EXTRACTION, CHARACTERIZATION AND APPLICATIONS OF PECTIN FROM ARTOCARPUS HETEROPHYLLUS (JACKFRUIT)

Dissertation submitted to

ST. TERESA'S COLLEGE (AUTONOMOUS), ERNAKULAM

Affiliated to

MAHATMA GANDHI UNIVERSITY

In partial fulfilment of requirement for the

AWARD OF THE DEGREE OF MASTER OF SCIENCE IN

HOME SCIENCE (BRANCH C)

FOOD SCIENCE AND NUTRITION

By

MALAVIKA MANOJ

Register No. AM23HFN008

DEPARTMENT OF HOMESCIENCE AND CENTRE FOR RESEARCH

APRIL 2025

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'Certified as bonafide research work'

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This is to certify that the thesis entitled ***"Extraction, Characterization and Applications of Pectin from Artocarpus heterophyllus (Jackfruit)"*** is an authentic record of the original research work carried out by **Ms. Malavika Manoj** with **Reg.No-AM23HFN008** under my Supervision and guidance during the academic year 2023-25.

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This is to certify that the dissertation entitled ***"Extraction, Characterization and Applications of Pectin from Artocarpus heterophyllus (Jackfruit)*** *is an authentic record of research work carried out by* ***Ms. Malavika Manoj*** *(Reg No:* ***AM23HFN008****), Department of Home Science, St. Teresa's College (Autonomous) Ernakulam, at the Doctor John's Biotech Center for Research and Development, Kottarakara under my supervision for the partial fulfilment of the degree of Master of Science in Food Science and Nutrition from Mahatma Gandhi University, Kottayam.*

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MALAVIKA MANOJ

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CHAPTER – 1

INTRODUCTION

Pectin is a heteropolysaccharide present in plant cell walls and used as gelling, thickening and stabilizing agent in food industry. Pectin structure mainly comprises of α- 1,4 glycosidic linkages or by alternating units of α-1,4 linked D-galacturonic acid and α-1,2 linked L-rhamnose residues. Traditionally, pectin which was commercially produced was extracted from the peels of citrus fruits and apple pomace etc. Currently there is growing interest in exploring the alternative sources of pectin which are sustainable and cost effective. Pectin is used for its potential health benefits in pharmaceuticals including its role as dietary fiber, lowering the cholesterol and its probiotic effects. Pectin has also applications in cosmetic industry, where it is used in the preparation of creams, lotions and other skincare formulations due to its stabilizing and emulsifying properties. Hence pectin is considered as a valuable compound for industrial application and research because of its versatility. There is increasing demand for natural hydrocolloids and thus exploring better alternative sources of pectin offers more scope in scientific fields like food industry, cosmetic industry and in pharmaceuticals.

Pectin extraction from jackfruit waste adds to economic development of farmers and also contributes to the environmental sustainability by developing value added products from jackfruit and thus reduces the agricultural waste.

Pectin extraction involves the process of isolating the polysaccharide from plant materials by various physical and chemical methods. The extraction process is very important in determining the yield, purity and quality of pectin and thereby directly determining the suitability of the extracted pectin in various food applications. The extraction process involves several steps, starting from the pre-treatment, extraction of the polysaccharide and the filtration and recovery. Each of these steps are carefully monitored and optimized to achieve better quantity and quality of pectin. The initial phase of pectin extraction is pre-treatment which involves the thorough cleaning of plant material to eliminate the impurities and then subjected to drying to reduce the moisture content. Drying can be achieved with the use of different methods such as sun drying, oven drying or freeze drying depending upon the material used for extraction.

The process of extraction involves the hydrolysis of plant material using the acidic solutions which helps to break open the complex cell wall structures and release pectin. Acidic conditions are found to be effective in disrupting the cellulose and hemicellulose which helps in the liberation of pectin from the plant matrix. Commonly used acids for pectin extraction from plant materials are citric acid, hydrochloric acid, sulfuric acid and nitric acid. The time taken for extraction depends upon the nature of raw material used and extraction method selected, which vary from as short as 30 minutes to several hours. After extraction the pectin is subjected to a process of purification/filtration and recovery. The extracted mixture is filtered to separate the alcohol soluble pectin from the alcohol insoluble residues by the use of alcohol. Ethanol and isopropanol are the two widely used alcohols for the purification of pectin. Alcohol helps in aggregation and separation of pectin from the solution by forming a gelatinous precipitate. This gelatinous precipitate is washed 2-3 times with alcohol to remove impurities and enhance the purity. Finally, the purified pectin is subjected to drying and is ground to fine powder. The powdered form of extracted pectin is then subjected to further characterization to assess the physicochemical properties.

Several factors such as pH of the extraction medium, temperature, time taken for extraction and solid-liquid ratio can influence the efficiency of pectin extraction. Generally acidic conditions are preferred as it enhances the solubilization of pectin, but excessive low pH levels may cause degradation of pectin structure, thereby compromising the quality. High temperature can increase the rate of extraction but this results in the thermal degradation if applied for long periods. Increase in extraction time helps to improve the pectin yield but this may also affect the pectin quality. Better extraction can also be achieved by high liquid volume, but this results in dilution of the extract. There are several research papers available regarding the extraction of pectin from other natural sources like peel of citrus fruits etc. Considering the case of jackfruit which is one of the largest fruits in the world and a major portion of the fruit is going as waste, utilization of these waste parts into valuable products like pectin offers more scope for the study. Based on the variety, stage of maturity, geographical location and extraction methods used, the composition of pectin from different jackfruits can be varied.

In food industry, jams are prepared by adding pectin which helps to facilitate gel formation and gives jams their characteristic semi-solid and spreadable consistency. Pectin is a gelling agent that interacts with acids and sugars to form a stable gel matrix. For trapping the fruit juice and pulp, ensuring a uniform texture and preventing separation of ingredients, the process of gelation is essential.

Pectin is commercially produced from citrus peels or apple pomace are subjected to a standardization process to achieve a consistent quality and performance in the production of jam.

Commercial pectin is available in different forms according to different sugar and acid requirements such as high methoxyl pectin and low methoxyl pectin.

Artocarpus heterophyllus also called as jackfruit, is a tropical fruit rich in nutrients and have a large biomass which is widely cultivated in south Asia. Kerala is considered as the hub of jackfruit in India, where the jackfruit production occurs abundantly. Jackfruit growing in Kerala are of different varieties including *varikka, koozha, aanjili, kadachakka, aathachakka* etc which is grown on the tree called *plavu.*

The Kerala varieties of jackfruit selected for the study are varikka and koozha. Varikka has firm flesh and koozha have soft flesh. These are widely grown in Kerala, for making different food products like jackfruit chips, flour, nectar, wine, roasted seeds, halwa etc. Even though jackfruit is one of the largest fruits in the world, it is of low cost, locally available, nutrient rich and easily grown in Kerala. The parts like flesh and seeds are regarded as edible portions and a majority of parts like rind, tandems and core are discarded as waste. These byproducts contain valuable substances like pectin, which makes jackfruit a good alternative source for extraction. The non-edible parts of jackfruit serve as a low-cost raw material for the extraction of pectin, thereby reducing the production cost and product cost which helps in better marketing of jackfruit pectin as an alternative for commercial pectin.

Studies have shown that the physicochemical properties such as high methoxyl content and degree of esterification, which are essential parameters for the application of pectin as gelling agent was found to be favorable in pectin extracted from jackfruit. This study focuses on the extraction, characterization and application of pectin from Artocarpus heterophyllus (jackfruit) two varieties- varikka and koozha. The physicochemical characterizations including the equivalent weight, methoxyl content, anhydrouronic acid content and degree of esterification are analyzed to determine the suitability of the extracted pectin for its industrial applications. The extracted jackfruit pectin was also added in the preparation of jam to assess the organoleptic qualities.

Relevance of the study

Food industries which process jackfruit generates biowaste such as rind, tandem and core. Due to limited research these parts of jackfruit remain underutilized and hence development of value-added products such as pectin from the underutilized parts of jackfruit adds more scope to this study. Pectin extraction from jackfruit waste contributes to the economic development of farmers and also adds to the environmental sustainability by developing value added products from jackfruit and thus reduces the agricultural waste. The extracted pectin from jackfruit can be standardized and developed into an industrial product enhancing its application in food industry for the production of various food products like jams and jellies. For analyzing the suitability of pectin in various industrial applications, evaluating the chemical composition of pectin from different varieties of jackfruit can provide more insights.

The main objectives of the study are;

- Extraction of pectin from Artocarpus heterophyllus (jackfruit) of two varieties – varikka and koozha
- Comparison of the extracted pectin from different varieties and parts of jackfruit – varikka and koozha
- Physicochemical characterization of the extracted jackfruit pectin for analyzing the suitability of the pectin in its industrial application
- Application of the extracted jackfruit pectin in preparation of jam and analyzing the organoleptic qualities.

CHAPTER – 2

REVIEW OF LITERATURE

The review of literature of the study entitled **"Extraction, Characterization and Applications of pectin from Artocarpus heterophyllus (Jackfruit)"** were discussed under the following headings;

2.1 Pectin- Structure and Properties

2.2 Extraction of pectin by various techniques and from different food items

2.3 characteristics and importance of pectin in industries

2.4 Artocarpus heterophyllus (Jackfruit) and its varieties

2.5 Addition of pectin in value added food products

2.1 Pectin- Structure and Properties

Pectin is a complex heteropolysaccharide found in cell walls of plants, which acts as cellular binder in fruits and vegetables (Constenla *et al*., 2003). The structure of pectin chains is made up of units of D-galacturonic acids joined together by α- 1,4 glycosidic linkages or by alternating units of α-1,4 linked D-galacturonic acid and α-1,2 linked L-rhamnose residues. The types of monosaccharides present in pectin are galacturonic acids, rhamnose, galactose and arabinose. Among these the galacturonic acids are the most dominating. The structure and characterization of pectin from same plant but different parts have significant differences in its monosaccharide composition, equivalent weight, degree of esterification etc. In homogalacturonan, galacturonic acid units can be partially methyl-esterified at the C6 position, while rhamnose units in rhamnogalacturonan-I may have side chains of neutral monosaccharides. These differences in composition and molecular structure enhance their physicochemical properties and broaden their technological uses. In the food industry, hence they serve as thickeners, emulsifiers, and stabilizers. (Melo *et al*., 2017).

In 1790 the molecule of pectin was first identified in apple pomace (Vauquelin, 1790). According to (Ciriminna *et al*., 2015) the majority production of pectin was from citrus peel (85.5%), apple pomace (14%) and sugar beet (0.5%). Pectin is soluble in water and insoluble in alcohol, hence an extraction process of the component from fruits can be made possible using alcohol extraction.

They exhibit property of gelling with water. Approximately 30% of pectin is present in the white portion of rind in lemons and oranges. It has an essential part to play in the initial growth of fruits. The degree of esterification is considered as the basis for the classification of pectin. The character, function, structure and properties of pectin from different fruits are different. Pectin has an average molecular weight of about 30-100 kDa. (Rowe *et al*., 2009).

Concerning the structure of polysaccharides (Diener *et al*., 2019) introduced a nomenclature recently where the primary structure of pectin is covalently sequenced and linked monomers of galacturonic acid and rhamnose which has repeating pattern. Pectin chains expose their functional groups and become hydrated in aqueous dispersion. Conformation of pectin chains in solutions is determined by properties like pH, temperature, ionic strength, presence of divalent ions and other properties like side chain structure, backbone structure and variations in linear density. The regular, intermolecular and spatial arrangement of the primary sequence into a helix defines the secondary structure of pectin. The repeating torsion angles of the α- 1,4 and α- 1,2 glycosidic bonds between monomers results in the formation of helical structure of pectin. Three dimensional structures like gels, complexes and viscous solutions are results of formation of higher-level structures due to ionic, hydrophilic or hydrophobic interchain interactions. Recently the exclusive reason for the morphogenesis of plant epidermis has found to be the conformation of pectin (Haas *et al*., 2020).

According to (Zdunek *et al*., 2021) adequate and convenient too for the evaluation of pectin conformation is atomic force microscopy, since it allows pectin molecule to relatively stretch straight forward in order to determine the stiffness and flexibility which is directly proportional to the measure of force extension curve.

Pectin shows difference in its molecular structure and is prone to physical, chemical and enzymatic changes (Freitas *et al*., 2021). Pectin structure contains different functional groups which can stimulate different functionalities and some modifications can make this polysaccharide for its novel applications due to modifications in physical-chemical properties like degree of esterification, molecular weight and formal charge (Freitas *et al*., 2020).

Enzymes which breakdown the pectin are mainly classified into three groups; the proto-pectinase which breakdown the insoluble protopectin into soluble polymerized pectin, the depolymerizing enzymes which breaks the α1→4 glycosidic linkages of pectin and finally the esterase which are involved in the de-esterification and de-acetylation of pectin. Complex heteropolysaccharides of molecular weight ranging between 25 and 360 kDa are observed in majority of the fruits and tissues of vegetables (Jayani *et al*., 2005).

According to FAO (food and agricultural organization) the pectin structure should contain ≥ 65% of galacturonic acid (Gal A) (Ngouemazong *et al*., 2015). The gelling property of pectin which is used in the food industries is directly depends on the degree of esterification of pectin. Higher degree of esterification exhibiting pectin gel around pH of 3.0 in the presence of sugar and lower degree of esterification exhibiting pectin gel in the presence of calcium ions under a wide range of pH with or without the presence of sugar (Sista Kameshwar *et al*., 2018).

It was estimated that at least 67 transferases including glycosyl-, methyl- and acetyl transferases was required for the biosynthesis of pectin. New developments identifies that these polysaccharides have role in both primary and secondary cell walls in understanding the pectin structure, function and biosynthesis. Diverse plant agronomical properties including plant biomass characteristics which is important for biofuel production was found to be affected by manipulation in synthesis of pectin. (Mohen, 2008). A decrease in the amount of covalently bound pectin, those supposedly located in the primary cell wall, which is mainly extracted with sodium carbonate is paralleled with an increase in the levels of soluble pectin. (Pose *et al*., 2011). To characterize the effect of pectate lyase and poly-galacturonase genes silencing of pectin nanostructure, atomic force microscopy (AFM) was used recently. Down regulation of pectate lyase and poly-galacturonase genes increased the length of pectin chains and their structural complexity. An increase in fruit firmness has been correlated with the magnitude of these changes (Pose *et al*., 2015).

According to (C Paniagua *et al*., 2017) branching of bound pectin decreases in ripen strawberry fruits. Strawberry fruit softening is contributed by the loss of nano structural pectin complexity. Pectin lyase, is an enzyme acting upon pectic substances which occurs as natural polysaccharides in primary cell walls and middle lamella of higher plants. The enzyme degrades pectin polymer directly by beta elimination mechanism. pectin lyase has potential applications in textile industry in retting and degumming of hemp, jute and flax (Yadav *et al*., 2009).

2.2 Extraction of pectin by various techniques and from different food items

Pectin have been extracted from various plant sources using a variety of techniques. To increase the extraction yield and enhance the quality of the final product, it is important to use an appropriate technique for pectin extraction. Conventional methods of extraction, like using mineral acids, and novel methods, like ultrasonic, microwave, and enzyme-assisted extraction with distinct mechanisms, are among the extraction techniques (Maric *et al*., 2018).

