PREPARATION AND SHELF LIFE STUDY OF HIGH ENERGY BISCUITS

by

ANUJ NIROULA

Food Technology Instruction Committee

Institute of Science and Technology

Tribhuvan University, Nepal

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A dissertation submitted to the Food Technology Instruction Committee in Tribhuvan University in partial fulfillment of the requirements for the degree of B.Tech. in Food Technology

by

Anuj Niroula

Food Technology Instruction Committee

Institute of Science and Technology

Tribhuvan University

Dharan, Hattisar, Nepal

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Tribhuvan University

Institute of Science and Technology

Food Technology Instruction Committee

Central Campus of Technology, Dharan

Approval Letter

This dissertation entitled *Preparation and shelf life study of High Energy Biscuits* presented by *Anuj Niroula* has been accepted as the partial fulfillment of the requirements for the B. Tech. in Food Technology.

Dissertation Committee

1. Chairperson ____________________________

(***Assoc. Prof.*** Basanta K. Rai*)*

2. External Examiner ____________________________

(***Dr***. Atul Upadhyay*)*

3. Supervisor ____________________________

(***Assoc. Prof.*** Pashupati Mishra*)*

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Anuj Niroula

Tribhuvan University

Institute of Science and Technology

Central Campus of Technology

Hattisar, Dharan, Nepal

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List of abbrevations

ANOVA	Analysis of Variance
BHA	Butylated hydroxyanisole
BHT	Butylatedhydroxyltoluene
BOPP	Biaxial oriented Polypropylene
CCT	Central Campus of Technology
EDTA	Ethylenediamine Tetraacetic Acid
EMOPs	Emergency Operations
FAO	Food and Agriculture Organisation
GMO	Genetically Modified Organisms
GMP	Good Manufacturing Practice
GMS	Glycerol Mono Sterate
HDPE	High Density Polyethylene
HEB	High Energy Biscuit
IDD	Iodine Defficiency Disorders
IR-EMOPs	Immediate Response Emergency Operations
LACERN	Latin American and Caribbean Emergency Response Network
LDPE	Low Density Polyethylene
NARC	Nepal Agricultural Research Council
NBS	Nepal Bureau of Standards
NGA	Nordihydro Guaiaretic acid
PG	Propyl gallate
PRROs	Protracted Relief and Recovery Operations
SMBS	Sodium Meta Bi Sulphite
SMP	Skimmed Milk Powder
WFP	World Food Program
WHO	World Health Organisation

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Abstract

High Energy Biscuits (HEB) is small baked bread or cakes that are supplemented with a premix of vitamins and minerals. HEB was prepared with 0, 5, 10, 15, 20 and 25 parts of soy flour with wheat flour to access the quality and acceptability of the biscuit. Thus prepared biscuits were subjected for 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 ($P \leq 0.05$) in overall acceptability among the samples. Sample C (wheat flour: soy flour :: 90:10) got the highest mean sensory score. The best ratio i.e. sample C was used for final preparation of biscuits and thus prepared biscuits were packaged in P1 (laminate PET + BOPP) and P2 (laminate PET + metallic BOPP) and stored at 40˚C. The samples were subjected for sensory analysis and chemical analysis in every two weeks for 12 weeks. There was no significant difference (F pr.>0.05) in mean sensory score of samples at the zero week and last week for the color, flavor, crispness and taste of the biscuit with respect to the packaging material, while the texture and overall appearance were found to have significant difference (F pr.<0.05). The samples showed significant difference (Fpr.<0.05) in all sensory parameters with respect to the time of storage. The acid value (AV), showed no significant difference between the consecutive studies although initial value of 0.12 and the final 0.29 and 0.35 for laminate PET + BOPP and laminate PET + metallic BOPP respectively were different significantly. The peroxide value (PV) showed significant difference after 4 weeks. The final 12 weeks values of AV, and PV were found to be 0.29 and 0.35 mg KOH/ gm oil in laminate PET + BOPP and laminate PET + metallic BOPP respectively where as 1.18 and 1.4 meqv peroxide/kg fat in laminate PET + BOPP and laminate PET + metallic BOPP respectively which was below the standard unacceptable level. The self-life study through the rate of increment of acid value and peroxide value, storage life up to 11.5 months and 15.5 months at 40˚C was determined for sample P1 (laminate PET + BOPP) and P2 (laminate PET + metallic BOPP) respectively. Thus soy flour can be successfully used up to 10 parts with wheat flour. The iron, magnesium, calcium and ascorbic acid content were higher than specified by WFP.

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PART I

Introduction

1.1 General introduction

Food is essential for human beings. People have different taste of foods and different food habits depending upon their religion, ethnic, geographical, climatic, seasonal, educational, economical, rural and urban situations (K.C., 2007). Baking is one of the oldest arts which has a long history and undoubtedly has a long future. It is hard to imagine a world evolving in which bakery does not play a part (Cauvain, 2003). Biscuit is kind of crispy dry bread, more or less hard, variously flavored and usually unleavened, prepared usually in small, flat, thin cakes (Smith, 1972).

Biscuits and biscuit like products have been made and eaten by man for hundreds of years (Tannahill, 1975). Biscuits are a traditional type of flour confectionery which were, and can still be, made and baked in a domestic kitchen. They are small baked products made principally from flour, sugar and fat. They typically have a moisture content of less than 4% and when packaged in moisture proof containers have a long shelf life, perhaps six months or more (Manley, 1983).

During ancient days Biscuit was one of the best and most suitable foods to be carried along during long voyages especially due to its higher shelf life and nutritive value. Those biscuits were called as ship's Biscuit; Walker's dictionary (London, 1848) defines: 'Biscuit is a kind of hard, dry bread, made to be carried to sea as a substitute of bread; it is a composition of fine flour, almonds and sugar.' Based upon these old themes, biscuits have gone through various modifications through new trials till this date. Nowadays various ranges of biscuits with distinct identical qualities and composition are found in the market. Biscuits are the low cost, processed food which offers good taste along with nutritional values at affordable price with convenient availability. Biscuits have in general, a good shelf life in comparison to most of the other snack items. Hence, it's obvious that it is convenient to be used as a travel snacks item (Shrestha, 1995).

High Energy Biscuits (HEB) is small baked bread or cakes that are supplemented with a premix of vitamins and minerals. This ready to eat food participates to the covering of urgent needs in the acute phase of an emergency situation during which population is not able to cook due to a lack of access to basic facilities like clean water, cooking equipment etc.(WFP, 2010). In 2007, WFP launched several projects in support of emergencies like the Peru Earthquake, the Belize and Jamaica Hurricane Dean and Bolivia and Mexico's Floods. In 2008 HEB biscuits have been used in Ecuador, Bolivia and Cuba.

Emergency Operations (EMOPs), covers the procurement and pre-positioning of HEB stocks. The underlying food aid strategy and mode of the implementation are regulated by the regional mechanisms implemented through the LACERN. So, in practice, the HEBs will be drawn down, distributed and replenished through specific interventions (IR-EMOPs, EMOPs, PRROs) which will include appropriate targeting and distribution modalities in their respective project implementation documents.

The name "High Energy Biscuit" itself indicates the richness of energy. Energy could easily be enhanced by producing a high fat content biscuit. But, singly the energy does not suffice HEBs. HEBs should fulfill all the essential nutrients in the acute phase of an emergency, during which population is not able to cook due to a lack of access to basic cooking and drinking facilities. HEBs should meet the technical specifications as specified by WFP.

1.2 Objective of the study

1.2.1 General objective

The general objective was to prepare and study shelf life of High Energy Biscuits.

1.2.2 Specific objective

a. To select the proper amount of soy bean to be used without beany flavor
b. To study the organoleptic changes during storage of HEB
c. To study the shelf life of HEB in various packaging material

1.3 Justification of the work

High Energy Biscuits (HEB) is supplemented with a premix of vitamins and minerals. This ready to eat food participates to the covering of urgent needs in the acute phase of an emergency situation during which population is not able to cook due to a lack of access to basic facilities clean water, cooking equipment etc. (WFP, 2010). HEB use is also extended to a complement food ration i.e. use as snacks to provide vitamins and minerals in regions/population where diet is subjected to nutritional deficiencies. HEB can be used also to prevent micronutrients deficiency of young children and school age children.

However, in Nepal, it is considered as the brand product for army and very little is learned about it by the consumers. There has been lot of controversy in Nepal regarding high energy biscuit and its micronutrient content and energy value. According to Subba R. (Nutritionist, Nepal Army), the armies, who are the daily consumers of High energy biscuit are monotonous of the only beany flavored biscuit produced till in Nepal. So a variety of flavor in HEB is also the present need. It is also noticed to have soya-bean off flavor during its storage and the shelf life is not attained as much as High energy biscuit should have (Personal communication, 2011).

This research was aimed in these two prospects. Soy bean flour was variably used with wheat flour to fulfill the 100 parts of flour, and Butter DRS flavor was used to prepare the high energy biscuit. These biscuits after sensory evaluation to various persons of Nepal Army, a best recipe was finalized and necessary analysis was carried out and packaged in two different wrappers i.e. P1 (laminate PET + BOPP) and P2 (laminate PET + metallic BOPP). These biscuits were analyzed in every 15 days for 3 months and the shelf life was then forecasted.

1.4 Importance

a. A new flavored high energy biscuit was prepared, that was acceptable to the daily users.
b. The shelf life of HEB was forecasted in two different types of wrappers.
c. It provides a duly information about the HEB, which is not known clearly by consumers.

1.5 Limitations of the study

a. Microbial analysis of the product other than total plate count and yeast and mold count could not be carried out due to time constraints.
b. The shelf life and changes in the biscuit during storage could be studied only up to 6 months due to time constraints.
c. The vitamins content in the biscuit after baking could not be analyzed. But use of vitamin premix as prescribed by producer for HEB to achieve desired result was followed.

PART II

Literature review

2.1 Biscuits

The word ‘biscuit’ is derived from the Latin ‘biscoctus’ or old French ‘bescoit’ word (Concise Oxford Dictionary, 1975), meaning twice cooked, a reference to the practice of first baking the product in hot oven and then transferring it to a cooler oven to complete drying process. Biscuits are very popular item prepared on regular basis and these too vary in size, shape, filling and type of recipe used. While they are simple to make, they require care, attention and understanding necessary to produce a standard product (Sultan, 1965).

Generally the term biscuit is used in the European countries and cookies in the USA. Biscuits and biscuit like products have been made and eaten by man for centuries (Hosney, 1986). Biscuits are ideal for nutrient availability, palatability, compactness and convenience. They differ from other baked products like bread and cakes because of having low moisture content, comparatively free from microbial spoilage and long shelf life of the product (Wade and Peter, 1988).

Although the first biscuits were dried-out rusks, useful as long-life food for sea journeys, early cooks making confections with fat and sugar would have found that if little dough pieces are baked in a typical hot oven and taken out when they have a good color and a stable structure they would not have been dry enough to be entirely crisp. Putting them back into a somewhat cooler oven to dry them out improved their eating qualities and also their shelf life. Baking from the start in a cooler oven for a longer period allows drying but results in less coloration and structure development. However, the term biscuit was applied originally to dried bread pieces. These were also sweetened and flavored with spices. Other products like our modern biscuits were made but called by more cake-like names for example: shortcake and shortbread etc. (Manley, 2000).

Now biscuits are made mostly in factories on large production plants. These plants are large and complex and involve considerable mechanical sophistication. Forming, baking and packing are largely continuous operations but metering ingredients and dough mixing are typically done in batches (Manley, 1998).

All flour confectionery is developed from human skills in baking and very much research has been directed to improving our knowledge of the science of what happens when flour is hydrated, mixed with other materials and baked. It is this research that has been the main driving force in the development of the biscuit industry. It is therefore correct to say that without science there is no innovation and without innovation there is no competitiveness (Manley, 2000).

2.2 Classification of biscuits

Biscuits are generally classified as hard dough and soft dough type of biscuits as per the protein composition of the flour used. In case of the hard dough biscuits the flour used should be the weakest possible to obtain and vice versa in case of soft dough biscuit (Smith, 1972). The soft dough group comprises all the sweet biscuits having many factors in common whereas the hard dough biscuits fall naturally into three sections: fermented dough, puff dough and the semi-sweet dough (Whitely, 1971).

2.2.1 Soft dough biscuits

Soft dough biscuits are generally sweet, thin and possess smooth surface with dimensions that are much more regular and consistence than the hard dough biscuits. Generally they contain higher fat content (20%-35%) and sugar (30%-45%) and are low in moisture. The higher gluten network development should be avoided which can be achieved by:

a) Weak flour
b) Lower moisture content
c) Short mixing time
d) Less aerating agents

Soft dough biscuits are less versatile because of the inherent nature of the dough. (Manley, 1983).

2.2.2 Hard dough biscuits

Hard dough biscuits generally contain less fat (10%-20%) and low sugar content (10%-18%). The dough adheres due to its higher water content and relatively lower fat content. This type of formulation produces an extensive gluten structure. The long mixing time develops the gluten and the mixer action stretches and orients the gluten strands to a point where much of the elasticity is destroyed. The water content varies in accordance with the flour strength, which might be as high as 20% of the flour weight (George, 1981). Further according to the variance in composition of one or more parameters hard dough biscuits can be further divided into:

2.2.2.1 Semi sweet biscuits

The flour used in this type of biscuits should be as weak as possible. Its higher water content and relatively low amount of sugar and fat produces an extensive gluten system and structure. Many flour formulations contain cornstarch or arrowroot to an extent of 10% of the flour weight in order to weaken the flour strength. Further to prevent excessive gluten development, a long mixing time followed by addition of sodium meta bi sulphite is carried out. Rapid cooling should be avoided as these types of biscuits are highly susceptible to checking (Dunn and Bailey, 1988).

2.2.2.2 Fermented dough biscuits

This type of biscuits include two groups the cream crackers and the soda crackers. Although these two types have variations within them in case of composition and process of manufacture, both of them have basic mode of production as fermentation. Studies show that the manufacture process of salt crackers is standardized whereas a lot of variations might be seen in the manufacture process of cream crackers. Ingredients commonly include flour of medium strength, protein (9.0-9.5) % , shortenings 12% for cream crackers to 14% for salt crackers, sugar basically is used as yeast food only, salt (2-3)%, malt for rapid fermentation due to its diastase activating effect (Smith, 1972).

2.2.2.3 Puff dough biscuit

Puff dough is special type of developed dough. In the course of dough piece forming a laminated structure must be developed and the layers are separated by soft, semi-solid, plasticized fat. The fat must be kept cool which means that the dough must also be cool or cold. Thus, puff dough must be mixed and must stand so as to maintain low temperatures (usually around 15°C or less). Iced water is used at dough mixing and a chilled room is required to hold the dough and in which to make the laminations (Manley, 1998).

2.3 Types of biscuits

2.3.1 Cream crackers

These have a simple recipe of flour, fat and salt, which is always fermented with yeast and the dough is laminated prior to cutting and baking. These have characteristic flaky and variously blistered biscuits (Manley, 1983).

2.3.2 Soda crackers

It is a square biscuit about 50 X 50 mm and 4 mm thick. The biscuits are produced with scrapless cutters so the edges are white and broken after baking. The fermentation is usually in two stages with a wet sponge lying for 18 hours followed by a dough stage, which is left to ferment for about 4 hours. It has alkaline reaction after baking hence the name soda crackers (Manley, 1983).

2.3.3 Savory crackers

These are variously salted, flavored and fat sprayed after baking. Depending upon their size, because they are made in a very wide range of shapes and sizes, they can be regarded as Savory snacks (Manley, 1983).

2.3.4 Water biscuits and matzos

Water biscuits have a simple recipe of flour, fat, salt and water in the ratio 100:6.5:1:29. The dough is undeveloped and crumbly or in balls after mixing. Matzos are a Jewish product and recipe is about 100 parts of flour to 38 of water (Manley, 1983).

2.3.5 Puff biscuits

Puff biscuits are all made from puffed dough in which there is a non-homogeneous distribution of fat. When this dough is laminated the fat causes discontinuities between the layers of dough and during subsequent baking these layers separated to give a flaky structure. The dough is not fermented and is invariably cold and under developed. Puff biscuits are eaten cold so the fat used must not have waxy tail after eating (Manley, 1983).

2.3.6 Short dough biscuits

These are made from cohesive dough that lacks extensibility and elasticity without the formation of gluten stands from the wheat flour. The texture of the baked biscuit is attributable to starch gelatinization and super cooled sugar rather than a protein or starch structure (Manley, 1983).

2.3.7 Deposited soft dough and sponge drop biscuits

Short dough, which are soft enough to be just pourable, are referred to as soft dough. The biscuits are rich in fat or based on egg whites whipped to stable foam (Manley, 1983).

2.3.8 Wafers

They are formed from a batter, which is baked between pairs of heated metal plates. The majority of wafer biscuits are based on the large flat sheets. The wafer sheet itself is baked from a simple batter containing little or no sugar. It is a tasteless product, which has a smooth surface and a very open cellular structure within (Manley, 1983).

2.3.9 Miscellaneous biscuit-like products

These include crisp breads, pizza bases, sausage rusk etc. **(**Manley, 1983).

2.4 Chemical composition of biscuits

Chemical composition of biscuit varies within the biscuit types due to their difference in the raw material composition, method of preparation, end purpose of the biscuit and various other factors. The major and most common difference between the biscuit types namely hard dough, soft dough and fermented dough biscuit is presented in Table 2.1.

Table 2.1 Chemical composition of biscuit

Type	Protein %	Fat %	Total sugar %	Other carbohydrate %	Moisture %	Salt and chemicals %
Soft dough	6.00	20.80	25.88	44.73	1.25	1.34
Hard dough	7.18	12.26	19.15	59.40	0.90	0.56
Fermented dough	7.20	15.00	7.20	67.00	1.50	2.10

(Source: Rao, 1991)

2.5 Raw materials for biscuit making

Mostly the common raw materials for biscuit making includes wheat flour, water, emulsifiers, sugar, salt. Raw materials can be divided into major and minor ingredients, those raw materials which are used in bulk and are a must for biscuit making are considered as major ingredients. For example, flour, water, sugar and fat are used in bulk in biscuit making procedure. Salt, skim milk powder (SMP), ammonium bi-carbonate, sodium bi-carbonate, coloring agents, flavoring agents, emulsifiers, fortifying agents, improvers etc. are used in small amounts and aren't a must for all sort of biscuits. These ingredients are used for developing the taste, texture, flavor and aesthetic value of the product. Therefore these minor ingredients are also known as the product improvers (Shrestha, 1995).

