ROASTING TIME-TEMPERATURE OPTIMIZATION FOR PREPARATION OF PEANUT BUTTER AND STUDY ON ITS SHELF LIFE

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Roasting Time-Temperature Optimization for Preparation of Peanut Butter and Study on its Shelf Life

A dissertation submitted to Department of Food Technology, Central campus of Technology, Tribhuvan University, in partial fulfillment of the requirements for the Degree of B. Tech. in Food Technology

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Approval Letter

This *dissertation* entitled *Roasting time-temperature optimization for preparation of peanut butter and study on its shelf life* presented by Anish Shrestha has been accepted as the partial fulfillment of the requirements for the B.Tech.Degree in Food Technology

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Abstract

The objective of this study was to develop peanut butter from roasted and grounded peanut. Raw peanut of variety *'Jayanti'* (Type-Spanish bunch) was collected from Sunsari district, Dharan and were roasted at three different temperatures (130°C, 140°C and 150°C) for three different time periods (10, 15 and 20 min) to optimize the process of butter making with 9 different samples. The raw peanut was sorted prior to roasting and grinding. The roasted-ground peanut paste was obtained in which ingredients namely lecithin (1%), salt (1%), sugar (1%) and oil (3%) were added. Oil was added to give final fat content of the butter not more than 55 % by weight. The peanut butter was then subjected for sensory analysis, proximate analysis and shelf life evaluation.

Based on the sensory evaluation, significantly best ($p<0.05$) peanut butter can be produced from preparing peanut paste by roasting peanut at 150°C for 10 min. The proximate analysis of peanut butter showed that it contained moisture content, fat content, total carbohydrate, crude fiber, crude protein, ash content and energy value of 5.8±0.04%, 52.70±0.45%, 19.28±0.05%, 5.47±0.5%, 38.67±0.07%, 5.67±0.80% and 641.5±3.57 kcal respectively. The shelf life of the peanut butter stored at ambient temperature (room temperature) was found to be 11 days, but the butter stored at refrigerated temperature was not spoiled by rancidity throughout the period of storage (55 days).

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List of Abbreviations

Abbreviations	Full Form
ANOVA	Analysis of Variance
AA	Amino acids
OA	Overall Acceptance
CCT	Central Campus of Technology
NARC	Nepal Agriculture Research Council
AOAC	Association of Official Analytical Chemists
EDTA	Ethylene Di-amine Tetra –Acetic acid
FDA	Food and Drug Administration
FSA	Food Standard Agency
USDA	United States Department of Agriculture
HVO	Hydrogenated Vegetable oil
LDL	Low Density Lipoprotein
MRP	Millard Reaction Products
PV	Peroxide value
AV	Acid Value
LSD	Least Significant Difference

Part I

Introduction

1.1 Background

Butter is perhaps the traditional spread developed since the inception of ancient food technology and its production technology has since not changed much. It is obtained by churning cream to a product consisting of unaltered fat globules and moisture droplets embedded in a continuous phase of butter fat. From the studies of Pearson (2007), butter contains butter fat, water and curd (consisting of casein, lactose and mineral matter). Another variety of spread is cheese which is mainly made from curd produced from the coagulation of souring milk. Souring is done by rennin, an enzyme obtained from the inner lining fourth stomach of the calf (Man, 2002).

General definition of spread include, but not limited to, spreads made from edible vegetable oil or animal fat or a combination of both such as margarine, cheese and butter and those obtained from fruits and vegetables such as jams, preserves and marmalades. Many researchers discuss the nutritional value of margarine and other spreads largely around two components. These are the total amount of fat and the types of fat (saturated fat, trans-fat) as components of the formulation. It has been concluded by some researchers that the saturated fatty acids in triglycerides contribute to elevated blood cholesterol levels, which on turn has often been linked to cardiovascular diseases (Keys *et al.*, 1965; Mensik *et al.*, 2003).

Trans-fats which do not occur naturally in vegetable fats are a consequence of partial hydrogenation of oils, requirement for some spread formulation procedure (Hayakawa *et al.*, 2000; Hu *et al.*, 1997; Willett *et al.*, 1993). According to Floter and Duijn (2006), many industries have gradually moved away from using partially hydrogenated oils since the mid 90s and now produce new spreads that contain less or no trans fats. Though it has been proven that the intake of cholesterol has less effect on high blood cholesterol level than saturated fat, the FDA of the United States of America has warned that healthy people should not consume more than 200 mg per day. However butter which contains high level of cholesterols which is rapidly consumed several times daily in the form of varied spreads. Though margarine may contain no cholesterol due to their preparation procedures, the

abundant saturated fat in margarine induces the bad type of cholesterol (LDL) in the course of human metabolism (Sebedio and Christie, 1998).

There are various types of spreads ranging from cheese, butter, margarine to fruit spreads (Keys *et al.*, 1965). Due to problems associated with consumption of cheeses and margarines, alternatives which can deliver the functionalities required in traditional spreads with less nutritional problems are being sought. In 1895 Dr. John Harvery Kellogg patented a process for creating peanut butter from raw peanuts as a protein substitute for patients without teeth (Anon, 2012). Peanuts (*Arachis hypogea* L.*)* are significant sources of proteins and fat, which contribute to solving world food shortages (Abdullah *et al.*, 1993). Approximately half of the total food use of peanuts is attributed to the production and consumption of peanut butter (Resurreccion, 1988). Other products include peanut beverages (Galvez *et al.*, 1990; Rubico *et al.*, 1987), peanut flour (Holt *et al.*, 1992), coffee whitener (Abdullah *et al.*, 1993), imitation cheese spreads (Santos *et al.*, 1989) and peanut paste (Muego-Gnanasekharan and Resurreccion, 1993).

Peanut roasting and the development of color and roasted flavor in peanuts has been a topic of research for some time. Typically, roast color is the most important quality control parameter in commercial processes. Peanuts, when heated, develop a unique, desirable, roasted peanut flavor. Roasting time and temperature play a role in the formation of peanut flavor and influence intensity of flavor and odor in roasted peanuts (Buckholtz *et al.*, 1980).

Peanut butter is a water-in-oil emulsion (Aryana *et al.*, 2000). The oil separates during storage, and the product needs to be remixed for better eating quality. This separation is a problem due to higher tendency of the product to become rancid. The stabilizer keeps the oil from separating from the peanut butter and improves texture, increases shelf life, and keeps the peanut butter fresh which most consumers prefer (Malupangue, 2005). Peanut butter is a semi-perishable product that is subject to a number of microbial, chemical and physical deteriorative changes, which affect the final quality of the finished product. The shelf life is greatly dependent on the quality of peanuts used and the conditions of the peanuts used for making the peanut butter. Deterioration of peanut butter arises from putrefaction of protein fraction caused by bacterial metabolism; darkening, which results from an interaction between sugar and protein in the product and; oxidative rancidity that develops in the unsaturated portion of oil when it is exposed to air (Woodroof, 1983a)

1.2 Statement of the problem

Peanut or groundnut (*Arachis hypogeal* L.), a member of the legume family, is an important food and oil crop. It is currently grown on approximately 42 million acres worldwide. It is the third major oilseed of the world after soybean and cotton (FAO, 2004). Within the last two decades, peanut has been grown extensively by farmers and promoted in parts of Nepal as cash crop by NARC when its demand as an oilseed increased gradually. Nepalese farmers are producing 1500 to 2500 kg of peanuts per ha of farm land brought under peanut cultivation. Rainfall, diseases and insects are the main factors that determine the productivity of peanuts in any given year. However, peanut roasting is carried out which have increased the shelf life compared to that of raw peanut and the roasted peanut are consumed as snack food which is becoming popular among youths in urban areas as well as among school children. Peanut can be processed into spreads and peanut butter but no any information is provided regarding the processing of such products in a standard method in Nepal. Peanut butter is extensively consumed in the western-world as bread spread but is not that familiar in Nepali market. Peanuts processing units in Kathmandu were found to be responsive to the taste of the consumers and are therefore well doing. But the peanut butter factories that are newly established in the Kathmandu valley are finding that they have to learn more in order to compete in the market. Therefore, study on shelf life of peanut butter is must for its valuable nutrition (Dhakal *et al.*, 2002). Roasting time-temperature optimization in preparation of peanut butter leads the spread to be of optimum quality in terms of color, flavor and energy.

1.3 Objectives of the study

1.3.1 General objectives

- To optimize roasting time-temperature of peanut for the preparation of peanut butter and evaluate its quality.

1.3.2 Specific objectives

- To evaluate the proximate composition of raw peanut and peanut butter.
- To study storage life of the peanut butter stored in under ambient and refrigerated temperature conditions on basis of PV values.

1.4 Significance of the study

Peanut is a nutritious legume grown abundant in Terai and Hill areas of Eastern Nepal and consumed directly by removing the shell of the groundnut. They are usually added as supplement to other foods such as snacks and may be used as spreads. However, not much has been done to process and store the legume and its processed products. Peanut spread has short shelf-life due to high fat content but has tremendous energy value. Short rancidity period affects sensory properties of peanut spread due to quick oxidation of fats, mainly unsaturated fatty acids. So the study of the shelf-life of the peanut product under this study helps to compare shelf-life in two different temperature conditions. Optimization of process to prepare peanut spread helps in variation of color and flavor development.

Thus, with change in shelf-life under different storage conditions of peanut butter, the freshness of peanut processed products as peanut butter can be determined. Consequently in this study, spread made from peanut has the potential of presenting the consumer an additional variety in which the legume can be consumed as well as this study helps in optimization of color and flavor characteristics roasted peanut with rich nutritional value and functional properties.

1.5 Limitations of the study

- Study of shelf-life of optimized product under different packaging materials could not be studied.

Part II

Literature review

2.1 History of peanut cultivation

Peanut cultivation is believed to have originated in Bolivia and surrounding countries in South America. Any warm, temperate region of the world has the capability of growing healthy, edible seeds. North and South American natives grew peanuts for some time before its written history. The seeds and techniques used by natives were taken to Europe during early colonization of the United States. During the 16th century, peanut growth and cultivation techniques quickly spread throughout Africa, Asia, and the Pacific Islands (Hammons, 1982).

There is no information or record which could enable us to make a definite statement as to when and where peanut was first introduced in Nepal. Similarly, how or through what medium peanut reached Nepal from its origin in America is not known. Elderly people in the villages told that peanut arrived in Nepal from India-with Nepali men who were travelling between India and Nepal for employment and the Indian nationals who came to work in Nepal's terai villages (Dhakal *et al.*, 2002).

Peanut reached East Asia from South America and from there it came to India-entering the country through the east coast of Madras along with the Spaniards (Sinha and Reddy, 1988). The fact that peanut came to East Asia with the Spanish people would allow to tentatively conclude that peanut must have been certainly entered Nepal through our terai in the hands of migrant farmers. Elderly Nepali villagers note that this crop can be grown in marginal lands also and with much less inputs than what is required for many other crops and that must have been one of the reasons for the initial adoption (Dhakal *et al.*, 2002).

2.2 Varieties of groundnut grown in Nepal

NARC has been a source of seed for the high yield varieties of peanut and they supply the seeds to any farmer who is eager to adopt their 'improved seeds'. According to NARC, some varieties of peanut found in Nepal are listed in the Table 2.1

Table 2.1 Different varieties of peanut found in Nepal

S.N.	Name of released variety	Year of release	Origin	Yield potential Mt/ha	Maturity (Days)	Recommendation Domain
1.	Baidehi	2005		3.3	115	Terai, Inner terai
2.	Rajharsi	2005		2.8	115	Terai, Inner terai
3.	Jyanti	1996		2.2	115	Terai, Inner terai & Mid hills
4.	Jyoti	1996		2.0	137-153	Terai, Inner terai & Mid hills
5.	Janak	1989	India	2.5	145	Terai, Inner terai & Mid hills
6.	B-4	1990	India	1.5	140	Terai, Inner terai & Mid hills

Source: NARC (2014)

2.3 Peanut growth

Peanut seed germination occurs when the soil temperature reaches 15.55ºC and the soil moisture is adequate for the seed to absorb 50% of its weight in water. With adequate moisture, a radical sprouts from the germinating seed a few days after planting. Food reserves are maintained in the cotyledon until the shoots emerge from the soil and begin to accumulate sunlight via photosynthesis. Peanuts are considered to be self-pollinating, with natural cross-pollination rates of less than 1% (Ketring *et al.*, 1982). The fruit of the peanut is a pod with 1 to 5 seeds that develop underground after the elongated pod with ovarian structure penetrates the soil 3-6 centimeters. Peanut plants produce bright-yellow complete flowers with male and female parts located in the axils of the leaves. Flowers normally appear 4-6 weeks after planting and plants continue to flower through the growing season. Depending on the variety, peanuts require anywhere from 100 to 150 days from planting to reach full maturity (Ketring *et al.*, 1982; Sanders *et al.*, 1982).

2.4 Harvesting, drying and storage of peanut

Ideally, peanuts are harvested when the majority of the pods elicit a veined surface, have a colored seed coat, and three-quarters of the seed show darkening on the inner surface of the hull. Mechanical digging, shaking, and windrowing follow in the harvesting process. Digging detaches the plant from the soil, shaking removes the excess soil, and windrowing inverts the plant, orienting its pods face upward allowing for curing. Drying is one of the most important aspects of peanut quality. If the seeds are not dried to safe moisture content (6-10%), quality deteriorates quickly and the probability of microbial invasion is increased. Artificial drying of the seed should be done relatively soon after harvesting. This prevents mold and aflatoxin formation, and the formation of off-flavors caused from fungal lipase action and oxidative rancidity by decreasing water content of the seed. Drying temperature should not exceed 16.66°C with a moisture reduction rate of 0.5% per hour (Sanders *et al.*, 1982).

Dried peanut seeds are normally stored as unshelled nuts at a relative humidity between the ranges of 60 to 70%. The seeds should be stored in such a way that pest and rodent invasion is not a problem. High temperatures during storage should also be avoided (Baker, 2002).

2.5 Nutrition of peanut

From a nutritional standpoint, peanuts contain many of the essential vitamins and minerals necessary for proper health. Peanuts also contain roughly 50% fat, the majority being unsaturated. In comparison to other nuts, such as pecans and walnuts, peanuts contain less total fat (Maga, 1991). Peanuts containing high (>70%) oleic acid (18:1), a monounsaturated fatty acid, may also be useful in dietary regimes designed to reduce blood cholesterol levels in postmenopausal women, without resulting in problems associated with oxidation of low density lipoproteins (O'Byrne *et al.*, 1997). Comparison of saturated fat in other foodstuffs is found in the Table 2.2

Table 2.2 Fat comparison of peanuts to other food products

Food (serving size)	Saturated Fat	Total Fat
Peanuts (28 g)	2 g	14 g
Peanut Butter (30 g)	2.5 g	14 g
Potato Chips (28 g)	3 g	10 g
Egg Salad (84 g)	4 g	19 g
American Cheese (28 g)	2.6 g	9 g
Hamburger (96 g)	7 g	17 g

Source:Baker (2002)

Vitamins and minerals present in peanuts are shown in the Table 2.3

Table 2.3 Vitamins and minerals present in peanuts and their uses in the body

Vitamins and Minerals	% RDI in one ounce serving of dry roasted peanuts	Uses in the Body
Vitamin E	25%	Vital antioxidant, which protects Vitamin A and the body's cells and tissues from damage. It is important for the immune system and may aid in the prevention of tumor growth.
Niacin	19%	Necessary for maintenance of healthy skin, the nervous system, and digestive tract.
Folate	10%	Important for development of new cells in the body, particularly during periods of growth and during pregnancy.
Thiamin (B1)	8%	Needed to ensure normal functioning of the nervous system, appetite, and digestion.

