant was then mixed with the cowpea milk after being sterilized. Flavoured and unflavoured cowpea milk samples were then inoculated with starter organisms, combination of *L. acidophilus* and *L. plantarum*. During the fermentation process there was a decrease in the final pH and an increase in the titratable acidity of the cowpea milk samples at the end of fermentation period. The cowpea milk samples were also inoculated using a single culture organism. The combination of the cultures is reported to be more favourable as there is a decrease in the pH and a greater increase in the titratable acidity. The crude protein, total ash, calcium, potassium and phosphorus values were reported to be greater on the cowpea yoghurt, as compared to the control and the commercial fan yoghurt. A decrease however is reported for lipid, moisture and crude fibre contents. The decrease obtained was estimated to be due to the loss of sample through the fine paste adhering to the cheese cloth. Therefore, the study showed that milk extracted from a plant source could be fermented with lactic acid bacteria to produce yoghurt-like product that is acceptable to the target consumers in terms of nutrition and flavour (Sanni *et al.*, 1999).

2.17 Conclusion

It is evident that the expansion to plant based milk yoghurt is crucial for several reasons including health, technical and economical. The need to shift from cow milk based yoghurt to plant based as an alternative has increased over the years. BGN contains a rich nutritional profile, and it can be utilized to close the malnutrition gap worldwide. The production of BGN powdered milk and yoghurt is feasible by employing drying technologies including spray drying and foam-mat drying process. Furthermore success production of powdered BGN milk and yoghurt will not only provide shelf stable products, but also promote the utilization of BGN.

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CHAPTER THREE

EFFECT OF SPRAY DRYING COMPARTMENT ON THE FUNCTIONAL, PHYSICAL, THERMAL AND NUTRITIONAL CHARACTERISTICS OF BAMBARA GROUNDNUT MILK POWDER

Abstract

Bambara groundnut milk (BGNM) was prepared using Bambara groundnut (BGN) flour. The seeds were pre-treated, dehulled then milled into flour. The BGN flour was re-hydrated with cold water at 25°C at a ratio of 1:10 for 2 h to make up slurry, which was processed accordingly (sieved, homogenized, pasteurized) to produce BGNM. The BGNM was dehydrated using the spray dryer process set at inlet temperature of 150°C and outlet temperature of 74°C. Total solids of BGNM to be spray dried was increased using different percentages (5%, 10%, 15%) of gluten free maltodextrin to obtain the optimal product yield. Bambara groundnut milk powder (BGNMP) as well as BGNM were subjected to chemical and functional analysis (water absorption index, water solubility index, wettability, emulsification capacity, bulk density, foaming capacity, foaming stability and colour measurement). The chemical properties of BGNMP and BGNM were investigated. Water absorption indices, fat absorption, bulk density, emulsification capacity, wettability, water solubility indices and foaming capacity ranged from 0.19 to 0.45 g/g, 1.22 to 1.98 g, 0.63 to 0.69 g/mL, 1.8 to 1.58 mL/g, 136.8 to 292.4 s, 85.18 to 90.25%, 0.29 to 34 mL, respectively. The foam stability at time intervals of 20, 40, 60 and 120 min ranged from 64.67 to 69.33 min, 63.00 to 67.33 min, 62.33 to 65.67 min, 61.33 to 63.00 min, respectively. BGNMP with 10% maltodextrin from the cyclone was significantly ($p < 0.05$) higher in WSI (88.80%) compared to BGNMP with 5% maltodextrin. BGNMP with 10% maltodextrin from the chamber was significantly ($p < 0.05$) in WAI (0.40 g/g) and FC (1.98 g) compared to BGNMP with 15% maltodextrin. BGNMP concentrated with 10% was preferred than the other BGNMP samples (5%, 15%). The effect of dryer compartments (CY and CH) had a significant difference ($p < 0.05$) in WAI, FA, BD and wettability, while EC, WSI and FC did not differ significantly ($p > 0.05$). There was a significant difference ($p < 0.05$) in protein, while there was no significant difference ($p > 0.05$) in total dietary fibre of BGNM and BGNMP. There was a significant difference ($p < 0.05$) in the colour characteristic of BGNMP samples. Colour characteristics ranged from 86.63 ± 0.34 to 94.03 ± 0.13 for lightness (L^*), 0.36 ± 0.01 to 1.31 ± 0.02 for redness (a^*), 6.99 ± 0.55 to 12.21 ± 0.16 for yellowness (b^*), 7.00 ± 0.55 to 12.26 ± 0.16 for chroma (c^*) and 84.16 ± 0.35 to 87.06 ± 0.57 for hue (h°). The colour difference ΔE between the different concentrated BGNMP samples was found to be acceptable.

3.1 Introduction

Legumes are an important source of protein, starch, fats, minerals and healthy protecting compounds as they are sourced from plants. Their nutrients play an important role in the traditional diet of many people in the world. Legume seeds such as BGN contain 200-250 g protein/kg; the protein is mostly rich in lysine (Schuster-Gajzago, 2004).

Bambara groundnut (BGN) is a legume seed that belongs to the family *Leguminosae*, subfamily *Papilionadeae*. BGN is an indigenous crop that produces an almost balanced food. It is tolerant to drought and is easy to cultivate and requires little demand on the soil. Thus the seed is not prone to the risk of crop failure. The seed is ranked the third most important legume after groundnut (*Arachis hypogaea*) and cowpea (*Vigna unguiculata*). Most researchers agree that BGN seeds are relatively high in lysine and are low in methionine and calcium. Other studies done also revealed that BGN contains 17.4-18% protein, 53.1-65% carbohydrates, 6.1% fat, 6.1% fibre, 3.4% ash (Baryeh, 2001). Yusuf *et al.* (2008) reported 1.8-4.3% moisture, 6-10.4 % fat, 18.5-20.7% crude protein, 4.4-6.3% ash, 3.0-3.3% crude fibre and 60-61.3% of carbohydrates. However, nutritive values of BGN differ slightly according to different researchers; Atiku *et al.* (2004) reported that the BGN seed contains 63% carbohydrates, 19% protein and 6.5% fat. The difference in nutrients could be due to different processing employed, including removal of hulls. Nonetheless these values indicate that BGN is a nutritious food.

Traditionally, BGN can be consumed in many different forms. It can either be fried or boiled with salt and eaten as a snack or can be pounded into flour and used in the preparation of soup, porridges and various fried or steamed African food products such as “akara”, “moi-moi” and “okpa” common in Nigeria. Local food drinks such as “kunu” and food stuff such as “tuwo” can be produced from BGN seed, as well as bread. Functional properties of BGN flours reported by Yusuf *et al.* (2008) showed water absorption capacity of 174%, oil absorption capacity of 150%, emulsion capacity of 78.5%, foaming capacity of 70% and foaming stability of 83%. Murevanhema & Jideani (2012) produced BGN milk and further evaluated its quality and sensory attributes. The success of developing BGNM by Murevanhema & Jideani (2012) encouraged the need to explore the production of BGNM powder. The functional properties of the BGN milk powder could find use as food ingredients. The objective of this chapter was to investigate the functional and nutritional characteristics of Bambara groundnut milk (BGNM) powder.

3.2 Materials and Methods

3.2.1 Source of materials and equipment

The BGN seeds were purchased from Trio trade, Johannesburg, South Africa. Disodium phosphate (CJP Chemicals, South Africa), sodium citrate (CJP Chemicals, South Africa), sunflower lecithin (DuPoint, South Africa), carrageenan (iota and kappa mixture) (CJP

Chemicals, South Africa) and maltodextrin (Tate & Lyle, South Africa) ingredients were used for the formulation of BGN milk. The analyses carried out in this chapter are outlined by Figure 3.1. Equipment used for the experiment was obtained from the Food Science and Technology Department and the Chemical Engineering Department of Cape Peninsula University of Technology.

3.2.2 Cleaning and processing of BGN seeds.

The BGN seeds were manually sorted to remove foreign objects. The seeds were steamed (Butcherquip Junior mini cooker, Model:CCB 0170, South Africa) for 1 h at 97°C, further soaked for 6 h, rinsed and then dried (Cabinet dryer, Model:1069616) at 50°C for 48 h according to the method reported by Murevanhema & Jideani (2012). The dried seeds were placed in zip lock bags and stored in the refrigerator (4-6°C) until when required.

3.2.3 Preparation of BGN flour

The dried seeds were dehulled to remove the skin, using a manual Corona Laders Y CIA A.8 extruder. Once the skins were removed, the seeds were manually separated, from the hulls. The seeds with skin were placed on baking trays and were continuously hurled in the air, separating the hulls from the seed and, once separated, hulls were removed by hand. The seeds were then ground to flour with sieve size 250 µm using a Trapp Animal Ration Shredder Hammer Mill Foliage TRF 400. The final BGN flour was placed into zip lock bags and stored at refrigeration temperature (4-6°C) until used.

3.2.4 Preparation of stable BGN milk

Throughout the production of BGNM, a multiple formulation system was attempted prior to obtaining the stable BGNM final milk. The initial step was to rehydrate the BGN flour with normal cold tap water at a ratio of 1:10. The solution was blended for 5 min using the Hallade SB-4 111262 warring blender and allowed to stand at room temperature (21 ± 2) for 2 h. The resultant slurry was filtered using 4 square fine cheese cloths folded twice, making 8 layers of cloth. The filtrate was allowed to stand for 10 min to allow the particles to settle at the bottom of container and the particles were discarded. The supernatant was followed by homogenisation using a Cadmach Ahmedabad-45 CMCM5 colloid mill. The solution was monitored for pH and total solids (TS), using the Crison, GLP 21 pH meter, Barcelona and a refractometer (Bellingham Stanley eclipse 45-06), respectively. After multiple experiments in order to produce stable milk, the supernatant was mixed with disodium phosphate (0.1%), sodium citrate (0.05%), powdered sunflower lecithin (0.1%), and carrageenan mixture of iota and kappa (0.03%). The solution was boiled at 100°C for 20 min on a hot plate stove, while stirring. The boiled solution was referred to as BGN “milk”. The milk was instantly transferred into sterile glass bottles and cooled to 4°C

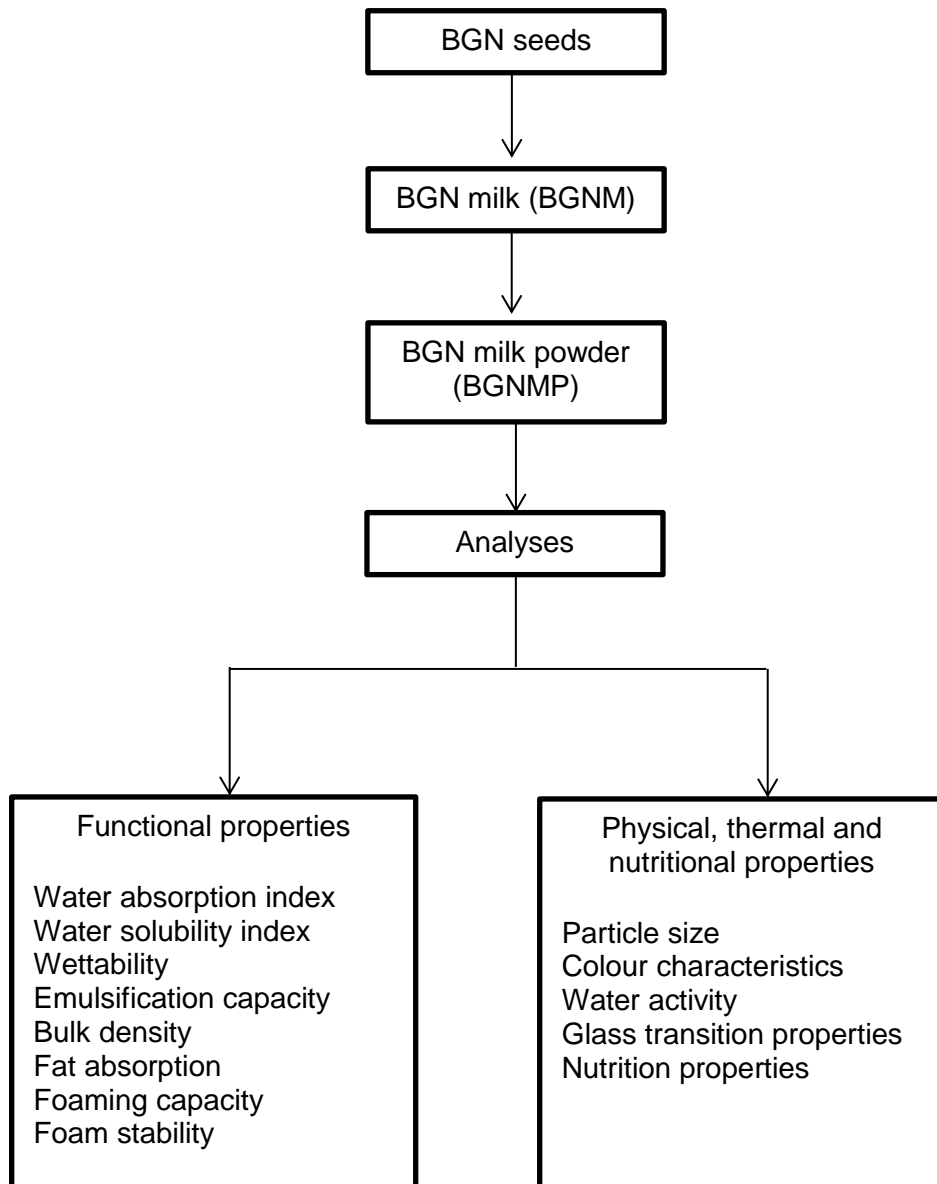


Figure 3.1

An overview of chapter 3

immediately. The BGN milk samples were stored under refrigeration temperature (4°C) and were further monitored for TS and pH.

3.2.5 Spray drying of the BGNM

BGN milk powder (BGNMP) was produced using an EDIBON SSPC Computer Controlled Spray Dryer with SCADA and PID control. Prior to spray drying, BGN milk described in section 3.2.4 was adjusted using corn maltodextrin powder with a dextrose equivalent (DE) of 20 as a drying carrier. Three levels of maltodextrin (5, 10 and 15%) were employed, elevating the TS of BGNM to 7, 11.5 and 15.5° Brix, respectively. The concentrated milk was pre-heated (60-80°C) and homogenised (85 rpm) for 20 min using a water bath prior to spray drying. The concentrated BGNM was fed through a silicon pump tube into the spray dryer by a peristaltic pump and atomized into small droplets by a 0.5 mm jet nozzle at an air pressure of 3 bars in a co-current air flow system. The inlet injection temperature (ST2) was set at 150°C and the outlet air temperature was automatically held at 70-74°C, dependent on the injection temperature and the flow rate. The flow rate of the liquid feed was set at 10% or 16 mL/min. The solution temperature was at 27.9°C and the room temperature was at 25.7°C. The flow of air was 42.9 m³/h. The final BGNM particles were collected from the cyclone and chamber (dryer compartments) and investigated for their functional and nutritional attributes.

3.3 Functional Characterisation of BGNM Powder

3.3.1 Water absorption index (WAI) and water solubility index (WSI)

WAI and WSI of the BGNM powder samples were determined using the method of Kaushal *et al.* (2012). BGNM powder (2.5 g) was dispersed in 30 mL of distilled water. The mixture was stirred using a glass rod, and cooked at 90°C for 15 min using a heating magnetic stirrer. The cooked paste was cooled to room temperature and transferred to tarred centrifuge tubes, and then centrifuged at 3000 g for 10 min. The supernatant was decanted into a tarred evaporating crucible dish for the determination of its solid content and the sediment was weighed. The weight of the dry solids was recovered by evaporating the supernatant overnight at 110°C. The analysis was done in triplicate and WAI and WSI were calculated by equations 3.1 and 3.2.

$$\text{WAI (g/g)} = \frac{\text{Weight of sediment}}{\text{Weight of dry sample}} \dots\dots\dots \text{equation 3.1}$$

$$\text{WSI (g/g)} = \frac{\text{Weight of dissolved solids in supernatant}}{\text{Weight of dry sample}} \dots\dots\dots \text{equation 3.2}$$

3.3.2 Wettability

Wettability was determined using the procedure described by Jinapong *et al.*, (2008) and carried out in triplicate. Distilled water (100 mL) with a temperature of $25 \pm 1^\circ\text{C}$ was poured into a 250 mL conical flask. A glass funnel was held over the edge of the conical flask, with the height between the bottom of the funnel and the water surface of 10 cm. A test tube was placed inside the funnel to block the lower opening of the funnel. BGNM powder (0.1 g) was placed around the test tube and the tube was lifted, while the stop watch started at the same time. Time taken for the powder to become completely wet, and for all the powder particles to penetrate the surface of the water, was recorded.

3.3.3 Emulsification capacity

The method of Lawal & Adebawale (2004) was used with slight modifications. A 2 g BGNM powder was mixed with 23 mL of distilled water. The mixture was mixed for 30 min using a vortex machine. Oil (Olive Pride Seed oil & Extra Virgin Olive oil) was added continuously at a rate of 1 mL/s from a burette until the emulsion break point was reached. The volume of oil added up to inversion point was noted and the emulsifying capacity (EC) was expressed as mL of oil per gram of sample. The emulsification determination was carried out at room temperature (21 ± 2) in triplicate.

3.3.4 Bulk density measurement

A sample of BGNMP (2 g) was placed into a graduated syringe and sufficient pressure was applied to pack the content in the syringe. The final volume of the sample in the syringe was recorded and bulk density was expressed as grams per millilitre (g/mL) (Parrott & Thrall, 1978).

3.3.5 Fat absorption (FA)

A sample of BGNM powder (1 g) was added to 5 mL of extra virgin olive oil in a 50 mL centrifuge tube. The content was mixed using a vortex for 15 sec every 15 min and after 20 min the tubes were centrifuged ($1600 \times g$, 25 min) at room temperature. Excess oil was decanted and tubes were weighed. FA was expressed as the amount of absorbed oil per gram of sample ($\text{g oil} \cdot \text{g}^{-1}$) (Wang & Toews, 2011).

3.3.6 Foaming capacity (FC) and foaming stability (FS)

Foaming properties of BGNM powder were determined according to the method reported by Kaushal *et al.* (2012). The dispersed sample (1.5 g in 50 mL distilled water), was homogenized using a homogenizer (Polytron n. Prof P.Williams) at high speed setting for 3 min. The blend was immediately transferred into a graduated cylinder and the homogenizer cup was rinsed with 10 mL of distilled water, which was then added to the graduated

cylinder. The volume was recorded before and after whipping. FC and FS were calculated using equations 3.3 and 3.4, respectively.

$$FC = \frac{\text{Increase in volume after homogenisation-Initial volume}}{\text{Initial volume}} \dots\dots\dots \text{equation 3.3}$$

FS =Foam volume changes recording at intervals of 20, 40, 60 and 120 min.....equation 3.4

3.4 Physical Characterisation of BGNMP

3.4.1 Particle size determination of BGNMP

The particle size and particle size distribution was analysed using a Leo 1430VP Scanning Electron Microscope (SEM) at Stellenbosch University. Prior to imaging the samples were mounted on a stub with double sided carbon tape. The samples were then coated with a thin layer of gold in order to make the sample surface electrically conducting. Beam conditions during surface analysis were 7 kV and approximately 1.5 nA, with a spot size of 150.

Particle sizes and particle size distribution of BGNMP was evaluated using images obtained from the SEM. The diameters of the particle sizes were measured individually according to the method of Tcholakova *et al.* (2004). BGNMP particle sizes were obtained in terms of volume-surface mean diameter ($d_{3,2}$) (Equation 3.5) and equivalent volume-mean diameter ($d_{4,3}$) (Equation 3.6).

$$d_{3,2} = \frac{\sum n_i d_{i3}}{\sum n_i d_{i2}} \dots\dots\dots \text{equation 3.5}$$

$$d_{4,3} = \frac{\sum n_i d_{i4}}{\sum n_i d_{i3}} \dots\dots\dots \text{equation 3.6}$$

3.4.2 Colour measurement of BGNMP

The colour of the BGNMP (3 g) was measured using a Konica Minolta Spectrophotometer CM-5 45°/0° standard, set at standard observer 10° and D65. The instrument was zero calibrated using a black tile ($L^* = 5.49$, $a^* = -7.08$, $b^* = 4.66$) and white calibration was performed using a white tile ($L^* = 93.41$, $a^* = -1.18$, $b^* = 0.75$). BGNMP samples were evenly placed in a petri-dish (30 mm diameter) and reflectance measured for $L^*a^*b^*$ and LCh colour scales. The L^* coordinate is lightness, 100 represents white and closer to 0 represents black Measurements for each sample were performed in triplicate at three different positions in the samples (one reading = average of three readings per rotated position), with the results recorded in L^* (lightness), a^* (chromaticity coordinate $+a^* =$ red and $-a^* =$ green), b^* (chromaticity coordinate $+b^* =$ yellow and $-b^* =$ blue), C^* (chroma) and h

(hue angle $0^\circ = +a^*$, $90^\circ = +b^*$, $180^\circ = -a^*$ and $270^\circ = -b^*$ and the total colour difference (ΔE) of the BGNMP was calculated using equation 3.7 (Bezerra *et al.*, 2013).

$$\Delta E = [(\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2]^{1/2} \dots \dots \dots \text{equation 3.7}$$

3.4.3 Water activity evaluation of BGNMP

Water activity (a_w) of BGNMP was measured using the Novasina Ms 1 Set Aw meter, which uses a cell protection filter to measure (a_w). Salt humidity standards of 53, 75 and 90% relative humidity were used to calibrate the measurement cell. BGNMP (5 g) was transferred into a sample dish and placed inside the Novasina analyser and the cell measuring protection filter was immediately closed. The a_w reading was observed after a period of 60 to 80 s. The test was performed in triplicates and the average was taken as the a_w value (Novasina General Catalogue, 2012)

3.5 Thermal Evaluation of BGNMP

3.5.1 Glass transition temperature assessment of BGNMP

The glass transition temperature (T_g) was determined using the method described by Shrestha *et al.* (2007a) with some modifications. A Perkin Elmer Pyris 1 Differential Scanning Calorimeter (DSC) 6000 equipped with Intracooler 2P was used to evaluate the glass transition temperature of Bambara groundnut milk powder (BGNMP) samples. The purge gas used was dry nitrogen (20 mL/min). Heat flow verification was performed using indium melting point (156.60°C) and zinc melting point (419.47°C) standards at heat scan rate of $10^\circ\text{C}/\text{min}$. About 10 mg of BGNMP was weighed into a 50 μL DSC aluminum pan and press sealed with a lid using DSC sample press. The thermal scanning of the samples was carried out alongside a sealed empty pan used as a reference. The thermal scanning of BGNMP was commenced from -20°C to 200°C at a rate of $10^\circ\text{C}/\text{min}$. The onset and endset values were calculated in the DSC thermo-gram as well as the T_g value was determined as half ΔC_p method at half the extrapolated change in specific heat using the pyris software. The analysis was carried out in triplicate.

3.6 Proximate analysis of BGNM powder

Protein determination was done using Kjeldahl ($N \times 6.25$), total fat according to AOAC (2005) method 996.06, total dietary fibre according to AOAC (1997) method 985.29, total sugar according to AOAC (2003) method 982.14, moisture and ash according to AOAC (2005) method 934.01 and 923.03, respectively. The carbohydrate was determined by difference and energy was determined according to Anon (2010).

3.7 Data Analysis

All measurements were carried out in triplicate. Results were expressed as mean \pm standard deviations. Multivariate analysis of variance was employed to establish differences between treatments. Separation of means where differences existed was determined by the Duncan's multiple range test (IBM SPSS 22).

3.8 Results and Discussion

3.8.1 Functional characteristics of BGNM powder as affected by maltodextrin concentrations

The descriptive statistics for the functional properties of BGNM powder as affected by maltodextrin concentration (5, 10 and 15%) from dryer compartments namely the cyclone (CY) and chamber (CH) is summarized in Table 3.1 and Table 3.2, respectively. A further statistical description of BGNMP on the functional properties with the effect of different dryer compartments is demonstrated in Table 3.3.

1 Bulk density

The BD properties of BGNM powder as affected by maltodextrin concentration (5, 10 and 15%) from dryer compartment CY is summarized in Table 3.1. The BD of BGNM powder ranged from 0.67 to 0.69 g/mL. The effect of 5, 10 and 15% concentration of maltodextrin on BGNMP did not differ significantly ($p > 0.05$) with respect to BD. The highest BD was obtained using the 15% concentration, while the lowest was obtained at 5 and 10% concentrations. The BD of BGNM powder from dryer compartment CH ranged from 0.63 to 0.67 g/mL (Table 3.2). The different maltodextrin concentration levels had a significant ($p < 0.05$) difference on BD. The bulk density of spray dried BGNM powder was higher as the maltodextrin concentration was increased. The most favourable results were obtained at BGNMP concentrated with 15%, than concentration of 10%.

Table 3.3 displays the effect of the drying compartment on the BD properties of spray dried BGNM powder. The BD of BGNMP from dryer compartments CY was 0.68 g/mL and that of CH was 0.65 g/mL. BGNM powders from different dryer compartments were significantly ($p < 0.05$) different from each other. The bulk density of a powder is defined as the mass of particles that occupies a unit volume of a container. It is determined by dividing the powder's net weight by the volume it occupied in a container (Barbosa-Canovas & Yan, 2003). It is a reflection of the load a sample can carry, if allowed to rest directly on another (Kaushal *et al.*, 2012). Bulk density of a mass of particles, which is measured with applied force or pressure, is known as packed or compact density (Onwulata, 2005). For this research, compact density methodology was applied. It is dependent on particle density, particle internal porosity, and arrangement of particles in the container (Sharma *et al.*, 2012).

The BGNMP with 15% maltodextrin found in dryer compartment CY and CH had the highest bulk density of 0.69 g/mL and 0.67 g/mL, respectively. This can be linked with what was reported by Chronakis (1998) and Knill & Kennedy (2004) stating, high DE maltodextrins have solubility, bulking, and bodying characteristics. Therefore, the higher the concentration, the higher the bulk density would be. Sharma *et al.* (2012) reported that the packed bulk densities of various milk powders, skim milk powders (SMP), whole milk powders (WMP), and fat free milk powders (FFMP) ranged from 0.44 to 0.88 g/cm³ (0.44 to 0.88 g/mL). This also suggests that the bulk density of BGNMP can be regarded as high or comparable to other commercial milk powders. The high BD of BGNMP found can be beneficial to producers when shipping powders over long distances because high bulk density powders reduces shipping volume, saves packaging and storage capacity.

