A DISSERTATION ON

Studies on storage stability of high fiber fat sugar reduced pea pod powder based cookies

SUBMITTED TO THE
DEPARTMENT OF BIOENGINEERING
FACULTY OF ENGINEERING & INFORMATION TECHNOLOGY
INTEGRAL UNIVERSITY, LUCKNOW

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FOR THE
DEGREE OF MASTER OF TECHNOLOGY
IN FOOD TECHNOLOGY

BY
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B.Tech.- M.Tech. Dual Degree Food Technology (X Semester)
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UNDER THE SUPERVISION OF

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DECLARATION FORM

I, **Shalini Singh**, a student of **B.Tech.- M.Tech. Dual Degree Food Technology** (V year/ X Semester), Integral University have completed my six months dissertation work entitled **"Studies on storage stability of high fiber fat sugar reduced pea pod powder based cookies"** successfully from **Integral University Lucknow** under the able guidance of **Mrs. Gazia Nasir**.

I, hereby, affirm that the work has been done by me in all aspects. I have sincerely prepared this project report and the results reported in this study are genuine and authentic.

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CERTIFICATE

Certificate that Ms **Shalini Singh** (Enrollment number 1800103037) has carried out the research work presented in this thesis entitled **"Studies on storage stability of high fiber fat sugar reduced pea pod powder based cookies"** for the award of **B.Tech.- M.Tech. Dual Degree Food Technology** from Integral University, Lucknow under my supervision. The thesis embodies results of original work and studies carried out by the student himself and the contents of the thesis do not form the basis for the award of any other degree to the candidate or to anybody else from this or any other University/Institution. The dissertation was a compulsory part of her **B.Tech.- M.Tech. Dual Degree Food Technology** degree.

I wish her good luck and bright future.

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TO WHOM IT MAY CONCERN

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I wish her good luck and bright future.

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Place: Lucknow **Shalini Singh**

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Abstract

The study assessed the impact of storage temperature and packing materials on the qualitative characteristics of cookies made with pea pod powder and controlled cookies made with wheat flour and barnyard millet flour. Low density polyethylene (LDPE) and aluminum pouches were utilized for the storage studies of the control cookies, while LDPE and aluminum pouches were employed at two different temperatures, namely 25°C and 38°C, for the cookies made with pea pod powder. Quality metrics including moisture content, free fatty acid content, peroxide value, hardness, microbiological analysis, and sensory evaluation of created goods were assessed every 15 days during the course of their three-month storage term. Cookies packaged in LDPE showed greater quality alterations than cookies packaged in aluminum pouches. In terms of overall acceptability, the sensory scores of cookies packaged in LDPE decreased more than those packaged in aluminum pouches. According to the findings of the aforementioned study, cookies can be stored in aluminum pouches for up to three months at two different temperatures.

Keywords Pea pod powder, Packaging materials, Shelf life stability, Free fatty acids, Peroxide value and Total plate count.

CHAPTER 1

INTRODUCTION

Green peas (*Pisum sativum L.)* are a significant crop in India and China produces the most of them globally. Both the green and the dried varieties are eaten all throughout the world. According to FAOSTAT's estimates for 2020, India produced about 5.703 million tons of peas. However, the outermost pod is frequently wasted and not effectively utilized when peas are processed into different food formats, such as frozen, dried, or canned. A hazard to both human and animal health can result from improper handling or disposal of the outer seed coat, which makes about 35–40% of the fresh weight of peas. Pea pods have a wide range of possible dietary uses and are a great source of fiber, carbs, and crude proteins. Their low calorific value of 210 kcal/100 g is a result of their low fat content, and they also contain large amounts of macronutrients like potassium, magnesium, iron, and zinc. After the peas are removed, about 35 to 40 percent of pea pods are wasted. According to studies by Mateos-Aparicio et al. (2010) and Fendri et al. (2016), these pea pods contain a range of nutrients, including total carbohydrates (24.34%), starch (6.81 g/100 g), crude fat (1.30%), protein (10-12%), glucose (12.0%), sucrose (8.0%), dietary fiber (58.0%), potassium (0.40 mg/100 g), calcium (3.04 mg/100 g), copper (0.06-1.08 mg/100 g), sodium (1.73 mg/100 g), magnesium (0.50 mg/100 g), iron (1.2 mg/100 g), and zinc (0.16-0.92 mg/100 g). Additionally, carotenoids, which are naturally occurring colorful pigments, are found in pea pods and have antioxidant properties (Narayanan et al., 2015). Proanthocyanidins, luteolin, apigenin, and quercetin glycosides, which may offer protection against chronic diseases, cancer, and other inflammatory-related disorders, are known to be present in peas, particularly in the seed coat (Shahidi and Yeo, 2018). Pea pods also include phenolic acids including cinnamic acids (5-caffeoylquinic acid, rosmarinic acid, and quinic acid) and benzoic acids (such as gallic acid). Additionally, pea pods contain polyphenols like flavonoids, including flavanones (naringenin and hesperidin), flavanols (epicatechin and catechin), flavonols (quercetin, quercetin 3-galactoside, and rutin), flavones (luteolin, diosmin, apigenin, and kaempferol 3-glucoside), and isoflavones (genistein and myricetin) (Castaldo et al., 2022).

As a small millet crop in Asia, barnyard millet, also known as Echinochloa spp., is becoming more significant (Renganathan et al., 2020). Echinochloa esculenta (Japanese barnyard millet) and Echinochloa frumentacea (Indian barnyard millet) are two well-known and frequently farmed species within the genus. The Echinochloa genus includes 20–35 annual and perennial species that are found all over the world and can flourish in a variety of agroclimatic and environmental situations. In areas where the climate and soil are unfavorable for rice growth, it has historically been the main cereal crop (Sood et al., 2015). Self-pollinating Echinochloa frumentacea is a member of the Poaceae family. In addition to Sanwa and Jhangora in Hindi, Shyama in Sanskrit, Oodalu in Kannada, Kuthiravaali in Tamil, Udalu and Kodisama in Telugu, Shamul in Marathi, Sama in Gujarati, Shamula in Bengali, and Swank in Punjabi are some of the other names for it. It is cultivated in various countries including India, China, Japan, Malaysia, Indonesia, Africa, and the United States of America (Anuradaha et al., 2014).

According to Kumar et al. (2000), it is mostly grown in the Indian states of Orissa, Maharashtra, Madhya Pradesh, Tamil Nadu, Bihar, Punjab, Gujarat, and the mountainous areas of Uttarakhand. From the Himalayan region in the north to the Deccan region in the south, India grows barnyard millet. It is a rainfed crop grown up to 2000 meters above sea level in the hills. It is frequently planted in hilly, marginal, or tribal locations where there are few alternatives for crop improvement on slopes and undulating fields. The genus, which includes the two domesticated species, also includes 20 to 30 annual and perennial wild species that are found all over the world, many of which can coexist peacefully with rice and thrive in moist or well-watered environments (Hilu, 1994). Due to its rich nutritional profile and potential health advantages, the grain known as barnyard millet has grown in popularity. It offers a balanced nutritional profile with 10.1% protein, 8.7% moisture, 3.9% fat, 6.7% crude fiber, 2.0% total fat, 68.8% carbs, and 398 kcal/100g energy (Ugare et al., 2014).Diverse businesses are working to create affordable and readily available solutions that match consumers' expectations about protein content and high fiber content in response to the increased demand for wholesome and nutritious food items (Thongram et al., 2016). Barnyard millet offers a chance to produce novel items such puffs, biscuits, noodles, pasta, and fermented foods (Thongram et al., 2016). The improvements In bakery processing, a lot of attention has been paid to creating goods that are simple to digest and suitable for those with diabetes, digestive problems, or cardiovascular concerns. Due to their high protein content, superior dietary fiber, and favorable absorption qualities in the human gut, millets, notably barnyard millet, have become attractive components for these baked goods (Dayakar Rao et al., 2017). Significant levels of healthy unsaturated fats, like oleic acid and linoleic acid, can be found in barnyard millet. As a result, Dayakar Rao et al. (2017) recommend it for those with diabetes mellitus and cardiovascular illnesses. A intriguing strategy for creating bakery goods that are not only nutrient-rich but also high in fiber is combining pea pods and barnyard millet. This combination has the potential to meet the global demand for such products (Dayakar Rao et al., 2017).

Natural sweeteners have grown significantly in prominence in recent years because of their many advantages. The Paraguayan herb called Stevia Rebaudiana is one such source of sweet substances. It contains steviol glycosides, which are allowed as food additives and are around 300 times sweeter than sucrose. According to rules established by the European Union, however, their use is restricted in some food categories (European Commission, 2011; Lemus-Mondaca et al., 2012). Stevia extracts have demonstrated a wide range of health-promoting qualities, in addition to antioxidative and antibacterial capabilities. It is especially advised for those with diabetes due to one of its most major health benefits—the capacity to lower postprandial glucose and insulin levels (Goyal et al., 2010; Madan et al., 2010). Because it is 100% natural, calorie-free, and 250–300 times sweeter than table sugar, stevia has many benefits over conventional sugar. It is advised for obese people since it is stable at high temperatures up to 200 °C, cannot ferment, improves flavors, prevents cavities, and all of the aboveThe leaves can also be used in their natural state, it is non-toxic, it is not addictive, and it can be used in baking and cooking (Panpatil and Polasa, 2008).Additionally, Stevia Rebaudiana leaves are used to make canned goods and confectionary items. Natural resources with a sweet taste but no negative effects on human health are in higher demand. One such plant is the Stevia Rebaudiana, which is widely used in the United States, France, South Korea, Canada, Russia, Ukraine, and China. For decades, these nations have exploited the 6-7% stevioside included in Stevia Rebaudiana leaves to produce candy, health drinks, and canned foods. It has been established that the principal sweet-tasting substance in Stevia Rebaudiana leaves, stevioside, is not mutagenic or carcinogenic subtances. Consequently, it has received widespread approval from healthcare groups. The Asteraceae family includes the perennial herb Stevia Rebaudiana, which is native to Paraguay and has been used there by the locals for more than 1,500 years.

The nutritional advantages of inulin and oligofructose are becoming more and more clear as scientific understanding grows (Roberfroid, 1997; Roberfroid & Delzenne, 1998). These compounds are well-known examples of active food components in the field of "functional foods" because they provide a distinctive combination of nutritional and technological advantages (Van Loo et al., 1999). There is a high demand for fat mimics or substitutes in food products in the effort to reduce calorie, fat, and cholesterol intake (Haque & Ji, 2003). When using such alternatives, it can be difficult to preserve desired sensory characteristics such breaking strength, crumb texture, moisture content, and mouthfeel (Zahn, Pepke, & Rohm, 2010). It has become clear that inulin, a carbohydrate-based fat substitute, is a good choice for offering low-calorie functionality. Around the world, inulin and oligofructose are utilized to increase the fiber content of many food products. They have been used to successfully replace fat in spreads, baked goods, and other products. The dietary fiber known as inulin is principally found in the roots of endive (Cichorium intybus L.), but it is also present in a number of other edible fruits and vegetables, including tomatoes, bananas, chicory, garlic, asparagus, and onions. The fructose units that make up the inulin used in the food industry are typically obtained from chicory and joined together by -(2, 1) bonds. Since human digestive enzymes are unable to hydrolyze inulin, it mostly survives until it enters the large intestine before being fermented by the intestinal microbiota (Roberfroid, 2007).

It is well known that increasing dietary fiber intake has a number of positive health effects, such as improved blood sugar and cholesterol control, defense against cardiovascular disease, regulation of intestinal function, promotion of gut health, and defense against colon cancer (Ktenioudaki & Gallagher, 2012).Along with bread and cakes, cookies are a common baked product that are eaten in big amounts every day and provide a practical approach to add dietary fiber and other beneficial substances to people's diets. Cookies often contain elements like flour, sugar, and fat in their final form, which results in a low water content. Researchers have explored various fiber sources to increase the fiber content in cookies, such as resistant starch (Brennan & Samyue, 2004; Laguna et al., 2011), inulin and beta-glucan (Brennan & Samyue, 2004), fibers from different fruits or wheat fiber (Bilgicli et al., 2007; Ajila et al., 2008; Laguna et al., 2014), and different wheat brans (Sudha et al., 2007; Reyes-Perez et al., 2013). The purpose of this study was to look into the alterations that pea pod powder and barnyard millet flour, which were used to make cookies, underwent after 90 days of storage at 25°C and 38°C. In addition, LDPE (Low-Density Polyethylene) and aluminum pouches were employed as packing materials. The study's objectives were to forecast the best packing material and establish the shelf life characteristics of the cookies. Moisture content, water activity, peroxide value, free fatty acid content, sensory analysis, microbiological analysis (total plate count, yeast and mold count, coliform count), color, and texture were all taken into consideration as response variables for the controlled cookies. Throughout the 90-day storage period, the cookies were examined every 15 days to determine how these parameters had changed.

