

**PROCESS AND PRODUCT CHARACTERISTICS OF A CANNED  
PEANUT SOUP BASE**

**BY**

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**THIS THESIS IS SUBMITTED TO THE DEPARTMENT OF  
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## DECLARATION

This is to certify that this thesis is the result of research work undertaken by Mwinmaalu Achillis Dongdem towards the award of the MPhil. Food Science degree at the Department of Nutrition and Food Science, University of Ghana Legon.

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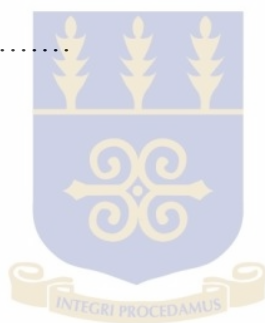
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## ABSTRACT

The most important processed form of peanuts in Ghana is peanut paste. The predominant use of this peanut paste is for the preparation of soup. This soup is a traditional delicacy enjoyed in most countries of West Africa. However, peanut soup preparation is time consuming and the soup has poor keeping qualities. Furthermore, the safety of peanut soup is of great concern because of perceptions of adulteration of the paste on the Ghanaian market and the high probability of aflatoxins contamination of peanuts. This study aimed to develop a process for a safe and consumer acceptable canned peanut soup base and to evaluate the product characteristics.

A suitable variety of peanuts (i.e. *Chinese* variety) was selected from among four most popular local varieties (Manipintar, Chinese, Sinkarzie and Bugla local). The selection was based on proximate composition, and variability in physical characteristics (seed dimensions and bulk density) as well as availability of the variety of peanuts. The manual color sorting method to minimize aflatoxin levels in peanuts was used and the aflatoxins levels monitored using HPLC procedures. To determine the optimum proportions of secondary ingredients required for the soup, eight (8) ingredient formulations were generated using mixture designs procedures (extreme vertices option) for the three secondary ingredients (pepper: 5 – 15g, Onion: 100 – 180g, tomato paste: 75 – 150g). The ingredient formulations were evaluated by 48 panelists in an optimization study and validated to obtain the most acceptable secondary ingredient formulation for peanut soup base. A 3 X 3 full factorial design for retort processing temperature (115, 120, 125°C) and processing time (40, 50, 60mins.) was used to determine the most adequate thermal process for a canned peanut soup base.

The physical characteristics of the peanuts showed significant difference among varieties ( $p < 0.05$ ). The *Chinese* variety was selected for the project because it is the predominant variety in major markets of Greater Accra Region of Ghana and also the most cultivated variety

among peanut farmers. Its seed dimensions - width and length - were also most variable (standard deviations  $\pm 0.62\text{cm}$  and  $\pm 2.27\text{cm}$ ) compared to the other varieties which made it a better choice to be milled into peanut paste rather than used for products that may require uniformity in seed dimensions. Total peanut weight loss due to the colour sorting was high (25%) however the total aflatoxin concentration of the *clean* sorted peanuts was below  $4\mu\text{g}/\text{kg}$ , which is one of the strictest maximum limit of total aflatoxins in peanuts acceptable in some Countries. The most suitable secondary ingredient formulation for peanut soup base was determined to be 12.30g pepper, 100.80g onion and 146.90g tomato paste per 300g of peanut paste. The limits of the thermal process for canned peanut soup base were established to range from  $122^{\circ}\text{C}$  to  $124^{\circ}\text{C}$  for 51 to 44mins respectively to achieve shelf-stability. The estimated sterilizing values ( $F_0$ ) under the thermal process conditions ranged from 0.04 to 1.92mins. The minimum thermal process treatment of  $115^{\circ}\text{C}$  for 40mins recorded the highest plate counts of 10,000cfu/g for APC and also 10,000cfu/g for Yeasts and Moulds Count. Peanut soup base subjected to the severest thermal process ( $125^{\circ}\text{C}$  for 60mins) yielded no observable growth (at dilutions  $10^2\text{ml}$ ) for both APC and Yeast and Moulds count.

Findings from this study show that a virtually aflatoxin-free peanut soup base, acceptable to consumers can be obtained by manually sorting peanuts, roasting to an appropriate colour, grinding into a smooth paste, mixing with predetermined proportions of secondary ingredients, and subjecting it to an established thermal process schedule.

## DEDICATION

I dedicate this thesis to my nephews Maximillian and Chrispine and my nieces Kesiah and Anthonia. May you grow in wisdom and stature.



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## CHAPTER ONE

### 1.0 INTRODUCTION

#### 1.1 Peanuts and peanut products

Peanuts (*Arachis hypogaea*) also known as groundnuts, believed to have originated from South America, was introduced by the Portuguese to West Africa (Nautiyal, 2002). They are cultivated in almost all the agro-ecological zones of Ghana largely as a subsistence crop (Atuahene-Amankwa *et al.*, 1990). In the last decade peanut production in Ghana has more than doubled, from 208,600 mt in 2000 (MOFA 2002) to 485,100 mt in 2009 (MOFA, 2010). There are many different varieties of peanuts cultivated in Ghana. Asibuo *et al.* (2008) characterized twenty varieties of peanuts that are cultivated in Ghana. Some of the most popular varieties include Manipintar, Chinese, and Sinkarzie which can be found in most Ghanaian markets.

Peanuts are an important source of income for farmers. They are also an inexpensive source of high quality dietary protein (22 to 30% protein) (Savage and Keenan, 1994). They contain 44 to 56% oil and are a rich source of minerals (phosphorus, calcium, magnesium, and potassium) (Savage and Keenan, 1994). They are a useful material in food fortification programs geared towards curbing protein-energy malnutrition (PEM) (Asibuo *et al.*, 2008). Peanuts are also a useful raw material in many food applications. They are eaten as whole seeds (freshly harvested, dried, boiled or roasted) or processed into peanut butter, oil, soups, stews and other products. According to Jolly *et al.* (2008), peanuts, processed as paste for making peanut soup is the highest (39.3%) form of peanut utilization in Ghana.

## 1.2 Developing convenience staples

The development of new products is an important business strategy for the expansion and survival of food industries. Food processors continue to innovate and develop new products to meet the changing needs and desires of consumers. In order to cut down production cost, improve the utilization of locally produced agricultural produce, as well as boost local consumer confidence and interest, it is important to develop competitive products using locally produced crops to satisfy the needs of consumers.

In Ghana as well as many other African countries, most traditional staples require long periods of time to prepare (e.g. fermented foods such as *kenkey*, *banku*). A great deal of time is spent on raw material preparation. However, a growing number of people are spending longer hours at work and away from home, and cannot afford the time, and the drudgery involved in the traditional process of preparing food. Consequently a niche market has evolved for convenience and ready to eat foods. To satisfy this market, research efforts have led to the development of some convenience staple products such as canned palm fruits extract, instant porridge flour, *fufu* flour and *banku* flour. These food products have found wide consumer acceptance locally and abroad. Another staple which can also be processed into an acceptable convenience staple is peanut soup, which is very popular among many West African consumers.

Peanut soup is eaten with other staples such as *tuo zaafi*, *boiled rice*, *rice balls*, *banku* and *fufu*. It is prepared using peanut paste, water, tomatoes, and a variety of spices including ginger and pepper and of course with meat or fish. The traditional method of peanut soup processing includes unit operations such as sorting of the peanuts to eliminate moldy, broken, shrivels and pegs, roasting, deskinning and milling into smooth peanut paste, ingredient

mixing and boiling (Sebigbon, 2010). In the traditional process the entire process could take a few hours.

In an attempt to reduce processing time and assure good quality peanut soup, the process of developing a canned peanut soup base was undertaken in this study. A prior survey conducted in Accra by Kotey (2010) indicated that 81% of working class consumers are willing to patronize a peanut soup base product. The survey also revealed that 55% of the consumers preferred a canned peanut soup base which blends peanut pastes, tomatoes and spices. The product was thus conceived as a concentrated blend of tomato paste, peanut paste, a spice-mix and water. The canned product should be reconstituted within a relatively short time into peanut soup with added meat or fish and water. In general, the concept of canning peanut soup base is purported at offering convenience because of the ease of handling and cooking.

### **1.3 Rationale**

The process of peanut soup making, starting from the raw peanuts, is cumbersome and time consuming. This poses a challenge to an increasing number of workers who spend long hours away from home. In addition to having poor keeping qualities, the safety of peanut soup is also of great concern. This is because peanut paste which is readily available in Ghanaian markets is perceived to be adulterated (Amaditor, 2010). Additionally, aflatoxin contamination of peanuts and peanut products has been of enormous concern globally because of their adverse (e.g. carcinogenic) effects on human health (Massey *et al.*, 1995). The human health hazard concern is obviously a distraction to the consumption of peanut soup as a staple and could pose serious threats to maximizing the utilization of peanuts as a food commodity, if not properly addressed. A canned peanut soup base, with guaranteed non-

detectable aflatoxins would earn consumer confidence and enhance consumer patronage of the product.

This study therefore seeks to develop a canned peanut soup base that will be safe and convenient for use by consumers. The development of the product will promote the consumption of peanut soup and assure its safety in terms of microbial and aflatoxins contamination. It will also maximize the utilization of peanuts and provide a ready market for peanut farmers.

#### **1.4 Main objective**

To develop a process for a shelf-stable canned peanut soup base and evaluate the product quality characteristics.

##### **1.4.1 Specific objectives**

The specific objectives were to:

- i. Select the most suitable peanut variety for making peanut soup base by determining the physico-chemical characteristics of four commonly cultivated peanut varieties in Ghana,
- ii. Evaluate the efficacy of the sorting process to minimize aflatoxins in peanuts for use in the canned peanut soup.
- iii. Determine the optimum ingredients formulation for canned peanut soup base.
- iv. Establish the thermal process variables (temperature and time) and evaluate the quality of the canned peanut soup base.

## CHAPTER TWO

### 2.0 LITERATURE REVIEW

#### 2.1 Origin and distribution of peanuts

Peanuts botanically known as *Arachis hypogaea* Linn belong to the fabaceae family. Peanuts mature between 90 to 135 days from the sowing date depending on the variety (Singh and Oswalt, 1995). They vary in testa colour (red, brown, purple, white), shape, size and length depending on the variety of peanuts and environmental factors (Ahmed and Mohammad, 1997).

According to Naturland (2000), peanuts have been cultivated domestically for over 4000 years. They derive their origin in Eastern Bolivia and spread to the surrounding South American countries including Brazil, Uruguay and North-Western Argentina (AFF, 2010). The Spanish traders took the crop to Europe from Brazil in 1784 (Smart, 1992). The Portuguese traders however, introduced peanuts from Brazil to West and East Africa and then to South-Western India in the 16th century (Nautiyal, 2002). Through slave trade the crop was taken much later to USA from West Africa (Bunting, 1958).

Peanuts are now widely cultivated in most parts of the world. They are cultivated in the tropical, subtropical, and temperate countries between 40°N and 40°S (Cummins and Jackson, 1982). The worlds' leading producers of peanuts are China, India, Nigeria and USA. In Africa, other important peanut growing countries include Sudan, Senegal, Chad, Congo and Ghana. Table 1 depicts the Food and Agriculture Organisation's (FAO) records of shell peanut production for some selected countries for the 2010 crop season.

**Table 1: FAO world rankings of shell peanut production for 2010 crop season.**

Country	Total Production (MT)	World Ranking
China	15,709,036	1
India	5,640,000	2
Nigeria	2,636,230	3
United States of America	1,885,510	4
Senegal	1,286,860	5
Indonesia	779228	7
Sudan	762500	8
Argentina	611040	9
Ghana	530887	10

Adapted from FAOSTAT (2010)

## 2.2 Peanut varieties

There are many varieties and cultivars of peanuts. Nurland (2000) broadly categorized peanuts into two (2) subspecies namely, the *hypogaea* and the *fastigiata*. The *hypogaea subsp.* is defined to include two botanical varieties the *hypogaea* (Virginia type) and *hirsuta* (Runner type) while the *fastigiata subsp.* includes the *fastigiata* (Valencia type) and *vulgaris* (Spanish type) base on their similarity in some characteristics (Singh and Oswalt, 1995). However, some other researchers preferably categorize peanuts into four distinct commercial/botanical varieties - *hypogaea*, *hirsuta*, *fastigiata* and *vulgaris*.

Morphologically, the subspecies *hypogaea* do not develop flowers on the main stem, but have alternate branching and flowering patterns (Asibuo *et al.*, 2008) and a longer vegetation period (Nurland, 2000; Singh and Oswalt, 1995). However the *hypogaea* (Virginia type)

type is less hairy with shorter branches compared to the *hirsuta* (Runner type). On the other hand, the subspecies *fastigiata* bear flowers on the main stem, exhibit sequential branching and flowering patterns and have a relatively shorter vegetation period (Asibuo *et al.*, 2008). The *fastigiata* (Valencia type) is however little branched compared to the *vulgaris* (Spanish type) (Singh and Oswalt, 1995).

The pods and seed sizes also differ among the four (4) identifiable commercial subspecies. While the Virginia-type peanuts have the largest pods and elongated seeds, the Runner-type peanuts have medium size seeds, Spanish-type peanuts have smaller round seeds, and Valencia-type peanuts have intermediate sizes and shapes (Singh and Oswalt, 1995; Kolan, 2010).

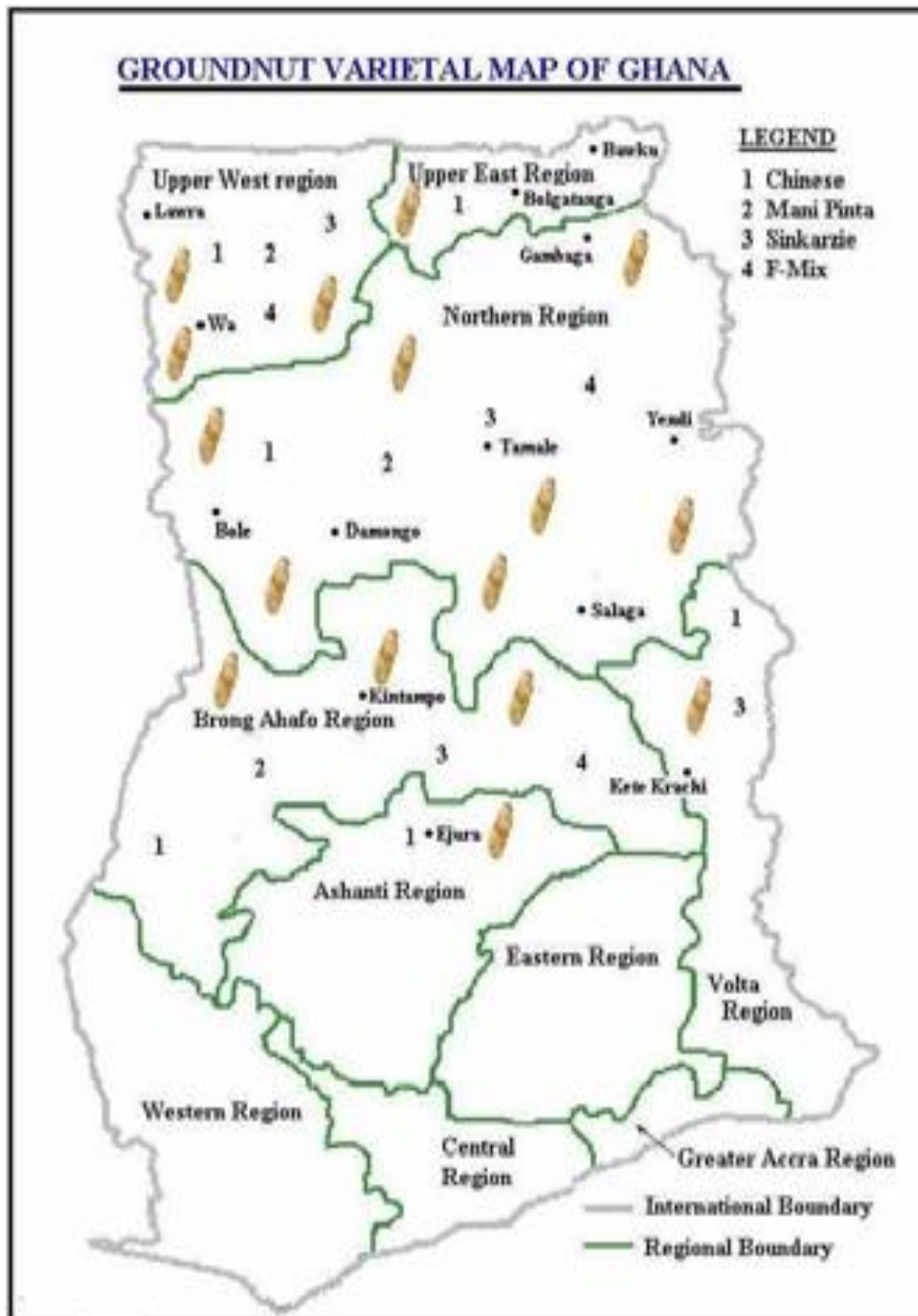
### **2.2.1 Peanut production and varietal distribution in Ghana**

Peanut production in Ghana has doubled in the last decade. The production of peanuts increased from 208,600 mt in 2000 (MOFA 2002) to 485,100 mt in 2009 (MOFA, 2010). This significant leap in production in 2009 has been attributed to increases in the area under cultivation and the average yield. The average yields of peanuts have increased to 1.5mt/Ha in 2009 from 1mt/Ha in 2002 although it still falls below the estimated potential 2mt/Ha. The area under cultivation also increased from 285,800ha in 2000 to a peak of 448,800ha in 2003/2005 but declined to 342,900ha in 2007/2009 (MOFA, 2010). FAOSTAT (2010) report of shell peanut production for the 2010 crop season peaked shell peanut production in Ghana at 530887mt.

Although peanuts are grown in all the agro-ecological zones of Ghana, the Northern sector of Ghana (Upper West, Upper East and Northern regions) produces the bulk of peanuts

consumed in the country (Atuahene-Amankwa *et al.*, 1990). The Northern sector falls within the Guinea savannah and Sudan savannah agro-ecological zones. The Northern region alone produces over 80% of the total peanuts produced in Ghana (Tsibey *et al.*, 2003). They are largely grown by small-scale farmers who dominate the agricultural sector of the country (MOFA, 2010).

In Ghana, Asibuo *et al.* (2008) identified 20 varieties and/or cultivars of peanuts and categorized them under *fastigiata* and *hypogaea* subspecies using their morphological features. Peanut varieties such as F-mix, Manipintar and Sinkarzie were classified under the *hypogaea* subspecies while Shitaochi (Chinese) and Kamaloo were classified under the subspecies *fastigiata*. Fig. 1 illustrates the varietal distribution of commonly cultivated peanut varieties in Ghana.



Source: Tsigbe *et al.* (2003)

**Fig. 1: Commonly cultivated peanut varieties and their distribution in Ghana.**

## **2.3 Importance of peanuts**

Peanut plays important roles in the economies of many countries. They are important as a cash crop and an important raw material for many industries. Their roles in meeting the nutritional needs of consumers and as an important agricultural crop capable of replenishing soil fertility cannot be overemphasized.

### **2.3.1 Nutritional significance**

Peanuts are an important source of inexpensive dietary nutrients. The nutritional composition of peanuts varies depending on the variety (Asibuo *et al.*, 2008), ecological factors (geographic and climatic factors) and the stage of maturity during harvest (Pickett, 1949; Brown *et al.*, 1975).

According to Asibuo *et al.* (2008), the carbohydrate content of 20 identified Ghanaian peanut varieties ranged from 19.02 – 27.16%. Ng and Dunford (2009) found that sucrose is the major sugar in peanuts (about 90% of the total sugars) while glucose and fructose were minor components. Peanuts are also known to be a good source of minerals. The potassium, sodium, calcium and magnesium contents of different Ghanaian peanut varieties were found to be significantly different (Asibuo *et al.*, 2008).

Asibuo *et al.*, (2008) reported that of the 20 Ghanaian peanut cultivars studied the protein content ranged from 18.92 – 30.53%. Peanuts are also reported to have a rich amino acid profile. Chopra and Sidhu (1967) analysis of the amino acid composition of peanuts indicated significant differences due to variety for total N, serine, glutamic acid, proline, alanine, leucine, tyrosine, phenylalanine, lysine, methionine, cystine and ammonia. Peanuts play an important role in meeting the protein needs of people especially in the developing countries.

Their good protein profile and content make them invaluable in many food fortification programmes purported at minimizing protein-energy malnutrition (PEM) (Asibuo *et al.*, 2008).

Peanuts are also an important source of dietary oil. The average oil content of peanuts is about 50% (dry matter) although widely reported significant difference may exist between different varieties or cultivars. Dwivedi *et al.* (1993) observed that the mean oil content of Virginia cultivars is slightly higher than the Spanish cultivars. The oil content and quality influence greatly the acceptability of peanut products. According to Ory *et al.* (1992), peanut oil generally contains 55–65% monounsaturated fatty acids (MUFA), 26–28% polyunsaturated fatty acids (PUFA), and 17–18% saturated fatty acids. A study of the fatty acid profile of peanut oil identified six fatty acids namely: palmitic (C-16:0), stearic (C-18:0), oleic (C-18:1), linoleic (C-18:2), linolenic (C-18:3), and  $\gamma$ -linoleic (C-18:2) acids (Shad *et al.* 2012). Ahmed and Young (1982) indicated that the amount of unsaturation in peanut oils is about 80% of the total fatty acid content of peanut oil. The unsaturated fatty acids are predominantly oleic acid (MUFA) and linoleic acid (PUFA). Hence the oleic acid to linoleic acid ratio is a strong indicator of the chemistry and quality of peanut oil (Shad *et al.*, 2012). The high level of unsaturation in peanut oils promotes human health by minimizing the risk of coronary heart diseases (Shad *et al.*, 2012).

Even though peanuts have a high energy density (5.9 kcal/g), epidemiological studies suggest an inverse relationship between the frequency of nut intake and body mass index (BMI) (Ellsworth *et al.*, 2001; Fraser *et al.*, 1992). High peanut intake was found to have limited effect on body weight gain (Alper and Mattes, 2002; O'Byrne, 1997). These observations have been attributed to the high levels of unsaturated fatty acids of peanut oils. The high

unsaturation has variously been explained to offer peanuts a strong satiety effect, potent appetite suppressor effect, and high fatty acid oxidation. Some reports suggest that the high satiety effect of peanut consumption provides a strong dietary compensatory effect which leads to a spontaneous cut in food (energy intake) at other times of the day and consequently a limited weight gain (Alper and Mattes, 2002; Fraser *et al.*, 1992). Similarly its appetite suppressor effect also leads to a subsequent reduction in energy intake during day (Greenberg *et al.*, 1990; Cox *et al.*, 2004). However, other accounts indicate that the high oxidation of UFA makes it less likely that they would be stored to contribute significantly to body weight (Jones, *et al.*, 1985). Some intervention trials have however failed to support this inverse association between frequent peanut consumption and BMI in unrestrained eating populations (Akuamoah-Boateng *et al.*, 2007).