2.5.1 Major ingredients

2.5.1.1 Flour

Flour is the basic raw material for biscuit making responsible for the major bulk of biscuit (Whitely, 1971). The flour used in biscuit and cracker vary in strength and baking characteristics (Bohn, 1956). Wheat grain is the only grain naturally capable of producing flour capable of being made into a low density baked product (Kent, 1983). Soy flour is used in dough due to its emulsifying property and higher level of protein content (Whitely, 1971).

a. **Wheat flour *(Maida):*** Wheat flour is unique among all the cereal flours in that it forms an elastic mass when mixed with correct proportion of water. This unique property is due to the presence of insoluble proteins, collectively called gluten. The gluten forming proteins (glutenin and gliadin) constitute about 75-80% of the total flour proteins (Mukhopadhyay, 1990). Glutenin gives solidity to the product whilst gliadin is the binding agent imparting the soft sticky character to the gluten. Gliadin is soluble in 70% alcohol and may be extracted from flour where as glutenin is soluble in alcohol and water (Ram and Nigam, 1979).

   Gluten is elastic, cohesive and rubbery and holds together and holds together the various ingredients of the dough. It has the property of holding the gases given off during fermentation and during baking. It sets in oven to form the firm, porous, open texture during baking which are necessary in the production of biscuits and crackers.

Thus gluten is the necessary framework, forming the sustaining wall of the whole structure of baked products (Bohn, 1956).

Wheat flour used for making biscuit should be the product obtained by milling cleaned hard or soft wheat or a combination of both types. Flour strength is usually defined by the percentage of protein present in the flour. Weak flour is casually accepted as the flour with low percentage of protein. Usually this protein is inferred to be gluten, which when the flour is made into a dough with water, will become very extensible under stress, yet when the stress is removed it will not fully return to its original dimensions. Further, the amount of stress required to facture the dough piece is less then that required under identical conditions when strong flour is used (Smith, 1972).

The strong flour protein has long links with few bonds while weak flour protein has short links with many bonds. During biscuit making weak and easy to stretch, soft wheat flour is found to be better (K.C., 1999). Beside the natural quality of flour, the modifications in the flour strength can be done by various treatments. Treatment of the flour with sulphur dioxide reduces the flour strength. Heat treated flour added to untreated flour is claimed to strengthen the flour. According to Kent (1983), improvers have some effect upon the nature and character of the gluten and cause it to behave, during fermentation, like the gluten of the stronger flour. The flour should be free flowing, dry to touch, should be creamy in color and free from any visible bran particles. It should also have a characteristic taste and should be free from musty flavor and rancid taste.

As suggested by Biscuit Bakers Institute book in Biscuit, Crackers and Cookies vol.1 (1972), the protein specifications for cookie flour is mentioned in Table 2.2.

Table 2.2 Protein specification for various flours

Type	Specifications	Protein (%)
Soft cookie flour	General sweet goods	7-8
Medium cookie flour	Cracker dough - up, rich cookies	8-9
Strong cookie flour	Cracker sponge	8.5-10

(Source: Arora, 1980)

The characteristics as required in flour is given in Table 2.3.

Table 2.3 Requirements for flour characteristics

S. No.	Characteristics	Requirements
1.	Moisture content	≤13.0%
2.	Gluten content on dry basis	≥7.5%
3.	Total ash on dry basis	≤0.5%
4.	Acid insoluble ash on dry basis	≤0.05%
5.	Protein (N×5.7) on dry basis	≥9.0%
6.	Alcohol acidity as H_2SO_4 in 90% alcohol	≤0.1%
7.	Water absorption	≥55%
8.	Sedimentation value	≥22%
9.	Uric acid (mg/100 g)	≤10
10.	Granularity	To satisfy the taste

(Source: Arora, 1980)

b. Soy flour

It is advantageous to use soy flour (SF) as a protein supplement and subsequently, research is ongoing in the blending of SF with various cereal grains (Boyacioglu, 2006). Soy flour plays a vital role in balancing the protein deficiency of our diet (Akubor and Ukwuru, 2005).

It is a major source of protein for dietary biscuits. Soya protein does not produce gluten. There are claims that biscuits using about 3 or 4% of soya flour based on the wheat flour content have better appearance, better eating quality and longer shelf life than those made without soya flour (Manley, 1998). TO obtain biscuits of high nutritional and organoleptic qualities, wheat flour could be substituted with 10% flour (Banureka and Mahacevan, 2009). But according to Riaz M. N. (1999), soy flour can be added at high levels (5-20%) in bakery to significantly improve their nutritional value and, at the same time, extend shelf life without adversely affecting spread ratios or other sensory qualities. The disadvantage of using soya flour as a source of emulsifier is that more water is needed in the dough to keep dough consistency at a level to allow good machining on production equipment which can make baking more critical (Stauffer, 2005).

Soy flour are considered an inexpensive source of high-quality protein (38% to 40%) that are abundantly rich in lysine, an essential amino acid, deficient in most cereal grains (Dhingra and Jood 2004; Shogren et al 2006). For these reasons, it is advantageous to use soy flour (SF) as a protein supplement and subsequently, research is ongoing in the blending of SF with various cereal grains (Boyacioglu, 2006). However, successful use of soy flour as low cost protein supplements in biscuit is challenging due to the unpleasant off-flavor compounds found in the soybeans. SPIs, in particular, can exhibit some flavors often considered to be unappealing for consumers, including cardboard and brothy flavors (Russell et al 2006). When added to baked products, SF and isolates contribute beany and astringent notes to bread (King et al, 2001). The beany note is also found in other soy-based products and has also been further characterized to be a raw soy and green flavor (Day N'Kouka et al, 2004). From a chemistry perspective, this beany flavor is associated with volatile compounds that develop due to flavor reversion of soybean oil, a phenomenon observed during oxidation of oils rich in linolenic acid (Liu, 1999). It is the presence of this sensory characteristic in general that is found to be unacceptable to some consumers, presenting a limiting factor for acceptance and widespread use of soybeans in the Western diet (Shogrenet al 2006; Heenan et al 2008). King et al (2001) have demonstrated that a 20% replacement of wheat flour with defatted regular and lipoxygenase free SF leads to significantly higher beany flavor.

2.5.1.2 Fat or shortening

Fat is one of the major ingredients in biscuit making. Shortening function of fat during biscuit making is a must step without which the baked product will be a solid mass held firmly together by strands of gluten (Griswold, 1962). Fat itself being insoluble in water prevents the extra cohesion of the gluten strands during mixing. The greatest attribute a shortening can possess is that it should have a plastic nature over a wide range of temperatures as it is likely to be encountered in its use for biscuit making (Smith, 1972).

The main action of the fat or shortening during mixing is to avoid the gluten forming proteins to come in contact with water by insulating the gluten forming protein molecules due to its hydrophobic nature. Hence, less tough dough with desired amount of gluten formation can be obtained. Thus shortened baked products possess less hard, crispier nature and can easily melt in mouth (Mukhopadhyay, 1990).

During mixing fat also helps in entrapment and retention of the air, which is highly necessary for a product for its good texture. Fat also lubricates the formed gluten molecules to distribute it to various sites during sheeting and hence preventing the agglomeration of the gluten molecules. Fat also plays a vital role in the softness, texture, palatability and keeping quality of the product (Menon, 1988-89).

In the earlier days of biscuit making animal lard was used for biscuit making which has now been totally replaced by hydrogenated vegetable oils. The molecules that exist in oil are built up of unsaturated fatty acid chains, some of which are loosely joined together by double bonds which are weak bonds hence, making the fat prone to easy oxidative rancidity of the oil. During hydrogenation the added hydrogen replaces the double bonds present to convert it to single bonds hence a stable solid fatty acid molecule is formed from weak bonded liquid fatty acid molecule (Smith, 1972).

Hydrogenated Vegetable fat are superior to the lards recently used in various aspects. Further in order to obtain the best product the hydrogenated vegetable oil to be used for biscuit making should possess the following properties:

a. It should possess good white to creamy color.
b. After keeping the fat at 50°c for 24 hrs and filtering, its color should be comparable with the control sample of oil.
c. The fat should have a smooth, uniform texture, free from any oil separation and large grains.
d. The fat should have a bland ‘clear’ odour and taste.
e. The fat should have a wide plastic range to suit particular production techniques and the product.
f. The crystalline structure of fat should be stable during mixing and after baking.
g. The fat should possess reasonable shelf life on its own without the addition of antioxidants.

h. The fat should be prepared from the blend of oils, which will not cause fat bloom during the storage of biscuit.

(Source: Mukhopadhyaya, 1990).

However, only shortening agent allowed in HEB is palm oil. Palm Oil has got natural antioxidant s called tocotrienols which helps in protecting body against ill effects of oxidation, giving long shelf life to the product then others. It does not have trans-fat i.e. zero trans-fat. It is composed of Palmitic acid and oleic acid. Palm oil is naturally semi solid at room temperature (melting point is 39°C) which makes it an excellent shortening for food application. Palmitic acid imparts creamy texture to the shortening which is important for baked products.

2.5.1.3 Sweetening agents

Sugar is another major ingredient in biscuit making. Sugar generally used in biscuit making is obtained from sugarcane and sugar beet. The sugarcane consists of 16-22% of sucrose while sugar beet consists of 8-9% of sucrose. During biscuit making various forms of sugar namely crystalline, pulverized, liquid, brown or soft sugar are used as per product requirement. Generally most commonly used form of sugar in biscuit making is pulverized sugar. The reason behind this may be due to its readily soluble characteristic which causes the palate to be deceived in sweetness. The crystalline size also has effect on sweetness, shortness and better spread capacity of biscuit (Whitely, 1971).

Use of crystal size varies according to the final product. Medium fine powdered sugar, with or without very fine granulation is more suitable for rotary dough; while a coarser sugar is tolerable in hard semi-sweet dough because of the larger quantities of water used, the longer mixing time and higher final dough temperature. Use of coarser sugar gives fissured tops or cracks which is desirable in case of crunches and ginger biscuits. Apart from these sugar types, lactose sugar from milk, and brown sugar which gives both color and delicious flavor to the product also are used.

Another type of sugar used in biscuit making is invert sugar syrup or simply invert syrup. Due to its lower caramelization temperature compared to sucrose the crust coloration of the biscuit takes on a browner appearance much quickly which is desirable in many products. If the crust coloration becomes too darker than requirement then a part of invert syrup can be replaced with glucose syrup. Experiments also show that use of invert syrup also reduces the baking time. Invert syrup also helps in preventing checking problem in biscuits. It also helps in moisture retention in biscuits. As a whole, sugar may be of any form helps in imparting sweetness, increasing tenderness, maintaining volume, crust color development, flavor improvement, moisture retention and proper spread of the biscuit (Smith, 1972).

2.5.2 The minor ingredients

2.5.2.1 Emulsifying agents

Emulsifying agents are surface-active agents promoting the formulation and stabilization of emulsions during biscuit making. It helps proper mixing of lipid and aqueous fraction and helps in maintaining good texture of the product. The unifying property of emulsion is due to the presence of a hydrophobic and a lipophilic group on the same molecule.Various recipes in biscuit making include those from high fat recipe to that of low fat recipe with low water and high water respectively. In the low fat recipes, process problems are associated with gluten development and dough machinability but in high fat recipes, there is more concern for the fat, to give maximum textural effects, dough stickiness and control of spread while baking (Manley, 2000).

Most commonly used emulsifiers are lecithin, eggs, mono and diglycerides etc. In creaming stage where the fat, sugar etc. are combined with all or no part of water, lecithin does exert an emulsifying action to give a smooth homogeneous mixture. Lecithin may be much more useful as an antioxidant also (Smith, 1972).

2.5.2.2 Leavening agents

Leavening are the gassing agents which causes the dough to spring off or puff up to give a porous open texture to the final product. Ammonium and sodium bicarbonate are the major chemical leaveners, while yeasts are the biological leaveners. Similarly, mechanical leavening can be done by incorporating the air within the dough matrix by mechanical agitation. Reaction of two or more chemicals also leads to production and incorporation of gas, mainly the reaction takes place between bicarbonates of ammonia as well as sodium with acidulants. To discuss about the major and most common leavening agent the baking powder, it should possess the following properties:

a. Maximum gas strength-greatest volume of gas for least weight of the product.

b. Proper balance of ingredients to prevent any impairment of the taste or appearance of the biscuit.
c. Innocuous ingredients and residues.
d. Optimum velocity of reaction to be susceptible to control.
e. Keeping quality under diverse and extreme conditions to remain unimpaired over reasonable periods of time.
f. Minimum cost of production, economical in use.

(Source: Smith, 1972)

The chemical reaction during use of chemical leaveners and acidulants is as given below:

$NH_4HCO_3 \xrightarrow{heat} NH_3 + CO_2 + H_2O$

Ammonium bicarbonate Ammonia Carbon dioxide Water

$(NH_4)_2CO3 \xrightarrow{heat} 2NH_3 + CO_2 + H_2O$

Ammonium carbonate Ammonia Carbon dioxide Water

The chemical equations for the reaction of soda and the commonly used acidulants are as below:

$NaHCO_3 + HX \longrightarrow NaX + CO_2 + H_2O$

Sodium bicarbonate Acid Carbon dioxide Water

$NaHCO_3 + C_4H_5O_6K \longrightarrow C_4H_4O_6NaK + CO_2 + H_2O$

Cream of tartar Sod. Pot. Tartarate

Both sodium and ammonium bicarbonate react with acidic ingredients if any, in the dough. Use of an excess ammonium bicarbonate makes the dough more alkaline and thereby may affect the protein structure of the dough. Use of an excess sodium bicarbonate will affect the crumb and crust color often with an accompanying unpleasant or soapy taste, unless any acidic material is used to neutralize the residual sodium carbonate (Bohn, 1956).

2.5.2.3 Milk solids

Milk and milk solids are considered to be the ingredients of value addition during biscuit making. Apart from increasing the nutritive value of the biscuit, milk and milk solids help in retention of flavors. Usually use of milk in biscuit making is done in SMP and full cream form due to its higher stability and easy storage facility. Milk solids when used in biscuit making have proved to enhance crust bloom and color, tenderness and texture without altering the symmetry and crumb color. The coloration may be due to the fact that the lactose in milk solid remains as lactose in the biscuit because it is not fermentable by yeast. Lactose helps in the formation of melanoids, the principle crust coloring substances, formed by the reaction of sugars and amino acids from the proteins under the influence of heat. Probably this reaction takes place in all biscuit dough baking (Smith, 1972).

According to USDEC, skim milk powder is an economical source of milk solids for developed bakery flavors. Flavor development occurs during the baking process when the amine group of the protein reacts with lactose and other reducing sugars as a function of the Maillard browning reaction. The protein and lactose are available for crust color development. High-heat skim milk powder provides structure and body to biscuits, resulting in a light biscuit with good height.

Higher milk flavor can be obtained by the use of condensed milk during biscuit making. Similarly among all the milk products butter is the potent product for better flavor development but due to economical aspect their use have nearly completely been replaced by butter flavors. Other milk products that are also sometimes used are cheese, whey, butter milk etc. (Shrestha, 1995).

2.5.2.4 Salt (Sodium Chloride)

Use of salt during biscuit making is not mainly to increase saltiness except in some salty biscuits. It helps to enhance the natural or other added flavors. Salt can reduce the sourness of acids and increase the sweetness of sugars in their effect in the palate (Fabriani, 1977).

In fermented dough salt helps to develop the gluten of the flour besides acting as a fermentation rate controller. Flours which lack a bit of ageing can be readily used with good gluten fermentation by use of a little bit higher dosage of salt. Salt to be used during biscuit making should be magnesium and calcium chloride free as the minerals may cause rancidity. Use of salt in the range of 1.0-1.5% of the flour weight is thought to be best but above 2.5% it may become objectionable or even nauseous (Mukhopadhyay, 1990).

2.5.2.5 Flavoring and coloring agents

Flavor is the quality of the thing that affects the sense of taste and smell. The majority of the flavors used in biscuit making are derived from natural sources and these are in many ways most satisfactory. To get good distribution in a dough, the flavor should be creamed with the sugar and shortening at the beginning of mixing. Except from the added flavors, flavor can also be obtained from the various ingredients such as nuts, fruits etc. Most commonly used flavoring agents are common salt, yeast, extracts, spices and essences (Whitely, 1971).

Coloring agents are mostly not added externally during biscuit making. Other ingredients like sugar, invert syrup, milk solids etc. provide color to the product mainly due to caramelisation. Coloring agents not only include the synthetic as well as natural colors only but various ingredients used during biscuit making also serve to provide appealing color to the product (George, 1981).

2.5.2.6 Water

Water is one of the most important ingredients during biscuit making. Quality of water used has a great effect on the product. Dissolved minerals and organic matters present in water can affect the flavor, color and physical attributes of the finished baked product (Smith, 1972).

The water used in the baking product should be potable and odorless if required, although no significant effect has been noticed due to the hardness, but demineralization is recommended if the mineral content is too higher which might cause a adverse in product color (Arora, 1980).

2.5.2.7 Anti-oxidants

Anti-oxidants act as a retarding or inhibiting agent in the onset of oxidation rancidity. As biscuit is rich in nutrients and fat content, it is highly prone to oxidative rancidity, so role of antioxidant is essential for prolonging the shelf life of the product. There are a number of naturally occurring substances as well as many man-made chemicals which possess anti-oxidant properties which can be used during biscuit making. Use of antioxidants should be done in the early stage of biscuit making as antioxidants cannot hide or remove the incipient rancidity (Ottaway, 1958).

Most commonly used antioxidants are, BHA (Butylatedhydroxyanisole), BHT (Butylatedhydroxyltoluene), PG (Propyl gallate), NGA (Nordihydroguaiaretic acid). Nearly all the added antioxidants are added with the shortenings for use.

An antioxidant should possess the following properties,

a. Non-toxic
b. Very little or effect on color, flavor or odour of the fat or the product.
c. Be readily incorporated- soluble in fat and oil.
d. Be effective in as low a concentration as possible.
e. Be stable to baking or frying temperatures.
f. Be stable to heat, even in alkaline media, such as biscuit dough.

Apart from all these major and minor ingredients sodium metabisulphite and potassium metabisulphite are used as conditioning agents. Special fortifying agents like protein, vitamins, fruits, nuts, chocolates etc. can also be mixed with biscuit (Smith, 1972).

2.6 General and mandatory requirements of biscuits as published by NBS

Biscuit should be properly baked, crisp and uniform in texture and appearance. They should not possess rancid flavor, fungal infection, off odour and any insect infestation. The mandatory standards as described by Nepal Bureau of Standards (NBS) is given in Table 2.4.

Table 2.4 The mandatory standard for biscuits

S. No	Characteristics	Requirements
1	Moisture	6.00% max
2	Acid insoluble ash (on dry basis)	0.1% max
3	Acidity of extracted fat (as oleic acid)	2.00% max

(Source: NBS, 1993)

For filled biscuits any of the fillers like jam, jellies, marshmallow, cream, caramel, figs, raisins etc can be used. The biscuits may be coated with caramel, cocoa or chocolates. Use of antioxidant as well as permitted preservative can be done not exceeding the maximum dosages.