2 *Emulsification capacity*

The emulsification capacity (EC) from the CY of BGNM powder as affected by maltodextrin concentration (5, 10 and 15%) is illustrated in Table 3.1. The EC of BGNM powder ranged from 1.28 to 1.58 mL/g. The effect of maltodextrin on EC did not differ significantly ($p > 0.05$). The EC of BGNM powder increased with decreasing maltodextrin concentration. The most effective EC was established at the maltodextrin concentration of 5%. The EC of BGNM powder at different maltodextrin concentrations from dryer compartment CH is depicted in Table 3.2. The EC of BGNM powder ranged from 1.18 to 1.20 mL/g. The effect of maltodextrin concentration had no significant ($p > 0.05$) difference, with regards to emulsification capacity. BGNM powder concentrated with 15% maltodextrin had a notable high emulsification capacity

Table 3.3 indicates the effect of the drying compartment on the emulsification capacity of BGNM powder. The EC of BGNMP from the CY was 1.45 mL/g and that from the CH was 1.20 mL/g. The BGNM powders from different dryer compartments (CH and CY) did not differ significantly ($p > 0.05$) with respect to EC. The emulsification activity describes the ability and capability of a protein to aid in the formation of an emulsion by being able to absorb to the interfacial area of oil and water in an emulsion. The emulsion stability normally reflects the ability of the proteins to impart strength to an emulsion in order to resist stress and changes and which affects the consistency combination of the interfacial area over a defined time period (Du *et al.*, 2014). Instability on the other hand is a result of protein present not sufficient enough to stabilize the fat globules; therefore particles are large, resulting in aggregation (Tangsuphoom & Coupland, 2008; Tangsuphoom & Coupland, 2009). It was observed that for BGNMP found in the cyclone, the increase in maltodextrin concentration lowered the EC and the increase in concentration had no increasing effect on EC. However, for BGNMP found in the chamber, the exact opposite effect was observed.

Table 3.1 Cyclone dryer compartment and maltodextrin effect on the functional properties of BGNM powder

Maltodextrin Concentration							
(%)	BD (g / mL)	EC (mL / g)	Wett. (sec)	WAI (mL / g)	WSI. (100 / g)	FA (g)	FC (mL)
5	0.67 ± 0.00 ^a	1.58 ± 0.33 ^a	256.6 ± 62.82 ^a	0.45 ± 0.05 ^a	85.15 ± 1.22 ^a	1.13 ± 0.23 ^a	0.31 ± 0.08 ^a
10	0.67 ± 0.01 ^a	1.50 ± 0.61 ^a	263.4 ± 111.81 ^a	0.26 ± 0.01 ^b	88.80 ± 1.30 ^b	1.33 ± 0.21 ^a	0.29 ± 0.04 ^a
15	0.69 ± 0.02 ^a	1.28 ± 0.25 ^a	292.41 ± 40.20 ^a	0.19 ± 0.01 ^c	88.05 ± 0.82 ^b	1.20 ± 0.05 ^a	0.30 ± 0.01 ^a

Mean values ± SD of triplicate determinations. Mean values in the same column followed by different letters are significantly ($p < 0.05$) different; WAI: Water absorption index; FA: Fat absorption; BD: Bulk density; EC: Emulsification capacity; Wett.: Wettability; WSI: Water solubility index; FC: Foaming capacity

Table 3.2 Chamber dryer compartment and maltodextrin effect on the functional properties of BGNM powder

Maltodextrin							
Concentration							
(%)	BD (g / mL)	EC (mL /g)	Wett. (sec)	WAI (g / g)	WSI (100 / g)	FA (g)	FC (mL)
10	0.63 ± 0.02 ^a	1.18 ± 0.10 ^a	235.8 ± 32.07 ^a	0.40 ± 0.04 ^a	86.80 ± 0.93 ^a	1.98 ± 0.06 ^a	0.34 ± 0.07 ^a
15	0.67 ± 0.00 ^b	1.20 ± 0.13 ^a	136.8 ± 12.40 ^b	0.20 ± 0.02 ^b	90.25 ± 0.25 ^b	1.76 ± 0.10 ^b	0.31 ± 0.01 ^a

Mean values ± SD of triplicate determinations. Mean values in the same column followed by different letters are significantly ($p < 0.05$) different; WAI: Water absorption index; FA: Fat absorption; BD: Bulk density; EC: Emulsification capacity; Wett.: Wettability; WSI: Water solubility index; FC: Foaming capacity

Table 3.3 Effect of dryer compartments on the functional properties of BGNM powder

Dryer Compartment	BD (g / mL)	EC (mL /g)	Wett. (sec)	WAI (g / g)	WSI (100 / g)	FA (g)	FC (mL)
CY	0.68 ± 0.02 ^a	1.45 ± 0.40 ^a	270.81 ± 69.2 ^a	0.30 ± 0.12 ^a	87.34 ± 1.92 ^a	1.22 ± 0.18 ^a	0.30 ± 0.05 ^a
CH	0.65 ± 0.02 ^b	1.20 ± 0.11 ^a	186.3 ± 58.42 ^b	0.31 ± 0.10 ^b	88.53 ± 58.42 ^a	1.88 ± 0.14 ^b	0.33 ± 0.05 ^a

Mean values ± SD of triplicate determinations. Mean values in the same column followed by different letters are significantly ($p < 0.05$) different ($p < 0.05$); WAI: Water absorption index; FA: Fat absorption; BD: Bulk density; EC: Emulsification capacity; Wett: Wettability; WSI: Water solubility index; FC: Foaming capacity, CH: Chamber; CY: Cyclone

The increase in concentration of maltodextrin showed an increase in the EC of BGNMP. However the difference was not significant ($p < 0.05$).

3 *Wettability*

The wettability of BGNM powder as affected by maltodextrin concentration (5, 10 and 15%) from dryer compartment CY is summarized in Table 3.1. The wettability of BGNM powder ranged from 256.6 to 292.41 s. The effect of 5, 10 and 15% concentration of maltodextrin on BGNMP did not differ significantly ($p > 0.05$) with respect to wettability. The wettability reached its peak at 15% and dropped at 5% maltodextrin concentration. The wettability of BGNM powder at different maltodextrin concentrations of 10% and 15% from dryer compartment CH are depicted in Table 3.2. The wettability of BGNM powder ranged from 136.8 to 235.80 s. The levels of maltodextrin concentration had a significant ($p < 0.05$) difference on wettability. BGNM powder concentrated with 10% maltodextrin showed the highest wettability as the milk powder was immersed in water for wettability proficiency. BGNM powder concentrated with 15% maltodextrin, despite the higher quantity had little wettability capabilities when immersed in water.

Table 3.3 describes the effect of the drying compartment on the wettability of spray dried BGNM powder. The wettability of dryer compartment CY was 270.81 s and that of CH was 186.30 s. BGNM powders from different dryer compartments were significantly ($p < 0.05$) different from each other. Wettability measures the time taken for the powder to sink below the water surface and absorb water, to be wetted, and to penetrate the surface at a given temperature (Westergaard, 2004; Baldwin & Pearce, 2005; Sharma *et al.*, 2012). The variation of maltodextrin concentration on BGNMP from dryer compartment CY showed no significant difference on its wettability, which means that all powders, had the same influence regarding water penetration. The time taken for BGNMP to be completely penetrated by water concentrated with 5, 10 and 15% ranged from 2-3 min. However, there was a significant ($p < 0.05$) difference for BGNMP from dryer compartment CH.

The increase in maltodextrin concentration reduced the time taken to completely wet and penetrate the milk powder particles. BGNMP (compartment CH) concentrated with 10% and 15% took exactly 3 min 55.8 s and 1 min 17 s, respectively. Sharma *et al.* (2012) reported that skim milk powder (SMP) wetted in less than 15 s is termed “instant” and further stated that whole milk powder (WMP) wetted within 30 – 60 sec are advantageous, but also added that for WMP, there is no requirement with respect to fast wetting. Westergaard. (2004) also reported that skim milk powders (SMP) are usually easily hydrated due to a reduced fat content of 0.03%. Hence, BGNMP cannot be considered as “instant” which then can be linked to the fact that it has a fat content of 1.6% [Section 3.8.4, page 77].

4 *Water absorption index*

The water absorption index (WAI) of BGNM powder as affected by maltodextrin concentration (5, 10 and 15%) from dryer compartment CY is summarized in Table 3.1. WAI of BGNM powder at different concentrations ranged from 0.19 to 0.45 g/g. The effect of maltodextrin concentrations (5, 10 & 15%) differed significantly ($p < 0.05$). At the lowest concentration (5%), the water absorption index was much higher, as the maltodextrin concentration was increased the water absorption capacity decreased. The WAI of BGNM powder from dryer CH shown in Table 3.2 ranged from 0.22 to 0.40 g/g. Increasing maltodextrin concentration significantly ($p < 0.05$) decreased WAI. The water absorption index was higher in BGNM powder concentrated with 10% maltodextrin than with 15%

Table 3.3 displays the effect of the drying compartment on the WAI of BGNM powder. The WAI of dryer compartments CY was 0.30 g/g and that of CH was 0.31 g/g. BGNM powders from different dryer compartments were significantly ($p < 0.05$) different from each other. The WAI determines the volume occupied by the powder after it swells in excess water and indicates the integrity of powder in aqueous dispersions (Kaushal *et al.*, 2012; Du *et al.*, 2014). The increase in maltodextrin significantly reduced the water absorption index of BGNMP in both dryer compartments CY and CH. BGNMP concentrated with 5% maltodextrin had the highest WAI and BGNMP concentrated with 15% concentration had the lowest WAI. Many of maltodextrin attributes introduced as a hydrocolloid to already complex systems are directly related to the product's texture properties (e.g. creaminess, thickness, smoothness, spreadability, pourability, flowability, brittleness and hardness) (Dokić *et al.*, 2004).

The observed trend of decreasing WAI of BGNMP with increased maltodextrin concentration can be linked to the hardness and brittleness associated with maltodextrin, which can retard the water absorption capabilities. This occurs when the maltodextrin is introduced to an already complex system (Dokić *et al.*, 2004). Furthermore water absorption involves the action of successful hydrogen bonding in a system between components (Sathe, 2012). Water is known to contain two hydrogen atoms, maltodextrin on the other hand is a starch hydrolysate having up to five hydrogen atom (Blamire, 2003; Knill & Kennedy, 2004). At lower levels of maltodextrin, water absorption capabilities may increase because the hydrogen atoms of water are not dominated by those of maltodextrin. However, higher levels of maltodextrin may result in more hydrogen atoms that dominate allowing the system to reach its saturation point, retarding water absorption capabilities. This may be used to explain the observed trend.

5 *Water solubility index*

The WSI of BGNM powder as affected by maltodextrin concentration (5, 10 and 15%) from dryer compartment CY is defined in Table 3.1. The water solubility index of BGNM powder

ranged from 85.2 to 88.8%. BGNMP concentrated with 10 and 15% maltodextrin did not differ significantly ($p > 0.05$), in water solubility index. However, BGNMP concentrated with 5% concentration was significantly ($p < 0.05$) lower in WSI compared to 10 and 15% concentrated BGNMP. The WSI of BGNM powder as affected by maltodextrin concentrations of 10% and 15% from dryer compartment CH was significantly ($p < 0.05$) different (Table 3.2). The WSI of BGNM powder ranged from 86.8 to 90.3%. BGNM powder concentrated with 15% maltodextrin was more soluble in water than BGNM powder concentrated with 10% maltodextrin. Therefore, WSI was most favourable at 15% concentration. Table 3.3 is an illustration of the effect of the drying compartment on the WSI of spray dried BGNM powder. The WSI of dryer compartments CY was 87.3% and that of CH was 88.5%. The BGNM powders from different dryer compartments (CH and CY) were not significantly ($p > 0.05$) different in WSI.

The solubility index of a powder is a measure of the degree to which it can be readily solubilized in water before use. This is linked to the amount of sediment obtained under defined conditions of mixing milk powders (Augustin *et al.*, 2003). The increase in maltodextrin concentration significantly increased the WSI of BGNMP in both dryer compartments CH and CY. WSI of BGNMP affected by 10 and 15% concentration was not significantly different from one another; however they were significantly different from BGNMP concentrated with 5%. The solubility of milk powders is dependent on pH and solubility is increased on the acid and alkaline side of this pH (Augustin *et al.*, 2003). Therefore the high solubility of BGNMP (range 85 to 90%) can be justified by the pH of BGN milk which ranged from 6.50 to 6.93, near to alkalinity.

6 *Fat absorption*

The fat absorption (FA) of BGNM powder as affected by maltodextrin concentration (5, 10 and 15%) from dryer compartment CY is given in Table 3.1. The FA ranged from 1.13 to 1.33 g. The effect of 5, 10 and 15% maltodextrin on BGNMP did not differ significantly ($p > 0.05$) with respect to FA. FA capacity was lowest at 5% concentration, at 10% the FA capability increased and at 15%, it decreased once again. Therefore, 10% concentration gave the highest FA capacity. The FA of BGNM powder from dryer CH ranged from 1.76 to 1.98 g (Table 3.2). The different maltodextrin concentrations were significantly ($p < 0.05$) different. FA was higher for BGNMP concentrated with 10%, than 15% concentration. The FA of dryer compartments CY was 1.22 g and that of CH was 1.88 g (Table 3.3). BGNM powders from different dryer compartments were significantly ($p < 0.05$) different from each other with respect to FA.

Fat absorption capacity is the ability of the milk powder protein to physically bind fat by capillary attraction. The mechanism of oil absorption has been attributed to the physical entrapment of oil within protein isolates and non-covalent bonds such as hydrophobic,

electrostatic and hydrogen bonds, which are the forces involved in lipid-protein interaction (Kaushal *et al.*, 2012; Falade *et al.*, 2014). The non-polar side chains of proteins are responsible for binding hydrocarbon side chains of oil (Kaushal *et al.*, 2012). BGNMP found in dryer CH was notably larger in particle size, as compared to BGNMP collected from dryer CY. This may have allowed the ease and frequent physical entrapment of oil within protein, which explains the higher FA for BGNMP collected from dryer CH (1.88 g) as compared to dryer CY (1.22 g). In addition both BGNMP were comparable to butter milk powder with fat absorption of 1.2g of oil/g of protein, which is rich in lecithin and used as a milk solid in the food industry (Sharma *et al.*, 2012).

7 Foam capacity

The foam capacity (FC) of BGNM powder as affected by maltodextrin concentration (5, 10 and 15%) from dryer compartment CY is demonstrated in Table 3.1. The FC of BGNM powder ranged from 0.29 to 0.31 mL. The effect of 5, 10 and 15% maltodextrin on BGNMP did not differ significantly ($p > 0.05$) with respect to FC. FC of BGNM powder was highest at concentration of 5% and lowest at 10% maltodextrin concentration. The FC of BGNM powder of 10% and 15% maltodextrin from dryer compartment CH is revealed in Table 3.2. The FC of BGNM powder ranged from 0.31 to 0.34 mL. The effect of maltodextrin (10, 15%) did not differ significantly ($p > 0.05$) in foaming capacity. However, a notable high foaming capacity of BGNM powder was observed at a concentration of 10%.

In Table 3.3 the effect of the drying compartments on the FC of spray dried BGNM powder is illustrated. The FC of the CY and CH was 0.30 mL and 0.33 mL, respectively. The BGNM powders from different dryer compartments (CH and CY) had no significant ($p > 0.05$) difference on FC. Foam capacity is the ability of a solution to produce stable foam. Proteins and surfactants present in the solution influences foam texture and are involved in retaining or improving stability of foams (Blasco *et al.*, 2011). Furthermore, protein is the surface active agent responsible for kinetic stability by reducing surface tension, thereby enabling formation of foams (Indrawati *et al.*, 2008). Therefore, the increase in maltodextrin may not be expected to result in an increased foam capacity, since maltodextrin has not been reported to be a source of protein. The variation of maltodextrin concentration had no significant difference in foam capacity of BGNMP found in both dryer compartment CY and CH.

8 Foam stability of spray dried BGNMP at different maltodextrin concentrations

Figure 3.2 depicts foam stability of BGNMP concentrated with maltodextrin in dryer compartment CY. The foam stability of BGNMP concentrated with 5, 10 and 15% maltodextrin at time intervals of 20, 40, 60 and 120 min ranged from 61.67 to 64.67 mL, 62.33 to 67.0 mL and 62.0 to 66.67 mL, respectively. There was no significant ($p > 0.05$) difference in the foam stability of BGNMP concentrated with 5% maltodextrin throughout the time intervals of 20, 40, 60 and 120 min. Foam stability of BGNMP concentrated with 10% and 15% concentration was significantly ($p < 0.05$) reduced between intervals of 20 to 120 min. BGNMP showed no significant ($p > 0.05$) difference in foam stability throughout the standing period of 120 min at 5%, while BGNMP concentrated to 10 and 15% showed foam to be reduced significantly ($p < 0.05$) throughout the standing period of 120 min. This could be because BGNMP concentrated with 5% initially did not reach fairly high foam stability as compared to 10 and 15% concentration and therefore after 120 min the change in foam stability was not seen as significant. This implies that the increase in concentration does not prolong the stability of BGNMP foam. Maltodextrin has been classified to hold multiple food functions one of which is fat binding (Chronakis, 1998). Nevertheless, Oetjen *et al.* (2014) stated that lipids have a large influence on foaming properties of milk and the presence of fat can adversely affect the stability of milk. However, skim milk produces rather stable foam. It is also stated that other lipids such as olive oil, which is a plant source improves the milk foam stability and maltodextrin used in this research is sourced from corn, which is also a plant source. Therefore, BGNMP with higher concentration of maltodextrin (15%) resulted in highest foam volume during foam formation however; BGNMP concentrated with 10% maltodextrin remained more stable over a period of 120 min.

The foam stability of BGNMP from the dryer compartment CH at different concentrations of 10 and 15% is summarized in Figure 3.3. The foam stability of BGNMP concentrated with 10 and 15% maltodextrin at time intervals of 20, 40, 60 and 120 min ranged from 61.33 to 69.33 mL and 63.0 to 69.0 mL, respectively. The foam stability of BGNMP from time interval of 20 to 120 min was significantly ($p < 0.05$) reduced at both concentrations. However, BGNMP concentrated with 10% maltodextrin was higher in foam stability over time. Figure 3.4 depicts foam stability of BGNM powder from different dryer compartments. Foam stability of BGNM powder from dryer compartment CY at intervals of 20, 40, 60 and 120 min was 66.11 mL, 64.22 mL, 63.11 mL and 62.00 mL, respectively and that of CH was 69.17, 67.33, 65.17 and 62.17 mL, respectively. The different dryer compartments were significantly ($p < 0.05$) different in foam stability. This may be due to the fact that BGNMP found in dryer CY differed from the powder found in dryer CH. This is because in the chamber (CH) heat intensity levels were high (150°C), compared to those of cyclone (CY) during the drying process. BGNMP found in dryer CY was visibly smaller, more brittle, had spreadability compared to powder found in dryer CH.

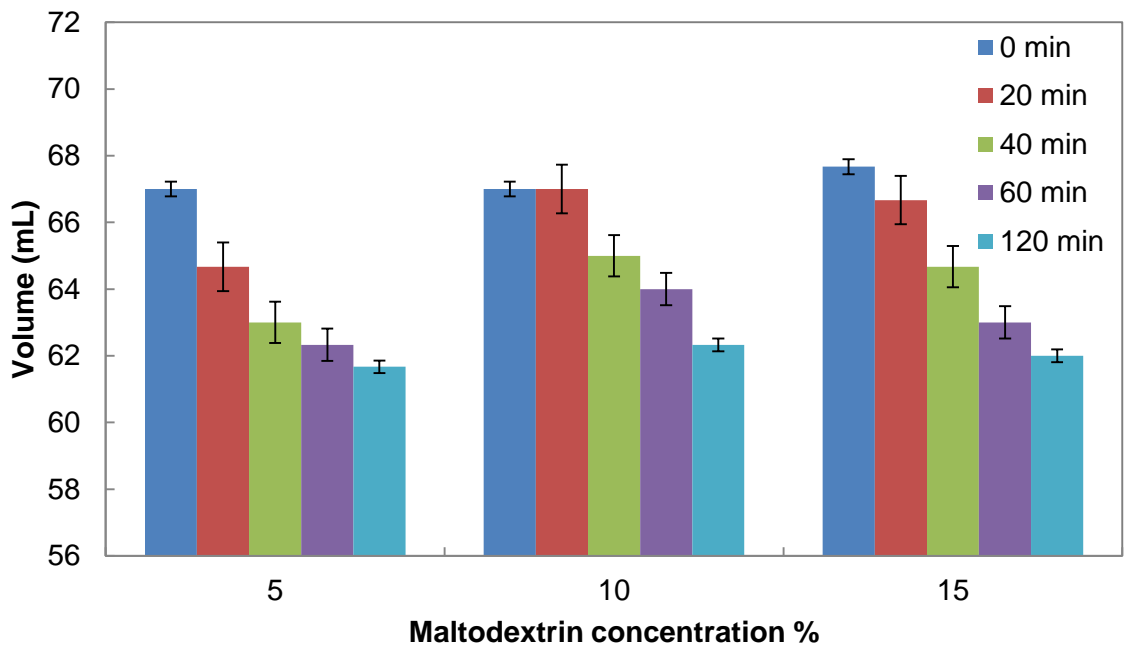


Figure 3.2 Cyclone and maltodextrin concentration effect on foam stability of Bambara groundnut milk powder (BGNMP).

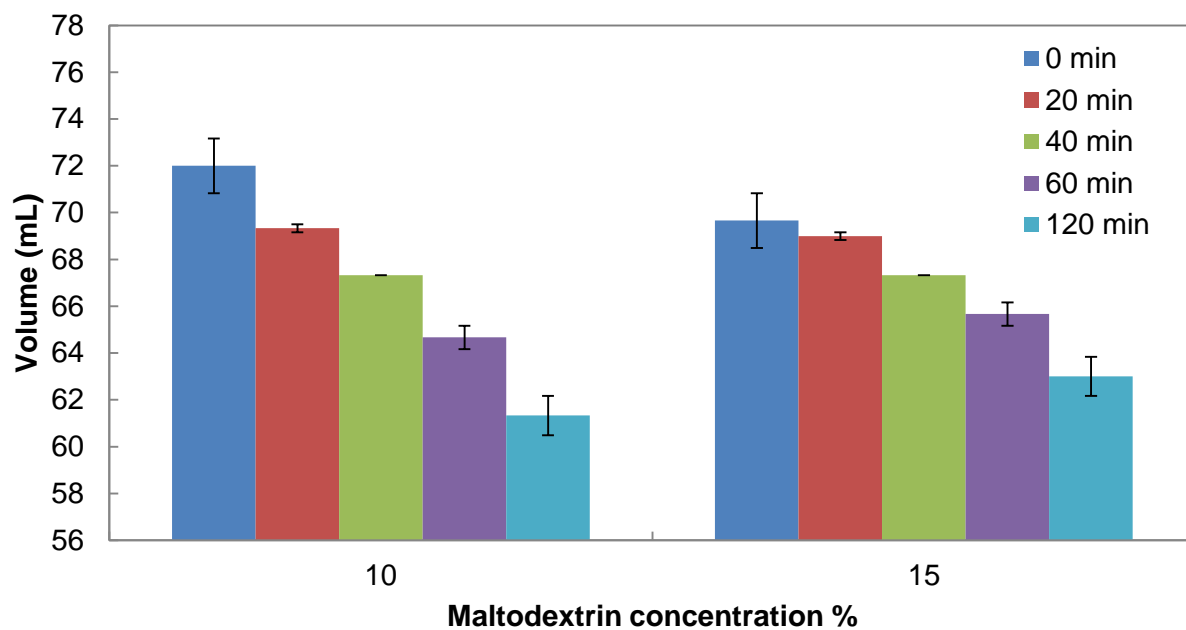


Figure 3.3 Chamber and maltodextrin concentration effect on foam stability of Bambara groundnut milk powder (BGNMP)

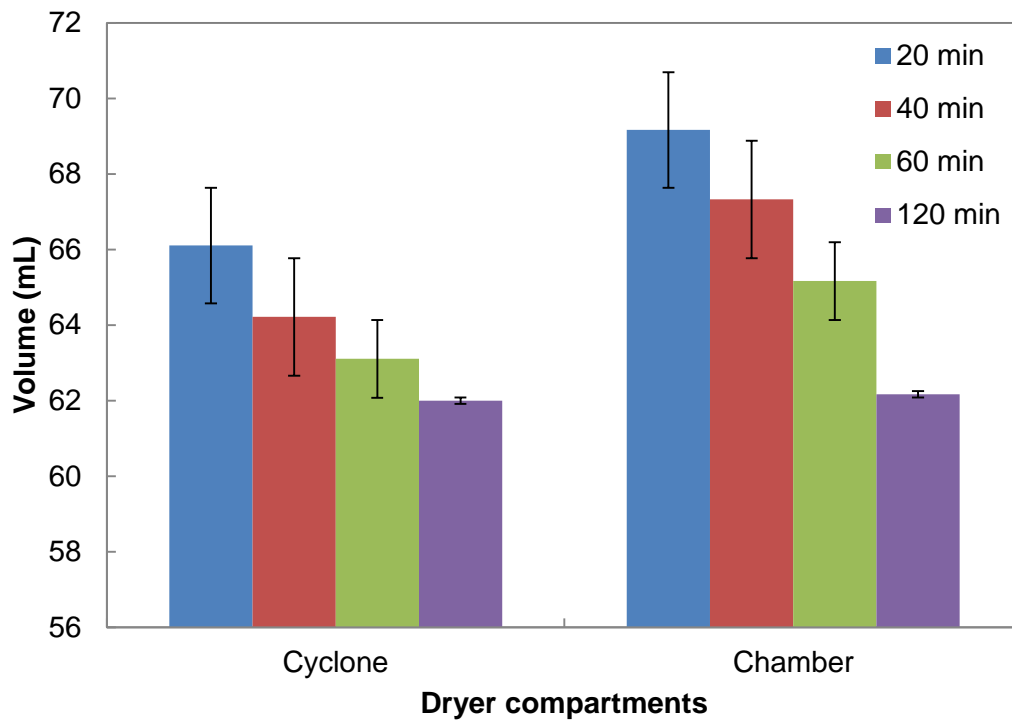


Figure 3.4 Effect of dryer compartment on the foam stability of Bambara groundnut milk powder (BGNMP)

3.8.2 Physical characteristics of Bambara groundnut milk powder

1 Particle size and particle size distribution of BGNMP

Descriptive statistics on particle size and particle size distribution of BGNMP is summarized in Table 3.4. The surface area mean diameter also known as the Sauter mean diameter ($d_{3,2}$) (Filippa *et al.*, 2012) of BGNMP concentrated with 5, 10 and 15% from the cyclone was 119.36, 105.39 and 157.14 μm , respectively. The surface area mean diameter ($d_{3,2}$) of BGNMP concentrated with 10 and 15% from the chamber was 112.57 μm and 84.05 μm , respectively. BGNMP at different concentration of 5, 10 and 15% from the cyclone did not differ significantly ($p > 0.05$) in the surface area mean diameter ($d_{3,2}$). BGNMP concentrated with 10 and 15% maltodextrin from the chamber also did not differ significantly ($p < 0.05$) in the surface area mean diameter ($d_{3,2}$). However, the effect of dryer compartments had a significant difference ($p < 0.05$) in the surface area mean diameter ($d_{3,2}$). BGNMP concentrated with 10% maltodextrin had the lowest surface area mean diameter ($d_{3,2}$) of 105.39 μm compared to BGNMP concentrated with 5% (119.36 μm) and 15% (157.14 μm) from the cyclone. Meanwhile, BGNMP concentrated with 10% had the highest (112.57 μm) surface area mean diameter ($D_{3,2}$) compared to BGNMP concentrated with 15% (84.04 μm) from the chamber. Particle size of BGNMP concentrated with 10% maltodextrin both from the cyclone and chamber were slightly similar with surface area mean diameter ($d_{3,2}$) of 105.39 μm and 112.57 μm , respectively.