### **2.3.2 Agricultural significance of peanuts**

Peanuts are an important cash crop and an essential source of income for peanut farmers the world over. They are also an important industrial raw material and are processed into a variety of food products such as roasted peanuts, peanut butters, peanut oil and other composite peanut products (Yao, 2004). Apart from being an important human food, the stalks, leaves, nuts and shells serve as animal feed. They are also used as materials for making compost fertilizer and for mulching (Yao, 2004). In crop rotation programmes they are an essential means of replenishing soil nitrogen (Konlan, 2010).

## **2.4 Peanuts utilization**

Peanuts are consumed in various forms depending on the food preferences and eating habits of consumers. The kernel/seed can be consumed raw, boiled, roasted, or made into peanut spreads, confectionaries and baked products (Yao, 2004). Oil is also extracted from peanut

for making cooking oils and other products such as soap, margarine, cosmetics, candles and creams (Singh and Diwakar, 1993).

Peanuts are processed into peanut milk. Usually, dehulled peanut seeds are soaked in 1% sodium bicarbonate solution for 16-18h and milled in an aqueous medium after draining of excess water (Singh and Diwakar, 1993). The wet mash is then steeped for 4-5h, and filtered through cheese cloth to extract the milk.

Also, they are processed into paste for further processing into other value added food products. The paste can be used to make peanut butter and spreads. Peanut butter is made by fine milling roasted groundnuts with added ingredients such as dextrose, salt, stabilizers (hydrogenated oil, lecithin) and antioxidants (Young and Heinis, 1989). In the USA, peanut butter is made of at least 90% peanuts and 10% added ingredients, with the oil content not exceeding 55% by weight (Weiss, 1983). However, products labeled peanut spreads contain less than 90% peanuts but their nutritional composition is equivalent to peanut butter with at least 24% protein (Young and Heinis, 1989).

In addition to these, peanuts can be used to make fermented products such as *oncom* and *tempeh* which are distinguished by the fungus used for the fermentation process (Singh and Diwakar, 1993). *Oncom* is made from peanut cake after pressing to remove the oil and fermented using either *Neurospora intermedia* (red *oncom*) or *Rhizopus oligosporus* (black *oncom*) (Singh and Diwakar, 1993). Peanut pastes are also used to make cakes which are fried in oil to produce *Kulikuli* which is eaten as a snack (Abbey *et al.*, 1989).

In Ghana and other West African countries peanut paste are used to prepare local delicacies such as peanut soups and sauces. According to Jolly *et al.* (2008) the highest form of peanut utilization in Ghana (39.3%) is for making peanut soups. Peanut soup also known as *Nkantinkwan* is prepared by boiling slurry of peanut paste with the addition of ground tomatoes and some spices together with meat or fish (Sebigbon, 2010). The other notable forms of peanuts utilization in Ghana include bread spreads (6.9%) and fresh boiled peanuts (4.5%) (Jolly *et al.*, 2008).

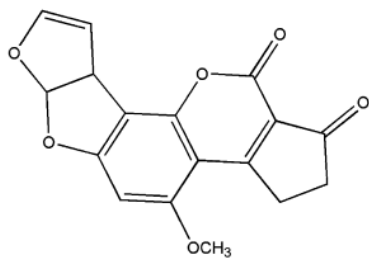
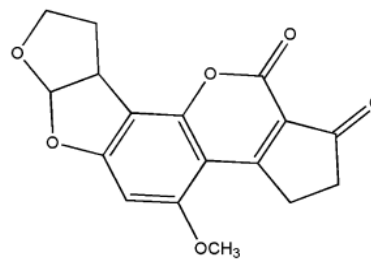
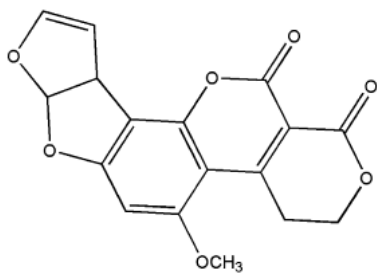
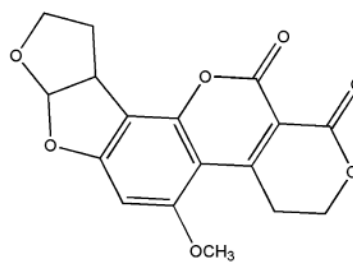
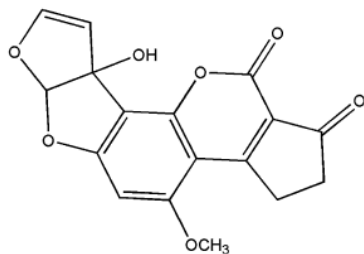
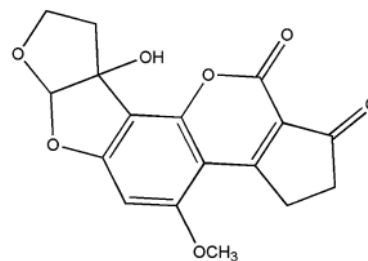
## 2.5 Aflatoxins in peanuts

Peanuts are among the agricultural produce susceptible to mycotoxin contamination. Mycotoxins are low molecular weight secondary metabolites produced by fungi which when ingested, inhaled or absorbed induce various degrees of toxicity to vertebrates, invertebrates, plants, and microorganisms (Bakole and Adebajo, 2003; Guo *et al.*, 2009). In terms of food hazards, mycotoxins are second to food-borne bacteria in humans but pose the greatest threat in livestock (Bakole and Adebajo, 2003). Five agriculturally important mycotoxins namely: aflatoxins, fumonisins, ochratoxin A, zearalenone and deoxynivalenol have been reported (IARC, 1993). The predominant mycotoxin in peanuts is Aflatoxin.

Aflatoxins (AF) were first described as ‘Turkey X disease’ in 1960 when 100,000 turkey pouts’ died in England following feeding with a peanut meal (Guo *et al.*, 2009). The meal was later identified to be infested with toxins produced by *Aspergillus flavus*, hence the name, A-Fla-Toxin (Blount, 1961).

### 2.5.1 Types of aflatoxins

There are several types of aflatoxins, but the four major types of naturally occurring aflatoxins are B<sub>1</sub>, B<sub>2</sub>, G<sub>1</sub> and G<sub>2</sub> (Guo *et al.*, 2009). There are also aflatoxins M<sub>1</sub> and M<sub>2</sub> which are hydroxylated metabolites of B<sub>1</sub> and B<sub>2</sub> respectively which are found in animal milk (Huang *et al.*, 2010). Although the fungi *A. parasiticus*, *A. niger* and *A. nomius* have been found to produce aflatoxins (Bakole and Adebajo, 2003), *A. flavus* is the commonest producer (Bradburn *et al.*, 1993). *A. flavus* produces aflatoxins B<sub>1</sub> and B<sub>2</sub>, while *A. parasiticus* produces G<sub>1</sub> and G<sub>2</sub> in addition to aflatoxins B<sub>1</sub> and B<sub>2</sub>. Aflatoxins are a major problem in the developing world and pose a great threat to the safety, quality and marketability of peanuts (Lubulwa and Davis, 1994). Fig. 2 shows the chemical structures of the six types of aflatoxins.

Aflatoxin B<sub>1</sub>Aflatoxin B<sub>2</sub>Aflatoxin G<sub>1</sub>Aflatoxin G<sub>2</sub>Aflatoxin M<sub>1</sub>Aflatoxin M<sub>2</sub>

Source: Huang *et al.* (2010)

**Fig. 2: Chemical structures of six types aflatoxins**

### **2.5.2 Aflatoxin concentrations in peanuts and peanut products**

Reports of aflatoxins levels in peanuts and peanut products in some surveys are quite alarming. Total aflatoxins concentrations in peanuts were reported to be in the range of 0.85-762.05 $\mu\text{g}/\text{kg}$  in Malaysia (Sulaiman *et al.*, 2007) and up to 2000 $\mu\text{g}/\text{kg}$  in Northern Nigeria (McDonald, 1964). Peanut cakes ('kulikuli') were also reported to have B<sub>1</sub> concentrations of 20- 455 $\mu\text{g}/\text{kg}$  in Oyo State, Nigeria (Akano and Atanda, 1990) while Adebajo and Idowu (1994) found aflatoxins levels above 30 $\mu\text{g}/\text{kg}$  in corn-groundnut snack ('donkwa'). In Ghana, Kotey (2010) reported total aflatoxins concentration of 2.59 – 26.02 $\mu\text{g}/\text{kg}$  in peanut soup from selected food joints on University of Ghana campus. The generally high concentrations of aflatoxins in peanuts have raised safety concerns and export problems with peanuts and peanut products from West African Sub-region considering the stringent regulatory limits by some countries.

The US Food and Drugs Administration has pecked aflatoxins maximum allowable limits at 20 $\mu\text{g}/\text{kg}$  for food and animal feed. Ghana Standards Authority's (GSA) limit for aflatoxins are 4 $\mu\text{g}/\text{kg}$  for peanut butter and butter crunches and 20 $\mu\text{g}/\text{kg}$  for cereals and legumes (FVO, 2007). The European Union however has enacted aflatoxins tolerance limit of 2 $\mu\text{g}/\text{kg}$  (B<sub>1</sub> only) and 4 $\mu\text{g}/\text{kg}$  (total aflatoxins) for nuts and nut products meant for direct human consumption (FVO, 2007).

### **2.5.3 Impact of aflatoxins on human health**

Aflatoxin in foods is a major worldwide public health issue. The toxic effects of aflatoxins on humans are dose and exposure dependent and could either lead to acute aflatoxicosis or chronic aflatoxicosis (Bryden, 1999). In 2004 in Kenya, the worst form of acute aflatoxicosis traced to the consumption of contaminated corn resulted in 125 deaths out of a total of 317

reported cases (CDC, 2004). According to Guo *et al.* (2009), prolonged intake of low doses of aflatoxins leads to chronic aflatoxicosis which results in cancer and suppression of immunological responses.

In Africa aflatoxin has been associated with the prevalence of hepatocellular cancer (Strosnider *et al.*, 2006). Exposure to aflatoxins facilitates the development of liver cancers in carriers of Hepatitis B and C (Williams *et al.*, 2004). Also, high exposure of children to aflatoxins may also cause stunted growth (Gong *et al.*, 2004). Studies on HIV positive people indicated an association between high levels of aflatoxins B<sub>1</sub> and vitamin A deficiency (Obuseh *et al.*, 2011).

#### **2.5.4 Aflatoxin detection and quantification**

There are different analytical techniques for the detection and quantification of aflatoxins in food commodities. Thin-layer chromatography (TLC), high-performance liquid chromatography (HPLC) and enzyme-linked immunosorbent assay (ELISA) are available analytical techniques. However these analytical methods vary in their sensitivity and accuracy (Bryden, 1999). According to Haung *et al.* (2010), TLC gives poor sensitivity, accuracy and separation of analytes while the possibility of false positive results also limits the usage of the rather rapid and more sensitive ELISA method. HPLC technique fitted with fluorescence detectors is the most commonly used method for analyzing aflatoxins. However this method is time consuming because of its larger column particle sizes (5µm) in addition to a tedious pre or post column derivatization process (Haung *et al.*, 2010). These shortfalls of HPLC method prompted a study by Huang *et al.* (2010) which established a rapid and more reliable method using ultra-high-performance-liquid chromatography tandem-mass

spectrometry (UHPLC-MS/MS). This technique is capable of simultaneous determinations of all the six types of aflatoxins in peanuts and peanut products.

### **2.5.5 Post-harvest strategies for controlling aflatoxin contaminations**

There are several post-harvest practices which can be adopted to prevent or minimize aflatoxin contamination of produce after harvest. These include adequate pod drying after harvest to moisture contents not beyond 8-9% proper storage in well ventilated dry structures, and avoiding insect and pest infestations (Price, 2005). Decontamination techniques through chemical degradation of aflatoxins using ammonia, bisulphates, ozone and hydrogen peroxide have also been investigated (Bryden, 1999). However, most of these deplete nutrient levels, produce unsafe products or are not economically viable. According to Hell and Mutige (2011), ammoniation is currently being used to decontaminate peanut meal intended for animal feed. Also, physical separation through sorting of shriveled, damaged, discoloured and immature nuts significantly reduce aflatoxin levels in peanuts (Whitaker, 1997). Some processing methods such as roasting also reduce aflatoxin contaminations in peanuts (Ogunsanwo *et al.*, 2004). However, total removal of aflatoxins through heat processing is impractical because of their high decomposition temperatures of 237°C to 306°C (Rustom, 1997). The usage of novasil® (a calcium montmorillonite clay additive) to bind aflatoxins in foods and prevent their absorption into the body has shown promise in controlling aflatoxins (Hell and Mutige, 2011; Wang *et al.*, 2008).

## **2.6 Peanut processing technologies**

After harvest, peanut pods are taken through some post-harvest processing technologies. They include screening, kernel sizing, blanching, colour sorting and roasting.

### **2.6.1 Screening and kernel sizing**

Screening describes the process of removing small, immature pods, loose shelled kernels (LSK) and other foreign material before shelling (Dorner, 2008). Screening of pods is best done using belt screens whose spacing can be adjusted to improve the efficiency of the screening process. Generally immature pods (usually small in size) and loose shelled kernels are associated with high levels of aflatoxins. A study by Dowell *et al.*, (1990) revealed a 35% reduction in aflatoxins contaminated lots of peanuts after screening.

Kernel sizing involves the passing of peanut kernels through a series of slotted screens after shelling. Kernel sizing is done to categorize and separate edible peanut kernels/seeds from smaller/immature seeds which are used for oil production (Dorner, 2008). Evaluation of the different categorizes of sized kernels revealed that as kernel sizes increase, aflatoxin levels decrease. High aflatoxin levels are usually associated with small and immature kernels (Whitaker, 1997). Cole *et al.*, (1995) observed that peanut sizing using a 16/84 by  $\frac{3}{4}$  in. slotted screen resulted in 29% aflatoxin reduction in peanut lots.

### **2.6.2 Blanching**

Blanching as used in the peanut industry is the process of removing peanut testa/skins from the shelled seeds (Whitaker, 1997). There are two types of blanching, the wet and dry blanching. Wet blanching involves the usage of warm water to brush peanut skins loosened by pushing them through sharp meshes (Naturland, 2000). During dry blanching process, peanut seeds undergo a light white roast to loosen the testa for easy deskinning. Dry blanching exposes discolourations and damages on blanched peanuts. The heat treatment darkens mould infested seeds or may cause some to retain their skins after blanching (Dickins and Whitaker, 1975). This ensures a better contrast against the white peanut background for

efficient colour sorting. It has been demonstrated that discoloured and damaged peanut seeds due to mould or insect infestation are usually associated with high levels of aflatoxins (Whitaker, 1997; Dorner, 2008).

### **2.6.3 Colour sorting**

The colour sorting process following blanching can be done using electronic sorters or manual sorters. In the United States, electronic sorters are used at large scale blanching facilities where peanuts rejected due to high aflatoxin contaminations are sent for processing to reduce aflatoxin levels (Whitaker, 1997). Batches of peanuts contaminated with aflatoxins of up to 300ppb can be reduced below 5ppb through blanching and electronic colour sorting (Ganzer, 1999). Whitaker (1997) also reported an average of 89.9% reduction in aflatoxin concentrations in a study over a five-year crop season through blanching and electronic colour sorting.

A manual sorting procedure established by Galvez *et al.* (2002) showed that aflatoxins could be reduced from 300ppb to nearly 0ppb by blanching at 140°C and handpicking discoloured and damaged peanuts. Blanching and colour sorting to eliminate aflatoxins lead to weight losses which vary depending on the level of aflatoxin contamination of the peanuts (Ganzer, 1999).

### **2.6.4 Roasting of peanuts**

Roasting is an important stage in the processing of many peanut products including peanut butter and peanut paste. Adequate roasting essentially extends the shelf life of peanuts and peanut products, facilitates subsequent processing and impacts desirable product

characteristics such as colour, flavour and aroma (Mendes *et al.*, 2001; Youn and Chung, 2012).

The positive effect of peanut roasting on shelf stability, flavour and colour has been attributed to the formation of Maillard Reaction Products (MRP) during roasting (Manzocco *et al.*, 2001). The melanoidins formed during Maillard reaction produce the desirable golden to dark brown colour of roasted peanuts which increase with increasing roast time and/or temperature. The increase in melanoidins formation during roasting has been associated with increased antioxidant capacity (oxidative stability) which prolongs the shelf-life of roasted peanut products (Cammerer and Kroh, 2009; Manzocco *et al.*, 2001). Also MRP such as alkylpyrazines which are formed during roasting are responsible for the desirable roasted peanutty flavour (Gross *et al.*, 2008).

These quality improvements attributed to roasting depend largely on the roast time and temperature variables (Mendes *et al.*, 2001). Therefore defining the roasting conditions is important to achieving optimum product characteristics. The selection of roast time and temperature variables depends on the degree of roast required (light, medium or dark), type of roaster, variety, maturity and moisture content of the material used (Mendes *et al.*, 2001).

Ayoola and Adeyeye (2000) observed that roasting decreases the proximate composition of peanuts. Reports have also suggested that roasting plays a role in enhancing the allergenicity of peanuts (Maleke *et al.*, 2000). Roasting of peanuts has been demonstrated to reduce aflatoxin concentrations in peanuts by 40 - 80% (Arzandeh and Jinap, 2011).

## 2.7 Thermal processing of canned foods

Thermal processing has been widely employed as a food preservation technique for extension of shelf-life and to ensure the safety of processed foods. The process of preserving foods in hermetically sealed containers through the application of an adequate thermal process to inactivate enzymes and destroy spoilage and disease causing microorganisms is known as canning (Ponce-Alquicira, 2004). The thermal destruction of microorganisms during canning operations depends on the number, the type and thermal resistance of microorganisms present, the physical characteristics of the product (i.e. water activity, viscosity of food, can shape and dimensions), and the chemical composition (i.e. preservatives, pH) (Ponce-Alquicira, 2004). The thermal process of low acid foods ( $\text{pH} \geq 4.6$ ) is usually designed to ensure the destruction of *Clostridium botulinum*: a highly heat resistant, rod-shaped, spore-former which survives under anaerobic conditions to produce the botulinum toxin (Awuah *et al.*, 2007). Although commercial sterility is achieved with the inactivation of the *C. botulinum* spores, other heat resistant spores such as *Clostridium thermosaccolyticum*, *Bacillus stearothermophilus*, and *Bacillus thermoacidurans* may still persist and cause spoilage under ‘abused’ temperature storage conditions (temperature  $> 30^{\circ}\text{C}$ ) (Awuah *et al.*, 2007).

### 2.7.1 Kinetics of thermal inactivation of microorganisms

Thermal processing of foods results in the inactivation of the microbial population. The kinetics of thermal inactivation of microorganisms have been established to follow a first order reaction which is represented at a given temperature as:

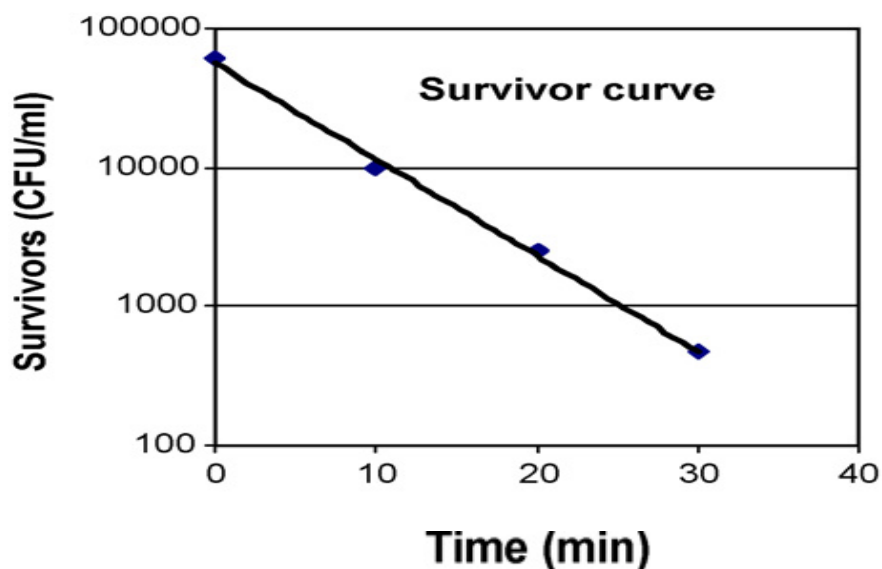
$$\frac{dN}{dt} = -kN \dots \dots \dots 2.1$$

where  $N$  is the microbial population,  $t$  is the time of heating at the specified temperature and  $k$  is the reaction constant (Maroulis and Saravacos, 2003).

The thermal destruction (inactivation) of microorganisms is represented by the  $D$  – value (decimal reduction time) at a particular temperature. The  $D$ -value is determined from the survivorship curve (Fig. 3) which is a plot of the number of microorganisms surviving a given heat treatment at a given temperature versus the heating time (Awuah *et al.*, 2007). The  $D$ -value refers to the time it takes to achieve 90% (one log cycle) reduction in target microorganisms at a constant temperature (Berry and Pflug, 2003). The  $D$ -value is mathematically represented as:

$$D = \frac{t_2 - t_1}{\log(A) - \log(B)} \dots \dots \dots 2.2$$

where  $A$  and  $B$  represent the survivor counts following heating for times  $t_1$  and  $t_2$  minutes (Awuah *et al.*, 2007).