2.7 Nutritive value of biscuit

Biscuit is a ready to eat good source of nutrient as it contains carbohydrates, fats, proteins, minerals and vitamins. Proteins are nutrients for growth and repair of tissues while carbohydrate and fat provide heat and energy. Similarly minerals provide nutrient for bone growth. Vitamins are responsible for normal metabolic activities and maintaining normal vitality of the body. Nutritive value of biscuit is given in Table 2.5.

Table 2.5 Nutritive value of biscuit (values per 100 g)

Weight per serving in gram	100 g
Calories (kcal)	480
Protein (g)	5.2
Fat (g)	20.2
Carbohydrate (g)	71.0
Calcium (g)	0.04
Phosphorous (g)	0.16
Iron (mg)	1.8
Vitamin A value (I. U)	-
Thiamine (mg)	0.03
Riboflavin (mg)	0.04
Nicotinic acid (mg)	0.8

(Source: Swaminathan, 1991)

2.8 Technology involved during biscuit making

Technology is the factor which enables easy doing of something by significantly reducing the labour, time, expenditure and increasing the quality. Technology is always beneficial until it is under control. Hence the skill to handle the available technology is the fore most need during an operation. Not only the technology controller or his department is responsible with the machinery control but with the total control from ingredient purchase to sales (Rao, 1985).

The technology of biscuit production is shown in Fig. 2.1.

Preparation of ingredients

↓ (Weighing, recipe calculation and fermentation of dough in case of fermented biscuits.)

Creaming and mixing

↓ (Performing is done for hard dough and fermented dough biscuits.)

Laying the dough

↓

Dough forming

↓ (Moulding in case of soft dough biscuit, Sheeting for other types)

Baking

(Temp. 176˚c to 204˚c for 4 to 6 minutes)

↓

Cooling

↓ (For 6 to 7 minutes)

Wrapping and packaging

↓

Labeling and Storage (RH 40 to 59% at room temp.)

(Source: KC, 1999)

Fig. 2.1 Flow sheet of manufacturing process of biscuit

2.8.1 Preparation of ingredients

2.8.1.1 Wheat flour

Flour is taken from the bottom of a silo, or after a sieving system, via rotary seal into a stream of air and is blown to a hopper which is mounted on a weighing system, probably above a mixer.

2.8.1.2 Fat soluble ingredients

GMS, Lecithin, BHA, Sugar and Coconut Powder are to be mixed with melted semisolid/liquid shortening.

2.8.1.3 Water soluble ingredients

Golden Syrup, Glucose Syruph, Malt and SMP are to be mixed with some water together and color (if used) and SMBS both separately in some water.

2.8.2 Metering of ingredients

Metering is probably the most important aspect of process control. Errors in metering may have an effect throughout the rest of the manufacturing process. In most plants insufficient is known about the precision of metering and deviations from standard are not recorded systematically so comparison with biscuit size and quality is difficult or impossible. So, it is important that metering is done properly. In most factories ingredients are metered to mixers by a combination of automatic (for bulk-handled materials) and manual (for small ingredients) methods. (Manley, 2000)

2.8.3 Dough mixing

Mixing is the major step during biscuit making. Properly mixed dough has a great influence in the final quality of the product. Mixing of the dough can be done in various ways as per requirement. Mixing in industries is carried out by use of electrical mixers, most commonly used mixers are two speed mixers. In top speed, the creaming up time is 3-5 minutes in two speed mixer types, while the flour should be mixed for 10 minutes on slow speed (Whitely, 1971).

In the context of biscuit doughs, biscuit sandwich creams and batters, the term ‘mixing’ covers a number of distinct operations. It includes:

a. the blending of ingredients to form a uniform mass
b. the dispersion of a solid in a liquid, or liquid in a liquid
c. the solution of a solid in a liquid
d. the kneading of the mass to impart development of gluten from flour proteins which have been hydrated at an earlier stage of the mixing
e. build up of temperature as a result of work imparted

f. aeration of a mass to give a lower density.

One or more of these actions is required in the formation of dough for the very many types of products that are called biscuits (Manley, 2000).

2.8.3.1 Creaming up method

This method of mixing up of dough mixing includes two steps, during first step the sugar and fat are blended together to fine dissolution after that other ingredients like milk powder, water, invert syrup, lecithin, color, essence, salt are mixed up for around 3-5 min. to form a homogeneous cream. Now the flour along with the aerating agents is mixed up with the cream and mixed at slow speed in the mixing machine for around 10 min. If other type of flour are to incorporated than care must be taken that they must be pre-mixed into the shortenings and the water before adding the other ingredients if the true attribute of thus added flour is to be achieved (Smith, 1972).

This type of mixing method holds the water in a more or less stable state so that it is prevented from making a wide spread attack on the flour to form any significantly higher amount of gluten network. Mostly short cake rotary and wire cut doughs are mixed by this method, in order to control flow and volume during baking. A significant factor in such mixings is the amount of water used (Whitely, 1971).

2.8.3.2 All in one method

As the name suggests, all the ingredients are mixed together and fed in the mixing machine. This method is straight forward where all the ingredients along with major part of water is fed into the mixing machine which some part of water is used to dissolve the aerating chemicals, flavors, colors and salt which is alter on mixed with the dough and is mixed until a satisfactory dough is produced. This type of mixing method is widely applied with hard, semi-sweet doughs. Due to the relatively higher water content in these doughs it results in very satisfactory gluten production and formation.

In fermented dough an important step apart from the above described methods is used which is known as punch back or knock back. This helps to break down the pockets formed by the carbon dioxide during fermentation. The accumulated carbon dioxide might otherwise be poisonous for the yeast cells (Smith, 1972).

2.8.4 Laying of dough

Lay time refers to the halt of the dough between mixing and machining. Lay time for various products is variable. Lay time for fermented dough is highly necessary and is long while in case of sulphited doughs it is avoided. A minimum of 15 minutes of lay off time should be given to the dough for the achievement of good surface gloss, color, and weight. It also makes the dough easily machinable (Bana *et.al*, 1996).

2.8.5 Forming and performing

The shaping or forming of the dough into various shapes and to the required thickness before baking is one of the major steps in biscuit making. In case of hard dough and fermented dough forming is done by using sheeters and laminators, which reduces the thickness of the dough to convert it into a thin sheet. Occluded air is eliminated from the dough. It also helps in proper spread of fat, salts which helps in producing a product with short eating and layered appearance.

Soft dough is directly fed into moulding or embossment disc which cuts them into required size, shape and appearance (Shrestha, 1991).

2.8.5.1 Sheeting, gauging and cutting

Of the various means of forming pieces for baking from a mass of dough, sheeting, gauging and cutting is the most versatile and commonly used method. The integrated set of machines that form dough pieces from a mass of dough are commonly referred to as a ‘cutting machine’. A cutting machine represents a straightforward mechanization of the old manual method whereby a mass of dough was rolled out and then dough pieces were cut with a cutter of the desired shape and size.

Within the sheeter the dough is compressed and worked to remove air and it is inevitable that some stresses are built up in the gluten structure. There is also a small increase in dough bulk density. Flanges at the ends of the rolls prevent the dough being extruded from the ends of the rolls and ensure that the emerging sheet is always of the desired width. The new sheet of dough then passes to one or more sets of gauge roll pairs which reduce the thickness to that required for cutting. Like the sheeter, there are flanges on one of the gauging rolls to prevent the dough extruding sideways and to maintain a full width of dough sheet. Sometimes, having been reduced in thickness, the sheet is folded or cut and piled up to form many laminations before being further gauged to a final desired thickness (Manley, 2000).

2.8.5.2 Rotary moulding

Principally, the dough is forced into moulds which are the negative shape of the dough pieces complete with patterns, name, type and docker holes. The excess dough is scraped off with a knife bearing upon the mould and thereafter the piece is extracted from the moulds onto a web of cotton canvas or other fabric.

Short dough may be sheeted, gauged and cut with an embossing type cutter in a similar way as for extensible dough, but the advantages of a moulder are:

a. It is not necessary to form and support a dough sheet
b. Difficulties of gauging are eliminated
c. There is no cutter scrap dough which must be recycled.

Until recently only short dough could be formed with a rotary moulder. That is because, as is explained below, the scraper knife drags back cohesive dough so the mould is not filled and the resulting dough piece is incomplete (Manley, 2000).

2.8.5.3 Extruding and depositing (Wire cutting)

These basically consist of a hopper over a system of two or three rolls which force the dough into a pressure/balancing chamber underneath. The rolls may run continuously or intermittently and may be capable of a short period of reverse motion to relieve the pressure and cause a suck back at the dies or nozzles at the base of the pressure chamber. This means that dough can be forced continuously or intermittently out of the pressure chamber.

The machine spans the width of the plant and is usually situated over the oven band. In the case of certain drier, wire cut dough and rout types (continuously extruded) which are subsequently cut into lengths before baking, the machine is over a normal canvas conveyor and not the oven band. Dough pieces formed on a conveyor may be spaced out as they are transferred onto the oven band.

Wire cutting gives attractive product appearance but production efficiency is not as good as with other methods of dough piece forming (Manley, 2000).

2.8.6 Baking

The baking and drying of dough is the essence of biscuit making. Baking involves heating the dough. The physics of heat transfer, heat flux, includes convection, conduction and radiation which are difficult concepts to appreciate and evaluate where only the temperature at selected places can be measured in an oven (Manley, 2000). Major part of heat transfer to the dough pieces is by radiation while the heat transfer by convection is very low as long as the air velocity in the tunnel is not higher than 5 feet per second, after which the heat transfer by convection tends to be higher. Apart from these three modes of heat transfer, high frequency heating is also used which has a higher rate of moisture removal (Petty, 1958).

During baking, the product is cooked, flavor and color is developed and the raw dough is converted into an edible snack named biscuit. The main objective of baking is to remove the moisture present in the dough pieces by gradual heating. The dough contains more than 25% moisture, a part of which is bound water present in the flour and other ingredients while other part is the free water added externally for dough making and easy machinability (Dutta, 1985). Every oven used till date consists of four basic parts:

a. A heat source
b. A base (sole or hearth), capable of being heated, on which the dough piece is placed.
c. A cover over the base, making up a chamber in which to retain the heat.
d. A closable opening through which the dough piece can be put into and taken from the baking chamber.

During baking the dough undergoes gradual changes physically as well as chemically.

Physical changes include:

a. Formation of a film crust on the dough.
b. Melting of the fat in the dough.
c. Gas release and volume expansion.
d. Conversion of water into steam.
e. Escape of carbon dioxide, other gases and steam.

Chemical changes include:

a. Gas formation.
b. Starch gelatinization.
c. Protein changes.
d. Caramelisation of sugar.
e. Dextrinization

(Smith, 1972)

Temperature in baking oven has different effect on the raw dough which is shown in Table 2.6

Table 2.6 Temperature related changes in biscuit during baking

Temperature (˚F)	Changes occurred
90-100	Top crust skin formation (Evaporation of surface moisture).
90-120	Evolution of CO_2 within crumb (Less solubility of CO_2).
90-150	Increase in volume due to CO_2.
90-210	Gas expansion (CO_2 and steam).
125-210	Starch gelatinisation (Biscuit structure).
170-190	Evaporation of alcohol, yeast action ceases.
170-250	Evaporation of alcohol, yeast action ceases.
350-400	Coagulation of protein (Irreversible).
370-400	Dextrinization (surface gloss).

(Source: Mukhopadhyay, 1990)

During baking it is necessary to have more steam in the oven than that derived from the moisture from the dough and the combustion of the fuel. Introducing steam into the baking chamber, either immediately at the entry of the dough pieces or at a point very early in their passage through the oven, helps to create a shiny crust formation, prevention of cracked crusts, increased volume and to some degree agitation of the oven atmosphere. The need of steam injection can be removed by using fast moving fans recirculating air at speeds of 2000 cu ft per minute. The dampers present at the ovens play a vital role in releasing the high positive pressure within the oven created due to high heat evaporation, similarly if high moisture cookies or biscuits are desired than the dampers at the last zone must be closed (Smith, 1972).

2.8.7 Cooling

Cooling is one of the most important part of biscuit production. As biscuits emerge from the oven they are very hot nearly at a range of 210-214˚F, very soft and moist. Therefore cooling of the biscuit prior to packaging must be done to maintain the proper structure of the biscuits. Immediately after the biscuits are released from the oven they possess a very high moisture content which signifies that the flour starch is still in some form of gelatinous paste and the dextrin still in partial solution. Similarly the sugar as well as fat will also be in its liquid form, protein although firmer than other ingredients is also pliable. Hence, almost all ingredients are in unset state.

Cooling helps in consequent loss of moisture and slowly sugars start to crystallise out and the dextrin grow tougher, then only the biscuit grow tougher and set. Cooling should be gradual and slow (Dutta, 1985).

Checking is the most common defect which occurs in the biscuits after they are prepared and not noticeable during processing. Checking refers to the hair like structure which might refer to weakening of the structure and breakage might occur after 24 hrs of packaging. Hard, semi-sweet biscuits are prone to checking rather than rotary moulded soft types which is due to its low fat and low sugar content leading to higher gluten development. As gluten has higher affinity towards water, it will extract it from the gelatinised starch present in the hot biscuit and so cause stresses to be set up. This problem is supposed to be further aided by rapid shrinkage of the biscuit due to rapid cooling. Hence, checking can be reduced or eliminated if the baking is slow and the cooling is gradual rather than rapid (Manley, 1983).

2.8.8 Secondary processing

In many cases biscuits which have been baked and cooled are subjected to further treatment in the form of coatings or sandwiching with flavored materials. These treatments are known as secondary processes. The additions include chocolate, fat-based creams, water icing, marshmallow, caramel toffee and jam or jelly. There may be a single addition such as chocolate or a combination of two or more materials. In the latter case each addition is usually made separately and a cooling or drying period allowed between each.

Secondary processing allows a much greater variety of flavors, textures and appearance to be achieved than by baking alone. The additions may result in a biscuit becoming a confectionery product and the materials used are more akin to the sugar confectionery industry than flour confectionery. It may be that chocolate enhances a biscuit or a biscuit fills out and enhances a chocolate product.

In-line secondary processing is not always the case and in many factories baked and cooled biscuits are collected in tins or trays to be coated or sandwiched, etc., at a later time.(Manley, 1983).

2.8.9 Packaging

Biscuits are low moisture content food. Their mandatory standards state them to be of low moisture content, mainly below 6%. The relative humidity of freshly baked biscuit is very low so in order to prevent rapid uptake of moisture from the atmosphere, the biscuits must be packed in a water vapor resistant packaging material. Packaging materials are those materials which contain the product within them providing necessary conditions and protection to the product inside to keep them safe and consumable over a long period of time. Packaging in case of biscuits must be close up together in order to provide a mutual reinforcing effect which prevents them from breakage. Packaging of biscuits at commercial level is generally done in triple laminates consisting of polyethylene, aluminum foil and paper (Paine and Paine, 1983).

The primary pack is the moisture proof unit which is to be offered for sale to the consumer. Secondary packaging into groups of 10, 20 or more in boxes or cases is for ease of storage and transportation, but as this packaging may have a significant effect on the mechanical protection afforded to the primary packs, it should be designed carefully (Manley, 2000).

2.8.10 Sensory perception of biscuits

Before launching the biscuit in the market, a panel of experts evaluate the overall acceptability of the final product which is known as sensory perception or evaluation. Many basis of evaluation include appearance, crispiness, crumb color, flavor and finally the overall acceptability. The evaluation is marked on the score cards which is later on discussed in order to find out the best product. Consumer research, case history research is also some of the factors that need to be included while drawing conclusions from the analysis (Dawadi, 2008).

2.9 Product specifications of High energy biscuits

2.9.1 Nutritional and micronutrient value:

As per WFP/WHO (2011), HEB shall contain following nutritional value per 100g dry matter:

Table 2.7(a) Nutritional value in HEB per 100g.

Paramaters	Value per 100g
Energy	450 kcal min
Moisture	4.5 % max
Protein	10-15 g (N*6.25)
Fat	15 g min
Sugar	10-15 g max
Fiber	2.3 g max

Table 2.7(b) Micronutrient rate and Chemical form

Micronutrient	Target	Chemical Form
Vitamin A-Retinol	250 mcg	Palmitate 250 CWS
Vitamin B1	0.5 mg	Thiamine mononitrate
Vitamin B2	0.7 mg	Riboflavin
Niacin	6 mg	Nicotinamide
Pantothenic acid	3 mg	Calcium d-pantothenate
Vitamin B6	1 mg	Pyridoxin hydrochloride
Folic Acid	80 mcg	Folic acid
Vitamin B12	2 mcg	Vitamin B12-1% in Manitol
Vitamin C	20 mg	Ascorbic acid coated
Vitamin E	1.9 mcg	Vit D3 100 CWS
Vitamin D	5 mg	Vit E 50 CES
Calcium	250 mg	Calcium Carbonate
Magnesium	150 mg	Magnesium Oxide
Iron	11 mg	Ferrous fumarate
Iodine	75 mcg	Potassium Iodate

The variation of the final product with respect to contents of fat shall not minus 5% and the moisture and fiber should not exceed 5% of the specified values. The permitted variation in premix content is -10 to +15 % for added vitamins and +/- 10% for added minerals.

Note: Variable levels of micronutrients (i.e. iron, zinc, calcium etc.) naturally present in raw materials may lead variable of micronutrients in finished product.

2.9.2 Microbiology

The microbiological contamination level for HEB is mentioned in Table 2.8

Table 2.8 Limit of microorganisms in HEB

Microorganisms	Maximum levels
Mesophyllic aerobic bacteria	10,000 cfu per g
Coliforms	10 cfu per g
Escherichia coli	0 per 10 g
Salmonella	0 per 25 g
Staphylococcus	0 per 10 g
Bacillus cereus	10 per g
Enterobacter sakazakii	0 per 10 g
Yeasts and moulds	100r g

Source: (WFP/WHO, 2011)

2.9.3 Safety requirements

HEB shall be free from objectionable matter, not contain any substances originating from micro-organisms or any other poisonous or deleterious substances, heavy metals or pesticide residues, in amounts which may represent a hazard to health (Table-2.9).