The volume or weight mean diameter also known as De Brouckere mean ($d_{4,3}$) (Horiba Scientific, 2014) of BGNMP with the effect of 5, 10 and 15% maltodextrin from the cyclone was 126.49, 113.51 and 162.35 μm , respectively. The volume mean diameter ($d_{4,3}$) of BGNMP concentrated with 10 and 15% from the chamber was 115.47 μm and 86.13 μm respectively. BGNMP at concentrations of 5, 10 and 15% from the cyclone did not differ significantly ($p > 0.05$) in the volume mean diameter ($d_{4,3}$). Additionally, BGNMP concentrated with 10 and 15% maltodextrin from the chamber also did not differ significantly ($p > 0.05$) in the volume mean diameter ($d_{4,3}$). However, the effect of compartments, displayed a significant ($p < 0.05$) difference in the volume mean diameter ($d_{4,3}$)

Kim *et al.* (2009a) reported high heat exposure promotes fast formation of crust. Moisture is evaporated rapidly resulting in reduced redistribution of components (less fat and protein to the crust). Hence the skin becomes hard and dry leading to wrinkled looking particles (Kim *et al.*, 2009a; Xiao & Gao, 2012). This explains the visibly hard surface of BGNMP from the chamber. Furthermore, lower heat exposure results in reasonable moisture evaporation prior to crust formation. Therefore, the dissolved components have enough time to migrate within the droplets, resulting in milk powder with smooth particle surface (Xiao & Gao, 2012). This explains the notably smooth surface particles of BGNMP from the cyclone. Moreover high heat exposures lead to uniform size and shape, while high heat exposure.

Table 3.4 Particle size and particle size distribution of Bambara groundnut milk powder

BGNMP			
Dyer compartment	concentration (%)	d_{3,2} (µm)	d_{4,3} (µm)
Cyclone	5	119.36 ± 10.78 ^a	126.49 ± 13.58 ^a
	10	105.39 ± 17.61 ^a	113.51 ± 16.29 ^a
	15	157.14 ± 0.58 ^a	162.35 ± 0.66 ^a
Chamber	10	112.57 ± 19.45 ^a	115.47 ± 20.96 ^a
	15	84.04 ± 5.88 ^a	86.13 ± 5.88 ^a

Mean values ± standard deviation of triplicate determinations. Mean values in the same column, within each dryer compartment followed by different letters are significantly ($p < 0.05$) different; BGNMP: Bambara groundnut milk powder; d_{3,2}: surface area mean diameter; d_{4,3}: volume mean diameter.

results in size variation. Therefore, explains the significant ($p < 0.05$) difference of dryer compartments in volume mean diameter value ($d_{4,3}$) and surface area mean diameter ($d_{3,2}$).

Determination of particle size in milk powders is one of the important physical properties. Particle size affects crucial milk powders aspects such as reconstitution and rehydration properties, including solubility and wettability. Larger, irregular in shape particles have been reported to induces wettability and exhibit good dispersibility because they provide more interstices space for wetting (Dairy Ingredient Application Programme, 2000). It was stated that spray dried milk powders have a diameter range of 10 to 250 μm (Sharma *et al.*, 2012) making BGNMP an exception as powdered milk.

Scanning electron microscopy (SEM) images were obtained to examine particle morphology of BGNMP (Figure 3.5). Fyfe *et al.* (2011) reported morphological characteristics of spray dried milk powder to be spherical particles containing vacuoles of occluded air. However, all BGNMP concentrations SEM images resembled a “buckled” morphological behaviour, which is in agreement with the statement made by Rogers *et al.* (2012). Stating that general morphology of uniform powder particles is “buckled” regardless of the temperature exposure or initial feed solid content. The authors further stated that low feed solid content causes particles to buckle extensively, while particles dried from a more concentrated feed show less signs of deformation from a sphere. This statement is in agreement with the obtained BGNMP SEM images, in which BGNMP with 5% maltodextrin (Figure 3.5 A) was more buckled compared to BGNMP with 15% maltodextrin (Figure 3.5 C), showing less signs of sphere distortion.

2 Colour characteristics of BGNM powder at different maltodextrin concentrations

The descriptive analysis for colour determination of BGNMP from CY is displayed in Table 3.5. The average values of lightness for 5, 10 and 15% BGNMP from compartment CY was 91.98, 98.85 and 94.03, respectively. BGNMP concentrated with 5 and 10% maltodextrin did not differ significantly ($p > 0.05$). However, further increase to 15% showed a significant ($p < 0.05$) difference in lightness. The redness of BGNMP concentrated with 5, 10 and 15% had mean values of 0.78, 0.36 and 0.62, respectively and the hue angles were 84.16°, 87.06° and 84.96°, respectively. Redness and hue angle of BGNMP concentrated with 5 and 15% maltodextrin did not differ significantly ($p > 0.05$). However, the redness and hue angle of BGNMP with 10% maltodextrin was significantly ($p < 0.05$) different from 5 and 15%. The yellowness of BGNMP concentrated with 5, 10 and 15% had mean values of 7.61, 6.99 and 7.03, respectively and mean chroma of 7.65, 7.0 and 7.06, respectively.

The yellowness and chroma of BGNMP were not significantly ($p > 0.05$) affected by

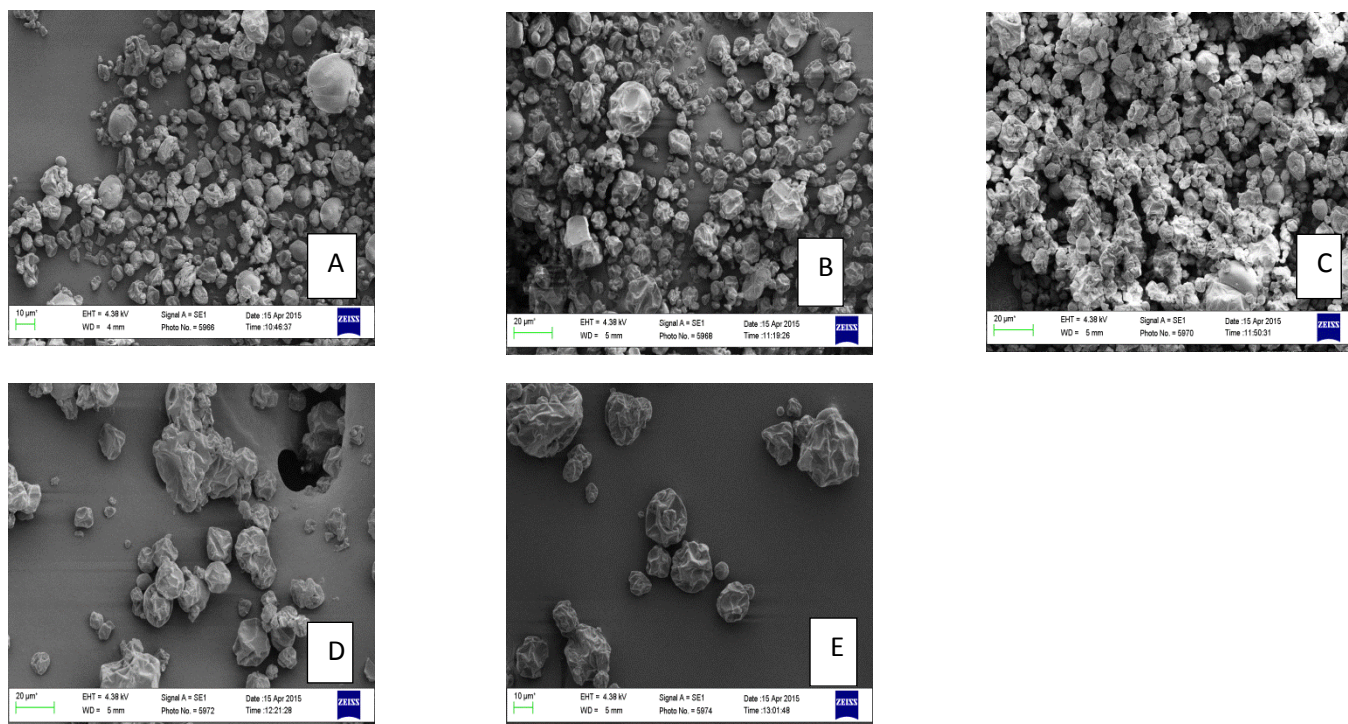


Figure 3.5 Scanning micrographs of Bambara groundnut milk powder (BGNMP). **A:** BGNMP with 5% maltodextrin in cyclone; **B:** BGNMP with 10% maltodextrin in cyclone; **C:** BGNMP with 15% maltodextrin in cyclone; **D:** BGNMP with 10% maltodextrin in chamber; **E:** BGNMP with 15% maltodextrin in chamber

Table 3.5 Cyclone and maltodextrin effect on the colour of BGNM powder

Maltodextrin concentration %	L*	a*	b*	C*	h°
5	91.98 ± 0.56 ^a	0.78 ± 0.09 ^a	7.61 ± 0.39 ^a	7.65 ± 0.39 ^a	84.16 ± 0.35 ^a
10	91.85 ± 0.93 ^a	0.36 ± 0.010 ^b	6.99 ± 0.55 ^a	7.00 ± 0.55 ^a	87.06 ± 0.57 ^b
15	94.03 ± 0.13 ^b	0.62 ± 0.05 ^a	7.03 ± 0.22 ^a	7.06 ± 0.22 ^a	84.96 ± 0.22 ^a

Mean values ± standard deviation of triplicate determinations. Mean values in the same column followed by different letters are significantly ($p < 0.05$) different; L*: Lightness; a*: Redness, b*: Yellowness; C*: Chroma, h°: Hue angle

the change in maltodextrin concentration. Table 3.6 shows descriptive statistics for BGNMP from compartment CH. The lightness, redness, yellowness, chroma and hue angle mean value of 10 and 15% BGNMP ranged from 86.63 to 90.93, 1.07 to 1.31, 0.11 to 12.21, 11.47 to 12.26 and 84.23 to 85.00, respectively. The lightness, redness, yellowness, chroma and hue angle of BGNMP concentrated with 10 and 15% found in compartment CH were significantly ($p < 0.05$) different.

BGNMP concentrated with 15% from both compartments CY and CH had a greater lightness than 5 and 10%. Increase in maltodextrin concentration had a significant increase in lightness, as the values were much closer to 100, which suggest lightness. The a^* and b^* of BGNMP were positive, which suggested that BGNMP is in the redness and yellowness colour space. The hue angles of the samples further suggested that the BGNMP is dominated by a yellowish colour as they are close to a hue-angle of 90° , which represents pure yellowness.

Table 3.7 shows descriptive analysis of the effect of dryer compartments on the colour of BGNMP. The lightness, redness, yellowness, chroma and hue angle of BGNM powder from dryer compartment CY was 92.62, 0.59, 7.21, 7.2 and 85.39, respectively and that of CH was 88.78, 1.19, 11.80, 11.86 and 84.23, respectively. The dryer compartments had a significant ($p < 0.05$) effect on the lightness, redness, yellowness, chroma and hue angle of BGNMP. BGNMP collected in compartment CY had greater lightness and higher hue angle than that collected in CH. The increase in maltodextrin concentration resulted in high lightness resolution of BGNMP. This trend can be justified by what was reported by Przetaczek-Roznowska *et al.* (2014) stating that maltodextrin subjected to thermal treatment results in desirable golden colour, due to caramelisation of short-chained fragment, which then can be perceived as lightness.

Colour differences between BGNMP

The colour differences (ΔE) between BGNMP samples with the different compartments taken into consideration ranged from 0.76 to 5.99. Colour difference ($\Delta E < 1$) can be defined as a not noticeable difference, where the observer does not notice the difference. Colour difference ($\Delta E = 1$) can be defined as a just noticeable difference (JND). Colour difference in the range of 4-8 is perceivable but accepted (Sharma, 2004), which entails that an observer notices the colour difference, and is considered acceptable.

The colour difference between BGNMP samples found in the cyclone concentrated with 5 and 10% maltodextrin were acceptable as they were < 1 . Colour difference between BGNMP with 10 and 15% maltodextrin and 5 and 15% found in the cyclone were also acceptable, though the difference was perceivable, as they had a ΔE value of 2.19 and 2.13 respectively.

Table 3.6 Chamber and maltodextrin effect on the colour of BGNM powder

Maltodextrin concentration						
%	L*	a*	b*	C*	h°	
10	86.63 ± 0.34 ^a	1.07 ± 0.06 ^a	12.21 ± 0.16 ^a	12.26 ± 0.16 ^a	85.00 ± 0.20 ^a	
15	90.93 ± 0.04 ^b	1.31 ± 0.02 ^b	11.39 ± 0.05 ^b	11.47 ± 0.05 ^b	84.23 ± 0.86 ^b	

Mean values ± standard deviation of triplicate determinations. Mean values in the same column followed by different letters are significantly ($p < 0.05$) different; L*: Lightness, a*: Redness, b*: Yellowness, C*: Chroma, h°: Hue angle

Table 3.7 Dryer Compartments Effect on the colour of BGNM powder

Dryer compartments						
	L*	a*	b*	C*	h°	
CY	92.62 ± 1.20 ^a	0.59 ± 0.20 ^a	7.21 ± 0.46 ^a	7.2 ± 0.47 ^a	85.39 ± 1.34 ^a	
CH	88.78 ± 2.37 ^b	1.19 ± 0.14 ^b	11.80 ± 0.46 ^b	11.86 ± 0.45 ^b	84.23 ± 0.86 ^b	

Mean values ± standard deviation of triplicate determinations. Mean values in the same column followed by different letters are significantly ($p < 0.05$) different; L: Lightness; a*: Redness; b*: Yellowness; C*: Chroma, h°: Hue angle

The colour difference between BGNMP found in the chamber concentrated with 10 and 15% maltodextrin was deemed perceivable and acceptable, with the value of 4.38.

Colour difference between the two dryer compartments (CY and CH) was considered perceivable and acceptable with colour difference of 5.99. Although the colour difference of the BGNMP samples from different dryer compartments was considered acceptable, only one BGNMP (10%) sample was preferred and selected. The colour differences of BGNMP samples are summarized in Table 3.8. It can be safely stated that the colour appearance of the produced BGNMP is acceptable.

3 *Water activity characteristics of BGNMP*

Water activity (a_w) of BGNMP concentrated with maltodextrin (5, 10 and 15%) is depicted in Table 3.9. The a_w of BGNMP concentrated with 5, 10 and 15% from dryer compartment; cyclone and chamber ranged from 0.45 to 0.57. The effect of dryer compartments; cyclone and chamber with respect to a_w of BGNMP concentrated with 10% maltodextrin, showed a significant ($p < 0.05$) difference. There was no significant ($p > 0.05$) difference with regards to a_w of BGNMP concentrated with 15% maltodextrin found in the cyclone and chamber.

The a_w of a food can be described as the vitality of water content in foods available for microbial growth (Fontana & Cambell, 2004). Water activity is amongst one of the most critical factors that control rate of food spoilage (Roos, 2001). Water in a food product exists in two forms namely; free water and bound water. Free water can be defined as the water that can be exhibited by pressing the food, which exist in pores of the food material. Bound water has been defined as water that remains unchanged, when a food is subjected to heat treatment, it exhibits no vapour and cannot be frozen below 0 (Park, 2008).

Food deterioration due to microbial growth (yeast and moulds to pathogens) occurs at a_w range of 0.6 to 1.0 (Roos, 2001; Fontana & Cambell, 2004). However, some enzymatic reactions, such as browning transpire at a_w range of 0.3 to 1.0 and increases rapidly at a_w of 0.6 to 0.8. BGNMP concentrated with 10% maltodextrin found in the chamber had the lowest a_w of 0.45 and BGNMP concentrated with 15% found in the chamber had the highest a_w of 0.57. All BGNMP had a_w activity less than 0.6, therefore, microbial spoilage of BGNMP may not be rapid.

3.8.3 Thermal properties of BGNMP

1 Glass transition temperature characteristics of BGNMP

The descriptive analysis for glass transition temperature measurements of BGNMP with different concentrations of maltodextrin (5, 10 and 15%) found in the cyclone is given in Table 3.10.

Table 3.8 Colour difference of dryer compartments and BGNMP samples concentrated with maltodextrin

Maltodextrin concentration %	ΔE	
	CY	CH
5-10	0.76	ND
5-15	2.13	ND
10-15	2.19	4.38

CY: Cyclone; CH: Chamber; ΔE : Colour difference; ND: not detected (There was no BGNMP concentrated with 5% maltodextrin obtained from the chamber).

Table 3.9 Maltodextrin and dryer compartment effect on the water activity (a_w) of Bambara groundnut milk powder

BGNMP Concentration (%)	Dryer compartments	
	CY	CH
	a_w	
5	0.56 ± 0.19	ND
10	0.54 ± 0.09 ^a	0.45 ± 0.08 ^b
15	0.57 ± 0.06 ^a	0.57 ± 0.01 ^a

Mean values ± standard deviation of triplicate determinations. Mean values in the same row followed by different letters are significantly ($p < 0.05$) different; CY: Cyclone; CH: Chamber; BGNMP: Bambara groundnut milk powder, a_w : water activity; ND: Not detected (There was no BGNMP concentrated with 5% maltodextrin obtained from the chamber)

Table 3.10 Cyclone and maltodextrin effect on the glass transition temperature measurements of BGNMP

Maltodextrin concentration (%)	Onset temperature (°C)	Glass transition temperature (°C)	Endset temperature (°C)	ΔC_p (J g⁻¹ K⁻¹)
5	45.02 ± 3.24 ^b	56.50 ± 1.57 ^b	73.75 ± 1.60 ^b	2.07 ± 0.29 ^a
10	39.27 ± 2.07 ^a	50.40 ± 2.14 ^a	66.22 ± 1.19 ^a	2.75 ± 0.28 ^b
15	47.54 ± 1.80 ^b	59.43 ± 2.61 ^b	75.36 ± 0.86 ^b	2.05 ± 0.12 ^a

Mean values ± standard deviation of triplicate determinations. Mean values in the same column followed by different letters are significantly ($p < 0.05$) different; BGNMP: Bambara groundnut milk powder

The onset temperature, glass transition temperature (T_g), endset temperature and heat capacity values for BGNMP found in the cyclone concentrated with 5% maltodextrin were 45.02°C, 56.50°C, 73.75°C and 2.07 J g⁻¹, 10% (39.27°C, 50.40°C, 66.22°C and 2.75 J g⁻¹), 15% (47.54°C, 59.43°C, 75.36°C and 2.05 J g⁻¹), respectively. BGNMP found in the cyclone with the highest maltodextrin concentration (15%) had the highest T_g (59.43). There was no significant ($p > 0.05$) difference between the BGNMP concentrated with 5 and 15% maltodextrin with respect to T_g . However, the effect of 10% maltodextrin on BGNMP showed a significant ($p < 0.05$) difference compared to BGNMP concentrated with 5 % and 15 %. The effect of maltodextrin with respect to T_g values of BGNMP found in the chamber is detailed in Table 3.11. The onset, glass transition, endset temperatures and heat capacity of BGNMP with maltodextrin concentration of 10% and 15% found in the chamber was 39.40°C, 49.62°C, 63.23°C, 1.36 J g⁻¹ and 45.39°C, 56.47°C, 72.92°C, 2.07 J g⁻¹ respectively. The effect of maltodextrin (10%, 15%) differed significantly ($p < 0.05$) in onset and endset temperature measurements.

Nevertheless, the effect of maltodextrin (10 and 15%) did not differ significantly ($p > 0.05$) with respect to the T_g of BGNMP. BGNMP found in the chamber with the highest maltodextrin concentration (15%) had the highest T_g of 56.47°C. Table 3.12 is a depiction of glass transition temperature measurement of BGNMP comparing the effect of dryer compartments on the T_g values of BGNMP. The onset, glass transition, endset temperatures and heat capacity of BGNMP for CY and CH were 43.95°C, 55.11°C, 71.77°C and 2.28 J g⁻¹ and 42.39°C, 53.47°C, 68.07°C and 1.71 J g⁻¹, respectively. The effect of dryer compartments showed a significant ($p < 0.05$) difference with respect to glass transition temperature measurement of BGNMP. Dryer compartment CY showed the highest T_g of 55.11°C.

Glass transition temperature can be defined as the temperature at which amorphous system changes from a glassy state to a rubber state (Tonon *et al.*, 2009; Abbas *et al.*, 2010; Yousefi *et al.*, 2011). Glass transition mainly occurs amongst amorphous food powders, causing stickiness and caking problems (Foster *et al.*, 2006). These changes usually arise when a food powder is exposed to temperatures above the powder's T_g value, induced by water activity and moisture content of the powder (Foster *et al.*, 2006; Shrestha *et al.*, 2007b). BGNMP contains a high amount of maltodextrin which has been reported to be an amorphous glucose polymer derived from starch (Grenby & Mistry, 2000; Descamps *et al.*, 2013). Although BGNMP contains maltodextrin which is reported to be amorphous, it unlikely to undergo the glass transition phenomena. This can be confirmed by the high T_g values (50.40 to 59.43°C) of BGNMP.

Table 3.11 Chamber and maltodextrin effect on glass transition temperature measurements of BGNMP

Maltodextrin concentration (%)	Onset temperature (°C)	Glass transition temperature (°C)	Endset temperature (°C)	ΔC_p (J g⁻¹ K⁻¹)
10	39.40 ± 1.98 ^a	49.62 ± 3.54 ^a	63.23 ± 2.86 ^a	1.36 ± 0.48 ^a
15	45.39 ± 2.88 ^b	56.47 ± 3.54 ^a	72.92 ± 1.88 ^b	2.07 ± 0.16 ^a

Mean values ± standard deviation of triplicate determinations. Mean values in the same column followed by different letters are significantly ($p < 0.05$) different; BGNMP: Bambara groundnut milk powder

Table 3.12 Effect of dryer compartments on the glass transition temperature measurements of BGNMP

Dryer compartment	Onset temperature (°C)	Glass transition temperature (°C)	Endset temperature (°C)	ΔC_p (J g⁻¹ K⁻¹)
CY	43.95 ± 4.24 ^a	55.11 ± 4.33 ^a	71.77 ± 4.36 ^a	2.28 ± 0.40 ^a
CH	42.39 ± 3.95 ^b	53.47 ± 3.54 ^b	68.07 ± 5.73 ^b	1.71 ± 0.50 ^b

Mean values ± standard deviation of triplicate determinations. Mean values in the same column followed by different letters are significantly ($p < 0.05$) different; BGNMP: Bambara groundnut milk powder; CY: Cyclone; CH: Chamber

3.8.4 Chemical composition of Bambara groundnut milk and Bambara groundnut milk powder

The term “milk” refers to cow’s milk. This definition excludes vegetable plant based milks such as “soy milk, almond milk and or peanut milk”. Plant based milk is referred to as a beverage according to its origin, for example “soy beverage”. Milk is generally considered an important source of nutrients, mainly protein (Pereira, 2014). The moisture (BGNM: 83.9; BGNMP: 8.5), ash (BGNM: 0.3; BGNMP: 2.1), total fat (BGNM: 0.3; BGNMP: 1.6), protein (BGNM: 0.9; BGNMP: 7.6), total dietary fibre (BGNM: 2.7; BGNMP: 3.4), total sugars (BGNM: 0.5; BGNMP: 4.5), carbohydrates (BGNM: 11.4; BGNMP: 72.4) and energy (BGNM: 219; BGNMP: 1419) are shown in Table 3.13. The chemical composition of the BGNMP and BGNM were significantly ($p < 0.05$) different except for the total dietary fibre. BGNM disclosed a higher moisture content of 83.9% significantly higher than BGNMP, this is because it contains high amount of water as it is in its liquid form.

This result is in agreement with other plant based milks, reported by Barros de Albuquerque *et al.* (2015) stated that peanut milk holds moisture content of 90.4% which is in the same range. It was observed that the moisture content of BGNMP was above that for standard milk powders (3-3.8%) for both skim and whole milk powder (Baldwin & Pearce, 2005). The elevated moisture content may be linked to the maltodextrin hygroscopic property. The high moisture content of BGNMP (8.5%), may adversely affect the shelf life of powders, however this may not be the case for BGNMP, as it has been found to hold a water activity of 0.45-0.54 [Section 3.8.2.(3), page 72]. The total fat, ash content, protein content, total sugars, carbohydrate and energy of BGNMP were significantly ($p < 0.05$) higher compared to that of BGNM. . This significant difference may be due to the fact that BGNM is in its diluted form, with high water content. Therefore, the present milk nutrients will be diluted.

BGNMP is expected to show a slight increase in the milk nutrients, because in dry weight basis, the nutrients are likely to be more concentrated. This is in agreement with the statement reported by Shrestha *et al.* (2007b), announcing that spray dried milk solutions, which involves rapid removal of moisture leads to concentration of various milk components (fat, protein, minerals). This is due to the migration of the milk components toward the surface, replacing the aqueous phase, therefore explains increase of BGNMP protein (7.6%). The BGNM protein of 0.9% was found to be lower than some plant based milk such as peanut and soy milk of 3.7% and 3.6%, respectively (Isanga & Zhang, 2009; Odo, 2003). However, it was comparable to that of hazelnut milk, which is 0.65% (Bernat *et al.*, 2014a). The fibre content of BGNM (2.7%) was found to be high compared to almond and hazelnut milk of 0.58% and 0.48%, respectively (Bernat *et al.*, 2014b). However the authors also reported the chemical composition of commercial vegetable milk, almond milk beverage

Table 3.13 Chemical composition (g /100 g product) of Bambara groundnut milk and Bambara groundnut milk powder

Proximate (%)	BGNM	BGNMP
Moisture	83.9 ± 0.01 ^a	8.5 ± 0.02 ^b
Ash	0.3 ± 0.01 ^a	2.1 ± 0.01 ^b
Total fat	0.3 ± 0.01 ^a	1.6 ± 0.01 ^b
Sat fats	0.1 ± 0.00 ^a	0.5 ± 0.01 ^b
Mono fats	0.1 ± 0.00 ^a	0.4 ± 0.01 ^b
Poly fats	0.1 ± 0.01 ^a	0.7 ± 0.00 ^b
Protein	0.9 ± 0.04 ^a	7.6 ± 0.02 ^b
Total dietary fibre	2.7 ± 0.38 ^a	3.4 ± 0.03 ^a
Carbohydrates	11.4 ± 0.23 ^a	72.4 ± 0.13 ^b
Of which total sugars	0.5 ± 0.19 ^a	4.5 ± 0.07 ^b
Energy Kj	219 ± 3.34 ^a	1419 ± 0.88 ^b

Mean values ± standard deviation of triplicate determinations. Mean values in the same row followed by different letters are significantly ($p < 0.05$) different, BGNMP: Bambara groundnut milk powder, BGNM: Bambara groundnut milk

to have dietary fibre of 0.4-2% expressed as 100 mL of liquid product (Bernat *et al.*, 2014a), comparable to that of BGNM (2.7%). Almond and chestnut milk commercial vegetable milk expressed as 100ml of liquid product were reported have 0.1-1.6% and 0.3% of total sugars respectively (Bernat *et al.*, 2014a), which again can be seen as comparable to BGNM (0.5%).