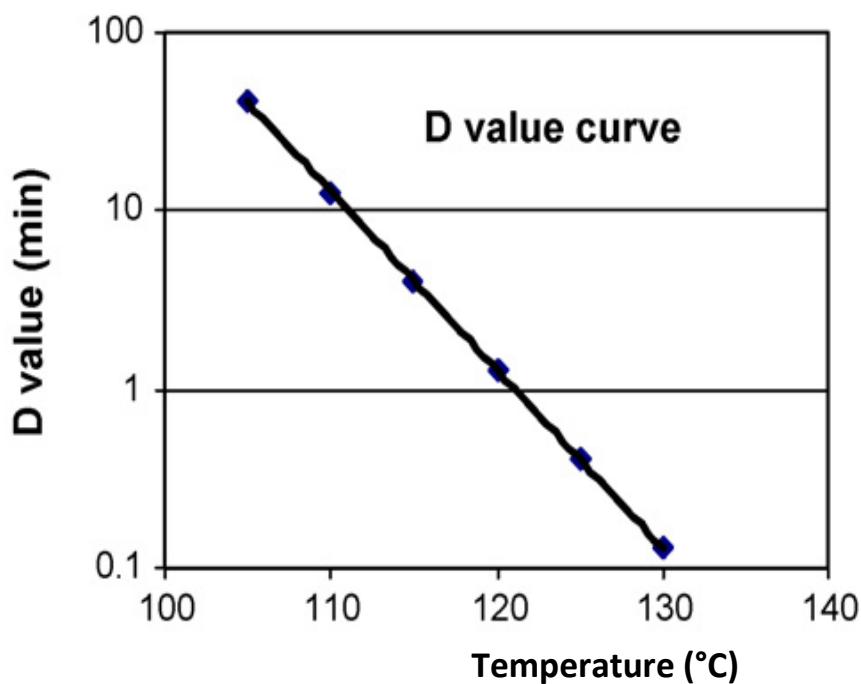
**Fig. 3: Typical survivor curve for determination of  $D$ -value**

Source: Awuah *et al.*, (2007)

Another important thermal inactivation parameter is the  $z$ -value. The  $z$ -value (temperature sensitivity) indicates the number of degrees rise in temperature to effect a 10-fold change in the  $D$ -value (Awuah *et al.*, 2007). The  $z$ -value which is obtained from the  $D$ -value curve (Fig. 4) has been defined mathematically as follows:

$$z = \frac{T_2 - T_1}{\log(D_1) - \log(D_2)} \dots\dots\dots 2.3$$

where  $D_1$  and  $D_2$  are  $D$ -values at temperatures  $T_1$  and  $T_2$ , respectively (Awuah *et al.*, 2007).



**Fig. 4: Typical D-value curve for the determination of  $z$ -value**

Source: Awuah *et al.* (2007)

### 2.7.2 Establishing a thermal process

The establishment of a thermal process is based on the lethality of the target organism at the cold point (slowest heating point) of the packaged product (Maroulis and Saravacos, 2003).

The lethality can be estimated by determining the process lethality/sterilizing ( $F_o$ ) value which is defined as the time required to cause log10 reductions in bacterial numbers at a given reference temperature. (Aquilar *et al.*, 2012). The  $D$  and  $z$  – values are important for the determination of the thermal process lethality. According to Holdsworth (1985), the sterilizing value ( $F_o$ ) can be determined using the general method or calculated using the formula method (mathematical method). The basic mathematical model (also known as the  $D$ - $z$  model) used to estimate the  $F_o$  of any thermal process with known time-temperature profile is based on the original works of Ball and Oslon, (1957) given by:

$$F_o = \int_0^t 10^{(T-T_{ref})/z} dt \dots \dots \dots 2.4$$

where  $F_o$  = the integrated lethality (at the slowest heating point) (mins),  $t$  = time of processing (mins),  $T$  = temperature ( $^{\circ}\text{C}$ ) at time  $t$ ,  $T_{ref}$  = reference temperature, usually  $121.1^{\circ}\text{C}$  (equivalent to  $250^{\circ}\text{F}$ ),  $z$  = slope of the logarithm of the decimal reduction time  $D$ , versus temperature for the specific organism (for *Clostridium botulinurn*  $z = 10^{\circ}\text{C}$ ).

An alternative to the  $D$ - $z$  model is the  $k$ - $E_a$  kinetic model (Arrhenius equation) which has also been established to represent the  $F_o$  value in terms of reaction kinetic constant (Aiba and Toda, 1967) i.e:

$$F_o = D_{121.1} \ln\left(\frac{N_0}{N}\right) = A \int_0^t e^{-E_a/RT} . dt \dots \dots \dots 2.5$$

where  $N_o$  is the initial number of organisms,  $N$  the number at time  $t$  and temperature  $T$ ,  $A$  is the Arrhenius constant;  $E_a$  is the energy of activation, and  $R$  is the gas constant.

Holdsworth (1985) observed that at a  $z$ -value of  $10^{\circ}\text{C}$  and a  $D$  value of 0.3mins a lot of low acid foods of commercial operations can be thermally processed at  $110\text{-}130^{\circ}\text{C}$  to produce a safe canned product. Generally, thermal treatment of low acid ( $\text{pH} > 4.6$ ) canned foods to achieve an  $F_o$  of 3 mins ( $z = 10^{\circ}\text{C}$ ) is considered the minimum thermal process ( $12D$  process) for a safe canned product based on the total destruction of *Clostridium botulinum* (Awuah *et al.* 2007; Berry and Pflug, 2003).

### 2.7.3 Effect of thermal processing on food quality attributes

Although thermal process time-temperature schedules are designed to achieve commercial sterility, the heat treatment can promote reactions that affect the quality of the food. Quality degradation involves subjective factors (e.g sensory attributes such as taste) and quantifiable factors such as nutrient degradation (Awuah, *et al.*, 2007). The changes in sensory and nutritional qualities of foods pose a challenge during the design of a thermal process since the target lethality must be achieved to ensure safety.

According to Holdsworth (1985), the need to optimize processing conditions arises because the kinetics of thermal degradation doubles for a  $10^{\circ}\text{C}$  rise in temperature while the rate of microbial inactivation increases 10-fold under similar conditions. The kinetics of nutrient and microbial degradations suggest that their  $z$  and  $D$  values differ (Awuah *et al.*, 2007). The  $z$  values for cooking and nutrients degradation ( $25 - 45^{\circ}\text{C}$ ) are generally higher than those for microbial destruction ( $z = 7 - 12^{\circ}\text{C}$ ) (Holdsworth, 1985). These differences in kinetics between the nutritional and microbial degradation are exploited to optimize thermal processes. Mansfield (1962) proposed a model for estimating the quality losses as *cook* or  $C$  values for low acid foods which is given by :

$$C = \int_0^t 10^{(T-T_{refq})/z_q} dt \dots \dots \dots 2.6$$

where  $z_q$  and  $T_{refq}$  represent the  $z$ - value and reference temperature for the most heat labile component component. A  $z_q = 33.1^\circ\text{C}$  and  $T_{refq} = 100^\circ\text{C}$  are used to determine the *cook/C* value designated as  $C_o$  (Awuah *et al.*, 2007).

The retention of optimum product quality during heat processing is best achieved using high temperature short time processes. However, Holdsworth (1985) indicated that HTST processes are only successful for thin films of liquid products. The more viscous or particulate foods present a barrier to the application of this principle to limit the destructive effects of prolonged heating on product quality. According to Awuah *et al.* (2007), vitamins are the most sensitive food components degraded by heat sterilization processes. However of the heat sensitive vitamins, thiamine has the most stable heat denaturation kinetics (Ryley and Kajda, 1994). Heat treatments of foods could also lead to browning (Maillard) reactions, modification of protein structures and the degradation of naturally occurring pigments such as chlorophylls , anthocyanins, carotenoids and betanins (Awuah *et al.*, 2007).

In conclusion, peanuts are an important crop widely distributed in the world. In Ghana, the varieties of peanuts cultivated are many. Although aflatoxin contamination of peanuts remains a big problem in peanut utilization, several technologies are available which can be adopted to reduce aflatoxins in peanuts for food applications. The manual colour sorting method developed by Galvez *et al.* (2002) offers a simple way to minimize aflatoxin levels peanuts. The principles of canning and thermal processing would be useful in the development of a canned peanut soup base to ensure microbial safety and shelf stability.

## CHAPTER THREE

### 3.0 MATERIALS AND METHODS

#### 3.1 Physico-chemical characterization of peanut varieties and selection of a suitable peanut variety for making canned peanut soup base.

Three peanut varieties (Manipintar, Chinese and Sinkarzie) were purchased from Nima market in Accra while the fourth variety (Bugla local) was obtained from Tamale central market in the Northern region. These four varieties were selected because they are the most commonly occurring and easier to obtain from the local markets.

##### 3.1.1 Determination of the physical characteristics of peanuts

###### 3.1.1.1 Seed dimensions

The seed dimensions measured were the seed length and seed width. The lengths and widths of five randomly selected peanut seeds were measured using vernier calipers (Nyalemegbe, 2007). The averages of the five measurements for each peanuts variety were calculated as the seed dimensions.

###### 3.1.1.2 Bulk density

The bulk density was determined as 100 peanut seeds weight. Peanut kernels/seeds were presorted to remove shriveled and damaged peanuts. The bulk density for each peanuts variety was determined as the weight of randomly selected 100 peanut seeds (Frimpong *et al.*, 2007). Determinations were done in quadruples for each variety of peanuts.

### **3.1.2 Determination of the chemical (proximate) composition**

Peanut samples were prepared according to AOAC method 935.52 (AOAC, 2005). The four peanut samples were coarse milled using a Straub plate grinding mill (model 4E, Straub cooperation, Philadelphia, USA) and stored in airtight glass container below 10°C for the chemical analyses. The analyses conducted included moisture content, crude protein, crude fat, crude ash and crude carbohydrates. All analyses were done in triplicates.

#### **3.1.2.1 Moisture content**

The moisture content was determined using the air oven method at 105°C for six (6) hours according to AOAC method 925.40 (AOAC, 2005).

#### **3.1.2.2 Crude fat**

The crude fat content was determined using the Soxhlet method according to AOAC (2005) method 948.22.

#### **3.1.2.3 Protein content**

The Kjeldahl method was used to determine the nitrogen content of the peanut samples as outlined in AOAC (2005) method 955.04C. The protein content was then calculated as the product of nitrogen content and a factor of 5.46 for peanuts.

#### **3.1.2.4 Crude ash**

The ash content was obtained as the weight of the inorganic residue after the carbonization (at 600°C) of the test portion of peanut flour in a muffle furnace (SF3, Stuart Sf, UK) to a constant weight of white ash (AOAC, 2005).

### **3.1.2.5 Crude Carbohydrates**

The crude carbohydrate content was calculated as the difference between the sum of the other components and 100% i.e.

$$\text{Crude Carbohydrates} = 100\% - (\% \text{protein} + \% \text{moisture} + \% \text{crude fat} + \% \text{crude ash})$$

### **3.1.3 Selection of a suitable peanut variety for making canned peanut soup base**

The most suitable variety of peanuts for making peanut soup base was selected based on the proximate composition, physical characteristics (seed dimensions) and a prior survey of three markets in Accra (Madina, Nima and Timber markets) to identify commonly sold peanuts. Literature on the availability and cultivation of the four varieties of peanuts was also considered in the selection of a suitable peanut variety for making canned peanut soup base.

## **3.2 Evaluation of the efficacy of a manual sorting method to minimize aflatoxins in peanuts for canned peanut soup**

The manual colour sorting method (Galvez *et al.*, 2002) was evaluated to assess the proportions (%) and the aflatoxin levels of the various peanut components that were sorted out. The sorting method was evaluated using the selected variety of peanuts (Chinese) with the overall aim of sorting to reduce aflatoxins in peanuts used for making peanut soup base.

### **3.2.1 Manual colour sorting to reduce aflatoxins in peanuts**

Peanuts (Chinese variety) were weighed (5kg) and manually sorted and the weights of the sorted out components (shriveled and damaged peanuts, and presorted peanuts) recorded. The presorted peanuts were put in perforated aluminum trays and blanched at 140°C for 25mins in batches of 1kg in an air oven with stirring every 10mins. The blanched peanuts were

colour sorted by removing discoloured seeds from the lots. The components sorted out after blanching were categorized as: hulls/skins, blanched peanuts, less than 50% discoloured peanuts (peanuts with seed length less than 50% discoloured), 50% or more discoloured peanuts (peanuts with seed length 50% or more discoloured), clean peanuts (peanuts with no discolouration). The sorted components were weighed and the proportions (%) of each component calculated. The sorted components were bagged and refrigerated at 5°C until used for aflatoxins analyses.

### **3.2.2 Aflatoxin analyses**

Aflatoxins analyses were carried out on six (6) of the sorted out components (section 3.2.1) of the Chinese variety using high performance liquid chromatography (HPLC) method. The components were: shriveled and damaged peanuts, hulls/skins, blanched peanuts, sorted out less than 50% discoloured peanuts, sorted out 50% or more discoloured peanuts, clean peanuts.

#### **3.2.2.1 Sample preparation**

The cone and quartering technique (HGCA, 2009) was used to sample test portion of each peanut component for aflatoxin analysis. 50g grams of sample was blended with 5g NaCl, and 200ml methanol:water (8:2) at low speed for 2mins and high speed for 1min in a Warring blender (Warring products division, Torrington USA). The slurry formed was filtered using whatman filter paper number 4. 10ml of filtrate was mixed with 60ml phosphate buffer saline (PBS) solution and the mixture eluted in an Easi-Extract Afla column packed with monoclonal antibodies specific to G<sub>1</sub>, B<sub>1</sub>, G<sub>2</sub> and B<sub>2</sub>. Column was rinsed with 10ml distilled water. Aflatoxins were then eluted from the column with 0.5ml methanol followed by 0.7ml methanol. The eluate was collected into 5ml volumetric flask and diluted with distilled water

to the mark. 2ml of the extract was filled into vial bottles and loaded onto the HPLC autosampler chamber for separation, detection and quantification. All reagents used were HPLC grade.

### **3.2.2.2 Detection and quantification of aflatoxin levels**

Waters HPLC system consisting of Waters 1525 Binary HPLC pump, Waters 2707 Autosampler, and Waters 2475 Multi  $\lambda$  Fluorescence Detector. Data acquisition and analyses was done using Breeze 2 software (manufacturers??) The waters HPLC was equipped with a reverse phase symmetry ( $C_{18}$ ) column (sperisorb55 ODS-1) of dimensions 25cm x 4.6mm x 5 $\mu$ m. The fluorescence detector operated at an excitation wavelength of 360nm and an emission wavelength of 440nm at temperature of 35°C. The mobile phase was a mixture of water:methanol:acetonitrile (60:30:20) at a flow rate of 1mLmin<sup>-1</sup> in an isocratic separation method. Linearity was estimated by injecting four Triology analytical laboratory aflatoxin standards: aflatoxin B<sub>1</sub> and aflatoxin G<sub>1</sub> at 2.0 $\mu$ g/ml and aflatoxin B<sub>2</sub> (B2) and aflatoxin G<sub>2</sub> (G2) at 0.5 $\mu$ g/ml. The limits of detection were determined to be: G1 and G2 = 0.13 $\mu$ g/kg; B1 and B2 = 0.15 $\mu$ g/kg. The injection volume used was 10 $\mu$ l.

## **3.3 Determination of the optimum ingredients formulation for peanut soup base**

### **3.3.1 Peanut soup ingredients preparation**

Based on preliminary interviews with commercial peanut soup makers, the soup ingredients used were tomato paste (Salsa), pepper (kpakpo shito), onions, peanut pastes, ginger, iodated salt (Anapuna), and water. The ingredients were purchased from Madina market.

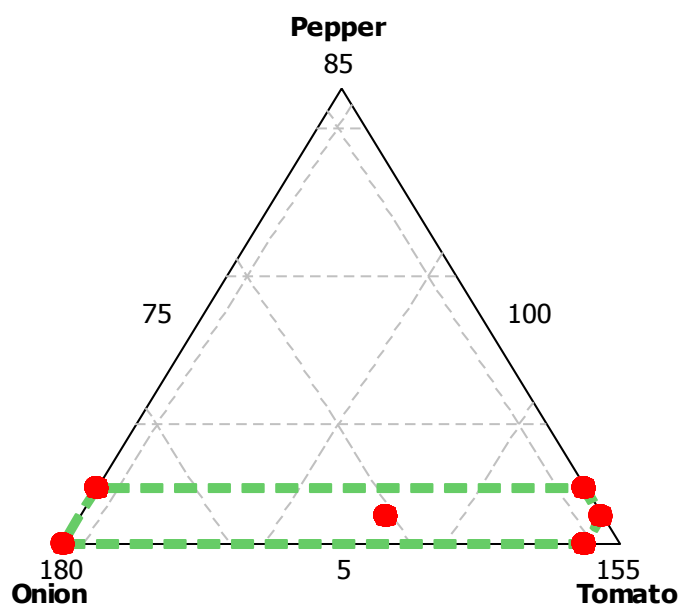
### 3.3.2 Experimental design for the optimization of peanut soup ingredients

A 3 component mixture design (extreme vertices design Fig. 5) was employed (Montgomery, 2001) and the experimental matrix and region generated using Minitab (version 14). The components were pepper, onion and tomato paste (Table 2) and different amounts of the ingredient combinations were generated (Table 3). The response variables that were determined on the groundnut soups included colour, spiciness, taste, consistency (thickness), smoothness, hotness, overall acceptability.

**Table 2: Mixture design components for peanut soup ingredients**

Ingredients	Low (g)	High (g)
Pepper	5.0	15.0
Onion	100.0	180.0
Tomato	75.0	150.0

Mixture total = 260.0



**Fig. 5: Experimental region of peanut soup ingredients' mixture design plot**

**Table 3: Amounts of ingredient combinations for peanut soup formulation**

Sample	Ingredients (g)		
	Pepper	Onion	Tomato paste
1	5	180	75
2	15	170	75
3	10	131	119
4	5	105	150
5	15	100	145
6	10	100	150
7	15	100	145
8	15	170	75

**Total ingredients mixture for each sample = 260.0g**

### 3.3.2.1 Preparation of peanut soups

Based on the extreme vertices design for 3 components, a total of eight formulations were generated using the Minitab software. Each of the eight (8) combinations of ingredients (Table 3) was used in addition to 300g peanut pastes, 15g iodated salt, 10g ginger and 2600ml of water. 300g of peanut paste was weighed into a cooking sauce pan. 200ml of water was added to the weighed paste and stewed for 5 minutes (till oil appeared on the surface of the paste) on the hot plate. 1800ml of water was added and stirred till the paste was totally mixed with the water. The weighed tomato paste (varied based on mixture combinations developed) was also added and mixed thoroughly. The pepper, onion (varied based on mixture combinations developed) and 10g of ginger were added and the mixture was allowed to stay on fire for fifteen (15) minutes at a temperature of about 70°C to 75°C. The onion, ginger and pepper were taken out and blended after the fifteen minutes with 200ml of water; sieved and re-blended with 200ml water; sieved again. Finally 200ml water was used to rinse the blender, poured into the sieve (with the blended content still in) and finally sieved into the

mixture. 15g salt was added and mixture allowed to boil for 25minutes at a temperature of 97°C to 98°C. The soup produced was allowed to simmer for another 25-30 minutes at a temperature of 80°C to 85°C.

### 3.3.3 Sensory evaluation of peanut soups prepared from mixture combinations

The 8 soups prepared from the mixture combinations were analysed for their attributes' acceptability (liking test) and overall acceptability of samples by a panel of 48 consumers using a 9-point hedonic scale (Table 4). The criteria for selection of panelists were that they must be consumers of peanut soup and above 18years. Peanut soups were warmed slightly served at a temperature of 35-40°C. Samples were coded using three digit numbers and the order of presentation to panelists randomized. The sensory evaluation ballot used is shown in appendix 7.1.

**Table 3: Hedonic (9-point) scale used for scoring product acceptability.**

Score	Interpretation
1	Dislike extremely
2	Dislike very much
3	Dislike moderately
4	Dislike slightly
5	Neither like nor dislike
6	Like slightly
7	Like moderately
8	Like very much
9	Like extremely

### **3.3.4 Determination of the optimum peanut soup ingredients**

Regression models of the sensory attributes of the peanut soup formulations were used to generate contour plots. The contour plots for each sensory attribute were overlaid based on the ranges of consumer acceptability for each attribute. The region in which all contours satisfied the criteria of acceptability was considered as the optimum region for the ingredient formulation. The optimized levels of ingredients were to be used for the formulation of the canned peanut soup base which is a concentrated form of the peanut soup.

### **3.3.5 Validation of ingredient formulation in the optimum region**

Four peanut soups made using two (2) ingredient formulations within the optimum region and two from outside of the optimum region were served to 50 untrained panelists for sensory evaluation. Sensory attributes of the peanut soups that were made using formulations from the optimum region and from outside the region were compared with predicted attributes using the regression models.

## **3.4 Establishing the thermal process variables (temperature and time) and evaluation of the quality of the canned peanut soup base.**

### **3.4.1 Experimental design for the thermal process**

A 3 x 3 full factorial design was used to determine the limits of the time and temperature schedules for the thermal process. The design variables were, time (40, 50 and 60 minutes) and temperature (115, 120 and 125°C). The response variables calculated or monitored included sterilizing values ( $F_o$ ), colour ( $L^*a^*b^*$ ) and total colour difference ( $\Delta E^*$ ). Table 5 indicates the thermal process combinations. The order of experimental runs was randomized.

**Table 5: Matrix of experimental run order and the thermal process temperature-time combinations**

Run order	Temperature (°C)	Time (mins.)
1	120	50
2	125	50
3	120	60
4	115	50
5	120	40
6	125	40
7	115	60
8	125	60
9	115	40

### 3.4.2 Preparation of canned peanut soup base

#### 3.4.2.1 Ingredients preparation for making peanut soup base

Ingredients combination used to prepare canned peanut soup base were the optimum ingredients combination which was established in previous experiment (3.3.4). Peanut paste was made from the selected peanut variety (i. e Chinese variety from section 3.1.2). The Chinese peanuts were presorted, blanched at 140°C for 25minutes and the discoloured peanuts manually removed to reduce aflatoxin levels. Roasting was done at 140°C for 60mins to obtain a medium roast colour ( $L^* 58 \pm 1$ ) using a oven (0825-178, Chalice Oven, Uk). The roasted peanuts were cooled under a fan and milled to paste using a disc attrition mill.

