

A DISSERTATION ON

**Shelf-life study of fresh cut pineapple pieces coated with a
nanoemulsion of *Trachyspermum ammi* L. & *Syzygium
aromaticum* essential oils and pectin**

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



**IN PARTIAL FULFILMENT
FOR THE
DEGREE OF MASTER OF TECHNOLOGY
IN FOOD TECHNOLOGY**

BY

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

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DECLARATION FORM

I, **Hina Khan**, a student of **B.Tech. - M. Tech Dual Degree Food Technology** (V Year/ X Semester), Integral University have completed my six months dissertation work entitled “**Shelf-life study of fresh cut pineapple pieces coated with a nanoemulsion of *Trachyspermum ammi L & Syzigum aromaticum* essential oils and pectin**” successfully from **Integral University, Lucknow** under the able guidance of **Dr. Rahul Sing, Assistant Professor and Dr. Roohi, Associate Professor, Integral University, Lucknow.**

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

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I wish her good luck and bright future.

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I wish her good luck and bright future.

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TO WHOM IT MAY CONCERN

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I wish her good luck and bright future.

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ACKNOWLEDGEMENT

I am deeply grateful for the divine blessings and guidance that have illuminated my path throughout this journey. I offer my heartfelt thanks to God for providing me with strength, wisdom, and resilience during the challenges of completing this dissertation work.

I would like to express my sincere gratitude and appreciation to all those who supported and guided me throughout the course of this thesis work.

First and foremost, I am deeply thankful to my advisor **Dr. Rahul Singh, Assistant professor, Department of bioengineering** for his unwavering guidance, insightful feedback, and continuous encouragement. Your expertise and mentorship have been invaluable in shaping the direction of my research. I am also thankful to **Mr. Vinay Kumar Pandey and Ms. Tripti Singh, Research Scholars, Integral University, Lucknow, Ms. Priyanka Dubey, Lab Assistant, Food Technology Lab** for their continuous support, motivation and untiring help during this project.

I extend my heartfelt thanks to my co-supervisor **Dr. Roohi, Associate professor**, for her valuable insights, constructive critiques, and for taking the time to review and provide feedback on my work.

My gratitude also goes to **Dr. Alvina Farooqui, Professor & Head, Department of bioengineering, Integral University, Lucknow** for creating an intellectually stimulating environment. The access to resources, seminars, and workshops has been crucial in expanding my horizons and refining my research skills.

I would like to express my sincere gratitude to my teachers **Dr. Ashish, Mrs. Poonam Sharma, Mrs. Gazia Nasir, Assistant professor, Department of Bioengineering** for their constant guidance, cooperation and support during my work.

I would also like to thank **Integral University, Lucknow** for providing the necessary resources, library facilities, and access to research materials that were crucial for the successful completion of this thesis.

I am indebted to my friends **Adiba Fatima, Ishana Khan, Shalini Singh and Ananya Singh** who provided me with stimulating discussions, shared resources, and offered moral support during the ups and downs of this journey. Your camaraderie made the challenges more manageable and the successes more meaningful. To my family, whose unwavering belief in my abilities and unconditional support have been a constant source of motivation, thank you.

Your patience during my long hours of work and your encouragement during moments of self-doubt have been crucial in sustaining my determination.

Lastly, I want to express my gratitude to all those who have supported me from the shadows, including mentors, role models, and authors whose works have shaped my intellectual growth. In closing, I am humbled by the collective efforts of those who have been a part of this journey. Your contributions have shaped this dissertation in ways beyond words. This work stands as a testament to your belief in me and your dedication to the pursuit of knowledge.

Place: Lucknow

Hina Khan

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LIST OF ABBREVIATIONS

CEO	Clove essential oil
AEO	Ajwain essential oil
rpm	Rotation per minute
°C	Celsius
TSS	Total soluble solids
TA	Titrateable acidity

ABSTRACT

The main objective of this study was to increase the shelf life and check the effect of physicochemical analysis on fresh cut pineapple at different storage conditions. Edible coating is used I fruits to extend the shelf life and improve the quality. To improve the edible coating of pectin and to make it more effective, clove and ajwain essential were also incorporated. The study was carried out to evaluate the ability of pectin based nanoemulsion coating to reduce and control the spoilage of fresh cut pineapple slices. The effect of clove and ajwain essential oil were also determined on the fresh cut pineapple. The nanoemulsion was ultrasonicated to ensure proper delivery on food surface. Fresh cut pineapple was coated in four different samples that is pectin solution(C2), pectin based nanoemulsion of clove essential oil(S3), pectin based nanoemulsion of ajwain essential oil(S4) and Pectin based nanoemulsion of clove and ajwain essential oil(S5), respectively. One control uncoated sample(C1) was also evaluated in comparison with other coated samples. Non coated sample shows deterioration at early stage, the color of uncoated sample also gets dark. Among the coated sample S5 pineapple sample have better color retention value, increased shelf life, reduced microbial growth, good antioxidant and antimicrobial activity. Therefore, 55 can be used for further future applications.

