

Reading Manual for Peanut Butter

Under PMFME Scheme



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Ministry of Food Processing Industries

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Abbreviations & Acronyms

Sr:No.	Abbreviations &Acronyms	Full Forms
1.	PM FME	Prime Minister's Formalisation of Micro Food Processing Enterprises Scheme
2.	MUFA	Monounsaturated fatty acids
3.	PFA	Paraformaldehyde
4.	LDL	Low Density Lipoprotein
5.	USD	United States Dollar
6.	CAGR	Compound Annual Growth Rate
7.	GAIC	Gujarat Agro Industries Corporation. Business
8.	FAO	Food and Agriculture Organization
9.	HACCP	Hazard Analysis and Critical Control Point
10.	VEG	Vegetarian
11.	FSSAI	Food Safety and Standards Authority of India

CHAPTER 1

INTRODUCTION

Groundnut or Peanut (*Arachis hypogaea* L.) is a cheap source of protein and thus, part of diet for many around the world. The chemical composition of peanuts reveal and average content of 26-28% of protein and 40-54% of oil by fresh weight, thus, making it an incredible source of valuable amount of energy nutrients. India is world's second largest producer of the peanuts, accounting to over 9 million tonnes of production in 2018 from nearly 8 million hectare of land under its cultivation. This production could be attributed to primarily five major states under its cultivation, namely, Gujarat, Tamil Nadu, Karnataka and Maharashtra that collectively produce about 90 percent of its production. Of this Gujarat and Andhra Pradesh are the front runners and account for more than half of the entire cultivation, followed by Maharashtra, Tamil Nadu and Karnataka (Talawar, 2004).

Peanut is one of the most popular and largest sources of edible oils. It produces high value oil and lipid components. The fat content in peanut contains monounsaturated fatty acids (MUFAs) (50%), polyunsaturated fatty acids (PUFAs) (33%) and saturated fatty acids (14%) (Settaluri et al., 2012). Major products obtained from peanuts are peanut oil, peanut butter and peanut meal. These peanut lipid products are considered as healthier options owing to the MUFA content in the peanut lipids. It helps in lowering the serum LDL cholesterol level by 14% and total body cholesterol level by nearly 11 %. It also helps to maintain the HDL cholesterol levels and reducing the triglycerides. The effects are comparable with that of olive oil based diet according to several studies. The overall impact thus, leads to better heart health and reduction in coronary heart disease. Peanut being a leguminous plant has good amount of protein than other nuts and are comparable with various healthy beans. Peanut protein content in peanut cake could be accounted to about 50% of the total cake weight (Zhao et al. 2012), with good biological value protein consisting of 20 amino acids, acting as major source of arginine. Peanut protein has good digestibility and unlike animal proteins also have bioactive components and high value fibres in its composition. Moreover, peanuts are excellent source of vitamin B complexes like Thiamine (B1), required for nervous and cardiovascular health. It also has significant content of B3, B5, B6 and B9. It also serves a major source of fat soluble vitamin particularly vitamin E owing to its

significant lipid content. Further, peanut provides magnesium (about 176 mg), calcium (54 mg), phosphorous (358 mg), iron (2.26 mg), zinc (3.31mg), selenium (7.5 mg) and other traces like copper and manganese to consumers. Peanut bioactive compounds are rich source of polyphenols, flavonoids and stilbene in various parts of the seeds. Compounds such as procyanidins, catechins and resveratrol, present in peanuts are responsible for anti-inflammatory activity and antioxidant activity (Zhao et al. 2012). Owing to these wide nutritional benefits associated with peanut and its components, peanut based products are increasing in demand with increasing health consciousness amongst consumers. Moreover, want for dairy and animal product alternatives are on the rise amongst consumer preferring vegan and vegetarian source of diet.

Peanut butter is one of the most sought after products of peanuts that was created during the late 18th century as a protein substitute for people with poor teeth. It was patented by Dr. John Harvey Kellogg as a nut meal product and was used for serving soldiers. Since then commercial production of peanut butter has flourished worldwide and now the product has become omnipresent and is consumed as part of staple diet. Moreover, it is found to be suitable product for every age group ranging from infants to elderly. Recent data shows beneficial health impact on malnourished infants when fed with peanut butter (Arya et al. 2016).

Peanut butter is usually made using roasting and crushing of raw peanuts into a creamy extracted product that is commonly consumed as spread over breads or as ingredient in various culinary preparations around the world. Its popularity has been increased recently in Indian urban population and thus, have started to appear on shelf of supermarkets in cities. Numerous variant of the product has been developed with basic peanut butter products by incorporating various flavors using fruits, cocoa solids and other such natural and artificial flavorings.

The present manual is designed to provide a comprehensive guidance about peanut butter preparation and processing in details, effect of processing on peanut butter and its properties, processing and packaging requirements and the product specifications, standards and safety requirements for peanut butter manufacture.

1.1 Present Status of National and Global Peanut butter market

The Indian peanut butter production is mostly concentrated in the peanut growing states, of which, Gujarat is leader in its manufacturing. The existing peanut butter manufacturing units are concentrated in this region and cater to the export to USA, Canada, Japan, Middle East, South East Asian and African Countries. Although the popularity of the peanut butter is variable around the world, most of which is consolidated in American and European countries, however, the trend of consumption has seen massive jump in developing nations as well. Most of this is mainly focused on urban population in countries like India (Dhamsaniya et al., 2012).

The global butter market currently is estimated to about 21.6 billion USD, and has grown at 3.3% CAGR during the current decade (2010-20) as against the global oil and fat market that has grown by 2.6% CAGR. The Indian butter market has even shown a bigger leap with growth at the rate of 8.6% CAGR from 2010 to 2020, while the Indian fats and oil market has grown at the rate of 6.7% CAGR, during the same period. Peanut butter is a dynamic product now piercing this butter market, owing to various growth drivers (GAIC, 2017).