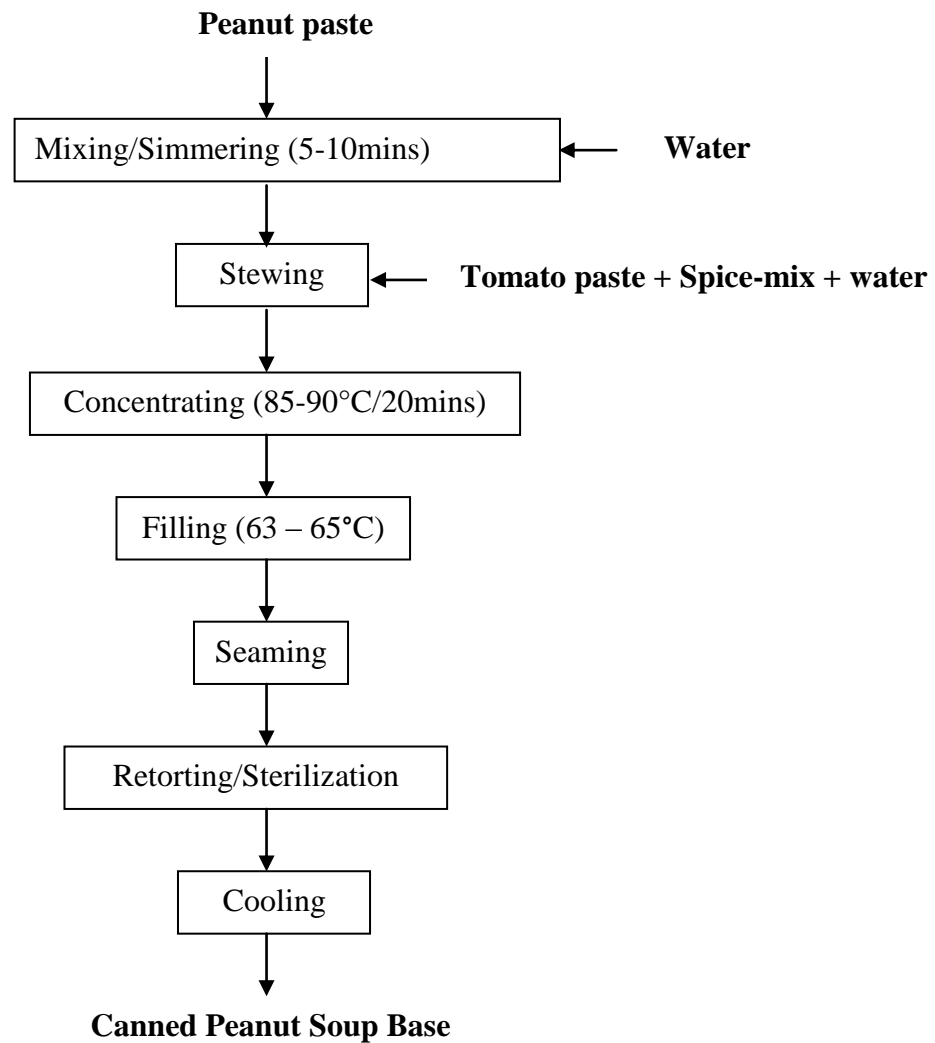
The onion, pepper and ginger were clean and used to prepare a spice-mix extract. The spice mix extract was prepared by boiling weighed amounts of ginger, pepper and onions (10g, 12.3g and 100.8g respectively per 300g of peanut pastes.) in 200ml water for 15mins. The boiled spices were ground in a binatone blender at low speed for 1min. and maximum speed for 2mins. and sieved. The residue was ground again in 100ml measured amount of water and sieved. The blender was rinsed with 100ml water and the content sieved. The filtrate obtained was used for the preparation of peanut soup base.

#### **3.4.2.2 Processing of canned peanut soup base**

Canned peanut soup base was prepared as shown in the process flow diagram (Fig. 6). Based on preliminary works the total volume of water for making peanut soup (2600ml per 300g peanut paste) was reduced to 780ml per 300g peanut pastes to make acceptable canned peanut soup base.

Peanut paste was weighed into a saucepan, mixed with water (300g paste: 200ml water) and simmered (for 10 minutes) till oil appeared on surface on a gas cooker. Tomato paste (Salsa tomato paste) was weighed (146.9g tomato pastes per 300g peanut pastes), added to the mixture in the saucepan and stirred to mix uniformly. The liquid spice-mix extract was measured in a measuring cylinder and added to the content of the saucepan. Finally the remainder of the amount of water needed to make up the total volume of water required (considering the volume of water added to the peanut pastes and the volume of the spice-mix extract) was added to the saucepan. The soup mixture was concentrated at 85-90°C for 20mins with continuous stirring to produce the peanut soup base. The formulated soup base was immediately filled at 70°C into lacquered tins of dimensions 99 x 118mm with

3.78% (30g) of added peanut oil to a net weight of 800g and seamed. The canned products were then retorted at different time-temperature combinations as shown in Table 5



**Fig. 6: Process flow for canned peanut soup base**

### 3.4.3 Heat penetration test of canned peanut soup base

For each thermal process schedule, two seamed cans were each perforated at one end to enable the collection of heat penetration data. A k-type heat insulated flexible thermocouple wire was fixed through the hole made in the perforated product such that it was positioned at the geometric centre of the can (Slowest Heating Point - SHP). The thermocouple wires were

connected to a precalibrated four channel data logging thermometer (General Tools and Instruments, NY 10013, New York). The initial product temperatures prior to retorting were kept within 63-65°C after seaming using a constant temperature water bath. The products were then retorted in a still vertical retort. The heat penetration data (product temperature) at the SHP from the time the steam was put on through to the end of air cooling of products were collected at a standard time interval of five (5) minutes. The processing temperature and time for the heating and cooling processes were also recorded every five (5) minutes. The maximum product temperature reached during thermal processing for each time-temperature combinations were also recorded.

#### 3.4.4 Determination of $F_o$ values

Estimates of  $F_o$  values for each time and temperature combination (Table 5) were determined using the trapezoidal integration method (Warne, 1988). The heat penetration data collected at standard time interval of five (5) minutes (section 3.4.2) were used. The process lethal rates ( $L$ ) at each product temperature ( $T$ ) were calculated using a reference temperature of 121.1°C and a  $z$  value of 10°C given by equation (1). The  $F_o$  values were calculated as the product of the standard time interval (5mins) and the sum of the lethal rates ( $L$ ) during the heating and cooling phases (equation 2).

$$L = \text{Log}^{-1} \frac{T-121.1}{10} \dots\dots\dots 1$$

$$F_o = 5 \sum L \dots\dots\dots 2$$

Where  $L$  - lethal rate,  $T$  – product temperature at SHP and  $F_o$  – sterilizing value

Equations 1 and 2 adapted from Warne (1988).

### 3.4.5 Determination of colour and total colour difference of peanut soup base

The colour ( $L^*a^*b^*$ ) of the peanut soup base was determined before and after retorting for each thermal process schedule as described in section 3.4.2. The total colour difference ( $\Delta E^*$ ) was calculated (using equation 3) considering the colour ( $L^{*t} a^{*t} b^{*t}$ ) of the unretorted peanut soup base as the target colour i.e:

$$\Delta E^* = \sqrt{((L^* - L^{*t})^2 + (a^* - a^{*t})^2 + (b^* - b^{*t})^2)} \dots \dots \dots 3$$

Where  $\Delta E^*$ - total color difference,  $L^{*t} a^{*t} b^{*t}$  – target colour of unretorted peanut soup base and  $L^* a^* b^*$ - colour of retorted canned peanut soup base.

Equation 3 source: Minolta (1991).

### 3.4.6 Determination of the limits of the thermal process for canned peanut soup base

Response surface contour plots of  $L^*$  values,  $F_o$  and  $\Delta E^*$  were overlaid to obtain the optimum region which defined the time and temperature limits for a shelf stable canned peanut soup base product.

### 3.4.7 Visual inspection of canned peanut soup base

The canned soup base products were inspected daily for any signs of bloating over the 7-day incubation period. Products were opened after seven days and the can lids inspected for any signs of darkening.

### 3.4.8 Microbial Analysis of thermally processed peanut soup base

Aerobic Plate Count (APC) and Yeasts and Moulds Count using the pour plate technique were done as described by Gilliland *et al.*, (1976). Plate Count Agar (PCA) was used for the

determination of APC while Potato Dextrose Agar (PDA) was used to enumerate yeasts and moulds.

### **3.5 Data analyses**

The means and standard deviations of measured variables were calculated using Microsoft Office Excel 2007. Analysis of variance (ANOVA) to compare treatment means (at  $p < 0.05$ ) and Multiple Range test of treatments found to be significantly different were done using Minitab (version 14). Mixture Regression models were developed to relate the effect of the mixture components on the response variables (sensory attributes) of peanut soup formulations using Statgraphics Centurion (version XV). The  $R^2$  were calculated to determine the adequacy of the models. Response surface plots and regression models were also developed and the R-squares and R(adj.)squares calculate to describe the effect of the thermal schedules on product attributes of the canned peanut soup base using Minitab (version 14). Paired t-test was also conducted to assess the effect of the thermal process schedules on the  $L^*$  (lightness) values of the peanut soup base before and after canning using Minitab (version 14).

## CHAPTER FOUR

### 4.0 RESULTS AND DISCUSSION

#### 4.1 Physico-chemical characterization of four selected peanut varieties

The physical and chemical properties of four Ghanaian peanut varieties namely, Manipintar, Sinkarzie, Chinese and Bugla local (Plate 1) were determined. These varieties were available in the major markets of Accra and could easily be obtained.

##### 4.1.1 Physical Characteristics

Physical characteristics determined were seed dimensions (seed length and seed width) and the bulk density (Table 6).

**Table 6: Physical characteristics of four peanut varieties**

Peanut Variety	Seed Width (mm)	Seed Length (mm)	Bulk Density (g/100seeds)
Manipintar	9.50 ± 1.07 <sup>a</sup>	14.88 ± 0.61 <sup>a</sup>	30.30 ± 0.75 <sup>a</sup>
Chinese	7.88 ± 0.92 <sup>ab</sup>	10.28 ± 2.27 <sup>b</sup>	20.22 ± 1.01 <sup>c</sup>
Sinkarzie (Red type)	8.32 ± 0.48 <sup>ab</sup>	14.52 ± 1.98 <sup>a</sup>	25.34 ± 0.09 <sup>b</sup>
Bugla local	7.38 ± 1.21 <sup>b</sup>	13.64 ± 0.73 <sup>a</sup>	23.79 ± 1.75 <sup>c</sup>

Values in columns with different superscripts are significantly different ( $p < 0.05$ )

Values are means of four replicates ± standard deviation



**Plate 1: Four locally cultivated Ghanaian peanut varieties**

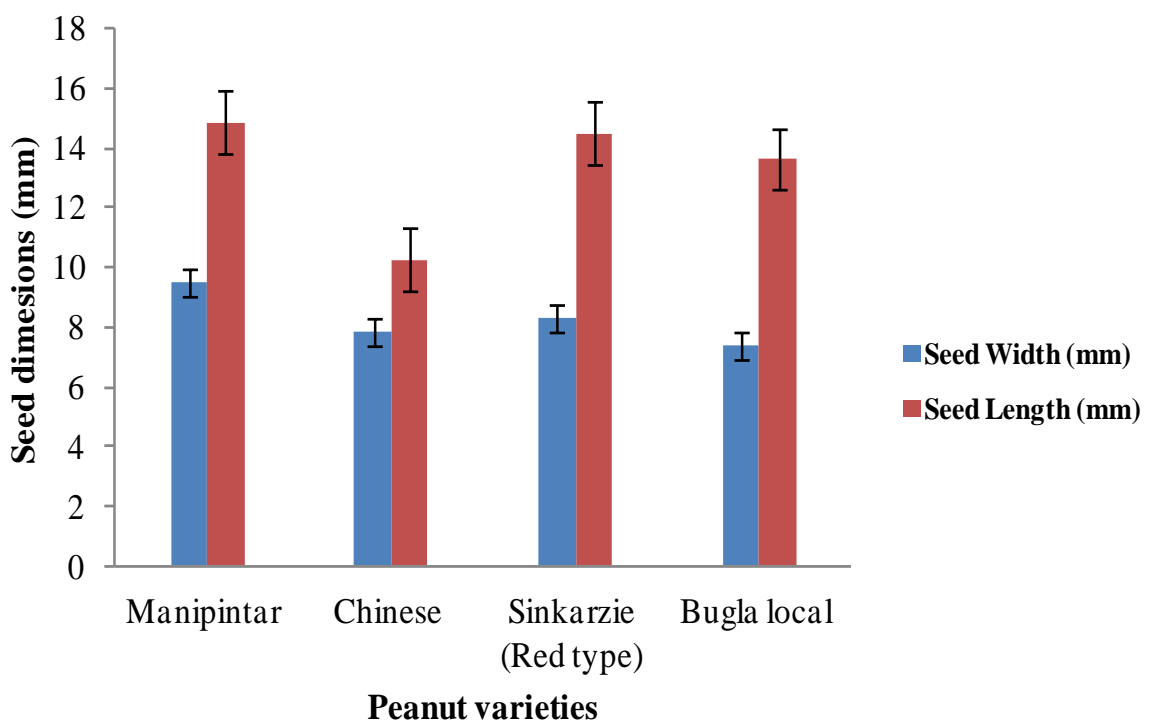
#### **4.1.1.1 Seed Dimensions**

The seed widths of the different varieties of peanuts ranged from 7.38 to 9.50mm (Table 6). Significant differences ( $p < 0.05$ ) in seed width were found among the varieties (Table 6). The Bugla local variety had the narrowest mean seed width while Manipintar had the widest (Fig. 7). Bugla local was also the most variable in terms of seed width (standard deviation of

1.21) while Sinkarzie was the least variable (standard deviation 0.48). The seed lengths of peanuts also differed significantly ( $p < 0.05$ ) among the varieties (Table 6). The Manipintar variety was the longest seeds (14.88mm), while Chinese were the shortest (10.28mm) (Fig. 7). The Chinese peanuts were the most variable (standard deviation 2.27) while Sinkarzie again was the least variable (standard deviation of 0.61) in terms of seed lengths. These dimensions compare very well with similar measurements that were conducted by Nyalemegbe (2007) for Manipintar and Chinese who reported seed lengths of 15.68mm and 10.60mm and seed widths of 9.65mm and 7.55 mm respectively. Nyalemegbe (2007) also reported significant variations in seed dimensions ( $p < 0.05$ ) between different peanut varieties.

The variations in seed dimensions indicate that different varieties of peanuts may have different shapes and sizes. Some peanuts may be more rounded (e.g. Chinese which shows least difference between seed width and length) than others. Variability (standard deviations) in seed dimensions for a particular variety could be indicative of the degree of uniformity in seed sizes. The varietal differences in seed dimensions could be attributed to genetic predispositions. However, environmental and climatic factors also affect peanut seed development (Ahmed and Mohammed, 1997). The measurement of seed dimensions can be useful in the classification of peanut seeds. Singh and Oswalt (1995) described the Virginia type peanuts as the largest pods with elongated seeds, the Runner-type peanuts as the medium size seeds, the Spanish-type peanuts as the smaller round seeds, and the Valencia-type peanuts as seeds with intermediate sizes and shapes. Hence the Chinese variety could be classified as Spanish-type while the Manipintar and Sinkarzie varieties could be classified as Virginia-type as suggested by Asibuo *et al.* (2008). The morphology of Bugla local peanut plant might be useful in determining its category although the seed dimensions suggest

intermediate sizes and shapes (Valencia-type). The measurement of seed dimensions is also important for the selection of screens during sizing or screening of peanuts. The dimensions are also useful in deciding the food applications such as in confectionery, or for use in salted peanuts or mixed nut products. In the case in which the Chinese variety is more variable in dimensions, it could best be used for products in which the seed dimension will not be of importance such as in groundnut paste/ soup manufacture.

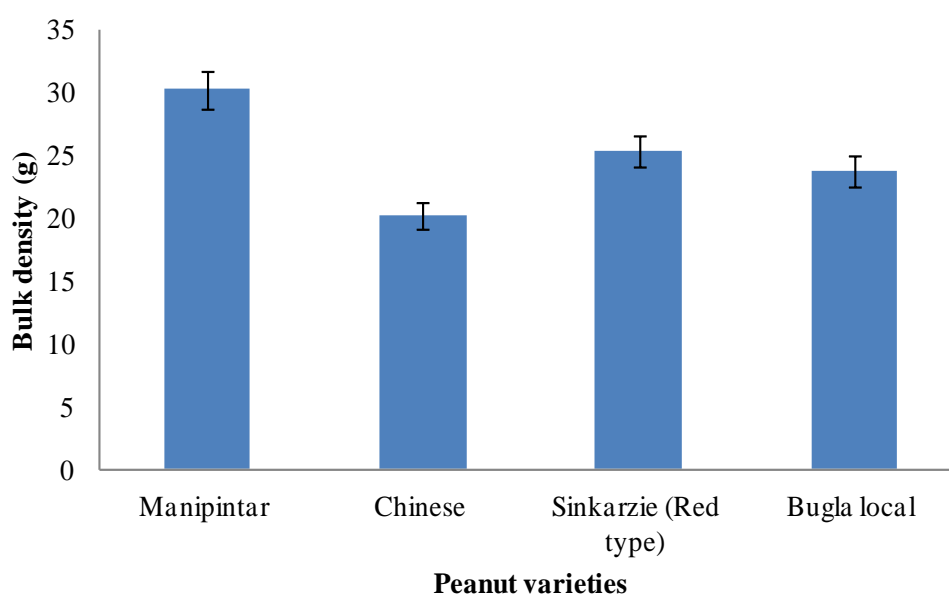


**Fig.7: Seed dimensions of four peanut varieties**

#### **4.1.1.2 Bulk density**

The bulk densities of the four varieties of peanuts were determined as the total mass of 100 peanut seeds. Significant differences ( $p < 0.05$ ) were observed among the four peanut varieties except between Sinkarzie and Bugla local which didn't show significant differences (Table 6). Fig. 8 indicates that the bulk density of peanuts varied from 20.22mm to 30.30mm

which depicted the least dense and most dense for Chinese and Manipintar varieties respectively. By observations, the peanut seed lengths showed trend similarities with the bulk densities among the four varieties. Similar observations were made by Frimpong *et al.* (2007) who found a moderate statistical correlation ( $r^2 = 0.75$ ) between seed lengths and bulk densities of 14 Ghanaian peanut varieties. The bulk density indicates to a greater extent the amount of food substance that each variety contains and may be useful in selecting peanut varieties by processors for some specific uses. This physical measure may also be useful in designing packages for peanut seeds by weights since less dense varieties may require larger volumes to meet specified package weights.



**Fig. 8: Bulk density of four Ghanaian peanut**

#### 4.1.2 Chemical Characteristics

The chemical characteristics determined included moisture content, fat content, proteins, total ash and total crude carbohydrates (Table 7). The chemical characteristics were expressed in dry matter bases (except moisture).

#### 4.1.2.1 Moisture content

The moisture content of the peanut varieties was determined as the mass of water in a given mass of peanut flour removed by evaporation. Table 7 indicates that significant differences ( $p < 0.05$ ) in moisture contents were found among the varieties. The moisture contents of the four varieties of peanuts ranged from 6.34 - 8.53%. Chinese variety had the least moisture content while Sinkarzie had the highest. Tables 6 and 7 reveal that the varieties with the highest moisture contents may not necessarily be the densest. The amount of water in peanuts could largely be affected by the extent of drying of the produce after harvest and the ability to properly store the produce throughout the storage life of the commodity. The moisture contents of foods, although a less reliable indicator compared to water activity, is an easy to measure constituent for predicting the susceptibility of foods to chemical and microbiological spoilage (Fenema, 1996). According to Singh and Oswalt (1995) it is important to dry peanuts to moisture contents of 6-8% to reduce the risk of mould infection and avoid high levels of aflatoxins.

**Table 7: Chemical characteristics of four peanut varieties**

Peanut Variety	Moisture (%)	Fat (%)	Protein (%)	Ash (%)	Carbohydrates (%)
Manipintar	$8.13 \pm 0.13^b$	$52.02 \pm 0.33^a$	$22.73 \pm 0.51^c$	$2.21 \pm 0.30^a$	$22.39 \pm 0.31^b$
Chinese	$6.34 \pm 0.05^d$	$50.02 \pm 0.40^b$	$25.77 \pm 0.63^a$	$2.08 \pm 0.12^a$	$21.73 \pm 0.16^b$
Sinkazie (Red type)	$8.53 \pm 0.12^a$	$52.74 \pm 0.28^a$	$25.02 \pm 1.38^{ab}$	$2.44 \pm 0.16^a$	$19.08 \pm 1.26^c$
Bugla local	$6.86 \pm 0.05^c$	$49.84 \pm 0.21^b$	$23.24 \pm 0.15^{bc}$	$2.27 \pm 0.04^a$	$24.18 \pm 0.38^a$

Values are means and standard deviations of three replicates

Values in a column with different superscripts are significantly different ( $p < 0.05$ )

Apart from values of the moisture content, other values are reported as % dry matter.

#### 4.1.2.2 Fat Content

The fat contents of the four peanut varieties show significant differences ( $p < 0.05$ ) between some varieties (Table 7). Sinkarzie was found to have the highest fat content (52.74%) while Bugla local had the lowest (49.84%). The fat levels were within the widely reported average range of 50% and the range of 33.6 – 54.95% reported by Asibuo *et al.* (2008) for some Ghanaian peanut varieties. However, Chinese peanut variety which is perceived and corroborated by Asibuo *et al.* (2008) to have the highest fat content among Ghanaian peanut varieties was rather lower than Sinkarzie and Manipintar. Pickett (1949) indicated that apart from genetic differences among varieties of peanuts, fat production is greatly influenced by environmental factors and the stage of maturity of the pods at harvest. Hence the observed deviation may be more of an environmental influence rather than genetic since these varieties were obtained from different places. The variations in fat contents of the different varieties of peanuts could be a useful indicator of their usage in food applications. The higher fat content varieties could be used preferentially for peanut oil production.

Fats being the predominant constituent of peanuts make them an invaluable source of dietary energy for consumers. They also play an important role in achieving the texture, mouthfeel and aroma of many peanut products (Mazaheri-Tehrani *et al.*, 2009). The high peanut fat content of peanuts coupled with their high proportion of unsaturated fatty acids (approximately 80%) makes peanuts and peanut products susceptible to rancidity (Ahmed and Young 1982).

#### 4.1.2.3 Proteins

The protein contents of the four peanut varieties were within reported range 18.92 to 30.93% (Frimpong *et al.*, 2007; Asibuo *et al.*, 2008) for some varieties of Ghanaian peanuts.