BGNM and BGNMP can be categorised as a low fat diet food having total fat of 0.3% and 1.6%, respectively. Saturated fats were identified in both BGNM (0.1%) and BGNMP (0.5%). Saturated fats are reported to be mainly sourced from animal foods, including red meat, poultry and full fat dairy products (CCI, 2007). However, rapeseed oil and sunflower oil were also reported to contain low levels of saturated fats (Atkins, 2011). Mert & Demirkesen (2016) also reported canola oil to contain 7.9% of saturated fats. One of the ingredients used in the development of BGNM and BGNMP was sunflower lecithin powder. Therefore, this can be used to explain the saturated fats identified in BGNM and BGNMP. Saturated fats have also been reported to elevate cholesterol levels however, only the ones mainly from whole dairy products (Miller *et al.*, 2000). In addition, based on the results BGNM has the potential of being acceptable vegetable milk.

3.9 Conclusion

BGNMP was successfully produced from BGNM by increasing the total solids using gluten free maltodextrin and spray drying it. BGNMP was collected from two different compartments namely; the chamber and the cyclone. Ordinarily the final powder to undergo analysis would be collected from the cyclone. However, both final powders found in chamber and cyclone were evaluated to investigate if they were significantly different. The functional attributes of BGNMP were optimal; they were in range with existing powders. BGNMP had fairly acceptable hydration properties (wettability, water solubility and water absorption index). Most importantly the solubility of both BGNMP from the different compartments was the same. High bulk density of BGNMP (0.67-0.69 g/mL) made it comparable to commercial milk powders (0.44-0.88 g/mL). BGNMP was a source of fibre (3.4%) and high in of protein (7.6%) considering it was not fortified with any protein powder concentrate powder. The moisture content of BGNMP was much higher compared to commercial dairy milk powders; however the water activity range (0.4-0.5) made BGNMP potential stable milk. The high moisture content may be due to the use of maltodextrin, which is known to be hygroscopic. Therefore, it is recommended that the drying carrier used to increase product solids to be carefully considered and monitored.

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CHAPTER 4

OPTIMISATION OF FOAM-MAT DRIED BAMBARA GROUNDNUT CULTURED DRINK (YOGHURT) POWDER PRODUCTION

Abstract

Bambara groundnut cultured drink (BGNCD) was prepared using Bambara groundnut (BGN) milk. BGNM was inoculated with yoghurt culture (*Lactobacillus bulgaricus* and *Streptococcus thermophilus*) (0.01% in 100 mL), after being heated to 45°C and was incubated at 35°C for 24 h to produce BGNCD. BGNCD was subjected to physicochemical, microbiological and rheological analysis. BGNCD nutrients composition includes total dietary fibre (3.6%), low fat (0.3%) and protein (1.2%). Titratable acidity and pH levels were 0.38% and 4.47, respectively. BGNCD lightness (L^*), redness (a^*), yellowness (b^*), chroma (c^*) and hue angle (h°) were 75.06, 2.29, 18.35, 18.50 and 82.89°; respectively. BGNCD showed no presence of pathogens (*E. coli*, and *S. aureus*) and had acceptable aerobic counts (3.56 log cfu/mL) as well as acceptable spoilage yeast and moulds (2.26 log cfu/mL). BGNCD exhibited viscoelastic properties that are of a yoghurt drink with $G' > G''$ and apparent viscosity of 0.26 Pa.s⁻¹, with R^2 , K and n values of 0.984, 1.95 and 0.41, respectively. The optimum formulation of gum arabic (6%) and methylcellulose (0.5%) was used to obtain foam-mat dried Bambara groundnut powdered yoghurt (BGNPY) with water solubility index (71.2%) and desirability of 0.956.

4.1 Introduction

Bambara groundnut (*Vigna subterranea*) is a seed crop that is widely cultivated in west and central Africa (Okpuzor *et al.*, 2010). The underutilized legume is mainly grown by women farmers to sustain their families (Mwale *et al.*, 2007). The legume has a variety of different cultivars, differentiated by shape of the hilum on the seed, size and shape of the seed and most importantly the colour of the testa (Linnemann, 1990; Mazahib *et al.*, 2013; Ani *et al.*, 2013). The seed poses multiple uses to the human kind. It can be used in the pharmaceutical industry, in which its oil is extracted for cosmetics, as well as environmental use, where it is used to feed livestock and improve soil fertility, due to its nitrogen and phosphorus content (Enwere & Hung, 1996). It can be used for human consumption at its different stages of maturation.

BGN has been reported in the production of a fermented pro-biotic beverage cultured using lactic acid bacteria (Murevanhema & Jideani, 2014). Traditionally lactic acid bacteria are associated with the preparation of fermented milk products. In addition literature reveals the drying of commercial fermented products using different drying process including the foam-mat drying process (Rajkumar *et al.*, 2007). Foam-mat drying is a simple process of drying liquid or solid foods by being mixed with a foaming agent to produce stable foam,

which undergoes air drying temperatures ranging from 50-80°C (Kandasamy *et al.*, 2012; Febrianto *et al.*, 2012). The drying process can be used to dry different foods such as, milk and beverages (Widyastuti & Srianta, 2011) including that from BGN.

A growing awareness of vegetable proteins such as BGN proteins, low in cholesterol has been established due to individuals who are lactose intolerant. In addition, fermented and non-fermented dairy products are often priced too high, for the low income earners (Isanga & Zhang, 2009). Therefore, there is a growing demand for affordable plant based products as an alternative to dairy products. BGN is a valuable crop and an increase in its consumption may improve protein intake and availability, considering its nutritive content. Feasibility of the production of BGNM has been studied by Brough *et al.* (1993) and Murevanhema & Jideani. (2012). Murevanhema & Jideani (2012) reported the shelf life stability of Bambara groundnut probiotic beverage (BGNPB) produced from four strains of *Lactobacillus*. However, not much has been reported on BGN powdered yoghurt. The objective of this study was to produce Bambara groundnut cultured drink (BGNCD) and further evaluate the physicochemical, rheological and microbiological attributes for the optimization of Bambara groundnut powdered yoghurt (BGNPY) production. For the sake of clarity in this chapter “milk” extracted from Bambara groundnut and cultured with yoghurt microflora will be referred to as BGNCD. Foam-mat dried BGNCD will be referred to as Bambara groundnut powdered yoghurt (BGNPY).

4.2 Materials and Methods

4.2.1 Source of the materials and equipment

The overview of the methodology employed in this chapter is shown in Figure 4.1. The BGN seeds were purchased from Trio trade, Johannesburg, South Africa. The yoghurt starter culture was purchased from Danisco, Cape Town and flavourants were obtained from Creative Flavours International, Cape Town. The total dietary fibre enzyme assay kit was purchased from Sigma-Aldrich, Johannesburg. Equipment used for the experiments was obtained from the Food Technology Department of Cape Peninsula University of Technology.

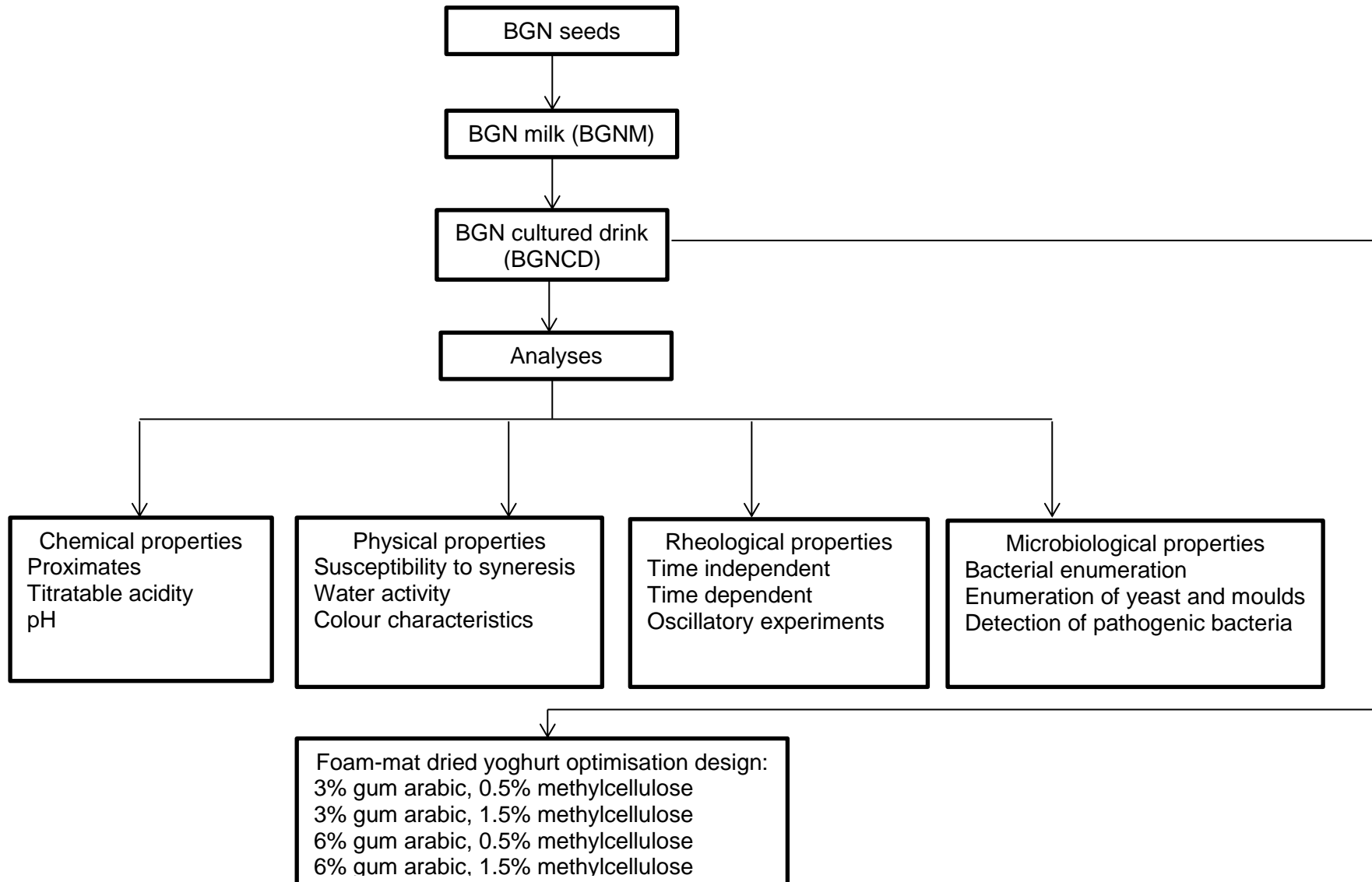


Figure 4.1

Chapter 4 methodology overview.

4.2.2 Production of Bambara groundnut cultured drink (BGNCD)

BGNM (400 mL) in 500 mL schott bottles was warmed to 45°C in a water bath for 15 to 20 min. The BGNCD was then aseptically inoculated with normal yoghurt culture (*Lactobacillus bulgaricus* and *Streptococcus thermophilus*) (0.01% in 100 mL) and incubated at 35°C using a water bath for 24 h. The fermented BGNM was immediately cooled in ice and stored under refrigeration at 3-4°C (Murevanhema & Jideani, 2012).

4.3 Chemical Analysis of Bambara Groundnut Cultured Drink

4.3.1 Proximate analysis of BGN cultured drink

Protein determination was done using Kjeldahl (N X 6.25), total fat according to AOAC (2005) method 996.06, total dietary fibre according to AOAC (1997) method 985.29, total sugar according to AOAC (2003) method 982.14, moisture and ash according to AOAC (2005) method 934.01 and 923.03. The carbohydrate was determined by difference and energy was determined according to Anon (2010).

4.3.2 Determination of pH and titratable acidity of BGN cultured drink

The pH of BGN cultured drink (BGNCD) was measured at room temperature (21 ± 2°C) using the Crison GLP 21 pH-Meter, Barcelona equipped with a glass electrode. The glass electrode was calibrated using buffer 7, 4 and 9 (Sanni *et al.*, 1999). BGNCD (9 mL) was transferred into a conical flask and 0.5 mL of phenolphthalein indicator solution (1%) was added. BGNCD was then titrated against standardised 0.1 M NaOH solution until a faint pink that persisted for 30 s appeared. Titratable acidity was calculated using equation 4.1 and was expressed as % lactic acid (CH₃-CHOH-COOH, MW = 90) (Olugbuyiro & Oseh, 2011).

$$TTA\% = \frac{\text{Titre value} \times M \times 90 \times 100}{\text{Volume of sample} \times 1000} \dots\dots\dots \text{Equation 4.1}$$

Where:

M = Molarity of 0.1 NaOH

90 = Molecular weight of lactic acid (CH₃-CHOH-COOH)

100 = Conversion factor to %

1000 = Conversion factor from milliliters to litres

4.4 Physical Analysis of Bambara Groundnut Cultured Drink

4.4.1 Measuring syneresis of the BGN cultured drink

After the cultured BGN drink was stored for 2 days at refrigeration temperatures (4°C ± 1), BGNCD susceptibility to syneresis (STS) was measured at room temperature (21 ± 2°C) by filtering 100 mL of BGNCD through MN 1674 Machinery-Nagel filter paper supported on a funnel. The contents on the filter paper were allowed to drain for 6 h and the whey collected

was measured and used as an index of syneresis (Isanga & Zhang, 2009). Analysis was carried out in triplicate. The following equation was used to calculate STS

$$\text{STS \%} = \frac{V_1}{V_2} \times 100 \dots \dots \dots \text{equation 4.2}$$

V_1 = Volume of BGN cultured drink whey collected after drainage

V_2 = Volume of BGN cultured drink

4.4.2 Water activity analysis of BGN cultured drink

Water activity (a_w) of BGN cultured drink (BGNCD) was measured using the Novasina Ms 1 Set a_w meter, which uses a cell protection filter to measure a_w . Salt humidity standards of 53%, 75% and 90% relative humidity were used to calibrate the measurement cell. BGN drinking yoghurt (10 mL) was transferred into a sample dish and placed inside the novasina analyser and the cell measuring protection filter was immediately closed. The a_w activity reading was observed after 60 to 80 s (Novasina General Catalogue, 2012). The test was performed in triplicate.

4.4.3 Colour measurement of BGN cultured drink

The colour of the BGNCD (2-3 g) was measured using a Konica Minolta Spectrophotometer CM-5 45°/0° standard, set at standard observer 10° and D65. The instrument was zero calibrated using a black tile ($L^* = 5.49$, $a^* = -7.08$, $b^* = 4.66$ and white tile ($L^* = 93.41$, $a^* = -1.18$, $b^* = 0.75$). BGNCD samples were placed in a clear coloured sample holder and reflectance measured for $L^*a^*b^*$ and LCh colour scales. The L^* coordinate is lightness, L^* values closer to 100 represents whiteness and closer to 0 is a representation of blackness. Coordinate a^* refers to green (-)/red (+) chromaticity and coordinate b^* refers to blue (-)/yellow (+) chromaticity (Bezerra *et al.*, 2013). Measurements for each sample were performed in triplicate at three different positions in the samples (one reading = average of three readings per rotated position), with the results recorded in L^* (lightness), a^* (chromaticity coordinate $+a^* = \text{red}$ and $-a^* = \text{green}$), b^* (chromaticity coordinate $+b^* = \text{yellow}$ and $-b^* = \text{blue}$), C^* (chroma) and h (hue angle $0^\circ = +a^*$, $90^\circ = +b^*$, $180^\circ = -a^*$ and $270^\circ = -b^*$).

4.5 Rheological Assessment of BGN Cultured Drink

The apparent viscosity of BGNCD was measured using an MCR 300 Paar Physical Rheometer (Discovery HR-1, Hybrid Rheometer). BGNCD was produced and stored under refrigeration at 4°C overnight, prior to analysis. The time independent, time dependent and

viscoelastic properties of BGNCD were then evaluated as described in section 4.5.1 and 4.5.2. All measurements were carried out in duplicate at 4°C.

4.5.1 Time independent rheological analyses

The Gauche *et al.* (2009) method was used to measure the flow behaviour of BGN cultured drink with slight modifications. BGNCD (25 mL) was carefully transferred into the rheometer measuring cup and allowed to equilibrate for 5 min. The change in apparent viscosity was measured as a function of increasing shear rate from 0.0 to 1000.0 s⁻¹. The analysis was set to run for a period of 300 s. The flow characterisation of the system was modelled by using the power law equation described as follows.

$$\tau = K\gamma^n \dots\dots\dots \text{equation 4.3}$$

Where (τ = shear stress), K = consistency efficiency, γ = shear rate and n = flow behaviour index which indicates the tendency of a fluid to shear thinning. Using this model, a graph of logarithm of shear stress against logarithm of shear rate was plotted to yield a straight line with a slope of $\tau = K\gamma^n$ and an intercept of $\log K$ (consistency index which serves as the viscosity index of the system).

4.5.2 Time dependent rheological analyses

The Gauche *et al.* (2009) method was employed to measure the flow of BGN cultured drink with slight modification BGNCD (25 mL) was carefully transferred into rheometer measuring cup and allowed to equilibrate for 5 min. The apparent viscosity was measured as a function of time from 0 to 300 s with constant shear rate of 50 s⁻¹, simulating the mouth.

4.5.3 Oscillatory analyses of BGN cultured drink

Oscillation amplitude sweep

BGNCD (25 mL) was transferred into a rheometer measuring cup and was allowed to equilibrate for 5 min. For amplitude sweep the strain was set at 0.01 to 12.03% at a constant angular frequency of 10.0 rad/s. To determine linear viscoelastic region, storage and loss moduli were plotted against frequency. Oscillation amplitude sweep describes the deformation strain for further analyses of frequency sweep (Guggisberg *et al.*, 2007).

Oscillation frequency sweep

BGNCD (25 mL) was carefully transferred into a rheometer cup. For frequency sweep a constant strain of 0.1% was adjusted at an angular frequency of 100.0 to 0.1 rad/s. The storage and loss moduli were plotted against angular frequency in order to determine the linear viscoelastic region of BGNCD (Guggisberg *et al.* 2007).

4.6 Microbiological Population in BGN Cultured Drink

The pour plate method was employed for the enumeration of microorganisms in BGN cultured drink. Counting of all typical colonies was performed using colony counter and a control was carried out for each analysis. All measurements were performed in triplicates. Only plates containing colonies from 25-250 were counted.

4.6.1 Bacterial enumeration in BGN cultured drink

BGN cultured drink (10 g) was weighed into a 90 mL sterile ringers' solution (10^{-1}) and mixed well. Then a series of dilutions (10^{-1} to 10^{-6}) were prepared by first transferring 1 mL aliquot of the 100 mL sample solution into a 9 mL sterile ringers' solution as the second dilution (10^{-2}), until last dilution (10^{-6}). For each dilution 1 mL aliquot was carefully and aseptically transferred into the base of a labelled sterile Petri-dish. Then separately, approximately 15 mL of pre-cooled plate count agar (PCA) and violet red blood agar (VRBA) was poured for the enumeration of mesophilic counts, and carefully swirled to mix well. Once all plates were allowed to solidify, they were incubated in an inverted position. PCA and VRBA plates were incubated at 37°C for 48 (Baylis, 2007).

4.6.2 Enumeration of yeast and moulds in BGN cultured drink

A series of dilutions for BGNCD were prepared as described in 4.6.1. Then approximately 15 mL of pre-cooled rose bengal chloramphenicol agar (RBCA) was poured into each petri-plate dish and mixed by gently swirling for the enumeration of yeast and moulds. Once solidified, plates were incubated at 25°C for 5 days in an inverted position (Baylis, 2007).

4.6.3 Detection of presumptive *Escherichia coli* species on Bambara groundnut cultured drink

The Da Silva *et al.* (2013) method was used for the detection of *E. coli* with slight modification. The detection of *Escherichia coli* (*E. coli*) was performed by employing the indole test. BGN cultured drink (1 g) was transferred into sterile 9 mL tryptone soya broth (TSB) to prepare an enrichment and was incubated at 37°C for 24 h. From the sample enrichment, 1 mL was aseptically transferred into a sterile 5 mL brilliant green bile broth (BGLB) containing a Durham tube positioned downward. From the BGLB mixture, 0.1 mL was transferred into a sterile 5 mL tryptone water prepared solution and both tubes containing BGLB and tryptone water mixtures were incubated at 44°C for 24 h. Thereafter, the BGLB containing tube was checked for the occurrence of a bubble inside the surface of the Durham tube. The presence of a bubble indicated positive for *E. coli* and absence of a bubble indicated negative for *E. coli*. Further confirmation was employed on occurrence of bubble by the addition of Kovac's reagent into the tube containing tryptone water mixture and

the formation of a red colour ring on the surface of the tryptone water tube after the addition of Kovac's reagent indicated presence of *E. coli*.

4.6.4 Detection of presumptive *Staphylococcus aureus* on Bambara groundnut cultured drink

The Lee *et al.* (2015) method was used with a slight modification employing the direct detection process. BGN cultured drink (1 g) was transferred into sterile 9 mL tryptone soya broth (TSB), mixed by vigorous shaking to make up enrichment and was incubated at 37°C for 24 h. After the incubation period, the sample enrichment was streaked onto a prepared mannitol salt agar (MSA) plate and further incubated for 24 h. Gram-stain was performed only on observed typical *S. aureus* colonies and further confirmed using the staphylase coagulase test for presence of *S. aureus*.

4.7 Optimisation of BGN Powdered Yoghurt Production Using Foam-mat Drying Process

BGN powdered yoghurt (BGNPY) was produced using the Krasaekoopts & Bhatia (2012) method with some modification. Foam-mat drying technique was performed employing four different formulations comprising of gum arabic (GA) and methylcellulose (MC). The independent variables and their levels of 2² factorial design are detailed in Table 4.1.

4.8 Statistical Analysis

Multivariate analysis of variance was used to determine differences between treatments for determining significant effects. The system behaviour was described by a linear interaction factorial regression model (equation 4.3), carried out using Design-Expert software (Trial version 9.06.2, Stat-Ease, Inc.).

$$Y = \beta_0 + \beta_1 + \beta_2 + \beta_{12} \dots \dots \dots \text{equation 4.3}$$

Where Y is the predicted response variable, β_0 is the intercept, β_1 and β_2 are the linear regression coefficients independent variables; gum arabic and methylcellulose, respectively and β_{12} is the interaction of gum arabic and methylcellulose.

The numerical optimisation was carried out in order to establish the values for GA and MC required for optimal foam-mat dried BGN yoghurt powder. The experimental data was fitted using a linear polynomial regression model. The quality of fit of model equation was evaluated using R², adjusted R², adequate precision, f-value and lack of fit. The significance of the regression coefficients was verified by determining the ρ -value.

Table 4.1 Independent variables and quantity used for the 2² factorial design for foam-mat drying BGN cultured drink.

Independent variables		
Treatment runs		
	Gum Arabic	Methylcellulose
1	3	0.5
2	3	1.5
3	6	0.5
4	6	1.5

4.9 Results and Discussion

4.9.1 Chemical properties of Bambara groundnut cultured drink

Chemical properties which were investigated in this study include proximate analysis, pH and titratable acidity parameters of BGNCD.

1 Proximate characteristics of Bambara groundnut cultured drink

The protein, total dietary fibre, ash, moisture, total fat, total sugar, carbohydrate and energy of BGNCD were 1.2%, 3.6%, 0.4%, 82.6%, 0.3%, 0.0% 11.9% and 233.8 kJ, respectively (Table 4.2). The high moisture content of BGNCD (82.6%) can be linked to the use of BGNM with moisture content of 83.9% [Section 3.8.4, page 77] to obtain the yoghurt. Furthermore, the initial processing to obtain BGNM involves the rehydration of BGN flour with water (1:10). These findings were similar to that of normal yoghurt from cow's milk with 88.1% moisture reported by Gauche *et al.* (2009) and peanut milk yoghurt with 78.8% moisture reported by Isanga & Zhang (2009).

The high fibre content (3.6%) can be explained by the high fibre content of the BGN seed ranging from 3.3% to 6.1% reported by Sirivongpaisal (2007); Yusuf *et al.* (2008) and Oyeleke *et al.* (2012). There were no total sugars detected in BGNCD, this can be attributed to the fermentation process, in which the present BGNM sugar (0.5%), [Section 3.8.4, page 77] were utilised by the yoghurt microflora to produce lactic acid (De Barabardere & De Baerdemaeker, 1999; Walstra *et al.*, 2005).

The total fat of BGNCD (0.3%) was visibly lower compared to other reported plant based yoghurts such as yam bean yoghurt (1.1%), peanut milk yoghurt (5.8%), (Amakoromo *et al.*, 2012; Isanga & Zhang, 2009) and in both studies the skin of the seeds were initially removed. The total fat of BGNCD was comparable to soy milk yoghurt (1.5-1.6%) reported by Rinaldoni *et al.* (2012). The low fat content of BGNCD renders it suitable to be low fat diet food. BGNCD protein (1.2%) was slightly lower when compared to other plant-based fermented milk such as, yam bean yoghurt protein (5.4%), and peanut milk yoghurt protein (5.2%) (Isanga & Zhang, 2009; Amakoromo *et al.*, 2012). The highly noticeable difference may be explained by the skimmed milk powder used to fortify peanut and yam bean milk for production of the yoghurt. The BGNCD carbohydrate (11.9%) was comparable to that of yam bean yoghurt (14.24%) reported by Amakoromo *et al.* (2012) however, slightly higher than of peanut milk (8.6%) (Isanga & Zhang, 2009).

2 The pH and titratable acidity of BGN cultured drink

The titratable acidity (TTA) and pH of Bambara groundnut cultured drink (BGNCD) was 0.35% and 4.47, respectively.

Table 4.2 Proximate composition of Bambara groundnut cultured drink.

Nutrients	Proximate (%)
Energy kJ	233.8 ± 1.53
Total fat	0.3 ± 0.02
Of which Saturated	0.1 ± 0.007
Monounsaturated	0.1 ± 0.00
Polyunsaturated	0.1 ± 0.02
Carbohydrates	11.9 ± 0.13
Of which Sugars	0.0 ± 0.00
Total dietary fibre	3.6 ± 0.15
Protein	1.2 ± 0.02
Ash	0.4 ± 0.01
Moisture	82.6 ± 0.01

Acidity and pH are two closely related terminologies, where pH is a scale used to measure the acidity or basicity in a solution, where the scale starts from 1 to 14. The scale of 1-6 represents acids, 7 is neutral and scale from 8-14 represents bases (alkaline). On the other hand acidity gives a qualitative indication of acids present in a solution (Hermansson & Syafii, 2015). The acidity of BGNCD was comparable to other (commercially available) drinking yoghurts such as holianda plain yoghurt, superyogo, holianda strawberry yoghurt and holianda pineapple/coconut yoghurt, with acidity of 0.31%, 0.33, 0.30 and 0.36%, respectively as reported by Olugbuyiro & Oseh (2011).