B6	4%	Produces and breaks down proteins in the body and manufactures red blood cells used to transport oxygen in the body.
Riboflavin (B2)	2%	Releases energy from the food we eat, helps skin stay healthy, and assists in the normal functioning of the eye.
Magnesium	12%	Important in the building of bones and teeth, creation of protein, transmission of nerve impulses, and maintenance of body temperature.
Copper	10%	Important for the formation of hemoglobin, health of bones, blood vessels, and nerves.
Phosphorous	10%	Component of all soft tissues that is fundamental to growth, maintenance and repair of bones and teeth.
Potassium	10%	Needed to ensure water balance in the body and creation of protein. It also helps release energy from nutrients and aids in nerve impulse transmission.
Zinc	6%	Aids in the formation of protein, wound healing, blood formation, taste perception, appetite, night vision, and general growth and maintenance of all tissues.
Iron	4%	Aids in the transport and distribution of oxygen in the body's cells.
Calcium	2%	Needed for the development and maintenance of healthy bones and teeth.

Source: Baker (2002)

Peanuts contain high levels of fiber, with naturally low sodium, are cholesterol free, and represent a good source of folic acid. Peanuts also have chemical characteristics that parallel recent discoveries in nutrition that have been found to be beneficial to human

health. Recently, resveratrol has received attention from the research community due to possible health benefits. Resveratrol was found in relatively moderate levels in muscadine grapes several years ago. Peanuts contain moderate levels of resveratrol, betastigmasterol, and behenic acid (Sanders and McMichael, 1999). The by-products of edible peanuts, such as the skin, contain behenic acid, which is used in cosmetics and shampoos (Baker, 2002).

2.6 Peanut storage, stability and oxidation

The term shelf life, in regard to peanuts or peanut oil, can be described as "the number of days before the onset of oxidative rancidity, a process which is generally induced in either the whole peanut or peanut oil by exposure to heat and air" (Mercer *et al.*, 1990). This definition implies that the shelf life of peanuts is only as long as the time it takes before the onset of oxidative rancidity. The keyword here is the term rancidity, where a food becomes rancid or inedible. Rancid can be defined as "having the unpleasant taste or smell of oily substances that have begun to spoil". Peanut oil stability is quite good when compared to other vegetable oils, partly due to the fatty acid composition. Traditionally, runner-type peanuts contain approximately 50% fat or oil, which consists of 41-67% oleic acid (18:1 n-9) and 14-42% linoleic acid (18:2 n-6). Because of the high amount of oil contained in a peanut, the quality can deteriorate quickly due to lipid oxidation, depending on a number of factors, such as the presence of oxygen, light, moisture, and high temperatures. A factor of interest involves the content and structure of the fatty acid constituents in the oil. As the amount of double bonds increase in a fatty acid, it is more susceptible to oxidation. Low levels of linolenic acid (18:3 n-6) in peanut oil are thought to be partially responsible for oxidative stability, where the rates of oxidation are approximately 1:10:100:200 for 18:0, 18:1, 18:2, and 18:3, respectively (O'Keefe *et al.*, 1993).

Lipid oxidation has been found to be a major source of the off flavors and decreased quality in peanuts. Painty and cardboardy off flavors are formed during lipid oxidation, making the product rancid and unacceptable to the consumer. The off flavors associated with painty (pent-2-enal) and cardboardy (2t, 2t-nonadienal) are formed over time from aliphatic aldehydes in lipid systems, along with various other aldehydes and ketones (Grosch, 1982).

The oxidation occurring at double bond systems is thought to occur for a number of reasons. Generally, autoxidation (the reaction with molecular oxygen through a self-

catalytic mechanism) is noted as the primary cause of the quality defects that occur in lipid systems, due to hydro-peroxide formation (Fig.2.1). This leaves the food product less acceptable because of off flavors and off odors created during oxidation (Nawar, 1996).

Autoxidation starts by a term described as initiation. Initiation can be caused by the decomposition of hydro-peroxides, metal catalysts, or light. Singlet oxygen, an excited state form of molecular oxygen, is thought to be the major contributing factor to initiation via free radical formation. This is commonly found in plant tissues containing chlorophyll, which is thought to be a sensitizer of singlet oxygen. Stable molecular oxygen (triplet oxygen) is not thought to attack the double bonds in lipid systems due to the rules of spin conversion, since the carbon, carbon double bonds in carboxylic acids are in singlet states. Once these free radicals are formed in the presence of lipids, attack at the double bond systems is evident (Nawar, 1996).

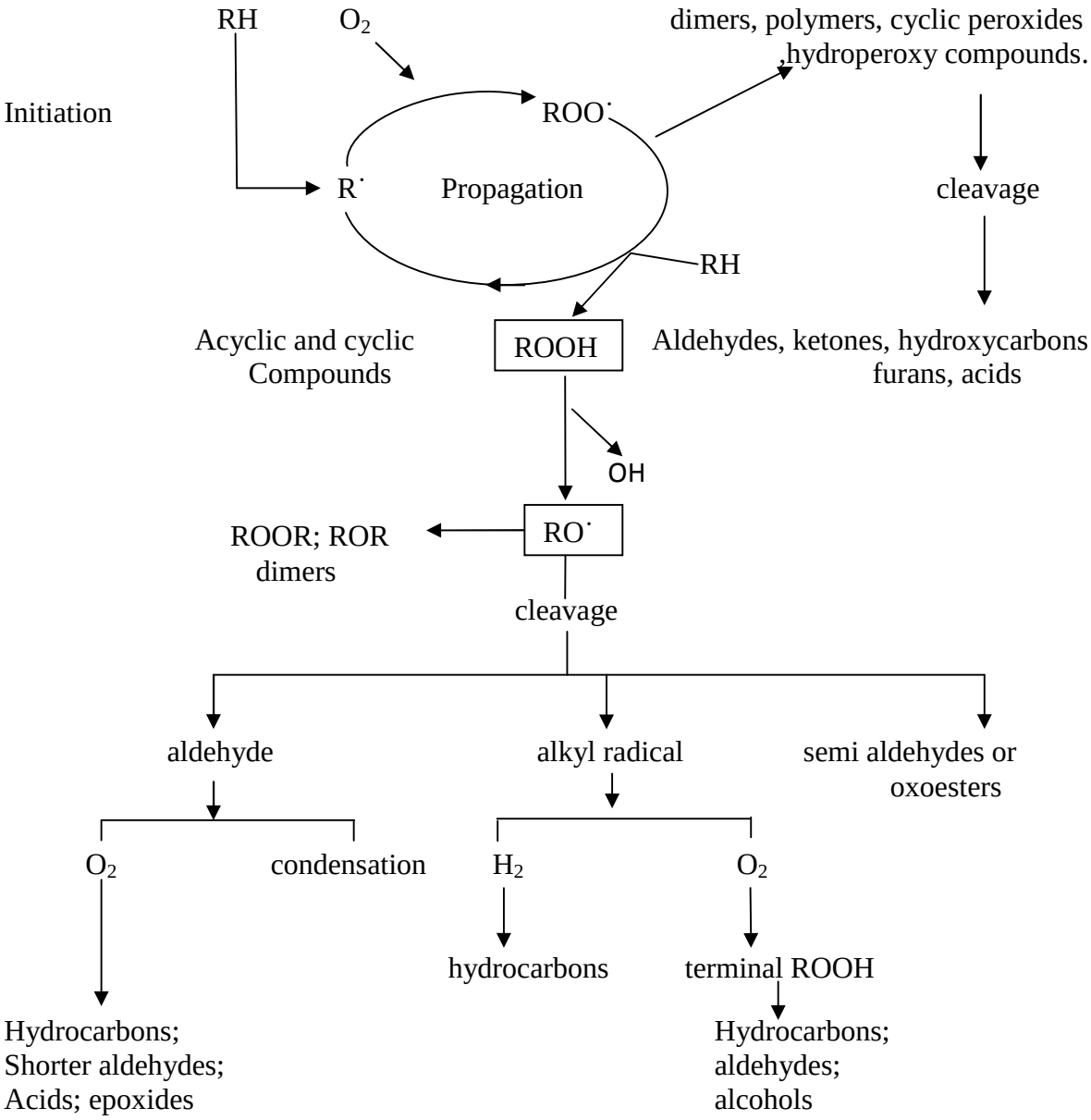

Fig. 2.1 Lipid autoxidation system in food

Source: Nawar (1996)

Propagation then takes place forming more peroxides and free radicals, increasing the rate of oxidation until the reaction must eventually slow or terminate due to the amount substrate available to form them (Nawar, 1996)

Factors influencing the oxidative stability of peanuts include the handling, processing, and environmental conditions occurring during distribution. These factors, as mentioned above, include light contact to the seed, oxygen levels present during storage, the presence of metal catalysts, the moisture surrounding the peanut seed, and the amount of heat the

seeds were exposed (O'Keefe *et al.*, 1993). Taking these factors into account is of great importance in peanut storage. Peanuts and peanut products would not be acceptable to consumers if they contained off flavors and/or off odors. Products of lipid oxidation, such as these, reduce peanut shelf life (Nawar, 1996).

Peanut processors and handlers can use the theories of lipid oxidation previously outlined to increase shelf life. By minimizing the amount of oxygen contact (possibly by flushing with nitrogen and/or vacuum sealing), the amount of light exposure (by using brown glass or packaging material with minimal light passage), the amount of heat contact after roasting, the contact to metals such as copper, zinc, or iron, and storage at proper water activities, peanuts can be acceptable for consumption for an increased period of time (Baker, 2002).

2.7 Formation of peanut flavor during roasting

Peanut roasting and the development of color and roasted flavor in peanuts has been a topic of research for some time. Typically, roast color is the most important quality control parameter in commercial processes. Roast color in peanuts is generally measured by light reflectance in a colorimeter, giving an L-value in a range from 80 (very light or no roast) to 30 (very dark roasted). The Hunter L-value of roasted peanuts used in high quality dry roasted peanuts and peanut butter falls in the range of 50-51 (Sanders *et al.*, 1989b).

Thermodynamic modeling of continuous, cross-flow roasters was established in the latter part of the 1990's (Landman, 1994; Landman *et al.*, 1994). This process describes the heat and mass transfer as a model for peanut roasting. Color development alone does not account for the flavor constituents of a particular variety of peanut, as the substrate for melanoidin and pyrazine development may be different from variety to variety, based on the sugars and free amino acids present in the peanut seed (Baker, 2002).

2.7.1 Pyrazine development

In roasted foods, pyrazines or alkylpyrazines are formed via Maillard reactions above 70ºC, and directly contribute to roasted or cooked flavors (Maga and Sizer, 1973). Formation of roasted flavor components in peanuts is especially apparent when peanut seeds are heated above 130°C (Landman *et al.*, 1994). Pyrazines play a role in the flavor when roasting peanuts, coffee beans, sesame seeds, barley, cocoa, popcorn, potato

products, rye crisp bread and beef. Below is an example of an unsubstituted pyrazine, a six-carbon ring with nitrogen atoms at the one and four positions:

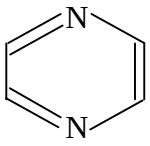

Fig. 2.2: An unsubstituted pyrazine

Source: Maga (1982)

Of all the products formed during the Maillard reaction, pyrazines are the most widely studied. These Strecker Degredation products account for well over 100 papers published since the 1950's. Many mechanisms for pyrazine formation have been postulated in models systems, but the complex nature of a food system, coupled with a multitude of reactions occurring simultaneously, do not allow scientists to measure all reactions taking place and their final products with currently available technology. This is to say that the final identified pyrazines formed during heating could come from several mechanisms, and any one particular postulated mechanism for their formation in peanuts is inconclusive (Baker, 2002).

Roasted flavor impact in peanuts is thought to come primarily from 2-methylpyrazine and isomers of dimethyl-pyrazine(Mason *et al.*, 1966; Warner *et al.*, 1996). The formation of pyrazine compounds are known to be caused from reactions involving amino acids and sugars via the Maillard reaction. The Maillard reaction, or carbonyl-amine browning reactions, was first proposed by Louis Camille Maillard in 1912. This complex scheme was further studied by Hodge (1953) and is still not fully elucidated. The fig.2.3 represents one of the many schemes for the formation of Maillard products.

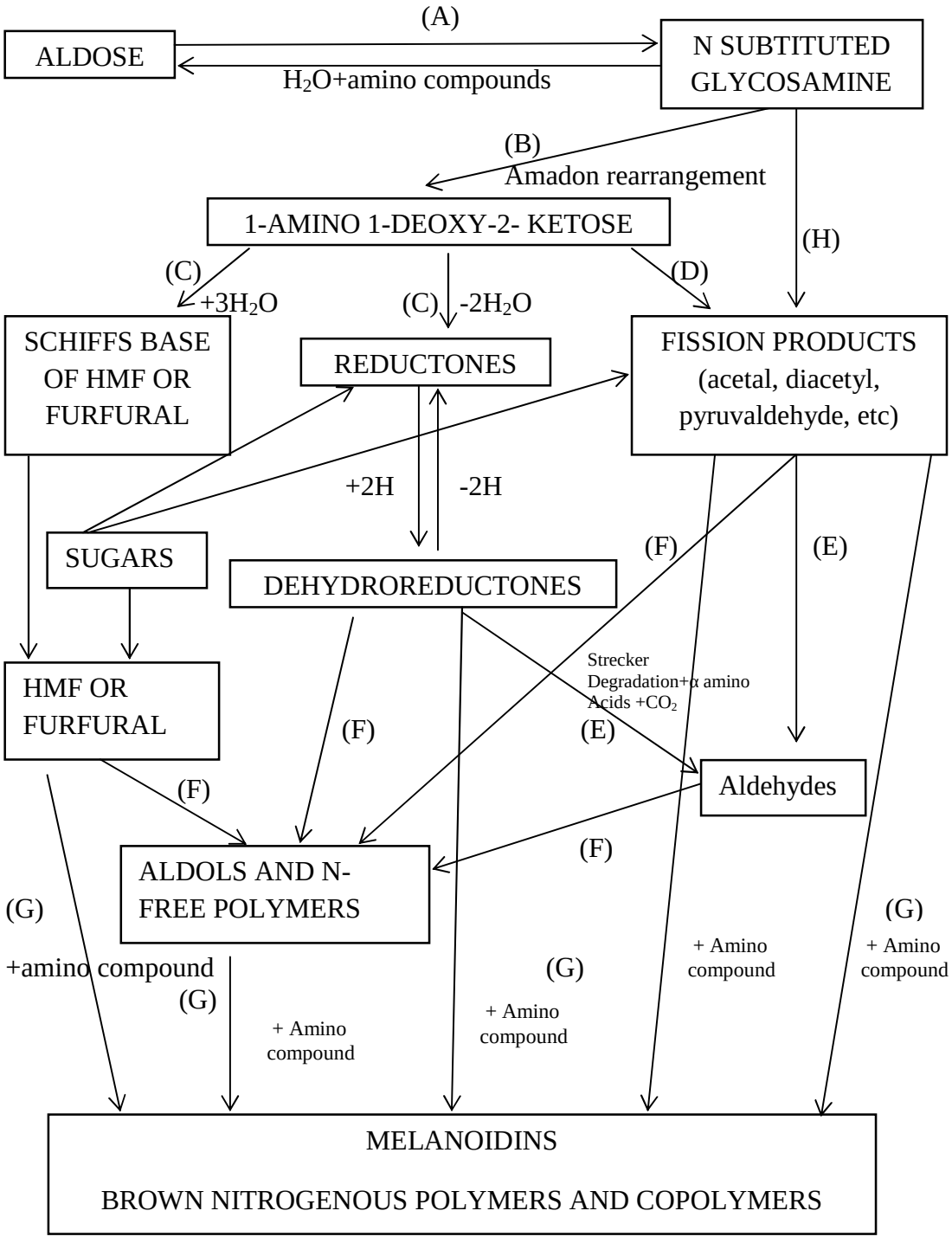

Fig.2.3 Maillard reaction pathway

Source: Fujimaki *et al.* (1985)

During heating of foodstuffs, amino-carbonyl precursors are formed by the liberation of free sugars and free amino acids. A reversible Schiff base is formed in the food matrix,

followed by the products of the irreversible Amadori rearrangement. The products of the Amadori rearrangement undergo a number of reactions, including dehydration, oxidation, cyclization, and scission (Fujimaki *et al.*, 1985).