Manipintar recorded the lowest average protein content of 22.73% while Chinese had the highest value of 25.77% (Table 7). Significant difference ( $p < 0.05$ ) in average protein contents were found among some varieties (Table 7). Chinese and Sinkarzie recorded higher protein contents (at least 25%) compared to Manipintar and Bugla local varieties. Similarly the two varieties also recorded fat contents of at least 50%. These two varieties could be considered in food fortification programmes purported at improving energy and protein density of deficient foods. Peanut proteins contribute significantly to the development of the nutty flavours and colour formation through Maillard reactions during roasting of peanuts (Grosso *et al.*, 2008).

#### **4.1.2.4 Carbohydrates**

The crude carbohydrate contents of peanuts were estimated as the percent difference after all the other chemical components (Table 7) were determined. The carbohydrate contents of the four varieties of peanuts ranged from the lowest of 19.08% for Sinkarzie to the highest of 24.18% for Bugla local. These values were within the range (19.02 -27.16%) reported by Asibuo *et al.* (2008) for some varieties of Ghanaian peanuts. Significant variations ( $p < 0.05$ ) in carbohydrate contents were observed among some varieties. Possibly, a combine effect of the protein and fat contents of the four peanut varieties influenced largely the average carbohydrate contents compared to taking each of these components individually as suggested by Frimpong *et al.*, (2007).

Peanut carbohydrates are important sources of energy and dietary fiber. They also contribute to the taste, of peanuts considering that 90% of the carbohydrates in peanuts are sucrose (Ng and Dunford 2009). Carbohydrates are also needed for the development of the characteristic

flavours and colours of roasted peanut products associated with Maillard reactions (Grosso *et al.*, 2008).

#### **4.1.2.5 Total Ash**

The mass of ash remaining after the combustion of biological substances is often used as the measure of the mineral content of the substance (Belitz *et al.*, 2009). The total ash content determined for the four varieties of peanuts didn't differ significantly ( $p > 0.05$ ) as observed in Table 7. Ash contents ranged from 2.08 to 2.44%. These were the lowest and highest average amounts for Chinese and Sinkarzie respectively. The major minerals such as calcium, potassium, sodium and magnesium as well as some trace minerals such as zinc, iron and manganese have all been found in Ghanaian peanuts to differ significantly (Asibuo *et al.*, 2008). Belitz *et al.* (2009) suggests that variations in the mineral contents of the same raw materials could be due to genetic and climatic factors as well as the farming practices, soil composition and maturity of the produce. Minerals in foods are important for their nutritional and physiological roles. However, some minerals (e.g. Cu, Fe) because undesirable colour changes, oxidize ascorbic acid, produce taste defects and off-flavours through fat oxidation in many food products (Belitz *et al.*, 2009).

#### **4.1.3 Selection of peanut variety for canned peanut soup base.**

The chemical and physical properties of peanuts provide useful information for the selection of peanuts for specific food applications. Among the four varieties of peanuts, Chinese variety was found to be the most variable in dimensions (Table 6) and could best be used for products such as peanut paste and peanut soup base where the seed dimension is not of importance.

The availability of raw materials is also an important criterion in the choice of materials for peanut soup base especially for purposes of commercialization. A preliminary survey and interactions with peanut distributors and sellers from three major markets (Timber, Madina and Nima markets) of the Greater Accra suggested that Chinese variety was available in the markets throughout the year. Ibrahim *et al.*, (2012) in a survey among farmers in the Northern region were over 80% of Ghanaian peanuts (Tsibey *et al.*, 2003) are cultivated also revealed that majority of the peanut farmers cultivated Chinese variety (50%) compared to Manipintar (38%), Bugla local (11%) and Sinkarzie (1%). The survey found that membership in a farm organization, the location of the farm and the duration of maturity of the peanuts influenced the choice of peanut variety cultivated by farmers (Ibrahim *et al.*, 2012).

## **4.2 Evaluation of the efficacy of manual sorting method to reduce aflatoxins in peanuts**

Peanuts are susceptible to aflatoxin contamination. In order to reduce aflatoxins in peanuts a manual colour sorting method was developed by Galvez *et al.* (2002) for use by small scale industry. However, concerns that the method is laborious, labour intensive and can lead to significant weight losses have been raised. The sorting method was evaluated to assess the proportions (%) of weight loss due to sorting and to determine aflatoxin concentrations of the sorted components.

### **4.2.1 Assessment of peanuts weight losses for the manual colour sorting method**

The manual colour sorting process involves major unit operations such as pre-sorting, blanching, and colour sorting. Table 8 shows the proportions (%) of the sorted peanut components using material and mass balance sheet.

**Table 8: Proportions (%) of the sorted components of peanuts (Chinese variety) using manual colour sorting**

Unit operation	Peanut components	Weight (g)	Proportion of sorted peanut components at each unit operation (%)	Proportion of sorted peanut components per total weight of unsorted peanuts (%)
	<i>Raw unsorted peanuts input</i>	5000		
Pre-sorting	Good pre-sorted peanuts	3723.7	74.5	74.5
	Shriveled and damaged peanuts	1273.5	25.5	25.5
	Loss (waste-husk, leaves)	2.8	0.1	< 0.1
	<i>Pre-sorted peanuts input</i>	3723.7		
Blanching	Hulls/Skins	152.3	4.1	3.0
	Blanched peanuts	3426.4	92.0	68.5
	Loss (moisture + other )	145	3.9	2.9
	<i>Weight of blanched peanuts input</i>	3426.4		
Colour Sorting	Discoloured peanuts	460.9	13.5	9.2
	Clean peanuts (no discolouration)	2962.5	86.5	59.3
	Other Losses	3.0	0.1	< 0.1

#### 4.2.1.1 Pre-sorting operation

The manual colour sorting method of reducing aflatoxin contamination of peanuts begins with pre-sorting. At pre-sorting, the raw unsorted peanut kernels/seeds are sorted to remove any leaves or debris, the shriveled and damaged peanuts from the good peanuts. Table 8 indicates that, for 5000g of peanuts pre-sorted, 74.5% were good while over 25% were the recorded total loss fraction. The loss was largely due to shriveled and damaged peanuts. The large proportions of shriveled and damaged peanuts suggest a bad harvest or poor storage

conditions of peanuts. The critical economic issue of concern is who bears the loss, the farmer or the peanut processor? Poor soil fertility and poor rainfall patterns are important factors which affect peanut pod development (Naturland, 2000). These could possibly affect adversely the quality of harvested peanuts. Jordan *et al.* (2011) and Hartzog and Adams (1973) suggested the addition of gypsum to calcium deficient soils (212kg/ha or less) for good pod development and prevention of excessive shriveling of peanuts. Naturland (2000) also recommends irrigation farming to avoid shriveling caused by drought stresses.

#### **4.2.1.2 Blanching operation**

The process of blanching in the peanut industry describes light roasting peanuts and dehulling to remove the skins/hulls (Whitaker, 1997). The blanched (dehulled) peanuts were 92.0% of the good presorted peanuts and 68.5% of the total raw unsorted peanuts (Table 8). The total weight losses at the blanching stage due to dehulling (hulls - 4.1%) and other losses (e.g. Moisture) was less than 8% of the presorted peanuts. The total loss in mass was however large (about 31%) when the raw unsorted peanuts were considered. Hence food processors could be advised to purchase presorted peanuts or buy raw unsorted peanuts of good quality so as to reduce weight losses due to shriveled peanuts.

#### **4.2.1.3 Colour sorting operation**

Colour sorting involves handpicking discolored peanuts from the blanched peanuts to obtain *clean* peanuts. The mass of the *clean* peanuts after the whole process of manual colour sorting was 86.5% of the blanched peanuts and 59.3% of the unsorted peanuts (Table 8). The Table 8 indicates that, the weight losses due to discolorations was about 13.5% of the blanched peanuts and 9.2% of the unsorted raw peanuts at the colour sorting stage. Discoloured peanuts are removed because they are usually associated with high aflatoxins

(Ganzer, 1999). Discolorations on peanuts are believed to be caused by insects and mould infestations (Whitaker, 1997). Peanuts become susceptible to insect and microbial infestations if their moisture contents exceed 8% (Price *et al.*, 2005). Hence rapid drying of harvested pods (using mechanical drying) and proper storage facilities could reduce losses due to discolorations which are caused by insects and microbial activities.

#### **4.2.2 Aflatoxin levels of the sorted Chinese peanut components**

Table 9 shows the aflatoxin concentrations of five selected components of the Chinese peanut variety sorted out using the manual colour sorting method. The batch of raw peanuts used had an estimated total aflatoxin level of 25µg/kg. Although the total aflatoxin concentrations in the unsorted peanuts were relatively low, the shriveled and damaged peanuts had very high total aflatoxin levels (310.92µg/kg). Aflatoxins B<sub>1</sub> and B<sub>2</sub> were the major contributors to the high levels of aflatoxins in the shriveled and damaged seeds with concentrations of 251.23 and 59.07µg/kg respectively. Aflatoxin G<sub>1</sub> was less than 1µg/kg in the shriveled and damaged peanuts. All the other components did not show detectable levels of aflatoxins except the *clean* peanuts which recorded a low amount of 0.17µg/kg for aflatoxins G<sub>1</sub>. Chromatograms depicting the levels of aflatoxin concentrations are shown in appendix 7.2

The low total aflatoxins concentrations for the unsorted peanuts compared to the shriveled and damaged peanuts could be attributed to the dilution effect due to presence of peanuts that were either very low in aflatoxins or had no aflatoxins at all. The very high aflatoxins concentrations for the shriveled and damaged peanuts were expected. Generally, small, immature and damaged peanuts are highly susceptible to aflatoxigenic mould infestations during pod development or poor postharvest storage conditions (Whitaker, 1997). According to Bakole and Adebajo (2003), *A. flavus* is the commonest of the aflatoxigenic moulds

which produces aflatoxins B<sub>1</sub> and B<sub>2</sub> while *A. parasiticus* produces G<sub>1</sub> and G<sub>2</sub> in addition to B<sub>1</sub> and B<sub>2</sub>. The results suggested that a lot of aflatoxin production were probably the result of *A. flavus* activities as reported by Bradburn *et al.* (1993) since B<sub>1</sub> and B<sub>2</sub> were the major aflatoxins detected.

The discoloured peanuts which recorded none detectable levels of aflatoxins was rather unexpected. Ganzer (1999) found a strong positive relationship between the amounts of discoloured and mouldy peanuts and the levels of aflatoxin. The observed deviation probably reveals that moulds that infect peanuts causing discolorations may not produce aflatoxins. The results support the suggestion that the presence of aflatoxigenic moulds may not necessarily lead to aflatoxins production because aflatoxin production in peanuts is influenced by conditions such as moisture content, environmental temperature, humidity, nitrogen stress and oxidative stress (Hell and Mutige, 2011; Guo *et al.*, 2009). The deviation could also be attributed to proper sorting out of all shriveled and damaged peanuts at the presorting stage since these could have passed onto the colour sorting stage and be sorted among the discoloured peanuts. The total aflatoxin level of 0.17µg/kg for the *clean* peanuts was within regulatory limits for most countries of the world including Europe (maximum limit 4µg/kg) and United States of America (maximum limit 20 µg/kg) (FVO, 2007).

**Table 9: Aflatoxin concentrations of sorted out components of Chinese peanuts obtained from Nima market**

Peanut components	Aflatoxins concentration ( $\mu\text{g}/\text{kg}$ )				Total aflatoxins concentration ( $\mu\text{g}/\text{kg}$ )
	B <sub>1</sub>	B <sub>2</sub>	G <sub>1</sub>	G <sub>2</sub>	
Shriveled and damaged seeds	251.23	59.07	0.62	ND	310.92
Less than 50% discoloured seeds	ND	ND	ND	ND	ND
More than 50% discoloured seeds	ND	ND	ND	ND	ND
Hulls/Skins	ND	ND	ND	ND	ND
Clean seeds (no discolouration)	ND	ND	0.17	ND	0.17

ND – No aflatoxins detected at the limits of detection: G<sub>1</sub> and G<sub>2</sub> = 0.13 $\mu\text{g}/\text{kg}$ ; B<sub>1</sub> and B<sub>2</sub> = 0.15 $\mu\text{g}/\text{kg}$   
%discoloration is done by visual observation considering seed length.

Total aflatoxins for Raw Unsorted Peanuts = 25 $\mu\text{g}/\text{kg}$

### 4.3 Determination of the optimum ingredient formulation for canned peanut soup base.

#### 4.3.1 Effect of ingredient mixture components on sensory attributes of formulated peanut soups

Table 10 shows the mean liking scores of the sensory attributes and the mean overall acceptability scores for the eight (8) formulated peanut soups.

**Table 10: Mean liking scores for sensory attributes of peanut soup samples**

Sample- P:O:T (g)	Colour	Thickness	Spiciness	Hotness	Taste	Smooth- ness	Overall Accept- ability
<b>5:180:75</b>	4.62±2.06 <sup>d</sup>	4.81±2.40 <sup>ab</sup>	4.28±2.28 <sup>ab</sup>	4.40±2.34 <sup>ab</sup>	3.94±2.05 <sup>a</sup>	5.94±2.23 <sup>a</sup>	3.54±2.33 <sup>a</sup>
<b>15:170:75</b>	4.02±1.88 <sup>d</sup>	4.28±2.20 <sup>a</sup>	5.02±2.51 <sup>bc</sup>	5.62±2.88 <sup>c</sup>	4.91±2.21 <sup>b</sup>	5.79±2.33 <sup>a</sup>	4.04±2.54 <sup>ab</sup>
<b>10:131:119</b>	5.62±2.92 <sup>bc</sup>	4.09±2.57 <sup>a</sup>	4.04±2.46 <sup>a</sup>	3.83±2.42 <sup>a</sup>	3.13±1.84 <sup>a</sup>	5.06±2.57 <sup>a</sup>	3.17±2.63 <sup>a</sup>
<b>5:105:150</b>	6.96±1.92 <sup>a</sup>	6.19±1.71 <sup>c</sup>	5.83±1.98 <sup>cd</sup>	4.94±2.21 <sup>bc</sup>	6.04±2.10 <sup>cd</sup>	6.17±2.21 <sup>a</sup>	5.37±2.90 <sup>c</sup>
<b>15:100:145</b>	5.89±2.10 <sup>b</sup>	5.51±2.34 <sup>bc</sup>	5.51±2.26 <sup>cd</sup>	5.09±2.26 <sup>bc</sup>	5.45±2.09 <sup>bc</sup>	5.68±2.28 <sup>a</sup>	4.85±2.79 <sup>bc</sup>
<b>10:100:150</b>	5.85±2.16 <sup>b</sup>	5.36±2.44 <sup>bc</sup>	6.38±2.01 <sup>d</sup>	5.57±2.41 <sup>c</sup>	6.32±2.03 <sup>d</sup>	5.89±2.10 <sup>a</sup>	5.48±2.83 <sup>c</sup>
<b>15:100:145</b>	4.79±2.38 <sup>dc</sup>	5.53±2.20 <sup>bc</sup>	5.66±2.13 <sup>cd</sup>	5.11±2.21 <sup>bc</sup>	5.70±1.93 <sup>bcd</sup>	5.49±2.16 <sup>a</sup>	4.74±2.52 <sup>bc</sup>
<b>15:170:75</b>	5.70±2.41 <sup>b</sup>	5.40±2.58 <sup>bc</sup>	5.89±2.13 <sup>cd</sup>	5.47±2.28 <sup>c</sup>	6.13±2.22 <sup>cd</sup>	4.81±2.50 <sup>a</sup>	5.54±2.86 <sup>c</sup>

Means are average scores of 50 panelists using a Hedonic scale from 1- dislike extremely to 9- like extremely.

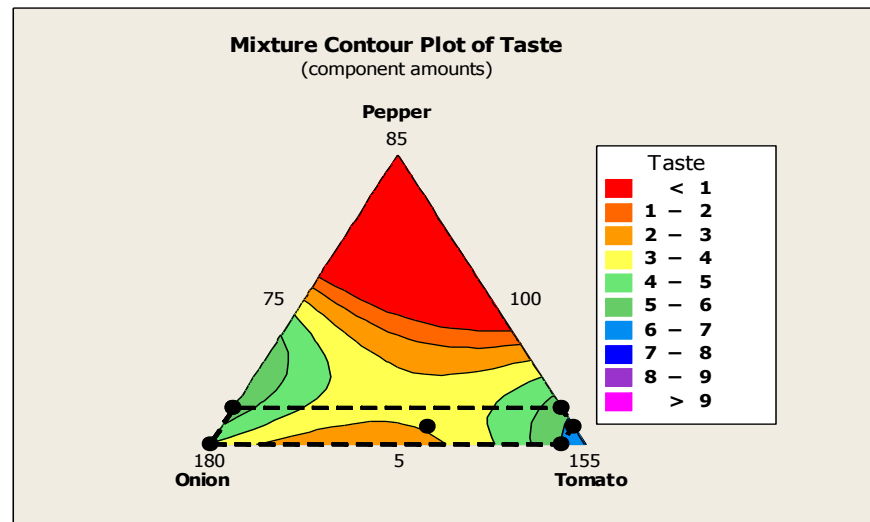
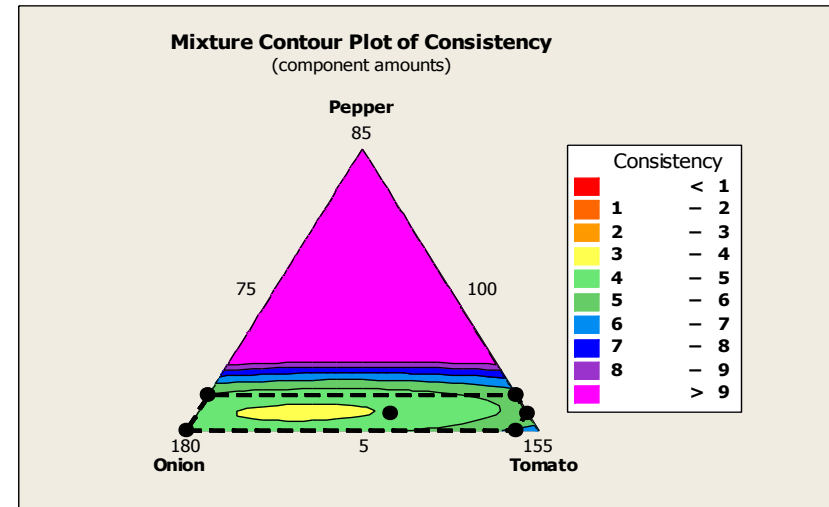
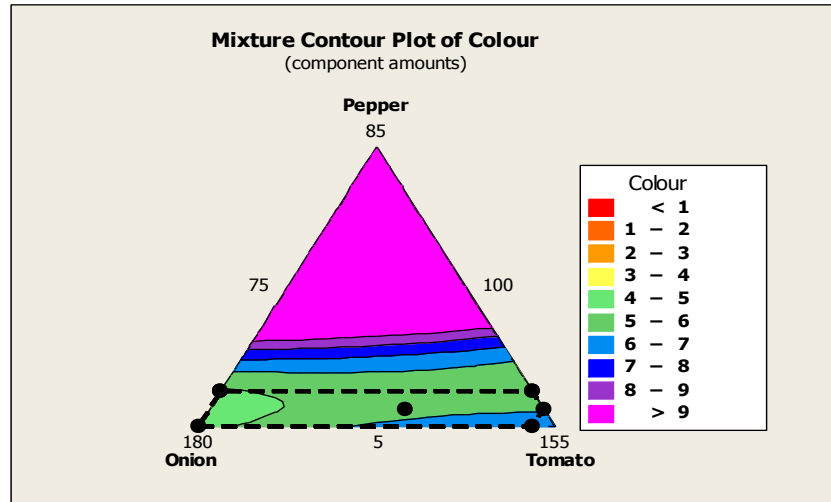
Values with different superscripts within the same column are significantly different ( $p < 0.05$ )

P – pepper, O – onion, T – tomato paste (Total secondary ingredients mixture = 200g per sample)

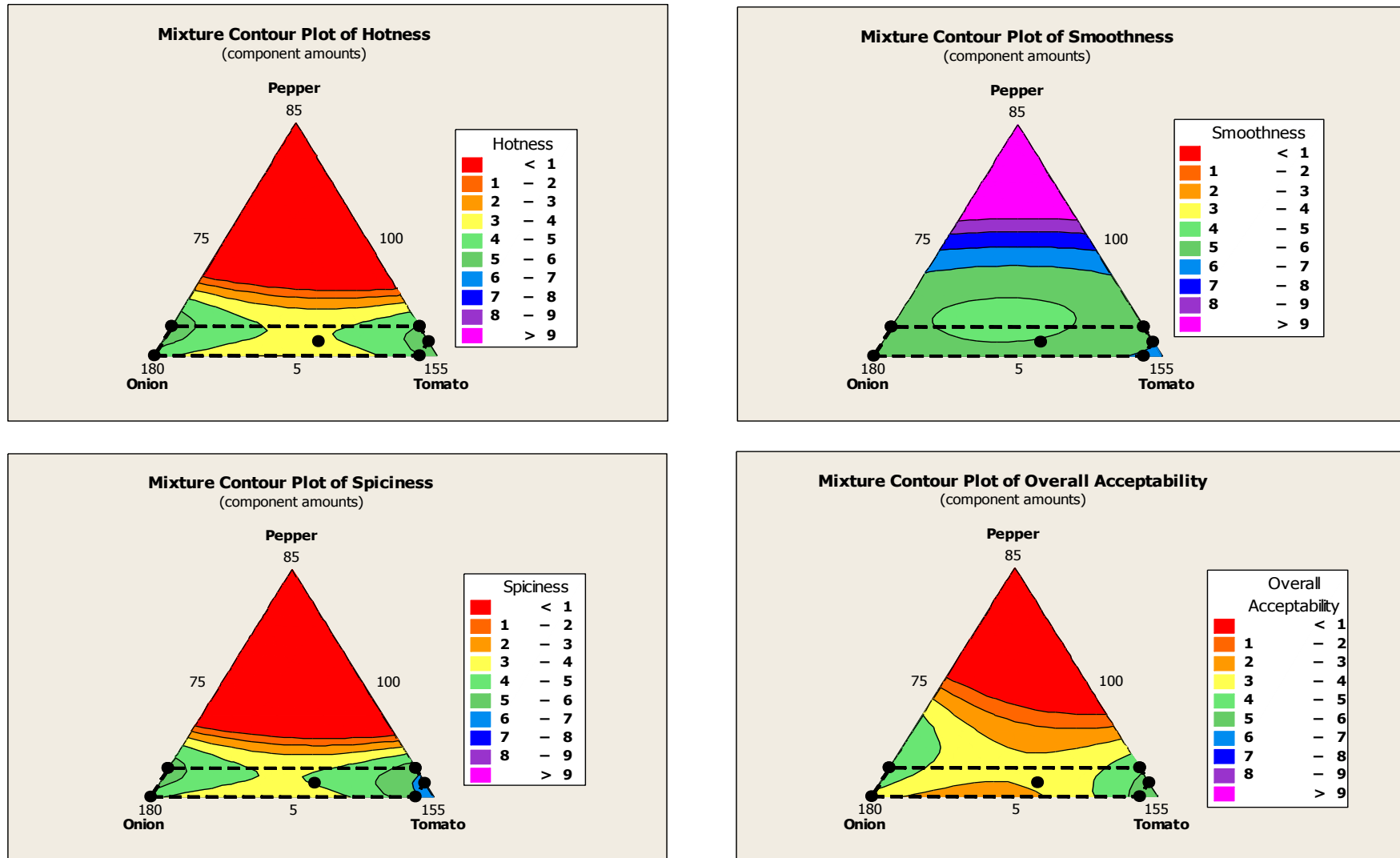
Generally the mean liking scores of the sensory attributes were lower than 7. Significant differences ( $p < 0.05$ ) were observed in the mean liking scores for colour, hotness, spiciness, taste, thickness and the overall acceptability of the formulated soup samples. However, the mean liking scores for smoothness of the soups did not differ significantly ( $p > 0.05$ ). The uniformity of the milling and grinding of all the ingredients used could have accounted for the no significant differences recorded for smoothness. The mean overall acceptable scores also showed significant differences among some formulations (Table 10). The mean overall acceptability scores were also generally low (less than 6) considering that a 9-point hedonic scale was used. There were only three ingredient mixtures (5:105:150, 10:100:150, 15:170:75) that scored higher than 5 for the overall acceptability of the formulated peanut soups. The significant variations in the mean liking scores among some formulations indicate that the mixture ingredients influenced greatly the sensory perception and acceptability of the formulated peanut soups. The rather low liking scores were probably because meat/fish and

seasonings were not used in the preparation of the soups. This was done to prevent any nuisance variables that could mask the effects of the mixture ingredients.