1.2 Objectives:

- To develop pea pod powder based fiber enriched fat- sugar reduced cookies
- To study the effect of packaging conditions on storage parameters of pea pod powder based fat- sugar reduced cookies and control cookies
- To study the effect of temperature on storage parameters of pea pod powder based fat- sugar reduced cookies and control cookies

CHAPTER 2

REVIEW OF LITERATURE

2.1 Peas

The scientific name for peas, Pisum sativum L, is a member of the Leguminosae family and is a nutrient-rich agricultural product. They are a widely grown crop all over the world because of their extraordinary capacity to tolerate freezing conditions (Kour et al., 2020; Tulbek et al., 2017). Just behind China, India is the second-largest producer of green peas in terms of production, and it is ranked tenth among vegetable crops. According to FAOSTAT, 2019; Mondor, 2020; Senapati et al., 2019; the annual global production of green peas and dry pea seeds is roughly 14.5 million tons and 22 million tons, respectively. Peas are a common delicacy that is eaten all across the world, both fresh and dried. According to Nasir et al. 2022, India alone produced 5.7 million tonnes of peas in 2020. However, the outer pod is frequently destroyed when peas are processed into different food types like frozen, dried, and canned. This outer seed coat is rich in fiber, carbohydrates, crude proteins, and macronutrients like potassium, magnesium, iron, and zinc. It makes up 35–40% of the fresh weight of peas. Due to its low fat content, it has a low calorie value of 210 kcal/100g (Nasir et al., 2022). Without proper use or disposal, the pea pods can serve as a breeding ground for infections, endangering both human and animal health. However, because they are a valuable source of nutrients, these pods could be used in food applications. Peas are additionally extensively available all around the world because to their abundance, affordability, and ease of cultivation. Their use has been linked to a number of health advantages, such as laxative, anti-cancer, anti-obesity, astringent, and cardio-protective actions (Zilani et al., 2017).

2.2 Pea pods

Substances with limited uses and frequent excretion during processing are referred to as by-products, which causes unwelcome environmental damage. The pods or peels of green peas, which make about 55% of the total volume of freshly harvested peas, are regarded as waste byproducts. The possible uses of these peapods have gotten little consideration and are often dumped as garbage. Peapods, on the other hand, are rich sources of phenolic antioxidants and natural food bioactive components, making them significant agro-industrial by-products (Guo et al., 2019; Hanan et al., 2020). Every year, almost 1 million tons of peapod trash are wasted in India alone. The synthesis of cellulolytic enzymes from peapod waste and its usage as animal feed have both been investigated in various studies (Vinay, 2018). Peapods made from mature, fresh green peas include a large amount of dietary fiber, protein, and calcium compared to by-products from broad bean pods and okra. They were successfully employed to make biscuits using 20% peapod powder, and the results were well-received (Garg, 2015).

2.2.1 Nutrient profile of pea pod

Pea pod by-product, also known as the leftover material from pea pods, is mainly made up of fiber, carbs, and crude proteins with very little fat. Potassium and magnesium are the key macroelements and iron and zinc are the main microelements in pea pods. The energy value of the pea pod matrix is reduced, measuring 210 kcal/100 g due to the low concentration of lipids present (Mejri et al., 2019). In a study on the antioxidant and hepatoprotective characteristics of a pea pod extract that was fractionated using bioassay-guided techniques, Seida et al. (2015) conducted a bioassay**.** There were concentrations of 61.27%, 14.6%, 0.79%, and 8% of carbs, crude protein, fat, and crude fiber in the by-product of the pea pod, respectively, on a dry basis. The by-products of pea also contained minerals like potassium, calcium, and magnesium at concentrations of 25.78%, 8.29%, and 6.82% respectively, as well as vitamins B1, B2, and C at concentrations of 1.61 mg, 0.4 mg, and 34.65 mg per 100 g, respectively. According to Mateos-Aparicio et al. (2010), these results are consistent with earlier literature publications on the composition of pea pod powder in terms of sugar concentration**.**

2.2.2 Carbohydrates present in the pea pod

2.2.2.1 Sugars

Sugar is a vital part of carbohydrates and can be found in many different foods, including fruits, vegetables, cereals, and milk. Additionally, flavored yogurt, sweetened beverages, baked goods, cereals, and other products frequently utilized in the food business all contain added sugars. There are many different types of carbohydrates, such as monosaccharides and polysaccharides, which have nutritional advantages and are used for a variety of things in the food industry. Sugars play a variety of tasks in food, in addition to adding sweetness (Zaitoun et al., 2018). These jobs include preservation, functioning as antioxidants, and increasing color, taste, and texture.Significant levels of soluble sugars, such as sucrose, glucose, xylose, and fructose, can be found in pea pods. According to research by Rudra et al. (2020), pea pods have the following amounts of sucrose, glucose, and xylose per 100 grams: 10.508, 1.07, and 1.786, respectively.

2.2.2.2 Dietary fiber

Insoluble and soluble dietary fiber can be divided into two groups. According to Fuentes et al. (2010), it is predominantly made up of carbohydrate polymers produced from plants, such as gums, pectin, cellulose, inulin, hemicelluloses, resistant starch, and a few non-carbohydrate components.Dietary fiber is a group of indigestible polysaccharides found non plants, such as gums, cellulose, pectins, hemicelluloses, oligosaccharides, and different lignified compounds, according to the Food and Agriculture Organization (FAO). These polysaccharides may exist naturally or may be triggered by mechanisms like enzymatic or chemical ones. For instance, while inulin requires fermentation by microflora to form short-chain fatty acids, which can aid in the prevention of gastrointestinal diseases, cellulose and hemicellulose directly encourage bowel motions (Liu et al., 2015).

2.2.3 Crude protein

By acting as intermediates and the building blocks for proteins, amino acids are essential for metabolism. To sustain optimal physiological activities, it is crucial for humans to consume an adequate number of essential amino acids of excellent quality (Resign et al., 2013). Nitrogen-containing proteins are essential for the growth and development of tissues. They provide a number of advantages, including the management of weight, augmentation of thermogenesis and satiation, improvement of high-density lipoprotein cholesterol levels, and promotion of bone mineralization (Hofman & Falvo, 2004). Numerous studies have demonstrated that unhealthy eating patterns and a lack of exercise greatly contribute to the increased prevalence of diabetes. Therefore, dietary changes can aid in the management or prevention of diabetes**.** Legumes, with their unique nutritional profile characterized by high fiber content, low glycemic index, and low fat content, offer a viable dietary intervention for diabetes control (Chaubey et al., 2019). Pea pods, in particular, are rich in proteins, making them the second major component after dietary fiber.

2.2.4 Fatty acid profile

Pea pods contain considerable levels of unsaturated fatty acids, with linoleic acid (C18:2n-6) and linolenic acid (C18:3n-3) being the main ones, according to research by Mejri et al. (2019). On the other side, the two main saturated fatty acids were found to be palmitic acid (C16:0) and stearic acid (C18:0). It was discovered that the abundance of unsaturated fatty acids was greater than double that of saturated fatty acids. The fatty acid composition of pea pods was different from that of Spanish samples, interestingly, with the former exhibiting a larger amount of C18:2 and C18:3 than C16:0.High h/H ratios and low levels of the atherogenic and thrombogenic indices (AI and TI) are suggested by the ratios of unsaturated to saturated fatty acids (UFA/SFA) found in pea pod lipids**.**

2.2.5 Minerals

The macro- and micro-elements found in pea pods are abundant. They are rich in potassium, calcium, and magnesium, with iron and zinc serving as the major microelements (Mateos-Aparicio et al., 2010).Pea pod, okara, and broad bean byproducts were the subjects of a comparative investigation by Mateos-Aparicio et al. (2010), and it was discovered that calcium is present in significant amounts in all three byproducts. Pea pod, broad bean, and okara had the greatest calcium availability among them. The research also showed that iron is the main constituent in pea pods, followed by broad bean and okara. Zinc is reported to be well bioavailable in legumes, including peas. Legumes can be used to absorb about 25% of the daily recommended zinc intake (Sandstorm et al., 1989). Male reproduction, cellular defense, and the generation of structural proteins all depend on zinc (Aghaei et al., 2014).

2.2.6 Bioactives present in the pea pod

The extraction of bioactive compounds has received substantial study and is regarded by numerous researchers as a successful method for reusing food processing by-products. When paired with current technological developments in molecular identification and separation techniques, this procedure is especially useful. To determine the antioxidant activity of pea pods and pea grains and to pinpoint their bioactive constituents using GCMS analysis, Saha et al. (2014) undertook a thorough investigation of both. The investigation found different chemicals in the pea grains and pods. Furthermore, chromatography methods revealed that the quantity of bioactive substances present in pea cotyledons was almost identical to that discovered in pea pods, indicating that eating pea pods is just as advantageous as eating pea grains (Saha et al., 2014). Mejri et al. (2019) conducted a study in Tunisia where they looked into the chemical makeup of pea pods (variety Basma). They measured the amounts of phenolic content, flavonoid content, and condensed tannins in the methanolic extract of the pods. It was discovered that each gram of extract has a phenolic content of 32 mg GAE (Gallic Acid Equivalent), a flavonoid content of 22 mg QE (Quercetin Equivalent), and a condensed tannin content of 48 g CE (Catechin Equivalent).

2.2.7 Pharmacological benefits

Recent research has illuminated the many pharmacological benefits connected to pea pods. This section seeks to explore these advantages in depth and give a thorough explanation.

2.2.7.1 Antidiabetic, renoprotective, and reproprotective activity

Due to the adverse effects of commercially available medications on the human body, researchers are becoming increasingly interested in investigating alternative treatments for people with type 2 diabetes (Arun et al., 2015). Mejri et al.'s (2019) study used a single intraperitoneal injection of 160 mg/kg bw of alloxan monohydrate to cause diabetes in mice following a 16-hour fast. The mice were given 5% glucose throughout the night to prevent hypoglycemia shock brought on by the drug's abrupt release of insulin. Blood samples from the mice's tail veins were collected after an 18-hour fast, and an Accu-Chek glucometer was used to determine their blood glucose levels. According to Bagri et al. (2009), diabetic mice had fasting blood glucose levels greater than 13 mM/L.The mice were separated into four groups, each with six mice, and were given oral administration of pea pod extract (PPE) once daily at a dose of 500 mg/kg through gavage. Both healthy and diabetic mice had their serum biochemical indicators evaluated. By measuring serum biochemical indicators like glucose, albumin, uric acid, urea, creatinine, total cholesterol, triglycerides, low-density lipoprotein (LDL), and high-density lipoprotein (HDL), the study also evaluated the anti-diabetic, kidney-protective, and reproductive-protective effects of PPE.

2.2.7.2 Hepatoprotective activity

According to Brockmoller and Roots (1994), the liver is a crucial organ in the human body that is involved in a number of metabolic activities. Numerous conditions, such as drug addiction, virus infections, alcohol intake, and exposure to chemical and biological toxins, can result in liver damage. According to Sturgill and Lambert (1997), these injuries can trigger autoimmune assaults against hepatocytes. Mejri et al.'s (2019) histological study on diabetic mice treated with alloxan revealed several types of liver injury, including lymphocyte infiltration, hepatocellular necrosis, central venous congestion, and sinusoidal dilatation. Interestingly, liver damage were dramatically reduced by oral administration of pea pod extract at a concentration of 500 mg/kg body weight. In another study, Seida et al. (2015) used a rat model of liver injury and oxidative stress generated by CCl4 to examine the hepatoprotective and antioxidant effects of a hydroalcoholic extract derived from pea (Pisum sativum L.) by-products.

2.2.7.3 Antibacterial activity

Previous studies have shown that plants with high flavonoid and polyphenol concentrations have antibacterial effects. In a recent scientific study, the antibacterial properties of a pea peel extract were assessed against a range of microorganisms, including Gram-positive Staphylococcus aureus, Gram-negative Escherichia coli, Pseudomonas aeruginosa, and Salmonella enterica. Both the methanolic and ethyl extracts shown strong antibacterial activity against a number of tested Gram-positive and Gram-negative bacteria. With a minimum inhibitory concentration (MIC) value of 350 g/mL, Pseudomonas aeruginosa showed the highest susceptibility to the ethyl acetate extract among them. On the other hand, according to Hadrich et al. (2014), the extract of ethyl acetate had the highest MIC value for E. coli (850 g/mL).

2.2.7.4 A-Amylase inhibition activity

The main enzyme, known as -amylase, is responsible for the early breakdown of starch during digestion. According to Tarling et al. (2008), it is essential in activating the starch hydrolysis process, which in turn results in the presence of glucose in our meals. Controlling postprandial hyperglycemia, a condition frequently linked to the onset of type 2 diabetes mellitus, requires proper management of this enzyme. According to a study by Hadrich et al. (2014), pea peel extract has the potential to control blood sugar levels by inhibiting the activity of -amylase.

2.2.8 Pea pod applications in food products

The different uses of pea pods are compiled in Table 1, and we shall go into more depth about each use below. In a study by Hanan et al. (2021), pea pod fiber powder was employed to extend the shelf life of buckwheat bread. The scientists found that adding pea pod powder to the bread decreased its extensibility, stickiness, resistance to extension, and work of adhesion. The synergistic effects of the soluble and insoluble fibers in pea pod powder improved the texture of the bread crumb while also leveling out some characteristics. Although the crumb's specific volume decreased, its moisture content increased. Overall, adding pea pod powder to bread improved its texture in a way that was pleasing to the consumer. Pea pod powder was used as one of the ingredients to improve the functionality of mayonnaise in a recent study by Rudra et al. (2020). By adding pea pod powder, the researchers were able to create an eggless mayonnaise with reduced fat content that was also enriched with dietary fiber and other micronutrients.