Odor and threshold ranges (in ppm) of pyrazines isolated from peanut, to date, are found in the following Table 2.4

Table 2.4 Odor and threshold ranges (in ppm) of pyrazines isolated from peanut

Pyrazines	Threshold (ppm)	Odor description
Methylpyrazine	60,000 – 150,000	Roasted
2,3-dimethylpyrazine	2,500	Roasted, Nutty
2,5-dimethylpyrazine	1,800 – 35,000	Roasted
Methoxypyrazine	700	Not distinct
2-methoxy-3 methylpyrazine	4	Nutty, Roasted Peanuts
Trimethylpyrazine	9,000	Roasted Chicken

Source: Maga (1982)

Peanuts, when heated, develop a unique, desirable, roasted peanut flavor. Roasting time and temperature play a role in the formation of peanut flavor and influence intensity of flavor and odor in roasted peanuts (Buckholtz *et al.*, 1980). According to Newell *et al.* (1967), free sugars and free amino acids were the major precursors to roast peanut flavor. These precursors form pyrazines and carbonyls via Maillard type reactions that contribute heavily to roasted peanut flavor (Mason *et al.*, 1966; Newell *et al.*, 1967; Walradt *et al.*, 1971). Alkylpyrazines formed in low moisture, amino acid-carbohydrate model systems, are thought to be the major constituents involved in roasted peanut flavor (Koehler and Odell, 1970).

Conversion of sucrose to fructose and glucose and/or formation of comparable reductones appear to be the major mechanism in regards to carbohydrates involved in this reaction (Mason et al., 1966). This was also seen in work done by Chiou and Tsai (1989), where glucose contents were higher after 10 minutes of roasting than in raw peanuts. Aspartic acid, asparagines, glutamic acid, glutamine, histadine, and phenylalanine are amino acids thought to be associated to the typical flavor in peanuts (Newell *et al.*, 1967). Peptide-2 (an unknown compound) was found to contribute greatly to roasted flavor, when hydrolyzed during roasting. This peptide, found in ample amounts in peanuts, is hydrolyzed when heated to form necessary amino acid reactants. Peptide-2 was also shown to increase as peanuts mature (Mason *et al.*, 1966).

Flavor and color development during peanut roasting is mainly affected by thermal processing conditions; however, other factors such as peanut variety, maturity, and post-harvesting conditions may also influence peanut flavor (Rodriguez *et al.*, 1989; Sanders *et al.*, 1989a). As sugars and free amino acids are the substrates for Maillard browning, their type and contents are important for the flavor and color formation during roasting. Immature peanuts typically contain higher concentrations of reducing sugars than mature peanuts, and thus they will roast faster than immature peanuts to the same color than mature peanut (Sanders *et al.*, 1989b). Mature peanuts typically contain higher amounts of free amino acids, which promote the development of roasted peanut flavor (Rodriguez *et al.*, 1989).

The breakdown of the polypeptide bonds and release of free amino acids from proteins and peptides during roasting allow free amino acids to participate in the Millard browning reaction by combining with the reducing sugars such as glucose and especially fructose that are also present. The amino acids, aspartic acid, glutamic acid, glutamine, histidine, asparagine, and phenylalanine have been established as precursors of typical roasted peanut flavor, whereas threonine, tyrosine, lysine, and arginine has been reported as precursors of atypical flavors (Newell *et al.*, 1967).

2.8 Factors influencing the shelf-life of peanut

Oxidation in food products leads to a reduction in the quality and shelf-life. Factors such as environmental stresses from harvesting or curing, exposure to light, heat and air can all cause different forms of oxidation (Burton and Ingold, 1986). Lipid oxidation is the main problem in the storage of high oil foods as it causes off-flavors and nutrient loss. The associated negative flavors in peanuts include rancidity, cardboard, stale, and other undesirable flavors (Warner *et al.*, 1996). Even under refrigerated conditions, foods can undergo lipid peroxidation by the mechanism of free radical chain reactions. Fig.2.4 depicts the main steps of free radical chain reactions during lipid oxidation: initiation, propagation and termination (Burton and Ingold, 1986). The initiation step is the spontaneous abstraction of a hydrogen atom from a lipid molecule and production of carbon centered radical $R^{\cdot}$. The propagation involves the direct addition of an oxygen molecule to a double bond to generate hydroperoxide compounds ($ROO^{\cdot}$). The $ROO^{\cdot}$ formed reacts with the organic material (RH) to produce ROOH and release $R^{\cdot}$. This propagation step is continued as $R^{\cdot}$ can be oxidized into a free radical ($ROO^{\cdot}$) and then released to produce more free radicals. This chain reaction is terminated when two free radicals produce a non-radical molecule (Wasowicz *et al.*, 2004).

Initiation

$$RH \longrightarrow \text{Production of } R^{\cdot}$$

Propagation

$$R^{\cdot} + O_2 \longrightarrow ROO^{\cdot}$$

$$ROO^{\cdot} + RH \longrightarrow ROOH + R^{\cdot}$$

Termination

$$ROO^{\cdot} + ROO^{\cdot} \longrightarrow \text{molecular products}$$

Fig.2.4 Steps of oxidation free radical chain reaction, where RH = lipid molecule and $R^{\cdot}$ = carbon centered radical (Burton and Ingold, 1986).

Both enzymatic and non-enzymatic catalyzed oxidation may occur in raw and roasted peanut during storage; however, considering the effect of enzyme inactivation during high

temperature roasting, the greater rate of lipid oxidation for roasted peanuts is probably caused by lipid oxidation catalyzed by metalloproteinase (Agbo *et al.*, 1992). Previous studies claimed particular transition metals, such as iron, may promote the lipid oxidation in food system (McClements and Decker, 2000).

The commonly used indicators of lipid oxidation are peroxide value (PV) and thiobarbituric acid reactive substances. As the primary reaction products of lipid oxidation are peroxides (ROOH), their concentration can be used as an indicator of the process of oxidation. The most commonly used method utilizes the ability of ROOH to liberate iodine (I_2) from potassium iodide(AOAC, 2010). The iodine formed can be reduced to iodide by adding the sodium thiosulfate ($Na_2S_2O_3$). The complete reduction of iodine to iodide can be reflected by the color change of solution from blue to colorless using starch as the indicator (Chang *et al.*, 2013). An issue in the use of the PV method lies in the fact that peroxides are primary products that are decayed in the latter stage of lipid oxidation. A lower PV may represent the very initial or the final stages of oxidation. An alternative method to measure the extent of lipid oxidation is to use the thiobarbituric acid test, which measures the products of hydroperoxide degradation. This method depends on the color products formed from the condensation of thiobarbituric acid with malonaldehyde, which is presumably formed in the oxidized lipids. This method is limited as other food components may react with thiobarbituric acid causing similar color formation (Kanner and Rosenthal, 1992).

Lipid oxidation is the major cause of the degradation of pleasant flavors and pres**e**nce of unpleasant off flavors, such as painty, rancidity, and cardboard, during peanut storage (Riveros *et al.*, 2010). The association between the peroxide value (PV) results and developed rancidities has been established (Agbo *et al.*, 1992). Both chemical and sensory analysis were used to assess peanut storability in shelf life tests (Agbo *et al.*, 1992; Riveros *et al.*, 2010). Additionally, positive correlations were determined between chemical analysis results (PV, conjugated dienes, and p-anisidine values) and oxidized and cardboard flavors, whereas negative correlation exists between roasted peanutty flavor and chemical analysis (PV, conjugated dienes, and p-anisidine value). In the literature, the shelf life of medium roasted peanut pastes prepared from normal peanuts was 128 days when stored at room temperature (23°C), based on a PV threshold of 10 meq O_2kg^{-1}, however, the shelf life of pastes made from high oleic peanuts was 300 days (Riveros *et al.*, 2010).

Flavor fade in peanuts has been defined as the loss of positive attributes associated with the flavor of fresh roasted peanuts such as roasted peanut and sweet aromatic accompanied by development of off flavors (such as, cardboardy, painty, and other oxidized flavors) during storage (Abegaz *et al.*, 2004).

The formation of off flavors is mainly due to the formation of various undesirable volatile aldehydes during storage, such as hexanal, heptanal, octanal, and nonanal. These sub-products of lipid oxidation may interact with roasted peanut flavor compounds, ultimately leading to loss of roasted peanut flavors. Also, large quantities of aldehydes formed during lipid oxidation may mask the roasted peanut flavors (Koehler and Odell, 1970; O'Keefe *et al.*, 1993; Warner *et al.*, 1996). In one study, tocopherols, peroxide value, and sensory attributes were tested in roasted peanuts after storage at 40 °C for 88 days (Silva *et al.*, 2010). Tocopherol contents of roasted samples decreased with storage time, but peroxide value increased with time, showing a negative correlation between the two. Positive sensory attributes of roasted peanuts decreased with storage time, while cardboardy, painty, and oxidized flavors increased. Many factors influence the lipid oxidation of peanut products, including variety, maturity, seed size, moisture content, water activity, processing, and storage and packaging conditions (moisture content of environment, processing, temperature, light, and oxygen) (Koehler and Odell, 1970; O'Keefe *et al.*, 1993; Riveros *et al.*, 2010; Sanders *et al.*, 1982).

It is known that high oleic peanuts are more resistant to lipid oxidation than normal peanuts, as the higher ratio of oleic acid to linoleic acid decreases the unsaturation of the lipids. Increase in moisture content resulted in decreased lipid oxidation rates up to the water activity of 0.40; however, further increases in water activity increased the oxidation rate as expected. Lipid oxidation occurs even at low temperatures, and the rate of oxidation reaction is accelerated with increased temperature (Evranuz, 1993). It has been found that the best strategy to control lipid oxidation of peanut paste is the prevention of oxygen contact with peanut pastes and utilizing low temperature for storage (Agbo *et al.*, 1992). Also, addition of some additives, such as a metal chelator (EDTA) or an antioxidant (mainly tocopherols), showed the potential to retard lipid oxidation reactions (Agbo *et al.*, 1992).

As previously described, the Maillard reaction is thought to generate compounds called Maillard reaction products with strong antioxidant properties (Amarowicz, 2009).

Enhanced antioxidant capacity resulted from the formation of Maillard compounds, such as melanoidins, and/or release of previously bound polyphenolic compounds, such as p-coumaric acid, which could protect tocopherols from heat degradation during roasting (Amarowicz, 2009). In the case of coffee roasting, the differences in the antioxidant activity of brewed coffee were highly dependent on the degree of roast (the darkness of roasted bean color) rather than on the type of coffee (Agbo *et al.*, 1992). The radical-scavenging activity of the non-phenolic fraction of brewed coffee increased with the degree of roasting along with the accumulation of MRP; however, dark roasted coffee showed reduced radical-scavenging activity compared to medium-roasted coffee. This was attributed to the degradation of the polyphenols formed via the earlier stages of Maillard browning (Baker, 2002).

The relationship between roasting conditions and storability is complex in the case of peanuts. Dry roasting was found to cause microstructure damage to the oleosomes, which allows the access of oxygen into the cell tissue, which can promote the lipid oxidation during storage (Perren and Escher, 2013). The damaged cell structure was seen to compromise the shelf life of roasted peanuts compared to the unroasted. Another aspect of the shelf life of roasted peanuts, roasting intensity, was considered the primary factor determining the storability. Although heat degradation of tocopherols during peanut roasting was observed, formation of MRP with high antioxidant capacities could protect the tocopherols from deterioration. Storage tests suggested tocopherol contents of roasted samples had a negative correlation with the peroxide value (Chang *et al.*, 2013). Both studies found that α-tocopherol showed the least stability in roasted peanuts compared to the other tocopherols under storage conditions.

Roasted peanuts showed enhanced total antioxidant capacity, which increased as peanuts were roasted to darker colors. Roasting to darker colors resulted in slower degradation of tocopherols during storage, attributed to higher amounts of MRP with antioxidant capacity formed during dark roasting that provide protection to the tocopherols (Davies and Labuza, 1997).

In one study, peanuts were roasted at 166°C from 0 to 77 min in a lab-scale oven and crude (unrefined) peanut oil was subsequently mechanically pressed and placed in open beakers at 85°C to accelerate oxidation. The most liable tocopherol, i.e. α-tocopherol, showed the greatest decreases in samples roasted for 7 and 21.5 min, whereas degradation

of this antioxidant was substantially less with higher roast intensities. Similar trends were observed for the total tocopherols. The final concentration of tocopherols in roasted peanuts or peanut oil was a balance between heat degradation and indirect heat stabilization via the formation of MRP with high antioxidant capacity (Davies and Labuza, 1997).

In a study by Chang *et al.* (2013), runner peanuts were roasted in a two zone gas fired roaster at 135 °C in the 1st zone and at 190 °C in the 2nd zone to a medium roasts. A storage study was conducted at 21±1 °C for 38 weeks for raw and roasted peanuts either under vacuum, or under ambient air conditions. When air was present, the peroxide value for the roasted peanuts reached 47meq/kg by 12 weeks. Under vacuum, lipid oxidation was significantly suppressed compared to storage when air was present. It was noted in that study that regardless of ambient air or vacuum condition, the roasted peanuts always had higher peroxide values than the unroasted raw peanuts (< 2 meq/kg), as roasting causes microstructure damages that promote lipid oxidation (Perren and Escher, 2013). In both studies, α-tocopherol was the least stable tocopherol isoform during roasting (Chang *et al.*, 2013; Perren and Escher, 2013).

2.9 Almond oil

Roghan Badam Shirin is commonly known as Badam Rogan, Badam Oil and sweet almond oil. The sweet almond kernels (especially BADAM MALAI GIRI imported from Afghanistan) are used for extracting Rogan Badam Shirin. Badam oil is the widely used for its health benefits internally as well as externally. Generally, Roghan Badam Shirin is popular for its beneficial effects on skin and hair. Badam oil is a rich moisturiser that works against dry skin and chapped lips on regular application. It also provides cardiovascular and neurological benefits. Badam Rogan is rich in antioxidants, vitamins, essential omega-6 acid. It also contains oleic and linoleic acid. The zero cholesterol in Badam Rogan makes it desirable for regular consumption.

Roghan Badam Shirin is light and mostly colourless. It can have a sweet odour or be odourless. In Ayurveda and Unani medicine, Badam Rogan is commonly used as external application for treating black spots, marks, blemishes, dark circles, hair fall, premature whitening of hair, dry skin and chapped lips etc. Oral intake of Roghan Badam Shirin is helpful in constipation, insomnia, memory loss, headache etc (Singh, 2015).

2.9.1 Nutrition facts and calories of almond oil

1 teaspoon (4.5 ml) Almond Oil (Badam Rogan) provides 40 Kcal, 1.76 mg Vitamin E, 12 mg phytosterols (mainly β-sitosterol). The fats in it contain 8% Saturated Fatty Acids, 66% Monounsaturated Fatty Acids and 26% Polyunsaturated Fatty Acids. It does not contain cholesterol and Trans fats (Singh, 2015).