The mixture contour plots (Fig. 9) for colour, taste and thickness (consistency) suggest that these three sensory attributes of the peanut soup formulations were liked most at higher proportions of tomato paste. The formulations with higher tomato proportions recorded higher than 5 mean liking scores for colour, thickness and taste within the experimental region. Peanut soup formulations that contained higher proportions of pepper and lower onions were also liked most in terms of the hotness and spiciness of the peanut soups (Fig. 10). The contour plot for overall acceptability (Fig. 10) shows that the peanut soups were preferred most at higher amounts of tomato pastes and pepper.



**Fig. 9: Effects of ingredient mixtures on likeness of peanut soup colour, consistency (thickness) and taste**



**Fig. 10: Effects of the ingredient mixtures on likeness of peanut soup hotness, smoothness, spiciness and overall acceptability**

Table 11 shows the fitted mixture regression models for the sensory responses. With respect to the regression models, hotness was the only attribute that was affected significantly ( $p < 0.05$ ) by a positive interaction of onions and pepper and a negative interaction of tomato paste and onions . The very high  $R^2$  values (91.57 - 99.58) indicated that the fitted models strongly explained the effects of the ingredient mixtures on the spiciness, hotness and taste of the peanut soups. Also, about 80% ( $R^2$ ) of the thickness of the peanut soups was accounted for by the fitted model. The models however weakly ( $R^2 = 65.64$  and  $66.67\%$ ) explained the effects of the ingredient mixture on peanut soup's colour and smoothness. It suggests that factors other than the mixture ingredients influence the liking of the colour and smoothness of soups. The models also revealed that the interaction terms of pepper and onion as well as pepper and tomato paste had the highest positive effect on the overall acceptability of the peanut soups. The fitted regression model could explain moderately ( $R^2 = 80\%$ ) the effects of the ingredient mixtures on overall acceptability of the peanut soups.

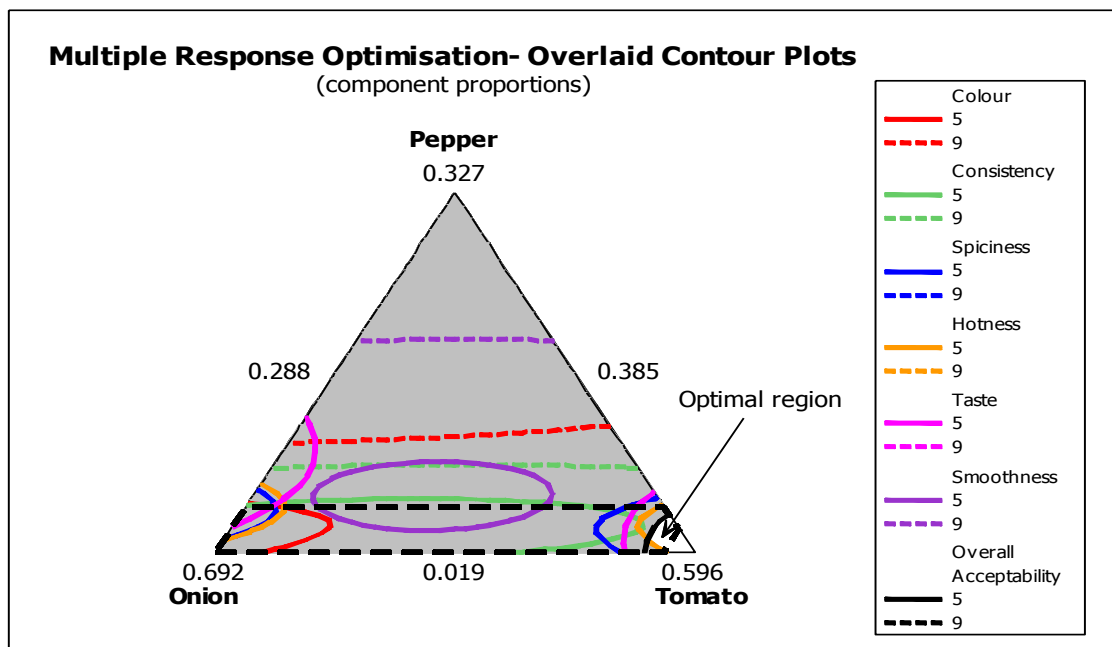
**Table 11 Mixture regression models of sensory attributes of the ingredient mixtures**

Model Component	Model coefficient for responses						
	Colour	Thickness	Spiciness	Hotness	Taste	Smoothness	Overall Acceptability
P: Pepper	726.20	1592.00	-962.70	-811.40	-281.50	207.50	-282.70
O: Onion	-0.80	12.00	16.00	15.50	21.30	12.00	15.00
T: Tomato	8.40	22.00	35.50	29.30	48.30	16.90	35.80
PO	-726.60	-1699.00	1118.70	943.60*	410.40	-237.10	395.00
PT	-883.00	-1777.00	944.70	811.90	161.30	-247.20	185.30
OT	16.70	-39.00	-93.40	-81.30*	-128.70	-33.70	-90.40
$R^2(\%)$	65.64	81.74	91.63	99.58	91.57	66.67	80.48

\*significant at  $p < 0.05$

### 4.3.2 Optimum peanut soup formulation for canned peanut soup base

The contour plots of all the sensory attributes and the overall acceptability were overlaid on each other to obtain the optimum region (Fig. 11). The maximum and minimum values for all attributes during optimisation were set as “9-like extremely” and “5-neither like nor dislike” respectively. The choice of mean liking of 5 was to ensure that every attribute was included in the optimization process. Two optimum peanut soup ingredient combinations that were selected from the optimum region were 7.5:102.2:150.3 and 12.3:100.8:146.9 which represent the proportions of pepper, onion and tomato pastes respectively. These were used to validate the peanut formulation for making the canned peanut soup base.



**Fig.11: Overlaid contour plots of peanut soup sensory attributes showing optimum region**

### 4.3.3 Validation of peanut soup formulation.

The validation process involved the selection of two optimal combinations of ingredients from the optimum region and two formulations outside the optimum region. Peanut soup samples were prepared using the four formulations and served to a consumer panel of 50. Table 12 shows the ingredient combinations for the validation process.

**Table 12: Ingredient combinations from the optimal and non-feasible regions used for validation of peanut soup**

Sample	Ingredient amounts (g)		
	Pepper	Onion	Tomato
1: OR 1	7.50	102.20	150.30
2: OR 2	12.30	100.80	146.90
3: NF 1	10.50	140.50	109.50
4: NF 2	14.20	160.30	85.50

OR- Optimal Region , NF- Non-feasible formulation

**Table 13: Mean liking scores of peanut soups during validation**

Sample	Responses (Sensory Attributes' Scores)						
	Colour	Thickness	Hotness	Smoothness	Spiciness	Taste	Overall Acceptability
<b>Predicted scores</b>	4.62-6.96	4.09-6.19	3.83-5.57	5.06-6.17	4.04-6.38	3.13-6.32	3.17-5.48
1: OR 1	6.68±1.72 <sup>a</sup>	6.72±2.20 <sup>a</sup>	5.48±2.08 <sup>a</sup>	5.78±2.16 <sup>a</sup>	5.36±2.42 <sup>a</sup>	5.96±2.47 <sup>a</sup>	5.92±2.42 <sup>a</sup>
2: OR 2	7.40±1.93 <sup>a</sup>	7.16±2.23 <sup>a</sup>	5.70±2.30 <sup>a</sup>	6.04±2.15 <sup>a</sup>	5.84±2.29 <sup>a</sup>	6.44±2.34 <sup>a</sup>	6.72±2.44 <sup>a</sup>
3: NF 1	5.74±2.05 <sup>b</sup>	5.40±2.28 <sup>b</sup>	5.50±2.36 <sup>a</sup>	6.14±2.24 <sup>a</sup>	5.36±2.45 <sup>a</sup>	5.42±2.50 <sup>a</sup>	5.80±2.45 <sup>a</sup>
4: NF 2	4.78±2.30 <sup>c</sup>	5.40±2.32 <sup>b</sup>	5.56±2.52 <sup>a</sup>	6.18±2.18 <sup>a</sup>	5.10±2.53 <sup>a</sup>	5.30±2.50 <sup>a</sup>	5.44±2.49 <sup>a</sup>

Means are average liking scores of 50 panelists using a Hedonic scale from 1- dislike extremely to 9- like extremely.

Values with different superscripts within the same column are significantly different ( $p < 0.05$ )

OR- Optimal Region, NF- Non-feasible formulation

The results of the validation study are shown in Table 12. Generally, samples prepared using formulations from the optimal region (OR1 and OR2) generally recorded higher mean liking scores for all the sensory attributes compared to those prepared from formulations outside the feasible region (NF1 and NF2). However, significant differences ( $p < 0.05$ ) in the sensory attributes were recorded for only the colour and thickness of the formulated peanut soups. Table 12 also reveals that formulation OR2 gave consistently higher mean liking scores for all the sensory attributes compared to the other three formulations. Generally the mean liking scores of most of the sensory attributes evaluated were within the limits of the predicted scores.

Similarly, the mean overall acceptability scores of peanut soup samples from the optimal region (OR1 and OR2) had higher mean acceptability scores (5.92 and 6.72) compared to those from the non-feasible region (5.44 and 5.80). However, no significant difference ( $p > 0.05$ ) in overall acceptability scores for the peanut soup samples was recorded. The mean overall acceptability scores for the four peanut soup formulations were slightly higher than the upper limits of the predicted scores (5.48). Formulation OR2 which gave the most desirable responses also recorded the highest mean overall acceptability score (6.72). Ingredients combination for formulation OR2 (Table 12) could give the most desirable peanut soup base.

#### 4.4 Establishing the thermal process variables (temperature and time) and quality evaluation of canned peanut soup base.

##### 4.4.1 Effect of the thermal process (time–temperature combinations) on heat penetration of canned peanut soup base

Heat penetration test was conducted to determine the temperature of the product at slowest heating point (SHP) during thermal processing. The maximum attainable temperatures at the SHPs are shown in Table 14 for each of the time-temperature combinations.

**Table 14: Maximum temperatures (°C) at slowest heating points (SHP) of canned peanut soup base retorted at different temperature-time combinations**

Temperature (°C)	Time (mins.)	Maximum temperature at SHP (°C)
115	40	94.2
115	50	97.6
115	60	99.8
120	40	97.8
120	50	101.6
120	60	104.5
125	40	104.4
125	50	102.5
125	60	110.5

Initial product temperature before retorting - 64±1°C

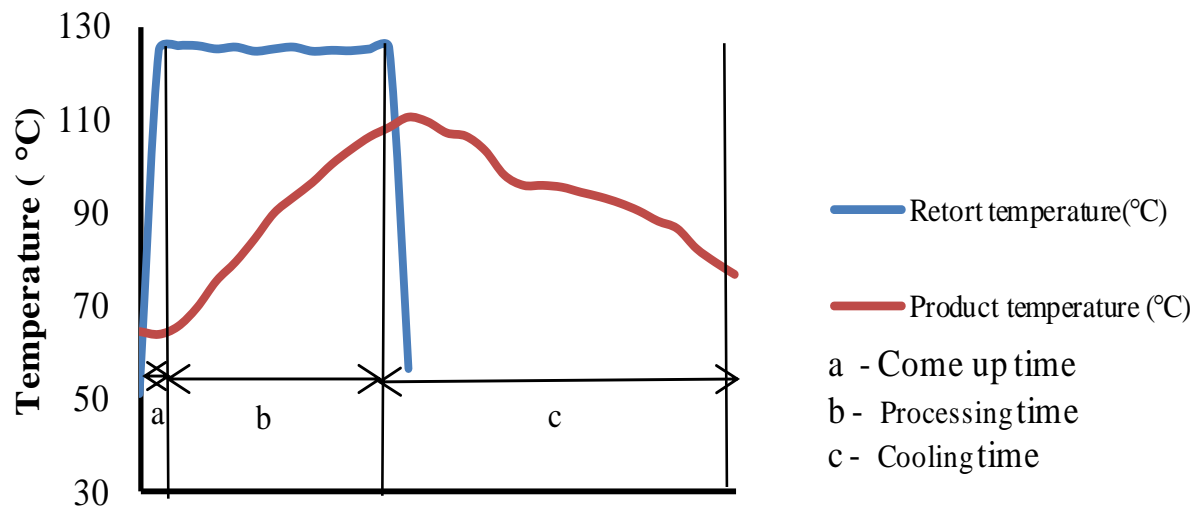
Table 14 shows that four (4) of the thermal process time-temperature combinations did not attain temperature of 100°C at SHP. Although the initial product temperature prior to retorting was at an average temperature of 64±1°C, none of the products attained their processing temperatures at SHP (Table 14). The highest recorded temperature at the slowest heating point was 110°C for the thermal process of 125°C for 60mins while the lowest of 94.2°C was recorded for thermal process of 115°C for 40mins. Generally the temperature at the SHP increased as the thermal process time and temperatures increased. However, the thermal process of 125°C/50mins had an unexpectedly lower maximum SHP. The deviation could be attributed to non-uniformity in the retort heating process or non-uniformity in the product heating characteristics (Smout *et al.*, 2000). Although the deviation could be treated as an outlier, its inclusion could yield a more robust thermal process design for the product since such deviations do occur in commercial thermal processing.

Fig. 12 depicts a typical heat penetration curve for the severest thermal process of 125°C for 60mins during heat penetration testing. The other heat penetration curves are shown in appendix.

7.3. The curves suggest that the rise in temperatures at SHP of the product was gradual compared to the retort temperatures which rose rapidly to the processing temperatures. The cooling phases also recorded very slow heat loss from the product after retorting resulting in longer cooling times.

These observations could be attributed to the viscous nature of the product and the type and dimensions (99 x 118mm) of the can used which probably reduced the rate of heat exchange at the SHP of the container. According to Awuah *et al.* (2007), viscous products tend to limit the transfer of heat by convection which is a much faster type of heat transfer compared to heat transfer by conduction. Other factors which affect heat penetration of canned products include the product

particle size and composition, the processing system used (type of retort), initial product temperature and the choice of thermal process (time and temperature combinations) (Berry and Pflug, 2003). Considering these factors either individually or in combination may be useful in attaining higher product temperatures at SHP during thermal processing.



**Fig. 12: Heat penetration curve for canned peanut soup base processed at 125°C for 60mins.**

#### **4.4.2 Effect of the thermal process (time-temperature combinations) on the sterilizing value ( $F_o$ ) of canned peanut soup base**

The sterilizing value ( $F_o$ ) depicts the total lethal effect or the severity of a thermal process when a reference temperature of 121.1°C and a  $z$  value of 10°C are used to establish the process (Berry and Pflug, 2003). Table 15 shows the calculated  $F_o$  values for the different time-temperature combinations of peanut soup base. The  $F_o$  values range from 0.04mins at 115°C/40mins to the highest of 1.9mins at 125°C/60mins. The estimated  $F_o$  values were less than the minimum  $F_o$  of

3mins recommended for low acid ( $\text{pH} > 4.6$ ) canned foods to achieve commercial sterility (Awuah *et al.*, 2007).

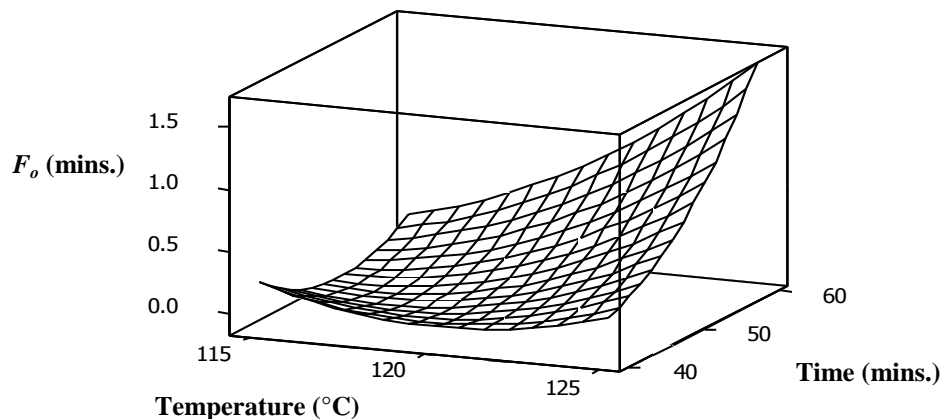
**Table 15: Sterilizing values for canned peanut soup base processed at different time-temperature combinations**

Temperature ( $^{\circ}\text{C}$ )	Time (mins.)	$F_o$ (mins.)
115	40	0.04
115	50	0.09
115	60	0.18
120	40	0.12
120	50	0.27
120	60	0.38
125	40	0.28
125	50	0.33
125	60	1.92

$F_o$  – sterilizing value.

Fig. 13 illustrates the effect of the processing time and processing temperature on  $F_o$ . High increase in  $F_o$  values were realized at only higher temperature and longer time processes. This could be attributed to the poor heat penetration rates of the products which resulted in low temperatures at the SHP for the lower temperature and shorter time thermal treatments. Generally, high lethal effects ( $F_o$ ) are achieved at SHP temperatures above  $100^{\circ}\text{C}$  (Warne, 1988). In order to achieve higher  $F_o$  values, the initial peanut soup base temperatures could be raised above  $65^{\circ}\text{C}$ . This could reduce the time it takes the product to reach high SHP temperatures and also enable the

product to be maintained at high processing temperatures for longer duration. The usage of cans with smaller dimensions and/or agitating retorts could improve the rate of heat penetration to achieve  $F_o$  of at least 3mins.



**Fig. 13: Effect of thermal process (time-temperature) on the sterilizing value ( $F_o$ ) value of the retorted canned peanut soup base.**

Regression modeling of the thermal processes indicates that none of the model components (linear, quadratic or interaction terms) had any significant effects ( $p > 0.05$ ) on the  $F_o$  values (Table 16). However, only the quadratic and interactions terms had positive effects on  $F_o$ . The  $R^2$  and  $R^2$  adjusted values were 85.3% and 60.8% respectively. This indicated that the thermal process model moderately explains the effect of thermal process on  $F_o$ . The low  $R^2$  values could be attributed to the deviation observed in heat penetration data since temperatures at SHP (Table 14) were used to calculate  $F_o$ . Optimization of the retort performance and minimizing product variability are two key processes that could improve the fitted model (Smout *et al.*, 2000).

**Table 16: Model coefficients for sterilizing values ( $F_o$ ) of the time-temperature combinations**

Model components	Coefficients	P-Values
Constant term	165.722	0.773
A:Temperature	-2.381	0.088
B:Time	-1.123	0.106
AA	0.009	0.461
BB	0.003	0.392
AB	0.008	0.131
R <sup>2</sup>	85.3%	-
R <sup>2</sup> -adjusted	60.8%	-

Model components with  $p < 0.05$  have significant effects on  $F_o$

#### 4.4.3 Effect of thermal process (time-temperature combinations) on colour (L\*a\*b\*) of canned peanut soup base.

Colour as perceived consist of three dimensions: lightness, hue and chroma (Minolta, 1991). The L\*a\*b\* colour system represents closely human sensitivity to colour where L\* is the lightness variable a\* and b\* are chromaticity (hue and chroma) coordinates representing green to red and blue to yellow respectively (Minolta, 1991). The L\*a\*b\* values of the peanut soup base retorted at different time-temperature combinations are shown in Table 17. The mean L\*a\*b\* values of the unretorted product (control) were L\* 58.79, a\* +9.95 and b\* +36.34. These were generally higher than the L\*a\*b\* measurements for the retorted peanut soup base which ranged from L\* 52.30 – 56.40, a\* +8.63 - +9.64 and b\* +30.04 - +34.94. The a\* and b\* values suggested that peanut soup base had shades of red and yellow coloration since the coordinate measurements are positive (Minolta, 1991). Measurements of the L\* are done from 0 - 100 which represent white to dark

colours respectively. The mean L\* values of the product ranged from a lighter colour of 56.40 to a darker colour of 52.30 for the thermal processes of 115°C for 40mins and 125°C for 60mins respectively (Table 17). The L\* showed consistently lower values as the processing temperature and processing time increased. These observations suggest that the colour of the peanut soup base was affected by the severity of thermal processes. A response surface plot of the L\* (Fig. 14) shows the effect of the processing time and temperature on the colour (L\*) of the peanut soup base.