Keywords: fresh cut pineapple: Clove essential oil: Ajwain essential oil: Pectin: Nanoemulsion

CHAPTER 1

INTRODUCTION

Freshly cut fruits are ready to eat (RTE) products which are very convenient for consumers. The call for fresh-cut fruits is growing day by day because of busy lifestyles, consumers need products which are ready to eat and healthy which save their time. Today both men and women of the family are working which increases the demand of such products. Keeping this in mind fresh cut fruits are preserved by different means to protect it from spoilage and contamination. Because of the strong and increasing demand and the actuality that fresh-cut fruits are observed as a quick, convenient, nourishing, healthy, and safe food, they are growing swiftly and became popular, mainly during the epidemic. These fruits provide several benefits, like quick and easy preparation, less waste generation at home, uniformity in quality, a smaller volume used for packaging and storage, and less expensive transportation costs. However, the peeling procedure of fruit can quickly degrade the quality of freshly cut fruit and cut down its shelf life (Suriati.,2022). The shelf life of fresh cut fruits is prolonged by means of edible coating. Pineapple is a perennial crop scientifically known as *Ananas comosus*. It is the member and part family of the Bromeliaceae that is farmed for its delicious and nutritious fruit in Both tropics and subtropics regions. It comes in third place after banana and citrus fruit produced globally. The primary producers are in Africa (South Africa and Nigeria), Asia (China, Indonesia, India, Philippines and Thailand), and South-Central America (Brazil and Costa Rica). Thailand, Costa Rica, Brazil, and the Philippines were the top four pineapple-producing countries in the world in 2012. Pineapples should be harvested when ready to consume because they are non-climacteric fruits. Consumers will be given a base flavor that is appealing if the fruit's skin color changes from green to yellow at the fruit's base, the soluble solids content is at least 12%, and the acidic character is not more than 1%. The fruit consists of fiber, copper, bromelain, manganese, vitamin C, vitamin B complex, calcium, zinc, and beta-carotene and is eaten fresh, dried, tinned, in juice, and in jams. The flesh is minimal in sodium and calories, free of fat and cholesterol. Consuming pineapple has several health advantages since it helps bones grow stronger, stimulates the immune system, and facilitates protein digestion. Its use is suitable for all phases of life due to its nutritious qualities, texture, and juiciness. Due to their shorter shelf lives and great susceptibility to structural or mechanical disfigurement, fully ripe

yellow mature fruit is frequently inappropriate for delivery to far-off markets; as a result, somewhat less mature and less ripen fruits are chosen. Immature fruit shouldn't be transported since it won't develop a pleasant flavor due to high acidity, low soluble content, and is more likely to suffer chilling damage. The industry for fresh cut pineapple is growing vastly (Lobo & siddiq.,2017). Fresh cut pineapples are generally packed in a plastic box and available in hypermarkets but have no flavor because of loss of components. Pineapple contains 86% water which can lead to loss of nutrients and contamination when sliced or cut. To prevent the loss of water-soluble vitamins and spoilage moisture, gases barrier is used which is called coating. The sliced pineapples are coated with edible coatings. There is also a lot of pineapple peel left as a waste product which can be used as a source of antioxidant, for the production of cosmetics, pharmaceutical products, food additives, edible films and edible coatings.