Cowpea milk yoghurt fermented with *Lactobacillus* species (*Lactobacillus plantarum* and *Lactobacillus acidophilus*) was reported to contain acidity and pH of 0.94% and 3.8, respectively (Sanni *et al.*, 1999). Rinaldoni *et al.* (2012) also reported the acidity range (0.60-0.68%) and pH range (4.6-4.8) for soy yoghurt fermented with normal yoghurt culture (*Lactobacillus delbrueckii subspecies bulgaricus* and *Streptococcus salivarius subspecies thermophilus*). Walstra *et al.* (2005) stated that the preferred yoghurt pH levels should range from 4.1 to 4.6. Furthermore, according to Codex Standards (2003) fermented milk and yoghurt should contain a minimum titratable acidity of 0.3% and 0.6%, respectively. Therefore, BGNCD can be deemed as a suitable fermented beverage.

4.9.2 Physical properties of BGN cultured drink

Physical properties investigated in this chapter include; susceptibility to syneresis, water activity and colour parameters.

1. Syneresis of Bambara groundnut cultured drink

The STS% of BGNCD (60.67%) was quite high. Syneresis is a common defect in fermented milk products; it is the shrinkage of gel, which then leads to whey separation (Sahan *et al.*, 2008; Gauche *et al.*, 2009). Isanga & Zhang (2012) stated that low fat yoghurts tend to have a higher degree of syneresis than high fat containing yoghurts. The high fat content combines with the already present protein in the system allowing the fat globules to participate as a copolymer with the protein to strengthen the gel network of yoghurt and thus reduce syneresis.

The authors further reported the STS% of peanut milk yoghurt (43.27%), with fat content (5.84%). Rinaldoni *et al.* (2012) examined the STS% of soy milk yoghurt (37.6 to 33.33%) with fat content (1.45 to 1.55%). Therefore, the high STS% of BGNCD (60.67%) can be attributed to its low total fat (0.3%) [Section 4.9.1(1), page 94]. Further, suggesting that the degree of syneresis of yoghurt is closely linked to available fats and protein in the system (Isanga & Zhang, 2012). It has been further reported that syneresis can also be induced by other factors such as slight disturbance/agitation during the initial stages of gel formation, wetted surface of yoghurt due to condensed water inside lid container containing

yoghurt and containers made of material to which formed gel does not stick to it (Walstra *et al.*, 2005).

2. *Water activity of Bambara groundnut cultured drink*

The water activity (a_w) of BGNCD was 0.85, which was expected to be as high due to the ratio of total water content of BGN milk used in the production of BGNCD. Water activity is the available water for microbial growth (Fonatana & Cambell, 2004). Hence, BGNCD with high a_w may be susceptible to spoilage microbial activity. However, proper cold refrigeration storage can slow down the bacterial activity, therefore extending the shelf life of BGNCD.

3. *Colour characteristics of Bambara groundnut cultured drink*

The lightness (L^*), redness (a^*), yellowness (b^*), chroma and hue angle of BGNCDY were determined to be 75.06, 2.29, 18.35, 18.50 and 82.89, respectively as depicted in Table 4.3. BGNCD can be declared as light due to the lightness (L^*) value of 75.06, which is close to 100 representing whiteness on the colour scale. The negative (-) a^* on the colour scale describes the colour greenness and a positive (+) a^* value describes the colour redness. The negative (-) b^* value defines the colour blueness and a positive (+) b^* value refers to the colour yellowness (Kneifel, 1992). BGNCD had a positive b^* (18.35) suggesting that BGNCD contained some yellowness and positive a^* (2.29), indicating redness characteristics of BGNCD. This can be attributed to the presence of β -carotene (10 μg) in BGN reported by Dansi *et al.* (2012). β -carotene is a carotenoid, a colour pigment responsible for the red, yellow and orange colour in plants (Fernandes-Garcia *et al.*, 2012). Furthermore, Aparicio & Harwood (2013) also stated that β -carotene contributes to the yellowish colour of foods, specifically vegetable oils.

The hue angle of BGNCD was 82.89° further signifying that the yoghurt is dominated by a yellow colour (Figure 4.1), as it is close to a hue angle of 90°, which indicates pure yellowness on the colour scale. Kumar & Mishra (2004) reported similar findings for mango soya fortified yoghurt, fermented with normal yoghurt bacteria (*Streptococcus thermophilus* and *Lactobacillus delbrueckii subsp. bulgaricus*), with Lightness (L^*) range of 63 to 79, which is comparable to the L^* of BGNCDY (75.06).

4.9.3 Rheological properties of Bambara groundnut cultured drink

1 *Time-independent rheological characteristics (shear dependent apparent viscosity-flow ramp)*

The time independent properties of BGN cultured drink (BGNCD) were investigated by measuring flow ramp shear dependent viscosity against shear rate.

Table 4.3 Colour characteristics of BGN cultured drink.

Colour parameters	Value
Lightness (L*)	75.06 ± 0.14
Redness (a*)	2.29 ± 0.01
Yellowness (b*)	18.35 ± 0.07
Chroma (c*)	18.50 ± 0.07
Hue (h°)	82.89 ± 0.01



Figure 4.2 Bambara groundnut cultured drink

The viscosity of BGNCD is described in the rheogram presented as Figure 4.3. BGNCD showed highest viscosity (0.26 Pas^{-1}) at shear rate (24.67 s^{-1}) and lowest viscosity (0.03 Pa.s) at shear rate (924.9 s^{-1}). Increase in shear rate resulted in a decrease in BGNCD viscosity. This behaviour reveals BGNCD as a non-Newtonian system with pseudoplastic properties. The viscosity (0.26 to 0.03 Pas^{-1}) of BGNCD was similar to that of Aryan yoghurt drink, manufactured in Turkey with milk using expolysaccharide producing cultures, with added viscous curd and salt water having a viscosity of 0.02 to 0.125 Pas^{-1} (Koksoy & Kilic, 2004). Figure 4.4 is a rheogram summarizing the viscosity of BGNCD against shear stress. The apparent viscosity of BGNCD was high (0.26 Pas^{-1}) at 6.37 Pa shear stress and decreased to 0.03 Pas^{-1} with an increased shear stress of 30.34 Pa . These observations once again proves BGNCD to be a shear thinning fluid material that flows more readily as it is stirred or sheared. However, at rest the fluid will behave as a solid, revealing to have Bingham plastic properties.

The mean consistency coefficient (K) and flow behaviour index (n) of BGNCD was 1.9453 and 0.4092, respectively. The coefficient of determination (R^2) was 0.984, which implies a good fitting of the power law model (Figure 4.5). The power law was used to describe the flow of BGNCD, of which gradient of a straight line is the flow behaviour index (n) specifying shear thinning trend of the fluid. The consistency coefficient (K) serves as a viscosity guide of a fluid system, higher values indicate a more viscous fluid (Rinaldoni *et al.*, 2012). Fluid food structures with flow behaviour index (n) below 1 indicate a shear thinning system (Rinaldoni *et al.*, 2012). With BGNCD having flow behaviour index of less than 1, further indicates its shear thinning properties.

Janhoj *et al.* (2008) reported flow characteristics of drinking yoghurt made from reconstituted skim milk powder cultured with lactic acid bacteria and with added hydrocolloid solution. The consistency coefficients (K) ranged from 0.02 to 0.2 Pas^n and flow index behaviour (n) ranged from 0.4 to 0.8 . The consistency coefficient (K) of BGNCD (1.9453 Pas^n) was much higher compared to that of drinking yoghurt reported by Janhoj *et al.* (2008) with 0.2 Pas^n , making BGNCD more viscous. Furthermore, the flow behaviour index (n) of BGNCD (0.4092 .) was comparable to that of drinking yoghurt (0.4 - 0.8) reported by Janhoj *et al.* (2008). Doogh, an Iranian traditionally fermented dairy drink was reported to have consistency coefficient (K) range 2.2 to 2.5 Pas^n (Hasheminya *et al.*, 2013) once again comparable to that of BGNCD (1.9453 Pas^n) Thus, increasing potential of BGNCD as a fermented drinking beverage.

2 Time dependent rheological properties (peak hold flow-apparent viscosity)

Time dependent flow reveals the original viscoelastic or structural properties of a fluid food system.

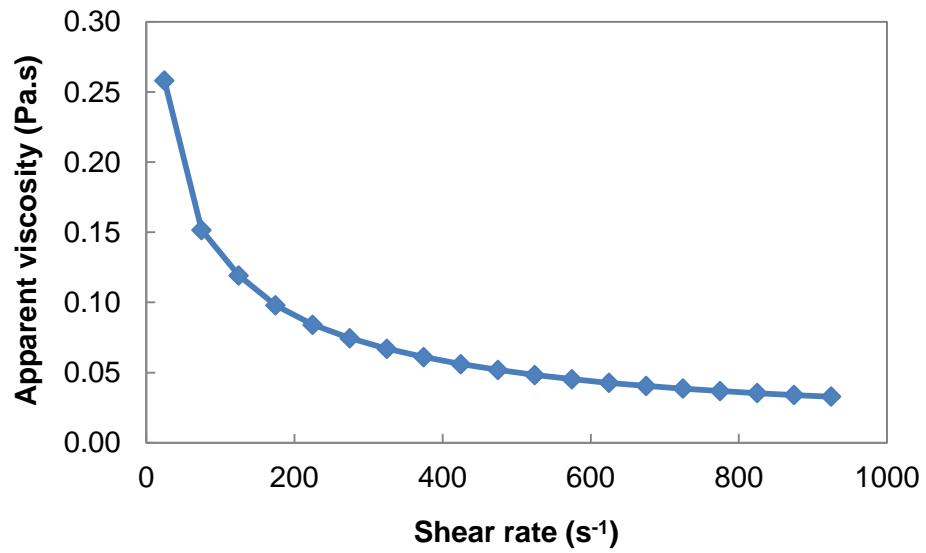


Figure 4.3 Apparent viscosity of Bambara groundnut drinking yoghurt (shear rate as a function)

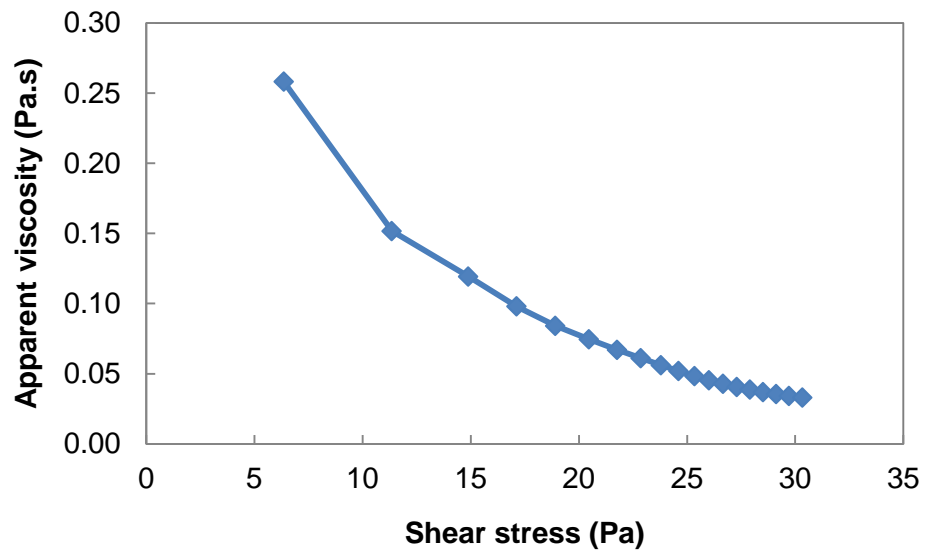


Figure 4.4 Apparent viscosity of Bambara groundnut drinking yoghurt (shear stress as function)

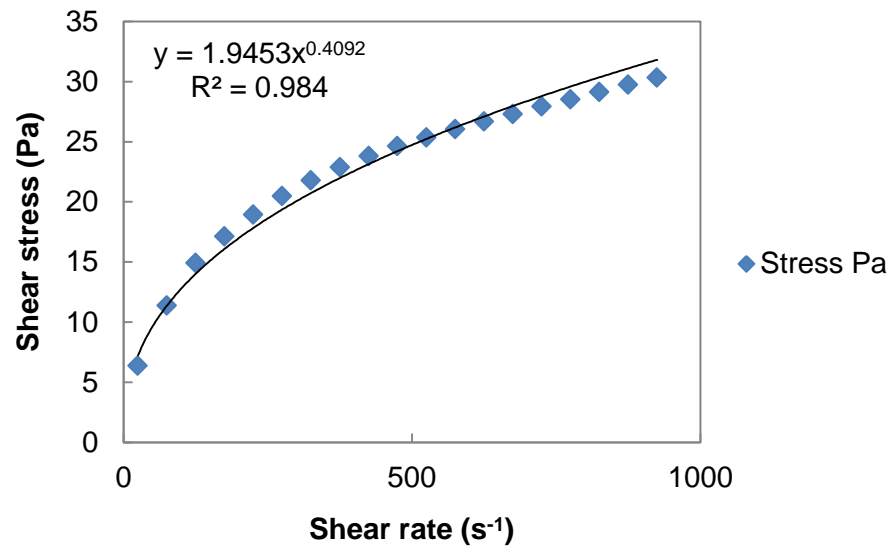


Figure 4.5 Flow behaviour of Bambara groundnut drinking yoghurt using power law model

The apparent viscosity of BGNCD was initially $0.2 \text{ Pa}\cdot\text{s}^{-1}$ at approximately 0 s and gradually decreased to $0.16 \text{ Pa}\cdot\text{s}^{-1}$ at 300 s indicated by Figure 4.6. The viscous properties of BGNCD showed a decreasing effect when shearing period was prolonged at a constant shear rate (50 s^{-1}), which is a simulation of shear rate in the mouth (Koksoy & Kilic, 2004). The above observed trend reveal BGNCD as a thixotropic system, as absence of shear results in higher viscosity and its application results in decreased viscosity.

3 *Oscillatory characteristics/Viscoelastic properties*

Oscillation Amplitude sweep

Oscillatory properties of BGNCD were investigated by measuring amplitude sweep viscoelastic properties against strain. The linear viscoelastic region (LVR) was stable at a strain amplitude of 0.188 for both moduli, elastic/storage modulus (G') and viscous/loss modulus (G'') of BGNCD. This means that the yoghurt can withstand stress of 0.188 Pa going down to 0.01 Pa (Figure 4.7). There was a visible gap between G' and G'' , where G' was greater than G'' ($G' > G''$), implying that the yoghurt was more elastic than it was viscous. Dynamic rheology (oscillation) measures the viscoelastic properties of a material, by applying stress or strain, which simulates the food material. The strain/stress should not be excessive enough to destroy the food structure completely (Qiao *et al.*, 2015). In amplitude sweep moduli are measured as a function of increasing strain, while frequency is fixed (Mehmet & Gunasekaran, 2002). Amplitude sweep was conducted to assess the LVR of BGNCD. In addition, a longer LVR indicates higher stability of BGNCD.

Oscillation Frequency sweep

The elastic/storage modulus (G') was greater than viscous/loss modulus (G''), ($G' > G''$). G' was greater by exactly 17.75 Pa compared to G'' , both at a constant strain of 0.1% (Figure 4.8). BGNCD had greater elastic properties than viscous properties. In frequency sweep, strain of a fixed amplitude is enforced on the BGNCD and the moduli (G' and G'') are evaluated over range of frequency to determine the nature of yoghurt material, resulting in a mechanical spectrum plot. These findings were similar to those of the Doogh yoghurt drink, with $G' > G''$, confirming presence of gel like structure (Azarikia & Abbasi, 2010). Furthermore, the lower the frequency levels food fluid system can withstand, the higher the stability of the food material, as time is defined as 1 over frequency ($t = 1/f$) (Mehmet & Gunasekaran, 2002). Therefore, at all frequencies should (G') be positioned above (G''), this implies that the structure of the system is stable. Thus BGNCD can be deemed as a stable fluid system as ($G' > G''$).

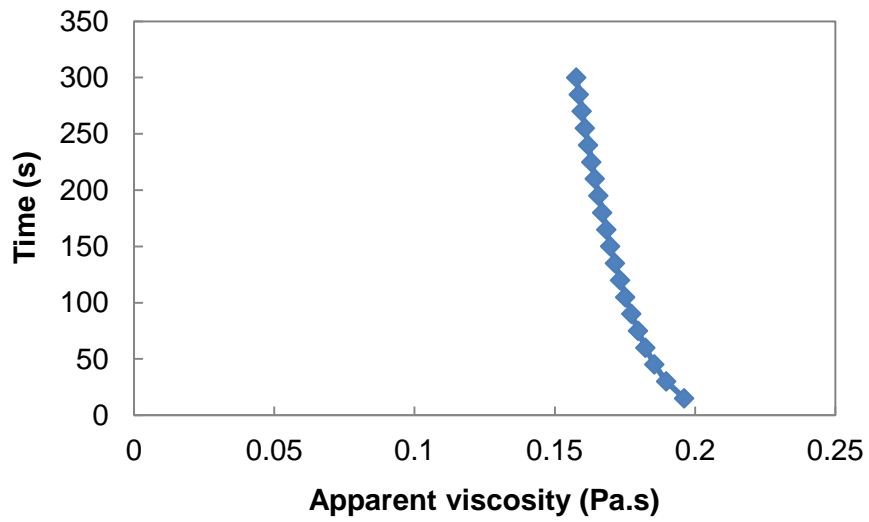


Figure 4.6 Apparent viscosity of Bambara groundnut drinking yoghurt, time as a function-Time dependent

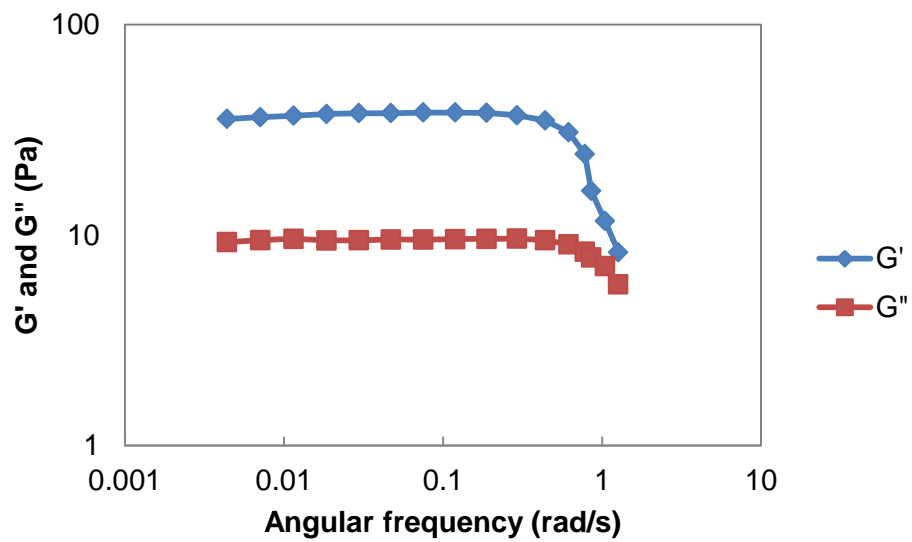


Figure 4.7 Amplitude sweep of Bambara groundnut drinking yoghurt.

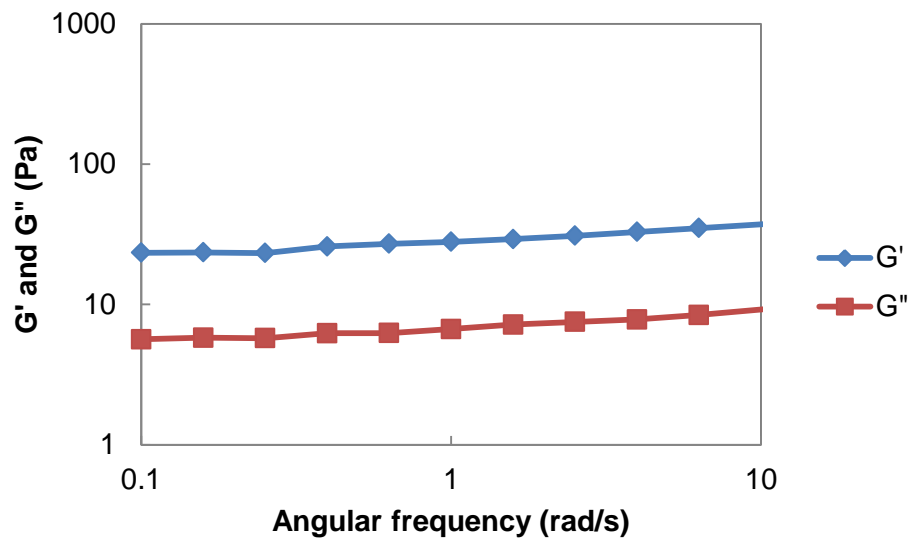


Figure 4.8 Frequency sweep of Bambara groundnut drinking yoghurt

4.9.4 Microbiological Properties of Bambara Groundnut Cultured Drink

1 Total viable counts in Bambara groundnut cultured drink

BGNCD had aerobic microorganism counts of 3.56 log cfu/mL. Total aerobic organisms are commonly used as indicator bacteria to evaluate the quality, safety and shelf life of foods (Odumeru *et al.*, 2002). They can grow and survive under oxygenated environments and are considered to be mesophilic bacteria, as they have wide growth temperature range of 20 to 45°C, but mainly grow optimally at 35 to 37°C (Walstra *et al.*, 2005; Russell, 2010). Excessive growth of total aerobic (spoilage bacteria) may lead to spoilage of BGNCD and restriction of its shelf life, resulting in undesirable sensory characteristics, such as loss of texture, development of off flavours and off colours (Luning & Marcelis, 2009). The department of health directorate (DOHD) recommends a colony count of < 50 000 cfu/g (4.69 log cfu/mL) in yoghurt. Ministry of food and drug safety bureau suggested 40 000 cfu/mL (4.60 log cfu/mL) for ground grain to dairy soy milk products. Thus, making BGNCD safe for consumption as a fermented beverage with only a total colony count of 3.56 log cfu/mL.

2 Yeast and moulds in Bambara groundnut cultured drink

Bacterial growth of spoilage yeast and moulds for BGNCD was 2.26 log cfu/ mL. Yeast and moulds form part of large group fungi and have the ability to produce mycotoxins, which pose a health hazard to humans, they are considered undesirable and a sign of poor hygiene (Gourama, 2005; Gadaga *et al.*, 2009). Walstra *et al.* (2005) stated that most moulds can start to grow at high osmotic pressure, under high acidity or at low water activity conditions. Fungi have a wide growth temperature range of 5 to 45°C, with optimum temperature range (25-28°C). Fungi can also grow within a wide pH range of 3-8.0, but favour an acid pH (pH of yeasts being 4 to 4.5 and moulds can grow from pH 2 to 8.5) (Da Silva *et al.*, 2013).

Spoilage moulds that generally grow on the yoghurt surface includes; *Penicillium*, *Aspergillus*, *Mucor*, *Rhizopus*, *Alternaria*, *Manilia* and *Absidia spp.* These growth conditions of yeast and moulds renders yoghurt and fermented foods a selective environment for their growth including BGNCD with acidity of 0.35% [Section 4.9.1(2), page 94] and a_w 0.85 [Section 4.9.2.(2), page 97]. Ledenbasch & Marshall (2009) further stated that yoghurts containing no more than 10 yeast cells, should have a shelf life of 3-4 weeks, when stored under appropriate conditions (5°C) and yoghurts having >100 cfu/g (2 log cfu/g) tend to spoil quickly. With BGNCD having spoilage fungi counts of 183.33 cfu/mL (2.26 log cfu/mL) makes it susceptible to rapid deterioration. On the other hand according to the Switzerland Specification guide, the limit contaminant of yeast and moulds combined for fermented milks is 3 log cfu/mL making BGNCD (2.26 log cfu/mL) within specification and safe for consumption.

3 *Total coliforms in Bambara groundnut cultured drink*

There was no coliform detected on BGNCD. Coliforms that occur in large numbers are a guide to presence of potential entero-pathogens (*E. coli*) in drinking water, soil and vegetables, indicating their microbiological quality (Rompré *et al.*, 2002; Yin & Ding, 2009). Coliforms can be described as aerobic (oxygen loving) and facultative anaerobic bacteria and in order for these organisms to multiply, they need to ferment lactose under gassy and acidic conditions at temperature range (35 to 37°C) (Rompré *et al.*, 2002; Da Silva *et al.*, 2013).

Consequently, the evaluation of total coliforms in BGNCD was necessary, as water was the main ingredient of BGNM, from which BGNCD was produced. BGNCD is acidic rendering itself a selective growth environment for coliforms microorganisms. However, the absence of lactose in BGNCD may be the inhibitory growth factor of total coliforms, which explains the absence of coliforms in BGNCD. BGNCD is within specification according to the Specifications and Standard for Foods (2011) (negative); East African Standards, (2006) (negative) for yoghurt; as well as Kenya Standards, (2013) (10 cfu/g) for fermented milks.

4 *Staphylococcus aureus sanitary properties of Bambara groundnut cultured drink*

S. aureus is a spoilage food borne pathogen, known to cause disease outbreaks to its development (*S. aureus* enterotoxin) in food stuff and consumption of it (Solano *et al.*, 2013; Jamali *et al.*, 2015;). Staphylococci have growth temperature range of 6 to 48°C, however optimum temperature of 37°C with pH range (4.0 to 9.8), optimum pH (6-7), while enterotoxin has temperature range of 10 to 46°C, with optimum temperature of 35 to 45°C, with optimum pH (5.15) and salt loving (10 to 20% NaCl) (Danielsso-Tham, 2013).

Therefore, that contributes to the growth of *S. aureus* and can be used to explain its non-detection in BGNCD. BGNCD has been previously reported to have a pH of 4.47 [Section 4.9.1. (2), page 95], with a storage temperature of 4-7°C, which makes BGNCD a non-selective environment for the growth of *S. aureus*. Lee *et al.* (2015) further reported that pathogenic *S. aureus* is associated with rich protein foods, rendering BGNCD less susceptible to its spoilage, with only protein content of 1.2% [Section 4.9.1.(1), page 94]. Additionally, presence of *S. aureus* is an indicator of poor hygiene (handling, pasteurization and unsuitable storage conditions). Pathogenic *S. aureus* has also been reported to be carried by humans (nose, throat and skin) (Di-Ciccio *et al.*, 2015), which further suggests that the absence of *S. aureus* implies good manufacturing practices (hygienic handling) were employed during the production of BGNCD.

5 *Escherichia coli* sanitary properties of Bambara groundnut cultured drink

There was no detection of *E. coli* in the produced BGNCD. *E. coli* is a facultative anaerobe food borne pathogen that can cause infectious diseases if ingested (Eslava *et al.*, 2003). *E.coli* is mainly found in faecal contaminated water. Presence of *E. coli* implies poor practices during handling of food material and determines the microbiological quality of the food (Sangadkit *et al.*, 2012; Bonadonna, *et al.*, 2007).

Enteric pathogenic *E. coli* has different strains, which include enterotoxigenic *E. coli*, enteropathogenic *E. coli*, enteroinvasive *E. coli*, Shiga toxin producing *E. coli*, which forms part of the ones known to be responsible for severe diarrhea in children and adults, diseases as well as death (Badri *et al.*, 2009). It can be anticipated that during the production of BGNCD, Good Manufacturing Practices (GMP'S) were employed as no *E. coli* strains were detected.