The primary reason for the increase in demand for peanut butter could be attributed to many factors like favourable demographic and lifestyle change. As the working population of India is increasing by 64% by current year (2021) and increasing purchasing capacity per capita, along with increasing urbanization. Worldwide consumers are becoming wary of the various health issues associated with the junk food consumption. These consumer population are looking for healthy alternative to the junk that would taste and feel just like the junk commodities. Moreover, the day to day activities of people are becoming strenuous and requirement of energy on the other hand have gone up. Peanut butter is a healthy alternative to the junk food providing required energy and other beneficial nutrients (Market Data Forecast, 2021). A market survey by Patel (2021) depicts the trend of peanut butter consumption across Indian cities (Figure 1).

With recent estimates India will become world's 5th largest consumer market by 2025, with the Indian middle class increasing by 583 million by this year. Other reasons include the improvement in the retail structure in India, with the industry becoming more organized. Big players like Reliance Fresh, Big Bazar, Easyday, etc. coming into picture availability of variety of consumer food products has grown. The retail market is expected to grow by 10%. Further,

the e-grocery segment in India has grown by 26% CAGR, owing to improved connectivity (GAIC, 2017).

The peanut butter is segment is based on the type of products and the channel of distribution. There are different types of products such as plain, regular, low sodium and sugar and flavoured. The market for plain or regular peanut is large, followed by low sodium and others. The segment like low sodium and flavoured peanut butter are growing market share. The recent prevailing conditions of COVID19 that has caused nationwide lockdowns, has influence even the peanut butter market, negatively. As with the case with most of the industries the production of peanut butter had hit. Apart from the hindrance in cultivation, transport and industrial production that has impacted all the sectors, the peanut butter market seem to have survived a greater loss, owing to consumers perspective about rising health benefits associated with consumption of healthy spread options. However, the exact impact of COVID19 over the peanut butter consumption patterns and consumer behaviour is still to be studied (Market Data Forecast, 2021).

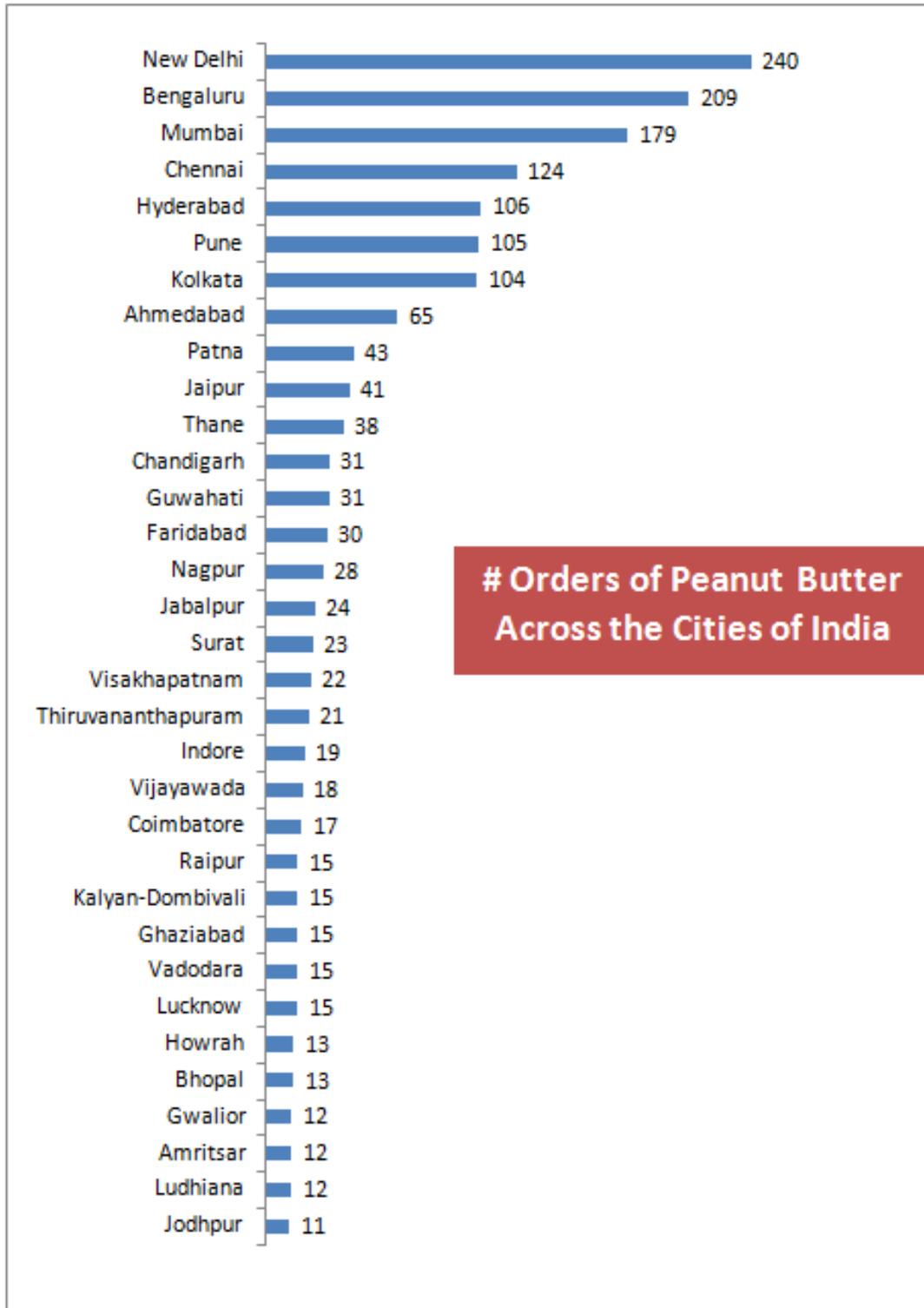


Figure 1. Trend of peanut butter consumption across Indian cities (Patel, 2021), values represents absolute number of orders received.