In majority of food processing industries, the fraction of discarded materials is very high (Laufenberg *et al*.,2003; Parfit *et al*.,2010) depending upon the method of harvest and location. Approximately 45% of total industrial organic pollution is generated by the food processing industry (Akerberg and Zacchi, 2000).

The results from the study of (Panchami et al., 2017) shows that there was an appreciable level of components such as reducing sugars, total sugars, nitrogen, phosphorous, potassium and organic carbon present in mango peel, citrus peel, banana peel and apple peel. About 24.5% of pectin can be extracted from citrus peels, 10.8% from apple pomace, 7.5% from mango peel and 2.5% from banana peel. From the 25.5%, about 24.5% of pectin can be extracted from citrus peel. 23.21% of galacturonic acid content was found in pectin extracted from passion fruit (de Moura *et al*., 2017).

The best possible source for the production of pectinase enzyme based on high pectin content among the fruit wastes tested was the citrus peels. Hydrochloric acid was used most commonly among the wide range of reagents used for the extraction of pectin as it is cost effective than other organic acids (Sudhakar and Maini, 1995).

The fruit wastes of orange, mango, apple and banana were used for pectin extraction. 300ml of 0.1 N HCl was added to 50g of ground fruit waste. This was subjected to boiling for 30 min, and then filtered under suction. Boiling water was added to the residue and filtrate was collected with 0.05 N HCl and 0.3 N HCl. The process was repeated and filtrates were collected. The pooled filtrate was made up to the volume of 500ml and 200ml from this was transferred into a beaker with 250ml of water. By adding 0.1 N NaOH using phenolphthalein indicator causes the neutralization of the acids presents in the filtrate and this was allowed to stand for 24 hours. Then 50ml of 1 N acetic acid was added and after 5 minutes 25ml of 1 N calcium chloride was also added. This was stirred and allowed to stand for an hour. For 2 minutes the contents were boiled and filtered through Whatman No.1 filter paper.

With the boiling water the final precipitate was washed until the filtrate was chloride free (Panchami *et al*.,2017).

The filter paper with calcium pectate was transferred to a pre-weighed dish and this was dried at 100◦C for overnight and cooled, then determined the dry weight for estimating the total pectin content (Manickam and Sadasivam,1996).

Citrus by-products are largely used to produce value added substances like pectin (Mamma *et al*., 2014). According to (Freitas *et al*., 2020) the peels of passion fruit, which is an industrial byproduct is rich in pectin. Passion fruit peels are generated in large quantities during fruit processing for the production of passion fruit juice and pulp. Several methods such as conventional extraction, enzyme-assisted extraction, extraction with subcritical fluids, UAE, MAE, UAME, S-MAE, HHP, DESs and NADESs were used for the extraction of pectin. The characteristics of these methods explains the effect of variables with emphasis on extraction of pectin from passion fruit peel (Liew *et al*., 2014).

From the extraction and characterization of pectin from citric waste by (Gama *et al*., 2015) showed that about 8% of pectin and 80% of carbohydrate which is high level was found in the residues of lime orange. With 8% of esterification, a higher yield of pectin of 78% was noticed at high heating time of 90 minutes, 6% of acid concentration and a temperature of 90◦C.

Pectin extraction from common fig skin was done (Gharibzahedi *et al*., 2019). Different extraction methods like hot water (HWE), microwave-assisted (MAE), ultrasound-assisted (UAE) and ultrasound-microwave assisted techniques were used to extract pectin from Ficus carica (common fig) skin. The results from the study shows that the yield of pectin after extraction were UMAE (11.7%) which showed the highest yield than MAE (9.26%), UAE (8.74%) and HWE (6.05%). A non-crystalline nature for pectin extracted by UMAE was showed on X-ray diffraction (XRD) analysis. By the use of HPLC photodiode array electrodes and FTIR spectroscopy revealed that the novel and conventional technologies for extraction do not change the monosaccharide composition and chemical structure of pectin. Pectin was extracted from peels of sweet lemon by microwave-assisted methods (Rahmani *et al*., 2020).

Citric acid is widely used as chelating agent extraction of pectin from mango peels by UAE method. A number of tropical and subtropical fruits by products have been found to represent potential sources of pectin. A study on extraction, characterization and application of pectin from tropical and subtropical fruits revealed that the extraction of pectin by conventional methods requires a lot of energy and mineral acids (Wang *et al*., 2016).

Studies have been undertaken in line with the sustainable development goals to assess the efficiency of non-conventional green extraction methods such as ultrasound-assisted, microwave-assisted and enzyme-assisted extraction to determine the yield and characteristics of pectin extracted from subtropical and tropical fruits (Picot-Allain *et al*., 2022).

Comparison between different techniques used for water-based extraction of pectin from orange peels which are rich in phenolic components (Rafiq *et al*., 2018) was done (Yeoh *et al*., 2008). For this study conventional and microwave assisted methods have been used for the extraction with different solvent pH, different types of solvent systems and different extraction periods. The highest total amount of pectin yield was from microwave extraction which was found to be 5.27% for 15 minutes extraction on a dry basis. Greater amount of material per unit time (%/min) was obtained after 5 minutes and this was the same amount as that extracted with 3 hours of Soxhlet extraction. From further investigations of microwave treatment at pH of 1.5, 2.0, 5.5 and 10.0 for 15 minutes revealed that the greater amount of pectin was extracted at strong acidic condition of pH 1.5. by studying the extraction period of 15 minutes and pH of 1.5 with solvent systems containing ethanol and EDTA (ethylenediamine tetra acetic acid) showed that it gives approximately double the amount of pectin extracted using distilled water.

A study was done on the analysis of pectin extracted from grape fruit peel (Mohamed, 2016), and grape pomace by the utilization of microwave technique and its influence on the pectin yield, molecular weight, galacturonic acid content and degree of esterification of pectin were analyzed by (Spinei *et al*., 2022). From this study, microwave power of 560 W and pH of 1.8 for 120 seconds were the optimal conditions for the extraction process. Pectin was extracted from the samples by MAE in optimal conditions and were analyzed by comparing with citrus and apple pectin based on FT-IR analysis. From the analysis, presence of different functional groups which attributed to the finger print region of pectin was established. The rheological behavior of pectin solutions showed good viscoelasticity. Thus, grape pomace was assumed to be a valuable source of pectin and from the results obtained by (Spinei *et al*., 2022) the extraction can be done by simple and quick techniques.

While analogous quality to conventional sources of pectin was maintained, Pectin was extracted from cocoa husk using solvents ammonium oxalate and acetic acid (Nazaruddin Ramli *et al*., 2011) with time of 60-180 min and pH ranging from 1.6-4.6.

Ultrasound-assisted extraction method is considered as a green and economically viable alternative to the conventional techniques used for food and natural products (Chemat *et al*., 2017). Aqueous pectin was extracted at a temperature of 60-90◦C, time of 60-180 min with liquid/solid ratio of 20-40 v/m from citrus medica peel (Pasandide *et al*., 2017).

On the laboratory analysis on extraction of pectin revealed that a higher yield of 17.92% was achieved with the sonication process of 25 minutes (Bagherian *et al*., 2011). Pectin extracted by ultrasound method had a Gal A content greater than 65% and DE greater than 50% (de Oliveira et al., 2016). By UAE method pectin was extracted from pomegranate peels (Moorthy *et al.*, 2015), sour orange peels (Hosseini, S.S. *et al*., 2019) and from tomato waste (Grassino *et al*., 2016). Pectin extracted from passion fruit peel under optimal conditions had a yield of 14.8g/100g of dried peel (Kulkarni *et al*., 2010).

2.3 Characteristics and importance of pectin in industries

Pectin has intrinsic biological activity and the structure of pectin differs according to the source and methods used for its extraction. It is regarded as a green option for the valorization of industrial residues of agriculture by providing high valuable products commercially (Freitas *et al*., 2021).

Pectin undergoes physical, chemical and enzymatic changes in which the functional groups present in its structure allows certain modifications in pectin thereby increasing its applications in food industry, agricultural industry, pharmaceutical industry and in biomedicines. Currently, pectin is used as an edible coating to protect foods, to produce nano particles, antimicrobial bio-based films. Healing agents and also in treatment of cancer. It is regarded as safe, easily available and non-toxic product which requires low production cost which makes more applications of the product in different sectors. Pectin has the ability to chelate metal ions because of its antioxidant capacity (Koubala *et al*., 2014).

The use of synthetic additives can be reduced by the addition of pectin to food emulsions as antioxidant thus, favoring numerous functionalities and can achieve clean label products. Pectin has the ability to form gel in the presence of calcium ions or in a solute with low pH which increases its importance in food sector. Ionic linkage and calcium bridges between the two carboxylic groups belonging to two different chains which is in close contact with each other results in gelation in low methoxyl pectin. A combination of hydrogen bonds and hydrophobic interactions between molecules results in gelation of high methoxyl pectin (Celus *et al*., 2018).

In food industry pectin is used in the production of jellies, jams, frozen desserts/foods and low-calorie foods as fat or sugar replacer. It is used to reduce blood cholesterol levels and gastrointestinal disorders in health sector. It also has applications in the production of edible films, foams, plasticizers and paper substitutes (Thakur *et al*., 1997).

Pectin and pectin derived oligosaccharides are significantly found to be used as an important ingredient in functional foods. The new applications of pectin as edible coating material in food stuffs due to its specific property to act like natural barrier for the exchange of gases, lipids, moisture and volatiles between the environment and the food thereby preventing microbial contamination (Vanitha *et al.,* 2019).

Pectin has a better solubility in pure water. When the dry powder of pectin is added to water it has a tendency to hydrate immediately and results in the formation of clumps. The properties such as viscosity, solubility and gelation of pectin are generally related. Factors which increase the strength of gel formation will increase the viscosity and decreases the solubility. Pectin is decomposed fast by de-esterification and by de-polymerization when dissolved (Sriamornsak, 2003). The decomposition rate depends upon the factors like temperature, pH, and water activity. The maximum stability is observed at a pH of 4. Certain protective effect is seen in pectin solution with sugar, while elevated temperature can make the degradation faster. Low pH and increase in temperature cause degradation by the hydrolysis of glycosidic linkages. De-esterification is also caused at low pH. Rapid de-esterification and degradation of pectin happens even at room temperature when the pH is alkaline. If stored under humid or warm conditions, powdered HM-pectin slowly lose their ability to form gels, where more stability is shown by LM-pectin (Rolin, 1993).

Ability to form gel with the presence of sugars and acids are one of the important characteristics off pectin HM-pectin, where LM-pectin forms gel with the presence of calcium ions. The hydrophobic interactions and hydrogen bonding are the two significant forces that causes the aggregation of pectin molecules (Oakenfull, 1991).

Sugar induces a competition for available water thereby reduces the hydration of pectin. The rate of gel formation is affected by one of the important physico-chemical characteristics of pectin that is degree of esterification. When the DE is above 72% which is high, causes rapid setting. The gelation mechanism in LM-pectin is well known as 'egg-box' mode (Grant *et al*., 1973).

Junction zones are created by the ordered and side chain associations of galacturonans, where the Gal A monomer's specific sequence in adjacent chains was linked intermolecularly by ionic and electrostatic bonds of carboxyl groups. Pectin has several uses in pharmaceutical industry, where these polysaccharides are used as bioactive agents for medical applications (Pawar *et al*., 2008). It was found to lower the cholesterol levels in blood in wide variety of subjects under experimental conditions (Sriamornsak, 2001). To have significant effect in cholesterol reduction at least 6g/day of pectin has to be consumed and less than 6g is not effective (Ginter *et al*., 1979). About 13% of reduction in serum cholesterol was reported by (Mietinnen, *et al*., 1977) with pectin consumption for 2 weeks treatment. The substance can act against poisoning with toxic cations as pectin is a natural prophylactic substance.

For removing lead and mercury from gastrointestinal tract and respiratory organs, pectin was found to be effective (Kohn, 1982) and these helps in drug delivery (Dos Santos *et al*., 2015). The intravenous injections of pectin reduce the coagulation time of blood drawn, thus helpful in controlling local bleeding and hemorrhage (Joseph, 1956). In infants and children, combinations of pectin with other colloids have been used to treat diarrheal diseases. Pectin may have a little antimicrobial action against E-coli under certain in-vitro conditions (Thakur *et al*., 1997).

Pectin was used as a carrier material in pharmaceuticals especially in colon-specific drug delivery systems to treat disease conditions like ulcerative colitis, crohn's disease and colon cancer. Pectin based delivery systems, especially the gel coating and ionotropic gelation have been manufactured with different techniques. These techniques and a safe toxicity profile of pectin made it an exciting and promising component for pharmaceutical industry for present and future applications. The pectin is used as a biopolymer in food and pharmaceutical industry because of its important characteristics like gel-formation, solubility etc. (Sriamornsak, 2001).

2.4 Artocarpus heterophyllus (Jackfruit) and its varieties

Artocarpus heterophyllus is a large widely grown tree belonging to the family Moraceae, which is commonly called as jackfruit in English (Prakash *et al*., 2009). Among the important genus of Artocarpus, jackfruit is the most useful and widespread tree. The tree is recognized by its fruits which are largest among the cultivated plants growing to medium size and reaches a height of 8 – 25 m (26 – 82 ft) (Elevitch *et al*., 2006).

It is widely grown in India and is native to the western ghats, Malaysia and was also found to be grown eastern and central Africa, south-eastern Asia, Florida, Brazil, Australia and in many specific islands (Rahman *et al*.,1999).

Jackfruit is regarded as the official state fruit of Kerala, where it is widely grown and is called as 'chakka' in Malayalam, in Tamil it is 'palaa', in Hindi it is 'kathal' or 'panas' and Gujarat it is 'phanas'. The jackfruit tree grows up to 10-15 meters in height and is commonly called as 'plavu' in Kerala. It grows at an altitude of 450-1,200 meter and is cultivated throughout the hot regions in India. The fruit is regarded as one of the largest and among the different species of tree's cultivated world-wide, jackfruit produces the heavier yield having a weight up to 35kg for an individual fruit. The tree is used for its fruit, timber, fodder and latex. Jackfruit has several uses where it is especially known for its health benefits, nutritional and agricultural benefits (Elevitch *et al*., 2006).

Artocarpus heterophyllus or Artocarpus heterophyllus Lam belongs to the mulberry family called Moraceae. The non-preferred scientific names of jackfruit are; Artocarpus brasiliensis Gomez, Artocarpus heterophylla Lam, Artocarpus maxima Blanco, Artocarpus philippinensis Lam, Polyphema jaca Lous, Soccus arboreus major Rumph etc. The ripen fruit flakes have a high nutritive value, where approximately 287-323 mg of potassium, 30.0-73.2 mg of calcium and 11-19 g of carbohydrates is present in every 100g of the ripen flakes.According to (Singh *et al*., 1963) jackfruit is referred to as 'poor man's food', because it has a major contribution to the food supply (Sim *et al*., 2003) to people and their livestock (Rahman *et al*., 1999). Both male and female was found to have inflorescence and hence it is called as monoicous tree (Bose, 1985). Cross-pollination causes the fertilization and the propagation of the tree is mostly through seeds (Morton, 1987).