Many organic compounds are extracted from pineapple to form composite edible films and edible coatings. Materials that can be possibly consumed by humans or that degrade naturally in the environment are referred to as edible films and edible coatings. Based on their purpose of use they can be divided into edible coatings, films, and sheets. In general, edible coatings are the liquid form of edible substances that may also include nanoparticles and other chemical and biological components. Wax coatings on fruit were the first edible coatings to be made commercially available in 1992. Edible coatings are now frequently employed on various fruits and vegetables, including tomato and aloe vera, as well as different types of fruits like apples, strawberries, bananas, and mangoes. Additionally, edible coatings on freshly cut fruits and vegetables are attracting a lot of attention because they protect them from various biological and microbial hazards that could cause food spoilage or different gastro-intestinal illnesses like diarrhea, acidity, abdominal cramps, and vomiting. Due to their essential characteristics against food related pathogens, enzymatic activity and lipid oxidation in packed food, such coatings and films play a significant role in extending the shelf life, maintaining physicochemical quality, as well as uplifting the microbiological character of packaged food. Since wastes are used as natural sources for edible coatings, these biodegradable compostable coatings are typically made from natural polymers like polysaccharides and proteins, which diminish the consumption of harmful chemicals and also boost the cost-effective packaging. Other benefits of such natural bio-compostable coatings include improving the quality and keeping quality of fresh foods and making it simpler to incorporate biological antibacterial agents into edible

coatings. Due to the effects of chitosan and other biological agents, the application of natural antimicrobial agents, such as green synthesized in chitosan edible coatings with nanoparticles and essential oils, has recently demonstrated a promising positive solution for the reduction of microbial contamination during shelf life. Pectin is found in fruits; it is a soluble fiber (polysaccharide). In baking and cooking, it serves as a thickening agent. In some scenarios, it is also utilized to form medications. Pectin helps to build volume in the stool and binds chemicals in the intestines. Additionally, it might lessen the quantity of cholesterol the body absorbs from food. Pectin is derived from citrus fruit and is also used to make edible coatings and films.

Pectin with essential oils of different varieties is used to form nanoemulsion composite coatings and films. Essential oil has bioactive, therapeutic and nutritive properties. (Suhag et al.,2020; paidari et al.,2021). The USFDA has classified essential oils as Generally Recognized as Safe having acronym GRAS, and because they are rich in bioactive contents and exhibit exceptional antioxidant characteristics and antimicrobial properties, they are used as food preservatives. However, Due to their excellent antibacterial and antioxidant activity, the use of essential oils as food additives has recently attracted increasing interest. These essential oils differ in terms of their biological activity, physical and chemical characteristics, and scent. For each individual application, it is crucial to choose the best one or combination. Clove and ajwain essential oil used in this study have antimicrobial and antioxidant properties with medicinal and health benefits. Clove and ajwain essential can be used in the form of nano emulsion coating because it increases the shelf life of fruits and vegetables, and it slightly affects the sensory characteristics of fresh cut fruits and vegetables (Chen et al., 2021; Pandey et al.,2022). Nanoemulsions are continuous phase emulsions that are dispersed in nanoscale size. These systems are not in equilibrium, they have kinetic stability, but lack thermodynamic stability. Oil in water (oil droplets in the aqueous phase) and water in oil (water droplets in the oil phase) nanoemulsions are the two basic forms of nanoemulsions. Different hydrophobic and hydrophilic compounds that are initially poorly soluble in the continuous phases can be solubilized using them. There are also advanced nanoemulsions, such as numerous emulsions of water in oil in water. Since around 1980, nanoemulsion has attracted more attention and has mostly been utilized to create nanoparticles. Since around 2000, direct uses of nanoemulsions in consumer goods, particularly cosmetics and pharmaceuticals, have grown in popularity.

After being transformed and included in the form of microcapsules, nanoemulsions function as a novel edible coating method that can work as a barrier to stop the release of natural substances into the environment. Applying these coatings to the food products' surfaces is simple and can be done in a variety of ways, including rubbing, spraying, and dipping. As they are formed from natural ingredients such as polysaccharides, proteins, organic and inorganic chemicals, and essential oils, these coatings have practical and therapeutic uses. The oil in water emulsions is widely used to form edible coatings among the many nanoencapsulating systems because they can easily be combined with ingredients of the highest quality and offer more scope for scaling up in the food industries (Tripathi et al.,2021).

The study has following objectives:

- To formulate essential oil based nanoemulsion edible coating.
- Coating fresh cut pineapple pieces with nanoemulsion- based edible coating solution.
- To study the shelf life and physicochemical properties of coated fruit at ambient temperature and refrigerated conditions.