CHAPTER- 2

PREPARATION OF PEANUT BUTTER

2.1 Selection of raw material for peanut butter manufacturing

A food product becomes desirable for consumption owing to various characters associated with it. Primary influencer of these characteristics could be attributed to the raw material selection. Peanuts are the main raw material for preparation of peanut butter and thus, most of these aforesaid characters could be attributed to its selection. The nutrition and flavour of the peanuts could be attributed to its composition. In peanuts the main component that is used in peanut butter manufacture are the oil and protein content in the peanut varieties. In comparison with the dairy butter products, that contains dairy fat in totality as the main component of the butter; the peanut butter contains of at least 20% of peanut protein, about 50% fat and rest all other nutrient components (Dhamsaniya et al. 2012). Although there are numerous varieties that could be used nationwide, however, there are few commercial varieties mainly cultivated in the major peanut growing states in India as shown in Table 1.

Table 1. Groundnut varieties available for use in India

State/Region	Varieties	Season
Gujarat	Akshay, Amrut, Apple, Avani 20, Bold, Desi, Dharnidhar, G-10, 11, 13, 17, 20, 22, 29, 30, 31, G-33, 37, 38, 39, 41, 47, G-555, G-99, Gavabeej, Gujarat II, Handeji, Israeli, J2, 20, 29, JV bold, Khedut, Kranti 93, Mandav 37, Prerna, Rohini, Super Bombay, Swastika 99, Swati, Sweta, T-33, TGS 26 and TG 41.	Kharif and rabi
Andhra Pradesh	Dharni, JL, K, K-6, Natu, Nati, Kadiri 2, 5, 6, Nagana, TAG-24 and Narayani.	Kharif
Karnataka	Ajaati, Badami, DTBTH, GAFA6, Ganagkaveri, GL, GL24, GI-6, GTBT, GPBD 4, Jowari, JL2, K6, KF6, M25, Shiggou, trupti and Kopargaon-1	Rabi

Rajasthan	Akshay , Algora, Archana, Ardgi, Awani 20, M-13, Fara10, 20, G10, 20 , G10 (Dharti), Gajrat, Galkot, GG13, GG 20, Girnar, Kadav, Lodha, Shankar, Mangal kalash, Matra, G21 and N-13.	Kharif and rabi
Maharashtra	G10, G 22, G 15, MH-1, JL 24, JL 22, JL-286, Unnati, Warna, Vikram, Korad, Kopergaon1 and 2, Phule, Vyas and TLG	Kharif and rabi
West Bengal, Orissa and Jharkhand	Vasundhara (Dh 101), TG 51, Vijetha (R 2001-2), Girnar 3 (PBS 12160)	
Punjab	M548, Girnar (PBS-24030), Utkarsh (CSMG 9510), GG 21, HNG 69, RG-510, HNG-123,	

Source: APEDA (2018) and DOD (2021)

The varieties selected for preparing peanut butter should yield good spreadability and firmness in the peanut butter texture. Different varieties could be analysed for different flavour profiles and could be combined to yield appropriate flavours (Dhamsaniya et al. 2012). Varieties mentioned in Table 1 have different cultivation characteristics, some of them are kharif varieties and some could be grown in both the seasons. Depending on this and the agroclimatic conditions specific varieties suitable for the region are selected, therefore some compositional variation would be prominent in the peanut butter products.

2.2 Peanut Butter Processing

Traditional process of groundnut oil mills, uses crushing of entire pod along with the groundnut shell to be processed with the kernel to obtain oil, this make the oil cake obtained unsuitable for further processing. However, when processed for peanut butter all the portions with exception of the outer testa could be used for human consumption (Dhamsaniya et al. 2012). The typical process of peanut butter manufacture involves simple unit operations, where, selected varieties of peanut cultivars solely or in combination are shelled and ground after removal of the testa (skin) to a pasty or buttery consistency. This preparation is admixed with other ingredient such as salt, sweeteners, herbs and spices and stabilizers. Other optional ingredients may include blending with vegetable oil, emulsifiers and whey. This preparation may

be proprietary. A flowchart of the complete process is as shown in Figure 2 and alternative process is shown in Figure 3