Table 2.1: Applications of the pea pods in various food products

Food product	Extract used	Analysis performed	Main findings	References
Bread enriched with buckwheat	PPP	Texture, extensibility, stickiness	Decrease in stickiness, work of adhesion, extensibility and resistance to extension	Hanan et al .,(2021)
Mayonnaise (eggless)	PPP	Sensory, texture, rheology	Acceptable range of PPP−7.5% enhanced stability and structural strength	Rudra et al ., (2020)
Instant Pea Soup Powder	PPP	Proximate, rehydration ratio, rheology	Acceptable range−12.5%	Hanan and Rudra ,(2020)

			Dietary fber levels improved from 7.47% in control to 13.25% on inclusion of PPP	
Biscuits	PPP	Proximate, water activity, sensory	Acceptable range of PPP−20%	Garg,(2015)
Bread	PPP	Physical characteristics, texture, color, sensory	Enhanced configuration curve and deformation energy Improved textural attributes such as springiness, adhesion and cohesion of the dough	Belghith et al., (2016)
Cake	PPP	Texture, color, sensory	Acceptability as a dietary fber source was>15% of PPP. Slight increase in hardness of the cake	Belghith et al ., (2016)

A useful cellulose material was successfully recovered from agricultural waste from pea (Pisum sativum L.) and broad bean (Vicia faba) pods in a study by Kassab et al. done in 2020. They used a variety of procedures to extract cellulose nanocrystals (CNC) and cellulose microfibers (CMF) from the pods, including bleaching, acid hydrolysis, and alkaline extraction. CMFB and CNCB were the names given to the extracted cellulose components from broad bean pods, whilst CMFP and CNCP were given to those from pea pods. The diameter of the pure CMFB was 14.67 2.29 mm, and its crystallinity was 70%. The diameter and crystallinity of pure CMFP, on the other hand, were 10.68 mm and 79%, respectively. Needles-like structures with diameters of 6.3 nm and 5.4 nm and lengths of 529 nm and 483 nm, respectively, were visible in the CNCB and CNCP. These measurements led to aspect ratios for CNCB and CNCP of 84 and 89, respectively.

2.3 Millets

Millets, which may have been the earliest cereal grain utilized for household uses, are a historical and vital source of nutrition for humans. They are classified as several kinds of small-seeded annual grasses found in five genera: Panicum, Setaria, Echinocloa, Pennisetum, and Paspalum in the Paniceae tribe, and Eleusine in the Chlorideae tribe. They are all members of the grass family Graminae. The origins of millet are numerous, coming from both Asia and Africa. Millets have long been a mainstay crop for populations in semi-arid tropical regions of Asia and Africa, where other crops find it difficult to grow. There are almost 6,000 different types of millet planted worldwide, and these grains have been cultivated since antiquity. Millets are still underused in many wealthy nations, despite their promise. However, there is a lot of room for improvement in the millet grain processing industry (Chandrasekara and Shahidi, 2010).

2.3.1 History

Ethiopia, where millet has been used since ancient times, is said to be where it first appeared (FAO, 1995). Even in the Bible, millet is mentioned as a component of unleavened bread (Leviticus 27:16). The ancient flatbread known as injera is made from finely ground millet, which is still a crucial food staple in Africa today (FAO, 1995). Consumption of millet has a lengthy history in both Asia and India. The Indian flatbread known as roti is made in these areas from ground millet seeds (FAO, 1995). Before the invention of potatoes and corn, millet developed into a major grain in Europe during the Middle Ages, especially in the nations of Eastern Europe (FAO, 1995). In the 19th century, the Setaria millet variety was introduced to the United States (FAO, 1995).

2.3.2 Millet production

The top twenty millet-producing nations, according to the Food and Agricultural Organization of the United Nations, are China, Burkina Faso, Mali, Sudan, Uganda, Senegal, Chad, Ethiopia, Nepal, Tanzania, USA, Pakistan, Myanmar, Ghana, Ukraine, and Angola. Around 60% of the world's millet is produced in South and East Asia, with 14% in Eurasia and Central Asia, 16% in Africa, and 10% in the rest of the world. India is currently the world's top millet producer, contributing 33–37% of the 28 million tonnes produced globally. Minor millets are farmed on 7 million hectares of land in India alone, producing 5 million tons of grains. According to Phanikumar (2010), the variety of millet varieties found in the dry plains of southern India is comparable to that found in Africa.India contributed 2.9 million tonnes of finger millet to the 37.0 million tonnes of millets produced worldwide between 1993 and 1994 (Agricultural Situation in India, 1993).

2.3.3 Nutrients profile

The coarse grain known as millet, also referred to as nutri-cereal, has many health advantages. It is distinguished by having more complex carbs, resistant starch, and slowly releasing sugars than other foods. Additionally high in fiber, millets have soluble fiber content that ranges from 3.4% to 6.5%. They typically have a low fat content of 1.1% to 5.0%. The B vitamins niacin, pyridoxine, and folic acid are particularly rich in millets. Millets are also a good source of calcium, iron, potassium, magnesium, and zinc. Millets outperform wheat and rice in terms of quantity and quality of nutrients (Kumar, 2010). About 8.0% of millets are protein, and 4.0% are fat. They include a lot of vitamins and minerals, calcium being particularly prevalent. Millets have a comparatively high dietary carbohydrate content.

2.4 Barnyard millet

A prominent tiny millet crop in Asia, barnyard millet, which is a member of the Echinochloa genus, has seen significant development in global output. The two main species of the Echinochloa genus, Echinochloa esculenta and Echinochloa frumentacea, are both widely farmed for use as human food and animal feed. This particular millet species is recognized for its nutritional composition, which includes significant amounts of protein, carbs, fiber, and crucial minerals like iron and zinc compared to other main grains. Additionally, because it is gluten-free and has a lower glycemic index, barnyard millet provides extra health advantages in the fight against conditions including obesity, diabetes, blood pressure control, cardiovascular illnesses, and celiac disease. Due to its low carbohydrate content, barnyard millet is a wonderful natural resource for the fast-paced modern lifestyle and promotes calm digestion. Linoleic acid, palmitic acid, and oleic acid are the three main fatty acids found in barnyard millet.

2.4.1 History and Origin

Historically grown in warm and temperate climates, barnyard millet is now most common in semi-arid tropics of Asia and Africa (Renganathan et al., 2020). In nations like India, China, Japan, and Korea, it is widely grown (Renganathan et al., 2020). From the Himalayan region in the north to the Deccan plains in the south, millet is grown throughout hilly terrain in India (Sood et al., 2015). The Echinochloa genus has 35 species, the majority of which are endemic to Southern or Eastern Asia and are referred to as barnyard millet (Upadhyaya et al., 2015). These species are found in milder and temperate climates. According to Renganathan et al. (2020), the Indian variation of millet is thought to have evolved simultaneously in India and Africa from an undomesticated species of Echinochloa, namely Echinochloa Colona (jungle rice). Despite being a very old crop, the genetic origin or source of barnyard millet is not yet known (Mitsui et al., 2020).

2.4.2 Chemical and Nutritional Composition

Barnyard millet grain is a highly nutritious crop that offers various health benefits. It is rich in protein, carbohydrates, fiber, and essential micronutrients such as iron and zinc (Ranganathan et al., 2020). Despite its numerous advantages in terms of agriculture and nutrition, barnyard millet has not been widely utilized. The grain contains approximately 6-13g of protein, 55-65.5g of carbohydrates, 2-4g of fat, 9.5-14g of crude fiber, and 3.8-4.5g of mineral matter. It also provides essential minerals like calcium (11-27.1mg), phosphorous (280-340mg), and iron (15-19.5mg). Starch, which accounts for 51-62% of barnyard millet’s composition, is its primary source of energy. Starch consists of two polymeric molecules, namely amylose and amylopectin, with amylose comprising about 20.0% (Vipin et al., 2021).

Table 2.2: Amino acid content

Amino acid	**Composition (mg / g)**
Isoleucine	288 – 372 mg
Leucine	725 – 762 mg

Lysine	106 – 136 mg
Methionine	131 – 133 mg
Cysteine	175 – 210 mg
Phenylalanine	362 – 430 mg
Tyrosine	119 – 150 mg
Threonine	35 – 263 mg
Tryptophan	63 mg
Valine	388 – 415 mg

Muthamilasaran et al., 2015 Lim et al., 2011

2.4.3 Antinutritional Content of Barnyard Millet

Barnyard millet contains certain compounds that are considered antinutritional, such as α-amylase inhibitor, trypsin inhibitors, phytate, and tannins. Phytates, also known as phytic acid, serve as a protective mechanism against oxidative stress by binding to iron involved in the Fenton’s reaction. Some phenolics and tannins in barnyard millet also act as antioxidants (Panwar et al., 2016).Humans cannot absorb phytate and lack the ability to hydrolyze it, which results in decreased bioavailability of minerals. Phytate, being a negatively charged ion, negatively affects the absorption of positively charged divalent and trivalent minerals like zinc, iron, calcium, and magnesium in pH-sensitive areas (Dhewa et al., 2021). The presence of antinutrients in barnyard millet forms complexes with essential dietary minerals such as zinc, calcium, and iron, leading to a significant reduction in their bioavailability and making them less usable by the human body (Sood et al., 2015). Consequently, reducing the level of phytic acid is desirable.

2.4.4 Antinutrients Reduction Strategies

Various processing methods have been employed to decrease the concentration of anti-nutrients in food, such as decortication/dehulling, soaking, germination, and fermentation. Decortication involves removing the outer covering of grains known as the pericarp, which used to be done manually but is now typically accomplished with the help of huller or milling machines. Decortication effectively reduces the levels of phytic acid and polyphenolic compounds in barnyard millet. De-hulling, which refers to removing the bran layer, also reduces the presence of anti-nutrients. The concentration of condensed tannins is likewise lowered through dehulling.

Recent research has shown that this method can decrease phytic phosphorus content by 23% in barnyard millet. Clearly, removing the pericarp and aleurone layer (outer parts) of grains decreases the content of anti-nutritional factors. Thus, decortication is a suitable method for eliminating phytates and tannins (Dhewa et al., 2021).Soaking is a simple method for reducing anti-nutrient content. Studies have reported that soaking grains in water for 12 to 18 hours is quite effective in reducing soluble phytic acid and proteolytic enzyme inhibitors.

2.4.5 Utililization of barnyard millet

Millets have been a long-standing staple food in Central America, Africa, and the Indian subcontinent. They form the basis of various traditional food items, including cakes made from fermented or unfermented dough, porridge or mudde, snacks, fried products, sweet or sour local beverages, non-alcoholic beverages, and grains cooked like rice by boiling (Taylor et al., 2017; Bhat et al., 2018). Millets are chosen based on their nutritional components and characteristics, typically varying with the seasons. These grains are excellent sources of both soluble and insoluble dietary fibers, present in the seed coat and the walls of endosperm cells, unlike rice, where the bran layers contribute to its fiber content (Kumar et al., 2016).Millet is highly valued for its nutritional and nutraceutical properties, offering a rich source of minerals, dietary fiber, and phenolic compounds. It provides various health benefits, including antimicrobial, antidiabetic, anticancer, antiatherosclerogenic, antioxidant, and anti-aging effects (Kumar et al., 2016). Among the different types of millets, barnyard millet stands out for its abundance of healthy nutrients (Taylor, 2017).

2.4.6 Application of barnyard millet

Various products made from barnyard millet have been successfully developed, demonstrating the value addition this millet brings. For instance, Nishad et al. (2017) conducted a study where they prepared cupcakes by incorporating barnyard millet at different concentrations. The sensory evaluation of these millet-based cupcakes indicated a high overall acceptability, and they exhibited a higher content of total dietary fiber. The crust and crumb structure, as well as the taste and flavor characteristics, scored between 81-84%. Notably, cupcakes made from millet flour showed the lowest glycemic index (GI) of 50.8 (Patel et al., 2015). Another study conducted by Patel et al. (2015) focused on preparing idli, a traditional Indian dish, using barnyard millet. The resulting products had a very soft texture, and the consistency of the fermented batter was thick. However, the taste and flavor quality were slightly bitter to bland, though the visual appearance showcased well-cooked and soft idlis. Bhosale et al. (2015) conducted a study to prepare kheer, a traditional Indian dessert, and evaluate its sensory quality. The experiment focused on sensory evaluation tests such as color, appearance, texture, flavor, sweetness, and overall acceptance.

2.5 Inulin

Inulin and oligofructose are functional ingredients found in various vegetables and fruits and can be derived from chicory roots through industrial processes. These ingredients offer a unique combination of nutritional properties and significant technological benefits in the realm of food production. When incorporated into food formulations, inulin and oligofructose can greatly enhance the sensory characteristics, improving both taste and mouthfeel across a wide range of food applications. Oligofructose, known for its high solubility, exhibits technological properties similar to those of sugar and glucose syrups. It is commonly used in conjunction with high-intensity sweeteners. On the other hand, inulin has lower solubility but excels in enhancing the stability of foams and emulsions. When used in the form of a gel in water, it demonstrates exceptional fat-like qualities. By replacing fat and carbohydrates with chicory inulin and oligofructose, food products can achieve a nutritionally enhanced profile without compromising on taste and texture.

2.5.1 Industrial production

During the early 1990s, researchers made several efforts to extract and purify inulin and oligofructose for their potential use as dietary supplements. Nowadays, these substances are commonly found in various food products. Initially, Jerusalem artichokes, dahlias, and chicory (Cichorium intybus) were considered for industrial production in temperate regions due to their high inulin content (over 15%). However, for various reasons, chicory has become the primary focus for processing (De Leenheer, 1996).Chicory roots, resembling small oblong-shaped sugar beets, are rich in inulin, with a consistently high content (over 70% on a dry basis) year after year. The production process involves extracting the naturally occurring inulin from chicory roots, a process similar to extracting sucrose from sugar beets through diffusion in hot water. Refining techniques borrowed from the sugar and starch industries, such as ion exchangers, are then employed.

2.5.2 Food applications

Both inulin and oligofructose are useful substances since they have both technological and nutritional benefits. One of their standout advantages is their capacity to boost both the nutritional content and organoleptic quality of food products (Franck & Coussement, 1997). In comparison to conventional dietary fibers, the addition of inulin or oligofructose to bread goods and breakfast cereals offers a major improvement.The inclusion of inulin or oligofructose makes a number of enhancements to bread goods and breakfast cereals. They help extruded foods and cereals become crisper and expand, giving them a more palatable texture. These fibers also provide these goods a longer shelf life, preserving their freshness for a longer time. Inulin and oligofructose also aid in maintaining moisture in breads and cakes, extending the time that they stay fresh. Inulin and oligofructose also have the benefit of being soluble, making it possible to include them into liquid systems like beverages, dairy products, and spreads. This solubility makes it possible to include fiber in these products while maintaining their texture and consistency. The use of inulin as a dietary fiber in tablets makes it a well-liked ingredient for nutritional supplements.