CHAPTER 2

REVIEW OF LITERATURE

Fruits are handled and prepared in a minimally processed manner to retain their real freshness without compromising and changing their quality or nutrition or diminishing their shelf lives. Fruits that have undergone minimum processing are referred to as fresh-cut fruits. They are created through cleaning, washing, trimming/peeling, coring, slicing, shredding, and packaging. Storage of freshly cut fruit is often done at relatively lower temperatures. Some benefits of these products include giving purchaser a variety of choices in a solo package, quick preparation, lessening household waste, clearly displaying the product's internal condition, ensuring quality in comparison to whole fruit, making it simple to assess the quality of the products purchased, uniform and consistent, lower product volume, lower transportation cost (Nicolau-Lapeña et al., 2021). They are frequently available in grocery stores, fast-food outlets, and catering businesses. Physiological, physical, and pathological issues are some other factors that must be considered while handling fresh-cut fruit after harvest. The natural barrier (peel) that shields whole fruits from deterioration, pathogens, or environmental factors is often removed during the preparation of freshly cut fruits, which increases their susceptibility to decay. Fruits' complex physiology makes it considerably harder to maintain their quality than it is for vegetables. Fresh-cut fruit's biggest drawback is its short shelf life, which is frequently shorter than two weeks (Putnik et al., 2017). Fresh fruits are vulnerable to chemical, physical, microbiological, and sensory alterations when chopped or only lightly treated. This has an impact on the product's marketability, lowers its nutritional value, and has been related to multiple foodborne outbreaks.

The style of packing, post-harvest processing, and cultivar type are only a few of the numerous other factors that must be taken into account [6]. Therefore, new methods are being developed to enhance the microbiological and sensory quality of fresh-cut fruits, including active packing, natural additives, ultraviolet light, high hydrostatic pressure, and ozone treatments. Food additives that prolong the storage life and has edible coating as a first (primary) packaging to conserve as well as maintain and retain the natural freshness and nutrient quality of the product are two ways to deal with issues brought on by minimal processing (Hasan et al., 2021; Paidari et al., 2021). Watermelon, Mango, rambutan, jackfruit, mangosteen, durian, orange, melon,

papaya, pineapple, and fruit mixes are some of the fresh fruits that are now available in the market.

The most well-known tropical fruit is the pineapple (*Ananas comosus*). The pineapple originated in America, most likely in Paraguay and Brazil. As a commercial fruit crop, it has spread all over the tropical areas and subtropical areas. Hawaiian Islands, Malaysia, Philippines, Thailand, Kenya, Mexico, Brazil, Ghana, Taiwan, Australia, Puerto Rico, India, and South Africa are major pineapple-producing nations. More than 8% of the world's pineapple crop comes from India. Assam, West Bengal, Karnataka, Meghalaya, Manipur, Arunachal Pradesh, Kerala, and Bihar are the top pineapple-producing states in India. The output of 15.3 t/ha is used to produce 1.53 million Tonnes of pineapple in India. the pineapple is categorized into three varieties Cayenne, Queen, and Spanish. A significant source of nutritional fiber, sugar, organic acids, vitamins, and minerals, pineapples are high in nutrients and primarily comprised of carbohydrates and water.

Table 2.1 Nutritional value of pineapple (per 100g)

Energy	48 kcal	Vitamin C	36.20 mg
Carbohydrates	12.63 g	Vitamin b complex	1.06 mg
Protein	0.54 g	Potassium	115.00 mg
Sugars	9.26 g	Iron	0.28 mg
Fat	0.12 g	calcium	13.00 mg

2.1 THE NEED FOR EDIBLE COATING

The pineapple's thick skin makes it challenging to break apart, and the vast amount of storage space increases the expense of transportation. Hence, Due to rising command for fresh, easy, minimally processed, convenient, non-additive, and nutritionally sound products, the global including local and international market for freshly cut commodities have experienced significant growth in recent years. Because of their nutritive content, freshness, practicality, and distinctive flavor, fresh cut pineapples are well-liked by consumers. However, peeling off and cutting the fruit can result in mechanical and structural damage that causes tissue juice and

pulp to exude, which encourages paling and browning due to enzymes on open surface, nutritional losses from softer tissues, and spoiling, greatly decreasing the fruit's nutritional and commercial worth. Therefore, increasing the life of storage of fresh-cut pineapple and preserving it has gained a lot of study interest. Since edible coating (EC) preservation technique is risk-free, eco-friendly, affordable, and easy to apply, it is thought to hold promise for enhancing the quality characteristics of fruits.