About three to seven months is needed for the developmental process to form the complete fruit (Ranasinghe *et al*., 2019) and it depends up on variations of pollination in different countries (Baliga *et al*., 2011). The colour of the bulbs can be cream, white, yellow, light or deep yellow, orange, saffron depending upon the variety (Jagadeesh *et al*., 2007).

(Tiwari *et al*., 2015) studied that change in fruit colour is caused by the conversion of chlorophyll, anthocyanin and carotenoid like pigments during the stage of ripening. The tree was found to provide many environmental services. It is a good component in a windbreak as border planting because of its high wind tolerance. It provides fallen fruit for livestock, shade and long-term timber when grown in pastures. The dense jackfruit cultivated in home garden is very ornamental and it provides a visual screen. The seeds of jackfruit are boiled, roasted or eaten as chestnuts and is used in baking by the addition in flour or cooked in dishes since it has high nutritional benefits. The fruit has either compound or multiple fruit (syncarp) with a green to yellow-brown outer rind portion which has hexagonal and conical carped apices which cover over a rubbery thick white to yellowish coloured wall. It has banana flavored flesh with acid to sweetish taste. The central fibrous part called as the core holds the heavy fruit together. Maturity of the fruits takes place from a period of 90-180 days. The seeds are enclosed in a thin layer of whitish membrane with oval to round shape having 2-3cm in length and 1-1.5cm in diameter. Seeds can be stored up to a month in cool, humid conditions since it is recalcitrant.

According to (Morton, 1965) the rainforest of western ghats in India is believed to be the indigenous place of jackfruit's origin. The fruit contains vitamin A, minerals and other nutrients and has economics benefits too (Akter *et al*., 2018).

A study by (Arora, 1997) suggested that it is important to coordinate the national, regional and local bodies to strengthen and appreciate the genetic resource activities in tropical fruit trees which is also a guiding to the process of research and development. In Bangladesh jackfruit is extensively grown in Dhaka, Sylhet and Chittagong region due to its wide genetic diversity and propagation of seeds. Cutting down of mature jackfruit trees for timber and clearing of land for agriculture and flooding are some of the reasons for the destruction of the species.

Addressing the feeding issues with the increase in population can be overcome with the cultivation, conservation and sustainable use of underutilized crops like jackfruit has major concerns. Some jackfruits have firm, crisp flesh and pronounced flavor were some have soft, mushy flesh sweet and insipid when ripe (Morton, 1965).

'Rudrakshi' is a small fruited variety with smooth flesh and rind of inferior quality (Singh et al., 1963). Waraka with firm rind less sweet and vela with soft rind adds to peni waraka or honey jack with sweet pulp, which are the two principle varieties of Ceylon have been identified (Macmillan, 1925). The variety kuru waraka has round small fruits. (Barrett, 1928) identifies vela as predominant variety in west India. It was believed that the cultivation of jackfruit in tropical countries, especially in southern part of Asia was done during the beginning of the Christian era. The state of Kerala in southern India and the Sri Lanka was known to consume the fruit in major quantities and it a standard part of their diet in some seasons of the year.

There is a great demand for both the fruit and the timber and thus it contributes to a source of income to farmers. At least a couple of jackfruit trees have been grown in backyards of almost all homes in Kerala and Sri Lanka. Even though the fruit has great popularity, there is only little scientific studies have been done regarding its nutritional and other potential benefits. The flesh and the seeds of jackfruit is edible and is consumed by preparing curries in boiled forms etc. while the fully ripen flesh of the fruit can be directly consumed as fruit. Products such as ice creams using pureed jackfruit, jams, jellies and marmalades have been developed by different countries all over the world. It has extensively used in traditional medicine because the parts of jackfruit including the fruits, leaves and barks are found to exhibit anticarcinogenic, antimicrobial, anti-inflammatory, antifungal, wound healing and hypoglycemic effects. Even though jackfruit has all these benefits, unfortunately in commercial scale processing it is still underutilized in regions where it is grown. (Thomas, 1980).

2.5 Addition of pectin in value added food products

According to (Vanitha & Khan, 2019) food processing industries uses food additives to blend two immiscible liquids to form the desired product. Such food additives are called as food emulgents or emulsifiers. These are surface active agents acting on the borders of immiscible layers. Pectin is one of the emulsifiers widely used in food industry. It is used in the production of low-fat sauces, sausages, low-fat meat batter etc.

The replacement of pork back fat with 15% pectin and 15% inulin was found to be effective in maintaining the emulsion stability and physicochemical properties of low-fat meat batter. From long tradition pectin was used as gelling agent in food industry. Under suitable conditions pectin forms different types of viscoelastic solutions. Because of the gelling property, pectin is used in preparation of jams, jellies and marmalades. Development of biodegradable packaging materials helped to identify pectin as an edible packaging polysaccharide. Because of the flexible nature, pectin serves as barrier against moisture, oil, aroma and reduces the oxidation and respiration in foods. Pectin as a packaging material helps to prevent highly perishable foods from enzymatic browning, off-flavor development, aroma loss, retards the lipid migration and reduces the attack of pathogens during storage. Pectin is used as a natural ingredient in healthy meat product formulations because of its functional properties such as gelling ability, water binding ability and emulsifying property. When added to meat products as gelling or thickening agent, pectin interacts with meat proteins (Sharefiabadi *et al*., 2020).

A layer between food components or on the surface of the food are called as films or coatings (Korkmaz *et al*., 2018). For the manufacturing of films with nutritional value, biodegradability and environmental compatibility (Maftoonazad *et al*., 2007), natural components such as carbohydrates and proteins are used. Pectin and its derivatives are used as biodegradable packaging as it is a natural component and act as an effective barrier, preventing moisture, oxygen and lipid thereby helps in preservation (Hoorfar, 2014). Kitchen wastes are used to produce green packaging materials from biopolymers like pectin (Sindhu *et al*., 2019).

Coatings and edible films which are pectin-based as alone or enriched with antioxidants and antimicrobial substances were tested on meat and meat products (Tural *et al*., 2017). As a carrier for additives with specific properties such as anti-browning, antimicrobial agents, texture enhancers, nutrients, flavors and probiotics, edible films were prepared (Falguera *et al*., 2011).

To inhibit food surface contamination, antimicrobials were incorporated into edible films in additional to the barrier effect against pathogenic and spoilage causing microorganisms. On the evaluation of effect of free nisin and nisin-loaded pectin nanoparticles on the growth rate of Listeria innocua in fermented pork meat at different fermentation conditions and temperatures for 96 hours were done and a significant inhibition effect on Listeria innocua by using both nisin and nisin-loaded pectin particles were observed (Borges *et al*., 2016).

(Krivorotova *et al*., 2016). Sensory evaluation of sausages with pectin casings were preferred over the gelatin/sodium alignate casings for sausage production (Liu *et al*., 2007). Positive scores in sensory evaluation were observed while replacing pork back fat with 35% olive oil and 0.45% pectin (Pappa *et al*., 2000).

Enhanced colour, sensory attributes and physical qualities were observed when fat content in Chinese sausage with substitution with 5% mango peel pectin. Pectin from plant sources were used in meat industry. Pectin from apple pomace was used in meat products such as buffalo meat sausage (Younis *et al*., 2015), chicken sausage (Yadav *et al*., 2016), and in fat reduced chicken sausage (Choi *et al*., 2016).

A study by (Islam *et al*., 2023) confirmed that the pectin extracted from jackfruit by-products was of good quality and had promising applications in improving and modifying the thickening properties of vegetable soups.

The natural antioxidants from lime peel residue extract were incorporated with pectin films made from lime peels. Combination of these with coconut water was developed as a plasticizer and was applied on a sachet in soybean oils to retard the rate of oxidation (Rodsamran *et al*., 2019). Compared to films prepared with glycerol as plasticizer, films prepared with coconut water had low water solubility and was found to be flexible. The incorporation of lime peel extract in pectin film increased the total phenolic content and thus enhances the antioxidant activity of the film. The oxidation of soybean oil was found to be retarded by the film for 30 days on storage. Hence the combination of coconut water and lime peel pectin and its extract as packaging film has excellent food application in industries. Biodegradable pectin films were developed (Melo *et al*., 2017) by cocoa puree reinforced with chitosan nanoparticles, which had potential application as food packaging material.

Low-methoxyl pectin was found to be a promising additive in phosphate free meat product formulations since low-methoxyl pectin forms gel in the presence of calcium ions (Ko *et al*., 2014). Pectin also enhances the water binding properties of phosphate free and phosphate reduced meat products (Cho *et al*., 2018). Pectin from orange peel stabilizes the casein networks as filler along with calcium and produces enhanced texture and improves the shelf life of yogurt.

According to (Huang *et al*., 2021) in food processing industry, pectin is a choice generally regarded as safe (GRAS) food additive in the preparation of salad dressings, jellies, jam, bread, fruity milk drinks, yogurt drinks and ice creams due to its excellent properties like emulsifying, stabilizing, moisture retention ability and cold dispersion. Leaving about 50% of its portion as waste, citrus are fruits are widely produced fruits and thus valorization of this waste with the use of advanced technology and research helps reduce pollution and increases the economic benefits (Sharma *et al*., 2022).

CHAPTER - 3

METHODOLOGY

The methodology adopted for the study titled **"Extraction, Characterization and Applications of pectin from Artocarpus heterophyllus (Jackfruit)"** is given under the following headings.

3.1 Sample collection and preparation

3.2 Extraction of pectin

3.3 Quantification of pectin

3.4 Physicochemical characterization of pectin

3.5 Functional properties of pectin

3.6 Food application of the extracted pectin

3.7 Assessment of organoleptic qualities

3.8 Food labelling of pineapple jam with pectin extracted from jackfruit

3.1 Sample collection and preparation

The sample selected was Artocarpus heterophyllus (Jackfruit) of two varieties; varikka and koozha belonging to the family of Moracea, as these are very popular in Kerala. These are one of the largest fruits in the world and it is an important evergreen tree in tropical regions, widely grown in Asia. It weighs 3.5-10 kg on average and maximum weight upto 25 kg. Jackfruit comprises of five different parts namely; fruit, rind, seed, strand and core. The bulbs (which is the flesh) and seeds are edible in nature.

Figure 3.1: Artocarpus heterophyllus (Jackfruit)

Figure 3.2: Collected samples of jackfruit (variety-varikka)

Figure 3.3: Collected samples of jackfruit (variety – koozha)

Samples were taken from both the varieties of varikka and koozha. The parts like flesh, core, tandems, and rind portion of both varikka and koozha were taken, for the comparison. Varikka has firm flesh and koozha has soft flesh. The samples taken from Artocarpus heterophyllus was first subjected for pectin extraction then characterization and finally food application. The sample collection of three jackfruits of the variety varikka were done from the Ernakulam market, which weighed about 1.62kg, 1.75kg and 1.76kg. The koozha variety was collected from household ettumanoor, Kottayam, which weighed about 1.5kg.

Preparation of samples

Fully ripen small sized three jackfruits of varikka variety were collected from the local market in Ernakulam district, and one koozha variety was taken from household area of Ettumanoor Kottayam, Kerala. It was first weighed, washed and cut open, then separated into different parts.

Each part was cut into pieces (tandems, peels, flesh, and core) and was subjected for hot air oven drying at 100◦C for 3-4 hours. The dried parts were grinded and sieved to get the powder. The sample powders of different parts were packed separately in low-density polyethylene bags, sealed and stored at a temperature of 18◦C further analysis.

(a) Core

(b) Tandem

(c) Flesh

(d) Rind

Figure 3.4: Jackfruit samples- (a) core, (b) tandem, (c) flesh and (d) rind

(a) Rind

(b) Tandem

(b) Core

(d) Flesh

Figure 3.5: Weighment of the samples before drying

Figure 3.6: Hot air oven drying

(a) Core

(b) Tandem

(c) Rind

(d) Flesh

Figure 3.7: Weighment of the dried samples

3.2 Extraction of pectin

The dried and powdered samples were taken for extraction process involving multidisciplinary steps. About 50gram of four samples of dried varikka and 50gram of four samples from koozha were taken for extraction. The extraction process was carried out using HCl. First the dried powdered sample was mixed with 0.1 N HCl. After mixing the powdered sample in the acid solution it was kept in a magnetic stirrer for 30 minutes at a speed of maximum and temperature of 70◦C. The process of microwave oven acid boiling was also tested for samples to check on the efficiency in extraction process. Then it was cooled to room temperature and was subjected to ultrasonication process at 37KHz for 30 minutes at 80◦C in an ultrasonicating machine which included three cycles. The extract after the ultrasonication was subjected to purification and filtration.

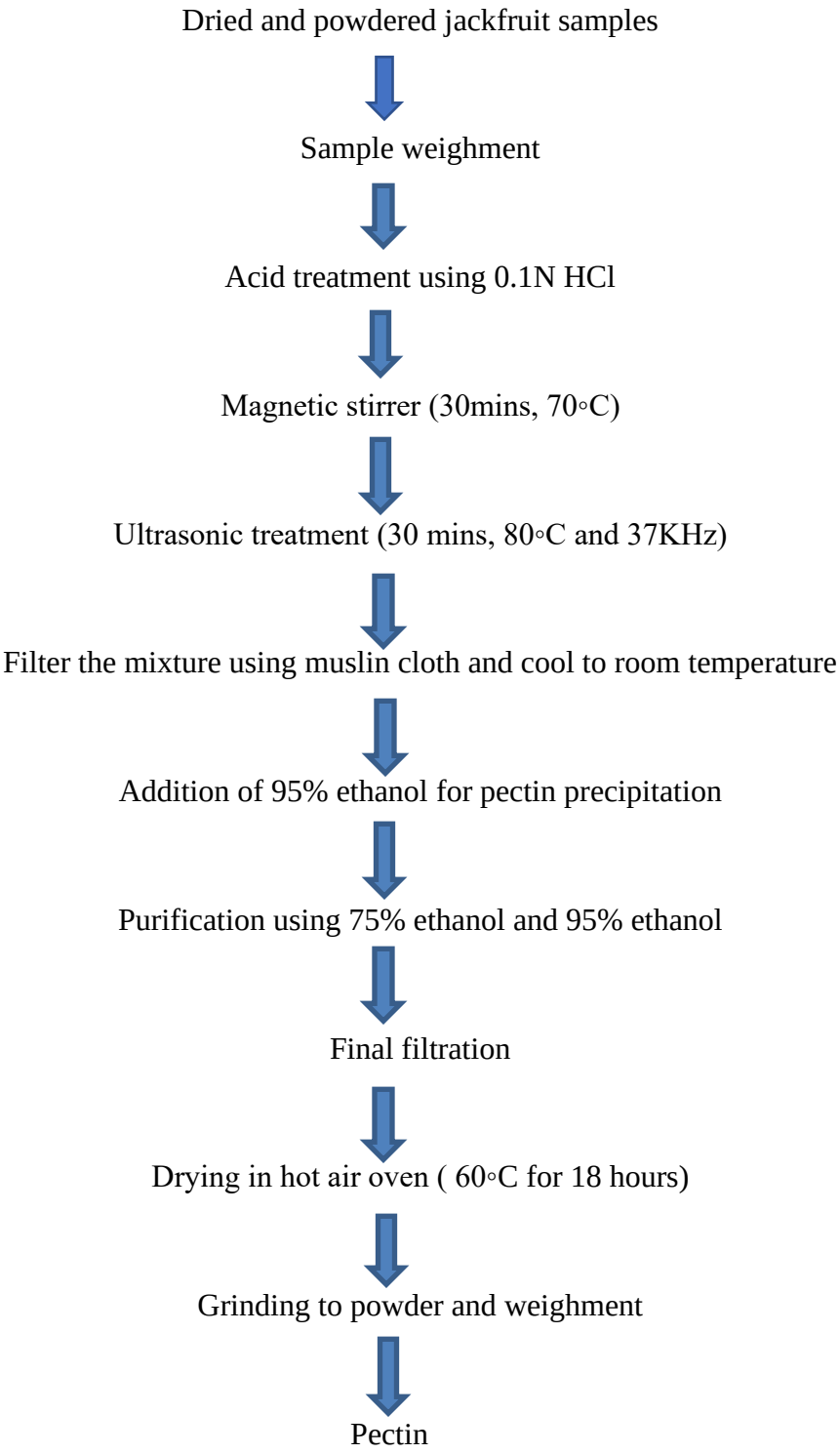
The extracts were filtered using muslin cloth and into the filtrate 95% ethanol was added. As the pectin is insoluble in alcohol the filtrate immediately forms thick and gel like precipitate. This can be taken as confirmation test for pectin. Then the formed precipitate was kept to cool in the refrigerator at -4◦C overnight. The cooled precipitate of pectin was subjected to washing with ethanol at various concentrations. The first washing was done using 75% ethanol for two times, then with 95% ethanol. Then the washed pectin was filtered and kept for drying in a hot air oven at 60◦C for 18 hours. Then the dried pectin was grinded to powder and weighed. The weighed pectin was stored in a polyethylene zip bag at room temperature for further analysis.