2.6 Stevia

The Rio Monday valley in the highlands of northern Paraguay in South America is where stevia, formally known as Stevia rebaudiana Bertoni, was first discovered (Katayama et al., 1976). It is also known as sweet weed, sweet leaf, honey leaf, candy leaf, and sweet herbs. Stevia has had tremendous growth in acceptance across the globe and is predicted to become a significant source of high-potency sweeteners (Khan et al., 2012). This extraordinary plant's leaves are naturally sweet, making it a great option for anybody watching their intake of sugar and carbohydrates. Stevia is a popular sweetener that has 0 calories and is a great alternative to sugar and other sweeteners. Approximately 32,000 hectares of land are being used to grow stevia, with 75% of the land being in China (Kenya, 2015). According to Megeji et al. (2005), it is grown for commercial purposes in China, Brazil, Paraguay, Central America, Thailand, and Korea. One of the biggest markets for stevia is Japan (Sumida et al., 1980).

2.6.1 Harvesting

The duration of the growing season, the type of land, and the diversity of crops all influence when it is time to harvest the crops. The first harvest may typically be completed four months or so after the crop is sown, with further harvests occurring every three months after that. Mid-September to late September, when the plants have grown to a height of 50 to 70 cm, is the ideal period for harvesting. Shorter days cause flowering, hence it is advised to harvest the crop right before flowering to get the most steviol glycosides in the leaves (Sumida, 1980). Using pruning shears to remove the branches and then removing the leaves is the simplest way to harvest. Since the stem ends contain a comparable amount of stevioside to the leaves, they can also be removed and harvested. Three commercial harvests are typically attainable each year. When cutting the plants at the ground level, it is best to leave around 10 cm of the stem portion uncut.

2.6.2 Chemical Components

A plant called stevia is used as a natural sweetener. Glycosides found in its leaves are what give the plant its sweet flavor. Stevia's abundance in phenols, flavonoids, and antioxidants is another factor that contributes to its health advantages (30). Stevioside (St) and Rebaudioside A (R-A), the two primary glycosides present in stevia, are renowned for their extreme sweetness. Rebaudioside A contributes for 2-4% of the dry weight of the leaves, while stevioside often makes up 5–10% (Kinghorn, 1987). Stevia also contains minor glycosides, flavonoid glycosides, coumarins, cinnamic acids, phenylpropanoids, and essential oils, in addition to Rebaudioside C (1-2%) and Dulcoside A & C. The majority of the sweetening glycosides found in stevia leaves include stevioside (3–10%), rebaudioside–A (13%), and Rebaudioside–B, C, and D (Singh & PPS, 2015). Eight phytochemical characteristics of stevia glucosides, including dulcosides A, rebaudiosides A–E, steviobioside, and stevioside, have also been found through studies (Kinghorn et al., 1984).

2.6.3 Uses

The leaves of the stevia plant have a delicious, sweet flavor that lingers in the tongue for a considerable amount of time (Maiti & SS, 2008). The bitter substances found in the veins encapsulate the sweet substances within the leaves. Although these leaves are frequently used to make sauces, their true brilliance is found in herbal drinks and direct ingestion. You can buy stevia leaves that have been ground or powdered in bulk or in handy tea bag form. They are used as a natural sweetener, color, and flavor enhancer in a number of delectable foods, including teas, salads, fruits, and coffee, among others. The by-product industry, which includes the residual plant components after the best leaves have been plucked for tea or extraction purposes, is another large market. This industry can grow by encouraging the use of these byproducts as fertilizers and additives for animal feeding items . Stevia has the potential to be used as a natural sweetener, a source of phytosterols, and a source of chlorophyll for medicines and dental care items .

2.7 Lecithin

A combination of phospholipids, glycolipids, and triglycerides make up the compound known as lecithin. The term "lecithin" is frequently used in biochemistry to refer specifically to pure phosphatidylcholine, a phospholipid that makes up the majority of the phosphate portion of egg yolks. It can also be extracted mechanically or chemically from soy and rice beans using hexane (Mertins, 2004). Lecithin is readily accessible as a dietary supplement for therapeutic reasons in a highly refined form. Lecithin is used commercially as an emulsifier and lubricant in a variety of industries, including the food and pharmaceutical industries. For example, it is used to make coatings for food goods and as an emulsifier in the production of chocolate. Because it is naturally found in cell membranes and may be completely digested, lecithin is regarded as a safe and non-toxic surfactant that is well tolerated by the body.

2.8 High fiber cookies

Food producers are actively looking for novel sources and carriers of these molecules as a result of the increased demand for functional foods and the quest for bioactive ingredients. More consumers are looking for goods that can support their continued physical and mental health. Dietary fiber is one of the many bioactive components of food, along with antioxidants, plant sterols, pro- and prebiotics, and vitamins (Rodrguez et al., 2006). Fiber's distinct functional characteristics, such as its capacity to bind water, exchange cations, and absorb fat, are determined by its unique structural makeup and chemical makeup (Thebaudin et al., 1997; Górecka, 2004). In recent years, there has been an increase in interest in discovering new sources of dietary fiber to add to foods with a variety of useful properties. Natural ingredients including oats, peas, maize, and grain bran are the main sources of fiber-rich products on the market. These can be included in food without being altered. There are, however, some products that must be altered in order to be put to use. Industrial by-products are used because they are practical, profitable, easy to access, affordable, and have the capacity to effectively manage post-production waste [Górecka and Anioa 1999, Borycka and Górecka 2001, Figuerola et al. 2005, Nawirska and Kwasniewska 2005, Nawirska and Uklanska 2008, Borycka and Górecka 2005, Anio and Górecka 2006].Dietary fiber concentrates can be used as meal supplements or as ingredients in other preparations that are high in fiber. Oats and barley are two examples of cereal-based preparations that contain a sizable amount of -glucans. In instance, cellulose and hemicelluloses, which constitute insoluble dietary fiber, are abundant in them.

Watermelon rind powder (WRP) is a wonderful component for preparing functional meals like cookies since it is a great source of dietary fiber and bioactive chemicals. A study was done to see how the quality of cookies would change depending on how much wheat flour was replaced with WRP or high-maize starch (HMS), which ranged from 10% to 30%. The findings showed that the dietary fiber content of the cookies increased as WRP or HMS were added in increasing amounts. Additionally, the total phenolic content, 2,2-diphenyl-1-picrylhydrazyl radical scavenging activity, and ferric reducing antioxidant power all rose in direct proportion to the amount of WRP in the cookies

2.9 Storage studies of cookies

Pea pod powder was substituted with wheat flour, barnyard millet flour, and other soft ingredients to create cookies that are high in fiber and low in calories. Powdered sugar was also substituted with stevia powder. The cookie recipe also called for butter, stevia powder, milk, baking powder, baking soda, lecithin, inulin, and salt. The aim of this study was to investigate the alterations that took place in controlled cookies produced from wheat flour, pea pod powder, and barnyard millet flour throughout the course of a 90-day storage period at 25°C and 38°C. In addition, LDPE (Low Density Polyethylene) and aluminum pouches were employed as packing materials. The goal of the study was to identify the best packing material and forecast the cookie's shelf life characteristics. Moisture content, water activity, peroxide value, free fatty acid content, sensory analysis, microbiological analysis (total plate count, yeast and mold count, and Coliform count), as well as color and texture, were some of the response characteristics taken into account. Over the course of the 90-day storage period, samples of the cookies were examined for these response variables every 15 days.

CHAPTER 3

MATERIALS AND METHODS

3.1 Materials and Equipments

3.1.1 Raw materials

The primary raw materials were purchased from a nearby shop and included pea pod powder, barnyard millet and wheat flour, butter, stevia powder, milk, baking powder, baking soda, lecithin, inulin, vanilla essence, and salt**.**

3.1.1.1 Pea pod powder

Peas of the ‘karishma’ variety were purchased from a local vegetable seller in Lucknow (India). The peas were manually removed from their shells, and the resulting pea pods were washed using flowing water. To eliminate the parchment layer, it was manually peeled off. In an industrial setup, the pea pods might undergo blanching at 100 °C for 2 minutes, followed by shelling to remove the parchment layer (Rudra et al., 2020). The pea pods were then dried in a tray drier (MSW206, Delhi, India) at 60 °C for 24 hours. Afterward, they were ground into a fine powder using a laboratory-scale grinder (Model: FX 11 Food Factory, Bajaj, India) at a speed of 18,000 rpm. The resulting powder was sifted through a 60 mesh sieve to obtain a fine powder.

3.1.1.2 Barnyard millet

Barnyard millet was procured from local shop. The barnyard millet was then ground in a grinder to obtain barnyard millet flour. Then flour was sieved through 600 mesh sieve to be used in cookies manufacturing.

3.1.1.3 Wheat flour

Wheat flour was procured from local shop. The wheat flour was sieved through 600 mesh sieve for use in cookies manufacturing.

3.1.2 Equipment and instruments:

The following equipments were used for preparation of cookies and their quality characteristics.

Table 3.1:Equipment with their specifications and functions

S.No.	Equipment	Specifications	Function
1	Electronic balance	Wensor (Capacity: 400g least count: 0.01g)	For Weighing cookies flour sample for making cookies
2	Hot plate	Category-MSW-314 Model : MSHP-21C	For heating the media to determine total plate count, yeast & mold count and coliform count
3	Microwave oven	LG Model: MC2149BB	Baking the biscuits
4	Texture Analyser	Stable micro systems TA	For measuring textural characteristics of biscuits
5	Hot air oven	MSW, 10A/VA, Jawahar Nagar P.B.No. 2151, Delhi-7 Model 52250	For measuring moisture content
6	Soxhlet apparatus	FOSS ST 243 SoxtecTM	For measuring fat content

7	Electric grinder	BAJAJ FX11 1.0 Litre, Stainless steel	For grinding pea pod powder and barnyard millet flour
8	Mould Steel	(Diameter: 0.54 cm)	For cutting cookies
9	Incubator	Dual display PID controller Display SV & PV Auto tuning feature	For storing cookies
10	Laminar flow	Vertical and Horizontal 220 volts / 50 Hz	For microbial analysis
11	Autoclave	400-440V	For autoclave media

Figure-3.1 Weighing Machine

Figure-3.2 Autoclave

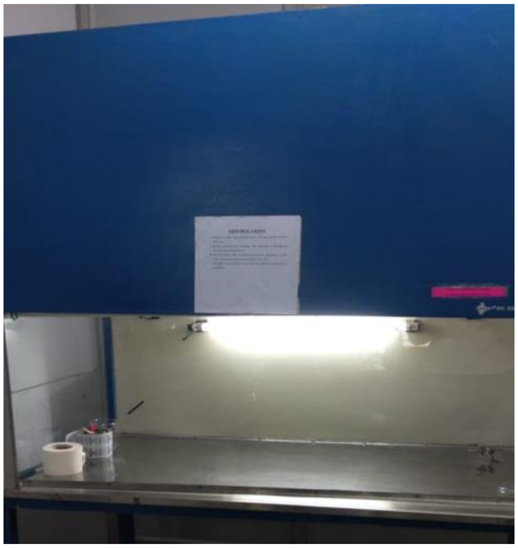

Figure -3.3 Air Laminar Flow

Figure- 3.4 Incubator

Figure -3.5 Texture Analyser

Figure -3.6 Hot air oven

Figure 3.7 : Laboratory hot plate.

3.1.3 Preparation of pea pod powder

In an industrial context, obtaining pea pod powder entails numerous procedures. The peas are first manually deshelled, removing the peas from their pods. Then, to get rid of any contaminants, these pea pods are washed under a stream of rushing water. The parchment covering the pea pods is then painstakingly removed. The pea pods may go through a process called blanching in an industrial setting, when they are heated to 100 °C for two minutes. The pea pods are softened during this blanching procedure, which also makes it easier to peel back the parchment coating (Rudra et al., 2020). After blanching, the pea pods are dried for 24 hours at 60 °C in a tray dryer (MSW206, Delhi, India). The moisture content of the pea pods is decreased as a result of the drying process. Using a lab-scale grinder (Model: FX 11 Food factory, Bajaj, India) with a speed of 18,000 revolutions per minute (rpm), the dried pea pods are ground into a powder. The ground pea pod powder is sieved using a 60 mesh sieve to create a fine powder. This sieve aids in removing any bigger particles and makes sure that the finished product has uniform particle size. For further examination and preservation, the sieved powder is subsequently kept in aluminum pouches at a temperature of 0 °C.

Procurement of peas

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Deshelling of peas

↓

Pea pods

↓

Removal of parchment layer

↓

Washing of pea pods

↓

Drying (Tray drier at 60°c for 24 h)

↓

Milling

↓

Sieving

Figure -3.6 Pea pod powder

3.2 Experimental Methodology

3.2.1 Preparation of pea pod powder based fiber enriched cookies

In the beginning, 'karishma' variety peas were bought from a neighborhood vegetable vendor in Lucknow, India. The pea pods were then rinsed in running water after being manually shelled. Manual labor was used to remove the top layer. According to Rudra et al. (2020), the pea pods could be blanched at 100 °C for two minutes in an industrial setting before being shelled to remove the outer covering. The pea pods were subsequently dried for 24 hours in a tray dryer (MSW206, Delhi, India) with the temperature set to 60 °C. The dried pods were then processed into pea pod powder using a laboratory-scale grinder (Model: FX 11 Food Factory, Bajaj, India) running at 18,000 rpm. The obtained powder was sieved through a 60 mesh sieve to create a fine powder (Rudra et al., 2020).