Liao et al., 2023 studied edible coating (EC) + modified atmosphere packaging (MAP), MAP, and EC treatments and their effect on fresh-cut pineapples at 4° C. Associated studies have shown that complex or combined coating are more effective than single-coating in protecting freshness. The quality, physiological aspect, and overall microbial count was measured in order to analyze the quality variations and effect. According to the findings, the edible coating + modified atmosphere packaging was more effective in preserving storage quality and prolonging the shelf life of freshly cut pineapples. Edible coating is one of the easiest techniques used to preserve and extend the shelf life of fresh cut fruits and vegetables. As they act as barrier to gases. The semi permeable barrier provided by edible coating reduces the transfer of gases, moisture, aroma, flavor, nutritive components and also lowers structural damage on fresh cut fruits. different coating materials are used for different kinds of fruits. Sometimes not only is a single coating material used to formulate edible coating but also coatings with different combinations are required to make edible coating for enhancing the quality and keeping life of product. Thus, careful studies are made to select coating material for each product so as to provide efficient barrier to gases and moisture.

Coating can be done by three ways that is brushing, dipping, and spraying. Dipping is the most effective way of coating because it provides uniformity and consistency to the product. The usage of synthetic and non-biodegradable coatings was reduced as a result of environmental protection awareness, while natural and biodegradable edible coatings with eco-friendly ingredients received increased attention. The most recent development in the food packaging sector is the use of eco-friendly materials. One of the natural polysaccharides utilized to create the edible coating is pectin. Pectin is extracted from several sources, although it is only used for research. Pectin is only manufactured in large quantities from a select few sources, including apple and citrus fruits. To extract pectin, the researcher uses a variety of sources,

such as various types of fruits and vegetables. Due to its hydrophilic properties, the pectin-based edible coating is typically employed on fruits and vegetables that have low moisture content. The quality and food safety, mechanical and resistive property, physical and structural property, and microbiological contamination in food products are all improved by pectin edible coating forms with some additional functional ingredients (Lazaridou & Biliaderis.,2020). There are numerous studies on the formation of a nanoemulsion using diverse substances, like essential oils, and pectin edible coating.

2.2 HISTORY OF EDIBLE COATING

Edible coatings and edible films have been used for centuries in the food industry and technology to preserve and prolong the life of food products. For instance, cellulose coating in meat casings and waxing done on fruits and vegetables. In China, edible coatings have been in use since the 12th century. The waxing of fruits was not created until 1922 and was first used commercially on fruits and vegetables. Hot-melt paraffin waxes were first made commercially available in the 1930s as edible coatings for fresh fruits like apples and pears. According to Erbil et al. (1986), covering peach surfaces with wax emulsions reduced water vapor and oxygen transport, reducing respiration rate and lengthening fruit shelf life. Oils or waxes and cellulose, according to Baldwin et al. (1995), prevent rotting and maintain the fresh-picked quality of tropical fruits. There have been numerous attempts to create additional materials that could be utilized to coat and change internal gas composition for temporary storage. An edible coating that is non-phytotoxic, tasteless, odorless, and excellent at preserving fruits was described. Sucrose fatty acid esters (SFAE), sodium carboxymethyl cellulose, and mono- and di-glycerides are all components of this coating substance. SFAE was initially created to function as an emulsifier. However, it has been proven that a coating of SFAE will delay the ripening of fruits. Since the 1980s, SFAE mixes sold under the brand names "TAL Pro-long" and "Semper fresh" have been used to coat fruits and vegetables. Zhang and Quantick (1997) proposed using marine invertebrate chitin and chitosan (deacetylated chitin) to create a translucent film that may be utilized as an edible coating on fruits and vegetables. Zein coating was put to the surface of tomatoes by Park et al. (1994), who claimed that the film coating prevented color change, weight loss, and loss of firmness during storage.