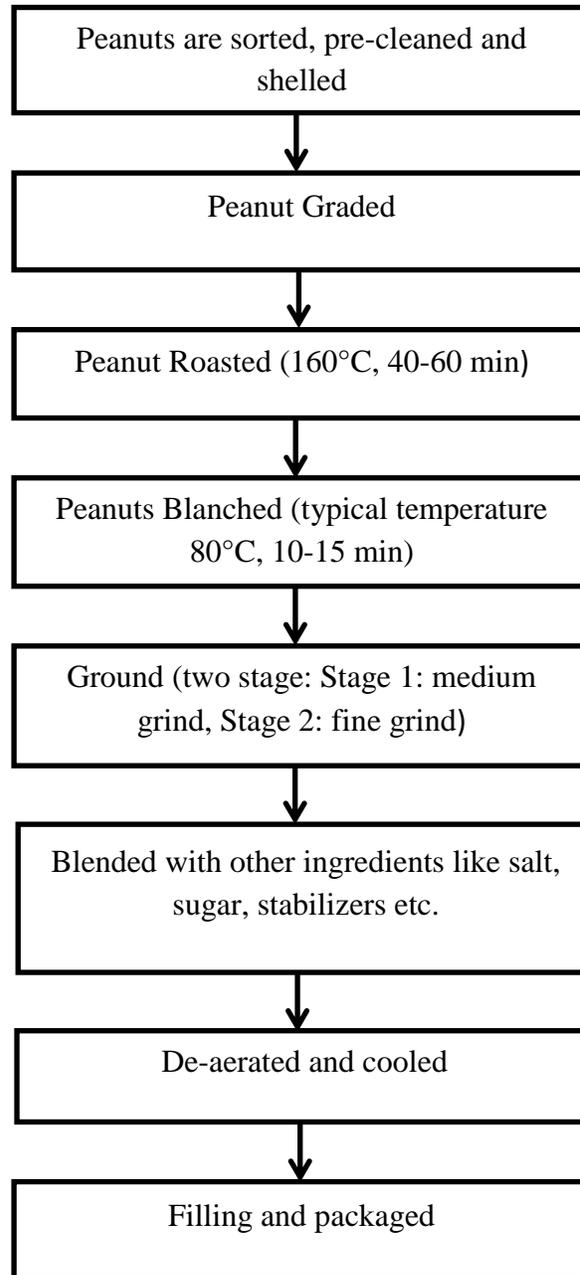


Figure 2: Flowchart for the peanut butter preparation process (Source: GAIC, 2017)

The typical process for manufacture of peanut butter consists of various stages as below:

1. Pre-cleaning, sorting and shelling of groundnut: Selected high quality peanut pods are sorted and shelled.
2. Grading: Shelled peanut pods are graded into various sizes, mostly complete, bold and big peanuts are chosen.
3. Roasting: the peanuts are then roasted at 160°C for about 40-60 minutes based on their initial moisture content. The final moisture content of the peanuts is brought to approximately 1% . This is primarily to reduce the water activity and thus, improve the shelf life of the peanuts.
4. Blanching: Post roasting the peanuts are cooled to room temperature. The peanuts are then subjected to blanching process, during which the outer testa (skin) is removed. Moreover, post blanching peanuts are inspected for discoloration such as blackening or greying of nuts.
5. Grinding: There are two stages of grinding of the peanuts. Stage 1 reduces the nuts into medium fractions in form of grits. These are further reduced to fine paste in the second stage. Moreover, other ingredients like sugar, salt, spices, stabilizers etc. are added at this stage and mixed thoroughly.
6. De-aeration: It is during the milling process that air gets incorporated into peanut butter and thus, during this stage the incorporated air is removed using vacuum aspiration. This process could be done in a combined grinding and vacuum machine.
7. Cooling: Post de-aeration, the ground peanut butter is now transferred to a stainless steel container, poured through hopper. Here the butter is mixed at intermediate stage and stored. The peanut is further stabilized and refrigerated in a rotating refrigerator known as votator.
8. Filling and packaging: The final products stabilized peanut butter is packaged into jars, either glass or polyethylene teraphthalate. Other commercial packs are also used to like sachets, etc. As peanut butter is high in lipid content, thus susceptible for lipid oxidation, the jars or containers are vacuum packed.