A nearby store provided the barnyard millet. After that, the barnyard millet was processed into flour using a grinder. When making cookies, flour was then sieved through a 600 mesh sieve.

A nearby store provided the wheat flour. For use in making cookies, a 600 mesh sieve was used to separate the wheat flour.

With a few slight modifications, cookies were produced using a technique similar to that mentioned by Nasir et al. in their work from 2020. Pea pod powder, barnyard millet flour, and wheat flour were the key ingredients utilized to make the cookies. Powdered sugar was changed to stevia powder to lower the calorie count. Other components were bought from a nearby store, including butter, stevia powder, milk, baking powder, baking soda, lecithin, vanilla essence, and salt. Sensory assessments made from earlier trials were utilized to determine the precise amounts of each ingredient used in the cookies. A microwave (model MC2149BB, LG, South Korea) was preheated to 180 °C for 2 minutes before baking the cookies. After 29 minutes of baking at the same temperature, the cookies were allowed to air dry.

Composition of pea pod powder based cookies

Table 3.2

Ingredients	Quantity (g)
Pea pod powder	5
Barnyard millet flour	10

Wheat flour	35
Lecithin	0.4
Stevia powder	0.4
Baking powder	1.5
Baking soda	0.5
Butter	26
Milk	10
Salt	0.3
Inulin	6.5
Powdered sugar	24

Cream the Amul butter (26g), inulin (6.5g), and powdered sugar (24g) for about a minute before using them to make cookies. Pea powder (5g) and barnyard millet flour (10g) should then be added to the mixture. Then manually stir in 35g of wheat flour, 1.5g of baking powder, 0.5g of baking soda, and 0.4g of lecithin powder for around 4 minutes. To physically shape the dough, use 10 ml of milk. Use a rolling plate and wooden roller that have been cleaned and dried to roll the dough to a thickness of about 7 mm. To cut out cookies that are all the same size, use a steel mold with a 5.4 cm diameter. Place the cookies on a baking sheet that is dry and clean.

For two minutes, preheat the microwave to 180°C. Bake the cookies on the baking sheet for 29 minutes at 180°C in the microwave. After baking, let the cookies cool for about 15 minutes in the air. Finally, to keep the cookies fresh, store them in aluminum pouches and LDPE (low-density polyethylene) packaging.

Figure 3.7: Flow chart of Pea pod powder based fiber enriched cookies

Pea pod powder, Barnyard millet flour, Wheat flour

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Mixing of all dry ingredients

↓

Creaming(mixing of sugar and butter)

↓

Mixing of cream with dry ingredients

↓

Preparation of small balls and roll out into sheets

↓

Cutting the sheets using cutter of dimensions 5mm

↓

Baking at 180°c for 29 minutes in preheated oven

↓

Cooling at 27°

↓

Packaging

↓

Storage at 25 C and 38 C temperature

Dry Ingredients

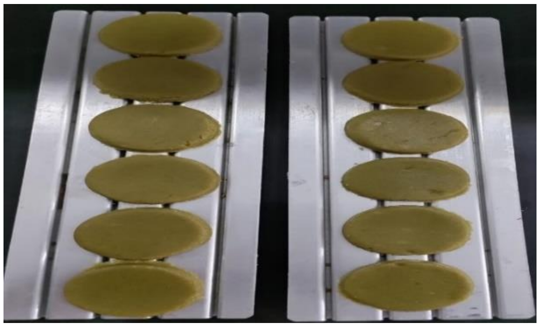
↓

Creaming(mixing of all dry ingredients)

↓

Dough making

↓

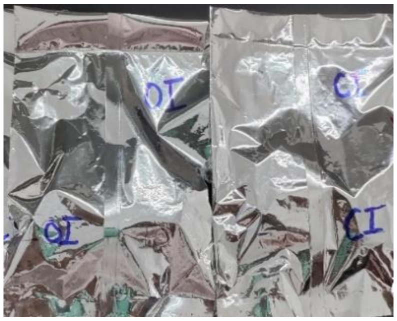


Cutting the sheets

↓

Baked Cookies

↓



Stored in Aluminium Pouches

Stored in LDPE Pouches

The control cookies were prepared by barnyard millet flour (10g), wheat flour (40g), butter (32.5g), powdered sugar (30g), salt (0.3g) baking powder (1.5g),baking soda (0.5g) lecithin powder (0.4g) but pea pod powder, Inulin and stevia were absent in these cookies.

Figure 3.8: Flow chart of controlled cookies (Barnyard millet flour)

Barnyard millet flour, Wheat flour

↓

Mixing of all dry ingredients

↓

Creaming(mixing of sugar and butter)

↓

Mixing of cream with dry ingredients

↓

Preparation of small balls and roll out into sheets

↓

Cutting the sheets using cutter of dimensions 5mm

↓

Baking at 180°c for 29 minutes in preheated oven

↓

Cooling at 27°c

↓

Packaging

↓

Storage at 25°c and 38°c temperature


Dry Ingredients

↓


Creaming (Mixing of all ingredients)

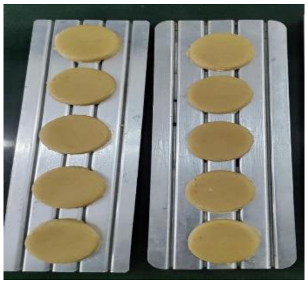
↓


Dough Making

↓

Sheeting the Dough

↓



Cutting the Cookies

↓

Baking the Cookies

↓

Stored in Aluminium Pouches

Stored in LDPE Pouches

3.3 Experimental Method

3.3.1 Free fatty acids content

The standardized procedure outlined by AOAC (2001) was used to determine the amount of free fatty acids in cookies. A flask containing 50 ml of benzene was filled with a 10-gram sample of ground stored cookies. For the purpose of extracting the free fatty acids, the mixture was left to stand for 30 minutes. The extracted solution was then transferred to a new flask, to which 5 ml of benzene, 10 ml of alcohol, and an indicator called phenolphthalein were added. A faint pink tint that lasted for 15 seconds occurred after the solution was titrated against 0.02 N KOH. Oleic acid served as a way to express the proportion of free fatty acids. The analysis was carried out three times.

$$\text{FFA}(\%) = \frac{282 \times 0.02\ KOH \times\ ml\ of\ alkali\ used \times 100}{(\%\ oleic\ acid\ )\ 1000 \times Wt.of\ sample\ taken} \times \text{dilution factor} \times 100$$

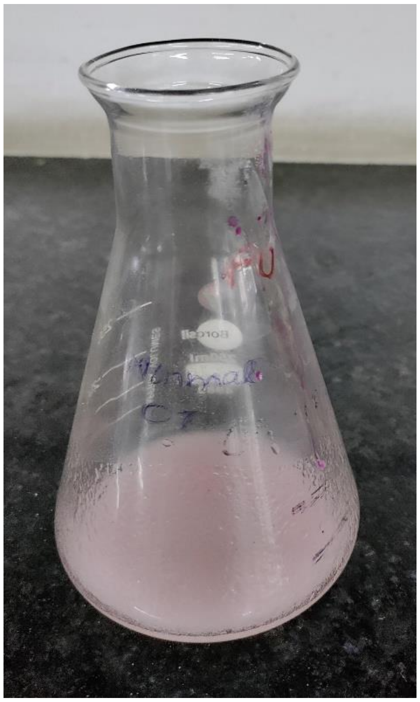


Figure 3.9: Endpoint observation

3.3.2 Peroxide value of cookies

Iodometric titration was used to calculate the peroxide value in accordance with the AOCS (2003) standard. A precisely weighed sample of 5 grams was added to a 250 ml iodine flask. A mechanical shaker was used to violently shake the liquid for an hour after adding 50 cc of chloroform. After that, a whatman no. 1 filter paper was used to filter the resultant combination. The filtrate was then transferred in 20 ml portions to additional iodine flasks. 30 ml of acetic acid and 1 ml of a saturated potassium iodide solution were put into this flask. After that, the flask was covered and kept in a dim environment for 30 minutes. The flask was taken out of the dark after this period of incubation, and 50 ml of distilled water was added. Utilizing starch (1%) as an indicator, the contents of the flask were titrated against a 0.02 N solution of sodium thiosulphate. The titration was continued until the solution's endpoint was colorless. It was noted how much sodium thiosulphate solution was used for the titration. The official AOCS approach from 2003 describes this procedure, which is widely acknowledged.

$$\text{PV (m eq .02/kg fat)}=\frac{Titre\ Value \times Normality\ of\ sodium\ thiosulphate \times 1000}{Wt\ of\ fat}$$

*Note: In case of samples, with less than 1g fat content, the PV values are expressed on sample Basis.

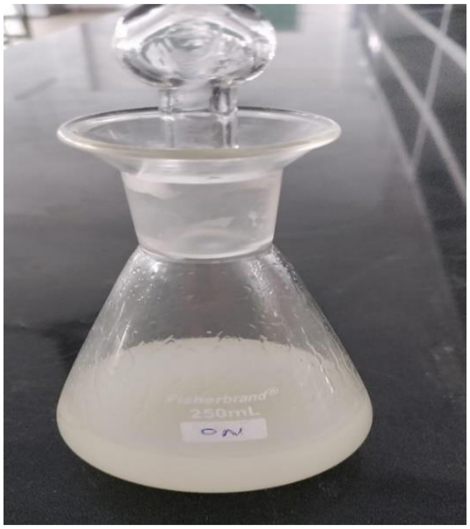


Figure 3.10: Endpoint observation

3.3.3 Proximate composition analysis

3.3.3.1 Moisture content of cookies

A sample's moisture content was calculated in accordance with the procedures provided in the AOAC (2012) standard. A Petri dish that had been previously dried and weighed was filled with approximately 5 grams of the sample. The sample was then dried until a consistent weight was obtained in a hot air oven set at 70 °C. The drying procedure was carried out three times, and the average result was noted. The AOAC (2012) method's formula was used to determine the percentage of moisture content.

$$\text{Moisture content (\%)} = ((W2-W3))/((W2-W1)) \times 100$$

W1 = weight of container with lid; W2 = Weight of container with lid and sample before Drying; and W3 = weight of container with lid and sample after drying

3.3.4 Texture Characteristics

3.3.4.1 Hardness

Using a TA.XT plus texture analyzer made by Stable Micro Systems in Godalming, England, the hardness of cookies' textures were examined. A 3-point bending rig (HDP/3PB) was used to gauge the hardness in accordance with the procedure described by Jambrec et al. in their 2013 study. Peak force, a measure of hardness, represented the highest resistance each cookie could muster against a rounded-edged blade—the point at which the sample began to crumble. On the other hand, the distance to peak force was used to define fracturability. According to [Levent and Bilgiçli in 2013], cookies with larger distance values were found to be more compressible and less prone to breaking

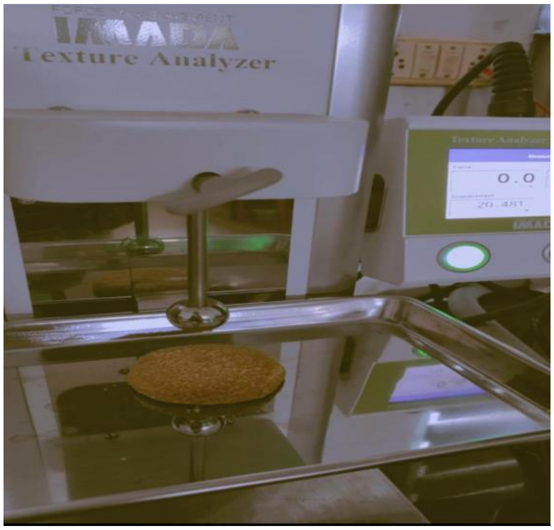


Figure 3.11

3.3.5 Sensory evaluation

The sensory qualities of the cookies were evaluated by a group of ten partially trained tasters using a 9-point Hedonic scale. The judges assigned ratings to a number of factors, including color and appearance, thickness, size, form, flavor, and general acceptance. The quality of the cookies was assessed using the average ratings given by the 10 reviewers (Avasthi, 2012).

3.3.6 Microbial examination of pea pod powder based cookies and controlled cookies

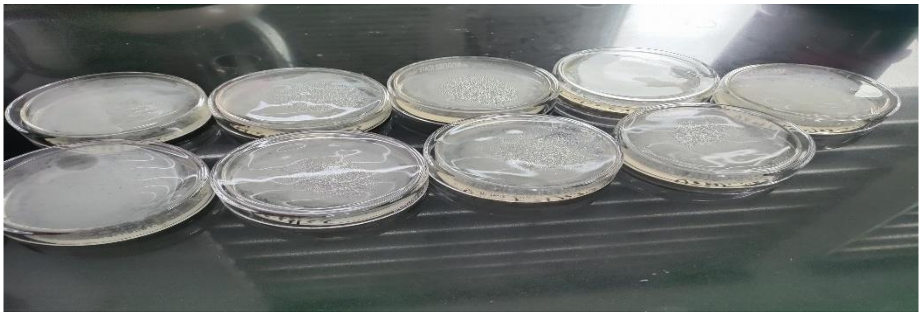
A critical component of evaluating the quality of food C products is microbial analysis. It works well as a protocol for quality analysis. To assess the microbiological quality, this study looked at the Total Plate Count (TPC), as well as the presence of yeast and mold. The enacted processes for the examination were as follows.

3.3.6.1 Total Plate Count (TPC)

Using Nutrient Agar, the researchers carried out a study to establish the total plate count (TPC) of cookies. Dilutions up to 10-4 were made, and a 0.1 ml sample was taken for isolation. The entire treatment was carried out in a laminar air flow environment under strict sterile guidelines. The results were then measured as colony-forming units per milliliter (CFU/ml) after the plates had been incubated at 37°C for 48 hours. At intervals of 1, 2, and 3 months, the TPC of the cookies was evaluated (Chandru et al., 2010).