2.10 Peanut butter

Peanut butter is made from ground dry-roasted peanuts, to which may be added seasoning and stabilizers (Chang *et al.*, 2013). A minimum content of 90% peanuts with less than 55% oil in peanut butter is required to meet the standards by U.S. FDA Standard of Identity (FDA, 2015). One challenge for natural peanut butter or peanut paste without stabilizers is the phase separation, i.e., the peanut oil separates from the solid phase of peanut paste and rises to the top of the container during storage. Stabilizers, primarily partially hydrogenated vegetable oils, have been added to commercial peanut butters to prevent phase separation. Additionally, salt is added as a flavor enhancer and sugar as a sweetener. As a soft, sticky, and easy to chew food product, the characteristic stickiness of peanut butter is mainly due to the high content of protein that pulls the moisture out of the mouth when chewed. On the other hand, the oil provides lubrication and counters the stickiness of peanut butter (Hartel and Hartel, 2008).

Peanut butter is a semi-perishable product that is subject to a number of microbial, chemical and physical deteriorative changes, which affect the final quality of the finished product. The shelf life is greatly dependent on the quality of peanuts used and the conditions of the peanuts used for making the peanut butter. Deterioration of peanut butter arises from putrefaction of protein fraction caused by bacterial metabolism; darkening, which results from an interaction between sugar and protein in the product and; oxidative rancidity that develops in the unsaturated portion of oil when it is exposed to air (Woodroof, 1983a).

There have been many studies to improve peanut butter as a food commodity. Most of these addressed such problems as (1) the prevention of oil separation on the surface, (2) improvement of smoothness and spreadability, (3) improvement of the consistency and

stickiness, (4) development of a type that can be blocked and sliced, (5) enhancement of flavor by the addition of optional ingredients, (6) effects of added fats, carbohydrates and stabilizers on the final quality, and (8) prevention of rapid deterioration of peanut butter during storage. All these problems define or set the limits of the shelf-life stability of peanut butter (Woodroof, 1983a).

Color along with other quality, safety and nutritional factors have achieved a more preeminent position in the minds of the consumers. This has necessitated a greater concern on the part of the food manufacturers in assessing the color of foods. Muego-Gnanasekharan (1992) reported that peanuts that are water blanched at 90ºC for 10 min are lighter in color. The color of peanut butter is basically affected by roasting time.

The most serious problem of natural peanut butter is the tendency of the oil to separate. Oil is released during the grinding of peanuts. The improvement of emulsion stability in peanut butter is characterized by the absence of two layers of oil and meal phase during ordinary conditions of storage, and improved texture, consistency, spreadability, flavor, color as well as nutritional value. Without stabilizers, the peanut meal settles at the bottom and forms a hard layer while the oil remains on top (Muego-Gnanasekharan, 1992). Several efforts have already been made to answer the problem. Among the solutions arrived at and researches done to address this particular problem were those cited by (Woodroof, 1983b), which include special grinding of roasted peanuts, the heat treatment of butter after packaging, and the incorporation in peanut butter of various substances, including water, honey, glycerin, mono- and di-glycerides, and vegetable oils hydrogenated to various degrees of hardness.

Some peanut butters were stabilized by incorporating into them a commercially hydrogenated peanut oil (M.pt. 148ºC) and iodine value of eight (Buckholtz *et al.*, 1998). Other commercial stabilizers incorporated in peanut oil, are hydrogenated peanut oil, and salt (Chiou *et al.*, 1991). Stabilizers used for peanut butter are partially or fully hydrogenated vegetable oils. Hydrogenated oils are usually suggested as stabilizing agents for peanut butter because of their efficient homogenization and crystallization. The use of unhydrogenated palm oil has also been studied for its stabilizing action on peanut butter (Hinds, 1995).

2.10.1 Manufacturing process of peanut butter

It is possible to buy raw peanuts instead of shelled peanuts or peanut pods. But it is advisable to install groundnut shelling plant to ensure quality of the all-important input which determines the ultimate quality of butter. The manufacturing process is briefly described hereunder.

2.10.1.1 Groundnut pre-cleaning and shelling

Good quality groundnut pods are sorted out and de-stoned before shelling them in openers.

2.10.1.2 Peanut grading

Shelled peanuts are graded according to sizes to ensure only big or bold peanuts are taken up for process.

2.10.1.3 Peanut roasting and blanching

This is a critical stage. Roasting is done at around 160ºC for 40-60 min depending upon the moisture contents. Roasting reduces water contents to around 1% which increases the shelf life of peanuts and helps develop flavor. After roasting, peanuts are cooled and then blanched (removal of outer red skin). After blanching each peanut is inspected to remove discolored (grey or black) nuts.

2.10.1.4 Grinding

Peanuts are then ground in peanut butter mill in two stages to produce fine and creamy butter. The outlet temperature is around 65-75ºC. All ingredients like salt, sugar and stabilizers are added during this process.

2.10.1.5 De-aeration

Air is incorporated into peanut butter during milling and subsequently it is removed in vacuum.

2.10.1.6 Cooling

A scraped surface heat exchanger is used for cooling. The outlet temperature depends upon the type of stabilizer used.

2.10.1.7 Filling and packing

Peanut butter is filled in Pet Jars or metal drums as per the instructions of the buyer. Immediately after filling, the jars are vibrated to remove any remaining air bubbles. After keeping jars or drums for around 35-40 h at around 20°C, the peanut butter sets completely and can be dispatched. The process flow chart is in Fig.2.5

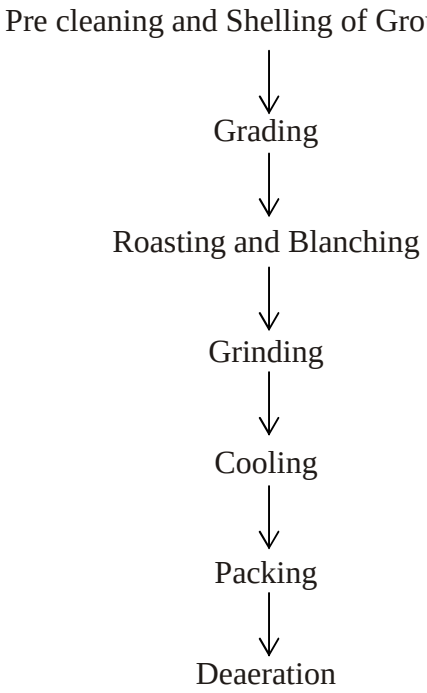

Fig. 2.5 Flow-chart for manufacturing process of peanut butter

Source:Hinds (1995)

2.11 Peanut butter and oil-separation

Peanut butter is a semi-perishable food, not readily susceptible to spoilage because of its low moisture content. The shelf life of peanut butter depends on the quality of peanuts used, method of curing and storage of the raw kernels, and the methods used in manufacturing and storing of the peanut butter (Woodroof, 1983b). According to Hinds (1995), oil separation is a concern in the stability of peanut butter because it usually indicates that the peanut butter may be rancid due to the exposure of the free oil to air and light. Many stabilizers for preventing oil separation have been developed (Chiou and Tsai, 1989). Chiou and Tsai (1989) studied the incorporation of between 1.5% to 2.0% glycerin

into peanut butter after grinding to prevent oil separation by forming an emulsion between the oil and solids in the peanut butter. However, the author did not state how long the peanut butter would avoid oil separation. A product composed of hydrogenated vegetable oil and salt that stabilizes peanut butter by reducing oil separation was developed (Woodroof, 1983b). Also reported is a similar product made from HVO and mono-glycerides derived from vegetable oils (Woodroof, 1983b). The recommended level of usage of the stabilizer is 1.0% to 2.0% by weight and it can be added into the grinder with other ingredients such as sugar and salt. Hydrogenated peanut oil is the stabilizer usually favored by the U.S. peanut butter industry for the prevention of oil separation (Woodroof, 1983b).

More recently, unhydrogenated palm oil was studied as a stabilizer for peanut butter (Hinds, 1995). The authors predicted that between 2.0% to 2.5% by weight of unhydrogenated, refined, bleached, and deodorized palm oil would prevent oil separation for more than a year at temperatures between 21 to 24°C. However, a verification of their prediction was not included in the study. Their indicator of stability was a maximum of 0.5% oil separation after 2 week of storage at 30 to 35°C. Their criteria used to establish stability was based on: (1) the U.S. Department of Agriculture (USDA) product specifications of a maximum of 0.5 ml free oil/jar of freshly manufactured peanut butter stored for 24 h at 30°C and (2) observations of commercial peanut butter, which should have remained stable for 1 year at 21 to 24°C, that showed 1% oil separation after 2 weeks of storage at 35°C (Hinds, 1995).

Expected shelf-life of a product is influenced by the environmental conditions under which the product will be stored and the amount of the initial quality that can be lost before the product can no longer be purchased by consumers. To determine shelf-life of a product, without waiting months or years, accelerated shelf-life testing is used. In accelerated shelf-life testing, the product is held under abuse conditions, including high temperatures or humidities to speed up the rate of quality loss. The most often used acceleration method is a combination of a higher humidity and temperature than the food to which it would normally be subjected (Vickers *et al.*, 2014).

2.12 Shelf life of peanut butter

Shelf life represents the period of time through which food products remain safe to eat and retain their essential sensory properties and comply with the label's nutritional declarations (Doughari *et al.,* 2007; Steele, 2004). Many products have a limited shelf life because as soon as they are produced, changes in their wholesomeness begin to occur and after some period, the product loses its wholesomeness and therefore must be removed from the shelf (Prescott *et al.*, 2002). Labuza (1982) explained the principal mechanisms involved in the deterioration of processed foods are related to the microbiological spoilage sometimes accompanied by pathogen development, chemical; and enzymatic activities causing breakdown of lipid, color, odor, flavor and texture and finally moisture and or vapor migration producing changes in texture, water activity and flavor.

2.12.1 Peroxide Value

Peroxide value indicates the degree of rancidity and provides a measure of the prospective life of the oil. It shows the influence of air, light and time on the oil and measures the amount of oxidation due to these factors at any specific time. It is generally thought that peroxides arise in vegetable oils from the attack by oxygen on the-CH group between carbon double bonds. These 2 hydrogen atoms are reactive and peroxides are formed. The breakdown of peroxides can be catalyzed by acids or heat and yields a number of other breakdown products, which contribute to the flavor or otherwise of oxidized oils. This might be due to the hydrolysis of triglyceride to free fatty acid which might be due to presence of moisture in the oil and activity of lipase enzyme coming from the contaminating microorganism.

Peroxide value measures the formation of intermediate hydro-peroxides in milli equivalents of active oxygen per kilogram of sample. Fresh oils have values less than 10 meq O_2 kg^{-1} (Pearson, 2007) and the value below 10 characterized the majority of conventional food grade oils (Codex, 1993).

2.12.2 Acid Value

Acid value measures the formation of FFA due to the production of lipase enzyme from microbial contamination present in 1 g of oil. According to Codex (1993), acid value for refined oils must be less than 0.6 mg KOH/g oil and for virgin oils must be less than 4 mg

KOH/g oil. FFA may be produced by the oxidation of double bonds of unsaturated fatty acid esters. In advanced stages of oxidation, FFA with low molecular weight was developed through the accumulation of acidic cleavage products and subsequently increased the acid value. This oxidation could have occurred with the aid of oxidative enzymes and the presence of a proportion of atmospheric oxygen in the headspace and incorporated into the oily food products. All fat containing foods is susceptible to spoilage through the auto-oxidation of unsaturated and polyunsaturated fats in the oil. The presence of FFA in oil is an indication of lipase activity or other hydrolytic action and the presence of the enzyme lipase. Therefore the increase in FFA level which is measured by the acid value suggests that as storage period increases the extent or proneness to rancidity also increases.

Part III

Materials and methods

3.1 Materials

3.1.1 Peanut

Out of several varieties of peanuts, *'Jayanti'* (Type-Spanish bunch) variety was selected because of high oil content and purchased from local market of Dharan.

3.1.2 Fat/Oil

Almond oil (*Hamdard* Rhogan badam Shirin) was selected and purchased from Gorkha department store of Itahari.

3.1.3 Emulsifier

Lecithin as emulsifier was provided by CCT laboratory.

3.1.4 Sugar/salt

Iodized salt produced by salt trading corporation was used and sugar from local grocery store was purchased.

3.1.5 Chemicals

All the chemicals required for the chemical assay were provided by CCT laboratory.

3.1.6 Glassware and apparatus

Glassware and apparatus like weighing balance, mixer/blender, heating oven (Tirano BDT 102, Baltra^{TM}), thermometers, conical flasks, beakers, aluminum foil, digestion and distillation flasks, Soxhlet apparatus, mortar and pestle were all provided by CCT laboratory.

3.2 Methods

3.2.1 Peanut grading

The un-shelled peanuts were graded for color, defects and spots.

3.2.2 Dry roasting

At first, threshold value for peanut roasting was determined and then the time-temperature variation was done within those threshold limits to obtain samples for sensory evaluation. The threshold value for peanut roasting was found to be within temperature range of 130-150°C and time range of 10-20 min for small scale of sample of 100 g for each roasting batch.

Dry roasting of the graded peanuts was done in pre-heated heating oven (Tirano BDT 102, Baltra™) in a batch of 100 g for each sample at different time-temperature variation.

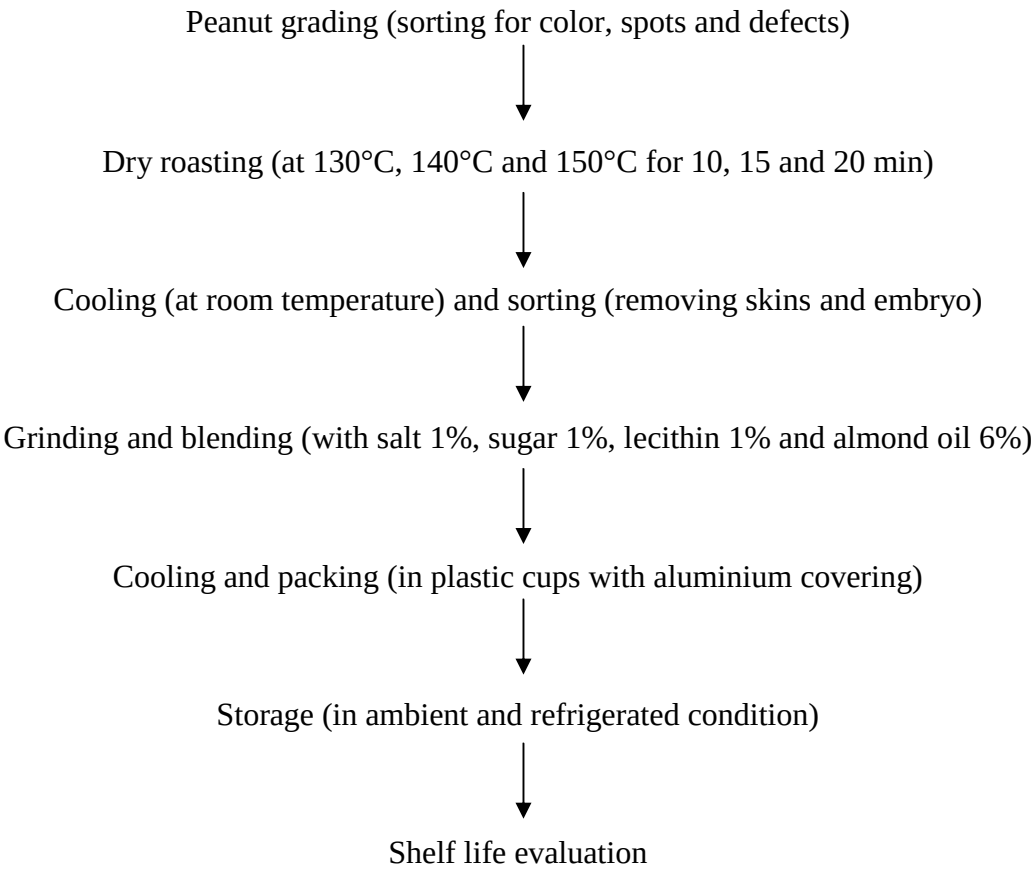

Fig.3.1 Flow chart for peanut butter preparation

Source:Hinds (1995)

On the basis of the threshold limits, experiment was conducted for preparation of peanut butter samples and coded as A, B, C, D E, F, G, H and I as given in Table 3.1

Table 3.1 Roasting time-temperature variation of samples for peanut butter preparation

S.N.	Samples	Roasting temperature(°C)	Roasting time(min)
1.	A	130°C	10
2.	B	130°C	15
3.	C	130°C	20
4.	D	140°C	10
5.	E	140°C	15
6.	F	140°C	20
7.	G	150°C	10
8.	H	150°C	15
9.	I	150°C	20

3.2.3 Cooling and sorting

At the exact time-temperature cooking was completed and the roasted peanuts were removed from heating oven as quickly as possible in order to stop cooking and produce a uniform product. The peanuts were then brought to a temperature of 30°C (room temperature).Sorting of roasted and cooled peanuts were done by removing skins and heart(embryo) of peanuts with hand rubbing.