4.9.5 Adequacy of linear interaction model for the effects of gum arabic and methylcellulose

The effect of gum arabic (GA) and methylcellulose (MC) using the linear polynomial regression model on WSI, WAI, and wettability for foam-mat dried BGN yoghurt was modelled. The linear model regression coefficients for functional properties of foam-mat dried BGN yoghurt are detailed in Table 4.4. The model p-value ranged from 0.0002 to 0.02 indicating that the linear models were significant ($p < 0.05$) for each response in explaining the variation between the independent and dependent variables. The predicted R^2 for WAI and WSI ranged from 0.752 to 0.797 and was in reasonable agreement with the adjusted R^2 range of 0.849 to 0.876 with a difference less than 0.2, respectively.

Furthermore, the adequate precision (AP) of WAI and WSI ranged from 8.930 to 10.89 respectively, which measures the signal to noise ratio indicating an adequate signal. A ratio greater than 4 is seen as desirable. Hence the linear polynomial regression model was an adequate fit to navigate the design space. The regression for the effect of gum arabic (β_1), methylcellulose (β_2) and gum arabic interaction with methylcellulose (β_{12}) on the WSI and WAI of BGNPY is given by the equation

$$Y = \beta_0 + \beta_1 + \beta_2 + \beta_{12} \dots\dots\dots \text{equation 4.4}$$

Table 4.4 Regression coefficients of predicted functional properties of dried BGN yoghurt with gum arabic and methylcellulose.

Coefficients	Dependent variables		
	WSI	WAI	Wett.
Linear			
β_0	73.94 ¹	1.19 ¹	62.05 ¹
β_1	0.12	-0.05	-2.16 ¹
β_2	-9.88 ¹	0.47 ¹	-4.62
β_{12}	0.50	0.06	
R^2	0.9098	0.8898	0.5541
Adjusted R^2	0.8760	0.8485	0.4550
Pred R^2	0.7971	0.7521	0.2073
CV (%)	2.28	9.40	8.46
Adeq precision	10.809	8.930	5.386

β_0 : Intercept; β_1 : Coefficient for the effect of gum arabic; β_2 : Coefficient for the effect of methylcellulose; β_{12} : Interaction of gum arabic and methylcellulose; WAI: Water absorption index; WSI: Water solubility index; Wett.: Wettability; ¹: significant at $p < 0.05$

On the other hand wettability showed only 2.6% chance that an f-value this large could occur due to noise. For this model a lack of fit value (0.00), implies lack of fit is not significant. Furthermore the predicted R^2 of 0.207 was not close to the adjusted R^2 (0.455), which exhibited a poor fit compared to WAI and WSI. Hence, for obtaining optimal response surface effects, the model with the highest R^2 was used (WSI).

4.9.6 The effect of gum arabic and methylcellulose on WSI, WAI and wettability of foam-mat dried BGN powdered yoghurt

1 Water solubility index (WSI) of foam-mat dried

The effect of methylcellulose on WSI of Bambara groundnut powdered yoghurt (BGNPY) was significant ($p < 0.05$), while gum arabic and interaction of gum arabic and methylcellulose did not have any significant effect ($p > 0.05$) on WSI of BGNPY. Gum arabic had a positive effect (+ 0.12111) by increasing the WSI of BGNPY. However, the intensity of increase was fairly low looking at the value (0.12111). An increase of methylcellulose in the system had a significant decrease on the WSI of BGNPY ($\beta_2 = -9.88$). Figure 4.9 is a depiction of the behaviour of gum arabic and methylcellulose on WSI. Methylcellulose (1.5%) gave WSI just below 62%. On the other hand methylcellulose (0.5%) had WSI of approximately 69%. The contour lines observed, linked to the methylcellulose axis suggest a positive significant effect it has on the WSI of BGNPY. The interaction of gum arabic (6%) and methylcellulose (0.5%) resulted in high WSI.

2 Water absorption index

The effect of methylcellulose on WAI of BGNPY was significant ($p < 0.05$) meanwhile the effect of gum arabic was not significant ($p > 0.05$) on the WAI of BGNPY. For WAI gum arabic had a negative effect ($\beta_1 - 0.04944$) by decreasing the WAI of BGNPY. The response surface for the interaction effect of methylcellulose and gum arabic on WAI is detailed in Figure 4.10. An increase in methylcellulose resulted in high WAI of BGNPY and a decrease in methylcellulose resulted in a reduced WAI. This was confirmed by the by the +positive value of methylcellulose ($\beta_2 = + 0.47333$). The highest WAI of 2.13 g/g for BGNPY was observed at methylcellulose (1.5%) and the lowest WAI of 1.31 g/g was observed at methylcellulose (0.5%). Furthermore, the contour lines connected to the methylcellulose axis on the diagram illustrates the significant effect of methylcellulose on the WAI of BGNPY.

3 Wettability

The analysis of variance for the response surface linear model indicated that methylcellulose had no significant effect ($p > 0.05$) on wettability of BGNPY.

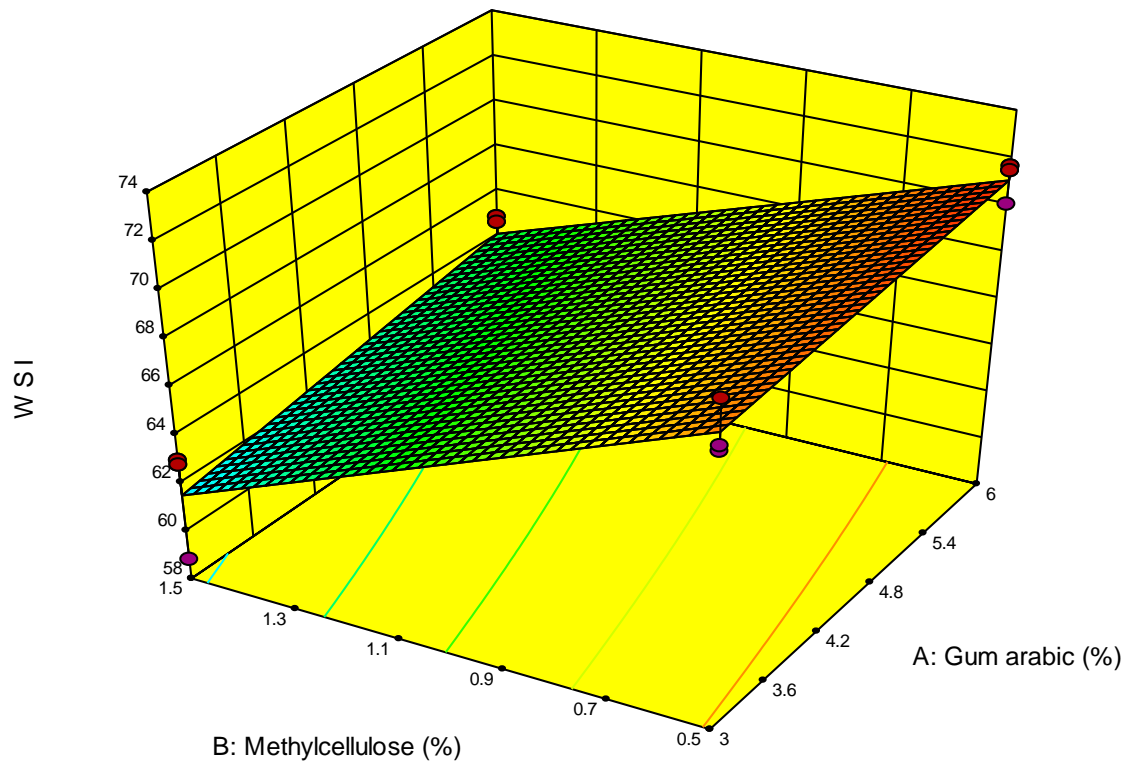


Figure 4.9 Effect of gum arabic and methylcellulose on the WSI of BGNPY-NR

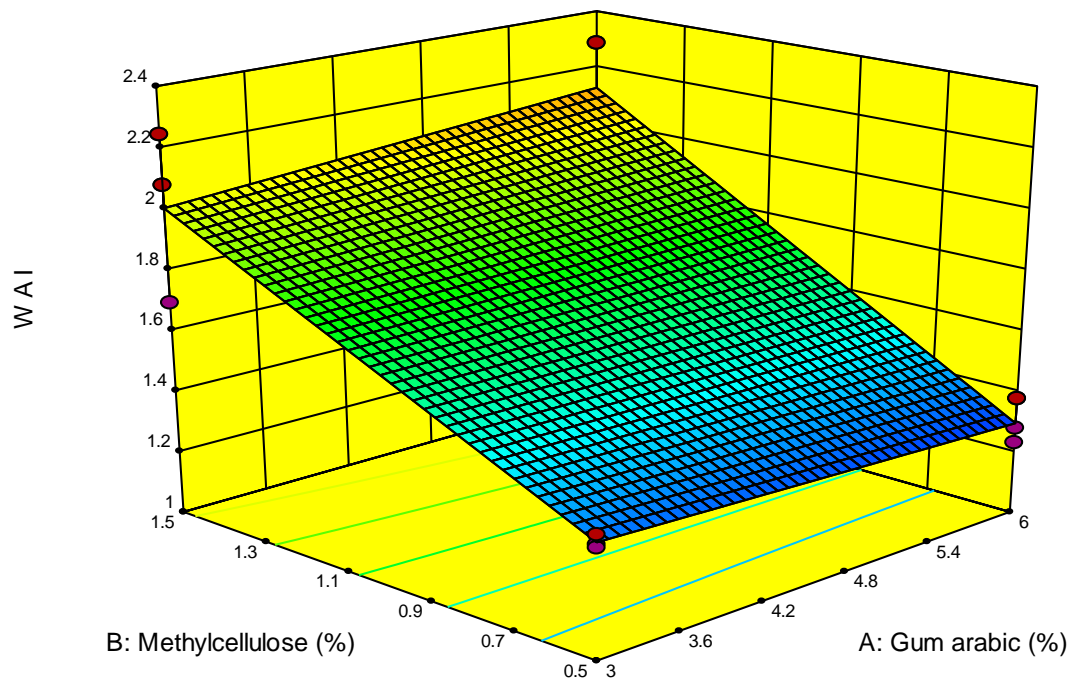


Figure 4.10 Effect of gum arabic and methylcellulose on the WAI of BGNPY-NR

While gum arabic showed a significant effect ($p < 0.05$) on wettability of BGNPY. Figure 4.11 is a representation of response surface for the behaviour of methylcellulose and gum arabic on wettability of BGNPY in a food system. The highest methylcellulose concentration (1.5%) in combination with the highest gum arabic concentration (6%) resulted in higher wettability, with wettability time of 42.2 s (Figure 4.10). In addition the least methylcellulose concentration (0.5%) combined with the least gum arabic concentration (3%) exhibited wettability time of 53.3 s.

The optimisation goal was to maximise water solubility index of gum arabic and methylcellulose. The optimal parameters were; gum arabic (6%) and methylcellulose (0.5%) with desirability of 0.956. Desirability is an objective function ranging from 0 to 1, 0 describing out of limit and 1 being at the goal.

4.10 Conclusion

Bambara groundnut cultured drink (BGNCD) was effectively produced from Bambara groundnut (BGN) flour by hydration of the flour with normal tap water. The final fermented beverage contained sufficient nutrients as edible yoghurt, with high total dietary fibre (3.6%), low fat (0.3%) and a protein content of 1.2%. The acidic and pH levels were 0.38% and 4.47, respectively. Physical attributes of BGNCD showed promising end results compared to other yoghurts specifically with syneresis of 60.67%, which of cause is dependent on the nutrients (fat), water activity of 0.85 and Lightness (L^*) of 75.06. Microbiological aspects were within the required specification with regards to the consumption of the yoghurt. BGNCD was determined to be pathogen free, with no detection of *E. coli* and *S. aureus* and low bacterial counts (aerobic organisms and spoilage yeast and moulds). The apparent viscosity and viscoelastic properties of BGNCD were comparable to other yoghurt drinks reported in the literature. Therefore, increasing the commercialisation potential for BGNCD as a fermented drink. Foam-mat dried BGNCD can be produced using; gum arabic (6%) and methylcellulose (0.5%).

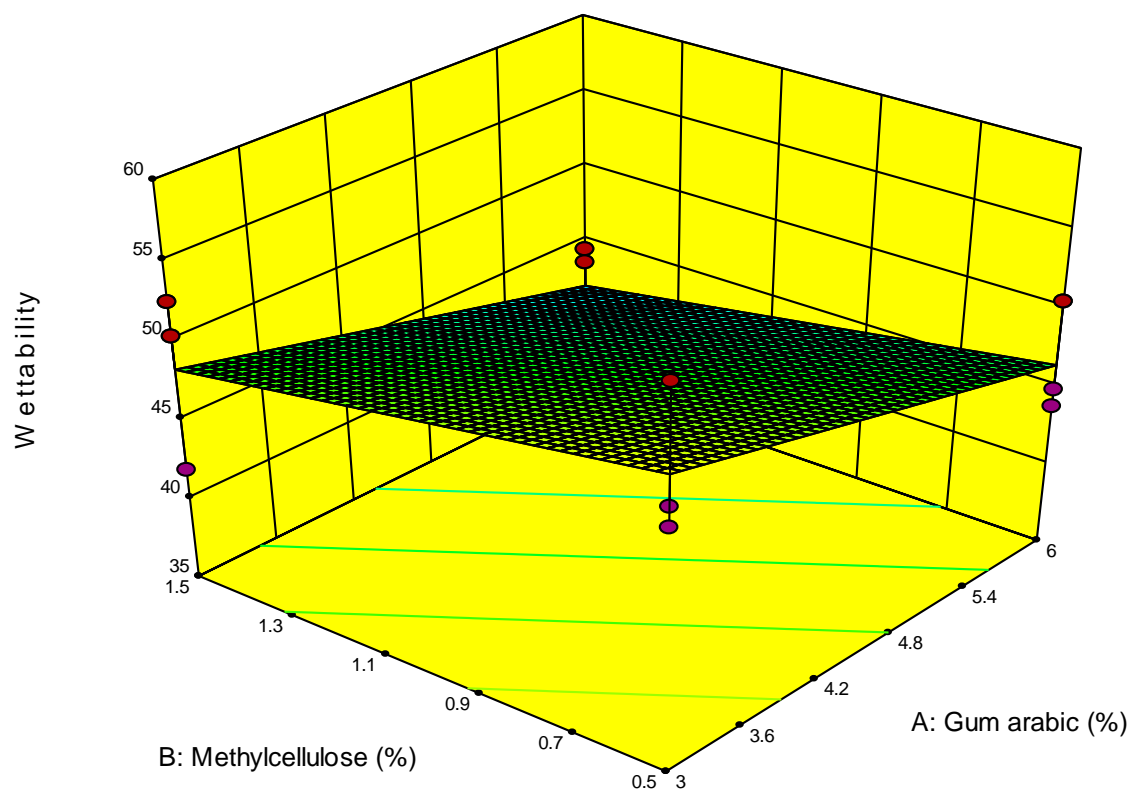


Figure 4.11 Effect of gum arabic and methylcellulose on the wettability of BGNPY-NR

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CHAPTER 5

FUNCTIONAL, NUTRITIONAL, THERMAL AND MICROBIOLOGICAL CHARACTERISTICS OF FOAM-MAT DRIED BAMBARA GROUNDNUT YOGHURT FROM BAMBARA GROUNDNUT MILK POWDER

Abstract

Bambara groundnut powdered yoghurt (BGNPY) was produced using Bambara groundnut milk powder (BGNMP) and Bambara groundnut milk (BGNM). BGNMP was reconstituted with water (1:5). The reconstituted BGN milk (BGNM-R) and original non-reconstituted BGNM (BGNM-NR) were inoculated with normal yoghurt culture; *Lactobacillus bulgaricus* and *Streptococcus thermophilus* (0.01 g/100 mL), while held at a temperature of 45°C and incubated for 24 h at 35°C in a water bath. The BGN yoghurts were then dehydrated using the foam-mat drying process with gum arabic (6%) and methylcellulose (0.5%) and dried at 50°C for 24h. The BGN powdered yoghurt from reconstituted milk (BGNPY-RM) and BGN powdered yoghurt from non-reconstituted milk (BGNPY-NRM) were evaluated for functional, nutritional and microbiological characteristics. Rehydration properties of the powdered yoghurts, including WAI and WSI did not differ significantly ($p > 0.05$). WSI and WAI ranged from 71.2 to 73.7% and 1.3 to 1.3%, respectively. The moisture, total fat, monounsaturated fats, polyunsaturated fats, saturated fats, protein, total dietary fibre, total sugars, carbohydrates and energy ranged from 9.0 to 9.1%, 1.4 to 1.6%, 0.3 to 0.4%, 0.6 to 0.6%, 0.5 to 0.6%, 4.8 to 5.7%, 3.9 to 4.3%, 2.1 to 2.2%, 75.3 to 76.0% and 1430 to 1433 kJ, respectively with no significant ($p > 0.05$) difference, while ash ranged from 2.29% to 2.52%, with a significant ($p < 0.05$) difference. Particle size and particle size distribution of BGNPY-R and BGNPY-NR had no significant ($p > 0.05$) difference, with surface-area mean diameter ($d_{3,2}$) and volume mean diameter ($d_{4,3}$) range of 103.14 to 112.40 μm and 105.46 to 114.86 μm for BGNPY-RM and BGNPY-NRM, respectively. There were no pathogenic bacteria, (*S. aureus*, *E. coli*) detected in BGNPY-RM and BGNPY-NRM.

5.1 Introduction

Yoghurt is a product of fermentation of milk sugars into lactic acid by the addition of yoghurt microflora containing *Streptococcus thermophilus* and *Lactobacillus delbrueckii* ssp. *bulgaricus*. In some countries less traditional microorganisms, such as *Lactobacillus helveticus* and *Lactobacillus delbrueckii* ssp. *lactis*, are sometimes mixed with the starter culture (Mckinley, 2005). Fermented milk products such as yoghurt were originally developed as a means of preserving the nutrients in milk, it was soon discovered that, by fermenting with different microorganisms, an opportunity existed to develop a wide range of products with different flavours, textures, consistencies and, more recently, health attributes (Mckinley, 2005). Yoghurt is claimed to have some health benefits due to its microflora. For

instance, *Lactobacilli* are reported to produce antibiotic like substances (acidolin, acidophin and lactcidin). It is further stated that the yoghurt culture have antagonistic behaviour towards undesirable microorganisms found in the intestines due to some metabolites produced during fermentation (Kumar & Mishra, 2004). The market now offers a variety of yoghurts to suit all palates (personal sense of taste and flavour) and meal occasions. Yoghurts come in a variety of textures (e.g. liquid, set, and smooth), fat contents (e.g. luxury, low-fat, virtually fat-free) and flavours (e.g. natural, fruit, cereal). It can be consumed as a snack or part of a meal, as a sweet or savoury food, and are available all year-round. This versatility, together with their acceptance as a healthy and nutritious food, has led to their widespread popularity across all population subgroups (Mckinley, 2005).

The versatility of the existence of yoghurt has also encouraged and promoted production of yoghurt originating from plant sources (milk legume) as a substitute to the animal milk. In Africa and the other developing countries, there is an inadequate supply and shortage of food protein because animal protein such as meat, milk and eggs are expensive and relatively difficult to acquire. There has been a constant search for unconventional legumes such as BGN as a new protein source and almost nutritionally balanced food material for their use as a functional food ingredients and nutritional supplement (Mckinley, 2005).

Bambara groundnut (BGN) is one of the underutilized legume seeds, which makes a complete food as it contains an important source of all nutrients classes, such as protein, oils, carbohydrates (starch), vitamins and minerals. It plays an important socio-economic role in the semi-arid regions of Africa. It is a rich source of protein that could help alleviate nutritional problems in these areas. The seeds can be used to produce vegetable milk that is comparable to other vegetable milks already in the industry such as soya milk, which then can be used to produce legume based yoghurt (Massawe *et al.*, 2005). Also, recently Murevanhema & Jideani (2014) reported the shelf life stability of BGN milk and BGN probiotic beverage (yoghurt). However, being a liquid, the shelf life could be short.

The need for dried yoghurt is gradually increasing in the industry, not only for an increased shelf life, but also to provide convenience during packaging, handling and transportation of dried yoghurts. Yoghurt powder is reported to be easier and cheaper to handle compared to liquid yoghurt because the weight and volume of the product are less in its dried form. Furthermore, Bambara groundnut milk powder (BGNMP) has been reported in chapter 3 of this study; however nothing is known about the possibility of yoghurt from BGNMP. The primary objective of this study was to produce BGN powdered yoghurt from reconstituted Bambara groundnut milk (BGNM-R) and non-reconstituted Bambara groundnut milk (BGNM-NR) employing the foam-mat drying technology and further investigate its functional, nutritional, thermal and microbiological properties.

5.2 Materials and Methods

5.2.1 Source of materials and equipment

Methylcellulose and gum arabic from acacia tree were purchased from Sigma, South Africa. Bambara groundnut milk (BGNM) and Bambara groundnut milk powder (BGNMP) produced in chapter 3 of this research project were used. Equipment used for the experiments were obtained from the Food Technology Department of Cape Peninsula University of Technology, South Africa. Figure 5.1 outlines the different analyses carried out in this chapter.

5.2.2 Reconstitution of BGNMP for BGN yoghurt preparation

BGNMP was reconstituted following the food component material balance equation method reported by Saravacos & Maroulis (2011). The food component material balance equation method is used to identify one or more food components; including total solids, soluble solids, water (moisture) (Saravacos & Maroulis, 2011). The moisture content of BGNMP before and after drying was measured using the AOAC moisture (934.01) method (2005). The identified water (moisture) loss after drying was then used to establish the reconstitution parameters (water and BGNMP). The reconstitution parameters (water, and BGNMP) were determined using equation 5.1.

$$(\text{Total component in}) - (\text{Total component out}) = \text{Total component accumulated} \dots \text{equation 5.1}$$

Where “total components in” represents initial product before processing (BGNM), “total component out” represents lost water through processing and total component accumulated represents final moisture content of BGNM. BGNMP (17.7 g) was added to water (82.3 mL) and blended, principally a ratio of 1:6.

5.2.3 Stability characterisation of Bambara groundnut milk powder using turbiscan

The stability of reconstituted BGNMP was analyzed using a Turbiscan vertical scan M.A 2000. Reconstituted BGNMP (7 mL) was transferred into a Turbiscan glass sample tube (zone 20-40 mm) and closed tightly with a cap. The tube was inserted inside the cell and the detection head analyser scanned the entire height of tube containing sample, acquiring transmission and backscattering data every 40 μm at 5 min intervals for 40 min at 20°C (Blijdenstein *et al.*, 2003; Blijdenstein *et al.*, 2004). The destabilisation was determined by means of scanning analysing detection head, for the identification of coalescence or flocculation (due to particle size variation), as well as sedimentation and creaming (due to particle migration).

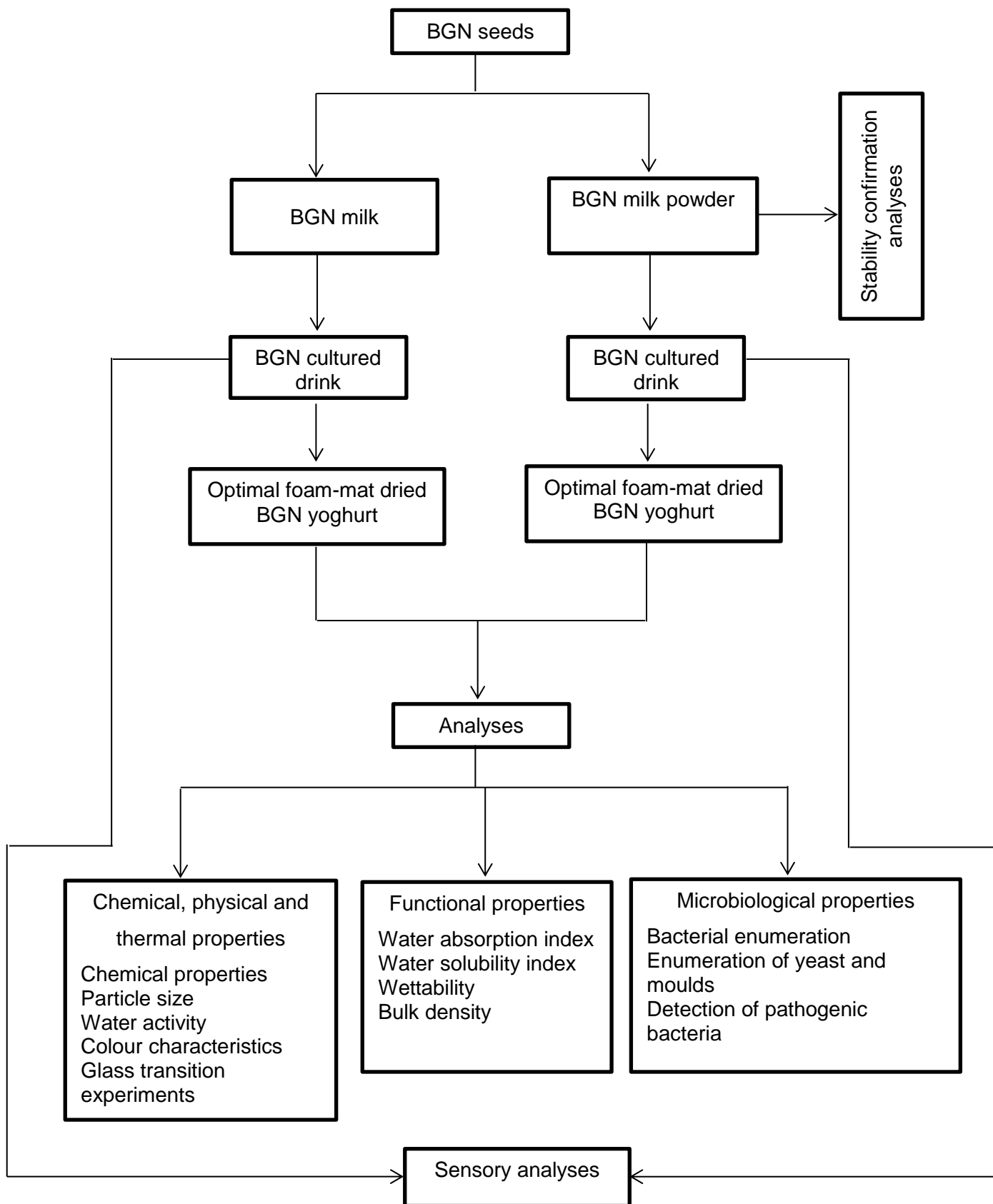


Figure 5.1 Chapter 5 methodology overview

The detection head passes light through the sample, which is received by the transmission detector, while the backscattering detector measures the light scattered backward by the sample. Measurements were done in duplicate.

5.2.4 Preparation of the BGN yoghurt

Reconstituted BGN milk (BGNM-R) and non-reconstituted BGN milk (BGNM-NR) (400 mL) in 500 mL schott bottles were warmed to 45°C in a water bath for 15 to 20 min. The BGNM was then aseptically inoculated with yoghurt microflora; *Lactobacillus bulgaricus* and *Streptococcus thermophilus* (0.01% in 100 mL) and incubated at 35°C in a water bath for 24 h. The fermented BGNM was immediately cooled in ice and stored under refrigeration at 3-4°C (Murevanhema & Jideani, 2012).