Table 2.9 Safety parameters

Toxins	Value
Arsenic (As)	< 0.10 ppm
Cadmium (Cd)	< 0.10 ppm
Lead (Pb)	< 0.02 ppm
Mercury (Hg)	< 0.20 ppm

Pesticide residues	
Carbamate	< 10 ppb
Organochlorine	< 10 ppb
Organophosphorus	< 10 ppb
Pyrethroid	< 10 ppb
Melamine	2.5 mg/kg max

Source: (WFP/WHO, 2011)

2.9.4 Additional requirements

a. Organoleptic: HEB shall have a pleasant smell and palatable taste.
b. Weight: One biscuits should weigh between 5g and 10 gm.
c. Shelf Life: HEB shall retain above qualities for 24 months from date of manufacture when stored dry at ambient temperatures prevalent in the country/state of destination.
d. Other Requirements
   - Free from radioactivity
   - Free from Genetically Modified Organisms (GMO) (if required by the contract)

   (WFP/WHO, 2011)

2.10 Raw materials specifications for HEB

2.10.1 Main ingredients

HEBshall be manufactured from fresh and good quality, free from foreign materials, substances hazardous to health, excessive moisture, insect damage and fungal contamination and shall comply with all relevant national food laws and standards (WFP/WHO, 2011)

Requirements for the raw materials are:

a) Wheat flour should conform to Codex STAN 152-1985.
b) Soy flour/soy protein should conform to Codex STAN 171-1989 (for soy) or Codex STAN 175-1989 (for soy protein).
c) Sugar should conform to Codex STAN 212-1999.
d) Skimmed milk powder should conform to Codex STAN 207-1999.
   - It shall be accompanied by a ‘melamine-free’ certificate.
   - Maximum level aflatoxin M1: < 0.5 mcg/kg milk (recommended methods ISO

14501/IDF 171:20071 or ISO 14674/IDF 190:20052).

e) Additives

- Lecithin shall be in proportion as specified in the Codex STAN 074-1981.
- Raising (Soda) agent as specified in the codex STAN 074-1981, the maximal value is determined by the GMP principles.
- Shortening: the only shortening agent allowed is palm oil as per codex STAN 074-1981.
- Artificial flavoring agents are not allowed except ethyl vanillin and vanillin: 7 mg/100 g

Raw materials must be stored under dry, ventilated and hygienic conditions. Only safe insecticides (i.e. phosphine) may be used for fumigation control. Where needed, fumigation must be performed by certified operators, as per SOP for contracted fumigation /WFP, revised Nov. 2003.

2.10.2 Vitamins and minerals

Complete premixes must be purchased from a WFP approved supplier. The composition of premix is presented in product specification. Suppliers of WFP's micronutrient premixes are DSM, Fortitech, Nicholas Piramal and Hexagon Nutrition or their authorized dealer (addresses of suppliers are in annex) Micronutrient premixes must be delivered to the processor of HEB with a complete Certificate of Analysis as well as with a Proof of purchase of premixes. The two documents must be presented with other documents for payment. Premixes must be stored in a dry, cool and clean place where the temperature is a maximum of 25ºC (WFP/WHO, 2011).

2.11 Packaging of HEB

To be used for WFP, HEB should be packaged in strong cardboard cartons suitable for multiple handling, individual packages of 100 g each or 250 g each (individual packages of 100 g or 250 g each packed in metalized laminate OPP 20micron/PR 3C/DRY/VMCPP 25micron), 100 of these or 40 of those respectively to be stuffed in one carton box of 10kg with necessary marking (WFP/WHO, 2011)

N.B. about 15-20 bags of silica gel of at least 1kg each should be placed in each container in order to absorb moisture. In addition craft paper should be laid to all sides of the container. Individual Biscuit Packaging should include the following Information (WFP/WHO, 2011):

a. Production date (month/year)
b. Best before date (month/year)
c. Nutritional information per 100g
d. "This product contains no lard"
e. "Not for individual sale"
f. "World Food Program + WFP logo"

2.12 Marking specification of HEB

Cartoons shall be marked at seller's expense with the following information, in letters measuring 1.0 to 1.5 cm on the cartons:

a. Net weight
b. Month and year of production
c. Full name or code of the production enterprise
d. Ingredients, nutritional information
e. WFP logo (in letters of 5 cm), or the Contract authority name/logo.

2.13 Supplementation in HEB

Supplementation is the term used to describe the provision of relatively large doses of micronutrients, usually in the form of pills, capsules or syrups. It has the advantage of being capable of supplying an optimal amount of a specific nutrient or nutrients, in a highly absorbable form, and is often the fastest way to control deficiency in individuals or population groups that have been identified as being deficient (Allen et al, 2006).

Vitamins are essential organic compounds that the body requires to maintain life and which it cannot synthesize itself, meaning they have to be in the food supply. Vitamins are either water-soluble (vitamin C and all the B vitamins) or fat soluble (vitamins A, D, E and K) (Wesley and Ranum, 2004)**.** Most of the vitamin losses ocur during baking, which is the most common process all wheat flour products go through. Although baking temperatures are high (over 200°C), the temperature inside the product is significantly lower, and over 70 percent of the vitamins remain unaltered (F. Hoffmann - La Roche, Unpublished Data, Basel). Minerals supplemented are calcium, magnesium, iron and iodine. Supplementation in HEB is carried commonly in the form of premixes. Supplements in HEB are described as follows:

2.13.1 Vitamin A

In vivo, this vitamin is generally found as the free alcohol or esterified with a fatty acid. The vitamin is available in pure form by chemical synthesis as vitamin A palmitate or the acetate, or recovered from molecularly recovered fish oil. It is a yellowish oily material which may crystallize into needlelike crystals (Parman and Salinard, 1981). Stable forms of vitamin A palmitate have existed for at least 30 years and have significantly increased the number and kind of foods that can be fortified with vitamin A, particularly cereal grain products. Commercial forms of dry vitamin A as palmitate or acetate are available embedded in a water-soluble matrix (e.g., gelatin, gum acacia, starch) and stabilized with antioxidants (*Klemm et al*., 2010).

Provitamins which are then converted to their active form, serve not only as nitrifying compounds but also as colorants and anti-oxidants. The most common of these is beta-carotene. Vitamin A is quite stable when heated to moderate temperatures in the absence of oxygen and light. Overall loss of activity during anaerobic heating may range from 5-50%, depending on time, temperature and nature of the carotenoids (Tannenbaum *et al*. 1987). In the presence of oxygen and light, there can be extensive loss of vitamin A activity through oxidation. The presence of trace metals accelerates this reaction.

In dehydrated foods, vitamin A and provitamin A are highly susceptible to loss by oxidation (Labuza *et al*., 1978). The extent of this loss depends on the severity of the drying process, protection provided by packaging materials and conditions of storage. Vitamin A in pure form is unstable in the presence of mineral acids but stable in the presence of alkali.

Naturally occurring vitamin A is insoluble in water but soluble in oil. In this form the vitamin has limited applicability. Vitamin A fortificants are commercially available in a wide range of forms adapted for use under various conditions. For mixing with dry products, a dry form of the fortificant was required with the appropriate size and density. Encapsulation of the vitamin in a more hydrophilic coat is commonly practiced in order to achieve a more water dispersible product. Two materials used in encapsulation are gum acacia and gelatin. These dry forms of the vitamin are also stabilized using tocopherols or phenolic antioxidants (Clarke, 1995).

2.13.2 Vitamin B1 (Thiamine)

Vitamin B_1, or thiamine, is a white crystalline solid with a characteristic yeast-like odor and a slight bitter taste. Thiamine is produced by chemical synthesis as the hydrochloride and mononitate salts. The hydrochloride is soluble to the extent of 50% in water as compared with 2.7% for the mononitrate (Bailey, 1991).

Thiamine is one of the most unstable vitamins. Its stability to heat and oxidation is greatest at a pH range of 6 and below. At higher values of pH it becomes increasingly unstable. Thiamine is susceptible to nucleophilicattack, therefore it is degraded by some mineral salts in aqueous foods (Clarke, 1995). Thiamin can be added as either thiamin mononitrate or thiamin hydrochloride. The mononitrate form is preferred because it is considered more stable. Both are white powders and add no color to the flour. There are no known functional problems in adding thiamin and the cost of thiamin fortification is not very high (Wesley and Ranum, 2004). Thiamine hydrochloride is the fortificant of choice in cases where dissolution in aqueous media is required. In most other cases the mononitrate is used due to its lower hygroscopicity. Thiamine is also commercially available in a coated form using mono- and di-glycerides of edible fatty acids (Clarke, 1995). However, antioxidant systems will help vitamin B1 remain heat stable (Baking Management, 2005)

2.13.3 Vitamin B2 (Riboflavin)

Vitamin B_2, riboflavin, is of an intense orange color and low water solubility. A commercially available more water soluble form of this vitamin is the sodium salt of riboflavin 5'-phosphate. Riboflavin is generally stable under most processing conditions, but is unstable in alkaline medium. It is very sensitive to light, particularly in the presence of ascorbic acid (Clarke, 1995).

2.13.4 Vitamin B6 (Pyridoxine)

Pyridoxine hydrochloride is a white powder and is not known to cause any problems when added to cereals. While whole grains are good sources of this vitamin, but refined wheat flour are not. But because it is found in a variety of foods, overt B6 deficiency is uncommon (Wesley and Ranum, 2004).

There is some suggestion that pyridoxine, along with folic acid and vitamin B12, can help lower homocysteine levels and thereby reduce the incidence of heart disease and stroke (Duell 1997; Jacques 2001; Kelly 2003).

Vitamin B_6, pyridoxine, is available commercially as the hydrochloride. Coated forms are also available as with all of the B-vitamins. This vitamin is quite stable to heat and atmospheric oxygen and heat, but degradation is catalyzed by metal ions (Clarke, 1995).

2.13.5 Vitamin B 12 (Cyanocobalamine)

Cereals contain no vitamin B12. It is present only in animal products. Deficiencies occur mainly in the elderly. The main justification for adding vitamin B12 is so that high levels of folic acid can be added without risk of masking B12 deficiencies (Ray, 2000) but it is also implicated along with some other B vitamins in reducing serum homocysteine levels (Bower, 1995).

Cyanocobalamin, the most important compound with vitamin B_{12} activity, is commercially available as a crystalline, dark red, hygroscopic powder. Human requirements for this vitamin are very low and it is commonly sold highly diluted by a carrier. In the preparations sold by Hofmann-La Roche, for instance, it can be purchased diluted with mannitol or a mixture containing modified starches, citrate, citric acid, benzoate, sorbic acid and silicon dioxide. The selection of preparation depends, of course, on the end use. In solution it is most stable between pH values 4-7. It is unstable to oxidizing and reducing agents and exposure to sunlight, but is fairly stable to heat (Bailey, 1991).

2.13.6 Niacin (Vitamin B 5)

Niacin is low in refined flours .is relatively expensive and could be added at a lower rate to wheat flour, or even excluded from flour fortification programs in wheat consuming populations. One reason for excluding Niacin is tryptophan, an amino acid in proteins that acts as a niacin precursor. When tryptophan content is considered even refined wheat flour becomes a fairly good source of niacin (Wesley and Ranum, 2004).

Niacin in the form of either nicotinic acid or nicotinamide can be used in nutrient addition to foods. At very high levels, nicotinic acid has been shown to cause unpleasant side effects such as flushing and 'pins and needles'. This has led to some preference for nicotinamide. Both forms of the vitamin are stable to atmospheric oxygen, heat and light in the dry state as well as in solution (Clarke, 1995).

2.13.7 Folic Acid

Folic acid is a yellow-orange, odorless, tasteless crystalline substance. It is moderately stable to heat and atmospheric oxygen. In neutral solution it is quite stable, but instability increases with a shift in pH in either direction. Folic acid is unstable to heat, light, sunlight, oxidizing and reducing agents (Clarke, 1995).

There is growing evidence that folic acid fortification will reduce the incidence of elevated homocysteine levels (Jacques, 1999), considered a major factor in cardiovascular disease and strokes. A study in Chile showed that the national folic acid fortification program reduced serum homocysteine levels in the elderly (Hirsch 2002).

To avoid any possible risk of adverse effects, folic acid fortification should be designed to limit regular daily intakes to a maximum of 1000 µg. Folic acid should be used in combination with vitamin B-12, especially in those countries with large proportion of elderly or where the consumption of eggs and milk products is low (Allen *et al*., 2003)

2.13.8 Pentothenic Acid

Pantothenic acid is a pale yellow, viscous, hygroscopic liquid which is very unstable. The most commonly used commercially available form is calcium pantothenate. This is a slightly hygroscopic white powder with no smell but a slightly bitter taste. Stability of this compound is greatest at pH values between 5 and 7(Clarke, 1995).

2.13.9 Biotin

Biotin is a white crystalline powder of low water solubility. It is generally commercially available in diluted form as the physiological requirement for this vitamin is so low. Hoffmann-La Roche sells a 1 % mixture of this vitamin with di-calcium phosphate di-hydrate. Biotin is fairly stable to heat, air and light (Clarke, 1995).

2.13.10 Vitamin C

Vitamin C or ascorbic acid is an odorless, white, crystalline compound which is stable in its dry form. Due to its high water solubility, losses due to leaching can be a problem in some processing procedures. Ascorbic acid is readily oxidized. In dehydrated citrus juices the degradation is dependent on both temperature and water activity. Other factors as well can influence the degradation behavior of vitamin C, these include salt and sugar concentration, pH, oxygen, metal catalysts and ratio of ascorbic: dehydroascorbic acid.

Vitamin C addition to foods is commonly practiced for reasons other than fortification. Commercially available forms of this vitamin include the free acid and the sodium and calcium salts of these, in powder as well as crystalline or granular form. For mixing with dry products, particle size and density are of course important considerations. A fat coated form of ascorbic acid is also available for enrichment purposes. Ascorbyl palmitate is a form of the vitamin used for purposes other than fortification. It is used as an antioxidant in fats and oils and has also emulsifying properties (Anon, 1985). Other areas of food processing for which vitamin C has application are the prevention of browning in fresh and canned fruit and vegetables, acidification, curing of meat and prevention of haze formation in brewed products (Borenstein, 1987).

2.13.11 Vitamin D

The principal forms of the vitamin are D_3 and D_2. They are white, crystalline fat-soluble vitamins, formed by irradiation of the appropriate sterol followed by purification procedures. These compounds are sensitive to oxygen and light, with the D_3 form of the vitamin being slightly more stable. Trace metals such as Cu and Fe act as pro-oxidants.

As with vitamin A, commercially available forms include fat-soluble crystals for use in high fat content foods, and encapsulated, stabilized versions of the fortificant, suitable for use in dry products to be reconstituted with water. As was stated for vitamin A, at the levels of water activity which exist in dehydrated foods, these fat-soluble vitamins are most susceptible to oxidative loss (Clarke, 1995).

Vitamin D is an important regulator of calcium metabolism. People who are exposed to sunlight will not need much vitamin D, but adult deficiencies have been shown to exist, with a higher prevalence in older subjects (Yan, 2000).

2.13.12 Vitamin E

Vitamin E is a slightly viscous, pale-yellow oily liquid obtained from molecular distillation of by-products from vegetable oil refining or by chemical synthesis. The naturally occurring form of the vitamin is the d-isomer. The synthetic compound is a racemic mixture of the d and 1 isomers. The 1-isomer doesn't have the full biological activity of the d-isomer, but due to the stability of the racemic mixture and the ease of purification, the IU of vitamin E has been defined as 1 mg dl-a tocopheryl acetate (Clarke, 1995).

Refined palm oil contains about 350-450ppm vitamin E, present as the RRR-tocopherol (30%) and tocotrienol (70%) isomers. In contrast, other oils such as corn, soyaand sunflower are good sources of the tocopherols but contain no tocotrienols (Sundram, 2003).

Vitamin E content in Crude palm oil is 708–1141ppm and Refined palm oil is 378–890ppm (Gapor 1990). The free alcohol form of the vitamin is highly unstable to oxidation and is therefore widely used in foods as an antioxidant to stabilize the lipid component of foods. Esterified forms of the vitamin, commonly the acetate, are much more stable. For this reason, fortificants are usually of this form. As with the other fat soluble vitamins, cold water soluble forms have been produced by encapsulation within a suitable matrix (Clarke, 1995).

2.13.13 Iron

Iron compounds used in food fortification are commonly classified according to their solubility. Selection of an appropriate iron fortificant for any given application is based on the following criteria: organoleptic considerations, bioavailability, cost and safety (Hurrell and Cook, 1990).

The use of more soluble iron compounds often leads to the development of off-colors and off-flavors due to reactions with other components of the food material. Infant cereals have been found to turn grey or green on addition of ferrous sulphate. Off-flavors can be the result of lipid oxidation catalyzed by iron. The iron compounds themselves may contribute to a metallic flavor. Some of these undesirable interactions with the food matrix can be avoided by coating the fortificant with hydrogenated oils or ethyl cellulose.

Bioavailability of iron compounds is normally stated relative to a ferrous sulphate standard. The highly water soluble iron compounds have superior bioavailability. Bioavailability of the insoluble or very poorly soluble iron compounds can be improved by reducing particle size. Unfortunately this is accompanied by increased reactivity in deteriorative processes.

Sodium iron EDTA is less well absorbed than ferrous sulphate from foods which contain few inhibitors to absorption. In the presence of these inhibitors, however, the EDTA complex is better absorbed. Sodium iron EDTA also participates to a lesser extent in deteriorative reactions. The use of this compound reduces the problem of precipitate formation in foods such as fish sauces and tea. The use of this compound is not advised in developed countries where the population already receives close to the recommended acceptable daily intake of EDTA (Hurrell and Cook, 1990).

The problem of low bioavailability of some of the less reactive forms of iron is often circumvented by the use of absorption enhancers added along with the fortificant. Examples of such enhancers are ascorbic acid, sodium acid sulphate and orthophosphoric acid.

2.13.14 Iodine

Iodine fortification is usually reserved for salt. There are no countries that currently require iodine to be added. However, there can be situations where salt iodization is not adequate and additional measures are needed to prevent the occurrence of IDD. Biscuit is normally made with 2% salt on a flour basis. This has been reduced in recent years due to concerns on high sodium levels, but it normally stays above 1.5%. U.S. standards of iodized salt is77 mg/kg (Wesley and Ranum, 2004).

The most commonly used compounds in the iodization of foods are the iodides and iodates of sodium and potassium. These are the additives allowed by Codex Alimentarius in the iodisation of salt. The iodide compounds (Bauernfeind, 1991) are cheaper, more soluble and have a higher iodine content (so that less is needed to achieve the same level of iodisation) than the corresponding iodates. Iodates are more stable under conditions of high moisture, high ambient temperature, sunlight, aeration and the presence of impurities. The use of iodate is therefore recommended for use in developing countries. Potassium iodide is well suited in cases where the salt is dry, free from impurities and has a slightly alkaline pH. Otherwise the iodide may be oxidized to molecular iodine and lost through evaporation. If excess water is present the iodide may be separated from the salt in the water film (FAO/WHO, 1991). Loss of iodide can be reduced through the addition of stabilizers such as 0.1% sodium thiosulphate and 0.1% calcium hydroxide combined or 0.04% dextrose and 0.006% sodium bicarbonate.

Calcium salts have been used with some report of off-flavor due to the calcium ions (Kuhajek and Fiedelman, 1973).

2.13.15 Calcium

The main calcium sources used in cereal fortification are calcium sulphate (gypsum) and calcium carbonate (limestone). Both are white and bland in flavor. Calcium sulphate is produced from mined gypsum by a precipitation process and is available either as the dihydrate with 23% calcium or the anhydrous form with higher calcium content (27%) but generally a higher price as well. Calcium carbonate used in cereal fortification is normally made by grinding limestone mined from very pure deposits.