2.3 ESSENTIAL OILS

Essential oils are made up of Terpenes, terpenoids, phenylpropenes, and other minor molecules, which are volatile liquids with distinctive aromas that are derived from plants. Due to their robust antibacterial activity and certain possible health benefits, such as their auxiliary characteristics in the treatment of rheumatism and their stimulant, carminative, and antispasmodic effects, they have drawn particular attention in the field of food preservation. Novel and new bio-degradable packaging substances based of natural biopolymers (polysaccharides, lipids, and proteins) have recently been developed. The antioxidant characteristics and antibacterial properties of essential oils can be increased by adding them, extending keeping life & lowering or inhibiting the growth of food-borne pathogenic organisms. The incorporation of EO with coating is intended to alter their functional properties, such as their water vapor permeability and antioxidant characteristics and antibacterial properties. Clove and ajwain essential oils have antibacterial, antifungal, therapeutic, and nutritional qualities. They are also high in antioxidants. They are used to combine coating materials with nanoemulsion. Eugenol and its derivatives make up the majority of the chemical components in clove essential oil (CEO), with small levels of α -humulene and β -caryophyllene. CEO is beneficial in a variety of industries, including food, biomedical, packaging, sanitary, cosmetics, and pharmaceuticals (Chen et al.,2017). CEO properties include antibacterial, antiseptic, antioxidant, pesticide, anticarcinogenic action and analgesic. CEO is frequently utilized in food as a spice, colorants, and natural preservative. Among other conditions, including high temperatures, low pressures, the presence of air and light, and others, essential oils have both volatile and labile chemicals which evaporate or dissolve effortlessly during production, utility, and storage, or while incorporation in food or packaging substance. The CEO's antibacterial property and antioxidant characteristics are severely constrained by its extremely evaporative and low number of water-soluble compounds, such as eugenol. Essential oils can be effectively protected from deterioration in severe settings by being encapsulated, which may also be used to extend the storage life of essential oils and give delivery systems a controlled application.

Numerous nanoencapsulation techniques for bioactive compounds have been studied (Assadpour & Mahdi.,2019). Iontropic gelation is very effective encapsulation method which produces great stability, a long usable life, a larger loading ability, good water dissolubility, and controlled delivery of the bioactive chemicals that were encapsulated. Essential oils are

complex mixtures of secondary metabolites from aromatic plants. Since ancient time clove oil has been used in food as a flavoring component and antimicrobial substance because of its biological properties, which include antifungal, antibacterial, antioxidant, and insecticidal properties. Additionally, it is used as an antiseptic in the treatment of oral infections. Clove essential oil prevents the formation of bacteria, yeast, and Mould (Pandey et al.,2022). In tryptone Soy Broth and cheese, it was proven to be effective against *Listeria monocytogenes* and *Salmonella enteritidis*. Due to the substantial volumes of eugenol, clove essential oils have natural antibacterial effects. Numerous gram-positive & negative bacteria, and yeast have shown that eugenol and the other phenolic components of clove interact with phospholipids in cell membranes and denature proteins, altering their growth and permeability. The utilization of essential oil(clove) helps to diminish the microbial growth of barely processed fruits. Pizato et al., (2022) studied the influence of chitosan coatings enriched with essential clove oil on quality of minimal treated strawberries in which treatments of different composition of chitosan and clove oil was given where chitosan with combination of essential oil(clove) was most effective than control samples. The application of chitosan and clove essential oil exhibited a slow growth of molds & yeasts, firmness and lower weight loss. The treatments thus proved to have potent effect to be applied as coatings for extension of product life as they lessen mass loss by approximately 86%, maintained chemical and physical characteristics, and bring down microbiological changes, mainly in mold and yeast growth.

The major component of ajwain essential oil is thymol, p-cymene, γ - terpinene, β - pinene, oleic acid, linoleic acid, palmitic acid. Steam distillation, solvent extraction, CO₂ extraction, maceration, enfleurage, cold press extraction, and water distillation are types of extraction techniques used in extracting ajwain essential oil from ajwain seeds. The quality of essential oils is dependent on the temperature and pressure used during the process of extraction. Thymol and ajwain seed oil are well known for their therapeutic benefits, including their antibacterial, carminative, antispasmodic, diuretic, antirheumatic, stimulant, expectorant, and Anti flatulent characteristics. Due to their environmentally friendly characteristics and GRAS (generally recognized as safe) status, natural antimicrobials, especially essential oils, are used as a potential replacement for synthetic preservatives. The possibility of encasing EOs in films, micro- and nanoparticles (MPs and NPs), micro- and nanocapsules (MCs and NCs), or nanocomposite materials has been raised. These methods boost AEO's stability in aqueous

conditions and, as a result, boost their bioavailability, minimize their negative effects, provide a controlled release for the encapsulated drug, protect them from the environment, or hide their pungent odor. To increase AEO's value, encapsulation has been used to increase its shelf life, improve its physicochemical stability, achieve controlled release, and suggest novel uses. Different encapsulation strategies using a variety of carriers have been developed and reported in research, with intriguing results.