MATERIALS USED

Table 3.1 Chemicals and apparatus

Chemicals used	**Apparatus used**
0.1 N HCl	Hot air oven (Technotech)
Ethanol 95% (Labogens 500ml)	Magnetic stirrer with hot plate (MH 2lt)
Ethanol 75% (Labogens 500ml)	Microwave oven (Morphy Richards 20MWS)
Distilled water	Ultrasonic machine (Johnson plasto sonic-ultra weld 500)
	Cup viscometer (B4 ISO 3944)

Flow chart of pectin extraction



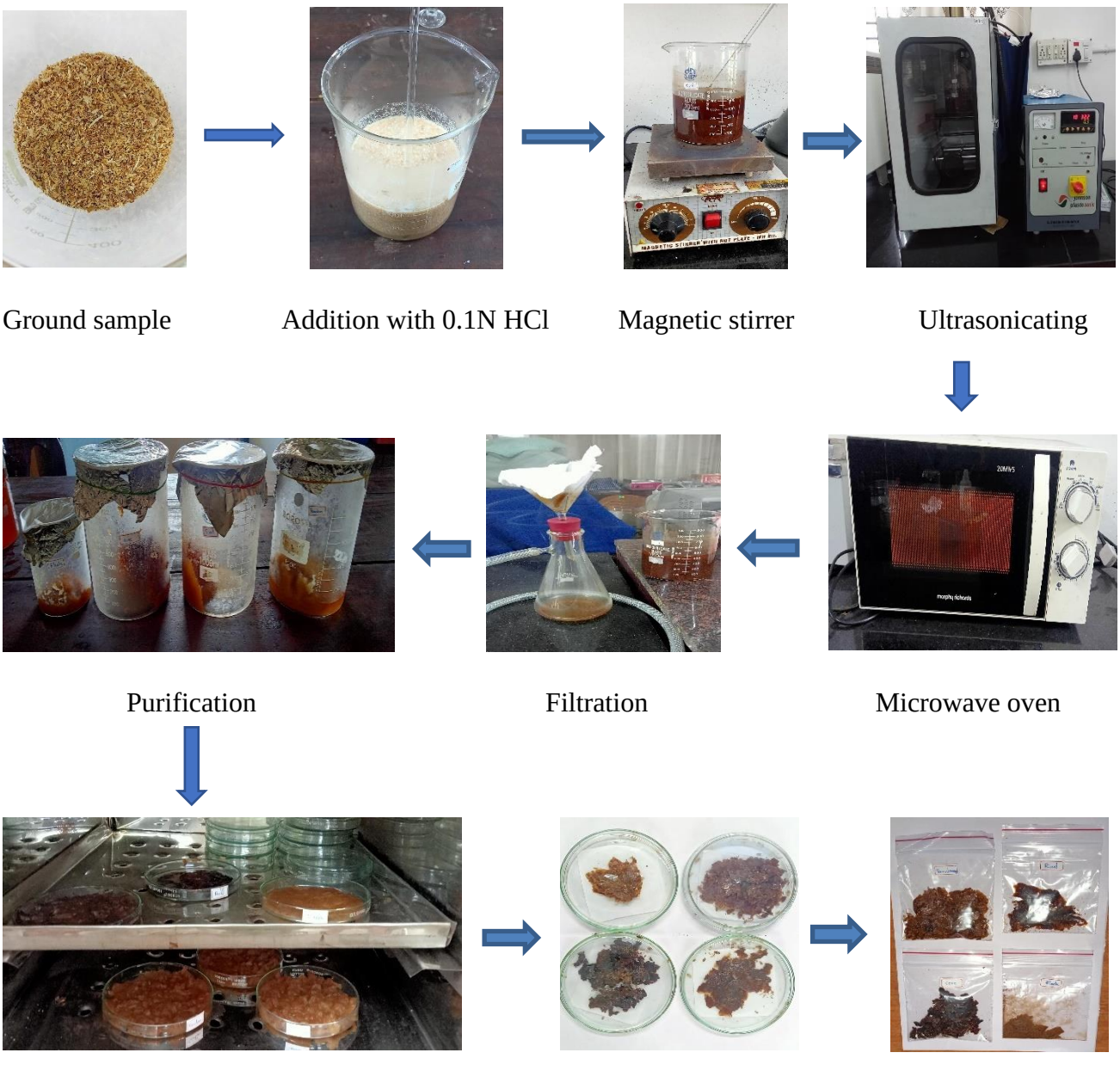


Figure 3.8: Pectin extraction process

3.3 Quantification of pectin

Pectin Yield

Yield of pectin was assessed from final extracted pectin by weighment method. The dried samples after the extraction process were weighed and the percentage of pectin yield was calculated. Pectin yield was calculated using the following equation;

Pectin Yield (%) = Amount of extracted pectin/ Initial amount of sample x 100

Or $P/Bi \times 100$

Where P is the amount of extracted pectin in gram,

Bi is the initial amount of powdered fruit samples.

Yield of pectin was assessed from final extracted pectin by weighment method. The dried samples after the extraction process were weighed and the percentage of pectin yield was calculated.

3.4 Physicochemical characterization of pectin

Characterization of pectin involves the identification properties of pectin such as degree of polymerization, Methoxyl content, Anhydrouronic acid content, Equivalent weight etc. and other properties including determination of ash and moisture content.

3.4.1. Moisture and ash content

The moisture and ash content of pectin from the four different parts of jackfruit varieties varikka and koozha were determined by methods using oven drying, and muffle furnace apparatus, respectively.

Determination of Moisture content

1gram of the sample (four different parts of both varikka and koozha) were taken for moisture content analysis. Weight of empty petri dish was taken. Then the sample was weighed in the same petri dish and was kept for heating in hot air oven for about 30 minutes. The heated sample was cooled by keeping it in a desiccator. Then the cooled sample was weighed and the weight difference was calculated to determine the moisture content present in the sample.

Determination of Ash content

About one gram of the sample (four different parts of both varikka and koozha) was taken for ash test. First, weight of empty crucible was taken, the weighed sample was transferred into it and weighed again. Then the crucible was kept in a muffle furnace to make the sample of pectin into ash. The ash was weighed and difference in the weight was calculated to determine the ash content.

3.4.2. Determination of equivalent weight

The equivalent weight of pectin is a measure of free galacturonic acid. It varies according to the type of extraction and depending upon the source of pectin. The equivalent weight of pectin can be determined by titration method. Eight samples of pectin were taken from parts of both the varieties koozha and varikka(core, tandems, rind and flesh). Sample of 0.5grams of the dried pectin was weighed and taken in a 250ml conical flask and it was moistened with 5ml of absolute ethanol. Added 0.1grams of sodium chloride into it and then added 100ml of distilled water. Six drops of phenolphthalein were added and then titrated it with 0.1 N NaOH taken in burette until the solution turns into a permanent pale pink colour. Equivalent weight was calculated using the following formula:

(a) **(b)**

Figure 3.9: (a)-Initial colour and (b)-final colour change during titration

$$\text{Equivalent weight(mg/mol)} = \frac{\text{Weight of Sample}}{\text{mL of Alkali} \times \text{Normality of Alkalki} \times 1000}$$

3.4.3. Determination of methoxyl content

Methoxyl content of pectin is the percentage of methoxyl groups attached to the galacturonic acid units, which indicates the degree of esterification. A high methoxyl content signifies higher degree of esterification and is noted as (HM) high methoxyl pectin and low methoxyl pectin as (LM). The determination of methoxyl content was carried out by saponifying the pectin and titrating the liberated carboxyl groups. The neutralized solution from equivalent weight determination was collected. To this solution 25ml of 0.25 N NaOH was added. The mixture was stirred thoroughly and let to stand for 30 minutes at room temperature. Then added 0.25 N HCl. This solution was titrated against 0.1 N NaOH in burette until the solutions turns into permanent pink colour. The percentage of methoxyl content was calculated using the formula;

$$\text{Methoxylcontent}(\%) = \frac{\text{mL of NaOH} \times \text{Normality of NaOH} \times 31}{\text{Weigh of Sample (g)} \times 1000 \times 100}$$

Where 31 is the molecular weight of the methoxyl group.

3.4.4. Determination of anhydrouronic acid content (AUA)

Anhydro-uronic acid content

It is an important parameter for determining the quality and suitability of pectin for applications such as jams, jellies and other food products. This parameter was often used as an indicator of pectin's purity and its gelling properties. To asses the anhydrouronic acid content, pectin was extracted from jackfruit (two varieties; varikka and koozha) and were analyzed using established methods such as gravimetry or titration.

The anhydrouronic acid content in extracted pectin was calculated by using the values of the equivalent weight and the methoxyl content. The AUA was calculated following the equation:

$$\text{Anhydro-uronic acid}(\%) = \frac{176 \times 100 \times 0.1}{\text{Weight of sample(g)} \times 1000 \times (z+y)}$$

Where, 176 g = molecular unit (1 unit) of AUA; z = mL (titer) of NaOH from equivalent weight determination; y = mL (titer) of NaOH from methoxyl content determination.

3.4.5. Degree of esterification

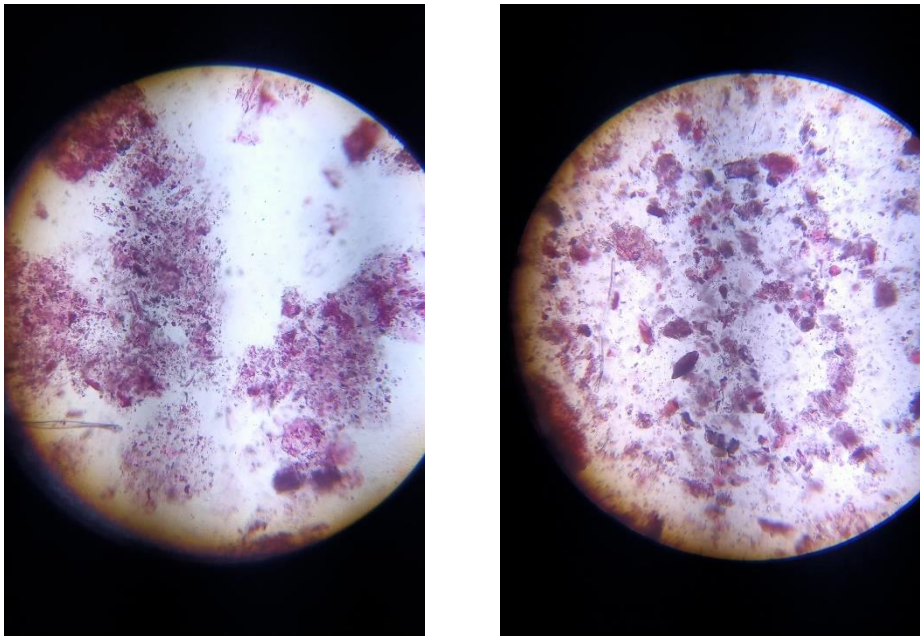
It is the percentage of esterified carboxylic groups in pectin. Pectin with a DE greater than 50% is high-methoxyl (HM) pectin, while pectin with a DE less than 50% is low-methoxyl (LM) pectin.

Degree of esterification was calculated using the formula;

$$\text{Degree of esterification (\%)} = \frac{176 \times \text{Methoxyl content (\%)}}{31 \times \text{AUA (\%)}}$$

3.4.6 Microscopic structure of pectin

The microscopic structure of the extracted pectin was observed using an electron microscope. A clean glass smear was taken. The extracted sample of pectin was made wet and gelled using distilled water. A drop of this sample was placed on the glass smear and stained using safranin dye. The prepared glass smear was covered with thin glass film and observed under electron microscope.



(a) Koozha **(b) Varikka**

Figure 3.10: Microscopic appearance of pectin

3.5 Functional Properties

Functional properties refer to the physical and chemical characteristics of food components that influence their behavior during processing, storage, and consumption. These properties go beyond basic nutritional value and play a crucial role in determining the texture, appearance, flavor, and stability of food products. Pectin, especially when extracted from jackfruit, demonstrates multiple functional properties that make it a valuable food ingredient. Its gelling ability is vital for products like jams and jellies etc. Its ability to bind water and form a viscoelastic gel matrix is influenced by factors such as pH, sugar concentration, and calcium ions. Additionally, pectin enhances the texture, consistency, and shelf stability of food products, making it a valuable ingredient in the food industry. The functional versatility of jackfruit-derived pectin underscores its potential as a sustainable and cost-effective alternative to commercially available pectin, particularly in regions where jackfruit is abundant.

3.5.1 Viscosity

The viscosity of extracted pectin was determined using viscometer. 0.5 grams of the sample powdered pectin was weighed and mixed with 100ml of distilled water to form a solution. This solution was heated to temperature of 40◦ C using a magnetic stirrer. Then the well dissolved solution was checked for temperature using a thermometer and at a temperature of 38◦C it was transferred into the viscometer cup and checked for viscosity. The time taken for the solution to fully pass through the cup or to fall down into the collecting beaker was noted. Viscosity was tested for the pectin extracted from core of both koozha and varikka.

Figure 3.11: Cup Viscometer (used for measuring viscosity)

3.6 Food application of the extracted pectin

The extracted pectin was subjected to food application in jam making as it has gelling properties. Pectin was added in pineapple jam. Pineapple was selected because of its low pectin content, seasonal availability and low cost and hence found to be a better option. Pineapple jam was prepared by adding the extracted pectin and two more jams were also prepared, one with the addition of commercial pectin and one with no added pectin. This was done to assess whether the natural pectin extracted from the jackfruit samples have same effect on addition to foods as like commercial pectin and natural pectin present in pineapple; thereby facilitating a comparison.