There is a considerable variation in the particle sizes available, from very fine to coarse.Because of the large bulk of the calcium source that needs to be added, it is not normally included in the fortification premix and its addition is done separately from the rest of the fortification. Calcium fortification has no effect on the color or taste of flour or bread, even at the high levels used.

Calcium carbonate has a slight pH raising and buffering action on flour. Added calcium is generally believed to be beneficial for yeast-leavened baking. Calcium fortification will greatly increase the ash content, making ash levels unusable as a way of measuring quality or extraction when enriched in flour (Wesley and Ranum, 2004).

2.13.16 Magnesium

Magnesium does not appear to be severely limiting in most diets. However, provision of calcium supplementation of foods without magnesium is highly controversial. A ratio of 4:1 or lower (by weight) of calcium to magnesium has been advocated. Although the overall risk and benefit of this addition cannot be evaluated at this time, the addition of 40–60 mg/d of magnesium to a supplement (as a food fortificant) is very unlikely to have side effects. Until further data are available demonstrating low magnesium intakes or inadequate magnesium status, the addition of magnesium to a complementary food supplement would be a safe but not a necessary step (Abrams and Atkinson, 2003).

2.14 Packaging

Packaging is defined as “a coordinated system of preparation of goods for shipment, distribution, storage and marketing at optimum cost, compatible with the requirement of the product” (Kennedy, 1955). Packaging is an essential part of processing and distributing foods. Whereas preservation is the major role of packaging, there are several functions for packaging, each of which must be understood by the food manufacturer (Potter and Hotchkiss, 2006).

According to Paine and Paine (1983), definitions of packaging;

a. A coordinated system of preparing goods for transport, distributions, storage, retailing and end-use.
b. A means of ensuring safe delivery to the ultimate consumer in sound condition at minimum cost.
c. A techno-economic function aimed at minimizing costs of delivery while maximizing sales (and hence profits).

2.14.1 Food packaging materials and forms

There are relatively few materials used in food packaging; metal, glass, paper and paperboard, plastics, and minor amounts of wood and cotton fiber. However, within each of these categories many types of packaging materials or combination of materials are available. In the case of polypropylene film alone, there are dozens of types of films and laminates varying in moisture permeability, gas permeability, flexibility, stretch, burst strength, and so on. Often, a new food product requires its own special package since optimum protection, economic considerations, and merchandising requirements change rapidly with variations in product composition, weight and form, and performance demands (Potter and Hotchkiss, 2006).

According to Potter and Hotchkiss (2006), packaging material are found in a wide variety of forms including the following: Rigid metal cans and drums; flexible aluminum foils; glass jars and bottles; rigid and semi rigid plastics can and bottles; flexible plastics made from many different films used for bags, pouches, and wraps; paper, paperboard, and wood products in boxes, pouches and bags; and laminates or multilayer in which paper, plastic, and foil are combined to achieve properties unattainable with any single component.

2.14.2 Types of container

The most commonly used containers for packaging of food products are: Pouches, Cartoons, Metal cans, Rigid plastic containers, Papers, Paperboard, Plastic materials like Cellophane, Polyethylene, Plastic films, Polypropylene, Laminating films, foils etc (Shah, 1993).

2.14.3 Special feature required for biscuit packing

According to Paine and Paine (1983), a good packaging material for biscuit must be:

a. Resistant to water vapor.
b. Non- tainting material with good grease resistance.
c. Should be strong enough to protect against any possible mechanical injury.
d. Should be opaque.
e. Easily printable.

The characteristics of the packaging materials are described in Table 2.10.

Table 2.10 Some characteristics of packaging materials

Component	Properties
Low density polyethylene	Moisture and vapor barrier, heat sealing medium
Paper	Stiffness, low cost, opacity, printable
Aluminium foil	Opacity, good water vapor and gas barrier
Oriented polyethylene terepthalate	Gas barrier, strength, grease resistant
PVC	Transparency, rigidity, gas barrier

(Source: Robertson, 1974)

The fat content is generally 15%. It is very susceptible to oxidative rancidity. HEB further is rich in oxidation sensitive vitamins and minerals. So the consideration that has to take in mind is the prevention from oxygen and light. Here is short description about these aspects of the products.

2.14.3.1 Reducing oxygen in the package

Open access to the atmosphere (oxygen) makes biscuits prone to oxidative rancidity as fat is a major ingredient used during biscuit making. It is the esthetic deterioration (undesirable characteristic change in flavor and odor). Destruction of essential fatty acids and development of toxic compounds may accompany rancidity. If the film is opaque, the light induced damage is essentially nil. Oxidative rancidity will develop at a rate which depends on the percentage of oxygen in the head space. Oxygen level above 2% in head space will be sufficient to allow rancidity development. For controlling this effect, inert filling is practiced e.g. nitrogen, carbon dioxide or vacuum pack is profitable for long time storage (Shah, 1993).

2.14.3.2 Reducing moisture vapor transfer

Moisture is the most important agent for hydrolysis of fat of the product. Up take of moisture by biscuit also makes them prone to microbial attack. So, the moisture barrier type material should be necessary. This property is simply tested by weighing the sample at certain interval and gain or loss in weight is observed, it indicates bad barrier properties (Shah, 1993).The moisture barrier properties of some important materials are shown in Table 2.11,

Table 2.11 Moisture barrier properties of some important materials.

S.N.	Film	Water vapour transmission rate (g/100in sq./24 hr/mm thickness)
1	Aluminum foil	0.01-0.02
2	Oriented polypropylene	0.2-0.4
3	Nitrocellulose-coated cellophane	0.7-1.0
4	Polymer coated cellophane	0.5-0.8
5	Unoriented polypropylene	0.6-0.9
6	Low Density Polyethylene	1.0-2.0
7	Polymer coated oriented poly propylene	0.3-0.4

Source: (Shah, 1993)

2.14.4 Plastics as packaging materials

The share of plastics in the packaging market has been growing at remarkable pace, partially replacing paper, glass, and metal. Because of their unique combination of properties, plastics have expanded the packaging industry to sophisticated levels. Plastics containers are light weight, breakage resistant, transparent, flexible, squeezable, moldable in complex shapes, easily colored and printed, retortable, sterilizable, reusable, and recyclable. Plastics have many positive tradeoffs within their array of versatile properties, including easy processing, good mechanical properties, large range of processing temperature, lowest density among packaging materials, and (for better or worse) they are permeable materials. In addition, plastics are economically competitive in cost with paper, glass, steel, and aluminium (Hernandez, 1997).

The British standard Institution has defined plastics as "a wide group of solid composite materials which are largely organic, usually based on synthetic resins or upon modified polymers of natural origin and possessing appreciable mechanical strength." At a suitable stage in their manufacture most plastic can be cast, moulded or polymerized directly to shape (Athalye, 1991).

Plastics can be divided into two main sub groups; Thermoplastics and Thermosets. Thermoplastics are those materials which can be heated and cooled repeatedly without Appreciable loss of mechanical and physical properties. Thermosets, on the other hand, soften once on the application of sufficient heat but harden on cooling (Athalye, 1991).

2.14.4.1 Thermoplastics

Among the very wide range of Thermoplastics commercially available today those that are used in large quantities in the field of packaging are given in Table 2.7;

Table 2.13 Thermoplastics in packaging

S.N.	Polymers	Major forms
1	LDPE	Films, lamination (extrusion) of paper foils etc.
2	HDPE	Films, woven sacks
3	Blow moulded products	Injection moulded products such as crates.
4	Polypropylene	Oriented PP Biaxially oriented PP (BOPP)
5	Metalized BOPP	Box strapping
6	Blister packaging	Corrugated boards
7	Polystyrene	Injection moulded containers and closures
8	Injection blow molded bottles	Thermoformed cups, tubes
9	Biaxial oriented films	Foams

Source: (Athalye, 1991).

2.14.4.2 Thermosets

These are rarely used in the field of packaging except in a small way for threaded closures made out of phenol formaldehyde and urea formaldehyde (Athalye, 1991).

Some of the distinct advantages that plastic materials offer (Athalye, 1991) are;

a. Plastics are light and less bulky than other packaging materials.
b. They have excellent barrier properties to moisture, odor, oxygen and other gases so that they can maintain the desired shelf-life for various products.
c. They are resistant to most chemicals that are non-toxic in nature and absolutely safe to use even in direct contact with food products, medicines etc.
d. Plastics can be processed into any desired shape or form such as films, sheets, bottles, tubes, pouches, crates etc.
e. They are safe in use as they do not break easily and the broken pieces are not harmful as those of glass and metal.
f. They save costs of storage and transportation because of lower volume and less secondary packaging.
g. They do not promote any bacterial growth and can be sterilized by all conventional methods and hence provide wide application in food, medical and chemical packaging.
h. Single serve packs for food items such as ketchups, condiments etc and small unit packs can be made available at low costs.
i. Plastic packages are temperature proof, break resistant, corrosion resistant and leak proof.
j. Plastic packaging does not possess any major disposal problems or environmental hazards since almost all of plastics can be recycled for reuse.

Although plastic packages have tremendous advantages, they have some limitations (Athalye, 1991). These include;

a. Some chemicals do attack particularly on some plastics.
b. They are not a total barrier to gases and water vapor, although some new barrier
c. Plastics have greatly improved oxygen, gas and odor permeability.
d. Abrasion resistance is not always adequate.

e. When stretched PVC/PET is used instead of glass for bottles, consumers may get the impression that there is less quantity because of reduced thickness.

2.14.5 Selection of proper packaging materials

Description of packaging materials is very complicated subject because in the modern development of plastic technology additional developments in design, use, merits and demerits are added day by day. Detail information is not possible here so on the basis of requirement of the product and the availability of packaging material and other technical constraints, only two types of packaging materials are preferred. So plastic packages that is going to be used in the work is described here in short, they are;

a. Poly Ethylene Terephthalate (PET)
b. Biaxial oriented Polypropylene (BOPP)

2.14.5.1 Poly Ethylene Terephthalate (PET)

PET has good sparkle and clarity. Density ranges from 1.38-1.395 gm/cc. Due to its high softening point of 245-270ºC, it performs well over a wide range of temperature ranging from -60 to +150 ºC, so it can be used for boil in the bag. It is not good for heat sealing and if this is required, it is usually coated with a layer of PE, BOPP etc. Film thicknesses range from thinner than 12 mcg for most polyster films to around 200 mcg for laminated composites. Main properties of PET are:

a. Good barrier properties
b. High Strength
c. Grease Resistance
d. Low and high temperature performance

(Kirwan and Strawbridge, 2003)

2.14.5.2 Biaxially oriented polypropylene (BOPP)

BOPP is the lowest density polymer (0.90g/cc) and extremely versatile because of its excellent process ability, mechanical and physical properties and high distortion resistance. Main properties of BOPP are,

a. They are good resistance to strong acid and alkalis.
b. Not affected by most solvent at room temperature.

c. It resists oil and grease and does not crack under any condition.
d. Good barrier of moisture and gases.

(Kirwan and Strawbridge, 2003)

For best WVTR and barrier properties, the preferences are (Athalye, 1991);

a. Aluminum foil and metallized films for the best barrier
b. PVDC and EVOH for the best non-foil barrier
c. PP, HDPE, LDPE for good protection
d. Polycoated paper for limited protection
e. Paper or permeable cellulose film for allowing the product to breathe.

2.14.6 Safety of food packaging

2.14.6.1 Migration from plastics

It is important to know that plastics are not completely inert to foods. Aside from the permeation of gases and vapors, it is also possible that components of the plastics can migrate to the food and would then be consumed with the food. This raise concern for the safety of some plastics. For this reason, all plastics used in food contact must have specific approval from regulatory agencies for the intended use. Food manufacturers must get written assurance from the plastic manufacturer that their container wrap meets all requirements for use in food contact (Potter and Hotchkiss, 2006).

2.14.6.2 Contamination

It is primarily which acts as a barrier to contamination of foods. Preventing recontamination of thermally processed low-acid food which are stored at room temperature is especially serious. Recontamination with pathogens bacteria such as *Clostridium botulinum*can lead to outbreaks of food-borne disease. One example occurred when fish had been processed in defective metal cans which contained small holes. Several people ended up with botulism, which is often fatal (Potter and Hotchkiss, 2006).

2.15 Shelf-life determination

The shelf-life of the product in the package under specified conditions can be confirmed by several methods, viz., (1) weight gain or loss method, (2) method based on testing the performance of the product, and (3) chemical (acid value, peroxide value) changes during storage. All these tests are related to water vapor and oxygen permeability of the packaging material, which in turn indicates the increase of acid value and peroxide value of the product overtime. All shelf-life assessment methods use accelerated and controlled conditions so that an accurate prediction of shelf-life can be possible within a short time (Kumar, 2001).

Peroxide value and free fatty acid tests are analytical methods typically used for fats and oils. If the fat must first be extracted from food products in order to run these tests, the following statements must be true to ensure valid results (Sewald and DeVries, 2000).

1. The fat extracted must be representative of the fat in the food.
2. No non-fat compounds, which would interfere with the test, should be extracted with the fat.
3. No active fat compounds can be either produced or destroyed during the extraction process.
4. The solvents used must be free of any active substances.

Storage temperature conditions should then be chosen which fit the product and give reliable results in a reasonable amount of time. Common temperatures used would be 20, 30, 40, and 55°C (68, 86, 104, and 131°F). A control, stored at 0°F, can also be used. The frequency of the analytical testing is the next important decision. The higher the storage temperature, the more frequent should be the testing. Weekly tests are common for most products (Sewald and DeVries, 2000).

PART III

Materials and methods

3.1 Raw materials

Himal chap wheat flour in the form of *maida* used for biscuit making was obtained from Salt Trading Co-operation, Sundhara, Kathmandu. Soy flour was obtained from local flour mill. Both wheat and soy flour was shifted to a 44 size mesh. Malaysian red palm oil, Malaysia was used as shortening agent. *Mayur* Brand Sugar obtained from Mahalaxmi Sugar Pvt. Ltd. ,Adarsanagar, Parsa, in the form of pulverized sugar was used. *Rahul* Skimmed milk powder (SMP) of Chitwan Milk Pvt. Ltd, Chitwan was used. Vitamin Premix and Mineral Premix produced by Hexagon Nutrition was used. Other raw materials like common salt, invert syrup, sodium and ammonium bi-carbonate, lecithin and Flavor were used from the regular store. P1 (laminate PET + BOPP) (12+25μ) and P2 (laminate PET + metallic BOPP) (12+25μ) were used for the packaging of the product.

3.1.1 Basis of formulation

The preparation of high energy biscuits was done on the basis of minimum (basic) nutrient requirement as specified by WFP (2006). Table 2.7 (a) and (b) shows the basic nutrient requirement for HEB.

3.1.2 Calculation of amounts of ingredients

For the formulation of HEB, the amounts of ingredients were calculated on dry weight basis. Legumes were taken as the source of protein, the cereals as the staple source. Mineral salts for mineral source, Sugar as a sweetening agent as well as flavoring agent, Ascorbic acid as the source of vitamin C, Oil as a source of energy and to maintain the level of essential fatty acids.

Amount of vitamin and mineral premix was added to achieve vitamins and mineral required level in HEB. Finally, from the calculation three formulae mixes was developed.
The recipe formulation for the HEB was carried out as given in Table 3.1.

Table 3.1 Recipe Formulation for High energy biscuit

Ingredients	Quantity (Parts)	A	B	C	D	E	F
Wheat flour	100	100	95	90	85	80	75
Soya flour (SF)	0, 5, 10, 15, 20, 25	0	5	10	15	20	25
Fat	18	18	18	18	18	18	18
Pulverised sugar	14	14	14	14	14	14	14
Milk powder (Full cream)	2.5	2.5	2.5	2.5	2.5	2.5	2.5
Salt	1.15	1.15	1.15	1.15	1.15	1.15	1.15
Sodium bicarbonate	0.9	0.9	0.9	0.9	0.9	0.9	0.9
Ammoniuim bicarbonate	2	2	2	2	2	2	2
Soya lecithin	0.2	0.2	0.2	0.2	0.2	0.2	0.2
Vitamin and Mineral Premix	0.2	0.2	0.2	0.2	0.2	0.2	0.2
Ascorbic acid	0.02	0.02	0.02	0.02	0.02	0.02	0.02
Flavor	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Water	10	10	10	10	10	10	10

3.2 Method of preparation

Raw materials were weighed as per the calculated recipe as given in Table 3.1. Then after creaming up of shortening, pulverized sugar, syrups, milk powder, part of water, lecithin and half of buckwheat flour was done till the mixture appeared as a creamy mass. The mixing was done in mixing machine at top speed for 3-5 minutes as per requirement. Addition of buckwheat flour early in the mixture was to enhance its naturally borne emulsifying capacity as described by Smith (1972).

Now to the creamy mass remaining buckwheat flour, wheat flour and chemical leaveners dissolved in water was added. Now the remaining water was also added and the mixing was carried out on two speed mixer on low speed for 10 minutes. The adequacy of mixing was checked by studying the adhering nature of the dough in the dye.

Now, thus prepared dough was left for 15 minutes for laying, in order to achieve maturity of dough for easy machinability and good surface gloss as described by Bana *et.al* (1966). The matured dough was then fed in the embossing dye or moulders directly.

The dough was then conveyed to continuous baking oven where it was baked for 5 minutes with the last damper open. Baked biscuits were now cooled for 20 minutes and then packed in poly propylene bags and stored in a cool and dry place for further analysis. The standard baking temperature profile set for the study was as given in Table 3.2.

Table 3.2 Baking temperature profile of the oven

Zone No.	1	2	3	4	5
Top Heat (ºC)	230	240	240	250	200
Bottom Heat(ºC)	230	250	250	210	200
Damper Position	Half- Open	Half-open	Closed	Closed	Closed

The method of preparation of biscuit was carried out as given in Fig 3.1.