**Table 17: Colour (L\*a\*b\*) of canned peanut soup base processed at different time-temperature treatments.**

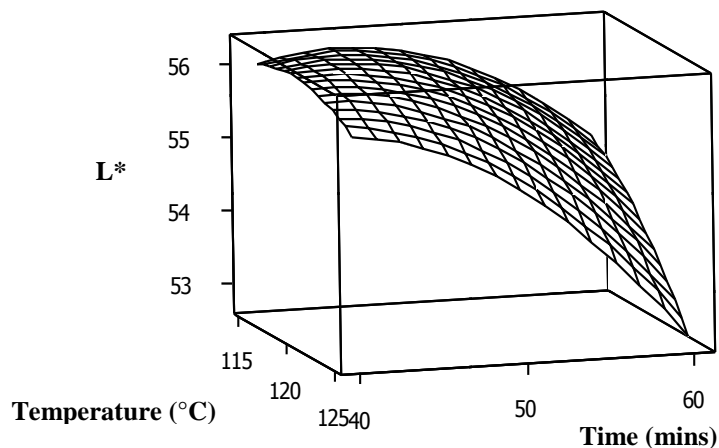
Temperature (°C)	Time (mins)	L*	+a*	+b*
115	40	56.40 ± 0.04	9.40 ± 0.03	34.54 ± 0.45
115	50	55.73 ± 0.05	9.64 ± 0.05	34.97 ± 0.17
115	60	54.71 ± 0.35	9.63 ± 0.11	33.89 ± 0.61
120	40	55.84 ± 0.12	9.31 ± 0.05	34.02 ± 0.13
120	50	55.43 ± 0.07	9.38 ± 0.11	33.63 ± 0.15
120	60	54.62 ± 0.22	9.05 ± 0.07	32.86 ± 0.37
125	40	55.73 ± 0.07	9.22 ± 0.04	33.84 ± 0.21
125	50	55.51 ± 0.23	8.63 ± 0.02	30.04 ± 0.48
125	60	52.30 ± 0.15	9.41 ± 0.33	32.28 ± 0.71
Control	-	58.79 ± 0.19	9.95 ± 0.09	36.34 ± 0.15

L\* - lightness from dark (0) to white (100), +a\* - redness, +b\* - yellowness

Control – Peanut soup base that was not retorted/sterilized

The response surface plot shows that the L\* values didn't change significantly at shorter processing times even when the temperature increased significantly. However, L\* values decreased as processing time increased at both low and high processing temperature. The

decreasing  $L^*$  values depicts darkening of the product colour as the severity of the thermal processes increased. The darkening of the product could be attributed to non-enzymatic browning (Maillard reactions) and also the charring of the canned product on the can lids. Peanut products brown at high processing temperatures because they have the basic chemical constituents – proteins and sugars – needed for Maillard reactions. Apart from the changes in colour which occur, Maillard reactions also lead to losses in amino acids - lysine and histidine (Awuah *et al.*, 2007).



**Fig. 14: Effect of thermal process (time-temperature) on the colour ( $L^*$ ) of canned peanut soup base**

Regression analyses were done to show the effects of thermal processing on  $L^*$  by fitting the  $L^*$  data to a regression model as shown in Table 18. The quadratic and interaction components of the fitted model for the thermal treatments had no significant effects ( $p > 0.05$ ) on  $L^*$ . However, the retort time and constant terms had significant effects ( $p < 0.05$ ) on  $L^*$ . The  $R^2$  and  $R^2$ -adjusted of

88.2% and 68.5% respectively indicated that the fitted model only moderately accounted for the observed thermal process effects on  $L^*$  values.

**Table 18: Model coefficients of lightness ( $L^*$ ) of peanut soup base processed at different time-temperature combinations**

Model Components	Coefficients for $L^*$	P-Values
Constant term	-127.988	0.000
A:Temperature	2.565	0.139
B:Time	1.562	0.031
AA	-0.009	0.658
BB	-0.006	0.281
AB	-0.009	0.287
$R^2$	88.2%	-
$R^2$ -adjusted	68.5%	-

Model components with  $p < 0.05$  have significant effects on  $L^*$

#### 4.4.4 Effect of thermal process (time-temperature combinations) on the total colour difference ( $\Delta E^*$ )

The total colour difference ( $\Delta E^*$ ) indicates the magnitude of colour change in the retorted products when compared to the unretorted product. The total colour differences ( $\Delta E^*$ ) were calculated from the measured  $L^*a^*b^*$  values of the retorted and the unretorted peanut soup products. Table 19 depicts the total colour difference between unretorted (control) and the retorted peanut soup base.

**Table 19: Total colour differences between unretorted and the retorted peanut soup base processed at different time-temperature treatments.**

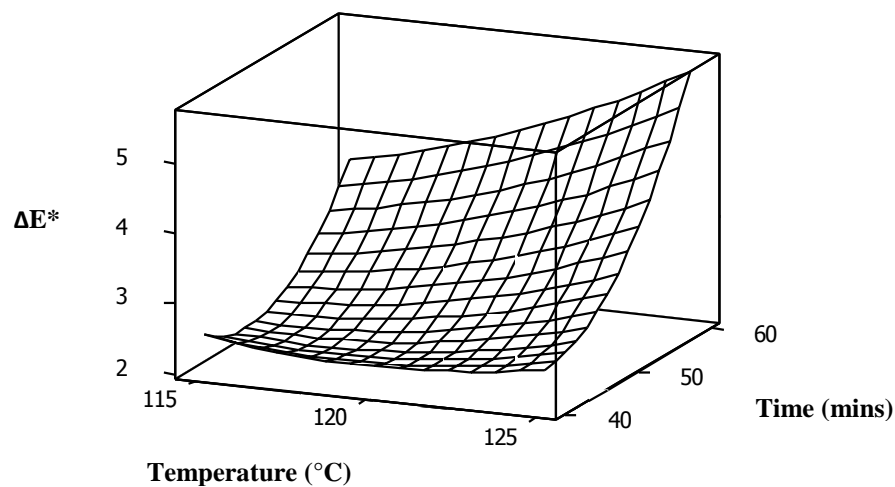
Temperature (°C)	Time (mins)	$\Delta E^*$
115	40	2.05
115	50	2.84
115	60	3.78
120	40	2.61
120	50	2.98
120	60	3.83
125	40	2.72
125	50	2.49
125	60	6.19

$\Delta E^*$  - Total colour difference,

Control – Peanut soup base that was not retorted/sterilized

Table 19 shows that the least colour difference of 2.05 was obtained at 115°C for 40mins whilst the highest colour difference (6.19) was recorded at thermal process of 125°C for 60mins. The effect of the colour change increases with the severity of the thermal processes.

Figure 15 displays the response surface plot depicting the effect of thermal process on the  $\Delta E^*$ . Generally, increases in temperatures at shorter times exhibit marginal increase in  $\Delta E^*$ .  $\Delta E^*$  values however increased steadily with time at both high and low temperatures. The processing time probably had higher effect on  $\Delta E^*$  values than processing temperature. Hence less product colour changes can be achieved at high temperature short time processes. These observations buttress the fact that optimum quality of heat labile food constituents is maintained when high temperature short time (HTST) operations are used (Holdsworth, 1985; Awuah *et al.*, 2007).



**Fig. 15: Effect of time-temperature combinations on the total colour difference ( $\Delta E^*$ ) of retorted canned peanut soup base**

Table 20 depicts that the fitted regression models could only moderately explain the observed effects of time-temperature processes on the  $\Delta E^*$  since the R-square and adjusted R-square values of 82% and 54.5% were not very high. The linear terms (temperature and time) were negative and did not have any significant effect ( $p > 0.05$ ) on  $\Delta E^*$ . The quadratic (AA and BB) and interaction (AB) terms of the fitted regression model though positive had no significant effect ( $p > 0.05$ ) on  $\Delta E^*$ .

**Table 20: Model coefficients for the total colour difference ( $\Delta E^*$ ) of retorted canned peanut soup base**

Model Components	Coefficients for $\Delta E^*$	P-Values
Constant term	175.643	0.024
A: Temperature	-2.312	0.275
B: Time	-1.697	0.052
AA	0.008	0.752
BB	0.008	0.289
AB	0.009	0.374
R <sup>2</sup>	82%	-
R <sup>2</sup> -adjusted	54.5%	-

Model components with  $p < 0.05$  have significant effects on  $\Delta E^*$

The colour of processed foods is an essential determinant of their acceptability by consumers. The vegetables which were used to make peanut soup base contain naturally occurring pigments such as chlorophyll and anthocyanins which could have been heat degraded to pyropheophytin and other brown pigments (Awuah *et al.*, 2007). Maillard reactions and/or burning of the product in the can during thermal processing could also have contributed to the increasing total colour differences with the severity of the thermal processing. The colour difference could be an important quality indicator in the optimizing processes since excessive browning of the product may not be desirable.

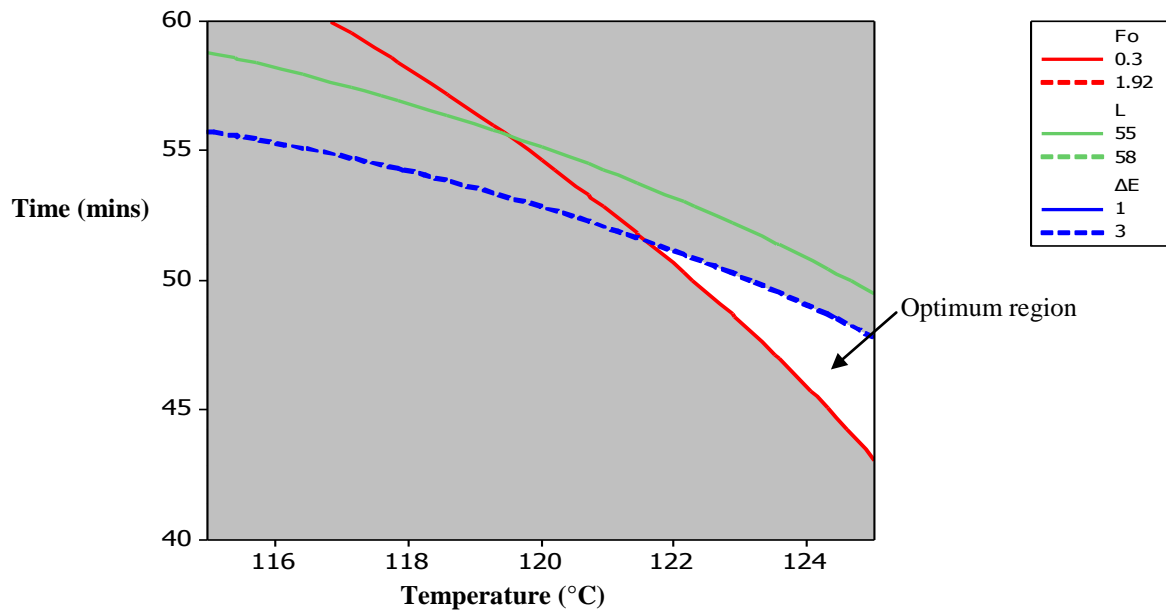
#### 4.4.5 Determination of the limits of the thermal process for canned peanut soup base

The thermal process for canned peanut soup was done by overlaying contours of three selected responses: the sterilizing value ( $F_o$ ), the total colour difference ( $\Delta E^*$ ) and the lightness ( $L^*$ ) of the peanut soup base. Based on the microbiological quality and the maximum temperatures of the product at the slowest heating point from the heat penetration data, a minimum  $F_o$  was fixed at 0.3mins while the maximum was set at 1.92mins. The lightness values were fixed at a minimum of 55 and a maximum of 58 while the total colour differences ( $\Delta E^*$ ) were fixed at a minimum of 1 and a maximum of 3. These colour indicators were selected to prevent excessive changes in product colour during retorting and to achieve a shelf stable product. The optimum thermal process obtained from the optimum region was determined to be from a temperature of 122°C to 124°C for 51 to 44mins. respectively (Fig. 16).

The plot revealed that the optimum thermal process was defined by the  $F_o$  and  $\Delta E^*$ . Thermal processing characteristics that affect  $F_o$  and  $\Delta E^*$  could be investigated to augment the thermal process design. Since the  $\Delta E^*$  could be affected by the charring/burning of the product on the can lids, stopping the lid darkening effect would pave the way for optimization of the thermal process based on the nutritional quality of the product.

Although the optimum thermal process could give a shelf stable product, its ability to prevent the survival of *Clostridium botulinum* spores cannot be guaranteed since  $F_o$  was less than 3mins. The addition of other ingredients to formulate peanut soup base could make the product susceptible to the growth of *Clostridium botulinum* spores. The adoption of an appropriate hurdle technology may prevent the survival of *Clostridium botulinum* spores even at low  $F_o$ . For instance, the

processing of some low acid canned products such as meat combines thermal processing and nitrite (anti-Clostridium spore) additives to ensure shelf-stability and safety (Heinz and Hautzinger, 2007).



**Fig. 16: Optimum region of time-temperature combination for a shelf-stable canned**

#### 4.4.6 Visual examination of canned peanut soup base processed

The addition of 3.75% (30g oil and 770g peanut soup base) of peanut oil to the peanut soup base during filling was expected to serve as covering oil to prevent a darkening of the product observed in the region of the headspace and on the can lid. Since oil is less dense it was expected to cover the surface of the product and prevent the product from adhering and drying up on the lid. Visual inspection of the 9 different thermal processes indicated that all the cans except the cans retorted at 115°C for 40 and 60mins. darkened on the lids (Plates 2 and 3). The results did not clearly suggest

that lower processing temperatures could prevent lid darkening since at 115°C for 50mins, lid darkening was observed.

Although the effect of higher percentage of covering oils could be investigated, the amount of the added oil may significantly affect the sensory attributes and acceptability of the product. The darkening of the product on lids gives the product an unattractive appearance when opened. Other methods such as thermal processing in agitating retorts could be investigated to assess their effect on lid darkening.

Also visual inspection of the canned peanut soup base products over the seven day storage period did not show any signs of bloating or spoilage for all the retorted products. The stability of the products could largely be attributed to high initial product temperatures ( $64\pm 1^\circ\text{C}$ ) at the start of retorting for each thermal process. The high initial temperatures ensured that pasteurization temperatures were reached much faster to destroy some heat labile enzymes and mesophilic microorganisms that cause food spoilage.



**Plate 2: Canned peanut soup base with 3.75% covering oil showing lid darkening effect after retorting at 115°C for 50mins**



**Plate 3: Canned peanut soup base with 3.75% covering oil without lid darkening effect after retorting at 115°C for 40mins**

#### **4.4.7 Microbial quality of canned peanut soup base processed at different time-temperature combinations**

Aerobic Plate Count (APC) and Yeasts and Moulds Count (YMC) were determined after seven (7) days of storage at room temperature (28-32°C) for each of the 9 thermally processed products.

Table 21 displays the time-temperature combinations used for retorting of the canned peanut soup base products and the resultant enumeration results.

**Table 21: Plate counts of canned peanut soup base processed at different time and temperature combinations**

Temperature	Time	APC (cfu/g)	Total Yeasts and Moulds Count (Cfu/g)
Control	-	15,000,000	12,000,000
115	40	10,000	10,000
115	50	200*	<100
115	60	450*	<100
120	40	8100	1200
120	50	100*	<100
120	60	<100	<100
125	40	250*	<100
125	50	<100	<100
125	60	<100	<100

American Public Health Association (APHC) rules were adopted in the calculation of plate counts

APC - Aerobic Plates Counts were mesophilic counts of plates incubated at 30°C

\* Estimated Aerobic Plate Counts (EAPC) for plates with less than 25 colonies at dilution of  $10^{-2}$

<100 refers to plates that had no observable growth at dilution of  $10^{-2}$

Control – Peanut soup base that was not retorted/sterilized

#### 4.4.7.1 Aerobic Plate Count (APC)

The APC (mesophilic count) of food products is a useful indicator of the level of microorganisms in foods (Maturin and Peeler, 2001). The unretorted peanut soup base had extremely high APC ( $15 \times 10^6$  Cfu/g) compared to the retorted products (Table 21). Generally APC were higher at shorter processing times and lower temperatures. The high APC of 8100 – 10,000cfu/g were recorded at

lower temperatures (115°C and 120°C ) short time processes (40mins). Products processed at time-temperature combinations of 115°C for 50mins and 60mins, 120°C for 50mins and 125°C for 40mins all showed growth lesser than the countable range 25-250cfu/g (APHA, 2001) at dilutions ( $10^{-2}$ ml). The higher temperature and longer time processes (120°C for 60 minutes and 125°C for 50 and 60mins.) showed no observable growths.

It is worth noting that due to unavoidable delays between filling and retorting of most of the products, some products were reheated to maintain the initial product temperature around  $64 \pm 1^\circ\text{C}$ . It is possible that counts higher than these could have been achieved for some of the treatments. This is because Prescott *et al.* (2002) observed that vegetative cells of mesophiles can be destroyed at temperatures between 60 - 70°C. None the less, the destructive effect of thermal processes on microorganisms which depicts a first order logarithmic reactions (Awuah *et al.*, 2007) largely accounted for the observed reduction in microbial loads at higher thermal processing combinations.

Although plate counts do not necessarily indicate the presence of a hazard, they give indications about the severity of thermal process, the microbial quality of the food and the shelf-life of the food products (IFST, 1997). The APC's for all the thermally processed peanut soup base were within the recommended Ghana Standards of  $10^3 - 10^4$ cfu/g (GS 955, 2009) that is required to produce a shelf stable canned product.

#### 4.4.7.2 Total yeasts and moulds count

Yeast and moulds are a diverse group of fungi that can grow in foods within a wider range of pH from 2 - 9 especially the moulds (APHA, 2001). Table 21 indicates that the total yeasts and moulds count was  $12 \times 10^6$ cfu/g for the unretorted product which was much higher than the retorted product. Higher yeast and mould counts of 10,000 cfu/g and 1200cfu/g were recorded for lower temperature shorter time processes of 115°C for 40mins. and 120°C for 40mins. respectively. The other thermal process treatments at 115-125°C for 50 - 60mins and 125°C for 40mins all showed no observable growth (>100cfu/g).

The high yeast and mould counts for the unretorted product probably explain why peanut soups ferment easily. The yeasts and moulds that cause spoilage of processed foods are the aerobes (APHA, 2001). According to Prescott (2002), most vegetative yeasts and mould cells and their spores can be destroyed at temperatures between 50 - 80°C. The survival of yeast and moulds in most processed products is usually attributed to underprocessing or post processing recontamination (Prescott, 2002). The presence of relatively high amounts of yeast and moulds in the peanut soup base processed at 115 and 120°C for 40mins could be attributed to the slow heat penetration due to the viscous nature of the products and high initial yeast and moulds count of the product before retorting. The high counts at 115 and 120°C for 40mins could probably lead to spoilage of the products with extended storage.

## CHAPTER FIVE

### 5.0 CONCLUSIONS AND RECOMMENDATIONS

#### 5.1 Conclusions

- i. The physical (seed dimensions and bulk density) and chemical characteristics (proximate composition) of different peanut varieties differ significantly. The high average crude fat (at least 50%), protein (23 - 26%) and carbohydrate (19 – 24%) contents of peanuts make them an essential source of dietary nutrients in food applications. Chinese peanut variety was the most suitable for making peanut soup base because of its predominance in the major markets of Accra compared to the other varieties. Also it was the most variable in seed dimensions (i.e. Standard deviations of seed width was  $\pm 0.92$ cm and seed length  $\pm 2.27$ cm) which preferably should be milled to peanut paste where kernel size are not very important.
- ii. The manual colour sorting is an effective method of minimizing aflatoxins levels in peanuts and peanut products to meet acceptable aflatoxin limits.
- iii. A canned peanut soup base acceptable to consumers can be formulated by blending 300g peanut paste, 146.9g tomato paste, 10g ginger, 12.3g pepper, 15g salt and 750ml of water.
- iv. Thermally processing canned peanut soup base at 122-124°C for 51-45 minutes respectively could give a relatively shelf stable canned peanut soup base of acceptable

colour and microbial quality. However, the severity of the thermal process may not ensure safety from botulism ( $F_o < 3\text{mins}$ ).

## 5.2 Recommendations

- i. The effect of initial product (fill) temperature and viscosity on the sterilizing value ( $F_o$ ) should be investigated.
- ii. The effect of thermal processing of the product on lid darkening and sterilizing value ( $F_o$ ) using an agitating retort should also be studied.
- iii. Shelf-stability studies to access shelf-life of the canned peanut soup base product should also be conducted.

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## 7.0 APPENDICES

### 7.1 BALLOT SHEET FOR EVALUATION OF PEANUT SOUP

DEPARTMENT OF NUTRITION AND FOOD SCIENCE

UNIVERSITY OF GHANA

#### BALLOT SHEET FOR EVALUATION OF PEANUT SOUP

PANELLIST NUMBER: \_\_\_\_\_ DATE: \_\_\_\_\_

#### **INSTRUCTIONS: Read carefully before you begin**

You have been provided with 3 coded samples of PEANUT (GROUNDNUT) SOUP. Please write the codes of the samples in the boxes provided in the order in which you have been presented and evaluate them from left to right.

Where tasting is involved, please rinse your mouth with the water provided after tasting a sample and **wait for about 15s before evaluating the next sample.** *You can re-taste the samples.*

ACCEPTABILITY: Please rank the intensity of your **liking** of each sensory attribute of each sample using the given scale:

Scale:

<b>1=dislike extremely</b>	<b>6=like slightly</b>
<b>2= dislike very much</b>	<b>7= like moderately</b>
<b>3= dislike moderately</b>	<b>8= like very much</b>
<b>4= dislike slightly</b>	<b>9=like extremely</b>
<b>5= neither like nor dislike</b>	

#### 1. COLOUR

Observe the colour of the groundnut soup you have been provided with.

Indicate your **LIKING** for *colour* of the soup by writing the appropriate rank score from the 9-point hedonic scale in the space below the code.

Sample code	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>
Rank	.....	.....	.....	.....

2. **CONSISTENCY (THICKNESS):** Using the spoon provided scoop a spoonful of the soup and observe the consistency as you gradually pour it back. Indicate your **LIKING** for *consistency* of the soup by writing the appropriate rank score from the 9-point hedonic scale in the space below the code.