3.6.1 Preparation of jam using - Trial and error method

The trial and error method was employed to determine the optimal conditions for preparing pineapple jam with standard method. Four jams were prepared, one bottle with the extracted varikka jackfruit pectin, one bottle with the extracted koozha jackfruit pectin, one with commercial pectin and another with no pectin. Sugar and vinegar were also added in the jam preparation.

Step 1: Initial jam mixture

Four pineapples were purchased from the Ernakulam market each weighed approximately 1kg. The pineapple was made into puree form. Equal volume of four separate puree were kept aside. Amounts of sugar taken were one cup for all four jams (typically 50-70% of the total weight of fruit). Amount of extracted pectin (ranging from 0.5-2.0% of the total weight of the fruit) were taken for the jam preparation. One table spoon of acid (vinegar) was added to all four jams to adjust the pH to 3.0-3.5, which is ideal for pectin gel formation.

Step 2: Heating and cooking

The pineapple puree was heated in a saucepan and bought to boil. During each trial, the cooking time was varied in between (from 15-20 minutes) and the temperature was monitored closely (typically aiming for a temperature of 105-110℃ to achieve the gelling point). When the boiling point was achieved, 1cup of sugar was added and stirred occasionally (typically 50-70% of the total weight of fruit) and dose of extracted pectin (ranging from 0.5-2.0% of the total weight of the fruit) were added except the fourth jam with no added pectin. About one tablespoon of acid (example; vinegar) was added to adjust the pH to 3.0-3.5, which is ideal for pectin gel formation. The jam mixture was continuously stirred to prevent burning.

Step 3: Testing for gelling

A small sample of jam was placed on a cold plate and allowed to cool for few minutes. The consistency was then tested using the wrinkle test (dragging a spoon through the jam to see if it holds the shape) or by checking the temperature at setting point (using thermometer to ensure it has reached 105℃). The jam was assessed for its gelling property, texture and spreadability.

Step 4: Adjustments and repeats

Based on the results from the initial trial, adjustments were made to the mixture if necessary. For example; if the jam did not set properly, the amount of pectin was increased or if jam was too firm then the pectin concentration was reduced. Similarly, temperature and time taken were also altered accordingly until the desired gelling and consistency achieved.

Step 5: Comparison with commercial pectin and natural jam

An alternative jam was also prepared using the same steps but by adding commercial pectin instead of the extracted pectin. The jam prepared using the extracted pectin with best consistency and gelling textures was compared with jam made using commercial pectin, which was assigned to be the control sample. A jam with no added pectin was also prepared to see whether the properties of jackfruit pectin or commercial pectin or natural pectin present within the fruit is better in quality.

Pineapple Jam Recipe

Ingredients

- Pineapple: 1kg
- Sugar: 2 cups
- Cinnamon: 2 Inch piece
- Acid (Vinegar/lemon juice): 1 tablespoon

Preparation

- Pineapple was first peeled off, cleaned and cut into small pieces. This was made into a puree by using a mixer grinder. The puree was poured into a sauce pan which has thick bottom and was kept for boiling.
- When it started to boil, sugar and cinnamon was added and mixed well in medium flame. Cinnamon was added for flavor. Stirring was done continuously for about 20 minutes.

- Then added acid (vinegar or lemon juice) and again stirred for 2 more minutes. Acid was added to prevent the crystallization of sugar on cooling.
- When the mixture turns to golden colour and appropriate thickness stow can be turned off and the mixture was kept for cooling.
- The prepared jam was transferred into a clean air tight glass jar for storage.

Figure 3.12: Commercial pectin used for the preparation of jam

(a) **(b)**

(c) **(d)**

Figure 3.13: Pineapple jam prepared using (a) extracted jackfruit pectin from varikka, (b) extracted jackfruit pectin from koozha, (c) commercial pectin, (d) no added pectin

3.6.2 Spreading Ability

Spreading ability of the jam is one of its functional properties which determines the quality of jam. Pectin present in fruits helps in gel formation when jam is prepared and increases its thickness. Hence spreading ability can be seen as a property to assess good quality and consistency of jams. The prepared jam samples were assessed for its spreading ability using bread & jam spreading method. All four samples of jam; (A) Jam prepared using pectin from varikka, (B) Jam prepared using pectin from koozha, (C) Jam prepared using commercial pectin and (D) Jam prepared with no added pectin were spread on a bread slice and observed for the spreading ability. Bread used for checking the spreading ability was purchased from Ashis bakers of Ashis super mercato, Ernakulam.

3.6.3 Assessment of organoleptic qualities

Sensory evaluation of the jam

The sensory evaluation was performed using hedonic scale; A 5-point scale to rate each attribute. A panel of participants (5-10 numbers) was selected to evaluate the jams. The scores were averaged to obtain a final sensory profile. The organoleptic qualities like taste, smell, appearance/colour, flavor/aroma and overall acceptance were evaluated. Sensory evaluation was done by the staff and research scholars of Dr Johns Biotech Laboratory and center for research, Kollam.

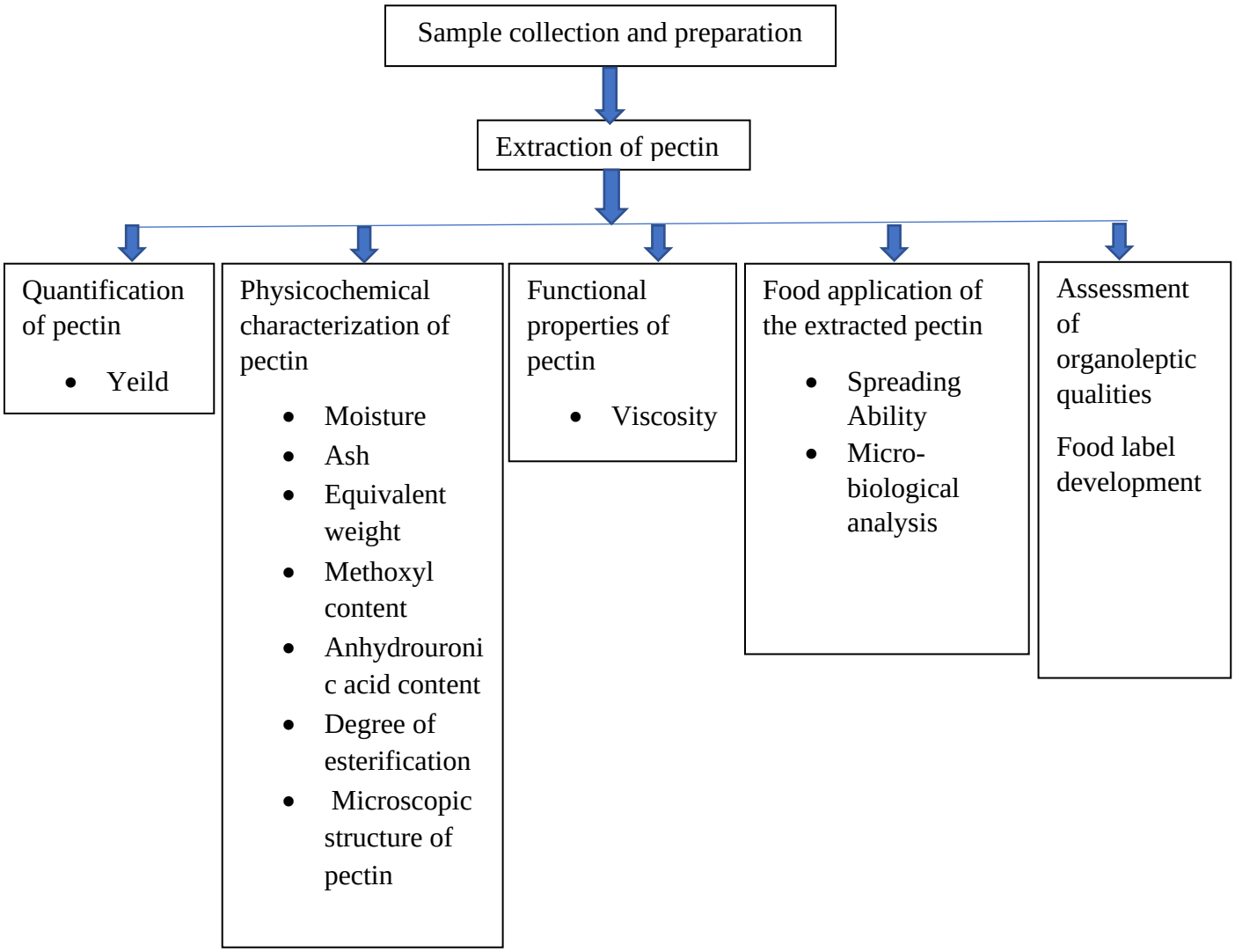
3.7 Microbiological analysis of the pectin added food sample (jam)

The microbiological analysis of the pineapple jam made with the addition of jackfruit extracted pectin was done by spread plate analysis. Three spread plates were prepared by initial sterilization by autoclaving at 121◦C for 15-20 minutes. The analysis was done to check on the growth of bacteria, fungi and total plate count. From the sample 0.1ml was pipetted over the center of the solidified agar medium and evenly spread over the surface of the medium. The plates were incubated under the optimum condition (37◦C for 24hours) following which the number of colonies developed were counted.

3.8 Food labelling of pineapple jam with pectin extracted from jackfruit

Food label was developed for the jam prepared with added jackfruit pectin aimed in creating awareness and food safety among consumers. The label was developed by following the FSSAI (Food Safety and Standards Authority of India) guidelines regarding the mandatory components required for packaging of foods. The label was designed using Canva. The label had two stickers, one to stick on to the front of the jam bottle which had main information like product name, caption and attractive designs. The second sticker was made to stick along the backside of the bottle which had information including, net weight, ingredients list, nutritional information, expiry date, storage instructions, manufacturer address, veg/non-veg logo etc. The jam was named as ‘Pine glow jam’. The label was developed with considering the regulatory standards and also adding the natural highlights of jackfruit by the attractive colour and theme. The animated image of pineapple and jackfruit was given to attract customers and especially kids. The labelling was not given to the jam bottles since further standardization procedures were required for the extracted pectin to remove the bitter taste.

Methodology overview



CHAPTER – 4

RESULT AND DISCUSSION

The results and discussion of the study entitled **'Extraction, Characterization and Applications of Pectin from Artocarpus Heterophyllus (Jackfruit)'** are discussed under the following headings;

4.1 Quantification of pectin

4.2 Physicochemical characterization of pectin

4.3 Functional properties of pectin

4.4 Food application of the extracted pectin

4.5 Assessment of organoleptic qualities

4.6 Food labelling of pineapple jam with pectin extracted from jackfruit

4.1 Quantification of pectin

Quantification of pectin was done to analyze how much of pectin was present in the selected fruit sample. Hence yield of pectin which was extracted from the jackfruit samples of two varieties varikka and koozha were done by calculating the total pectin yield.

4.1.1 Calculation of pectin yield

Pectin yield (%) represents the amount of pectin present in the samples after the extraction process. The calculation was done using the formula;

Pectin Yield (%) = Amount of extracted pectin/ Initial amount of sample x 100

Or $P/Bi \times 100$

Where P is the amount of extracted pectin in gram,

Bi is the initial amount of powdered fruit samples.

The results obtained are given in the table;

Table 4.1: Pectin yield (%) from different parts of jackfruit

SI No.	Jackfruit part taken as sample	Pectin yield (%)
1.	Core (koozha)	15.68
2.	Core (varikka)	11.87
3.	Tandem (koozha)	12.3
4.	Tandem (varikka)	10.98
5.	Rind (koozha)	6.24
6.	Rind (varikka)	10.98
7.	Flesh (koozha)	1.72
8.	Flesh (varikka)	2.55

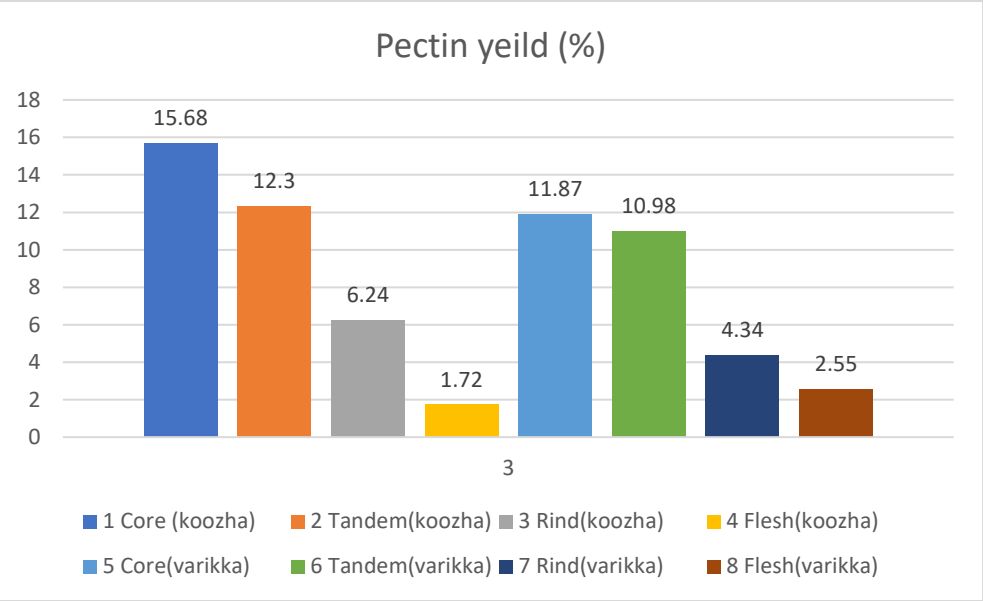


Figure 4.1: Pectin yield (%) from different parts of jackfruit

The total yield of pectin from 50gram of four samples from the jackfruit variety varikka and four samples from the variety koozha were calculated. As per the results the highest pectin yield was found in the core part of jackfruit for both the varieties. Second most yield was obtained from the tandems. The rind had pectin yield lesser than tandems. Lowest pectin yield was found in the jackfruit flesh.

Among the core part, the highest pectin content was found in the variety koozha (15.68%). For a comparison, core part of variety varikka was also taken which also showed good amount of pectin of about (11.87%) after the extraction process. From the overall results, the pectin yield from core, tandem and rind parts of the variety koozha was greater than that of varikka, but the pectin yield from the part flesh of the variety varikka (2.55%) was greater than that of koozha (1.72%). For an industrial level extraction of pectin from jackfruit, an extraction process from the whole jackfruit is recommended as it more economical and time saving and also gives better yield.

4.2 Physicochemical characterization of pectin

Physicochemical characterization involves the qualitative analysis of extracted pectin from the two varieties of jackfruit- varikka and koozha. The different important physicochemical characters of pectin including the moisture and ash content, equivalent weight, methoxyl content, anhydrouronic acid content, the degree of esterification and microscopic structure were analyzed.

4.2.1 Moisture and Ash content of pectin

Moisture content refers to the amount of water present in a material. It is usually expressed in percentage (%). Moisture content analysis is important for predicting the behavior of pectin during its processing, storage and consumption.

Ash content of a material is the amount of inorganic residue present or left behind after the process of burning the organic matter at high temperature. The inorganic residue generally comprises of minerals. Ash content of foods are expressed in percentage (%). Determination of moisture and ash content have application in food science, material science and combustion studies.