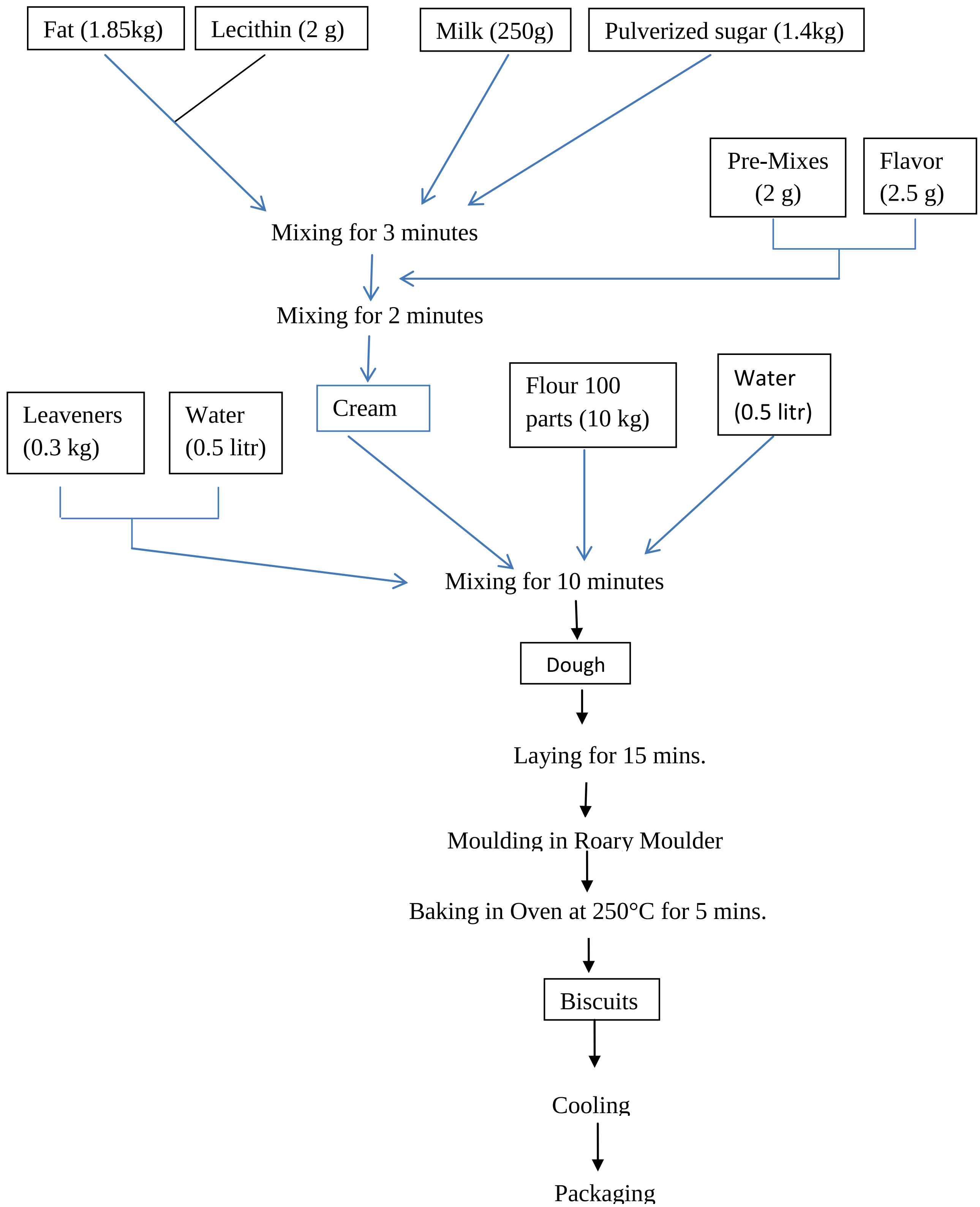


Fig 3.1 Flow chart for the production of biscuit

3.3 Evaluation of prepared HEB

3.3.1 Sensory evaluation

Sensory evaluation was performed by 9 point hedonic scoring test (9 = like extremely, 1= dislike extremely) for color, texture, flavor, taste and overall acceptance from 0 weeks to 12 weeks in every 2 weeks. The evaluation was carried out by 10 panelists comprising of members of Nepal Army, each of them the regular consumer of HEB. Sensory evaluation was carried out in individual both with adequate light and free from obnoxious odors. Each panelist was provided with 5 samples coded random numbers and evaluation card (Appendix A). They were provided with potable water for rinsing between the samples. Verbal communication among the panelist was prohibited. They were asked to evaluate the samples individually using score card.

3.3.2 Physiochemical analysis of product.

3.3.2.1 Moisture content

Moisture content was determined using hot air oven (Ambassador, working temperature 0 to 300ºC, UK) as per Ranganna (2001).

3.3.2.2 Crude protein

Crude protein was determined by Micro Kjeldahl method as per Ranganna (2001).

3.3.2.3 Crude fat

Fat content in the product was determined by soxhlet extraction method as per Ranganna (2001).

3.3.2.4 Total ash

Total ash was determined by ashing in electric muffle furnace (Ambassador, working temperature-900ºC, UK) as per Ranganna (2001).

3.3.2.5 Crude fiber

Crude fiber was determined as per Ranganna (2001).

3.3.2.6 Total carbohydrate

Total carbohydrate was determined by difference method.

Total carbohydrate (%) =100-(moisture + protein + fat + crude fiber + ash) %

3.3.2.7 Calcium

Calcium content was determined by precipitation method as per Eagan *et al*. (1981).

3.3.2.8 Magnesium

Magnesium content was determined by Spectrophotometric method as per Ranganna (2001).

3.3.2.9 Iron

Iron content was determined by Spectrophotometric method as per Ranganna (2001).

3.3.2.10 Vitamin C

Vitamin C content was determined as per K.C. and Rai (2007).

3.3.3 Evaluation of microbiological quality

3.3.3.1 Total plate count (TPC)

TPC was carried out using the method of AOAC (2005).

3.3.3.2 Yeast and mold

Yeast and Mold was determined according to AOAC (2005).

3.3.5 Shelf life prediction

Following chemical and sensory analysis were carried out in each interval of 15 days.

3.3.1 Chemical analysis

3.3.1.1 Acid value

Acid value was determined according to Ranganna (2001).

3.3.1.2 Peroxide value

Peroxide value was determined according to Ranganna (2001).

3.4 Cost calculation

The cost of the best HEB formulations was calculated including a profit of 10% (Appendix D).

3.5 Data analysis

Data on sensory analysis were tabulated for comparison and were graphically represented using Microsoft Excel-2007. Data were statistically processed by GenStat Discovery Edition 3, GenStat Procedure Library Release PL15.2, Version 7.22 DE (Copyright 2008, VSN International Ltd) for Analysis of Variance (ANOVA). Means of the data were separated whether they are significant or not by using LSD (least significance difference) method at 5% level of significance.

PART IV

Results and Discussion

The wheat flour and soy flour were collected and mixed with other ingredients to formulate HEB of 0, 5, 10, 15, 20 and 25 parts of soya flour to fulfill 100 parts of flour. Finally, from the calculation six formulae mixes was developed on the basis of nutrient requirement as specified by WFP, from the used raw material. Proximate composition of the flour as well as biscuit was carried out. The best product among the six variations was determined by carrying out sensory evaluation and the detailed nutritional value of the best product was analyzed. The proximate composition of soya flour as well as wheat flour was obtained as given in Table 4.1.

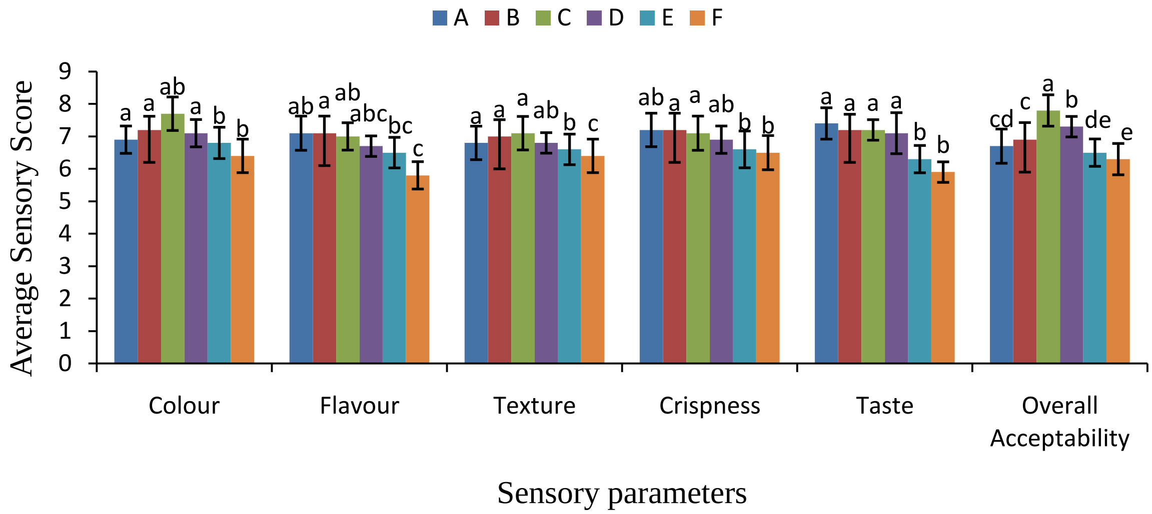
Table 4.1 Proximate composition of wheat and soy flour

Parameters in %	Wheat flour*	Soy flour*
Moisture	12.80 (0.38)	5 (0.45)
Crude protein	9.90 (0.25)	43.15 (0.59)
Crude fat	1.50 (0.23)	0
Crude fibre	0.45 (0.16)	9.6 (0.49)
Total ash	0.51 (0.31)	4.56 (0.69)
Carbohydrate	75.29 (0.45)	21.05 (1.1)
Gluten content	9.10 (0.59)	0

The values are the means of triplicate samples and the values in the parenthesis are standard deviation.

4.1 Organoleptic quality of the products

The prepared six HEB formulas were subjected to sensory evaluation. The samples were provided to 10 semi trained panelist. The semi trained panelists evaluated for various parameters of HEB namely, color, flavor, texture, crispness and overall acceptability. The panelists were requested to provide scores in the score sheets as per their perception. Data were analyzed statistically and best product was found out. The statistical representation of the sensory analysis is given in figure 4.1.



The similar alphabet above the error bars indicate that the samples are not significantly different ($p>0.05$) and the error bar represents the standard deviation.

Fig. 4.1 Average sensory score for six different formulas

The ANOVA at 95% level of confidence ($p > 0.05$) showed that the formula- A, B, C, D, E and F were significantly different from each other in sensory attributes.

4.1.1 Color

The average sensory score for color was 6.9, 7.2, 7.7, 7.1, 7.0 and 6.6 for sample A, B, C, D, E and F respectively. The analysis of variance showed that in case of color, sample A, B, C and D showed significant difference with sample E and F ($p>0.05$).

The soy enrichment in biscuits had significant effect in color, as was observed by Akubor and Ukwuru (2005). The color of the biscuit changed from creamy to dark brown, with an increase in sensory score from 6.9 to 7.7 and then decrease up to 6.6, when soy flour was enriched from 0% to 25%. The biscuit with 10 parts soy flour had the highest sensory score. The darker color may be due to Maillard reaction between reducing sugar and protein (Dhingra and Jood 2000). In this case, an increase in protein content, especially the higher lysine, from soy flour probably caused the darker crust color (Bertram, 1953).

4.1.2 Flavor

The average sensory score for flavor was 7.1, 7.1, 7.0, 6.7, 6.5 and 5.8 for sample A, B, C, D, E and F respectively. The analysis of variance showed that in case of flavor, sample A, B and C and sample E and F showed significant difference with each other while sample D showed significant difference with sample F only ($p>0.05$).

This is similar to the findings of Ayo et al (2007).who observed no significant difference on sensory score for flavor on increasing the soybean flour up to 15 parts level indicating better flavor rating, and thereafter it decreased at 20 and 25 parts level soy flour enrichment. Flavor of biscuit decreased from 7.1 to 6.4 with increase in proportion of soy flour from 0 parts to 25parts. This could be due to the beany flavor of soy flour (Grewal, 1992).

4.1.3 Texture

The average sensory score for texture was 6.8, 7.1, 7.0, 6.7, 6.5 and 6.4 for sample A, B, C, D, E and Flavor respectively. The analysis of variance showed that sample B and C and sample E and F showed significant difference ($p<0.05$), however sample A and C were not significantly different with either of the samples ($p>0.05$).

This is similar to the findings of Akubor and Ukwuru (2005) who observed that there were no significant differences in texture of biscuit up to 10 parts soy enriched and un-enriched biscuits. The texture of the crust was related to the external appearance of the biscuit top which implies smoothness or roughness of the crust. The texture of crust was decreased from 7.1 to 6.4 with the increase in proportion of soybean flour from 5 parts to 25 parts in the biscuits. There was increase in texture of biscuit from 0 parts to 10 parts soy flour enrichment and then decreased when soy flour proportion was increased as obtained by Ugwuona (2009). This may be due to increase in percentage of protein with increase in proportion of soy flour, that results in lower dough hydration and less consistency of dough resulting crumbly biscuit as observed by Maache-Rezzoug et al (1998).

4.1.4 Crispness

The average sensory score for crispness was 7.1, 7.3, 7.1, 6.9, 6.6 and 6.5 for sample A, B, C, D, E and F respectively. The analysis of variance showed that in case of crispness, sample

A, B and C and sample E and F showed significant difference with each other. However, sample D was not significantly different with any of the samples ($p>0.05$).

The sensory score showed no significance difference in crispness, with soy flour up to 15%, however gradual decrease in the crispness was observed. This might have been due to the effect of the oil in the soy flour (Oluwamukomi et al, 2010).

4.1.5 Taste

The average sensory score for taste was 7.4, 7.2, 7.2, 7.0, 6.3 and 5.9 for sample A, B, C, D, E and F respectively. The analysis of variance showed that in case of taste, sample A, B, C and D and sample E and F showed significant difference with each other ($p>0.05$).

This is similar to the findings of Banureka and Mahendran (2009) who observed that there were no significant differences in taste of biscuit up to 15 parts soy enriched and un-enriched biscuits. The score for taste is decreased from 7.4 to 6 with the increase in the level of substitution of soy flour. Biscuit containing 25 parts soybean flour was rated poorest in taste. The biscuit with 0 parts soy flour has the highest mean value and 25 parts soy flour added biscuit has the least mean value. This change in taste may be due to combined effect of Maillard reaction between reducing sugar and protein (Dhingra and Jood, 2000) and beany-flavor of soybean (Rastogi and Singh, 1989).

4.1.6 Overall acceptability

The average sensory score for overall acceptance was 6.7, 6.9, 7.8, 7.3, 6.6 and 6.4 for sample A, B, C, D, E and F respectively. Sample C was significantly different from other samples and had the highest sensory score.

As obtained by Banureka and Mahendran (2009), from the overall acceptance rating, 10% soybean flour incorporated biscuit obtained the highest preference compared to others. At this level of soy flour incorporation, biscuits had higher scores for all the sensory attributes evaluated. Above and below this level, biscuits received lower sensory scores with the least score at 25 parts soy flour incorporation. This may be because of higher sensory scores for other sensory attributes at this level. The higher levels from 10 parts of soy flour let to the increase in the acceptability of the product declined due to compact texture of the crumb and the strong flavor (Dhingra and Jood, 2004).

However, biscuit with 15 parts soy flour was also moderately liked and was acceptable as was found by Ayo et al (2007).This is because the soy and wheat protein interact by covalent and non covalent bonds producing more soluble protein aggregates yielding better sensory profile (Pablo et al.,2005).

4.2 Analysis of the best product

The chemical analysis of the best product (Product C) found from sensory analysis was carried out. The result is tabulated in Table 4.2.

Table 4.2 Analysis result of HEB.

Parameters	Amount
Moisture (%)	0.97 (0.15)
Protein (% db)	12.25 (0.19)
Fat (% db)	15.55 (0.52)
Ash (% db)	0.06 (0.02)
Crude fibre (% db)	2.08 (0.2)
Sugar (%db)	13.26 (0.35)
Total Carbohydrate (% db)	58.17 (0.73)
Energy (Kcal./100g)	538.43 (18)
Calcium (mg/100g)	280 (11)
Ascorbic Acid (mg/100g)	22.25 (0.19)
Magnesium (mg/100g)	154.54 (13)
Iron (mg/100g)	13.02 (0.54)

The values are the means of triplicate samples and the values in the parenthesis are standard deviation.

4.3 Microbiological quality of product

Total Plate Count (TPC) and Yeast and Mold count of the product as received by the microbiological assay are shown in Table 4.3.

Table 4.3 Microbiological assay of the products

Parameters	cfu/g
TPC	<1000
Yeast & Molds	48

4.4 Conformance of best product with WFP specification

The analytical results of the best product showed conformance with the WFP specification. However, some toxicological test analysis and vitamin analysis could not be performed due to laboratory and time constrictions.

4.5 Shelf life evaluation of the HEB

The shelf life of the HEB was studied for 12 weeks. The product was packed in P2 (laminate PET + metallic BOPP) and P1 (laminate PET + BOPP) bags and stored at room temperature. The acid value and peroxide value of extracted fat of the product was evaluated from the date of manufacture up to 12 weeks, and sensory evaluation was done in each week till 12th week to check the acceptability.

4.5.1 Changes in acid value

In general, the acid value is the indication of free fatty acid content in the product. The increment in the free fatty acid of the product was found increased with storage time and also depends on packaging material. After twelve weeks of storage, the acid value increment was found minimum in P2 (laminate PET + metallic BOPP) while maximum oxidation of fat obtained in P1 (laminate PET + BOPP). The acid value (AV) of the product was observed to be 0.12 at initial which reached 0.29 in P2 (laminate PET + metallic BOPP) and 0.35 in P1 (laminate PET + BOPP) within 12 weeks. The value of extracted fat acidity obtained was far below the unacceptable level of maximum 1 mg KOH/ gm oil till the last date of analysis. The changes in the acid value is shown in figure 4.2.

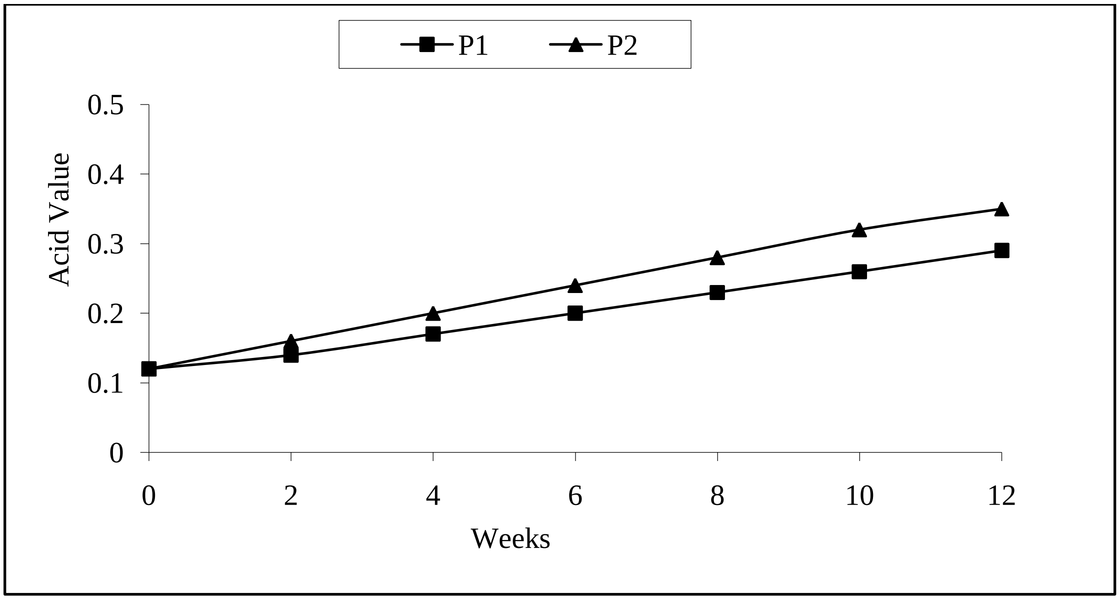


Fig. 4.2 Changes in acid value during storage

From the result of statistical analysis (Appendix C), variation in packaging material had no significant effect on acid value but storage time had significant effect on acid value of product. According to Hamilton (1983), rancidity can be the subjective organoleptic appraisal of the off flavor quality of food. Rancid off-flavors are concerned with the changes that result from reaction with atmospheric oxygen i.e. oxidative rancidity or by hydrolytic reactions catalyzed by lipases from food or from microorganisms. The change of acid value increment of the sample could easily be evaluated by Fig. 4.2.