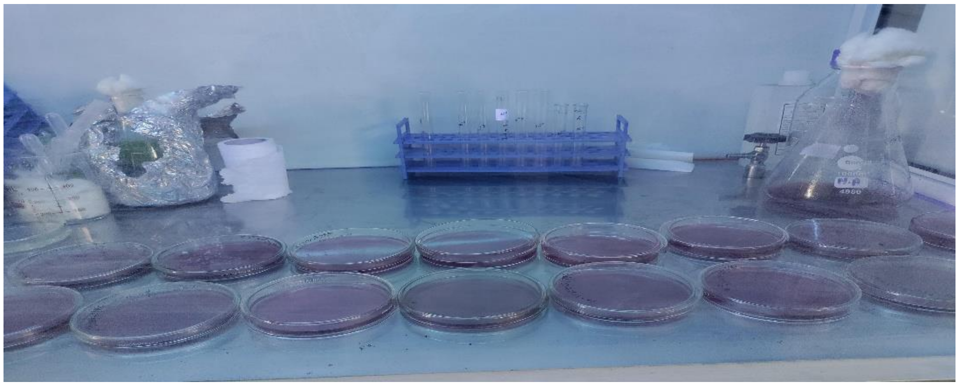
3.3.6.2 Yeast and Mould

Cookies were tested for yeast and mold using Potato Dextrose Agar (PDA) and the streak plate technique for isolation. Sterilizing the media and pouring it into plates was the first step. An aliquot of 0.1 ml of the material was utilized for streaking on the plates after it had been diluted up to a 10-4 concentration. For 48 hours, the plates were incubated at a temperature of 37°C to promote the growth of yeast and mold. Colony Forming Units (CFUs) per milliliter were counted and reported as the analysis's outcome following the incubation period.



3.3.6.3 Coliform count

A modified version of the method given in the compendium of methods for the microbiological examination of foods (AMPH, 1992) was used to assess the microbial safety, specifically the presence of coliforms, in the items. The original approach was modified to meet our unique needs and circumstances.



CHAPTER 4

RESULTS AND DISCUSSION

In this chapter we have discussed about the outcomes of the study carried out to determine the effect of different packaging parameters to retain the qualitative and quantitative properties of pea pod powder based fat sugar reduced cookies.

4.1 Storage stability studies of cookies

In the current study, controlled cookies made from wheat and barnyard millet flour were stored for 90 days at two different temperatures (25 and 38 degrees Celsius), using two different types of packaging (aluminum pouches and low density polyethylene), and under two different temperature conditions. The goal was to identify the best packing material and foresee the cookie's shelf life features. The cookies' moisture content, peroxide value, free fatty acid levels, hardness, microbiological analysis (total plate count, yeast and mold count, coliform count), and sensory ratings were all analyzed as response parameters in the study. Throughout the 90-day storage period, samples of cookies were examined every 15 days. The results are described below:

4.1.1 Effect of packaging material and storage on characteristics of cookies

4.1.1.1 Change in moisture content of cookies across storage

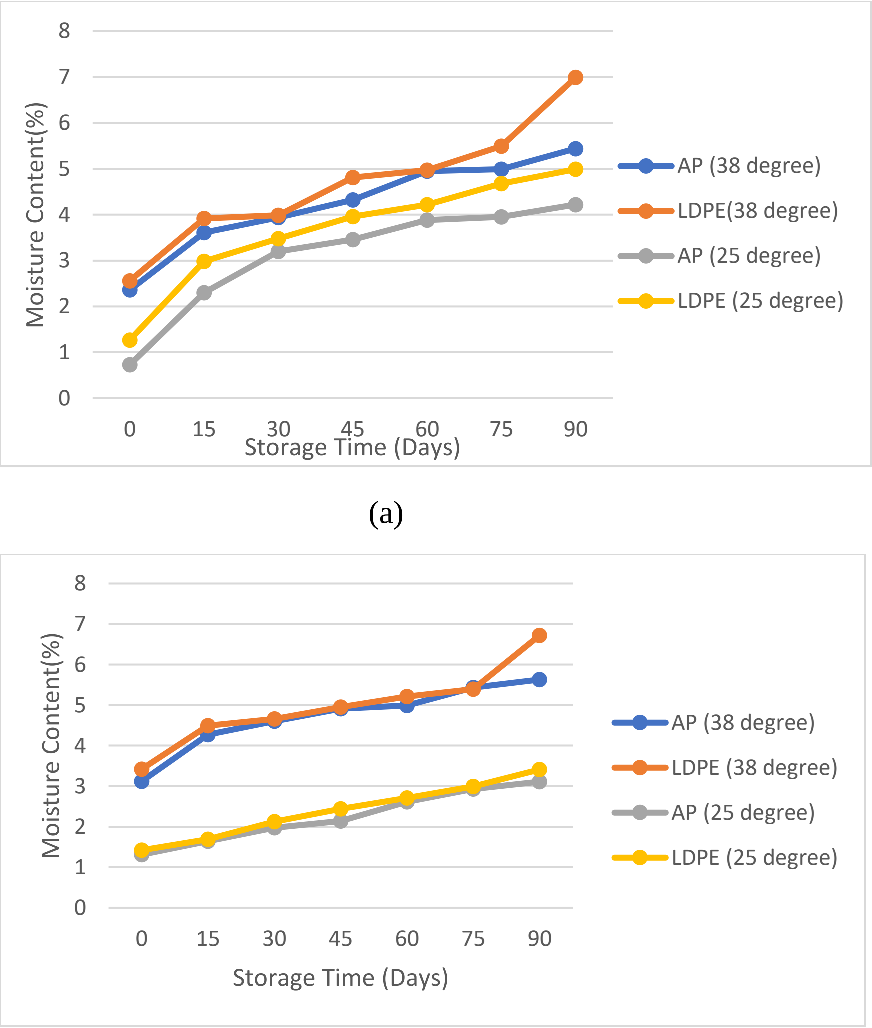
According to studies, a product's moisture content has a significant role in determining how stable it will be in storage. Throughout the storage period, the created cookies' moisture levels were checked on a regular basis. Table 4.1 shows the variation in moisture content between the controlled cookies and the cookies made with pea pod powder. Throughout storage, regardless of the packaging materials or storage temperature, the moisture level of both types of cookies rose. According to Krik and Sawyer (1991) and Rehman and Shah (1999), this shift could be ascribed to the flour's hygroscopic qualities, the packaging utilized, the interaction between storage and packing, and the storage temperature. At a storage temperature of 38°C, the controlled cookie's moisture content increased dramatically from 2.36 g/100g to 5.44 g/100g in aluminum pouches and from 2.56 g/100g to 6.99 g/100g in LDPE pouches. The moisture content of controlled cookies, however, increased from 0.73g/100g to 4.22g/100g in aluminum pouches and from 1.27g/100g to 4.99g/100g in LDPE when they were stored at 25°C. The moisture content of cookies made with pea pod powder increased from 3.12g/100g to 5.63g/100g in aluminum pouches and from 3.42g/100g to 6.72g/100g at 38°C in LDPE. And the pea pod powder-based cookies climbed from 1.31g/100g to 3.11g/100g when kept at 25°C in aluminum pouches, and for LDPE, they increased from 1.42g/100g to 3.41g/100g. It is clear from Table 4.1 that cookies stored in LDPE at a temperature of 38°C experienced the highest increase in moisture, whereas cookies stored in aluminum pouches at a temperature of 25°C experienced the lowest change. This difference in moisture increase could be attributed to variations in the water vapour transmission rates of the packaging materials used. However, according to IS7836 Indian Standards, the change in moisture content of the created cookies was within the standard allowed limit of 9% (Agraha-Murugkar and Jha, 2011).

Table 4.1 Effect of storage periods and packaging materials on moisture Content of Cookies

Cookies	Storage days	38°		25°	
		Aluminium pouches	LDPE	Aluminium pouches	LDPE
Controlled Cookies	0	2.36	2.56	0.73	1.27
	15	3.61	3.92	2.30	2.98
	30	3.94	3.99	3.2	3.48
	45	4.32	4.81	3.46	3.96
	60	4.95	4.97	3.88	4.22
	75	4.99	5.49	3.95	4.68
	90	5.44	6.99	4.22	4.99
Pea pod powder based cookies	0	3.12	3.42	1.31	1.42

	15	4.27	4.49	1.64	1.69
	30	4.60	4.66	1.97	2.12
	45	4.91	4.95	2.14	2.44
	60	4.99	5.21	2.61	2.71
	75	5.43	5.39	2.93	2.99
	90	5.63	6.72	3.11	3.41

Note-(LDPE)Low density polyethylene, (AP) Aluminum pouches.



(a)

(b)

Graph 4.1: Effect of storage periods and packaging materials on moisture Content (a) Control Cookies (b) Pea pod powder based cookies

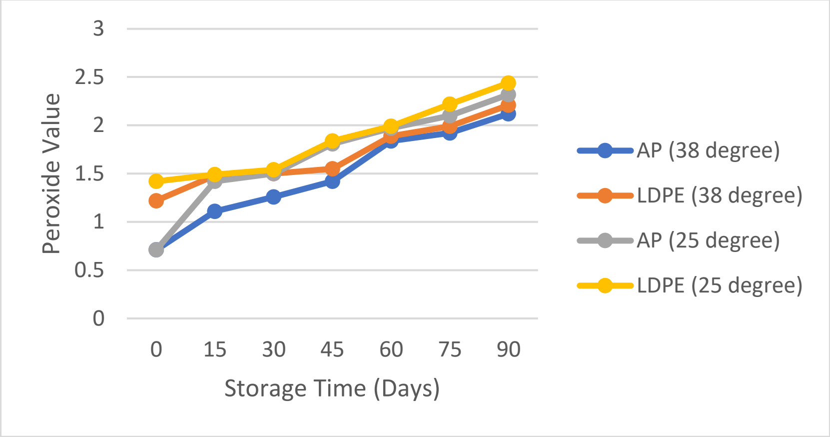
4.1.1.2 Change in peroxide value of cookies across storage

A key measure for assessing food product shelf life is its peroxide value. It is a quantitative measure of how rancid food products are. Table 4.2 shows the variation in peroxide value of produced cookies over a 90-day storage period stored in various packaging materials at various storage temperatures. Controlled cookies stored in aluminum pouches and LDPE stored at 38°C saw significant increases in the peroxide value from 0.71 mEq 02/kg fat to 2.12 and 1.22 mEq 02/kg fat to 2.21 mEq O2/kg fat, respectively, while cookies stored at 25°C saw significant increases from 0.71 mEq 02/kg fat to 2.32 and 1.42 mEq 02/kg fat to 2.44, respectively. According to Gahlawat and Sehgal (1994), the increase in peroxide values during storage is likely caused by peroxidation of the double bonds in unsaturated fatty acids, which break down to form secondary oxidation products that may suggest rancidity.Aluminum pouches at both temperatures showed the least change in peroxide value among the packaging materials. Controlled cookies and cookies made with pea pod powder showed similar trends of change in peroxide value throughout storage. Although the PV of the generated cookies dramatically rose, they still had much lower peroxide values than the permissible level (10 10mEqO2/kg fat) recommended by Aylward (1999). The impact of oil seed incorporation on the storage stability of produced instant cereal mix was examined by Vidhyasagar et al. in 1991. In contrast to what was seen in the current investigation, the study demonstrated a significantly higher peroxide generation. Similar results were found by Rao (2000) for modak (4.8mEqO2/kg fat) and Prakash et al. (1991) for khakra (3.7mEqO2/kg fat), which supported the findings of the current study. Similar results were found by Rao (2000) for modak (4.8mEqO2/kg fat) and Prakash et al. (1991) for khakra (3.7mEqO2/kg fat), which supported the findings of the current study. In their study, Lohia and Udipi (2015) also found that after 14 days of storage, the peroxide value increased from 5.12 mEq O2/kg fat to 9.94 mEq O2/kg fat, which is greater. The storage in polyethylene bags at ambient temperature may be the cause of this low shelf life.