The results obtained for moisture and ash content of extracted pectin from the varikk and koozha jackfruit have been given in the table;

Table 4.2: Moisture and Ash content of extracted pectin

Sl.No	Sample	Moisture content (%)	Ash content (%)
1	Core (koozha)	9.8	4.6
2	Tandem (koozha)	9.3	4.9
3	Rind (koozha)	9.1	6.1
4	Flesh (koozha)	10.3	1.6
5	Core (varikka)	9	4.6
6	Tandem (varikka)	8.7	4.3
7	Rind (varikka)	7.6	5.3
8	Flesh (varikka)	9.6	1.3

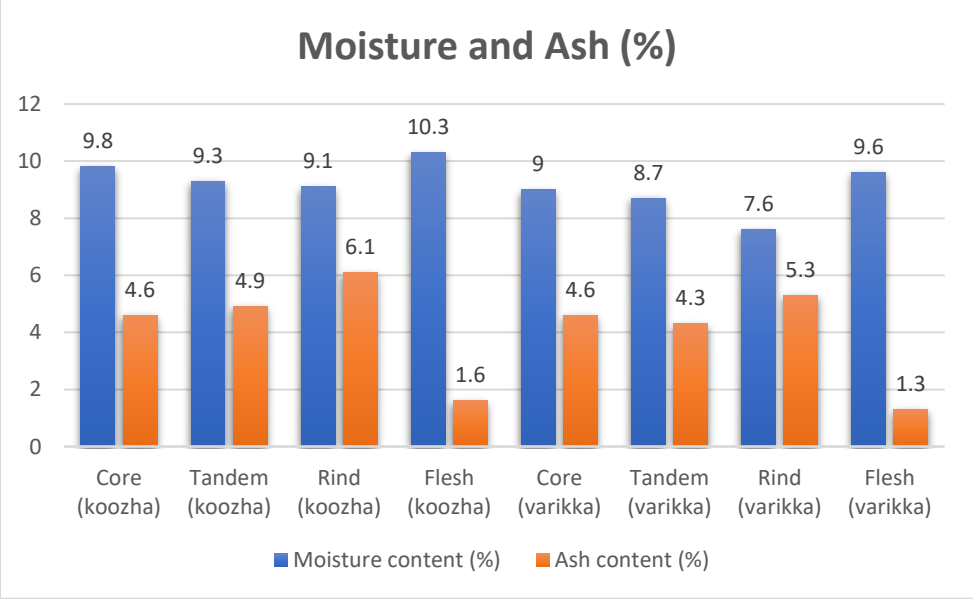


Figure 4.2: Moisture and Ash content (%) of pectin from different parts of jackfruit

The moisture and ash content of pectin from the four parts (core, tandem, rind and flesh) of varikka and koozha were determined by oven drying, and muffle furnace apparatus, respectively. From the moisture analysis it was found that the flesh of koozha had the greater moisture content of 10.3% followed by the core of koozha with 9.8%, tandem of koozha with 9.3% and the least moisture content in koozha was from the rind with 9.1%.

In the case of varikka the highest moisture content was found to be in the part flesh with 9.6%, followed by the core with 9%, tandem 8.7% and the rind with 7.6% which was the least. we can interpret that the moisture content is very low after all the drying process and hence can recommend a higher shelf life for the extracted pectin. Low moisture content helps to prevent microbial contamination and also as the water content is low, the pectin is found to be concentrated which has potential applications in food industry.

The Ash content was analyzed by weighing an empty crucible after in which one gram of the weighed sample was added and again weighed. This was charred and made into ash in muffle furnace after which the final weight of the sample was noted. From the values obtained it was found that the rind part of koozha with 6.1% had the highest value for ash content and the flesh of varikka had the least value for ash with 1.3%. In the case of koozha; the ash content was high in rind with 6.1% followed by tandem with 4.9%, core with 4.6% and the flesh with 1.6%. In the case of varikka; the ash content was found to be high in the rind with 5.3% followed by the core with4.6%, tandem with 4.3% and the flesh with 1.3% which was the least. This value indicates the inorganic residues remains after the combustion. Such inorganic residues implies that the extracted pectin is highly pure. As low as the ash level, then higher is the purity of pectin. The acceptable value of ash content for food grade pectin is commonly considered below 1%. Since the extracted pectin has ash value ranges between 1-6%, it is said to be good quality and has potential applications in both food industry and research field.

4.2.2 Determination of Equivalent weight

The equivalent weight of the extracted pectin from different parts of both the varities; varikka and koozha were done using titration method. Equivalent weight was calculated using the following formula:

$$\text{Equivalent weight (g)} = \frac{\text{Weight of Sample}}{\text{mL of Alkali} \times \text{Normality of Alkalki} \times 1000}$$

The results obtained were given below;

Table 4.3: Equivalent weight (mg/mol) of extracted pectin

Sl.No	Sample	Equivalent weight (mg/mol)
1	Core (koozha)	413.22
2	Tandem (koozha)	471.69
3	Rind (koozha)	442.47
4	Flesh (koozha)	537.63
5	Core (varikka)	458.71
6	Tandem (varikka)	480.76
7	Rind (varikka)	427.35
8	Flesh (varikka)	543.47

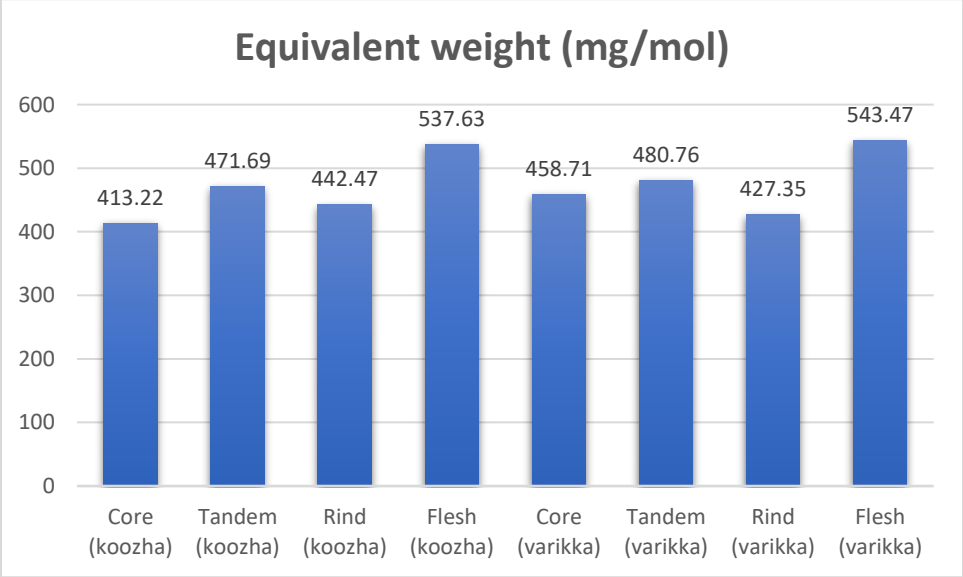


Figure 4.3: Equivalent Weight (mg/mol) of extracted pectin from different parts of jackfruit

Different parts of both varikka and koozha variety of jackfruit was taken for equivalent weight determination. From the results it was found that the core of varikka with 543.47mg/mol had highest equivalent weight compared to koozha with 537.63%, which indicates only a minor difference between the values. This indicates that only few free carboxylic acid groups are available for neutralization or we can say varikka has high molecular weight than koozha.

In case of koozha; the highest value for equivalent weight was in flesh with 537.63mg/mol followed by tandem with 471.69mg/mol, rind with 442.47mg/mol and the core with 413.22mg/mol, which was the least. In case of varikka; the highest equivalent was found to be in flesh with 543.47mg/mol followed by tandem with 480.76mg/mol, core with 458.71mg/mol and the rind with 427.35mg/mol. The koozha had lowest value of equivalent weight than varikka which indicates that it has more free carboxylic acids available which helps in greater reactivity that is advantageous in certain applications like low sugar or calcium sensitive formulations. On the other hand, the highest equivalent weight in varikka helps in strong gel forming properties.

4.2.3 Determination of Methoxyl content (%)

Methoxyl content of pectin is the percentage of methoxyl groups attached to the galacturonic acid units, which indicates the degree of esterification. A high methoxyl content signifies higher degree of esterification and is noted as (HM) high methoxyl pectin and low methoxyl pectin as (LM). Methoxyl content of pectin was calculated using the formula;

$$\text{Methoxylcontent}(\%) = \frac{\text{mL of NaOH} \times \text{Normality of NaOH} \times 31}{\text{Weigh of Sample (g)} \times 1000 \times 100}$$

Where 31 is the molecular weight of the methoxyl group.

The results obtained are given below;

Table 4.4: Methoxyl content (%) of extracted pectin

Sl.No	Sample	Methoxyl content (%)
1	Core (koozha)	4.18
2	Tandem (koozha)	6.97
3	Rind (koozha)	7.44
4	Flesh (koozha)	8.83
5	Core (varikka)	5.73
6	Tandem (varikka)	6.91
7	Rind (varikka)	7.30
8	Flesh (varikka)	8.68

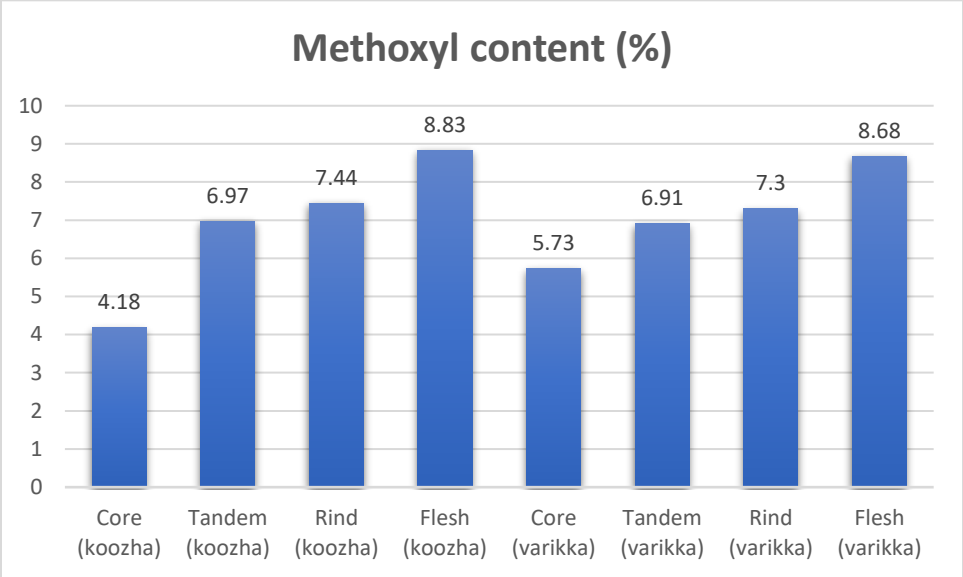


Figure 4.4: Methoxyl Content (%) of pectin extracted from different parts of jackfruit

From the results obtained it was found that the flesh of jackfruit had the highest methoxyl content (8.83%). In case of koozha; the methoxyl content was high in the flesh with 8.83% followed by the rind with 7.44%, tandem with 6.97% and the core with 4.18% which was found to be the least. In case of varikka also the flesh had the highest methoxyl content value with 8.68% followed by the rind with 7.3%, tandem with 6.91% and the core with 5.73%. Among the two varities; the flesh of koozha had the highest methoxyl content value. According to the results obtained, the methoxyl content of the extracted pectin were in the range of between 4-8%. This suggests that extracted pectin comprises of both high methoxyl and low methoxyl content based on its extraction conditions. The high methoxyl content of pectin which is greater than 7% is suitable for application in sugar acid gelation, whereas the low methoxyl content of pectin which is less than 7% is useful in forming calcium ions. The varience in the values was influenced by the extraction conditions such as pH, temperature and alcohol precipitation etc.

4.2.4 Determination of Anhydrouronic Acid Content (%)

Anhydrouronic acid content is one of the important parameters of pectin which determines its suitability to use in food industries as gelling, thickening and stabilizing agent. This parameter is often used as an indicator of pectin's purity and its gelling properties.

The AUA was calculated following the equation:

$$\text{Anhydro-uronic acid (\%)} = \frac{176 \times 100 \times 0.1}{\text{Weight of sample(g)} \times 1000 \times (z+y)}$$

Where, 176 g = molecular unit (1 unit) of AUA; z = mL (titer) of NaOH from equivalent weight determination; y = mL (titer) of NaOH from methoxyl content determination.

The results obtained are given in the table below;

Table 4.5: Anhydrouronic Acid Content (%) of Extracted Pectin

SI. No	Sample	AUA (%)
1	Core (koozha)	52.09
2	Tandem (koozha)	53.15
3	Rind (koozha)	56.67
4	Flesh (koozha)	52.80
5	Core (varikka)	51.39
6	Tandem (varikka)	52.09
7	Rind (varikka)	57.72
8	Flesh (varikka)	52.00

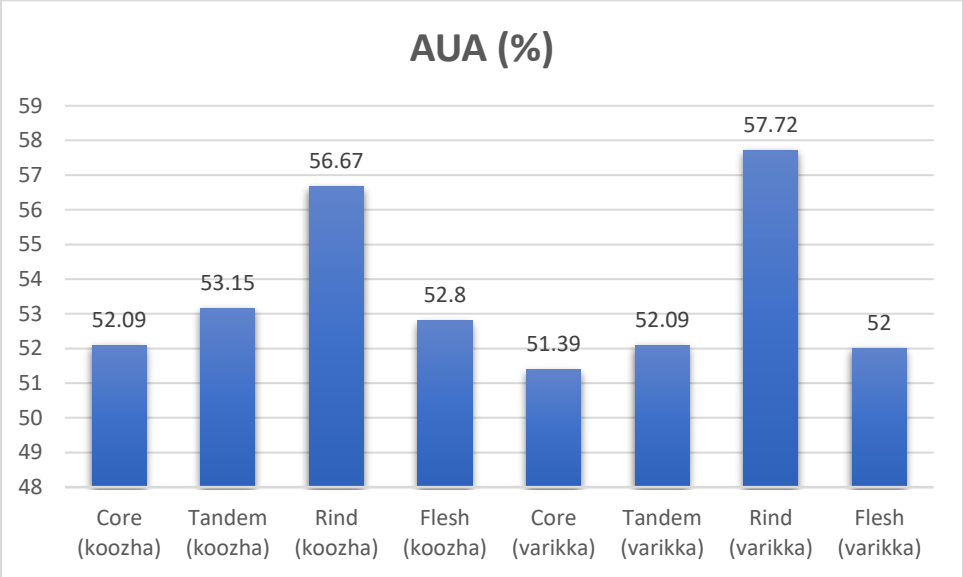


Figure 4.5: Anhydrouronic Acid Content (%) of extracted pectin from different parts of jackfruit

As per the results obtained, the anhydrouronic acid content was high in the rind portion of both the varieties of jackfruit and among the two varieties the rind of varikka had the highest anhydrouronic acid content with 57.72%. In koozha; the anhydrouronic acid content was high in rind with 56.67% followed by tandem with 53.15%, flesh with 52.8% and the core with 52.09%.