4.5.2 Changes in peroxide value

Peroxide value is a very sensitive indicator of the early stages of oxidative deterioration of fats and oils. PV therefore provides a means of predicting the risk of the development of flavor rancidity. After twelve weeks of storage, the peroxide value increment was found minimum in P2 (laminate PET + metallic BOPP) while maximum increment of peroxide was obtained in P1 (laminate PET + BOPP).

The peroxide value (PV) of the product was observed to be 0.87 at initial which reached 1.18 and 1.4 in P2 (laminate PET + metallic BOPP) and P1 (laminate PET + BOPP) at 40ºC respectively within 12 weeks. The value of PV obtained was far below the unacceptable level of maximum 3 meqv peroxide/kg fat. The change in peroxide value of the product is shown in Fig. 4.3.

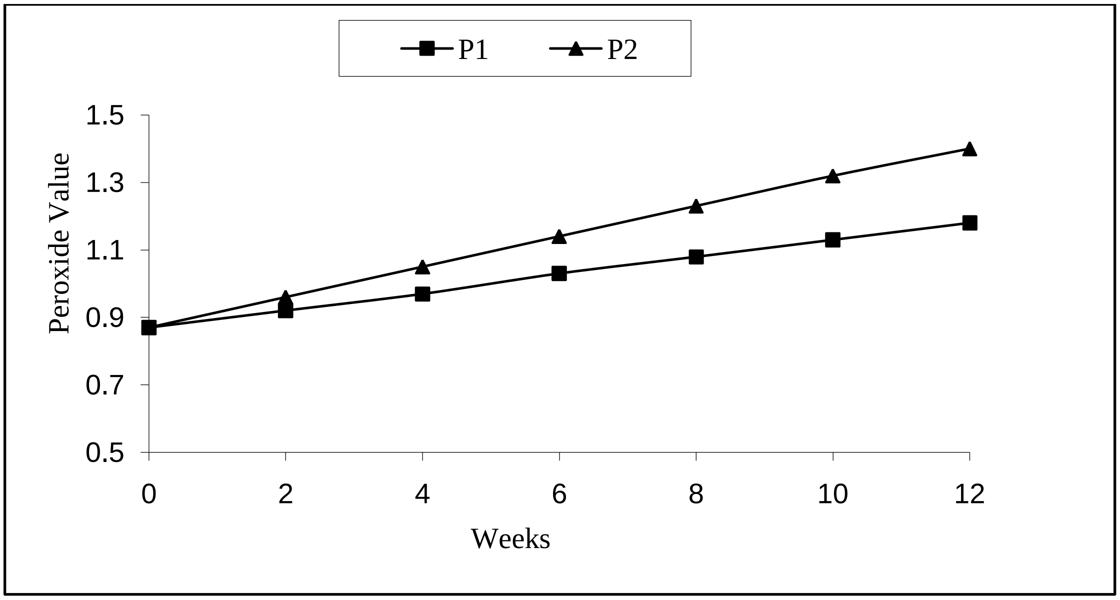


Fig. 4.3 Changes in peroxide value during storage

According to Hudson (1983), peroxide values are very sensitive indications of the early stage of oxidative deterioration. Though peroxides and hydro-peroxides of oils and fats are themselves tasteless, their presence is a sure indicator of flavor deterioration to come, since they are comparatively unstable. They will break down inevitably at ambient temperature to yield a range of off-flavors in the form of small molecules, especially carbonyl compounds.

4.5.3 Sensory analysis

4.5.3.1 Changes in color

Average sensory scores given by the 10 panelists were tabulated in Table C.1.9 and Table C.1.10 and the changes in color of the product was used for the comparative study between two packaging materials stored at 40ºC temperature with increase in storage time.

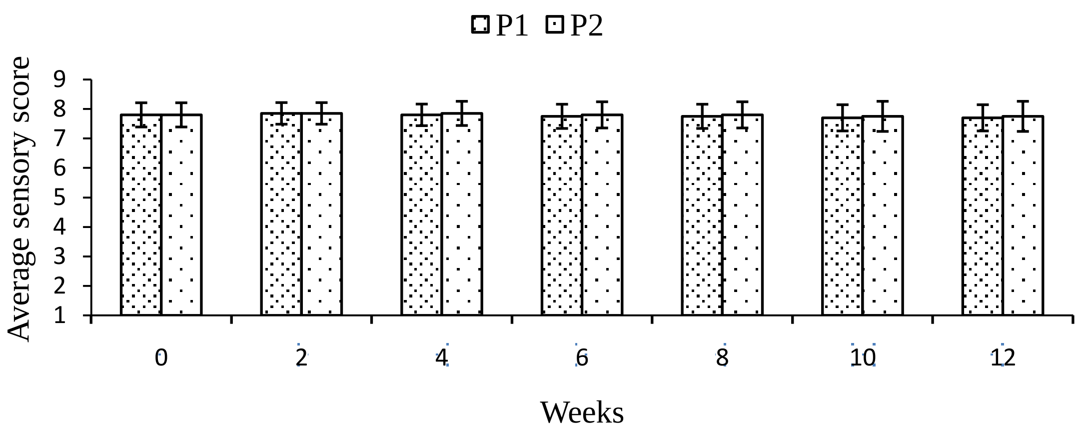


Fig. 4.4 Changes in color during storage at 40ºC temperature

Color, which can be judged by the eye, is the first importance in food selection (Ranganna, 2001). In the study (Fig. 4.4) the mean sensory for color at first was found to be slightly increased then afterward were found to be decreasing within the time. From the result of statistical analysis (Table C.1.3) it can be concluded that variation in packaging material had no significant ($p>0.05$) effect on the color of the product, but the storage time significantly affected ($P<0.05$) its color.

4.5.3.2 Changes in flavor

Flavor is the second important attributes of the product. Flavor embraces the senses of taste, smell and feeling. As far as human beings are concerned, it is generally agreed that the sense of taste is limited to sweet, sour, salty and bitter.

Average sensory scores given by the 10 panelists were tabulated in Table C.1.9 and Table C.1.10 and the changes in flavor of the product was used for the comparative study between two packaging materials stored at 40ºC temperature with increase in storage time. From the analysis (Fig. 4.5), it can be summarized that there is gradual reduction of mean sensory score of all the samples at both the condition. From the result of statistical analysis (Table C.1.4) it can be concluded that variation in packaging material had no significant ($p>0.05$) effect on the mean sensory score for flavor of the product, but the storage time significantly affected ($P<0.05$) its flavor.

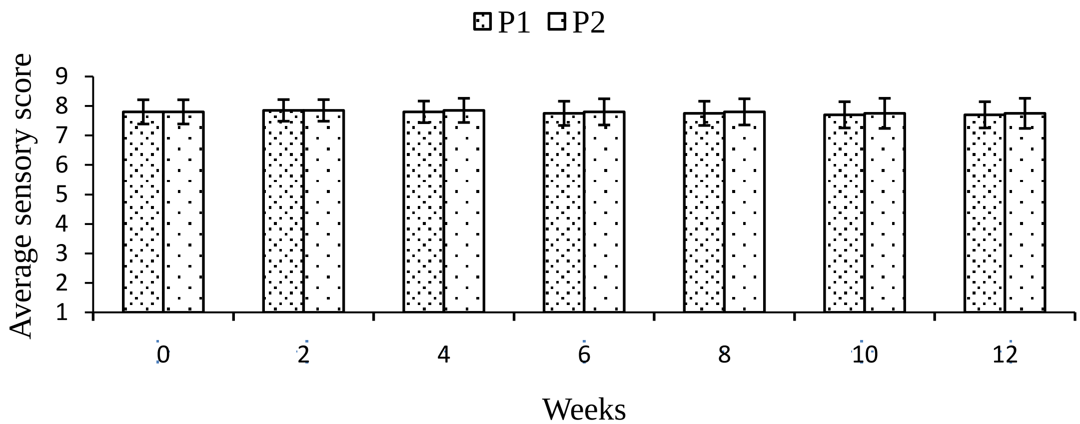


Fig. 4.5 Changes in flavor during storage at 40ºC temperature.

The major factor responsible for the flavor changes in the product are all the chemical parameters simultaneously occurred during the period of storage. Increased moisture content can lower the flavorings ability of product and similarly formation of free fatty acid and peroxides can give the off-rancid flavor to the product.

4.5.3.3 Changes in texture

Texture is another important sensory attribute, which evaluates the product with perception of tongue and acceptance of body.

Average sensory scores given by the 10 panelists were tabulated in Table C.1.9 and Table C.1.10 and the changes in texture of the product was used for the comparative study between two packaging materials stored at 40ºC temperature with increase in storage time.

From the analysis (Fig. 4.6) it was found that there is also gradual decrease in sensory score for texture. From the statistical analysis (Table C.1.9) it was found that, variation in packaging material had no significance difference in taste ($p>0.05$) but only the effect of storage time.

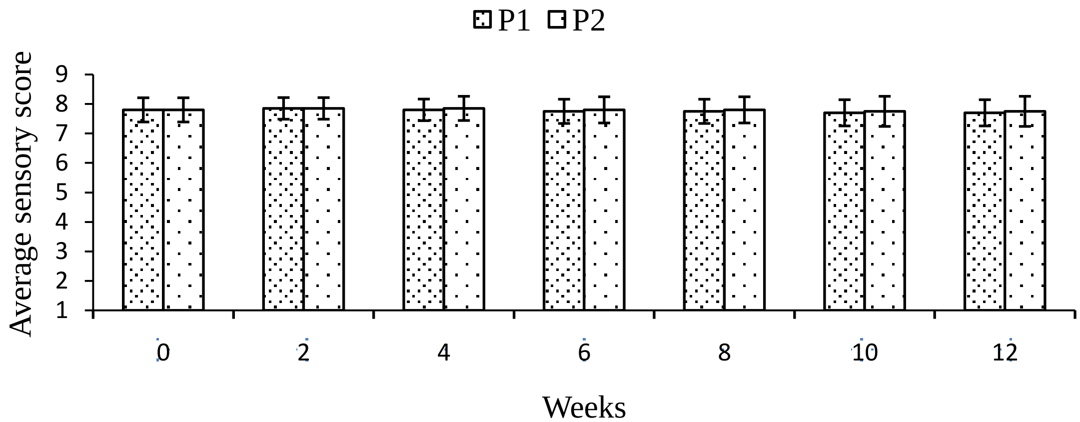


Fig. 4.6 Changes in texture during storage at room temperature.

4.5.3.4 Changes in crispness

Crispness is another important sensory attribute, which evaluates the product with bite and perception of tongue.

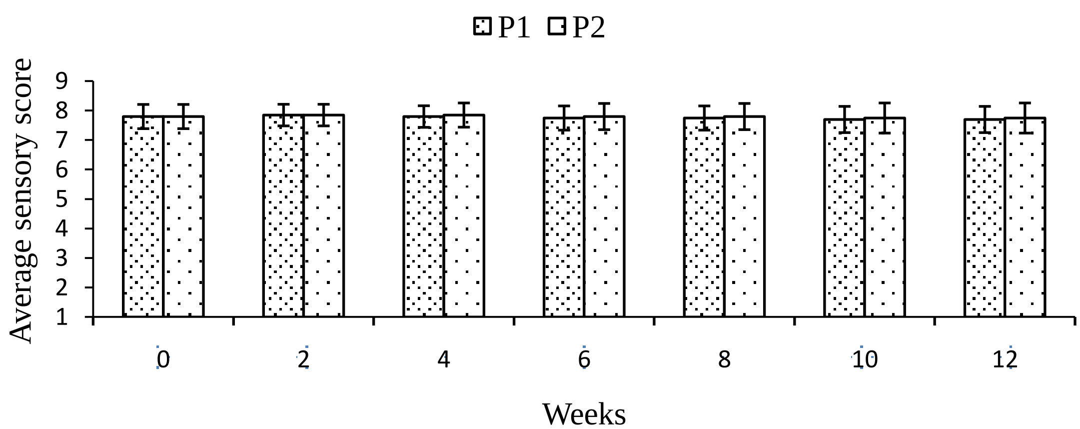


Fig. 4.7 Changes in Crispness during storage at 40ºC temperature.

Average sensory scores given by the 10 panelists were tabulated in Table C.1.9 and Table C.1.10 and the changes in taste of the product was used for the comparative study between two packaging materials stored at 40ºC temperature with increase in storage time.

From the analysis (Fig. 4.7) it was found that there is also gradual decrease in sensory score for crispness. From the statistical analysis (Table C.1.8) it was found that, variation in packaging material had no significance difference in crispness ($p>0.05$) but only the effect of storage time.

4.5.3.5 Change in taste

Taste is the primary factor which determines the acceptability of any product, which has the highest impact as far as market success of product, is concerned. Taste evaluates the product with perception of tongue and acceptance of body.

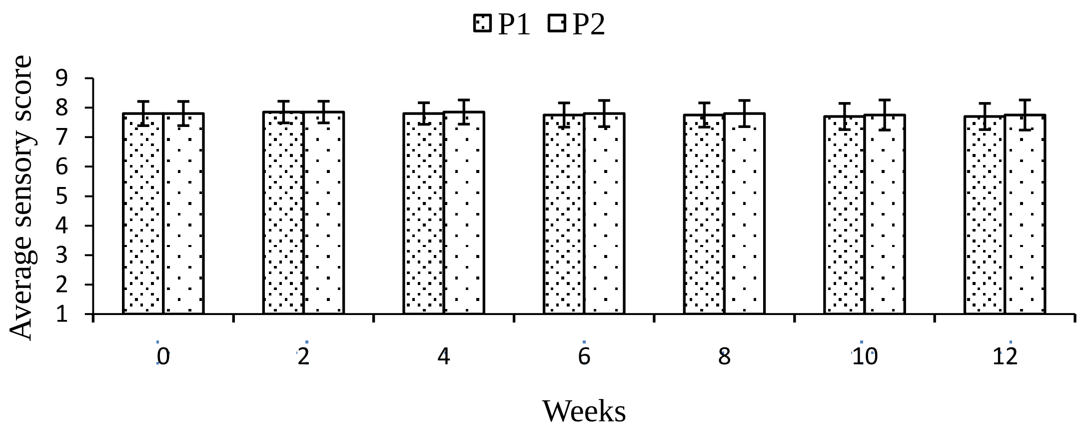


Fig. 4.8 Changes in taste during storage at 40ºC temperature.

Average sensory scores given by the 10 panelists were tabulated in Table C.1.9 and Table C.1.10 and the changes in taste of the product was used for the comparative study between two packaging materials stored at 40ºC temperature with increase in storage time.

From the analysis (Fig. 4.8) it was found that there is also gradual decrease in sensory score for flavor. From the statistical analysis (Table C-1.7) it was found that, variation in packaging material and storage time had no significance difference in taste ($p<0.05$).

4.5.3.6 Change in overall acceptability

Overall acceptability includes many implications, which is the important parameter in organoleptic estimation.

Average sensory scores given by the 8 panelists were tabulated in Table C.1.9 and Table C.1.10 and the changes in taste of the product was used for the comparative study between two packaging materials stored at 40ºC temperature with increase in storage time.

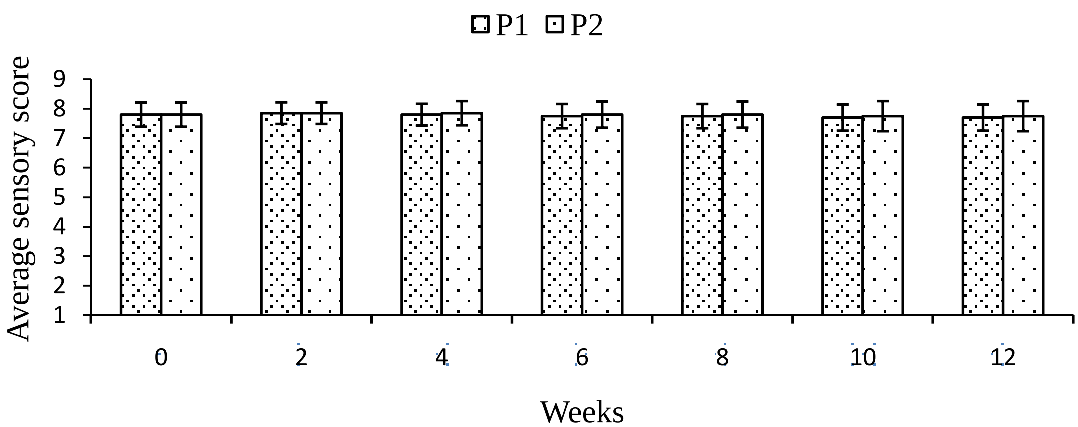


Fig. 4.9 Changes in overall acceptability during storage at 40ºC temperature.

From the analysis (Fig. 4.9) it was found that there is also gradual decrease in sensory score for overall acceptance. From the statistical analysis (Table C-1.8) it was found that, variation in packaging material and storage time had significance difference in overall acceptability ($p<0.05$).

The initial hedonic ratings of the biscuits are very much likely to predict the long term acceptability of biscuits. Goldman (1994) conducted a repeated consumption test of breakfast cereals and found the hedonic scores to be consistent. Kremer et al (2001) also observed that for food items that were eaten repeatedly, the difference between ratings for the first time the food was eaten was minimal compared to subsequent ratings. Findings of Leon et al (1999) for the study using five varieties of biscuit were consistent when using hedonic categorization. In another study, Sulmont-Rosse et al (2008) demonstrated that repeated exposure can produce experimental boredom leading to low success. Therefore, the single evaluation each person is to be preferred. However, mean of the panelists has been considered in the work.

4.6 Rate of increase in the AV and PV and the projected shelf life

The probable shelf life of the HEB was projected by studying the observed increase pattern of the values. The rate of increase in the AV was calculated to be 0.0102 mg KOH/ gm oil per day in case of P2 (laminate PET + metallic BOPP), while 0.011 mg KOH/ gm oil per day in case of P1 (laminate PET + BOPP). This rate of increase in the AV will take 62 weeks (15.5

months) in P2 (laminate PET + metallic BOPP) and 46 weeks (11.5 months) in P1 (laminate PET + BOPP) to cross the unacceptable value of AV which is 1 mg KOH/ gm oil per day.