3.2.4 Grinding and blending

The roasted, cooled and sorted peanut samples of 100g were subjected to the grinder for coarse grinding and blending along with salt and sugar each 1%, lecithin 1% and almond oil 3%to make the total fat content not to exceeding 55%. Then, each prepared sample was subjected to mortar and pestle for fine grinding.

3.2.5 Cooling and packaging

The blended peanut butter samples were cooled to room temperature and packed in plastic cups along with aluminum foil covering.

3.3 Sensory analysis

Sensory analysis of prepared nine samples of peanut butter were done by 9 point hedonic rating test (Ranganna, 2007) as shown in Appendix A. Samples were compared on the basis of taste, flavor, spreadibility and overall acceptance. The sensory panelists were teachers of food technology of CCT. Peanut butter samples were kept in plastic cups for analysis. Bread loafs were provided for evaluation of spreadaibility of the samples.

3.4 Statistical analysis

The determination of all the readings was conducted in triplicates. The data were analyzed by using statistical tool Genstat Discovery Edition 3 (2008) at 5% level of significance. The means were compared using LSD methods.

3.5 Analytical methods

Lab analyses of the optimum product were carried out including proximate analysis using (Ranganna, 2007) and AOAC (1990) methods and the shelf life studies were based on peroxide values using methods described by Kirk and Sawyer (1991).

3.5.1 Proximate analysis of raw material and peanut butter

The raw peanut was ground in grinder (Bakeman^{TM}) and the grounded peanut along with the optimum peanut butter sample were taken for proximate analysis. Moisture, protein, crude fat, crude fiber and ash content were determined using standard methods. All analyses were carried out in triplicates.

3.5.1.1 Determination of moisture content

The moisture content was determined by using hot air oven method as described byRanganna (2007).2 g of each sample was transferred into a previously dried and weighed dish (W_1). The dish and its content (W_2) were placed in hot air oven (Tirano BDT 102, Baltra^{TM}) which was thermostatically controlled at 105°C for 6hrs. The dish was

removed placed in a desiccator and weighed and subsequently put back into the oven, reheated, cooled and weighed until constant weight (W_3) was attained. The loss in weight was reported as the percent moisture content which was calculated according to the formula:

$$\text{Moisture content} = \frac{W_1\text{-}W_3}{W_2\text{-}W_1} \times 100$$

Where:

W_1 is the weight of dish

W_2 is the weight of dish and wet sample

W_3 is the weight of dish and dried sample

3.5.1.2 Determination of crude protein

Crude protein was determined using macro Kjeldahl procedure (AOAC, 1990). 2 g of each sample was weighed into a digestion flask and 0.5 g of selenium based catalyst added. A volume of 25ml concentrated H_2SO_4 was added and the flask agitated to wet the entire sample. The flask was placed in the digestion burner and heated slowly for 8 hrs until the entire solution was clear. The sample was then cooled to room temperature and the digested solution transferred into a 100 ml volumetric flask and made to the mark. A volume of 25 ml of 2% boric acid was pipetted into a 250 ml conical flask and 2 drops of mixed indicator (20 ml bromoceresol green and 4 ml of methyl red) added. The liquid from the steam trap was drained and the stopcock which drains the steam trap left opened. The conical flask and its sample were placed under the condenser. A measured volume of 10 ml of the digested sample was transferred into the steam jacket and 20 ml of 40% NaOH was added to the decomposition flask. Distillation was run for 30 s and the distillate was titrated with 0.1 M HCL solution. To calculate the percentage total nitrogen, the formula listed below was used and the 6.25 factor was used to convert nitrogen to crude protein.

$$\text{Moisture content} = \frac{100\times (V_A\text{-}V_B) \times N_A \times 0.01401 \times 100}{w \times 100}$$

Where:

V_A is the volume in ml of standard acid used in titration

V_B is the volume in ml of standard acid used in blank

N_A is the normality of acid (HCL) and

w is the weight in grams of the sample.

3.5.1.3 Determination of crude fat

Soxhlet extraction method was used in the crude fat determination (AOAC, 1990). A mass of 2 g of the dried samples from moisture determination were transferred into a cellulose thimble. A ball of glass wool was placed in the thimble. Anti bumping granules were added to a previously dried 250 ml round bottom flask and weighed. A volume of 150 ml of petroleum spirit was added and apparatus assembled. A quick fit condenser was connected to the Soxhlet extractor and refluxed for 4 h on high heat on the heating mantle. The flask was then removed and evaporated on a steam bath. The flask with the fat was heated for 30 min in an oven (Baltra ^{TM}) at 103^0C, cooled in a desiccator and weighed.

$$\text{Moisture content} = \frac{\text{Weight of flask and fat-Weight of flask} \times 100}{2}$$

3.5.1.4 Determination of crude fiber

The defatted sample from crude fat determination (AOAC, 1990) was transferred to a 750 ml Erlenmeyer flask, 0.5 g asbestos added followed by 200 ml of boiling 1.25 % H_2SO_4. The flask was immediately placed on hot plate and connected to the condenser. After 30 min the flask was removed and the contents filtered through linen cloth in funnel and washed with boiling water until washings were no longer acidic. The charge and asbestos were washed back into the flask with 200 ml boiling 1.25 M NaOH. The flask was connected to the condenser and boiled for 30 min. The contents were filtered through linen cloth and washed thoroughly with boiling water. The residue was transferred to Gooch crucible and the remaining particles washed into the crucible with 15 ml alcohol. The crucible and its contents were dried for 1 h at 100°C, cooled in a desiccator and reweighed. The crucible was ignited in an electric furnace at 600°C for 30 min, cooled and reweighed. The percentage crude fiber was calculated using the formula.

$$\text{Crude fiber} = \frac{(X-Y)}{W} \times 100$$

Where:

X is the weight of crucible and dried sample before ashing

Y is the weight of the crucible and sample after ashing

W is the weight of the sample used in the fat determination

3.5.1.5 Determination of ash content

Ash content of the sample was determined according to AOAC (1990). A mass of 2 g sample was transferred to a previously ignited and weighed crucible and placed in a preheat furnace at 600°C for 2 h. The crucible was removed, allowed to cool in air and placed in a desiccator whilst still hot. The sample was allowed to cool and weighed. The % ash was calculated by the formula:

$$\text{Ash content} = \frac{\text{C-A}}{\text{B-A}} \times 100\%$$

Where:

A is the weight of the crucible

B is the weight of crucible and raw sample

C is the weight of crucible and dried sample

3.5.1.6 Determination of carbohydrate

Total carbohydrate was determined by subtracting the amount of ash, protein and fat from total dry matter.

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

3.5.1.7 Determination of energy

Energy content of the products was calculated by Atwater's method (AOAC, 1990)

$$\text{Energy} = \text{protein}\times 4 + \text{carbohydrate}\times 4 + \text{fat}\times 9$$

3.5.2 Shelf life Analysis

For shelf-life studies, the optimum sample was divided into two, one stored under refrigeration (0 to 4°C) and the other at ambient conditions (room temperature of 25 to 30°C). The sample was then subjected for estimation of shelf-life by calculating changes in its acid value and peroxide value for 55 days.

3.5.2.1 Determination of Acid Value

In this method (Kirk and Sawyer, 1991), 10 g of fat/oil extracted by soxhlet apparatus from the sample was taken in 250 ml conical flask and 50 ml of neutral alcohol was taken. By adding two drops of phenolphtalenein indicator and swirling the contents, the flask was taken into a hot plate and the mixture was warmed to about 70ºC. The warm mixture was titrated with 0.1N NaOH to persistent pink color and the titration was carried out in triplicate.

$$\text{Acid Value} = \frac{\text{ml of alkali} \times \text{strength of alkali} \times 56.1}{\text{Wt. of sample (g)}}$$

3.5.2.2 Determination of Peroxide Value

Peroxide value (Kirk and Sawyer, 1991) was determined by first extracting oil from peanut butter as described by (name). In this method, a volume of 100 ml chloroform was added to 50 g sample, stoppered and shaken thoroughly with a Griffin Flask Shaker (M009B) for 30 min and the procedure repeated five times. The extract was collected in a conical flask and left in the fume chamber for most of the chloroform to evaporate and the oil was siphoned using a syringe. The peroxide value was then determined by weighing 3 g of the extracted oil into a stoppered conical flask and 10 ml of chloroform added and the fat dissolved by vortexting after which 15 ml of glacial acetic acid and 1 ml fresh saturated aqueous potassium iodide was added. The flask was then stoppered and shaken for 1 min and placed in the dark for 5 min, topped with 75 ml of distilled water, mixed and titrated with 0.01 M sodium thiosulhate solution using 1% soluble starch solution as indicator. The peroxide value was obtained using the equation.

$$\text{Peroxide Value (mg KOH/g)} = \frac{(V\text{-}V_0)T\times 10^3 \text{mEq } O_2/\text{kg}}{M}$$

Where:

$(V - V_0)$ is the titre value,

T is the exact molarity of the sodium thiosulphate solution

M is the mass in grams of the sample

Part IV

Results and discussion

Process optimization for the preparation of peanut butter was conducted by determining the threshold value for peanut roasting at first and then the time-temperature variation was done within those threshold limits to obtain samples for sensory evaluation. The threshold value for peanut roasting was found to be within temperature range of 130°C to 150°C and time range of 10 to 20 min for small scale of sample of 100 g for each roasting batch. On the basis of the evaluated threshold limits, experiment was conducted for preparation of peanut butter samples A (130°C for 10 min),B (130°C for 15 min),C (130°C for 20 min),D (140°C for 10 min),E (140°C for 15 min),F (140°C for 20 min),G (150°C for 10 min),H (150°C for 15 min) and I (150°C for 20 min). For each sample, salt 1%, sugar 1%, lecithin 1% and almond oil 3% were added.

4.1 Proximate analysis of raw peanut

Table 4.1 Proximate composition (in dry basis) and energy values of raw peanut.

Parameters	Value (db)
Moisture (%)	7.06±0.18
Fat (%)	46±0.03
Total Carbohydrate (%)	12.9±0.02
Crude Fiber (%)	3.7±0.03
Crude protein (%)	26.54±0.53
Ash (%)	3.8±0.06
Energy Value (Kcal)	571.76±2.47

Raw peanuts contained low moisture in comparison to fruits like avocado which had reduced shelf-life due to growth and proliferation of microorganisms according to Adegoke *et al.* (1992). The moisture content of raw peanut (7.06 %) was comparable to the result

obtained by Boli *et al.* (2013)and other low moisture cereals and legumes such as soyabean (8.1 %), Bengal gram and rice according to Swaminathan (2003).

The fat content of raw peanut (46 %) was comparable to the fat content obtained byIngale and Shrivastava (2011) in JL-24 variety of peanut i.e. 46.2 %. The carbohydrate content of raw peanut (12.9 %) was less than the data provided by (Settaluri *et al.*, 2012) i.e. 21.51 %. Akaninwor and Arachie (2002)reported that the carbohydrate content of most fruits and seeds gave values ranging from 2.97 to 4%. This result showed that peanut contained higher amount of carbohydrates than most of the fruits.

The crude fiber content of raw peanut was found to be 3.7 %. This was comparable to that obtained by FSA (2007) for banana and guava, which were 3.4 and 3.6% respectively. This value is also comparable to the values reported by Boli *et al.* (2013). The crude protein content of raw peanut was found to be 26.54% which is greater than the values given by Atasie *et al.* (2009). Fruits such as avocado contained crude protein value of 1.72. Fruits have been reported to provide comparatively little protein (Hawthorn, 1981). The ash content of raw peanut was found to be 3.8 % which was less compared to the data reported by Boli *et al.* (2013) i.e. 4.86 to 5.82 %.

4.2 Sensory evaluation of peanut butter

The samples were subjected to sensory analysis to the trained and semi-trained panelist and the results were depicted statistically. The time-temperature variation on peanut roasting showed significant difference on the organoleptic quality of peanut butter.

4.2.1 Color

The mean sensory score for color were found to be 5.7, 5.8, 6.8, 6.9, 6.4, 6.2, 8.4, 6.0 and 5.6 for samples A, B, C, D, E, F, G, H and I respectively. Statistical analysis showed that the variation in time-temperature of peanut roasting for preparation of peanut butter samples had significant effect in the color of peanut butter ($p<0.05$). LSD indicated that the color score of sample G (roasting at 150°C for 10 min) was significantly different from the rest of the others and also had the highest score. The result showed that the color of the peanut butter grew darker with the increase in the temperature at fixed time. Color development in peanut butter is dependent on the creation of brownish-colored polymeric compounds known as melanoidins formed via Maillard browning products that correspond

directly to color development in foods. As the temperature of roasting increases (for a given time), the final color appears darker due to the formation of higher molecular weight melanoidins formed during heating (Ames *et al.*, 1994). The color scores of sample C and sample D were not significantly different from samples E and F but were significantly different from samples A, B, G, H and I. Likewise, the color score of sample I was significantly different from samples C, D, E and G but were not significantly different from samples A, B, F and H.

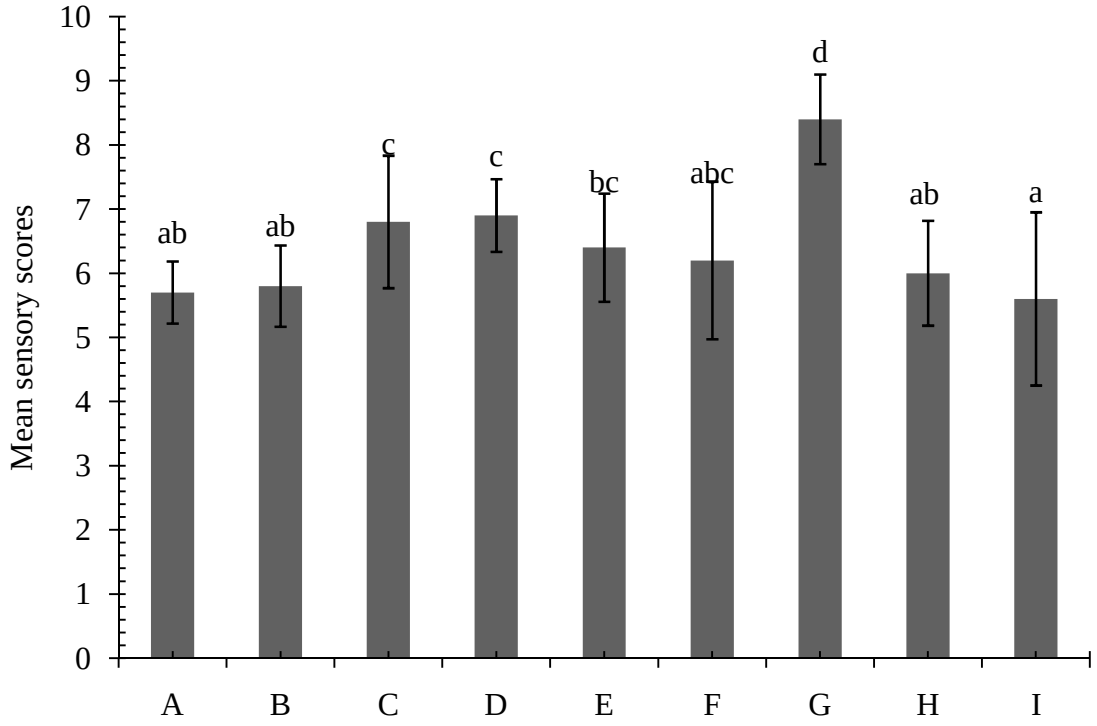

Fig.4.1 Sample mean sensory score for color

The mean sensory score for color is represented in Fig.4.1. Values on top of the bars bearing similar superscript are not significantly different at 5% level of significance. Vertical error bars represents ±standard deviation of the scores given by panelists.