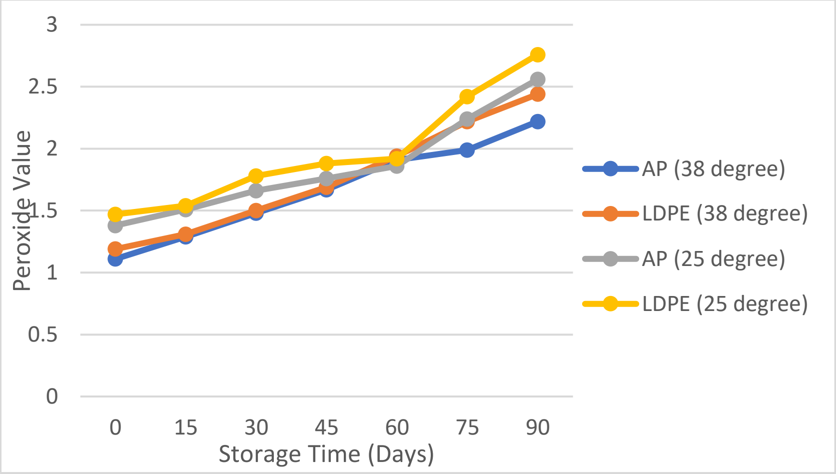
Table 4.2 Effect of storage periods and packaging materials on peroxide value of Cookies (meq. Peroxide/Kg)

Cookies	Storage days	38°c		25°c	
		Aluminium pouches	LDPE	Aluminium Pouches	LDPE
Controlled cookies	0	0.71	1.22	0.71	1.42
	15	1.11	1.48	1.42	1.49
	30	1.26	1.50	1.50	1.54
	45	1.42	1.55	1.81	1.84
	60	1.84	1.89	1.97	1.99
	75	1.92	1.99	2.10	2.22
	90	2.12	2.21	2.32	2.44
Pea pod powder based cookies	0	1.11	1.19	1.38	1.47
	15	1.29	1.31	1.51	1.54
	30	1.48	1.50	1.66	1.78
	45	1.67	1.69	1.76	1.88
	60	1.91	1.94	1.86	1.92
	75	1.99	2.22	2.24	2.42
	90	2.22	2.44	2.56	2.76

Note-(LDPE)Low density polyethylene, (AP) Aluminum pouches



(a)



(b)

Grapg 4.2: Effect of storage periods and packaging materials on Peroxide value (a) Control Cookies (b) Pea pod powder based cookies

4.1.1.3 Change in FFA content of cookies across storage

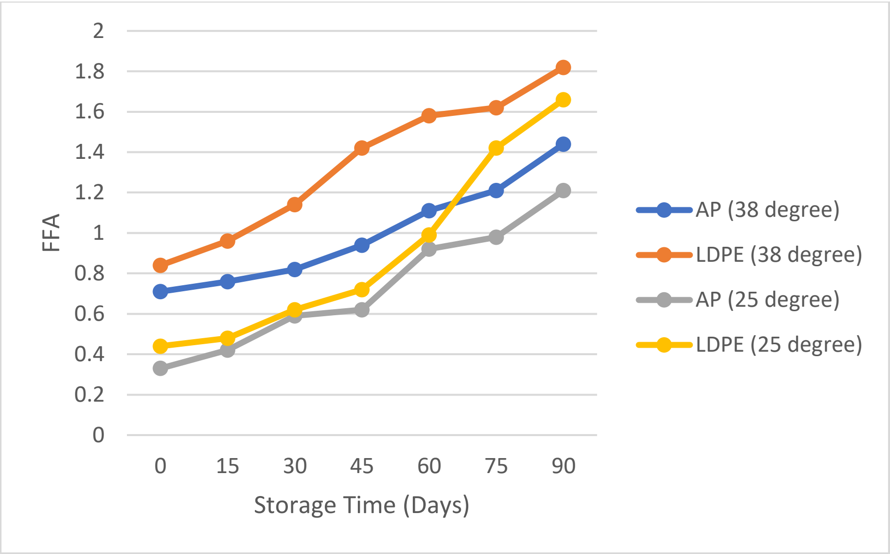
A product's lipid content may be a factor in the sensory quality of the product degrading over time. Triglycerides are hydrolyzed chemically or enzymatically to yield a combination of free fatty acids, glycerol molecules, diacyl glycerol molecules, and monoacyl glycerol molecules (Frankel, 2005). The rate at which this reaction takes place is significantly influenced by a number of variables, including the presence of oxygen, moisture, temperature, and the type of packaging employed (Manzocco and Lagazio, 2009; Speer and Kolling, 2006). Numerous volatile chemicals are created by the oxidation of FFA, which causes the loss of beneficial qualities including freshness (Frankel, 2005). Table 4.3 illustrates how packaging materials and storage temperature affect FFA content.After 90 days, the FFA content of controlled cookies stored in aluminum pouches increased from 0.71 to 1.44 Mg/100g at 38°C, which was lower than controlled cookies packed in LDPE (1.82 mg/100g), while controlled cookies stored at 25°C in aluminum pouches increased from 0.33 to 1.21 Mg/100g, which was also lower than controlled cookies packed in LDPE (1.66 mg/100g). However, storage at 25°C revealed a smaller range of FFA in both varieties of cookies kept in various types of packaging. Regarding the packing materials, cookies stored in LDPE containers had a higher FFA concentration than aluminum pouches, which may be due to the increase in moisture in the corresponding packaging materials. The variance in the materials used to create the formula mixes may be the cause of the discrepancy in the FFA content of different enteral formulae. According to Molteberg et al. (2014), the activities of lipases and lipolytic acyl-hydrolases may be responsible for the increase in total quantity of FFA during storage.

Table 4.3 Effect of storage periods and packaging materials on FFA content of Cookies (mg KOH/g)

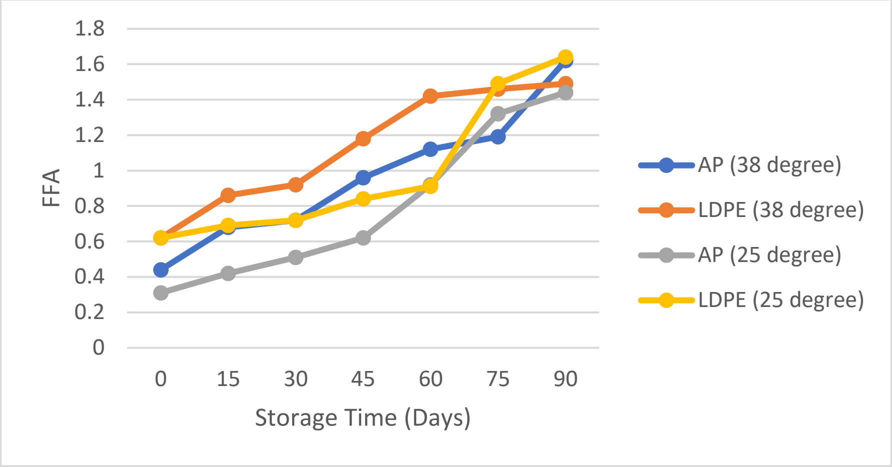
Cookies	Storage days	38°c		25°c	
		Aluminium pouches	LDPE	Aluminium Pouches	LDPE
Controlled cookies	0	0.71	0.84	0.33	0.44
	15	0.76	0.96	0.42	0.48
	30	0.82	1.14	0.59	0.62
	45	0.94	1.42	0.62	0.72
	60	1.11	1.58	0.92	0.99
	75	1.21	1.62	0.98	1.42

	90	1.44	1.82	1.21	1.66
Pea pod powder based cookies	0	0.44	0.62	0.31	0.62
	15	0.68	0.86	0.42	0.69
	30	0.72	0.92	0.51	0.72
	45	0.96	1.18	0.62	0.84
	60	1.12	1.42	0.92	0.91
	75	1.19	1.46	1.32	1.49
	90	1.62	1.49	1.44	1.64

Note-(LDPE)Low density polyethylene, (AP) Aluminum pouches



(a)



(b)

Graph 4.3: Effect of storage periods and packaging materials on FFA (a) Control Cookies (b) Pea pod powder based cookies

4.1.1.4 Change in Hardness of cookies across storage

The primary criterion for evaluating the general quality of finished goods like cookies is hardness. Tables 4.4 illustrate how the texture of controlled cookies and cookies made with pea pod powder varies during the course of storage at room temperature. The hardness of cookies packed in LDPE decreased as storage time increased, but the decline was greater in LDPE-packed cookies than in cookies packaged in aluminum pouches. The moisture content, water activity, and other physicochemical features of the product composition, i.e. interactions between constituents like fat, sugar, and flour, all have an impact on how hard dry or dehydrated meals are. Temperature had less of an impact on crispness than did water activity. Carter et al. (2015) found that cookie samples kept their crispness and texture at water activity values below essential water activity values. Product texture is significantly influenced by moisture redistribution and migration, physical changes to the key components of cookies, and interactions among them as they become increasingly diverse during storage. Depending on the type of product, there are different values for water activity and moisture content that correspond to customer rejection. According to Hough et al. (2001), biscuits lose their crispness during storage when their water activity values fall between 0.43 and 0.60. Charunuch et al. (2007) showed comparable outcomes for rice snacks that had been kept for four months.

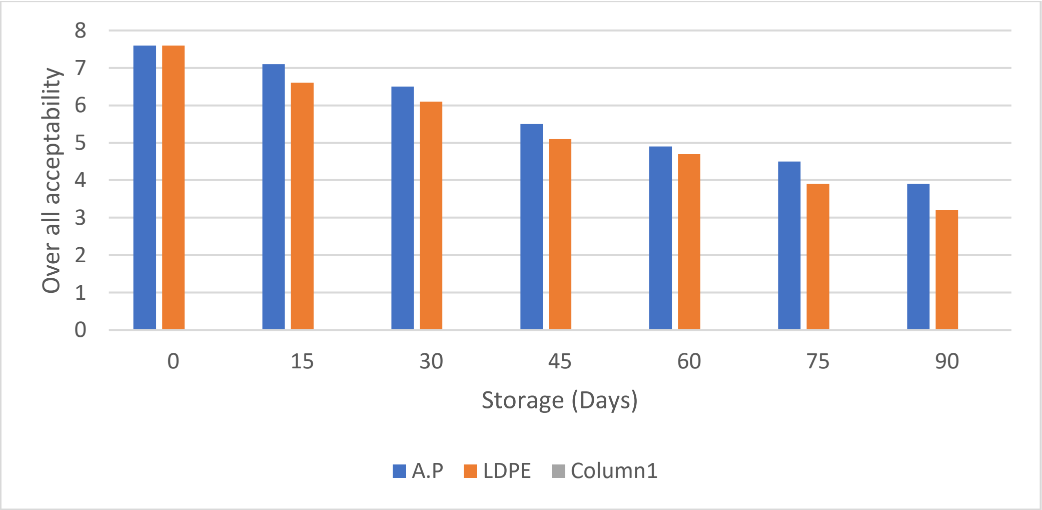
Table 4.4 Effect of storage periods and packaging materials on hardness of Cookies

Cookies	Storage days	38°c		Cookies	
		Aluminium Pouches	LDPE	Aluminium Pouches	LDPE
Controlled	0	99.3N,0.841nm	76.4N,0.22nm	49.8N,1.39nm	86.5N,1.223nm
	15	102.6N,2.665nm	55.2N,1.21nm	102.1N,2.707nm	75.3N,1.224nm
	30	103.0N,2.109nm	44.3N,1.11nm	109.9N,4.627nm	66.4N,4.626nm
	45	104.2N,2.208nm	42.3N,1.22nm	103.1N,2.108nm	55.1N,2.192nm
	60	105.1N,2.211nm	36.57N,1.33nm	105.2N,2.412nm	44.1N,2.412nm
	75	107.2N,2.312nm	34.24N,122nm	107.4N,2.413nm	34.1N,2.605nm
	90	108.4N,2.412nm	32.12N,1.44nm	109.5N,3.211nm	33.2N,2.312nm
Pea pod powder based cookies	0	45.9N,1.194nm	86.2N,0.11nm	38.5N,0.642nm	39.1N,1.129nm
	15	67.7N,1.477nm	74.3N,024nm	77.0N,1.116nm	39.2N,2.144nm
	30	87.6N,2.712nm	66.1N,1.22nm	103.4N,2.590nm	38.1N,3.144nm
	45	95.5N,2.812nm	54.2N,2.11nm	104.2N,2.209nm	37.4N,3.211nm
	60	100.1N,4.272nm	32.2N,2.12nm	105.4N,2.208nm	35.3N,4.272nm
	75	102.7N,2.552nm	31.1N,1.44nm	107.2N,2.512nm	34.4N,2.111nm
	90	104.2N,3.442nm	30.2N,1.62nm	109.8N,3.411nm	32.5N,3.224nm

Note-(LDPE)Low density polyethylene, (AP) Aluminum pouches

4.1.1.5 Overall acceptability (sensory characteristics)

For determining the product's acceptance and commercial relevance, sensory investigations are crucial. Packaging material had a noticeable impact on the general acceptance of both controlled cookies and cookies made with pea pod powder. Cookies made with pea pod powder and packaged in aluminum pouches had the highest average overall acceptability, while cookies packaged in LDPE had the lowest average overall acceptability. Up to 90 days after storage, there was no discernible change in the overall acceptability of the cookies, but after that point, a discernible decline in acceptability was discovered. The lowest average overall acceptability was found at 90 days of storage, which subsequently decreased to the lowest value. The best average overall acceptability was found up to 15 days of storage duration. The general acceptability of cookies packaged in LDPE decreased more noticeably than that of cookies packaged in metal pouches. The general shift in the sensory acceptability of the samples may have been brought about by changes in the items' color, flavor, and texture. According to Puyed et al.,2010). this may be because of increased moisture uptake, a high peroxide value (index of rancidity), and a subsequent loss of crispiness.



Graph 4.4: Effect of storage periods and packaging materials on overall acceptability of Cookies

4.1.1.6 Change in total plate count of cookies across storage

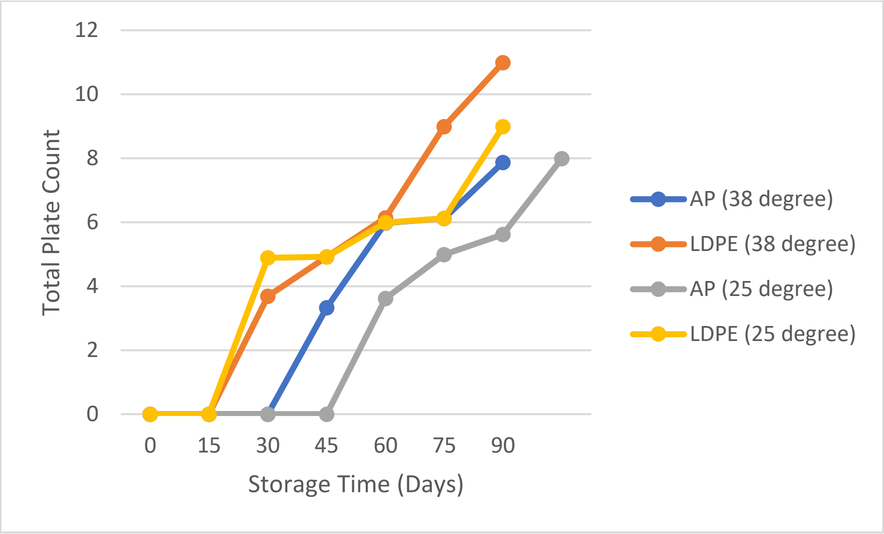
The most crucial factor determining a product's safety is its microbiological safety. Table 4.5 shows the microbiological quality of the developed cookies in terms of total plate count (TPC). It displays the outcomes of the microbiological examination of control and pea pod powder-based cookies stored at 38°C and 25°C, respectively, during storage. It is possible that there was a lag period for the bacteria to acclimatize to the new environment because there was no discernible variation in total plate counts across 0, 15, or 30 days at the two different temperatures. After 30 days of storage at two temperatures, the total plate counts in the cookies increased, whereas it took around 30 days at 38°C and 25°C. Considering that bacteria expanded more quickly at a 25°C storage temperature. Regardless of the materials utilized for packaging or the storage conditions, a considerable rise in TPC of the created cookies was observed. Cookies stored at 25°C showed a larger increase in TPC over the course of a 90-day storage period. Compared to aluminum pouches, cookies kept in LDPE containers exhibited a higher rise.