Similarly, the rate of increase in the PV was calculated to be 0.0579 meqv peroxide/kg fat per day in case of P2 (laminate PET + metallic BOPP), while 0.0625 meqv peroxide/kg fat per day in case of P1 (laminate PET + BOPP).

This rate of increase in the PV will take 82 weeks (20.5 months) in P2 (laminate PET + metallic BOPP) and 48 weeks (12 months) in P1 (laminate PET + BOPP) to cross the unacceptable value of PV which is 3 mg KOH/ gm oil per day. Although this is just a theoretical value, a slight increase in the shelf life of the HEB.

4.7 Cost of product

The cost of most preferred HEB (sample C) is NRs. 11.91 per 100gm including overhead cost and profit of 10%. (Calculation is given in Appendix D).

Part V

Conclusions and recommendations

5.1 Conclusion

On the basis of the study carried out, the following conclusions were drawn.

a. High energy biscuit prepared by using wheat: soy flour in the ratio 90:10 was found to be the best in terms of sensory analysis and showed that it meets the minimal nutritional requirements of a person during catastrophic conditions as specified by WFP.
b. The sample packaged in two different packaging materials i.e. P1 (laminate PET + BOPP) and P2 (laminate PET + metallic BOPP) at 40ºC temperature, gradual increase of acid value and peroxide value. However the values were far below the standard unacceptable range during the study for 12 weeks. Similarly decrease in color, flavor, taste and texture sensory scores was found with storage time.
c. The total cost of HEB was calculated and found to be NRs. 11.91 per 100gm.

5.2 Recommendations

This study can be further continued with the following recommendations,

a. Study of the vitamin retention after baking should be carried out.
b. HEB can be a distinct nutritional supplement. Evaluation of nutritional quality (e.g. PER, NPU, Digestibility etc) of the product should be studied.
c. Study of the amino acid profile and fatty acids composition of the prepared products
d. If HEB could be produced on an industrial scale for open market, children with severe acute malnutrition will be greatly benefited in its use due to its comparatively low cost with rich nutrients content.
e. Shelf life study using different packaging material and its optimization.
f. The shelf life of the HEB was found to be satisfactory in P2 (PET+ metallic BOPP). Further studies on combination of packaging material can be done eg. metalized laminate OPP 20micron/PR 3C/DRY/VMCPP 25micron.
g. Study for various flavored HEB production.

PART VI

Summary

Biscuits are the low cost, processed food which offers good taste along with nutritional values at affordable price with convenient availability. Biscuits have in general, a good shelf life in comparison to most of the other snack items. It is no more only viewed as a luxury tea item snack, but as an essential daily food composition of an average Nepalese household. Since biscuit is a kind of dry food having a long shelf life, the problem of deterioration is very low in comparison to other bakery products. Biscuits owing to their shelf- life can be beneficial for feeding programs and other immediate catastrophic conditions.

The present work is mainly focused on the preparation of High Energy Biscuit, used for as food at emergency situations and at other times as nutritional supplement, considering the problems of malnutrition and micronutrient deficiency and popular snack item amongst people.

Wheat flour and soy flour was screened through sieve of pore size 0.046mm diameter and proximate analysis was done. The moisture, fat, crude fiber, ash, protein, and gluten content were found to be 12.80, 1.50, 0.45, 0.51, 9.90, 9.10 and 5.0, 19.5, 9.6, 4.56, 43.15, 0 gm per 100gm of wheat flour and soy flour respectively. The wheat flour and soy flour were mixed in different proportions to make up 100 parts of flour i.e. A (100% wheat flour), B (95:5), C (90:10), D (85:15), E (80:20) and F (75:25). The other ingredients palm oil, pulverized sugar, salt, lecithin, invert sugar, sodium and ammonium bicarbonate and butter flavor were taken constant. The six different biscuits 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 ($P \leq 0.05$) in overall acceptability among the samples. Sample C (wheat flour: soy flour :: 90:10) got the highest mean sensory score. According to Dhingra and Jood (2004), higher levels from 10% of soy flour lead the acceptability of the product to decline due to compact texture of the crumb and the strong flavor. Proximate analysis of sample C was then carried out and the moisture, fat, crude fiber, protein, and ash content of HEB were found to be 0.97, 15.55, 2.08, 12.25 and 0.06 respectively.

The best ratio i.e. sample C was used for final preparation of biscuits and thus prepared biscuits were packaged in P1 (laminate PET + BOPP) and P2 (laminate PET + metallic BOPP) and stored at 40˚C. The samples were subjected for sensory analysis in every two weeks for 12 weeks. There was no significant difference (F pr.> 0.05) in mean sensory score of samples at the zero week and last week for the color, flavor, crispness and taste of the biscuit with respect to the packaging material, while the texture and overall appearance were found to have significant difference (F pr.< 0.05). The samples showed significant difference (F pr.< 0.05) in all sensory parameters with respect to the time of storage.

The chemical analysis i.e. AV and PV of the samples P1 (laminate PET + BOPP) and P2 (laminate PET + metallic BOPP) was done and the data were statistically analyzed by using two way ANOVA (no blocking) at 5% level of significance. This showed that both the packaging material shows the significant difference in AV of the product whereas there was no significant difference in packaging material in case of PV of the product. Similarly, storage time also showed the significant effect on the acid value, peroxide value, flavor and flavor and texture of the product. From the self-life study through the rate of increment of acid value and peroxide value, storage life up to 11.5 months and 15.5 months at 40˚C was determined for sample P1 (laminate PET + BOPP) and P2 (laminate PET + metallic BOPP) respectively. The rate of chemical changes was quite slower in sample packaged by P2 (laminate PET + metallic BOPP) as well as most of the sensory attributes were also preserved.

The cost of the product was calculated on the basis of raw materials, ingredients used and 10% overhead cost of total ingredients and was found to be NRs. 11.91 per 100gm. These costs vary with time and season.

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Appendices

Appendix-A

1. Sensory Evaluation Card

Sensory Evaluation Sheet

Of High Energy Biscuit

Prepared By: AnujNiroula

Purpose: Dissertation for the *partial fulfillment of the requirements for the degree Of* Bachelor’s Degree in Food Technology (B. Tech. Food)

Name of Panelist:

Age:

Dear Panelist, you are given 6 sample of HEB, please conduct the sensory analysis based on the following parameter using the table given

Sample	Color	Flavor	Taste	Texture	Overall acceptance
A					
B					
C					
D					
E					
F					

Like Extremely	9
Like Very much	8
Like Moderately	7
Like Slightly	6
Neither like nor dislike	5
Dislike Slightly	4
Dislike Moderately	3
Dislike very much	2
Dislike Extremely	1

Comment (if any)

………………………………………………………………………………..…………………

………………………………………………………………………….........

…………………...

Signature

Thank You!!!

Appendix-B

1. Sensory evaluation of the product

Table B.1.1 Two way ANOVA (no blocking) for color

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Formulation	5	9.483	1.8967	9.47	<.001
Panelist	9	2.483	0.2759	1.38	0.227
Residual	45	9.017	0.2004		
Total	59	20.98			

Since F pr. < 0.05, there is significant difference between the samples so LSD testing is necessary.

Table B.1.2 LSD for color

Sample	Mean score	LSD at 0.05	Mean difference				Remarks
A	6.9	0.4032	B-A=	0.3	A-F=	0.5*	>LSD*
B	7.2		C-B=	0.5*	B-D=	0.1	<LSD
C	7.7		C-D=	0.6*	B-E=	0.4	
D	7.1		D-E=	0.3	B-F=	0.8*	
E	6.8		E-F=	0.4	C-E=	0.9*	
F	6.4		C-A=	0.8*	C-F=	1.3*	
			D-A=	0.2	D-F=	0.7*	
			A-E=	0.1			

(* = Significantly different)

Table B.1.3 Two way ANOVA (no blocking) for for crispness

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Sample	5	4.8833	0.9767	4.14	0.004
Panelist	9	1.0833	0.1204	0.51	0.859
Residual	45	10.6167	0.2359		
Total	59	16.5833			

Since F pr. < 0.05, there is significant difference between the samples so LSD testing is necessary.

Table B.1.4 LSD for crispness

Sample	Mean score	LSD at 0.05	Mean difference				Remarks
A	7.1	0.4375	A-B=	-0.2	A-F=	0.6*	>LSD*
B	7.3		B-C=	0.2	B-D=	0.4	<LSD
C	7.1		C-D=	0.2	B-E=	0.7*	
D	6.9		D-E=	0.3	B-F=	0.8*	
E	6.6		E-F=	0.1	C-E=	0.5*	
F	6.5		A-C=	0	C-F=	0.6*	
			A-D=	0.2	D-F=	0.4	
			A-E=	0.5*			

(* = Significantly different)

Table B.1.5 Two way ANOVA (no blocking) for flavor

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Sample	5	12.6	2.52	12.51	<.001
Panelist	9	2.9333	0.3259	1.62	0.139
Residual	45	9.0667	0.2015		
Total	59	24.6			

Since F pr. < 0.05, there is significant difference between the samples so LSD testing is necessary.

Table B.1.6 LSD for flavor

Sample	Mean score	LSD at 0.05	Mean difference				Remarks
A	7.1	0.4043	A-B=	0	A-F=	1.3	>LSD*
B	7.1		B-C=	0.1	B-D=	0.4	<LSD
C	7		C-D=	0.3	B-E=	0.6*	
D	6.7		D-E=	0.2	B-F=	1.3*	
E	6.5		E-F=	0.7*	C-E=	0.5*	
F	5.8		A-C=	0.1	C-F=	1.2	
			A-D=	0.4	D-F=	0.9*	
			A-E=	0.6*			

(* = Significantly different)

Table B.1.7 Two way ANOVA (no blocking) for taste

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Sample	5	18.15	3.63	16.87	<.001
Panelist	9	1.8167	0.2019	0.94	0.502
Residual	45	9.6833	0.2152		
Total	59	29.65			

Since F pr. < 0.05, there is significant difference between the samples so LSD testing is necessary.

Table B.1.8 LSD for taste

Sample	Mean score	LSD at 0.05	Mean difference				Remarks
A	7.4	0.4178	A-B=	0.2	A-F=	1.5	>LSD*
B	7.2		B-C=	0	B-D=	0.1	<LSD
C	7.2		C-D=	0.1	B-E=	0.9	
D	7.1		D-E=	0.8	B-F=	1.3	
E	6.3		E-F=	0.4	C-E=	0.9	
F	5.9		A-C=	0.2	C-F=	1.3	
			A-D=	0.3	D-F=	1.2	
			A-E=	1.1			

(* = Significantly different)

Table B.1.9 Two way ANOVA (no blocking) for Texture

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Sample	5	3.2833	0.6567	3.46	0.01
Panelist	9	2.35	0.2611	1.37	0.228
Residual	45	8.55	0.19		
Total	59	14.1833			

Since F pr. < 0.05, there is significant difference between the samples so LSD testing is necessary.

Table B.1.10 LSD for texture

Sample	Mean score	LSD at 0.05	Mean difference				Remarks
A	6.8	0.3926	B-A=	0.3	A-F=	0.4*	>LSD*
B	7.1		B-C=	0.1	B-D=	0.3	<LSD
C	7		C-D=	0.2	B-E=	0.5*	
D	6.8		D-E=	0.2	B-F=	0.7*	
E	6.6		E-F=	0.2	C-E=	0.4*	
F	6.4		C-A=	0.2	C-F=	0.6*	
			A-D=	0	D-F=	0.4*	
			A-E=	0.2			

(* = Significantly different)

Table B.1.11 Two way ANOVA (no blocking) for Overall Acceptability

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Sample	5	15.2833	3.0567	16.74	<.001
Panelist	9	3.0833	0.3426	1.88	0.08
Residual	45	8.2167	0.1826		
Total	59	26.5833			

Since F pr. < 0.05, there is significant difference between the samples so LSD testing is necessary.

Table B.1.12 LSD for Overall acceptability

Sample	Mean score	LSD at 0.05	Mean difference				Remarks
A	6.7	0.3849	B-A=	0.2	A-F=	0.4*	>LSD*
B	6.9		C-B=	0.9*	D-B=	0.4*	<LSD
C	7.8		C-D=	0.5*	B-E=	0.4*	
D	7.3		D-E=	0.8*	B-F=	0.6*	
E	6.5		E-F=	0.2	C-E=	1.3*	
F	6.3		C-A=	1.1*	C-F=	1.5*	
			D-A=	0.6*	D-F=	1*	
			A-E=	0.2			

(* = Significantly different)

Appendix C

1. Shelf life of the product

Table C.1.1 Two way ANOVA (no blocking) for AV

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Storage time	6	0.0645	0.01075	43.83	0.001
Packaging material	1	0.004829	0.004829	19.69	0.004
Residual	6	0.001471	0.000245		
Total	13	0.0708			

Since F pr. < 0.05, there is significant difference between the samples at different storage time and between two packaging material.

Table C.1.2 Two way ANOVA (no blocking) for PV

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Storage time	6	0.278686	0.046448	14.69	0.002
Packaging material	1	0.044579	0.044579	14.1	0.009
Residual	6	0.018971	0.003162		
Total	13	0.342236			

Since F pr. < 0.05, there is significant difference between the samples at different storage time and between two packaging material.

Table C.1.3 Two way ANOVA (no blocking) for color

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Packaging material	1	0.0016071	0.0016071	4.5	0.078
weeks	6	0.0371429	0.0061905	17.33	0.001
Residual	6	0.0021429	0.0003571		
Total	13	0.0408929			

Since F pr. > 0.05 for packaging material, there is no significant difference between the samples at different packaging material. But F pr. < 0.05 for different storage time, therefore there is significant difference between the samples at different storage time.

Table C.1.4 Two way ANOVA (no blocking) for flavor

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Packaging material	1	0.0016071	0.0016071	4.5	0.078
weeks	6	0.0121429	0.0020238	5.67	0.027
Residual	6	0.0021429	0.0003571		
Total	13	0.0158929			

Since F pr. > 0.05 for packaging material, there is no significant difference between the samples at different packaging material. But F pr. < 0.05 for different storage time, therefore there is significant difference between the samples at different storage time.

Table C.1.5 Two way ANOVA (no blocking) for texture

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Packaging material	1	0.0028571	0.0028571	8	0.03
Weeks	6	0.0135714	0.0022619	6.33	0.02
Residual	6	0.0021429	0.0003571		
Total	13	0.0185714			

Since F pr. < 0.05, there is significant difference between the samples at different storage time and between two packaging materials.

Table C.1.6 Two way ANOVA (no blocking) for crispness

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Packaging material	1	0.0016071	0.0016071	4.5	0.078
Weeks	6	0.0142857	0.002381	6.67	0.018
Residual	6	0.0021429	0.0003571		
Total	13	0.0180357			

Since F pr. > 0.05 for packaging material, there is no significant difference between the samples at different packaging material. But F pr. < 0.05 for different storage time, therefore there is significant difference between the samples at different storage time.

Table C.1.7 Two way ANOVA (no blocking) for taste

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Packaging material	1	0.0007143	0.0007143	1	0.356
Weeks	6	0.0142857	0.002381	3.33	0.084
Residual	6	0.0042857	0.0007143		
Total	13	0.0192857			

Since F pr. > 0.05, there is no significant difference between the samples at different storage time and between two packaging material.

Table C.1.8 Two way ANOVA (no blocking) for overall acceptance

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Packaging material	1	0.0044643	0.0044643	15	0.008
Weeks	6	0.0267857	0.0044643	15	0.002
Residual	6	0.0017857	0.0002976		
Total	13	0.0330357			

Since F pr. < 0.05, there is significant difference between the samples at different storage time and between two packaging materials.

YTable C.1.9 Average sensory score for sample packed in P1 (laminate PET + BOPP)

Weeks	Color	Flavor	Texture	Crispness	Taste	Overall Acceptability
0	7.7(0.48)	7(0.44)	7.1(0.44)	7.1(0.44)	7.2(0.52)	7.8(0.42)
2	7.8(0.42)	7.05(0.31)	7.1(0.55)	7.1(0.44)	7.3(0.57)	7.85(0.36)
4	7.75(0.36)	7.05(0.51)	7.1(0.44)	7.1(0.44)	7.25(0.55)	7.8(0.42)
6	7.7(0.31)	7(0.44)	7.05(0.31)	7.05(0.51)	7.25(0.55)	7.75(0.44)
8	7.7(0.48)	7(0.44)	7.05(0.51)	7.05(0.31)	7.2(0.52)	7.75(0.44)
10	7.65(0.47)	6.95(0.31)	7(0.55)	7(0.45)	7.2(0.52)	7.7(0.48)
12	7.6(0.44)	6.95(0.31)	7(0.55)	7(0.45)	7.15(0.36)	7.7(0.48)

Values in the Parenthesis are the standard deviation.

Table C.1.10 Average sensory score for sample packed in P2 (laminate PET + metallic BOPP)

Weeks	Color	Flavor	Texture	Crispness	Taste	Overall Acceptability
0	7.7(0.48)	7(0.44)	7.1(0.44)	7.1(0.44)	7.2(0.52)	7.8(0.42)
2	7.8(0.42)	7.05(0.31)	7.1(0.55)	7.1(0.44)	7.3(0.57)	7.85(0.36)
4	7.75(0.36)	7.05(0.51)	7.1(0.44)	7.1(0.44)	7.3(0.57)	7.85(0.36)
6	7.7(0.31)	7.05(0.31)	7.1(0.55)	7.1(0.44)	7.25(0.55)	7.8(0.42)
8	7.7(0.48)	7(0.55)	7.1(0.44)	7.05(0.51)	7.25(0.55)	7.8(0.42)
10	7(0.44)	7(0.55)	7.05(0.31)	7.05(0.31)	7.25(0.55)	7.75(0.44)
12	7.65(0.47)	7(0.44)	7.05(0.51)	7.05(0.31)	7.2(0.52)	7.75(0.44)

Values in the Parenthesis are the standard deviation.

Appendix-D

Table D Cost calculation of the product (Formula C).

Particulars	Cost (NRs/kg)	weight (gm) (wet basis)	Cost (NRs/100gm)
Wheat flour	30	70	2.1
Soybean	70	11	0.77
SMP	445	0.2	0.089
Shortening (palm Oil)	155	14	2.17
Sugar	70	13	0.91
Ammonium Bicarbonate	400	1.4	0.56
Sodium Bicarbonate	200	0.7	0.14
Others		3	0.5
Vitamin & Mineral premix	8500	0.2	1.7
Raw material Cost		113.5	8.939
Processing and labor Cost			
(10% of raw material cost)			0.8939
Packaging cost			1
Profit (10%)			1.08329
Grand Total Cost			11.91619

Cost of raw materials varies with season.