4.2.2 Flavor

The mean flavor scores for samples A, B, C, D, E, F, G, H and I were found to be 6.1, 5.9, 6.8, 7.0, 6.4, 5.9, 7.7, 6.1 and 6.0 respectively. Statistical analysis showed that the variation in time-temperature of peanut roasting had significant effect in flavor of peanut butter ($p<0.05$).LSD at 5 % level of significance indicated that the sample G was significantly

different from rest of the entire sample and also had the highest score. It may be due to development of flavor compounds on high temperature roasting of peanut but high temperature roasting along with increasing time might have resulted to burnt flavor and was unacceptable as compared to sample G by the panelist. Roasting time and temperature play a role in the formation of peanut flavor and influence intensity of flavor and odor in roasted peanuts (Buckholtz *et al.*, 1980). The flavor scores of sample C and D were significantly different from samples A, B, F, G, H and I but were not significantly different from sample E. The flavor scores of sample A, B, F, H and I showed that they were significantly different from samples C, D and G but were not significantly different from sample E.

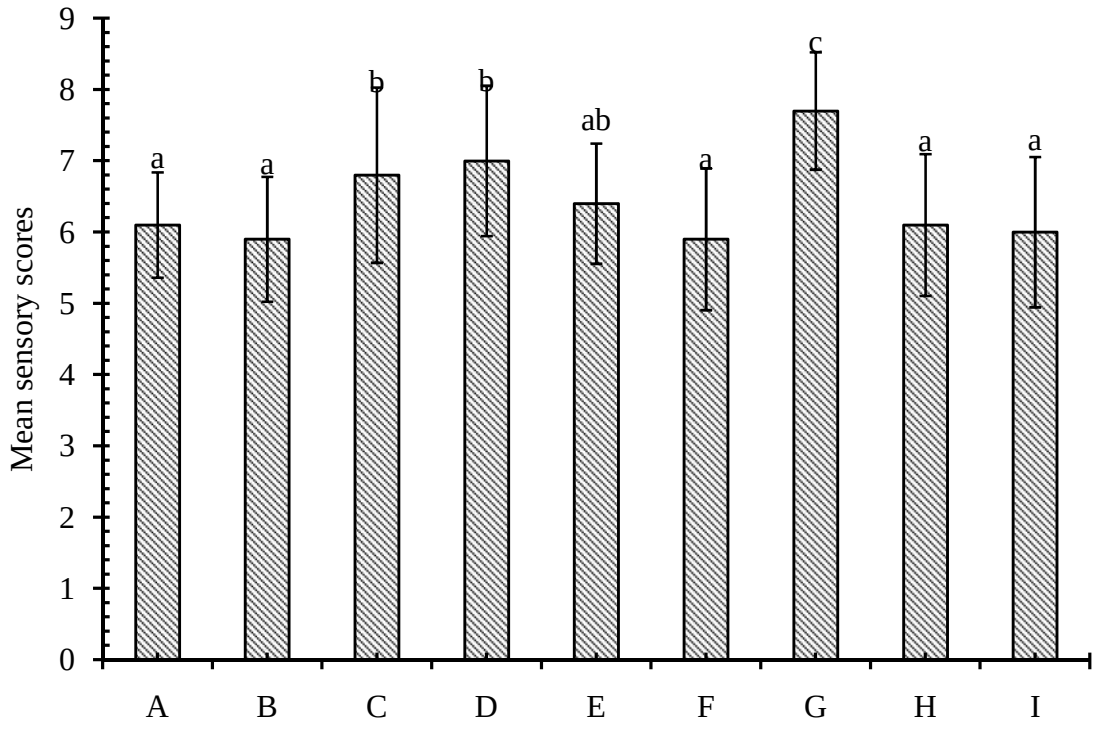

Fig.4.2 Sample mean sensory score for flavor

Values on top of the bars bearing similar superscript are not significantly different at 5% level of significance. Vertical error bars represents ±standard deviation of the scores given by panelists.

4.2.3 Taste

The average taste scores were found to be 5.9, 5.9, 6.3, 7.0, 6.4, 6.2, 7.7, 6.4 and 6.2 for samples A, B, C, D, E, F, G, H and I respectively. Statistically there was significant effect on taste of peanut butter due to time-temperature variation of peanut roasting ($p<0.05$). LSD between the samples indicated that the samples A, B, F and I were significantly different from samples D and G. Similarly sample G was significantly different from rest of all other samples except sample D and also had highest score. The sample G was prepared by roasting peanut at temperature 150°C for 10 min. It shows that higher temperature roasting imparts good taste at less time of roasting and higher temperature of roasting of peanut for increasing time of roasting might have resulted to bitter taste.

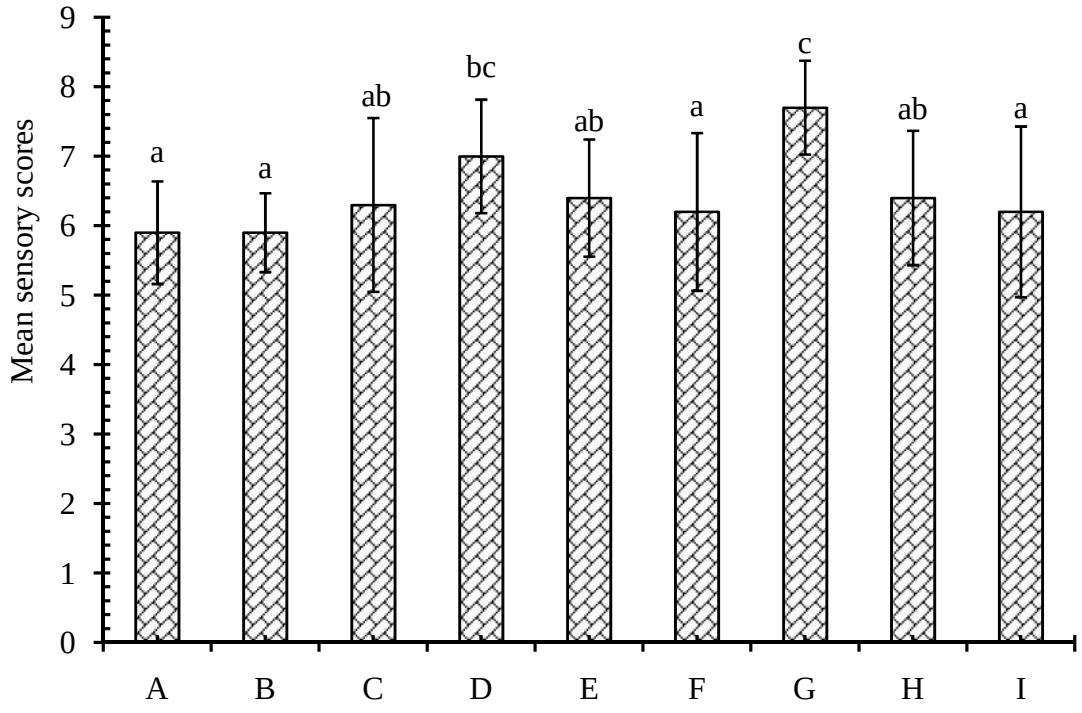

Fig.4.3 Sample mean sensory score for taste

Values on top of the bars bearing similar superscript are not significantly different at 5% level of significance. Vertical error bars represents ±standard deviation of the scores given by panelists.

4.2.4 Spreadibility

The average spreadibility score were found to be 6.5, 6.4, 6.7, 7.0, 6.3, 6.1, 7.6, 6.3 and 5.8 for samples A, B, C, D, E, F, G, H and I respectively. Statistical analysis showed that the time-temperature variation of peanut roasting had significant effect on spreadibility of peanut butter ($p<0.05$). LSD indicated that the score of sample G were highest of the entire samples and were significantly different from rest of all other samples except sample D. It might be due to combined effect of optimum time and temperature of peanut roasting leading to smooth grinding of the paste on incorporation of the oil. The score of sample I was significantly different from samples A, C, D and G. All other samples were not significantly different.

The mean sensory score for spreadibility is represented in Fig.4.4. Values on top of the bars bearing similar superscript are not significantly different at 5% level of significance. Vertical error bars represents ±standard deviation of the scores given by panelists.

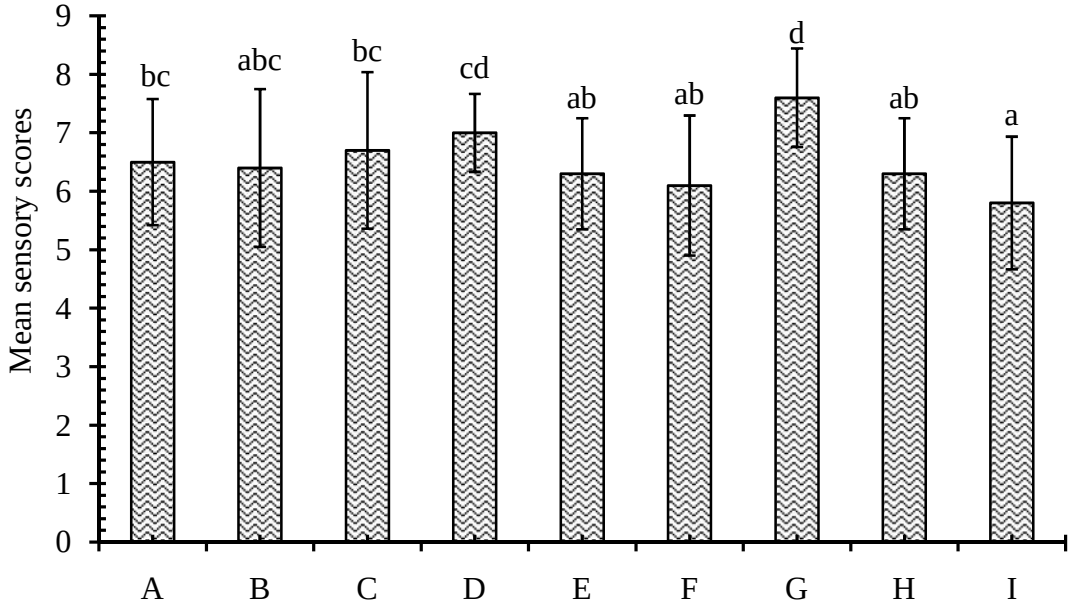

Fig.4.4 Sample mean sensory score for spreadibility

4.2.5 Overall acceptance (OA)

The mean sore for overall acceptance were found to be 6.25, 6.45, 6.5, 6.9, 6.35, 5.85, 7.6, 6.2 and 5.75 for samples A, B, C, D, E, F, G, H and I respectively. Statistical analysis showed that the variation in time-temperature of peanut roasting had significant effect ($p<0.05$) on the overall acceptance of peanut butter. LSD indicated that the overall acceptance scores of sample G were significantly different from rest of samples except sample D. Sample I overall acceptance score were significantly different from samples C, D and G but were not significantly different from scores of samples A, B, E, F and H. Sample G had highest and sample I had lowest overall acceptance score.

The mean sensory score for OA is represented in Fig.4.5. Values on top of the bars bearing similar superscript are not significantly different at 5% level of significance. Vertical error bars represents ±standard deviation of the scores given by panelists.

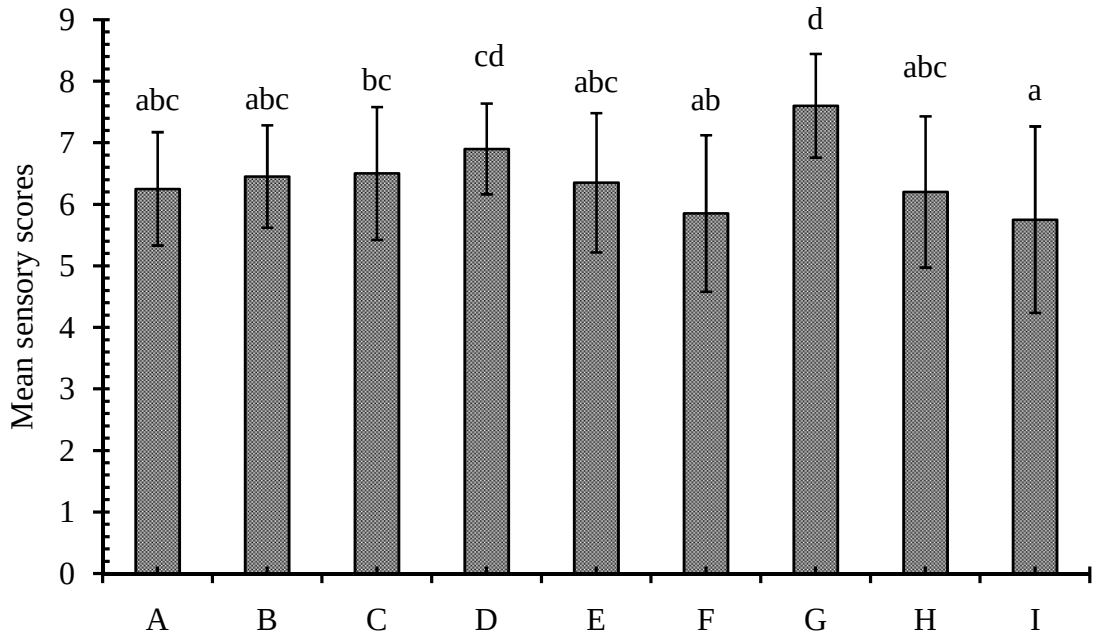

Fig.4.5 Sample mean sensory score for OA

Color, flavor, taste and spreadibility have a combined effect on the overall acceptance the peanut butters. Sample G was significantly superior in terms of color, flavor, taste and

spreadibility compared to rest of peanut butter samples. Hence, based on the sensory evaluation of peanut butter samples, optimum quality peanut butter ($p<0.05$) can be prepared by time-temperature variation for peanut roasting at 150°C for 10 min.

4.3 Proximate analysis of the peanut butter

Table 4.2 Proximate composition (in dry basis) and energy values of peanut butter.

Parameters	Values (db)
Moisture (%)	2.8±0.04
Fat (%)	49.32±0.45
Total Carbohydrate (%)	13.28±0.05
Crude Fiber (%)	3.8±0.50
Crude protein (%)	27.37±0.07
Ash (%)	4.23±0.80
Energy Value (Kcal)	606.48±3.57

Peanut spread contained low moisture in comparison to fruits like avocado which had reduced shelf-life due to growth and proliferation of microorganisms according to Adegoke *et al.* (1992). Thus, shelf-life of peanut products was expected to have longer shelf-life in comparison to products of high moisture food like avocado. The low moisture content of the peanut spread compared with the raw peanut maybe due to the addition of other solutes such as salt and extraction of butter from peanut. Prescott *et al.* (2002), reported that by adding solutes, water can be made less available. The water content or moisture of food affects its physical as well as chemical properties such as the structure, appearance and taste of the food product. These properties become important in determining the food's susceptibility to spoilage, shelf life and the processing conditions required.