In varikka the anhydrouronic acid content was high in the rind with 52.72% followed by tandem with 52.09%, flesh with 52% and the core with 51.39% which was the least. The obtained values suggests that the pectin has potential applications as thickening and stabilizing rather than gelling. Generally, AUA content of pectin greater than 65% is considered ideal for gelling properties. Hence further purification of the extracted pectin with addition of alcohol helps in improving its gelation properties.

4.2.5 Determination of Degree of Esterification (%)

The percentage of esterified carboxylic groups present in pectin are called as degree of esterification. Pectin with a DE greater than 50% were regarded as high-methoxyl (HM) pectin, while pectin with a DE less than 50% was low-methoxyl (LM) pectin.

Degree of esterification was calculated using the formula;

$$\text{Degree of esterification (\%)} = \frac{176 \times \text{Methoxyl content (\%)}}{31 \times \text{AUA (\%)}}$$

The results of degree of esterification of the extracted pectin is given below;

Table 4.6: Degree of Esterification (%) of extracted pectin

Sl. No	Sample	Degree of Esterification (%)
1	Core (koozha)	45.55
2	Tandem (koozha)	74.45
3	Rind (koozha)	74.53
4	Flesh (koozha)	94.94
5	Core (varikka)	63.30
6	Tandem (varikka)	75.31
7	Rind (varikka)	71.80
8	Flesh (varikka)	94.59

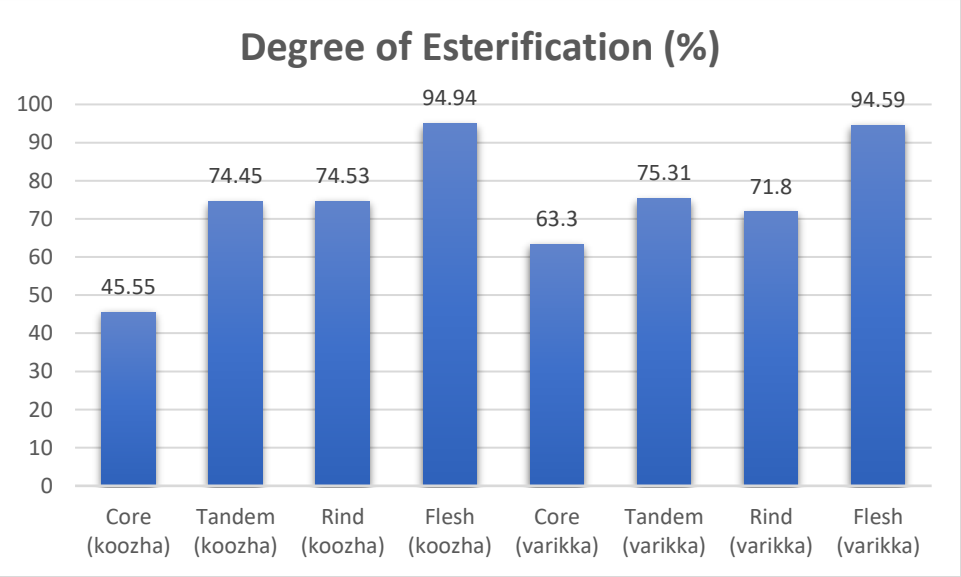


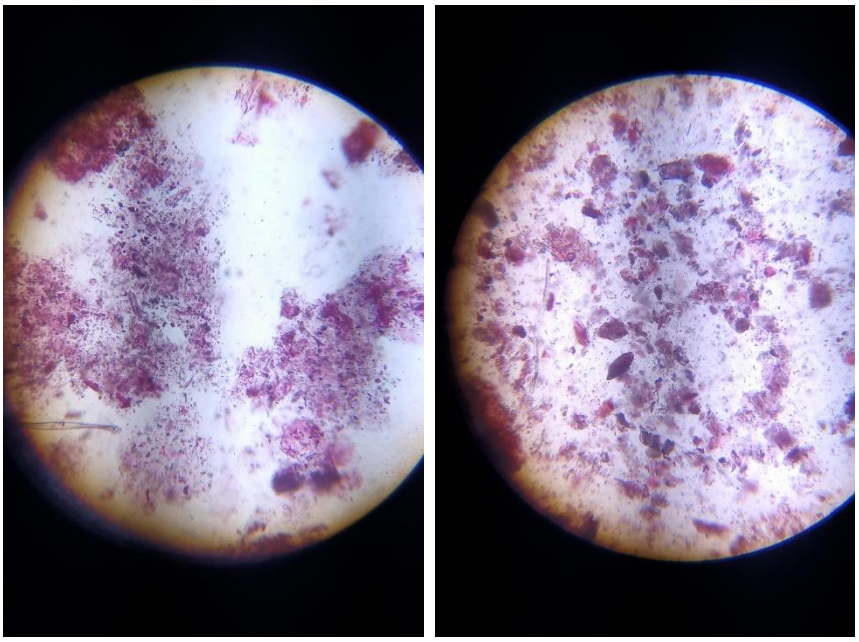
Figure 4.6: Degree of Esterification (%) of extracted pectin from different parts of jackfruit

The degree of esterification of pectin extracted from both the varieties koozha and varikka were done and it was found that the flesh of both varieties had the highest value for degree of esterification. In the case of koozha; the degree of esterification was highest in the flesh with 94.94% followed by the rind with 74.53%, tandem with 74.45% and the core with 45.55% which was found to be the least.

In the case of varikka the degree of esterification was found to be high in the flesh with 94.59% followed by the tandem with 75.31%, rind with 71.8% and the core with 63.3%. From the obtained results the values of pectin ranges from 45-94% where, values above 50% of degree of esterification indicates that the pectin has high methoxyl content and gels in the presence of sugar and acids which is commonly used in jams and jellies. Low degree of esterification values which is below 50% suggests that it forms calcium induced gels, which is suitable for low sugar applications. The mixed nature of the extracted pectin from different parts of jackfruit helps in enhancing its versatility in food applications.

4.2.6 Microscopic Structure of the extracted pectin

The results of microscopic analysis of pectin structure from samples-varikka and koozha are given below;



(a) koozha **(b) varikka**

Figure 4.7: Microscopic structure of jackfruit pectin- (a) koozha and (b) varikka

The samples of pectin extracted from Artocarpus heterophyllus (jackfruit) was diluted with distilled water, then stained and was examined under the light microscope. The structure of pectin appeared to be granular and in polygonal shapes and in different sizes. The structure appeared as pink colour due to the stain from the safranin.

4.3 Functional Properties of Pectin

4.3.1 Viscosity

The measure of resistance of a fluid to flow is called as viscosity. In simple words it is the property of fluids to easily flow which depends upon the thickness, temperature etc. The viscosity of the extracted pectin was analyzed using the cup viscometer (B4 ISO 3944). The pectin was dissolved in 100ml water and the time taken to flow in seconds under room temperature was noted. Results thus obtained are depicted below;

Table 4.7: Viscosity of the extracted pectin from different parts of jackfruit

SI No	Sample	Weight of the sample (g)	Volume of water (ml)	Time of flow (seconds)	Temperature (Degree Celsius)
1	Core (koozha)	1	100	15.09	37.9◦C
2	Tandem (koozha)	1	100	14.56	37.9◦C
3	Rind (koozha)	1	100	15.23	37.9◦C
4	Flesh (koozha)	1	100	14.49	37.9◦C
5	Core (varikka)	1	100	13.08	37.9◦C
6	Tandem (varikka)	1	100	12.53	37.9◦C
7	Rind (varikka)	1	100	13.21	37.9◦C
8	Flesh (varikka)	1	100	12.45	37.9◦C

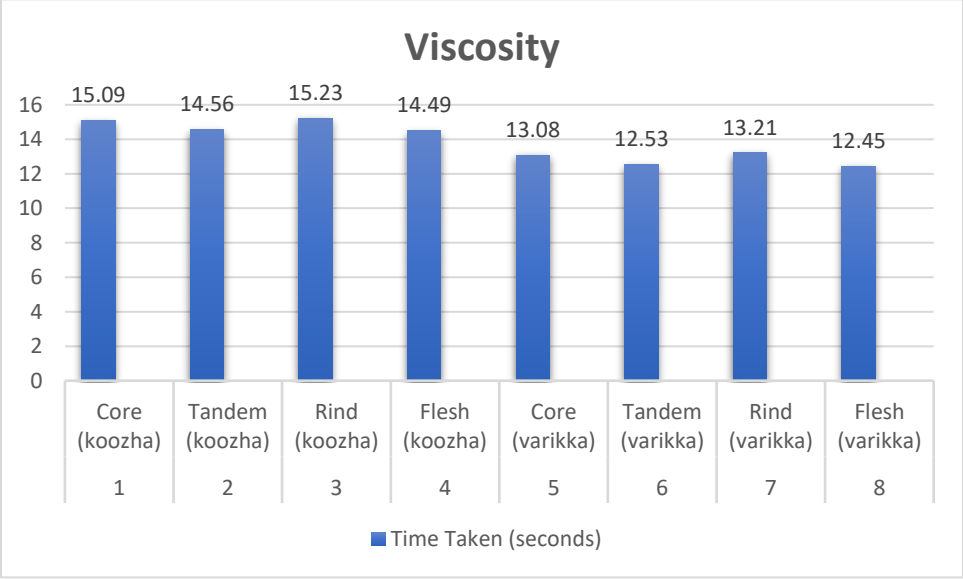


Figure 4.8: Viscosity analysis of the extracted pectin from different parts of jackfruit

From the values obtained the viscosity was highest in the pectin extracted from the rind of both the varieties of jackfruit. In the case of koozha; the highest viscosity was found in the rind with a time of flow of 15.23 seconds followed by core with time of 15.09 seconds, tandem with time of 14.56 seconds and the flesh with time of 14.49 seconds to flow through the viscometer.

In varikka, the highest viscosity was exhibited by the rind where the time of flow was 13.21 seconds followed by the core with 13.08 seconds, tandem with 12.53 seconds and flesh with 12.45 seconds which was the least. As the viscosity increases, then the time of flow of the liquid through the cup viscometer also increases. The viscosity property exhibited by the pectin extracted from jackfruit aids in better thickening applications

4.4 Food application of the extracted pectin

The food application of the extracted jackfruit pectin was done in pineapple jam. Four samples of pineapple jams were prepared. The prepared samples were;

A- Jam with extracted pectin from varikka
B- Jam with extracted pectin from koozha
C- Jam with commercial pectin
D- Jam with no added pectin

(a) **(b)**

(c) **(d)**

Figure 4.9: Pineapple jam prepared using (a) extracted jackfruit pectin from varikka, (b) extracted jackfruit pectin from koozha, (c) commercial pectin, (d) no added pectin

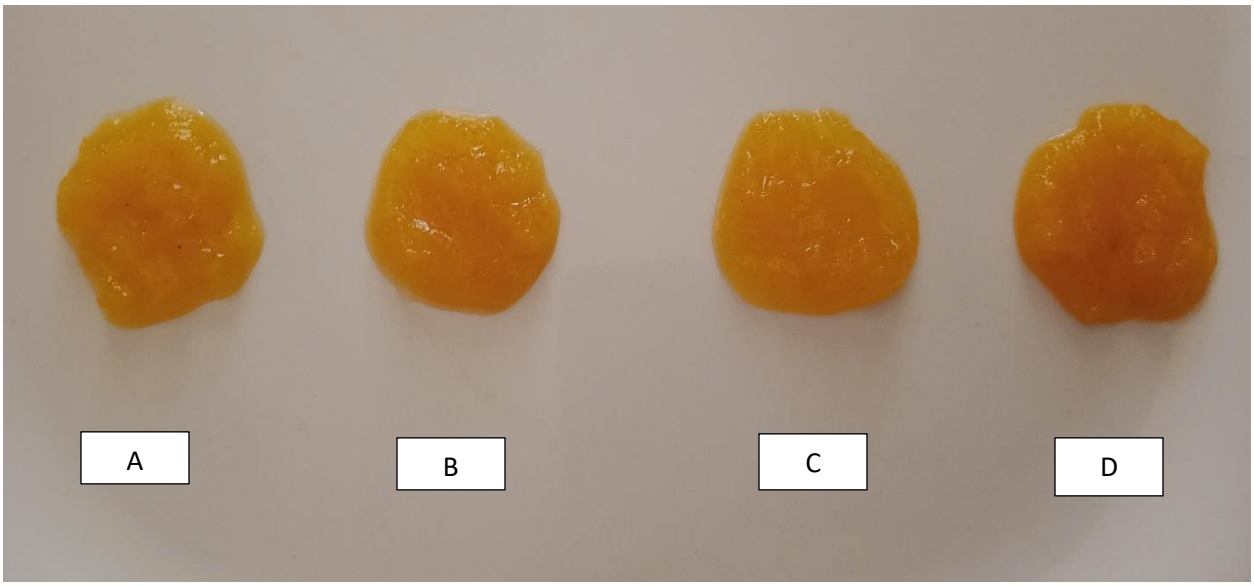


Figure 4.10: Prepared jam samples

4.4.1 Spreading Ability

Spreading ability of the jam is one of its functional properties which determines the quality of jam. Pectin present in fruits helps in gel formation when jam is prepared and increases its thickness. Hence it can be seen as a property to assess good quality and consistency of jams. The spreading ability of pectin was assessed by applying the prepared pineapple jam on bread. The results are given under the table below;

Table 4.8: Spreading ability of prepared jam samples

SI No	**Sample**	**Spreading Ability**
1	Jam with extracted pectin from varikka	Good spreading
2	Jam with extracted pectin from koozha	Good spreading
3	Commercial pectin added jam	Good spreading
4	No pectin added jam (natural)	Good spreading

(a) Varikka-pectin added **(b) koozha-pectin added**

(c) Commercial-pectin added **(d) No pectin added**

Figure 4.11: Spreading ability of -(a) Jam with pectin extracted from varikka, (b) Jam with pectin extracted from koozha, (c) Jam with commercial pectin, (d) Jam with no pectin

The spreading ability of the prepared jam samples were assessed to assess the functioning property of the extracted pectin, commercial pectin and natural pectin. From the assessment it was found that all the four samples of jam showed good spreading ability, the jam samples uniformly spread over the bread and had uniform thickening and binding texture. As there was no difference seen between the spreading ability of the jam prepared using the extracted pectin from jackfruit, commercial pectin and natural pectin, the pectin extracted can be recommended to have use in jam preparation.