Table 4.5 Effect of storage periods and packaging materials on total count of Cookies

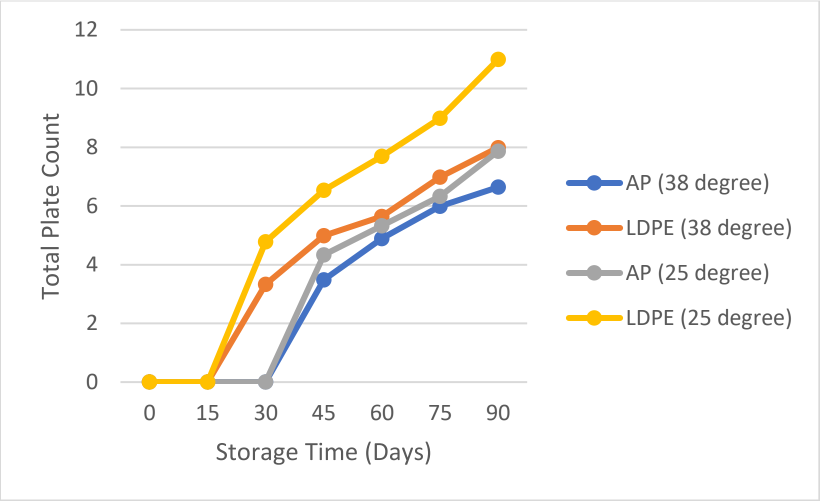
Cookies	Storage days	38°c		25°c	
		Aluminium pouches	LDPE	Aluminium Pouches	LDPE
Controlled cookies	0	ND	ND	ND	ND
	15	ND	ND	ND	ND
	30	ND	3.69	ND	4.89
	45	3.33	4.92	3.62	4.92
	60	5.98	6.14	4.99	5.99

	75	6.12	8.99	5.62	6.12
	90	7.87	10.99	7.99	8.99
Pea pod powder based cookies	0	ND	ND	ND	ND
	15	ND	ND	ND	ND
	30	ND	3.33	ND	4.78
	45	3.48	4.99	4.33	6.53
	60	4.89	5.64	5.33	7.69
	75	5.99	6.98	6.33	8.99
	90	6.64	7.99	7.87	10.99

Note-(LDPE)Low density polyethylene, (AP) Aluminum pouches,ND- Not detected



(a)



(b)

Graph 4.5: Effect of storage periods and packaging materials on Total plate count (a) Control Cookies (b) Pea pod powder-based cookies

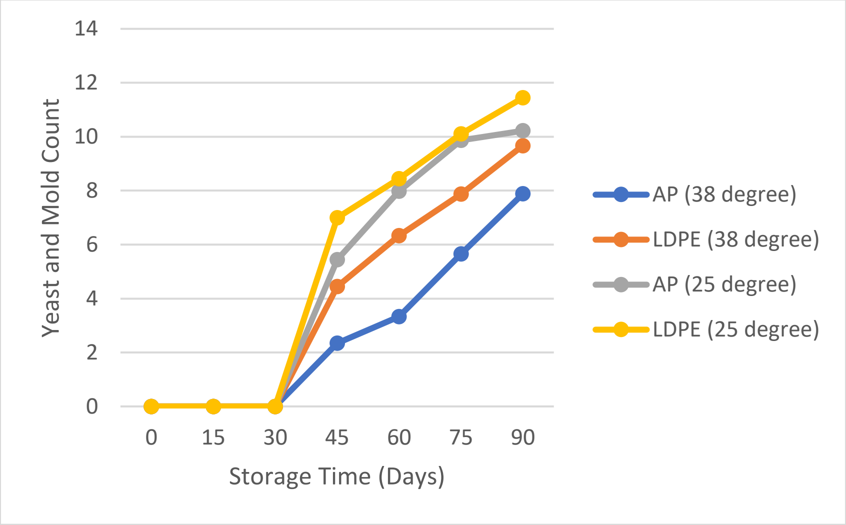
4.1.1.7 Change in yeast and mold count of cookies across storage

The findings of the microbiological study of control cookies and cookies made with pea pod powder after storage at two different temperatures, namely 38°C and 25°C, are shown in Table 4.6. It is possible that there was a lag period for the bacteria to adapt to the new environment because there was no discernible variation in the yeast and mold count during 0, 15, and 30 days at two different temperatures. After 30 days of storage at two temperatures, the amount of yeast and mold in the cookies increased, whereas at 38°C and 25°C, it took roughly 30 days. This is because microorganisms developed more quickly at a 25°C storage temperature. No matter what kind of container was used or what temperature the cookies were stored at, a noticeable increase in the yeast and mold count was seen. Cookies kept at 25°C exhibited a larger growth in yeast and mold count over the course of 90 days of storage. Compared to aluminum pouches, cookies kept in LDPE containers exhibited a higher rise.

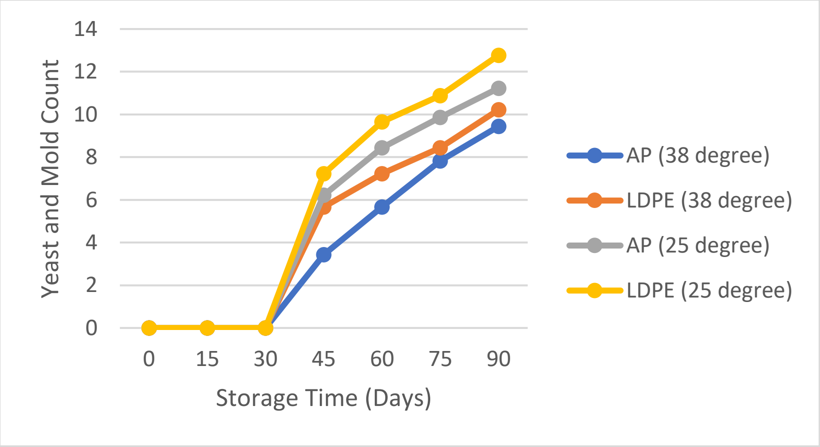
Table 4.6 Effect of storage periods and packaging materials on yeast and mold count of Cookies

Cookies	Storage days	38°c		25°c	
		Aluminium pouches	LDPE	Aluminium pouches	LDPE
Controlled cookies	0	ND	ND	ND	ND
	15	ND	ND	ND	ND
	30	ND	ND	ND	ND
	45	2.34	4.44	5.44	6.99
	60	3.33	6.33	7.98	8.44
	75	5.66	7.87	9.87	10.11
	90	7.88	9.67	10.22	11.44
Pea pod powder based cookies	0	ND	ND	ND	ND
	15	ND	ND	ND	ND
	30	ND	ND	ND	ND
	45	3.44	5.66	6.22	7.22
	60	5.66	7.22	8.44	9.66
	75	7.82	8.44	9.87	10.88
	90	9.44	10.22	11.23	12.77

Note-(LDPE)Low density polyethylene, (AP) Aluminum pouches, ND- Not detected



(a)



(b)

Graph 4.6: Effect of storage periods and packaging materials on Yeast and mold count (a) Control Cookies (b) Pea pod powder based cookies

4.1.1.8 Change in Coliform count of cookies across storage

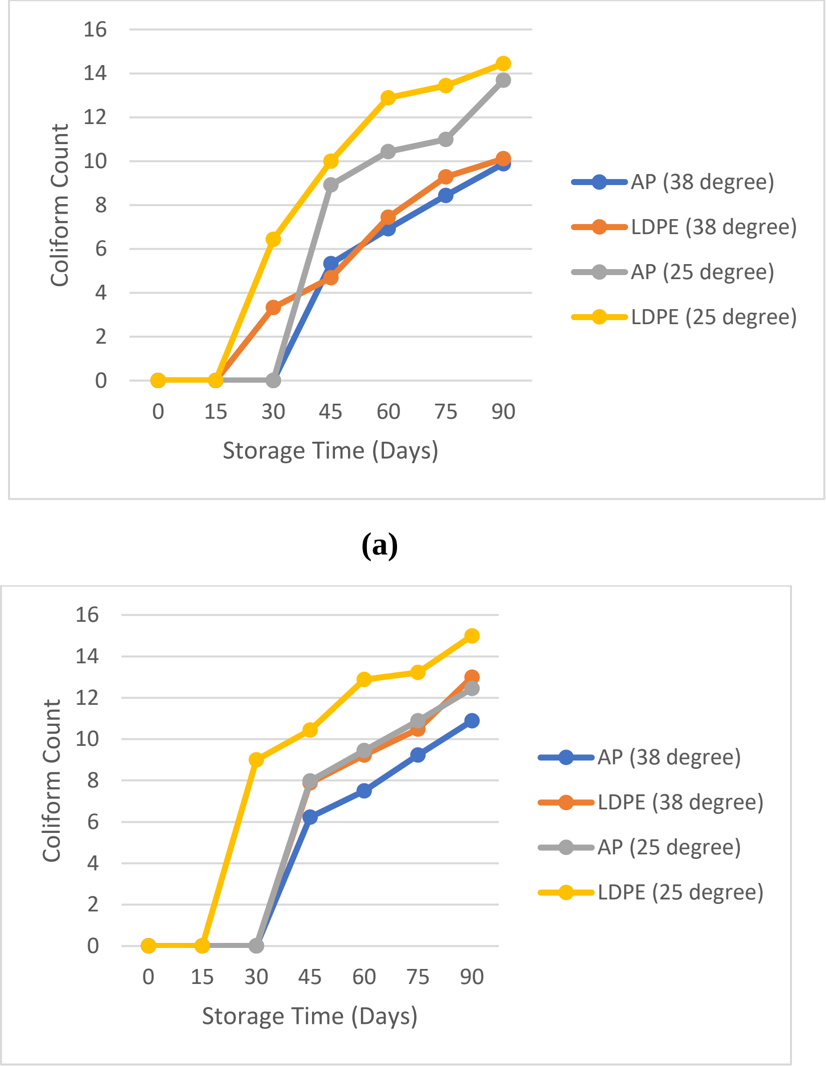
Table 4.7 displays the coliform count and microbiological quality of the generated biscuits. It displays the outcomes of the microbiological examination of control and pea pod powder-based cookies stored at 38°C and 25°C, respectively, during storage. During 0, 15, and 30 days at two different temperatures, there was no discernible difference in the number of coliforms, indicating that there was a lag period for the bacteria to adjust to the new environment. After 30 days of storage at two temperatures, the number of coliforms in the cookies grew, whereas it took around 30 days at 38°C and 25°C. Considering that bacteria expanded more quickly at a 25°C storage temperature. In spite of the materials utilized for packing and the storage temperatures, a considerable increase in coliform count was observed in the created cookies. The coliform count of the cookies kept at 25°C increased more rapidly over the course of storage for 90 days. Compared to aluminum pouches, cookies kept in LDPE containers exhibited a higher rise.

Table 4.7 Effect of storage periods and packaging materials on coliform count of Cookies

Cookies	Storage days	38°c		25°c	
		Aluminium pouches	LDPE	Aluminium pouches	LDPE
Controlled cookies	0	ND	ND	ND	ND
	15	ND	ND	ND	ND
	30	ND	3.33	ND	6.44
	45	5.33	4.68	8.92	9.99
	60	6.92	7.44	10.44	12.88
	75	8.44	9.28	10.99	13.44
	90	9.88	10.11	13.68	14.44
Pea pod powder based cookies	0	ND	ND	ND	ND
	15	ND	ND	ND	ND
	30	ND	ND	ND	8.99
	45	6.22	7.88	7.98	10.44
	60	7.49	9.22	9.44	12.88

	75	9.22	10.48	10.88	13.22
	90	10.88	12.99	12.44	14.99

Note-(LDPE)Low density polyethylene, (AP) Aluminum pouches, ND- Not detected



(a)

(b)

Graph 4.7: Effect of storage periods and packaging materials on Coliform count (a) Control Cookies (b) Pea pod powder based cookies

CHAPTER -5

Summary and Conclusion

This study showed that the storage and packaging of controlled cookies and cookies made with pea pod powder greatly influenced their quality features. Both packaging saw a rise in moisture content, free fatty acids, and peroxide value of the cookies they contained, but LDPE had a decrease in hardness and sensory ratings. At 38°C, the free fatty acid content of controlled cookies grew from 1.44 to 1.82, while at 25°C, it increased from 1.21 to 1.66. In contrast, the free fatty acid content of cookies made with pea pod powder reduced from 1.62 to 1.49. Even though the amount of free fatty acids increased during storage, the rise was within acceptable bounds. According to the moisture content values, the cookies, whether they were made using pea pod powder or a control method, were both stable and immune to microbial deterioration for up to 90 days. Cookies packaged in aluminum pouches had the highest average overall acceptability, whereas controlled cookies and cookies made with pea pod powder and packaged in LDPE had the lowest average overall acceptability.

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