Alternative process for manufacture of peanut butter

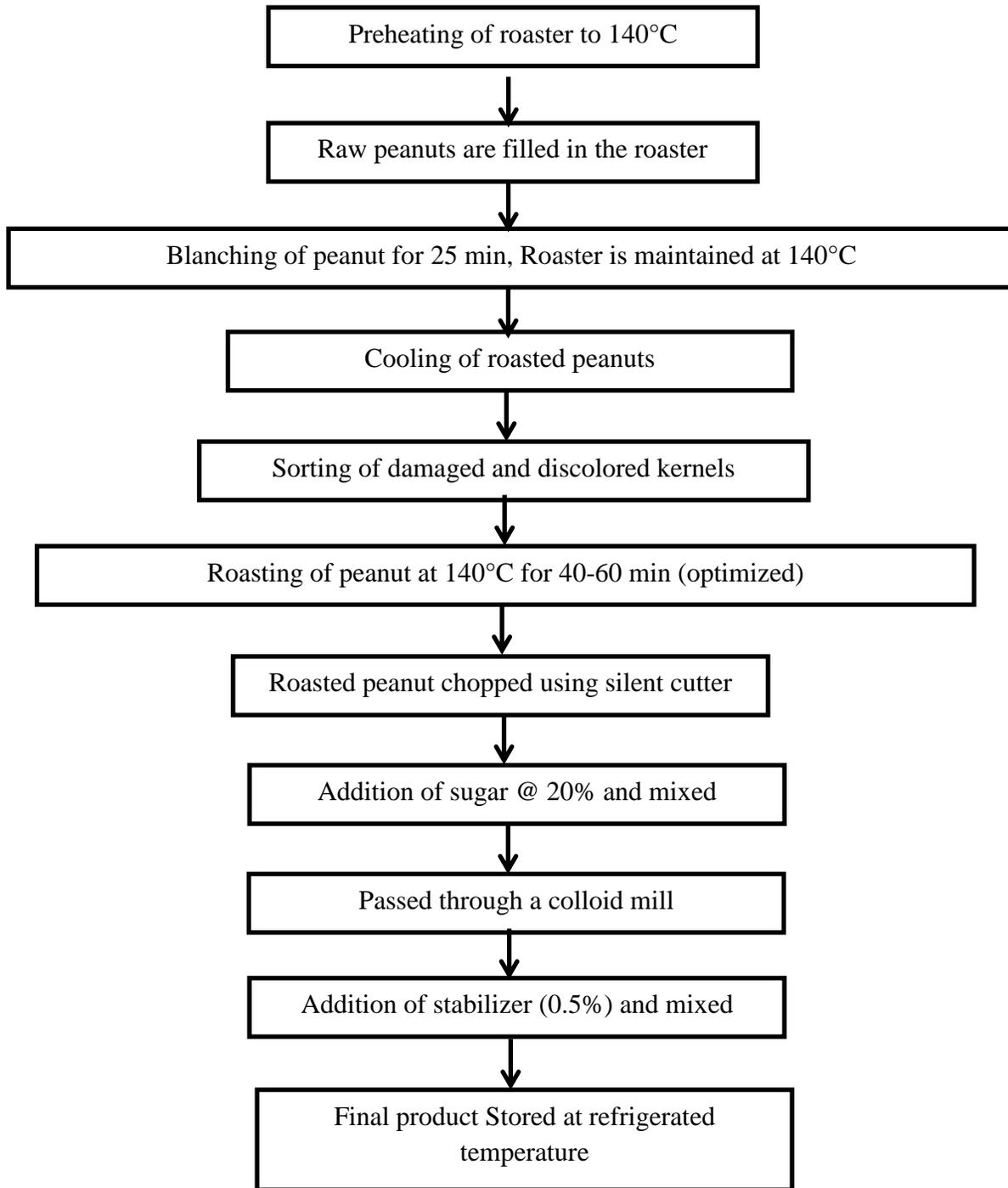


Figure 3: Alternative peanut butter preparation process (Source: Galvez et al. 2006)

2.3 Effect of Processing on Peanut Butter

Peanuts are susceptible to attacks from various pests, namely Bean leaf roller (*Lamprosema indicata*), long horned grasshopper (*Phaneroptera furcifera*), tiger moth caterpillar (*Dasychira mendosa*), June beetles (*Leucopholis irrorata*), saint fac grasshopper (*Atractomorpha psittacina*), etc. among other. However, the most prominent attack is seen by mould *Aspergillus flavus* that has lasting impact on the peanut and its products like peanut butter. It is absolutely necessary to process peanuts in order to reduce its impact on peanut products. *Aspergillus flavus* contamination, leads to formation of aflatoxin if the nuts are insufficiently dried post-harvest. The infection is observed both pre and post-harvest conditions. Aflatoxin affects atleast 25% of the world's peanut production and entering into the value chain. Owing to its drastic health impact such could lead to lasting impact on revenues as well. The maximum acceptability of aflatoxin is reported to be not more than 20 µg/kg of the commodity in the United States, while it is 4 µg/kg by European Union. Storage and processing of peanut had shown positive reduction in aflatoxin (Martey et al. 2020).

Other infections like various strains of salmonella have been studied in peanut butter. They have reported survival data of infecting organism in the peanut butter samples during the storage of 24 weeks. However, when thermally processed these infections have shown dramatic reduction, thus, thermal processing of peanut butter is a plausible solution for such infestations in peanut butter (Burnett et al. 2001). Further emerging techniques like high pressure processing has also shown promising results in reducing the infections of salmonella amongst consumers, when peanut butter samples are treated adequately (Grasso et al. 2010).

Apart from reducing infection and effect of intoxication, processing of peanut when converted to peanut butter has been studied to affect the composition and nutrition gained through the product. Chun et al. (2003) studies the effect of manufacturing processes on the vitamin E content of the peanut butter. They used peanut of runner type and estimated the peanut butter samples for their tocopherol content among raw peanut and peanut butter samples. They showed that addition of oils and stabilizers to the roasted peanuts contributes to addition of 4% of the alpha-tocopherol content in the final product. Moreover, retention of 95% of the total tocopherols are reported during the peanut butter manufacturing process. Tocopherols are primary antioxidants that are responsible for curbing free radical damage to the consumer's cells.

Peanut and its products have been topic of research over peanut allergenicity. Various works has been done on understanding the effect of processing like thermal treatments on the allergic.

Along with thermal processing various other technologies for processing of peanuts to reduce the allergic components have been studied in the recent past. High hydrostatic pressure is one such technology that modifies the biological protein structures that are capable of developing an immune response upon peanut consumption. HPP could work better in synergy with other technologies. Therefore, HPP and thermal processing in combination is a better technology to reduce the allergenicity of peanut. Other technologies like irradiation of peanuts before using for preparation of peanut butter could reduce the allergic reactions

2.4 Quality Attributes and it's Testing

The shelf life of the peanut butter is highly dependent on the various physical, chemical and microbiological changes that occur in the product during storage. Major factor responsible for the deterioration is degradation of peanut proteins due to putrefaction, particularly attributed to microbial action. Darkening of the product is resultant of browning reactions caused by protein and sugar interaction and rancidity is common owing to proportional unsaturated fatty acid content, caused when exposed to air. Several attempts have been made to improve the shelf life of the product by addressing the issues by preventing oil separation, improving the spreadability, texture, stickiness and consistency of the product. Moreover, new product types are being developed to improve industrial handling like bulk blocks and commercial sliced products. Flavor enhancement by adding various ingredients is another aspect of product range enhancement. The time and temperature of roasting to achieve the desirable traits of texture and color is quiet essential and thus, various studies are directed towards them. The hazard analysis and critical control point analysis for the peanut butter plant is as shown in Figure 4.