4.4.2 Microbiological analysis of the jam prepared using extracted pectin

Table 4.9: Microbiological analysis of jam prepared with extracted pectin

Sample	Total plate count (CFU/ml)	Fungi	Bacteria/Coliforms
Jam (with addition of extracted pectin from jackfruit)	58	Absent	Absent

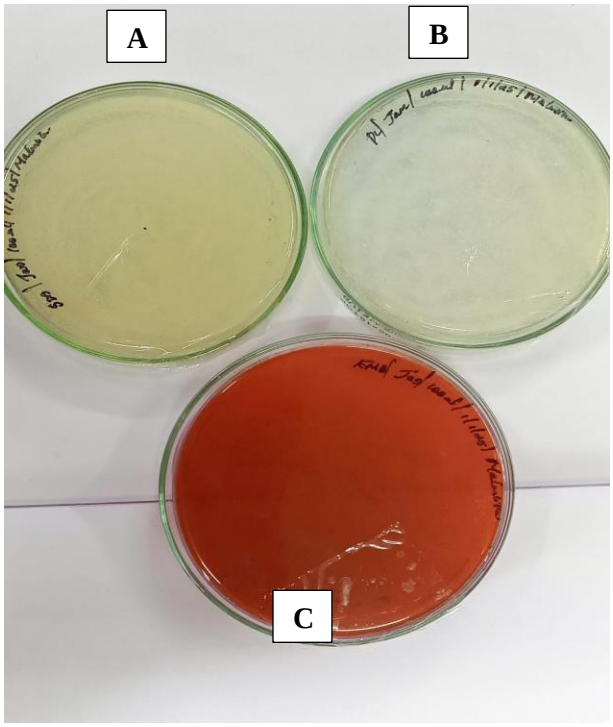


Figure 4.12: (A) TPC, (B) fungi and (C) bacterial plates of the food sample

The microbiological analysis of the jam prepared using pectin extracted from jackfruit was analyzed by preparing plates for bacteria, fungi and TPC, where it was found that there was no growth of bacteria and fungi in the sample plates. The total plate count was also found to be less than 60 CFU/ml. The analysis was done determine the growth of microbes in the prepared jam samples.

4.5 Assessment of organoleptic qualities

For assessing the organoleptic qualities like appearance/colour, taste, texture, smell/aroma and overall acceptance of the prepared jam, sensory evaluation was done**.** An overall mean value of all the five sensory attributes was also calculated. The results of sensory evaluation was discussed under the table below;

Table 4.10: Mean values from the sensory evaluation of the prepared jam samples

Sample	**Appearance/ Colour (5)**	**Taste (5)**	**Texture (5)**	**Smell/ aroma (5)**	**Overall acceptance (5)**	**Mean value (5)**
Jam with extracted pectin from varikka	4.6	3.26	4.13	4.23	3.6	3.9
Jam with extracted pectin from koozha	4.6	3.31	4.1	4.3	3.8	4.0
Jam with commercial pectin	4.46	3.96	3.9	4.43	4.03	4.1
Jam with no pectin added	4.5	3.96	3.93	4.36	4.06	4.1

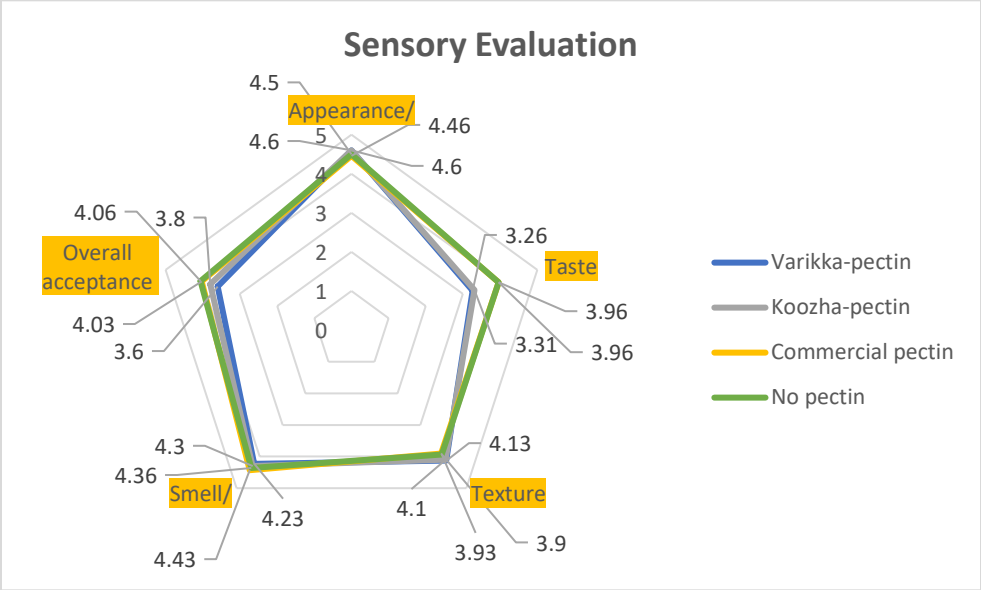


Figure 4.13: Sensory evaluation of the prepared jam samples

From the results obtained by doing sensory evaluation of the prepared samples of jam it was found that there was mixed opinion about the organoleptic qualities like appearance, taste, texture, smell and overall acceptance. For the jam prepared with pectin extracted from the variety varikka got mean value of 4.6 for appearance, 3.26 for taste, 4.13 for texture, 4.23 for smell and got an overall acceptance of 3.6. For the jam prepared with pectin extracted from koozha got the mean values for sensory evaluation as 4.6 for appearance, 3.31 for taste, 4.1 for texture, 4.3 for smell and 3.8 for overall acceptance. For the jam prepared by adding the commercial pectin was found to have the mean values for sensory evaluation as 4.46 for appearance, 3.96 for taste, 3.9 for texture, 4.43 for smell and 4.03 for overall acceptance. For the jam prepared with no added pectin got mean values as; 4.5 for appearance, 3.96 for taste, 3.93 for texture, 4.36 for smell and 4.06 for overall acceptance.

Considering each attribute, the highest mean value for overall acceptance was reported for the jam prepared with no added pectin, for smell it was for jam made with commercial pectin, for the texture the highest mean value was reported for both the jams made with extracted pectin from jackfruit. The taste was reported to be best in the jam made with commercial pectin and no added pectin, which showed same values as 3.96. The appearance or the colour of the jams made were better accepted was of both the jams made with pectin extracted from jackfruit. Considering the overall mean value, the jam made using commercial pectin and without pectin was found to be more accepted with a mean value of 4.1. Jam made with koozha got an overall mean value of 4.0.

Among the four jams prepared, jam made with varikka pectin got the least overall with 3.9 as mean value for sensory attributes. Pectin extracted from koozha had shown good texture and appearance than other samples only a slight bitterness in taste and had jackfruit aroma, which can be rectified by further standardization process. A slight bitterness was experienced in the jam prepared using the extracted pectin because of the acid treatments and further extraction conditions carried out during the extraction. Hence to reduce the bitterness of the extracted pectin, a standardization step which was found to be used in the manufacture of commercial pectin was recommended by the treatment of pectin with sucrose and calcium carbonate. Hence from the study it was found that the pectin extracted from jackfruit, especially Koozha has potential application in food as gelling and thickening agent.

4.6 Food labelling of pineapple jam with pectin extracted from jackfruit

Food label was developed for the jam prepared with added jackfruit pectin aimed in creating awareness and food safety among consumers. The label was developed by following the FSSAI (Food Safety and Standards Authority of India) guidelines regarding the mandatory components required for packaging of foods. The label was designed using Canva. The label had two stickers, one to stick on to the front of the jam bottle which had main information like product name, caption and attractive designs. The second sticker was made to stick along the backside of the bottle which had information including, net weight, ingredients list, nutritional information, expiry date, storage instructions, manufacturer address, veg/non-veg logo etc. The jam was named as ‘Pine glow jam’. The label was developed with considering the regulatory standards and also adding the natural highlights of jackfruit by the attractive colour and theme. The animated image of pineapple and jackfruit was given to attract customers and especially kids. The labelling was not given to the jam bottles since further standardization procedures were required for the extracted pectin to remove the bitter taste.

Figure 4.14 Food label developed for jackfruit pectin added jam

CHAPTER – 5

SUMMARY AND CONCLUSION

The study entitled **"Extraction, Characterization and Applications of Pectin from Artocarpus Heterophyllus (Jackfruit)"** was done to extract pectin from parts like rind, core, tandem and flesh of jackfruit varities- varikka and koozha and to analyze the physicochemical characteristics to ensure the eligibility of pectin for the application in foods. The study evaluates the yield, quality and functional characteristics of the pectin extracted from jackfruit which is an underutilized fruit and also explores the feasibility of using jackfruit as an alternative source for pectin. The aim of the study was;

To extract pectin from Artocarpus heterophyllus (two varieties- *varikka* and *koozha*)

Physicochemical characterization of the extracted pectin from Artocarpus heterophyllus

Food application of the extracted pectin in preparation of jam

Assessment of organoleptic qualities of the jam prepared using the extracted pectin from Artocarpus heterophyllus

The results of the study are summarized below;

- Highest pectin yield was found to be in the core part of both the varieties of jackfruit. Among the varieties varikka and koozha; koozha had the highest pectin yield of 15.68%. Lowest pectin yield was found in the flesh of both varieties ranging from 1.72 - 2.55%.
- Analysis of pectin extracted from jackfruit for moisture content indicated that; among the four parts of jackfruit, pectin extracted from the flesh had high moisture content with koozha having 10.3% and varikka having 9.6%. Moisture content was less in the rind of both the varieties when compared to other parts of jackfruit.
- Ash content was high in rind portion of koozha with 6.1% and varikka with 5.3% and was low in flesh of koozha with 1.6% and varikka with 1.3% when compared to other parts.
- Equivalent weight of extracted pectin was having the highest value in flesh of both the varieties; with koozha having 537.63g and varikka having 543.47g. Lowest value for equivalent weight in koozha was in the core part with 413.22g and in varikka was in the rind with 427.35g.

- Methoxyl content was high in the flesh portion of both the varieties. Flesh of koozha had methoxyl content of 8.83% and flesh of varikka with 8.68%. Methoxyl content was low in core of koozha with 4.18% and varikka with 5.73%.
- Anhydrouronic acid content was found to be high in the rind portion of both varieties of jackfruit. Rind of varikka had highest AUA content of 57.72% and in koozha, the AUA content was 56.67%. Lowest AUA content was found to be in the core, of koozha with 52.09% and varikka with 51.39%.
- Degree of esterification was high in the flesh of both varikka and koozha jackfruits. In the flesh of koozha the degree of esterification was 94.94% and in the flesh of varikka the degree of esterification was 94.59%. Among the four parts of jackfruit of both varieties, degree of esterification was found to be low in core of koozha with 45.55% and core of varikka with 63.3%.
- From the microscopic observation of the extracted pectin sample, the jackfruit pectin appeared to be in granular and polygonal in shape with different sizes.
- Viscosity analysis of extracted pectin showed that the pectin extracted from koozha were more viscous than varikka. The time of flow of pectin solution under room temperature through the cup viscometer was about 15.23 seconds in pectin from rind of koozha followed by 15.09 seconds taken by core of koozha. Pectin extracted from parts like tandem (koozha:14.56 seconds and varikka:13.08 seconds) and flesh (koozha:14.49 and varikka:12.45 seconds) had low viscosity.
- Spreading ability of the four prepared samples (jam prepared from jackfruit pectin: varikka and koozha, commercial pectin and jam prepared with no added pectin) didn't show much difference. All the four samples of jam had good spreading ability.
- Pineapple Jam was prepared using extracted pectin from jackfruit to find out its food application.
- Microbiological analysis of the jam prepared using the extracted pectin from jackfruit showed a Total plate count of 58 CFU/ml and absence was reported for the plates for fungi and coliforms.

- Sensory evaluation results showed that the jam with no pectin added had better appearance with 4.5 rating. Better taste was reported for jam with commercial pectin and no added pectin with 3.96 rating. Texture was better reported in jam prepared using extracted jackfruit pectin with 4.1 rating. Smell was reported better in the jam prepared with commercial pectin with 4.4 rating. Overall acceptance was better reported in jam with no added pectin.

CONCLUSION

Pectin is a heteropolysaccharide which is naturally present in fruits and is used as gelling, thickening and stabilizing agent in foods. Jackfruit is an evergreen tree widely grown in Kerala. Processing of jackfruit for several products in industrial level generates waste products such as rind, tandems and core. Hence extraction of pectin from these waste parts offers scope for its utilization in food industry. The study was successful in extracting pectin from different parts of two varieties of jackfruit. The underutilized core of both the varieties of jackfruit had the highest pectin yield, especially in koozha having pectin yield of 15.68%. The physicochemical characterization of pectin; like equivalent weight, methoxyl content, Anhydrouronic acid content and degree of esterification was observed to have different values for pectin extracted from different parts which implies that the pectin from same variety of jackfruit has different physicochemical characters. Microscopic examination showed granular and polygonal shape of pectin. Viscosity was observed to be high in koozha derived pectin.

The extracted pectin was successfully applied in jam preparation. The sensory evaluation for the organoleptic qualities showed good overall acceptability and the texture of jackfruit derived pectin was reported better for its spreading ability. Microbial analysis of jam sample with jackfruit derived pectin confirmed its safety for consumption with safer levels of Total plate count and showed absence of fungi and coliforms. Overall, the study points out the potential of using jackfruit waste parts for its better utilization as products like pectin. As per the results obtained from the study, the extracted jackfruit pectin was found to be a better alternative for commercial pectin and further research on optimizing conditions for better extraction and applications in field of food science and pharmaceuticals will increase the functional property and commercial viability of jackfruit derived pectin.

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APPENDIX

SENSORY EVALUATION OF THE JAM

Thank you for participating in the sensory evaluation of jam samples developed as part of my thesis work, titled *" Extraction, Characterization and Applications of Pectin from Artocarpus heterophyllus (Jackfruit)."* This evaluation involves four variations of jam: one prepared with by adding pectin extracted from varikka jackfruit, one with pectin extracted from koozha jackfruit, one with commercial pectin, and one with no added pectin. Your feedback on taste, texture, aroma, appearance, and overall acceptability is crucial in assessing the potential of jackfruit pectin as a natural gelling agent in food products. Please take a few moments to share your honest opinions by filling out the form below. Your valuable insights will contribute significantly to this research.

1. **Jackfruit pectin (varikka) added jam**

(5- Very good, 4- Good, 3- Fair, 2- Average, 1- Poor)

Sl no.	**Name of the evaluator**	**Appearance/ Colour (out of 5)**	**Taste (out of 5)**	**Texture (out of 5)**	**Smell/ Aroma (out of 5)**	**Overall Acceptance (out of 5)**
1						
2						
3						
4						
5						
6						
7						
8						
9						
10						
11						
12						
13						
14						
15						

2. **Jakfruit pectin (koozha) added jam**

(5- Very good, 4- Good, 3- Fair, 2- Average, 1- Poor)

Sl no.	**Name of the evaluator**	**Appearance/ colour (out of 5)**	**Taste (out of 5)**	**Texture (out of 5)**	**Smell (out of 5)**	**Overall acceptance (out of 5)**
1						
2						
3						
4						
5						
6						
7						
8						
9						
10						
11						
12						
13						
14						
15						

3. **Commercial pectin added jam**

(5- Very good, 4- Good, 3- Fair, 2- Average, 1- Poor)

Sl no.	**Name of the evaluator**	**Appearance/ colour (out of 5)**	**Taste (out of 5)**	**Texture (out of 5)**	**Smell (out of 5)**	**Overall acceptance (out of 5)**
1						
2						
3						
4						
5						
6						
7						
8						
9						
10						
11						
12						
13						
14						
15						

4. **Jam with no added pectin**

(5- Very good, 4- Good, 3- Fair, 2- Average, 1- Poor)

Sl no.	**Name of the evaluator**	**Appearance/ colour (out of 5)**	**Taste (out of 5)**	**Texture (out of 5)**	**Smell (out of 5)**	**Overall acceptance (out of 5)**
1						
2						
3						
4						
5						
6						
7						
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