Fat is one of the major constituent in peanut spread. The fat content of the peanut butter was reported to be 49.32 % which is higher than the value given by Ozcan and Seven (2003). The value is greater than cheddar cheese (FSA, 2007) but less than avocado spread which had higher fat value in raw avocado itself. The fat content of the peanut spread was

significantly increased compared to the raw peanut due to the addition of almond oil in the butter. The carbohydrate content of peanut spread was 13.28% indicating the addition of sugar and emulsifier resulted in significantly higher carbohydrate in the peanut spread. The value of carbohydrate in peanut butter is comparable to data given by Boli *et al.* (2013). FSA (2007) reported negligible values for carbohydrates in both butter and margarine.

The crude fiber content in the peanut spread increased to 3.8 %. This is similar to the value for guacamole reported by the USDA (2005). Fruits such as avocado spread contained crude protein value of 0.39%. Fruits have been reported to provide comparatively little protein (Hawthorn, 1981). The crude protein value of peanut spread was found to be 27.37% which is comparable to the value given by Boli *et al.* (2013).

It can be deduced that peanut spread is a good source of protein and should be used in conjunction with other foods as protein supplements. The ash content of peanut spread was found to be 4.23 %. The relatively higher ash value in the processed product in this project might be as a result of the salt.

4.4 Shelf life of peanut butter

Polhemus (2005) stated that in predicting shelf life, the largest number of weeks for which the degrading parameter has reached 90% must be considered. The degrading parameters in this case were acid values and peroxide values. Among these peroxide and acid values were observed for 55 days for both ambient (room temperature of 25 to 30°C) and refrigerated temperature (0 to 4°C) conditions. The PV value was found to be 10.2 when rancid flavor occurred after 11 days in non-refrigerated temperature. According to Codex (1993), rancid flavors occurs in oily products after 10 meq O_2/Kg. The result resembled with Codex (1993), but according to Codex (1993), rancidy occurs in virgin oily product after 4 meq O_2/Kg. This result got contrary to the result of peanut butter on the basis of acid value as the AV value was very low (0.028 mg KOH/Kg) when rancidity occurred in non-refrigerated temperature. This may be due to conversion of hydrolytic reactions to oxidative reactions in the peanut butter which is also shown by the logarithmic relation of PV and AV values of peanut butter as shown in Fig.4.9 and Fig.4.10 during its storage period.

PV value was thus selected to evaluate shelf-life of the product. Thus the shelf-life of the peanut butter in non-refrigerated temperature was found to be 11 days. But shelf life of

the peanut butter in refrigerated condition could not be evaluated as the rancidity did not occurred in the refrigerated temperature during the course of study (55 days) and the PV values did not exceed the threshold value of rancidity as given by Codex (1993)

4.4.1 Peroxide value of peanut butter

The peroxide values of peanut butter obtained from the samples at different length of days within 55 days of storage period at ambient and refrigeration temperature is shown in Fig.4.3.

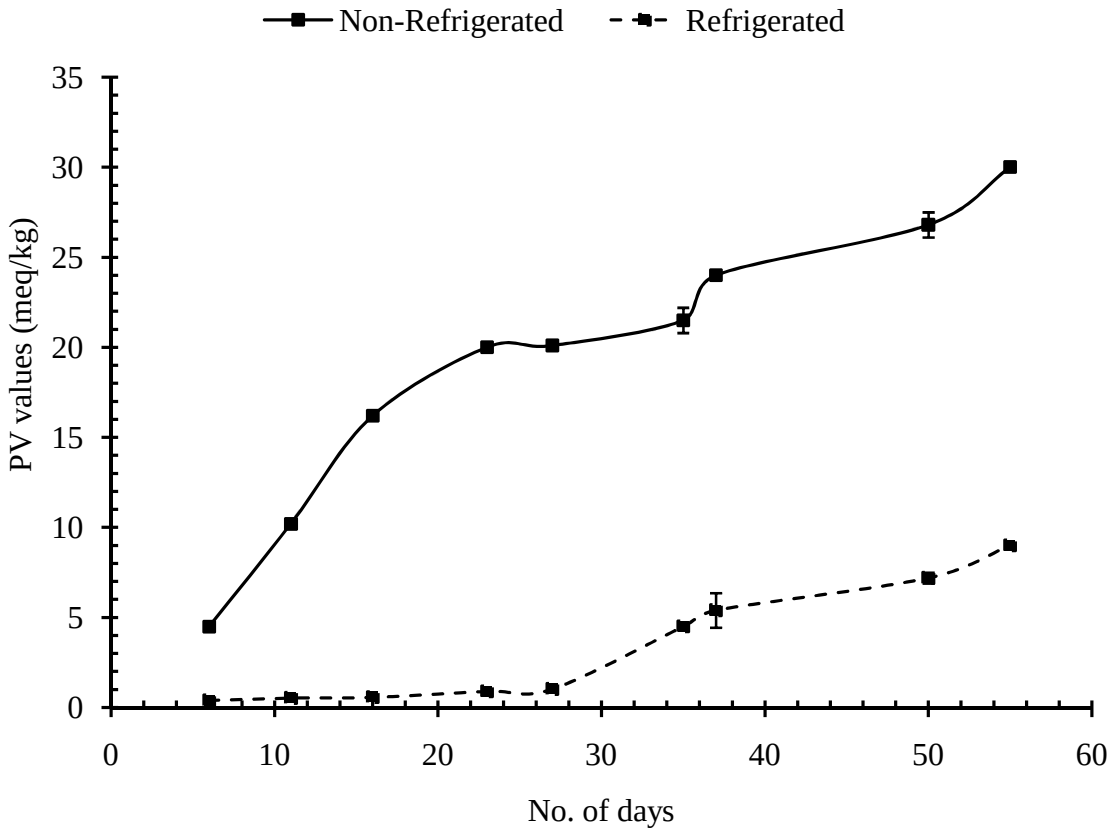

Fig.4.3 PV values of peanut butter under ambient and refrigeration temperature

*Error bars in the curve represent standard deviation of PV of triplicates at respective storage condition

It was observed that there was a steady and rapid rise in peroxide values of sample with the increase in length of days stored under ambient conditions as compared to slow increase in peroxide values for samples stored under refrigeration conditions. The highest PV is 9 meqO_2/kg at 55 days of storage at refrigerated condition indicating, still, the

freshness of the samples according to Pearson (2007). The data obtained from this experiment were to be expected since the rate of increase of peroxide concentration for the refrigerated samples were lower than the rate of increase for the ambient samples.(Pearson (2007)) stated that a rancid taste was noticeable when the peroxide value exceeds 10 meq O_2/Kg in oily products in ambient temperature. The limits of peroxide value ambient samples were considered at 10 meq O_2/Kg for this experiment .Similarly,(Pearson (2007)) stated that a rancid taste was noticeable when the peroxide value exceeds 10 meq O_2/Kg in oily products in refrigerated temperature. The limits of peroxide value refrigerated samples were considered at 10 meq O_2/Kg for this experiment.

4.4.2 Acid value of peanut butter

The acid values of peanut butter obtained from the samples at different days of sampling within 55 days of storage period at ambient and refrigeration temperature are shown in Fig.4.7

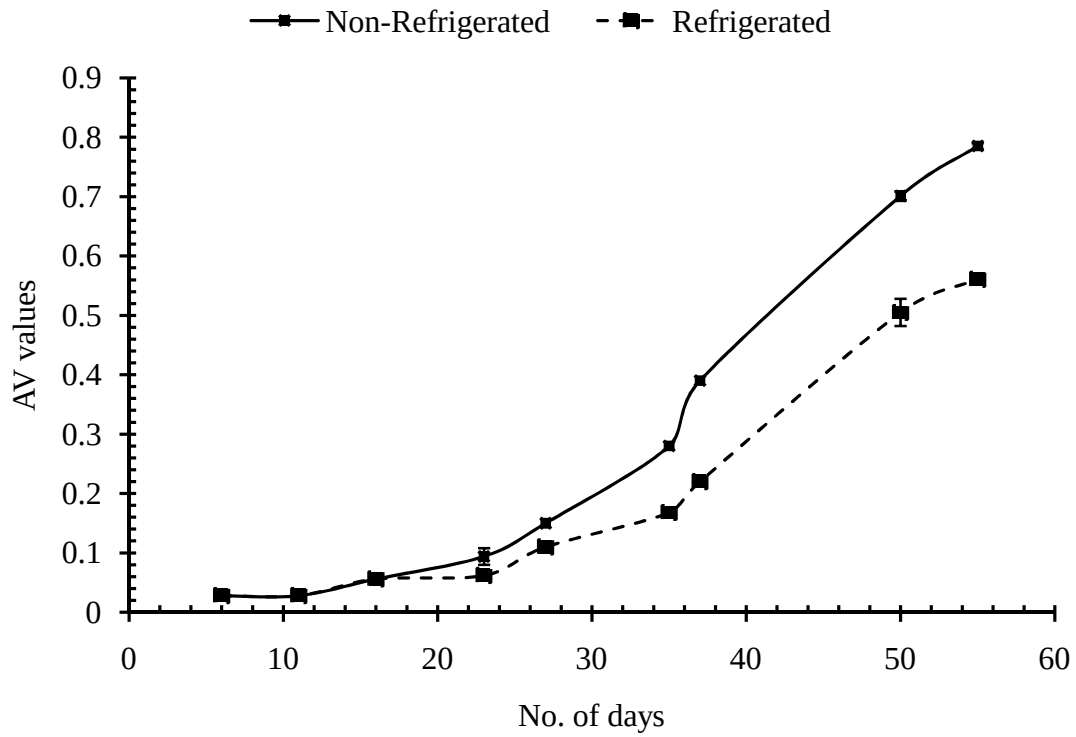

Fig.4.7 Acid values of peanut butter under ambient and refrigeration temperature

*Error bars in the curve represent standard deviation of AV of triplicates at respective storage condition.

It was observed that as the number of storage days increased, there was a rise in acid values with a steady rise for samples stored under ambient conditions and a slower increase for samples stored under refrigeration conditions. The data obtained from this experiment were to be expected since the rate of increase of lipid oxidation for the refrigerated samples were lower than the rate of increase for the ambient samples.

4.4.3 Comparison between acid value and peroxide value of peanut butter

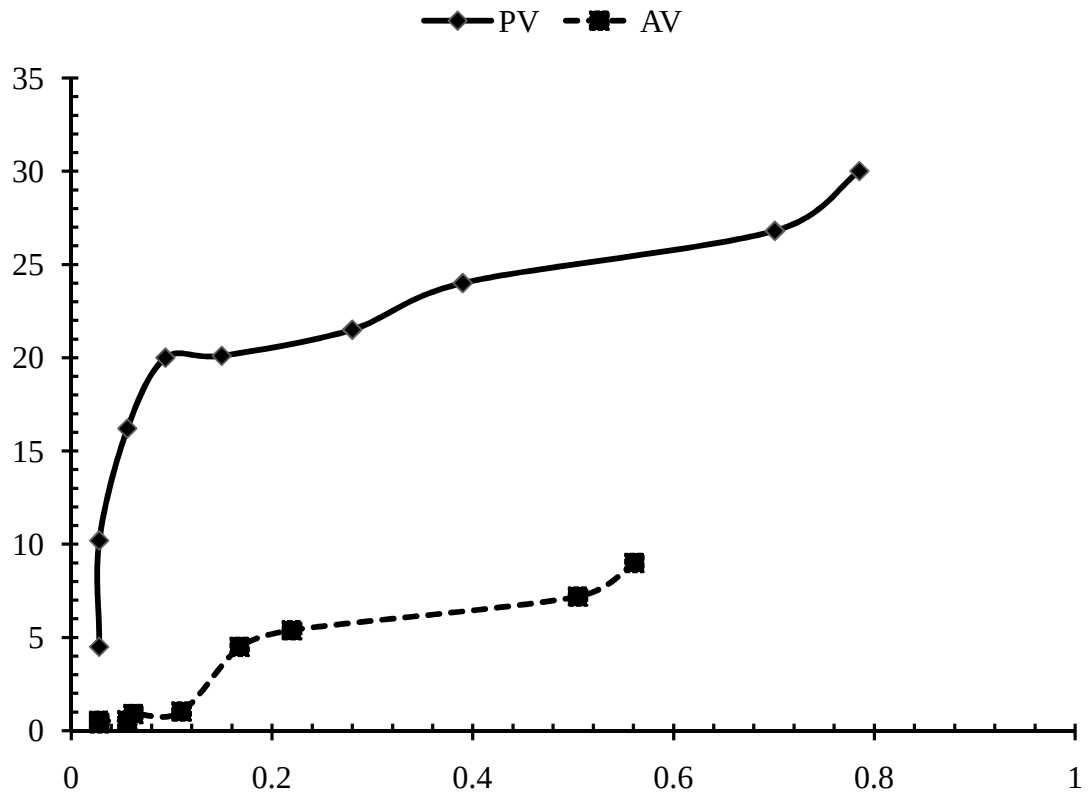

Fig.4.8 Comparison between acid value and peroxide value of peanut butter stored under different storage conditions

The primary environmental conditions that effect hydrolytic rancidity in stored fats are temperature and moisture (Allen and Hamilton, 1994). Hydrolysis is accelerated by increasing temperature with every 10º C rise in temperature; the rate of hydrolysis is expected to double. This is an important factor to consider when storing fats and oils, as even a small rise in storage temperature increases the liberation of FFA from the oil or fat, thus a having a catalytic effect on hydrolytic rancidity (Gustone and Padley, 1997).

But, to detect the rancidity in fats and oils, it is more common to use methods of determining oxidative rancidity than methods of determining hydrolytic rancidity (Pomeranz and Meloan, 1994). In the literature by (Patterson, 1989), it is explained that increase in FFA content of an oil indicates that (i) oil was stored inappropriately and (ii) oxidation has occurred. Since, hydrolytic rancidity accompanies oxidative rancidity (Williamson, 1998) and AV values are lagging shortly behind PV values in this study, it can be concluded that the liberated FFA were gradually oxidized, which is also shown in logarithmic relation (R^2=0.899 for non-refrigerated condition and R^2=0.883 for refrigerated condition) between AV and PV values of peanut butter in this study as shown in the Fig. 4.9 and Fig. 4.10.