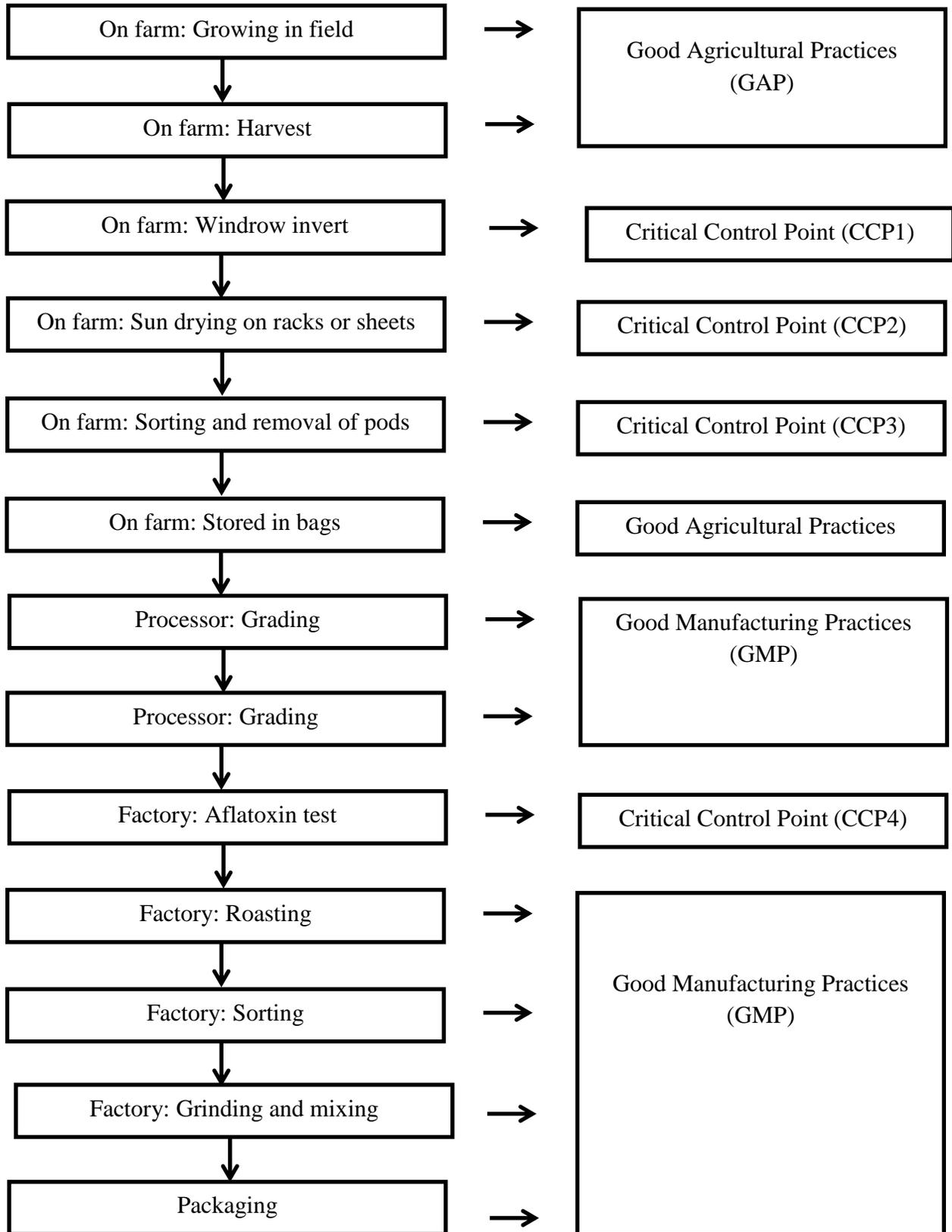


Figure 4: The hazard analysis and critical control point analysis for the peanut butter plant(FAO, n.d.)

2.5 Value Addition and Byproducts Processing

Prior survey has shown that peanut butter's popularity has only increased worldwide recently. Although peanut butter is itself an finished product and is consumed popularly in its primary product form (non-flavoured), there are multiple variants of the product available and are researched. The primary classification of peanut butter is done mainly based on its grade and texture. The textural classification of peanut butter is classified as follows.

1. Smooth peanut butter: a fine particle size and no perceptible graininess
2. Regular: defined grainy texture with perceptible peanut particles (dia: 1/16 inches)
3. Chunky: partial fine and grainy (dia larger than 1/16 inches)

Another classification as per U.S. grade classification is Grade A, US Standard, and US Sub-standard.

Galvez et al. (2006) developed a choco-peanut butter spread product. Figure 5 depicts the flowchart of processing of choco-peanut spread. Other products with stabilized peanut butter are prepared with the addition of roasted cassava flour by Palomar et al. (2006). In this process the roasted cassava flour is added to the roasted peanuts before the grinding process.

Attempts have also been made to incorporated fruit purees in peanut butter based products(Swanson and Munsayac, 1999) Peanut butter is also studied to be used as ingredient in other food products like cookies (Lathrop et al. 2014) and ice cream (Ting Hung et al. 2015).