5.2.5 Production of BGN powdered yoghurt using foam-mat drying process

BGN powdered yoghurts (BGNPY) were produced using the Krasaekoopts & Bhatia (2012) method with some modification. Foam-mat drying technique was performed employing the optimised formulation. Gum arabic (6%) and methylcellulose (0.5%) were used as foaming agents. BGN yoghurts [Section 5.2.4, page 129] were mixed with the foaming agents, blended using a Silverson Model L4R Homogenizer for 10 minutes and transferred into Teflon metal trays. The trays containing the foamed mixture were dried (Cabinet dryer, Model 1069616) at 50°C for 24 h. The dried yoghurt flakes were ground into fine powder using a Royals Worcester mortar and pestle. The fine yoghurt was placed in sealed zip lock bags and stored till further analysis. The dried BGN “yoghurt” produced from reconstituted BGN “milk” was referred to as Bambara groundnut powdered “yoghurt” from reconstituted “milk” (BGNPY-RM). While, dried BGN “yoghurt” from non-reconstituted (fresh) “milk” was referred to as Bambara groundnut powdered “yoghurt” from non-reconstituted “milk” (BGNPY-NRM).

5.3 Proximate Analysis of Bambara Groundnut Powdered Yoghurt

Protein determination was done using Kjeldahl (N X 6.25), total fat according to AOAC (2005) method 996.06, total dietary fibre according to AOAC (1997) method 985.29, total sugar according to AOAC (2003) method 982.14, moisture and ash according to AOAC (2005) method 934.01 and 923.03. The carbohydrate was determined by difference and energy was determined according to Anon (2010).

5.4 Functional Properties of Bambara Groundnut Powdered Yoghurt

5.4.1 Water absorption index (WAI) and water solubility index (WSI)

WAI and WSI of the BGN powdered yoghurt from reconstituted milk (BGNPY-RM) and BGN powdered yoghurt from non-reconstituted milk (BGNPY-NRM) were determined using the

method of Kaushal *et al.* (2012). BGNPY-RM and BGNPY-NRM (2.5 g) were dispersed in 30 mL of distilled water, using a glass rod, and cooked at 90°C for 15 min in a heating metal. The cooked paste was cooled to room temperature and transferred to tared centrifuge tubes, and then centrifuged at 3000 *g* for 10 min. The supernatant was decanted for the determination of its solid content into a tared evaporating crucible dish and the sediment was weighed. The weight of the dry solids was recovered by evaporating the supernatant overnight at 110°C. WAI and WSI were calculated using equations 5.1 and 5.2.

$$\text{WAI (g /g)} = \frac{\text{Weight of sediment}}{\text{Weight of flour sample}} \dots\dots\dots \text{equation 5.1}$$

$$\text{WSI (g /g)} = \frac{\text{Weight of dissolved solids in supernatant}}{\text{Weight of flour}} \dots\dots\dots \text{equation 5.2}$$

5.4.2 Wettability

Wettability was determined using the procedure described by Jinapong *et al.* (2008). Distilled water (100 mL) at a temperature of 25 ± 1°C was poured into a 250 mL glass beaker. A glass funnel held on a retort stand was set over the beaker with the height between the bottom of the funnel and the water surface of 10 cm. A test tube was placed inside the funnel to block the lower opening of the funnel. BGNPY-RM and BGNPY-NRM weighing 0.1 g was placed around the test tube and the tube was lifted, while the stop watch was started at the same time. Time taken for the powder to become completely wet, and for all the powder particles to penetrate the surface of the water, was recorded.

5.4.3 Bulk density measurement of BGN powdered yoghurt

BGNPY-RM and BGNPY-NRM each weighing 2 g were placed into graduated syringes and sufficient pressure was applied to pack the content in the syringes. The final volume of the sample in each syringe was recorded and bulk density was expressed as grams per millilitre (g/mL) (Parrott & Thrall, 1978).

5.5 Thermal Characterisation of BGN Powdered Yoghurt

5.5.1 Glass transition temperature of BGN powdered yoghurt

The glass transition temperature (*T_g*) was determined using the method described by Shrestha *et al.* (2007a) with some modifications. A Perkin Elmer Pyris 1 Differential Scanning Calorimeter (DSC) 6000 equipped with Intracooler 2P was used to evaluate the glass transition temperature of BGNPY-RM and BGNPY-NRM samples. The purge gas used was dry nitrogen (20 mL/min). Heat flow verification was performed using indium melting point (156.60°C) and zinc melting point (419.47°C) standards at heat scan rate of 10°C/min. About 10 mg of BGNPY-RM and BGNPY-NRM were weighed into a 50 µL DSC aluminum

pan and press sealed with a lid using a DSC sample press. The thermal scanning of the samples was carried out alongside a sealed empty pan used as a reference. The thermal scanning of BGN powdered yoghurt samples was commenced from -20°C to 200°C at 10°C/min in triplicates and the onset, endset values were calculated in the DSC thermo-gram as well as the Tg value was determined as half ΔCp method at half the extrapolated change in specific heat using the Pyris software.

5.6 Physical Characterisation of Bambara groundnut Powdered Yoghurt

5.6.1 Particle size measurement of BGN powdered yoghurt

The particle size and particle size distribution was analysed using a Leo 1430VP Scanning Electron Microscope (SEM) at Stellenbosch University. Prior to imaging the samples were mounted on a stub with double sided carbon tape. The samples were then coated with a thin layer of gold in order to make the sample surface electrically conducting. Beam conditions during surface analysis were 7 kV and approximately 1.5 nA, with a spot size of 150.

Particle sizes and particle size distribution of BGNPY-RM and BGNPY-NRM were evaluated using images obtained from the SEM. The diameters of the particle sizes were measured individually according to the method of Tcholakova *et al.* (2004). BGNPY particle sizes were obtained in terms of volume-surface mean diameter ($d_{3,2}$) (Equation 5.3) and equivalent volume-mean diameter ($d_{4,3}$) (Equation 5.4).

$$d_{3,2} = \frac{\sum n_i d_{i3}}{\sum n_i d_{i2}} \dots \dots \dots \text{equation 5.3}$$

$$d_{4,3} = \frac{\sum n_i d_{i4}}{\sum n_i d_{i3}} \dots \dots \dots \text{equation 5.4}$$

5.6.2 Water activity evaluation of BGNPY-RM and BGNPY-NRM

The water activity (a_w) of BGNPY-RM and BGNPY-NRM was measured using the Novasina Ms 1 Set A_w meter, which uses a cell protection filter to measure a_w . Salt humidity standards of 53, 75 and 90% relative humidity were used to calibrate the measurement cell. BGNPY-R and BGNPY-NR (5 g) were transferred into a sample dish and placed inside the Novasina analyser and the cell measuring protection filter was immediately closed. The a_w reading was observed after period of 60 to 80 s. The test was performed in triplicate (Novasina General Catalogue, 2012).

5.6.3 Colour measurement of BGN powdered yoghurt

The colour of the BGNPY-RM and BGNPY-NRM each weighing 3 g were measured using a Konica Minolta Spectrophotometer CM-5 45°/0° standard, set at standard observer 10° and D65. The instrument was zero calibrated using a black tile ($L^* = 5.49$, $a^* = -7.08$, $b^* = 4.66$)

and white calibration was performed using a white tile ($L^* = 93.41$, $a^* = -1.18$, $b^* = 0.75$). BGNMP samples were evenly placed in a petri-dish (30 mm diameter) and reflectance measured for $L^*a^*b^*$ and LCh colour scales. The L^* coordinate is lightness, 100 represents white and closer to 0 represents black. Measurements for each sample were performed in triplicate at three different positions in the samples (one reading = average of three readings per rotated position), with the results recorded in L^* (lightness), a^* (chromaticity coordinate $+a^* = \text{red}$ and $-a^* = \text{green}$), b^* (chromaticity coordinate $+b^* = \text{yellow}$ and $-b^* = \text{blue}$), C^* (chroma) and h (hue angle $0^\circ = +a^*$, $90^\circ = +b^*$, $180^\circ = -a^*$ and $270^\circ = -b^*$ and the total colour difference (ΔE) of the BGNPY-RM and BGNPY-NRM was calculated using equation 5.4 (Bezerra *et al.*, 2013).

$$\Delta E = [(\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2]^{1/2} \dots \dots \dots \text{equation 5.4}$$

Colour difference (ΔE) < 1 can be defined as a not noticeable difference, where the observer does not notice the difference completely. Colour difference value of 1 can be defined as a just noticeable difference (JND), colour difference in the range of 4-8 is termed perceivable (Sharma, 2004), but accepted, which entails that an observer notices the colour difference, and is considered acceptable.

5.7 Microbiological Measurements of Bacterial Load in BGN Powdered Yoghurt

The pour plate method was employed for the enumeration of microorganisms in BGNPY-RM and BGNPY-NRM. Counting of all typical colonies was performed using the BOECO CC1, BOE 515700, Germany colony counter and a control was carried out for each analysis. All measurements were performed in triplicate. Only plates containing colonies from 25-250 were counted, a standard countable range for enumerating colony forming units on an agar plate using the pour plate technique without introducing error (Baylis, 2007).

5.7.1 Bacterial enumeration in BGNPY-RM and BGNPY-NRM

BGNPY-RM and BGNPY-NRM (10 g) were weighed into a 90 mL sterile ringers' solution (10^{-1}) and mixed well. Then a series of dilutions (10^{-1} to 10^{-6}) were prepared by first transferring 1 mL aliquot of the 100 mL sample solution into a 9 mL sterile ringers' solution as the second dilution (10^{-2}), until last dilution (10^{-6}). For each dilution a 1 mL aliquot was carefully and aseptically transferred into the base of a labelled sterile Petri-dish. Then separately, approximately 15 mL of pre-cooled plate count agar (PCA) and violet red blood agar (VRBA) was poured for the enumeration of mesophilic counts, and carefully swirled to mix well. Once all plates were allowed to solidify, they were incubated in an inverted position. PCA and VRBA plates were incubated at 37°C for 48 h (Baylis, 2007).

5.7.2 Enumeration of yeast and moulds in BGNPY-RM and BGNPY-NRM

A series of dilutions for BGNPY-RM and BGNPY-NRM were prepared as described in 5.7.1. Then approximately 15 mL of pre-cooled rose bengal chloramphenicol agar (RBCA) was poured into each petri-dish and mixed by gently swirling for the enumeration of yeast and moulds. Once solidified, plates were incubated at 25°C for 5 days in an inverted position (Baylis, 2007).

5.7.3 Detection of presumptive *Escherichia coli* species on Bambara groundnut powdered yoghurt

The Da Silva *et al.* (2013) method was used for the detection of *Escherichia coli* (*E. coli*) with slight modifications. The detection of *E. coli* was performed by employing the indole test. BGNPY-RM and BGNPY-NRM (1 g) was transferred into sterile 9 mL tryptone soya broth (TSB) to prepare an enrichment and was incubated at 37°C for 24 h. From the sample enrichment, 1 mL was aseptically transferred into a sterile 5 mL brilliant green bile broth (BGLB) containing a Durham tube positioned downward.

From the BGLB mixture, 0.1 mL was transferred into a sterile 5 mL tryptone water prepared solution and both tubes containing BGLB and tryptone water mixtures were incubated at 44°C for 24 h. Thereafter, the BGLB containing tube was checked for the occurrence of a bubble inside the surface of the Durham tube. The presence of a bubble indicated positive for *E. coli* and absence of a bubble indicated negative for *E. coli*. Further confirmation was employed on occurrence of bubble by the addition of Kovac's reagent into the tube containing tryptone water mixture and the formation of a red colour ring on the surface of the tryptone water tube after the addition of Kovac's reagent indicated presence of *E. coli*.

5.7.4 Detection of presumptive *Staphylococcus aureus* on Bambara groundnut powdered yoghurt

The Lee *et al.* (2015) method was used with a slight modification employing the direct detection process. BGNPY-RM and BGNPY-NRM (1 g) was transferred into sterile 9 mL tryptone soya broth (TSB), mixed by vigorous shaking to make up enrichment and was incubated at 37°C for 24 h. After the incubation period, the sample enrichment was streaked onto a prepared mannitol salt agar (MSA) plate and further incubated for 24 h. Gram-stain was performed only on observed typical *S. aureus* colonies and further confirmed using the staphylase coagulase test for presence of *S. aureus*.

5.8 Sensory Analysis of Bambara Groundnut Cultured Drink Produced from Reconstituted BGN Milk and Non-reconstituted BGN Milk

Consumer sensory analysis was conducted with 48 untrained panellists. The sensory evaluation test was conducted in a sensory laboratory at room temperature ($22 \pm 1^\circ\text{C}$). Each panellist was accompanied to their individual booth, to minimise destruction. The panellist consisting of students and staff were served two samples of BGNCD produced using non-reconstituted Bambara groundnut milk (BGNM-NR) and the other using reconstituted Bambara groundnut milk (BGNM-R). The BGN cultured beverages were prepared 24 h before sensory evaluation. Each sample (40 mL) was identified by a three-digit random number and was served cold ($4-6^\circ\text{C}$). The samples were presented in white polystyrene foam cups placed onto green and black coloured plastic trays alongside one another. A cup of water was provided to reset the palate before and between tastings. The panellists were requested to give their consent for participation in the study, by signing the provided consent form before evaluating the samples. The panellists were provided with a score sheet which consisted of two-coded samples with a 5-point hedonic scale ranging from 1= dislike extremely to 5 = like extremely. The panellists were instructed to rate each sample individually on its own merit on the five point hedonic rating scale for appearance, colour, taste, aroma, texture and overall acceptability

5.9 Data Analysis

All measurements were carried out in triplicate and results were expressed as mean \pm standard deviations. Multivariate analysis of variance (MANOVA) was employed to determine mean differences among treatments. Duncan's multiple range test was used to separate means where differences existed (IBM SPSS 23).

5.10 Results and Discussion

5.10.1 Stability of reconstituted Bambara groundnut milk

Figure 5.2 is the Turbiscan profile of reconstituted BGNM indicating the stability curves as backscattering (BS) flux percentage (%). The BS% of the reconstituted milk was approximately 25.6% presented on the y-axis, which also represented the solids in reconstituted BGNM solution. The reconstituted milk was quite stable observed on the x-axis (10 mm-60 mm), indicated by the straight curve of multiple lines overlaying on top of each other. This implies that the particles were evenly dispersed in the solution.

Stability is studied to evaluate the homogeneity of an emulsion or solution, which further identifies creaming, sedimentation, coalescence and the flocculation mechanism occurring in that solution (McClements, 2004). The creaming instability can be identified by concentration of particles migrating to the top of the solution, where as in sedimentation particles settle to the bottom of the solution. Flocculation is the aggregation of particles and

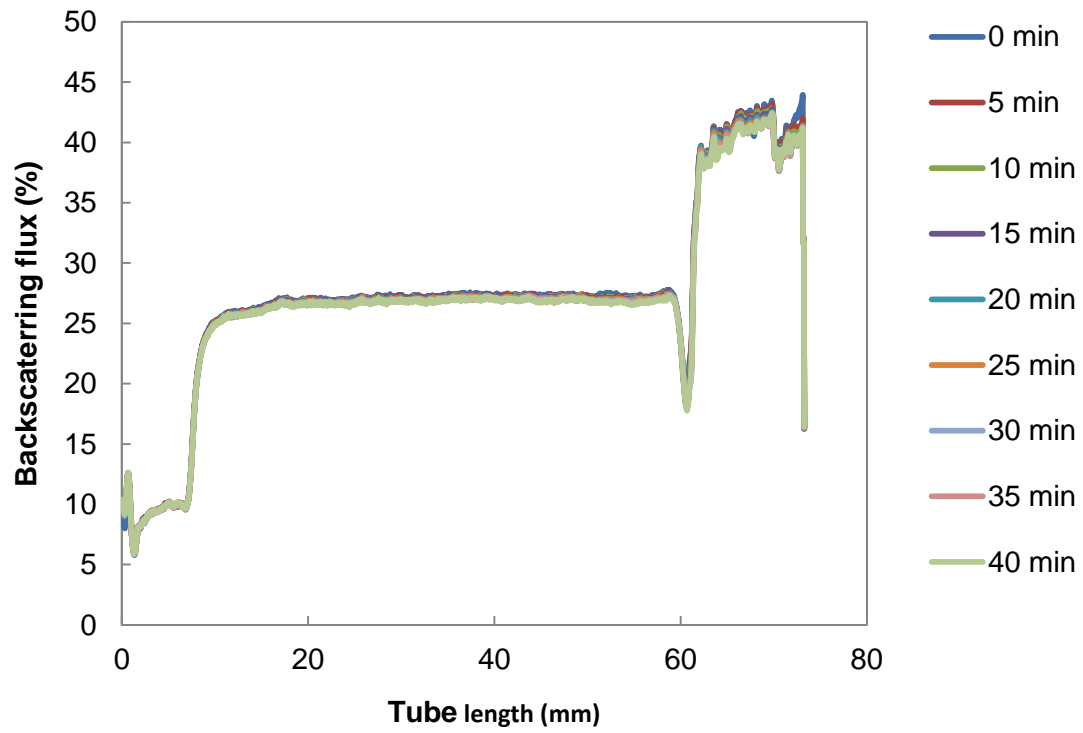


Figure 5.2 Turbiscan backscattering multiple stability scan profile of reconstituted BGN milk.

coalescence involves merging of two particles into one large particle (Robins *et al.*, 2002; McClements, 2004). Liang *et al.* (2014) reported that the creaming and sedimentation mechanism is said to occur due to the gravitational and or centrifugal force separation that out weights the diffusion of droplets or particles. The stability of reconstituted BGN milk can then be attributed to what was reported by Klinkesorn *et al.* (2004) that texture modifiers used as ingredients increased the viscosity of the continuous phase emulsion, thereby slowing down gravitational separation of droplets. It was further stated that the stabilizing action of maltodextrins is by viscosity modification or gelation of an aqueous continuous phase. Which explains the stability of the reconstituted BGN milk, as maltodextrin was one of the main ingredients.

5.10.2 Chemical composition of Bambara groundnut powdered yoghurt

The chemical composition of BGNPY from non-reconstituted milk and BGNPY for reconstituted BGN milk is displayed in Table 5.1. The moisture, ash, total fat, monounsaturated fats, polyunsaturated fats, saturated fats, protein, total dietary fibre, total sugars, carbohydrates and energy of BGNPY-NRM: BGNPY-RM are 9.1; 9.0%, 2.3; 2.5%, 1.4; 1.6%, 0.3; 0.4%, 0.6%; 0.6%, 0.5; 0.6%, 5.7; 4.8%, 4.2; 3.9%, 2.1; 2.2%, 75.3; 76.0% and 1430; 1433 kJ, respectively. The chemical composition of the BGNPY-RM and BGNPY-NRM were not significantly ($p > 0.05$) different, except for their ash content ($p < 0.05$). BGNPY-RM had a higher ash content (2.52%), compared to BGNPY-NRM (2.29%). On the other hand, BGNPY-NRM had a noticeably higher moisture (9.1%), protein (5.7%) and total dietary fibre (4.2%) content compared to BGNPY-RM, which was not significant. The total dietary fibre content of BGNPY-RM (3.9%) and BGNPY-NRM (4.2%) were observed to be fairly high.

This may be attributed to the foaming agents used, gum arabic and methylcellulose. Soluble fibres, which are known for their good solubility in water, have been reported to include mucilage and gums (botanical, animal and microbial). Gum arabic and methylcellulose are classified as botanical gums (Tosha & Yada, 2010; Dhingra *et al.*, 2011), which were incorporated in the manufacturing of BGNPY-RM and BGNPY-NRM. Furthermore, methylcellulose is derived from highly purified forms of cellulose (Manthey & Xu, 2009), known to form part of insoluble dietary fibre (Dhingra *et al.*, 2011). Therefore, the addition of these gums may have contributed to the high total dietary fibre. The fairly high protein of BGNPY-RM (4.8%) and BGNPY-NRM (5.7%) may also be associated with the gum arabic incorporated into the system. William & Phillips (2001) stated that gum arabic contains small amounts of protein, which is the important part of its structure.

Table 5.1 Chemical composition (g/100 g product) of Bambara groundnut powdered yoghurt from reconstituted and non-reconstituted Bambara groundnut milk

Nutrient	Proximate (%) ¹	
	BGNPY-NR	BGNPY-R
Moisture	9.1 ± 0.03 ^a	9.0 ± 0.01 ^a
Ash	2.3 ± 0.01 ^a	2.5 ± 0.01 ^b
Total fat	1.4 ± 0.14 ^a	1.6 ± 0.13 ^a
Monounsaturated fats	0.3 ± 0.04 ^a	0.4 ± 0.02 ^a
Polyunsaturated fats	0.6 ± 0.06 ^a	0.6 ± 0.03 ^a
Saturated fats	0.5 ± 0.04 ^a	0.6 ± 0.08 ^a
Protein	5.7 ± 0.20 ^a	4.8 ± 0.28 ^a
Total dietary fibre	4.2 ± 0.13 ^a	3.9 ± 0.08 ^a
Carbohydrates	75.3 ± 0.58 ^a	76.0 ± 0.29 ^a
Of which total sugars	2.1 ± 0.05 ^a	2.2 ± 0.20 ^a
Energy kJ	1430. ± 1.42 ^a	1433 ± 4.94 ^a

Mean ± standard deviation of triplicate determinations, Mean values in the same row followed by different letters are significantly different ($p < 0.05$), BGNPY-NR; Bambara groundnut powdered yoghurt from fresh BGNM, BGNPY-R; Bambara groundnut powdered yoghurt from reconstituted BGNMP

5.10.3 The functional characteristics of BGNPY-RM and BGNPY-NRM.

The WAI, WSI, wettability and BD of BGNPY-RM and BGNPY-NRM was 1.27 g/g; 1.31 g/g, 73.30%; 71.22%, 15.70 s; 46.70 s and 0.87 g/mL; 0.82 g/mL, respectively. There was no significant ($p < 0.05$) difference on the WAI, WSI and BD between the powdered yoghurts. However, a significant ($p < 0.05$) difference was observed in wettability between the two powdered yoghurts as shown in Table 5.2. BGNPY-RM was higher in wettability (15.70 s) compared to BGNPY-NRM (46.70 s). Koç *et al.* (2014) reported the functional properties of spray dried dairy yoghurt powder, with wettability (374 s), solubility index (68.7%) and dispersibility (351 s). BGN powdered yoghurts showed much higher rehydration properties, with WAI (1.27 g/g; 1.31 g/g), WSI (73.30%; 71.22%) and wettability (15.70 s; 46.70 s) for BGNPY-RM and BGNPY-NRM, respectively.

This may be linked to the gum arabic (6%) incorporated into the milk in production of BGN yoghurt powders, reported to be readily soluble in water (Ward *et al.*, 2005). Furthermore, it was also stated that methylcellulose is non-ionic, an active molecule that has no electric charge (not affected by the hardness of water), but can also act as an emulsifier due to its hydrophilic as well as hydrophobic capabilities (Fernandez *et al.*, 2003). In addition Koç *et al.* (2014) stated that, the lower the particle size, the lower the solubility of powders and vice versa. In addition Koç *et al.* (2014) reported a dairy spray dried yoghurt powder with particle size (93.053 μm) and wettability (374 s). Whilst the particle size and wettability of BGNPY-RM and BGNPY-NRM were 105.46 μm , 114.86 μm and 15.7 s and 46.7 s, respectively [Section 5.10.5, page 138 and Section 5.10.3, page 136]

Furthermore, the BD of BGNPY-RM (0.87 g/mL) and BGNPY-NRM (0.82 g/mL) were higher compared to the BD of spray dried dairy yoghurt (0.538 g/mL) reported by Koç *et al.* (2014). This may be associated with different processing techniques employed to obtain the yoghurt powders, as BGN powdered yoghurt was foam-mat dried and further manually milled to a fine powder. The BD differences may also be attributed to the different yoghurt compositions.

5.10.4 Thermal characteristics of BGN powdered yoghurt

The onset temperature (BGNPY-RM; 38.20°C; BGNPY-NRM; 49.72°C), glass transition temperature (T_g) (BGNPY-RM: 48.88°C; BGNPY-NRM: 61.01°C), endset temperature (BGNPY-RM: 59.70°C; BGNPY-NRM: 73.69) and heat capacity (BGNPY-RM: 1.46; BGNPY-NRM: 1.09) is given in Table 5.3. There was a significant ($p < 0.05$) difference in the onset, T_g and endset temperature of BGNPY-R and BGNPY-NR, but no significant ($p > 0.05$) difference in heat capacity of BGNPY-R and BGNPY-NR. BGNPY-NR had the highest T_g of 61.01°C compared to BGNPY-NR (48.88°C), which implies that BGNPY-NR is more heat stable than BGNPY-NR.

Table 5.2 Functional properties of BGNPY-RM and BGNPY-NRM

Powdered yoghurt	WAI (g/g)	WSI (100/g)	Wett. (s)	BD (g/mL)
BGNPY-RM	1.27 ± 0.09 ^a	73.30 ± 1.47 ^a	15.70 ± 3.97 ^a	0.87 ± 0.04 ^a
BGNPY-NRM	1.31 ± 0.08 ^a	71.22 ± 0.84 ^a	46.70 ± 3.59 ^b	0.82 ± 0.04 ^a

Mean values ± SD of triplicate determinations. Mean values in the same column followed by different letters are significantly ($p < 0.05$) different, BGNPY-NR; Bambara groundnut powdered yoghurt from fresh BGNM, BGNPY-R; Bambara groundnut powdered yoghurt from reconstituted BGNMP, WAI = Water absorption index, WSI = Water solubility index, Wett. = Wettability, BD = Bulk density

Table 5.3 Glass transition temperature of BGNPY-RM and BGNPY-NR

Powdered yoghurt	Onset temperature (°C)	Glass transition temperature (°C)	Endset temperature (°C)	ΔC_p (J g⁻¹ K⁻¹)
BGNPY-RM	38.20 ± 0.82 ^a	48.88 ± 2.11 ^a	59.70 ± 3.31 ^a	1.46 ± 0.33 ^a
BGNPY-NRM	49.72 ± 2.59 ^b	61.01 ± 3.53 ^b	73.69 ± 0.30 ^b	1.09 ± 0.23 ^a

Mean values ± standard deviation of triplicate determinations. Mean values in the same column followed by different letters are significantly ($p < 0.05$) different, BGNPY-NR; Bambara groundnut powdered yoghurt from fresh BGNM, BGNPY-R; Bambara groundnut powdered yoghurt from reconstituted BGNMP

The stability of a milk powder is closely associated with its physical state, which includes physical changes such as sticking and caking resulting in collapsing of the structure. Deformed physical stability of milk powders adversely affects their quality and shelf life (Hogan *et al.*, 2010). The phenomenon of stickiness and caking occurs due to plasticisation of milk powder surface particles by water, allowing inter-particle binding and formation of clusters. When the powder is in its glassy stable state, it has high viscosity and the contact time between particles is prolonged. A drastic decrease in viscosity over the Tg range reduces the contact time between particles, causing inter-particles to join resulting and stickiness and caking (Roos, 2001).

The Tg phenomena (glassy state to rubbery state) powders only occurs when powders are exposed to temperatures above their Tg value (Foster *et al.*, 2006). Therefore, both BGNPY-R and BGNPY-R are less susceptible to physical deformation of caking and stickiness and can be deemed stable as their Tg is above room temperature, which is their recommended storage temperature, thus having the potential of an increased shelf life.