Sample code	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>
Rank	.....	.....	.....	.....

3. **TASTE: Take a spoonful of soup and taste** Indicate your **LIKING** for *taste* of the soup by writing the appropriate rank score from the 9-point hedonic scale in the space below the code.

Sample code	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>
Rank	.....	.....	.....	.....

4. **SPICINESS: Take a spoonful of soup and taste for how spicy it is.** Indicate your **LIKING** for *spiciness* of the groundnut soup by writing the appropriate rank score from the 9-point hedonic scale in the space below the code.

Sample code	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>
Rank	.....	.....	.....	.....

5. **SMOOTHNESS: Take a spoonful of soup and comment on how it feels in your mouth** Indicate your **LIKING** for *smoothness* of the soup by writing the appropriate rank score from the 9-point hedonic scale in the space below the code.

Sample code	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>
Rank	.....	.....	.....	.....

#### 6. OVERALL ACCEPTABILITY

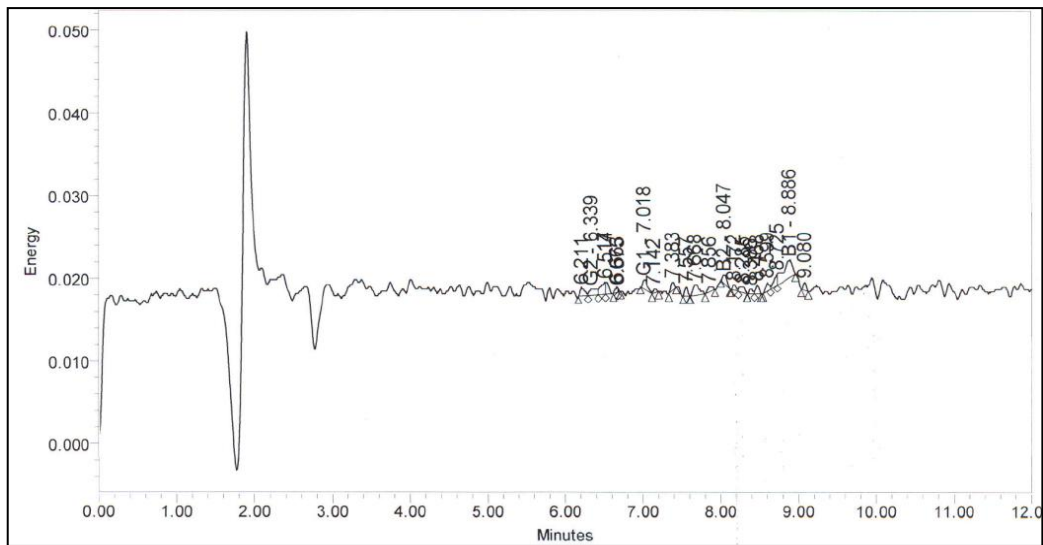
Overall, how much do you **LIKE** each of the groundnut soup samples you have been provided with?

Sample code	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>
Rank	.....	.....	.....	.....

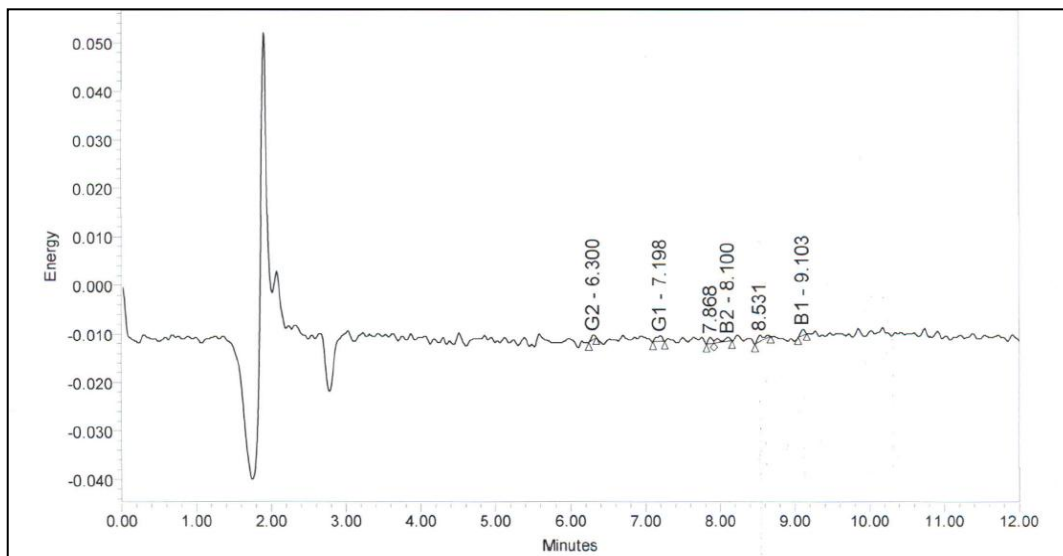
*COMMENTS:*

.....

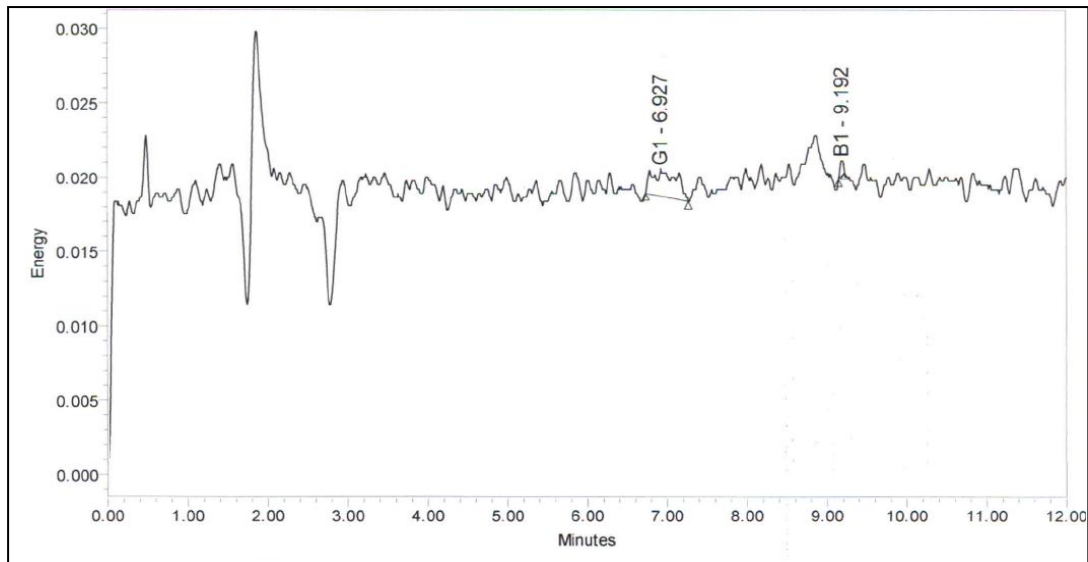
## 7.2 CHROMATOGRAMS FOR AFLATOXIN LEVELS OF SORTED PEANUT COMPONENTS



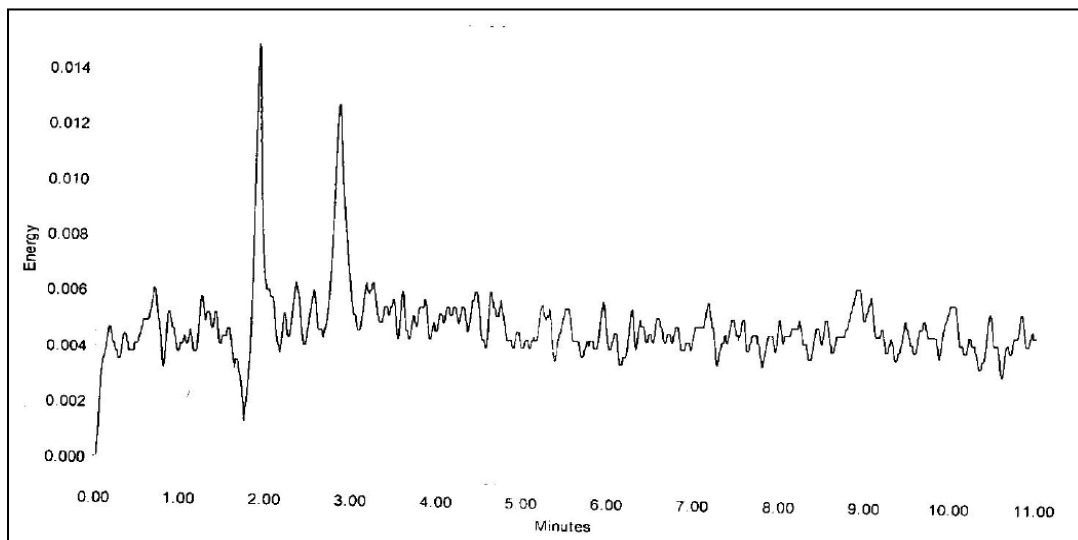
### 7.2.1. Aflatoxin levels of Less than 50% discoloured peanuts sorted using manual colour sorting method



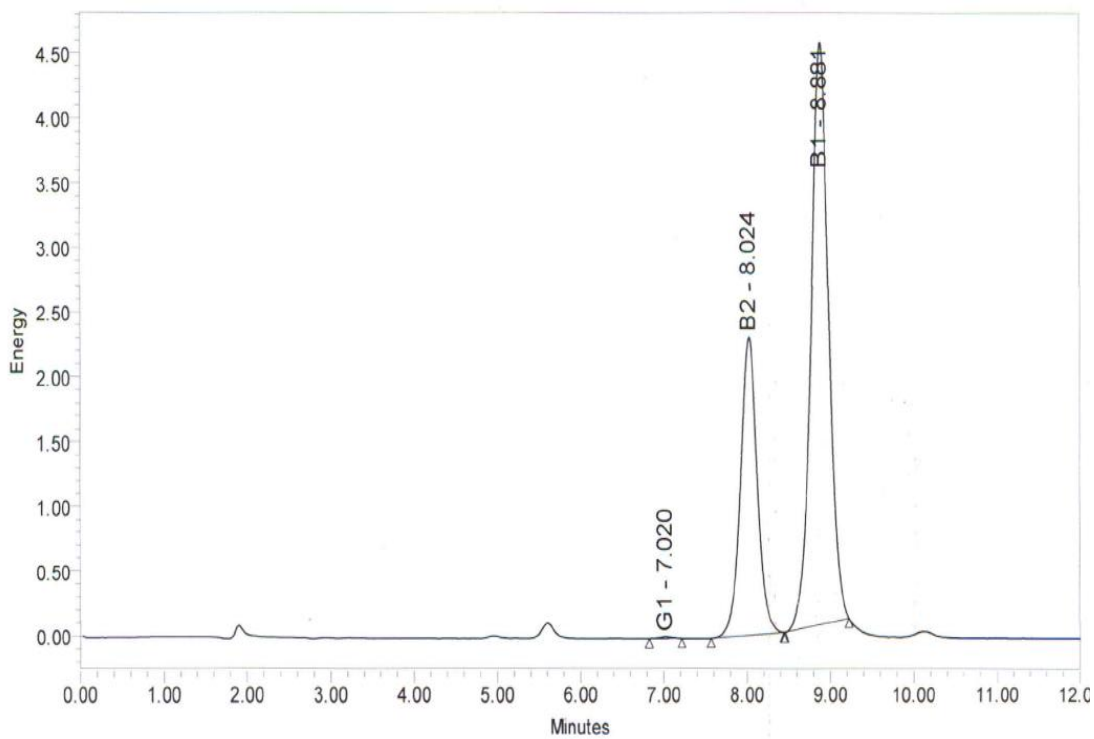
### 7.2.2 Aflatoxin levels of Chinese peanut skins/hulls



**7.2.3 Aflatoxin levels of clean (no discolouration) Chinese peanuts sorted using manual colour sorting method**

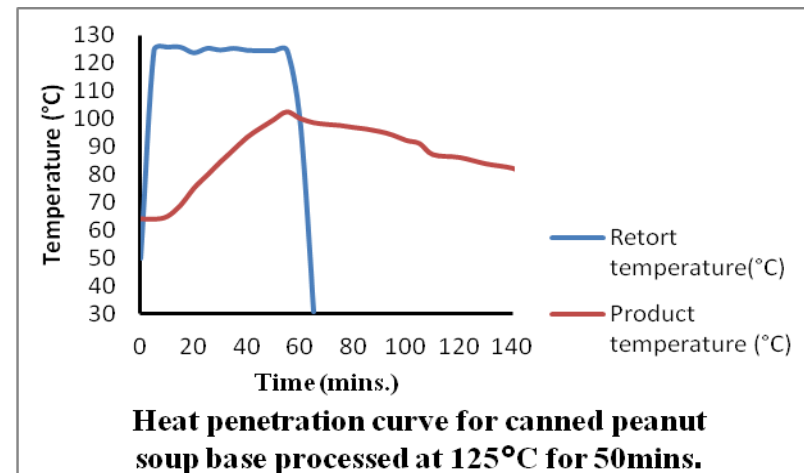
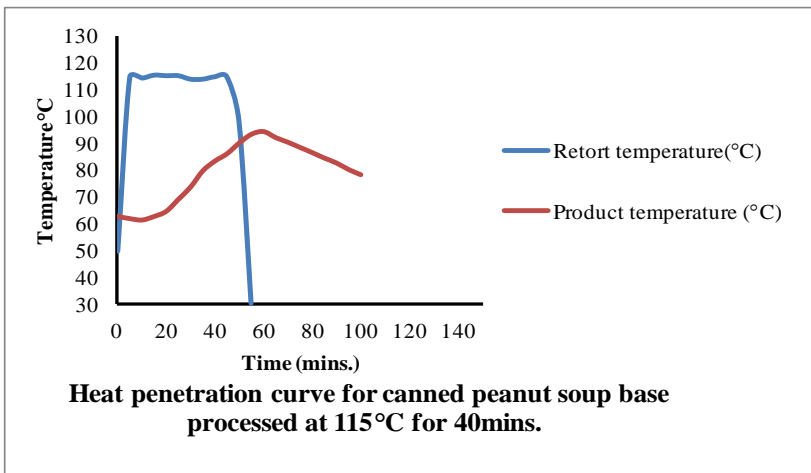
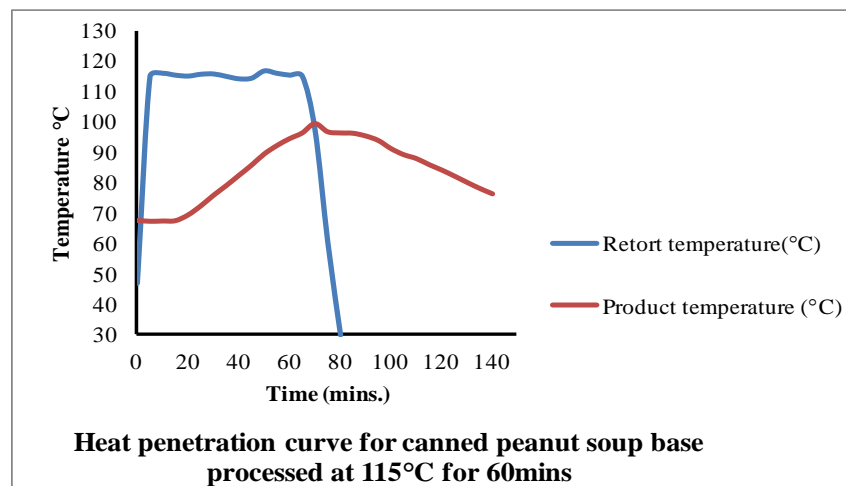
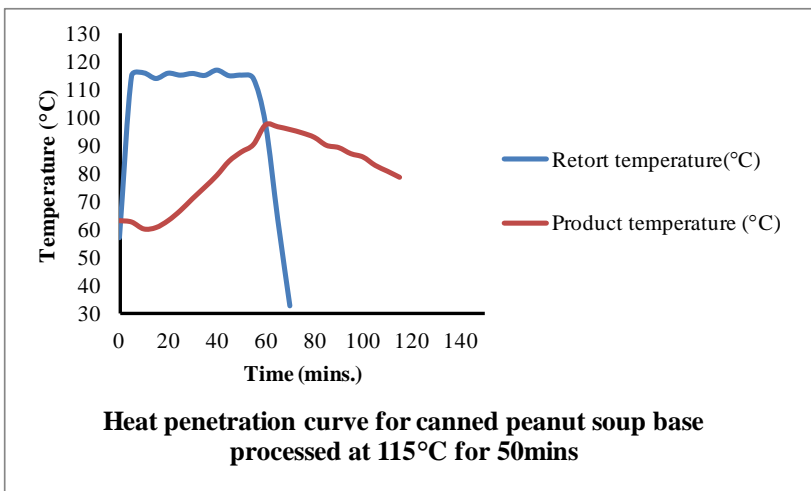


**7.2.4 Aflatoxins levels of more than 50% discoloured Chinese peanuts sorted using the manual colour sorting method**

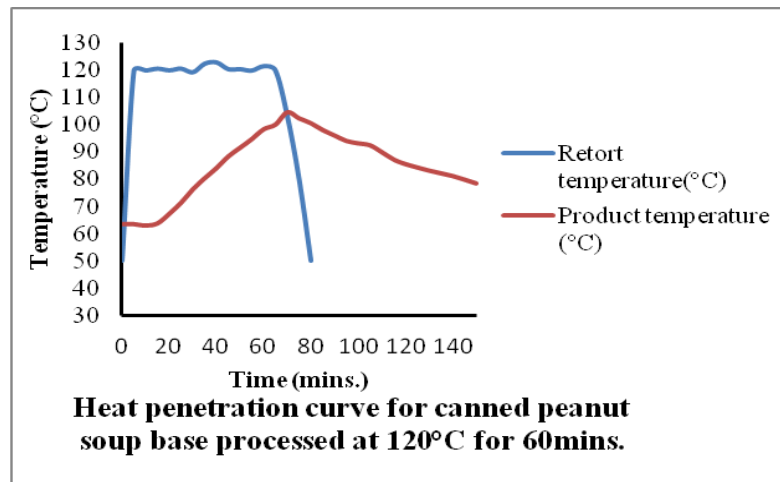
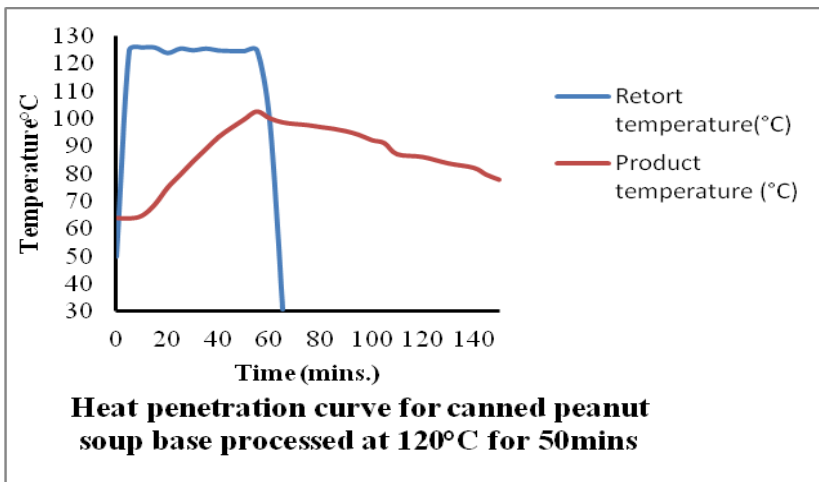
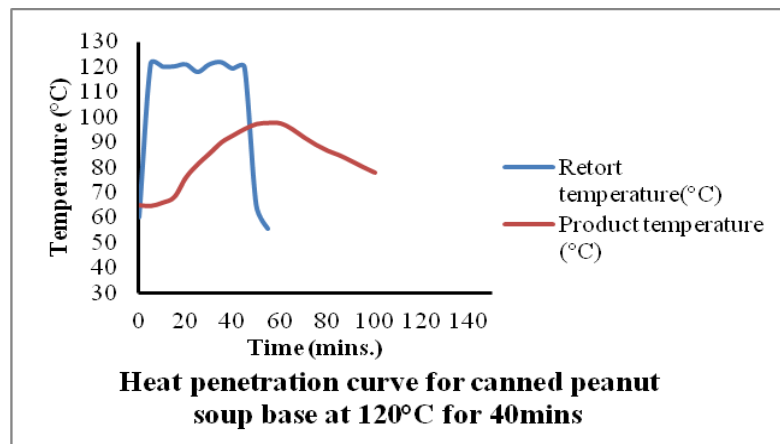
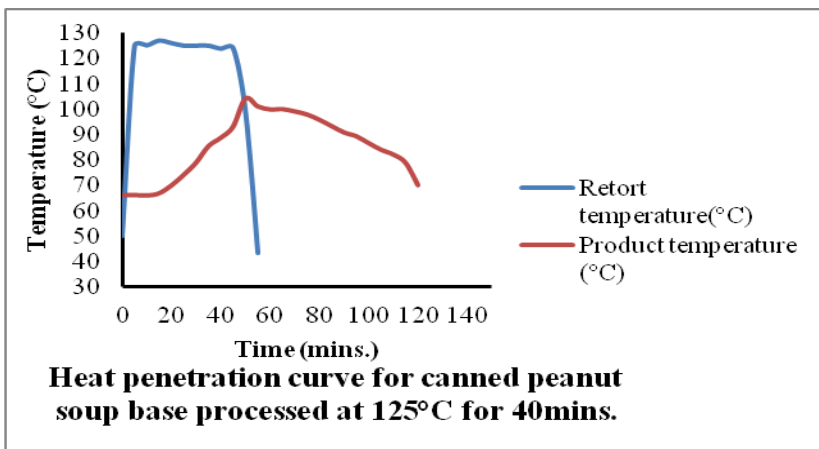


**7.2.5 Peaks depicting aflatoxin levels in sorted shriveled and damaged Chinese peanuts**

**APPENDIX 7.5 HEAT PENETRATION CURVES THERMAL PROCESSING OF CANNED PEANUT SOUP BASE**



**7.3.1 Heat penetration curves of canned peanut soup base processed at 115°C for 40, 50 and 60mins and 125°C for 50mins.**



**7.3.2 Heat penetration curves of canned peanut soup base processed at 120°C for 40, 50 and 60mins and 125°C for 40mins**