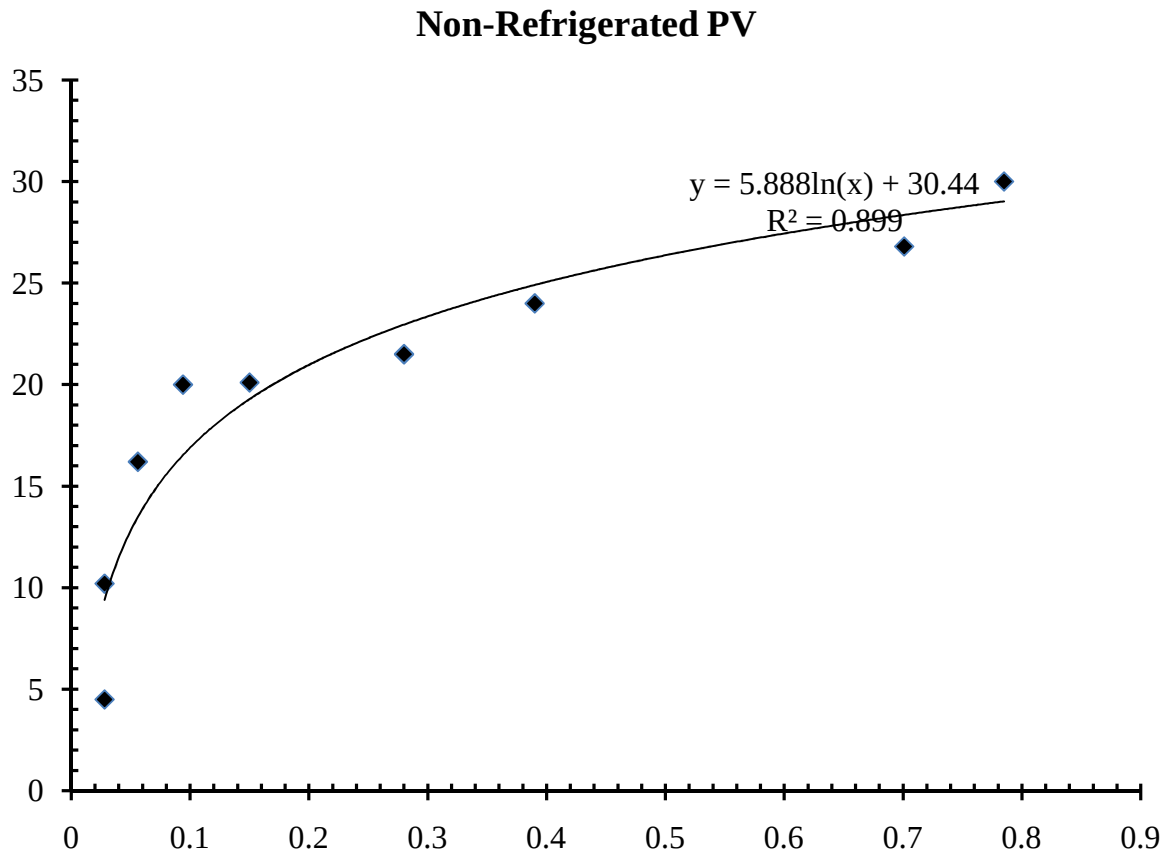

Fig.4.9 Correlation between AV and PV values of stored peanut butter at non-refrigerated condition

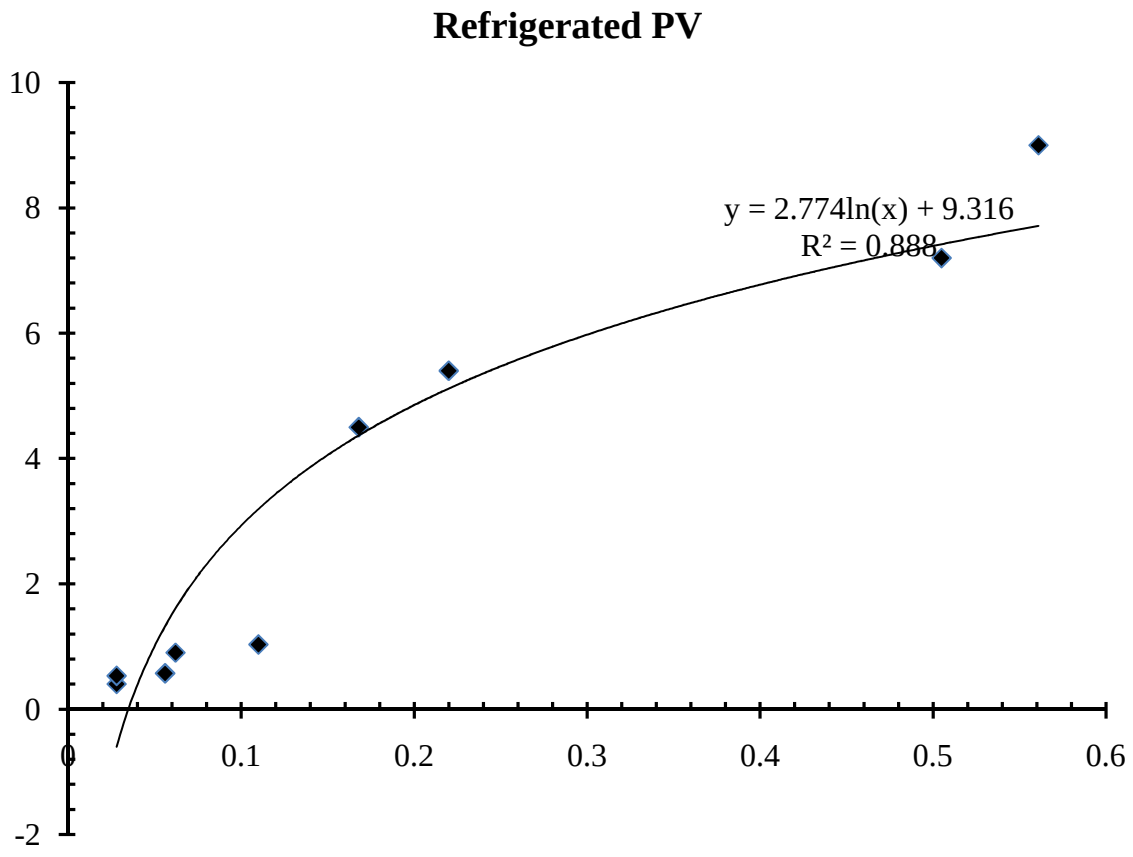

Fig.4.10 Correlation between AV and PV values of stored peanut butter at refrigerated condition

4.5 Cost calculation

The cost of 1 kg optimum peanut butter was evaluated as NRs. 600 which is shown in Appendix C

PART V

Conclusions and recommendations

5.1 Conclusion

On the present work, optimization of the preparation process and quality evaluation of peanut butter was done. From the research, the following conclusions were made:

a. Peanut butter prepared by roasting peanut at temperature of 150°C for 15 min was found to be best in terms of sensory analysis.
b. From the proximate analysis, moisture content of peanut butter was decreased compared to raw peanut due to roasting and addition of other solutes such as salt and sugar.
c. Total carbohydrate content of peanut butter increased due to addition of sugar and emulsifier.
d. The increase in ash content of peanut butter compared to raw peanut was due to addition of salt.
e. The shelf life of peanut butter was found to be 11 days for ambient (room) storage temperature of 25 to 30°C. While rancidity had not occurred up to 55 days of storage at refrigeration temperature of 0 to 4°C when 10 meq O_2/kg was considered threshold for the rancidity.
f. The overall cost of 1kg optimum quality ($p<0.05$) peanut butter was calculated to be Rs. 600 which was comparatively less than that of market peanut butter i.e., NRs. 150/200g.

5.2 Recommendations

Based on the current study, following recommendations can be made:

a. Different varieties of peanut could be studied for the preparation of peanut butter.
b. Study of amino acid profile and fatty acid profile of the product could be studied.
c. Shelf-life of the product using different packaging materials could be studied.

Part VI

Summary

Peanut butter is made from ground dry-roasted peanuts, to which may be added seasoning and stabilizers. A minimum content of 90% peanuts with less than 55 percent oil in peanut butter is required to meet the standards by U.S. FDA Standard of Identity. One challenge for natural peanut butter or peanut paste without stabilizers is the phase separation, i.e. the peanut oil separates from the solid phase of peanut paste and rises to the top of the container during storage. Stabilizers, primarily partially hydrogenated vegetable oils, have been added to commercial peanut butters to prevent phase separation. Additionally, salt is added as a flavor enhancer and sugar as a sweetener.

Raw peanut of variety *'Jayanti'* (Type-Spanish bunch) was collected from Sunsari district, Dharan and were roasted at three different temperatures (130ºC, 140ºC and 150ºC) for three different time periods (10, 15 and 20 min) to optimize the process of butter making with 9 different samples (A, B, C, D, E, F, G, H and I). The raw peanut was de-shelled, washed and sorted prior to roasting and grinding and paste was obtained by the addition of ingredients namely lecithin (1%), salt (1%), sugar (1%) and almond oil(3%) to give final fat content of the butter not more than 55 % by weight. The nine different samples were prepared and subjected to sensory evaluation. The data obtained were statistically analyzed using two way ANOVA (no blocking) at 5% level of significance which showed there exists significant difference ($p<0.05$) in overall acceptability among the samples. Sample G (roasted at 150ºC for 10 min.) got the highest mean sensory score. The sample G was thus subjected for estimation of shelf-life under different storage temperatures, namely refrigerated and ambient by calculating changes in its acid value and peroxide value for 55 days. The shelf- life of the peanut butter stored at ambient temperature was found to be not more than 11 days, but the butter stored at refrigerated temperature was not spoiled by rancidity throughout the period of storage of 55 days. The price of 1kg good quality peanut butter thus prepared at cost Rs. 600 which is cheaper than market value (R.s.150/200g)

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Appendices

Appendix A

Specimen card for sensory evaluation

Hedonic rating test

Name of panelist: …… Date:……

Product: Peanut butter

Please taste the sample and check out how much you like or dislike. Use the appropriate scale to show your attitude by giving the point that best describes your feeling about the sample.

Sample	Color	Taste	Flavor	Spreadibility	Overall acceptance
A					
B					
C					
D					
E					
F					
G					
H					
I					

Give points as follows:

Like extremely9	Like slightly6	Dislikemoderately3
Like very much8	Neither like nor dislike5	Dislike verymuch2
Like moderately7	Dislike slightly4	Dislike extremely1

Comments:……
……

…….

Signature

Appendix B

Two way ANOVA (no blocking) for sensory analysis of peanut butter

Codes	Quality Attributes				
	Color	Flavor	Spreadability	Taste	Overall Acceptability
A	5.600^{ab}	6.100^{a}	6.500^{bc}	5.900^{a}	6.250^{abc}
B	5.800^{ab}	5.900^{a}	6.400^{abc}	5.900^{a}	6.450^{abc}
C	6.800^{c}	6.800^{b}	6.700^{bc}	6.300^{ab}	6.500^{bc}
D	6.900^{c}	7.000^{b}	7.000^{cd}	7.000^{bc}	6.900^{cd}
E	6.400^{bc}	6.400^{ab}	6.300^{ab}	6.400^{ab}	6.350^{abc}
F	6.200^{abc}	5.900^{a}	6.100^{ab}	6.200^{a}	5.850^{ab}
G	8.400^{d}	7.700^{c}	7.600^{d}	7.700^{c}	7.600^{d}
H	6.000^{ab}	6.100^{a}	6.300^{ab}	6.400^{ab}	6.200^{abc}
I	5.600^{a}	6.000^{a}	5.800^{a}	6.200^{a}	5.750^{a}
LSD	0.7441	0.6973	0.6446	0.7528	0.7298

Table B.1 ANOVA for color of peanut butter

Source of variation	Degree of freedom	Sum of squares	Mean squares	Variance ratio	F probability ratio
Sample	8	60.9556	7.6159	10.94	<.001
Panelists	9	14.8444	1.6494	2.37	0.021
Residual	72	50.1556	0.6966		
Total	89	125.9556			

Table B.2 L.S.D for color

Samples	Mean Scores	LSD at 0.05
A	5.7^{ab}	0.7441
B	5.8^{ab}	
C	6.8^{c}	
D	6.9^{c}	
E	6.4^{bc}	
F	6.2^{abc}	
G	8.4^{d}	
H	6.0^{ab}	
I	5.6^{a}	

Table B. 3 ANOVA for flavor of peanut butter

Source of variation	Degree of freedom	Sum of squares	Mean squares	Variance ratio	F probability ratio
Sample	8	30.4000	3.8000	6.21	<.001
Panelists	9	31.6556	3.5173	5.75	0.021
Residual	72	44.0444	0.6117		
Total	89	106.1000			

Table B.4 L.S.D. for flavor

Samples	Mean Scores	LSD at 0.05
A	6.1[a]	0.6973
B	5.9[a]	
C	6.8[b]	
D	7.0[b]	
E	6.4[ab]	
F	5.9[a]	
G	7.7[c]	
H	6.1[a]	
I	6.0[a]	

Table B.5 ANOVA for spreadability

Source of variation	Degree of freedom	Sum of squares	Mean squares	Variance ratio	F probability ratio
Sample	8	22.3556	2.7944	5.34	<.001
Panelists	9	56.4556	6.2728	12.00	<.001
Residual	72	37.6444	0.5228		
Total	89	116.4556			

Table B.6 L.S.D. for spreadability

Samples	Mean Scores	LSD at 0.05
A	6.5^{bc}	0.6446
B	6.4^{abc}	
C	6.7^{bc}	
D	7.0^{cd}	
E	6.3^{ab}	
F	6.1^{ab}	
G	7.6^{d}	
H	6.3^{ab}	
I	5.8^{a}	

Table B.7 ANOVA for taste

Source of variation	Degree of freedom	Sum of squares	Mean squares	Variance ratio	F probability ratio
Sample	8	26.2222	3.2778	4.60	<.001
Panelists	9	20.6667	2.2963	3.22	0.002
Residual	72	51.3333	0.7130		
Total	89	98.2222			

Table B.8 L.S.D. for taste

Samples	Mean Scores	LSD at 0.05
A	5.9[a]	0.7528
B	5.9[a]	
C	6.3[ab]	
D	7.0[bc]	
E	6.4[ab]	
F	6.2[a]	
G	7.7[c]	
H	6.4[ab]	
I	6.2[a]	

Table B.9 ANOVA for overall acceptability

Source of variation	Degree of freedom	Sum of squares	Mean squares	Variance ratio	F probability ratio
Sample	8	24.8556	3.1069	4.64	<.001
Panelists	9	47.6694	5.2966	7.90	<.001
Residual	72	48.2556	0.6702		
Total	89	120.7806			

Table B.10 L.S.D for overall acceptability

Samples	Mean Scores	LSD at 0.05
A	6.25^{abc}	0.7298
B	6.45^{abc}	
C	6.5^{bc}	
D	6.9^{cd}	
E	6.35^{abc}	
F	5.85^{ab}	
G	7.6^{d}	
H	6.2^{abc}	
I	5.75^{a}	

Appendix C

Cost Evaluation of Peanut Butter

Cost evaluation of the best peanut butter sample i.e. sample prepared by roasting peanut at 150°C for 10 min was carried out for 1kg

Parameters	Quantity	Rate	Amount (NRs)
Raw Peanut	1	NRs. 200/kg	200
Almond oil	60ml	NRs. 250/50ml	300
Iodized Salt	10g	NRs. 25/kg	0.25
Sugar	10g	NRs. 85/kg	0.85
Lecithin	10g	NRs 1190/kg	11.9

Quantity of peanut butter produced: 1070 g

Expenses required to produce 1.07 kg peanut butter: NRs. 513

Total overhead cost: Rs. 128.25/- (25% of total cost)

Total cost: NRs. 641.25/-

Therefore, price of 1 kg of peanut butter is NRs 600.

Appendix D

Photo Gallery

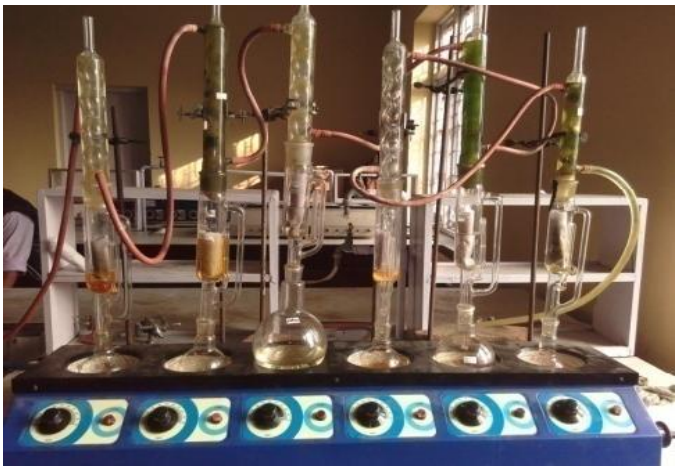

Plate 1: Fat extraction by Soxhlet apparatus

Plate 2: Crude protein determination (Kjeldahl)

Plate 3: Sensory panelist (1)

Plate 4: Sensory Panelist (2)