5.10.5 Physical characteristics of BGN powdered yoghurt

In this study the physical properties evaluated included particle size, water activity and colour parameters, of Bambara groundnut powdered yoghurt. These attributes of BGN powdered yoghurt are discussed in the following sections.

1 Particle size characteristics of BGN powdered yoghurt.

The surface area mean diameter ($d_{3,2}$) of BGNPY-RM (103.14 μm); BGNPY-NRM (112.40 μm) and volume mean diameter ($d_{4,3}$) of BGNPY-RM (105.46 μm); BGNPY-NRM (114.86 μm) are illustrated in Table 5.4. There was no significant ($p > 0.05$) difference in the surface area mean diameter ($d_{3,2}$) and volume mean diameter ($d_{4,3}$) of BGNPY-RM and BGNPY-NRM. This implies that perceptibly, the particle size and particle size distribution of the BGN yoghurt powders was similar.

Koç *et al.* (2014) reported volume mean diameter of a spray dried dairy powder of 3.053 μm , which was quite lower than that of BGNPY-R (105.46 μm) and BGNPY-NR (112.40 μm). This difference may be due to different composition and processing technologies. Scanning electron micrographs (SEM) of BGNPY-R and BGNPY-NR are shown on Figure 5.3. The surface morphology of BGNPY-RM and BGNPY-NRM resembled surface characteristics of a freeze dried powder. The surface morphology of the BGN powdered yoghurts differed compared to spray dried powders which are reported to be spherical, with a smooth surface, less collapsed structure and solid like with presence of large dents or slightly dimpled surfaces (Fyfe *et al.*, 2011).

Table 5.4 Particle size and particle size distribution of BGNPY-RM and BGNPY-NR

Powdered yoghurt	d_{3,2} (µm)	d_{4,3} (µm)
BGNPY-RM	103.14 ± 17.80 ^a	105.46 ± 19.13 ^a
BGNPY-NRM	112.40 ± 6.96 ^a	114.86 ± 7.75 ^a

Mean values ± standard deviation of triplicate determinations. Mean values in the same column followed by different letters are significantly ($p < 0.05$) different; BGNPY-NR: Bambara groundnut powdered yoghurt from fresh BGNM; BGNPY-RM: Bambara groundnut powdered yoghurt from reconstituted BGNMP; d_{3,2}: Surface mean diameter; d_{4,3}: Volume mean diameter

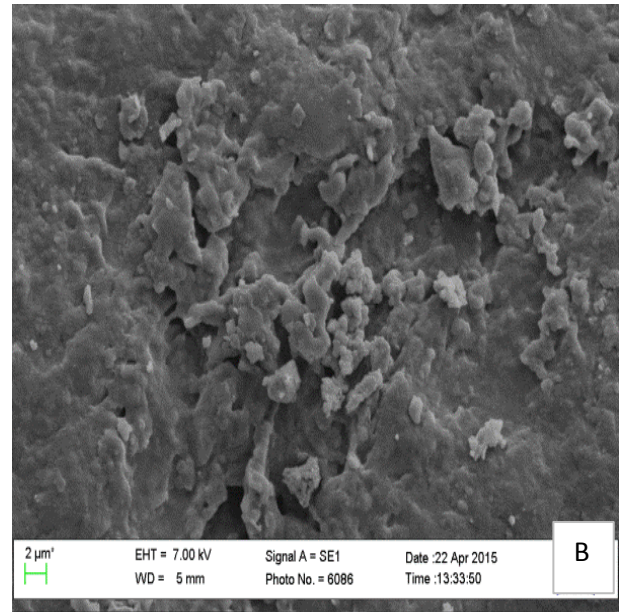
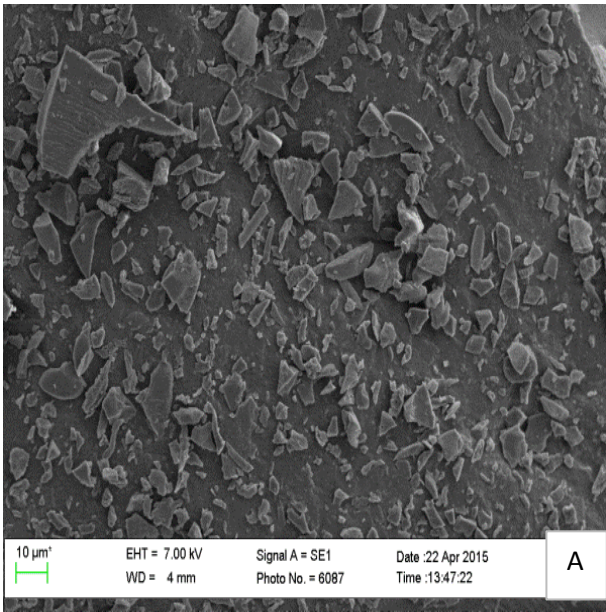


Figure 5.3 Scanning electron micrographs of Bambara groundnut powdered yoghurt – **A:** BGNPY-RM, **B:** BGNPY-NRM

This may be due to the fact that like freeze dried powders, BGNPY-RM and BGNPY-NRM were obtained in a cake or flake like form and required manual grinding into a fine powder. BGNPY-RM (Fig. 5.3 A) had a rough surface, with many small cracks, resembling broken glass or flake like appearance. Meanwhile, BGNPY-NRM (Fig. 5.3 B) had smoother surface compared to BGNPY-RM. This may be linked to the fact that BGNPY-RM was produced from BGN milk powder diluted with water, which may have resulted in reduced total solids. On the other hand BGNPY-NRM was produced from fresh BGN milk likely to have higher total solids. In addition Fyfe *et al.* (2011) reported that low solid feed results in an increased indentation and collapsed surface structure whereas high solid feed results in a smoother surface. However, Koç *et al.* (2014) stated that a porous surface is expected to be advantageous for improved wetting and solubility. The surface structures of BGNPY-RM and BGNPY-NRM were more porous than smooth, thus increasing the potential hydration properties of BGN powdered yoghurts.

2 *Water activity of BGN powdered yoghurt*

The water activity (a_w) of BGNPY-R and BGNPY-NR was 0.46 and 0.40, respectively. The a_w of the BGNPY-R and BGNPY-NR differed significantly ($p < 0.05$). The exact same processing was employed to obtain the yoghurt powders. However, processing was carried out on different days; therefore the difference in water activity may be due to variation in relative humidity of the environment, which may have an effect on moisture content of sample being dried. In addition BGNPY-R was produced from reconstituted BGN milk powder and BGNPY-NRM was produced from fresh BGN milk. In the process of reconstituting various factors such as temperature and vigorous agitation through mixing may be linked to the difference in water activity of the BGNPY yoghurts. Fontana & Campbell (2004) also stated that temperature may cause water activity fluctuation to a food material due to changes in water binding, dissociation of water and sample matrix.

3 *Colour characteristics of BGN powdered yoghurt*

The descriptive analysis for colour determination of BGNPY-RM and BGNPY-NRM is displayed in Table 5.5. The lightness (L^*) redness (a^*), yellowness (b^*), chroma (C^*) and hue angle (h°) for BGNPY-RM and BGNPY-NRM was 85.63; 85.61, 2.81; 1.86, 5.42; 4.98, 6.11; 5.32 and 62.60°; 69.55°, respectively. There was no significant ($p > 0.05$) difference on the lightness of BGNPY-R and BGNPY-NR; while the redness, yellowness, chroma and hue angle of BGNPY-R and BGNPY-NR were significantly ($p < 0.05$) different. The powdered yoghurts (BGNPY-R and BGNPY-NR) were observed to be light, with lightness value of 85.63 and 85.61 respectively, which is much closer to $L=100$ indicating lightness on the colour scale. BGNPY-R and BGNPY-NR had hue angle of 60.60° and 69.55°, respectively.

Table 5.5 Colour characteristics of BGNPY-RM and BGNPY-NRM

Powdered yoghurt	L*	a*	b*	C*	h°
BGNPY-RM	85.63 ± 0.67 ^a	2.81 ± 0.16 ^a	5.42 ± 0.06 ^a	6.11 ± 0.10 ^a	62.60 ± 1.23 ^a
BGNPY-NRM	85.61 ± 0.25 ^a	1.86 ± 0.09 ^b	4.98 ± 0.21 ^b	5.32 ± 0.21 ^b	69.55 ± 1.09 ^b

Mean values ± standard deviation of triplicate determinations. Mean values in the same column followed by different letters are significantly ($p < 0.05$) different, BGNPY-NR; Bambara groundnut powdered yoghurt from fresh BGNM, BGNPY -R; Bambara groundnut powdered yoghurt from reconstituted BGNMP, L* = Lightness, a* = redness, b* = yellowness, C* = Chroma, h° = Hue angle

This suggested that BGNPY had a slight yellowish colour as the hue angles were closer to hue angle of 90°, which signifies pure yellowness on the colour scale. BGNPY-R with a chroma of 6.11 can be said to be more saturated than BGNPY-NR with chroma of 5.32.

Kumar & Mishra (2004) reported colour characteristics of a mango soy fortified yoghurt powder with soy milk, buffalo milk and mango pulp. The yoghurt powder had L*(79.41), a*(4.91) and b*(23.70) colour characteristics. The L* co-ordinate was comparable to that of BGNPY-RM (L* = 85.63) and BGNPY-NRM (L* = 85.61) respectively, although BGN yoghurt powders were lighter. BGN yoghurt powders also had slight redness and yellowness colour characteristics indicated by the positives a* and b* as with soy mango fortified yoghurt powder. The mango soy fortified yoghurt with b* of 23.70 indicated a more yellowness compared to BGNPY-RM (b*=5.42) and BGNPY-NRM (b*= 4.98). The colour difference (ΔE) between BGNPY-RM and BGNPY-NRM was determined to be 1.04. This implies that the colour difference between the powdered yoghurts is perceivable, as Sharma (2004) described a colour difference of 1 as a just noticeable difference (JND).

5.10.6 Microbiological Characteristics of BGN Powdered Yoghurt

1 Total plate count in BGNPY-RM and BGNPY-NRM

BGNPY-RM had aerobic bacterial counts of 15.23 log cfu/mL, while BGNPY-NRM had cluster of colony growth. Presence of aerobic organisms can be an indication of poor hygiene during processing, which may result in reduced shelf life (Odumeru & Belvedere, 2002). However, total plate count colonies are also an indication of collective enumeration of all mesophilic bacteria including lactic acid bacteria, yeast and moulds and total coliforms.

2 Total coliforms in BGNPY-RM and BGNPY-NRM

There were no coliforms detected in BGNPY-RM, while BGNPY-NRM had bacterial counts of 3.03 log cfu/mL. Total coliforms bacteria are often used as indicator for the presence of pathogenic bacteria; their occurrence suggests external contamination (Yin & Ding, 2009). According to SSFF (2011) a fermented vegetable powdered beverage should not have growth of total coliform counts, thus, making BGNPY-NRM unsafe for consumption.

3 Yeast and moulds in BGNPY-RM and BGNPY-NRM

There was no detection of spoilage yeast and moulds in BGNPY-RM however, BGNPY-NRM had yeast and moulds counts of 3.59 log cfu/mL. Yeast and moulds are described to be psychrotrophic troublesome spoilage organisms, which can produce large amounts of extracellular hydrolytic enzymes that can break down protein, carbohydrates and fat molecules. This may have an adverse effect on the shelf life of a product (Ledenbach & Marshall, 2009). BGNPY-RM and BGNPY-NRM showed little growth of yeast and moulds,

which may signify a longer shelf life period for BGN powdered yoghurt under appropriate storage conditions.

4 *Lactic acid bacteria in BGNPY-RN and BGNPY-NRM*

The starter viable counts of BGNPY-RM before drying was 7.65 log cfu/mL and after drying was 7.20 log cfu/mL. While BGNPY-NRM had 8.28 log cfu/mL starter counts before drying and 5.22 log cfu/mL after drying. The number of microbial counts was higher in fresh yoghurt, compared to the powdered yoghurt. The foam-mat drying process decreased the lactic acid bacterial counts of BGNPY-RM and BGNPY-NRM. It is known that heat treatment conditions have the capacity to inactivate microbial organisms (Rascon-Diaz *et al.*, 2012) due to the metabolic injury of cells (Kumar & Mishra, 2004). The decrease in the lactic acid bacterial counts may be due to the drying conditions of 50°C for 24h, due to extended drying duration.

On the other hand, the observed survival of starter counts may be due to the presence of *S. thermophilus*, a gram positive cocci organism, which is known to be heat stable, because of the additional bonds (disulphide linkages, hydrogen bonds, hydrophobic bonds and ionized group interactions) providing that stability to the secondary and tertiary structure of protein (Kumar & Mishra, 2004). Rascon-Diaz *et al.* (2012) reported viable bacteria from spray dried natural yoghurt and yoghurt incorporated with mixture of carrageenan and guar gum both made from cow's whole milk and non-fat dried milk, inoculated with 1% of active cultures (*L. bulgaricus* and *S. thermophilus*). Natural spray dried yoghurt and yoghurt incorporated with carrageenan and guar gum had total viable count of 8.39 log cfu/mL and 10.27 log cfu/mL after drying, respectively. This was much higher than that of BGNPY-RM (7.2 log cfu/mL) and BGNPY-NRM (5.22 log cfu/mL) after drying. However, may be linked to inoculation levels (0.01% /100 mL) in BGNPY-RM and BGNPY-NRM.

5 *Staphylococcus aureus sanitary characteristics of BGNPY-RM and BGNPY-NRM*

There was no detection of *S. aureus* pathogenic bacteria in BGNPY-RM and BGNPY-NRM. *S. aureus* is known to be associated with foods that require manual handling as well as food that have been insufficiently heated (Lee *et al.*, 2015). Reconstituted BGNM and non-reconstituted BGNM used to prepare the BGNPY-RM and BGNPY-NRM were pasteurised by heating to 100°C for 20 min. Furthermore, reconstituted BGNM was obtained through spray drying at high temperatures of 150°C [Section 3.2.5, page 48]. It can be said that the production of BGNPY-RM and BGNPY-NRM incorporated adequate heat treatment. In addition *S. aureus* is reported to occur at water activity range of 0.83-0.9 (Danielsson-Tham, 2013; Lee *et al.*, 2015), and the water activity of BGNPY-NR and BGNPY-R ranged from 0.40 to 0.46, respectively [Section 5.10.5 (2), page 141]. Therefore, these conditions inhibit the growth of *S. aureus*, a pathogenic bacterium.

6 *Escherichia. coli* sanitary characteristics of BGNPY-RM and BGNPY-NRM

E. coli pathogenic bacteria were not detected in BGNPY-R and BGNPY-NR. The presence of *E. coli* strains in the human gut is known to cause intestinal infections including diarrhoea, haemorrhagic colitis and neonatal meningitis (Badri *et al.*, 2009). Sangadkit *et al.* (2012) stated that the detection of *E. coli* may also suggest contamination from direct or indirect faecal origins of humans. Therefore, the absence of *E. coli* in BGNPY-RM and BGNPY-NRM implies good manufacturing and sanitation practices during its production. Furthermore, BGNPY-RM and BGNPY-NRM may be ingested without causing infection in the human gut due to *E. coli*.

5.10.7 Sensory characteristics of Bambara groundnut cultured drink

The demography of the panellists who partook in the sensory study is detailed in Table 5.6. There were 48 panellists consisting of 22.9% staff and 77.1% students, which of 4.2% were international students and 95% local students. There were 39.6% males and 60.4% females, with the following age categories; less than 20-29 years (87.5%), 30-39 years (10.4%) and 40 and above (2.1%). Multivariate analysis indicated the rating amongst panellist for appearance, colour, aroma, taste, texture and overall acceptability differed significantly ($p < 0.05$). The difference was highly expected because the panellists comprised of individuals from diverse backgrounds. The panellists mean ratings for appearance, colour, aroma, taste, texture and overall acceptability of BGNCD from fresh BGN milk and BGNCD from reconstituted BGN milk powder were 3.31;3.65, 3.31;3.42, 3.37;3.98, 2.67; 2.67, 2.85; 3.31, and 2.90; 3.23, respectively illustrated in Figure 5.4.

The two BGNCD samples were significantly ($p < 0.05$) different with respect to appearance, texture and overall acceptability, while the colour, aroma and taste were not seen as significantly ($p > 0.05$) different. The appearance of BGNCD from reconstituted BGNMP (3.65) was significantly ($p < 0.05$) higher as compared to that of BGNCD from fresh BGNM (3.31). Appearance of the yoghurt has been described to have a smooth, creamy body (Lee & Lucey, 2010). According to the panellists, BGNCD from reconstituted BGNMP was perceived as superior with respect to creaminess and smoothness, compared to BGNCD from fresh BGNM. BGNCD from reconstituted BGNMP was again significantly ($p < 0.05$) higher with regards to texture than BGNCD from fresh BGNM. Dairy Australi/NCDEA (2013) stated that texture of yoghurt is affected by a number of factors including viscosity, fattiness and firmness. BGNCD from reconstituted BGNMP was judged to be more viscous and firmer than BGNCD from fresh BGNM, which is highly dependent on the gel formed strength of a yoghurt. Lee & Lucey (2010) further elaborated that the gel strength of yoghurt may be affected by the total protein and fat content of a formulation system, which has an impact on its final total soluble solids. The superior viscous and firmer texture of BGNCD from reconstituted BGNMP (3.31) may be due to the higher protein (7.6%) and fat (1.6%)

Table 5.6 Demography of panellists

Item	Frequency (%)
Gender	
Male	19 (39.6)
Female	29 (60.4)
Age	
< 20-29	42 (87.5)
30-39	5 (10.4)
40 & above	1 (2.1)
Staff or student	
Staff	11 (22.9)
Student	37 (77.1)
International student	
Yes	2 (4.2)
No	46 (95.8)

*Numbers are frequency and percentage in bracket

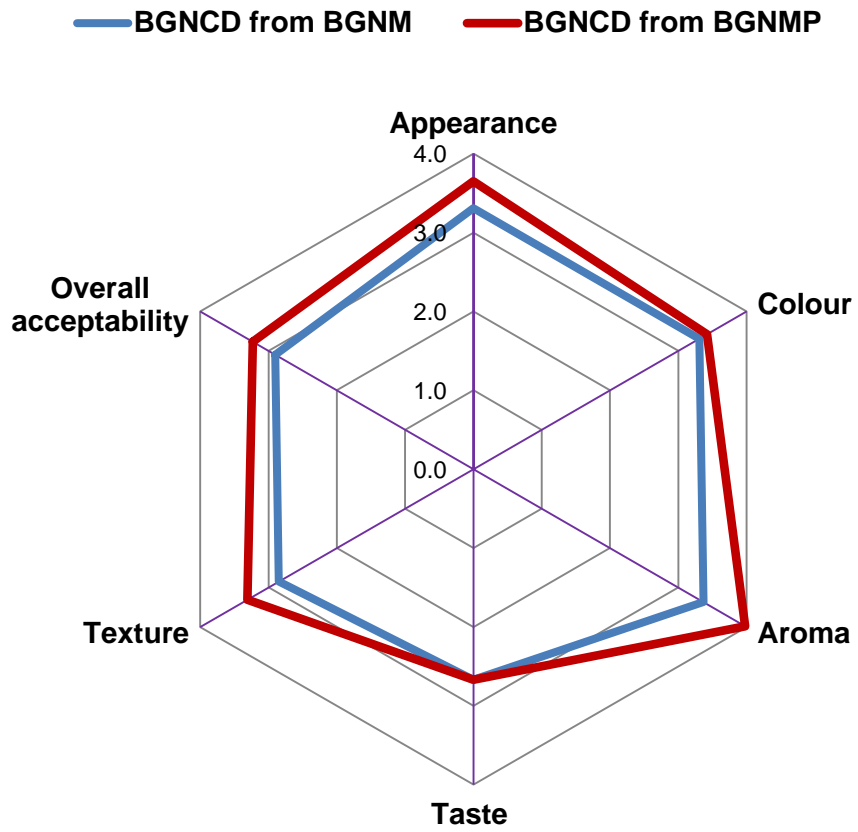


Figure 5.4 Mean sensory scores of Bambara groundnut cultured drink made from fresh Bambara groundnut milk and reconstituted Bambara groundnut milk powder.

of BGNMP as compared to that of BGNM protein (0.9%) and fat (0.3%) [Section 3.8.4, page 77].

The overall acceptability of BGNCD from reconstituted BGNMP (3.23) was rated higher compared to that of BGNCD from fresh BGNM (2.90). The overall acceptability of yoghurt has been reported to be affected by physical attributes such as syneresis and perceived viscosity (Lee & Lucey, 2010). BGNCD from reconstituted BGNMP had higher ratings for appearance and texture, which may be attributed to the consistency or viscosity of yoghurt. This resulted in resistance to flow in the mouth, perceived amount of grease/fat in the mouth and the solid compact sensation that holds its shape in the mouth. Therefore, making BGNCD from reconstituted BGNMP more superior than BGNCD from fresh BGNM in its overall acceptability. The colour, taste and aroma of the yoghurt were seen as the same. This is because the flavourant and sweetener (sugar) introduced into the samples were identical with respect to quantity and type. Out of the 48 panellists who partook in the sensory study, 56.3% found BGNCD made from fresh BGNM to be valid with respect to its flavour. The remaining 43.7% described BGNCD from fresh BGNM to be thin, runny watery with regards to consistency. The panellists (43.7%) further described BGNCD from fresh BGNM to lack sweetness, volume, mouth feel and to have a beany taste. Panellist (60.4%) ranked BGNCD from reconstituted BGNMP acceptable. The other panellists (39.6%) defined BGNCD from reconstituted BGNMP to lack sweetness, too sour, to have a beany taste and after taste. On the other hand some members of the panellist found BGNCD from reconstituted BGNMP to have a nice thick texture and aroma.

5.11 Conclusion

BGN powdered yoghurt was produced from BGN milk and BGN milk powder employing the foam-mat drying processing technology. The powdered yoghurt showed favourable rehydration properties necessary for reconstitution, with WSI of 73.3% (BGNPY-RM) and 71.2% (BGNPY-NRM). The powdered BGN yoghurts also exhibited a fairly satisfactory nutritional profile, with total dietary fibre of 3.9% and 4.2%, protein (4.8% and 5.7%) and total fat (1.6% and 1.4%) for BGNPY-R and BGNPY-NR, respectively. However, it is recommended that the physical properties of powdered yoghurt including texture, gel structure and flavour be assessed closely, as this study did not focus on those aspects.

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CHAPTER SIX

GENERAL SUMMARY AND CONCLUSIONS

The functional and nutritional characteristics of Bambara groundnut (BGN) milk powder as an ingredient in yoghurt production were investigated in this study. The following objectives were identified in this study:

1. Production and characterisation of Bambara groundnut milk powder (BGNMP) with a view to establish its use in BGN yoghurt.
2. Evaluation of the physicochemical, rheological and microbiological characteristics of Bambara groundnut cultured drink (BGNCD).
3. Optimisation of BGNCD from BGNMP for the production of BGN powdered yoghurt employing the foam-mat drying process with a view to establish its functional and nutritional characteristics.

The first objective was achieved as BGNMP was successfully produced using the spray drying process employing maltodextrin as a drying carrier at three levels (5, 10 and 15%) and consequently characterised for functional and nutritional properties. BGNMP was characterised by high water solubility index, which is an important feature for milk powders as poorly soluble powders can cause difficulty during processing resulting in economic losses. Furthermore, the bulk density and particle size distribution of the BGNMP was comparable to various commercial skim milk powders. Bulk density and particle size are economically and commercially important properties of milk powders therefore; making BGNMP a good competitor to other milk powders in the market. The low water activity of BGNMP indicated its overall potential stability. BGNMP was high in protein and a good source of fibre which is important for its use in BGN yoghurt. The hypothesis that, the production of BGNMP from BGN seeds will be possible was accepted.

The second objective was achieved by investigating the physicochemical, rheological and microbiological properties of BGN cultured drink. The high fibre, low fat and low total sugars of BGNCD indicates its potential as a low fat diet food. The pH and acidity of BGNCD indicated its potential of being a fermented drink. The elastic (G') properties of BGNCD was positioned above viscous (G'') properties which is important in a yoghurt indicating presence of a gel structure and suggest that the structure of BGNCD system was stable. The absence of pathogenic bacteria in BGNCD revealed its potential long life storage. Therefore, nutritional, rheological and microbiological properties of BGNCD revealed its potential as a cultured drink.

The third objective which included the optimisation of BGNCD for the production of BGN powdered yoghurt from BGN milk and BGNMP employing the foam-mat drying process was achieved using combination of gum arabic (6%) and methylcellulose (0.5%). The high glass transition temperature and low water activity of BGN powdered yoghurts indicated their

resistance against caking and stickiness and overall potential stability. BGN powdered yoghurts were characterised by high bulk density, which is beneficial when considering packaging, handling and storage costs. Both BGN powdered yoghurts showed high water solubility indices, wettability and lightness which is beneficial for its incorporation in dry mixes for convenience foods including dry dessert mixes, soup bases and instant drink mixes.

The following conclusions can therefore be drawn from this study:

1. BGN milk powder can be produced from BGN seeds
2. BGN milk powder has beneficial functional and physical properties which are comparable to commercial milk powders
3. BGN milk powder can be successfully used as an ingredient to produce BGN cultured drink as alternative to normal milk
4. BGN cultured drink produced using BGN milk powder was acceptable to consumers
5. BGN powdered yoghurt can be produced employing the foam-mat drying method; however physical attributes including texture and taste require some improvement.
6. Literature information from this study was contributed to a review article titled "Foam-mat Drying Technology: A Review".

APPENDICES

**Appendix A: Manuscript Accepted for Publication in Critical Reviews in Food Science
& Nutrition titled; Foam-mat Drying Technology**

Appendix B: Peer reviewed research paper titled; Effect of spray drying compartments on the functional, physical, thermal and nutritional characteristics of Bambara groundnut milk powder

Appendix C: Book of Abstracts – Research output presented at national and international conferences

1. **Hardy, Z.** & Jideani, V. A. (2014). Foam-mat Drying Technology: A Review. Unmoderated poster presentation. In: Book of Abstracts-Research That Resonates. IUFOST 17th World Congress of Food Science and Technology & Expo MP.3 Montreal, Canada.
2. **Hardy, Z.** & Jideani, V.A. (2014). Stability of Bambara Groundnut (*Vigna subterranean*) Milk. Poster presentation. In: Book of Abstracts-Research and Innovation for sustainable development. p 101. U6 Consortium 2nd International Conference, Cape Town, South Africa.
3. **Hardy, Z** & Jideani, V.A (2015). Functional and nutritional characteristics of Bambara groundnut (*Vigna subterranean*) milk powder. Oral presentation. In Programme and Book of Abstracts-Product Development/New Products. 21st SAAFoST Biennial International Congress and Exhibition, 2015, Durban, South Africa.
4. **Hardy, Z** & Jideani, V.A (2015). Production of Bambara groundnut cultured beverage. Pre-Conference Workshop for Students/Project Idea Description for Commercialisation. Eighth Annual International Conference, Entrepreneurship Education for Economic Renewal, 2015, Science Park, Vaal University of Technology, Sebokeng, South Africa.

Appendix D: Approved Ethics Clearance