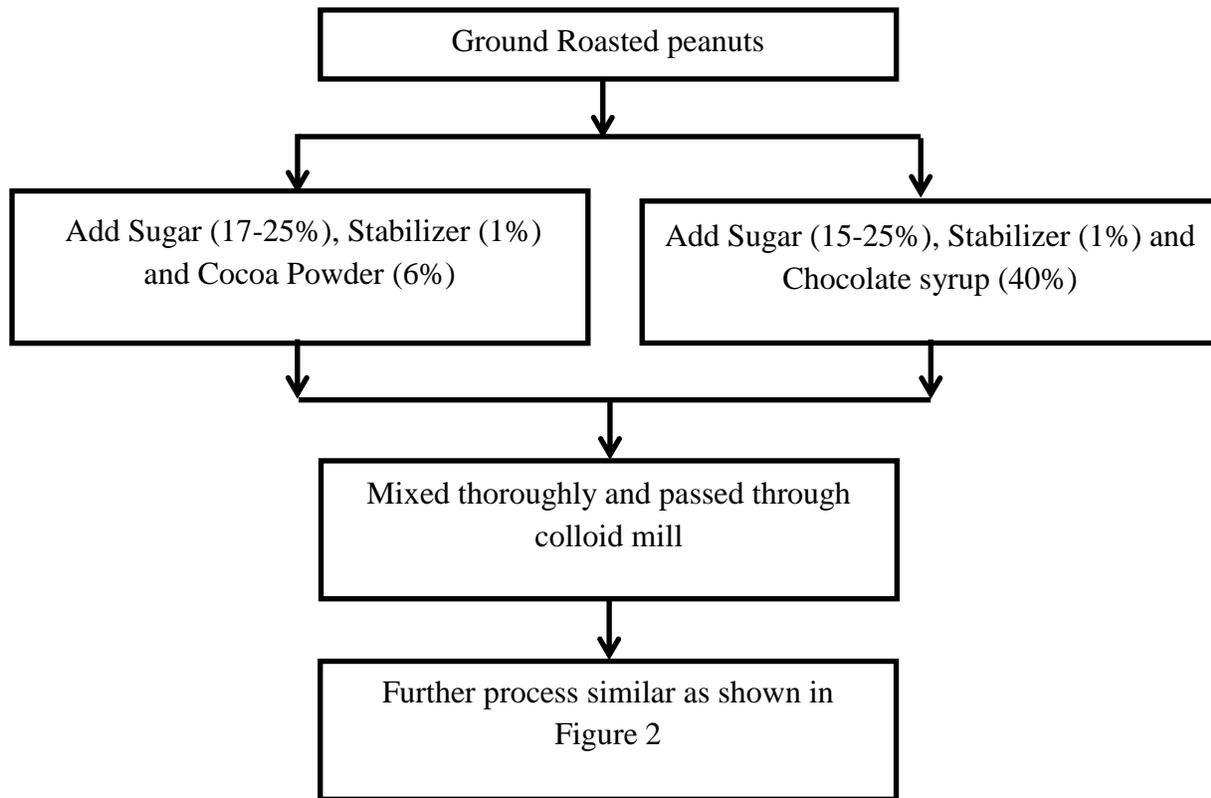


Figure 5: Variation in process flow for preparation of choco-peanut spread (Galvez et al. 2006).

Additives in Peanut Butter

To enhance the product stability, aesthetic appeal and shelf life, peanut butter is added with some additives in the commercial process. Following is a brief account of these additives.

1. **Stabilizers/Emulsifiers:** Peanut butter is incorporated with partially hydrogenated or unhydrogenated palm oils, to reduce oil separation and improve texture and stability of the product over period of time. Traditionally industries have used these vegetable oils for stabilization of peanut butter and its products. Moreover, peanut butter added with vegetable oils has found to have more acceptable texture. Commonly vegetable oils or stabilizers are added during the grinding of the peanuts. Further, the oil separation from the product also depends on the degree of grinding of the raw material. Finer the grind more is the oil separation and coarser.

Another set of additives added as emulsifiers are mono and di glycerides (MDG) although seems to be fats they are used as emulsifiers. They are sourced from oilseeds like palm, sunflower and soybean. They act as stabilizers as well in these foods, to improve the consistency and texture of the foods. However, as they may contain trans fatty acids they are often considered to be unhealthy and thus it becomes quite essential to label them on the peanut butter products, although the quantity is too low (about 0.5 g).

2. **Preservatives:** Common preservatives used in peanut butter are sodium salts of benzoates, as owing to the low moisture content in the peanut butter product the possibility of bacterial contamination in general is low. However, moulds are most probable contaminants of the product. Therefore, sodium benzoates prevent the product from mould growth and also retain freshness. It is classified as class II preservative. Butylated hydroxyanisole (BHA) is yet another preservative, majorly considered as antioxidant that was used to preserve mould growth. BHA and similar synthetic preservatives are banned in various countries like UK, Japan and European nations, owing to their cancer causing effects (International Agency for Research on Cancer, 1986). Mostly BHA, BHT and TBHQ are added to foods with higher fat content, to prevent oxidation of these products.

CHAPTER – 3

PACKAGING OF PEANUT BUTTER AND TECHNOLOGY

3.1 Packaging requirements of product

As per the requirement of FSSAI (2011), the containers or utensil used for preparation, packaging or to store the product should not be rusty, and should be properly tinned, depending on type of metal iron, copper or aluminium. The general guidelines for plastic containers as specified under the regulation should be followed. For canned product the container should be packed and sealed properly, container not dented or rusted, perforated and seam distortions and the cans should not be leaking. The packaging used for edible oil and fat product should conform with the FSSAI regulations, if packaged in tin plate as per prime grade quality in BIS Standards No. 1995/13955/9025/13954 and IS NO. 10325/10339.

Labelling requirements: prepackaged products should have particulars with declaration in English/Hindi (Devnagari script)/any other native language. Label should not contain misleading information that is likely to have false impressions. The prepackaged product should conform to the labelling requirement as per section 2.2.1. as specified in FSSAI (2011) labelling requirements, with information as stated below:

1. Name of food,
2. List of ingredients
3. Time of manufacturing
4. Flavouring agent
5. Net weight
6. Nutritional information with amount of nutrients
7. If added with hydrogenated fats should be declared as such.
8. Health/nutritional claims if any should be stated
9. Declaration of veg or non-veg components (if any) with specified symbols (sizes as specified in regulations)

10. Declaration of food additives: any acid regulators, antifoaming, anticaking agents, antioxidants as stated in above section, color, emulsifiers, stabilizers or other ingredients as permitted in the product (stated in Chapter 2).
11. Batch/Code/Lot identification
12. Date of production/manufacture
13. Best Before and use by Date
14. Country of origin

Other specific requirement for edible oil/fat should be labeled with expressions of purity as (Super/Extra/Micro/Double/Ultra refined) and content of cholesterol (if part of product). The peanut butter product should be provided with label requirement of blending with palmolein and groundnut oil content with percentage (FSSAI, 2011).

3.2 Plant layout, machines and equipment

3.3 Following list of equipment are suggested for typical peanut butter processing plant

Sr. No.	Name of equipment	Suggested power requirement (kW)
1.	Feeding Machine	0.55
2.	Hoister	0.75
3.	Continuous roaster machine	24
4.	Elevator	0.75
5.	Continuous cooling machine	5
6.	Peeling machine	0.74
7.	Selection belt	0.75
8.	Storing and feeding machine	0.55
9.	Grinder machine	5.5
10.	Paste pump	2.2
11.	Mixing tank	2.2
12.	Vacuum tank	2.2
13.	Storage tanks	-
14.	Control cabinet	-
15.	Pipelines	-

Adopted from (Gelgoog Company, 2020)

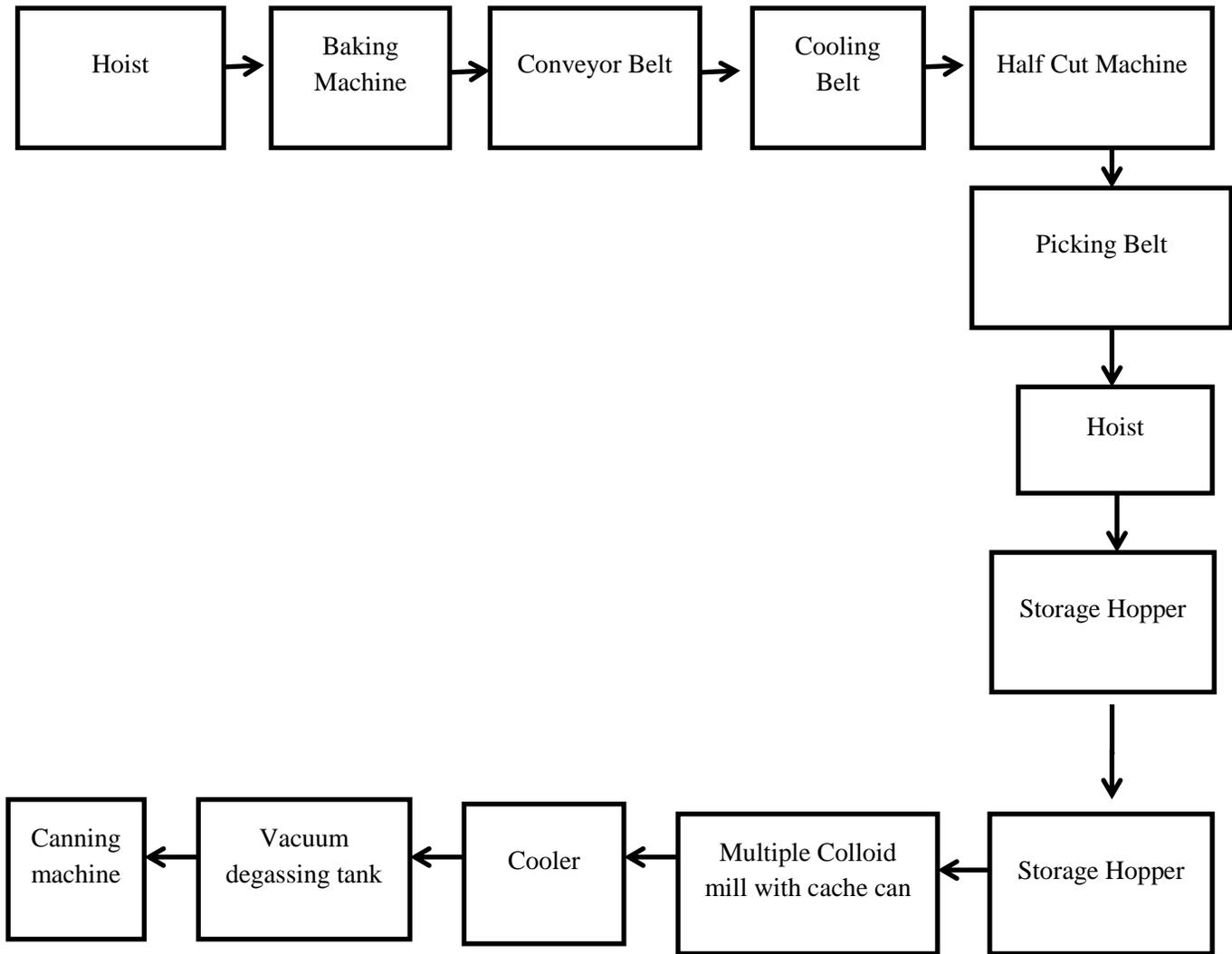


Figure 3: Typical plant layout for peanut butter manufacture

CHAPTER- 4

PRODUCT SPECIFICATIONS, STANDARDS AND FOOD SAFETY REGULATIONS

4.1 FSSAI standards for peanut butter

According to FSSAI, peanut butter is defined as “a cohesive, comminuted food product from clean, sound, shelled peanuts or groundnuts (*Arachis hypogaea* L.) by grinding roasted mature kernels removed of the seed coat.” Further, addition of sugar, edible oils and fats and liquid glucose could be added as per regulations. Table 2 states the specifications as per FSSAI for the peanut butter.

S. No.	Parameters	Limits
1.	Moisture	Not more than 3.0 percent by weight
2.	Fat	Not less than 40.0 percent by weight (db)
3.	Protein	Not less than 25 percent by weight (db)
4.	Total ash	Not more than 5.0 percent by weight (db)
5.	Acid value of extracted fat	Not more than 4.0
6.	Salt as NaCl	Not more than 2 percent by weight

Adopted from FSSAI (2020)

Following considerations have to be met as per regulations.

1. The product should not contain argemone oil.
2. Food additives as specified in FSSAI regulation Appendix A could be added as permitted.
3. The product should comply with food safety regulations (2011) for toxins, contaminants and residues
4. Food Safety and Hygiene standards as prescribed by Schedule 4 of the FSSAI, Licensing and registration of business regulation should be thoroughly followed.

5. The microbiological standards of the product should meet as prescribed by the Appendix B of the FSSAI regulations.
6. The packaging and labelling requirements should comply with the provisions as laid by FSSAI (2011) are discussed in the Chapter 3.
7. Sampling and sample analysis: the specific protocols for the analysis and sample are given in Manual of Methods of Analysis of Food (FSSAI, 2011).

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