The final products could be emulsions, complexes, micelles, liposomes, particles, or capsules. While some studies have emphasized on its activity against certain strains of bacteria, the majority have focused on its augmentation of stability and bioavailability as well as preservation of its valuable biological qualities throughout processing and storage. Babolanimogadam et al., (2020) demonstrated the effects of coating of chitosan as a base with ajwain essential oil (AEO) on extension of shelf-life of chicken breast in refrigerated condition. The AEO was incorporated with chitosan in varying concentrations and combinations to coat chicken breast by chitosan solution, The microbial properties of chicken breast, such Enterobacteriaceae, total aerobic psychotropic bacteria, *Pseudomonas* spp, and total aerobic mesophilic bacteria were studied during 13 days of period of storage. The results conclude that using AEO has exceptional effects on the depletion of all given groups of microorganisms when compared with the un-coated control group. In the present study, 1 percent v/v AEO treatment was the most effective and potent in microbial groups throughout the storage period of 13th days. In conclusion, this study demonstrates that chitosan coating containing 1% AEO has better antimicrobial activity and it can be used to extend the shelf-life of meat and chicken products.

2.4 NANOEMULSION BASED EDIBLE COATING

Commercially utilized immiscible liquids, water and oil, produce water-in-oil or oil-in-water nanoemulsions. Little spheroids of water are spread in oil containers to create water-in-oil nanoemulsions, whereas tiny drops of oil are scattered in an aqueous or water phase to create oil in water nanoemulsions. The emulsifying agent used for coating in water in oil nanoemulsions are often hydrophobic or lipophilic, whereas hydrophilic emulsifier is utilized for oil in water nanoemulsions. Due to the ability to include various lipophilic chemicals with antimicrobial property and antioxidant effects within a hydrophilic system, oil in water

nanoemulsions is perfect for creating edible coatings. A high liquifying fat/oil and a hydrophilic in nature surfactant existing in an aqueous or water phase are typically homogenized at a higher degree than the defrosting temperature of lipids to form an oil-in-water nanoemulsion. The system is then cooled below the lipid phase's melting point, which causes the formation of oil droplet crystals (Assadpour et al., 2019). Nano-emulsions, also referred to as ultrafine emulsions, mini emulsions, and submicron emulsions are colloidal systems that are heterogeneous in nature are made up of at least two immiscible liquids (non-mixable liquid) that are distributed into one another as tiny drops in the 20 to 500 nm range. At sizes between 50 and 200 nm, they are transparent, and at sizes up to 500 nm, they have a milky appearance. One of the two immiscible liquids must have an aqueous character, whereas the other must have an oleaginous character. Influenced by the particle size of the drops, nanoemulsions might have an optically clear or somewhat murky appearance.

Measurement of transmission yields the turbidity (τ) representation of opacity. Along with affecting optical properties and stability, droplet size also affects rheology and release behavior. Because of this, nanoemulsions are better suited for various applications than microemulsions. Nanoemulsions' tiny particle size has numerous significant advantages over other types. According to Nasr et al. (2022), nanoemulsions aid in preserving stability and enhancing physicochemical qualities. Due to their quick digestion, nanoemulsions give pharmaceuticals an aqueous state that renders their water insoluble and increases their bioavailability. As oil-in-water nanoemulsions are protected from assault by both air and water, they play a critical role in reducing the absorption changeability and in protecting against hydrolysis and oxidation. Nanoemulsion distribution can boost a drug's efficacy while reducing the overall dosage and negative effect (Garcia et al., 2022). Aqueous, oily, and emulsifying phases make up a nanoemulsion. The aqueous phase is mostly created using water. In addition to these, polar molecules are also utilized as co-solvents (simple alcohols and polyols), acids, minerals, and bases. Essential oils, triacylglycerols, diacylglycerols, monoacylglycerols, mineral oils, fat replacements, waxes, free fatty acids, lipophilic chemicals, vitamins, and weighing agents are among the non-polar ingredients utilized to prepare the oil part formulation. Numerous lipophilic substances, oil of essential components, mineral oils, waxes, replacements of lipids, vitamins soluble in oil, and weighing agents can also function as oil phases. Due to their availability, affordability, nutritional value, and functional properties,

long-chain triacylglycerols are the most appropriate nanoemulsion formulations. The sole aqueous phase of a nanoemulsion is formed by a polar solvent and a cosolvent. The oil containing phase physico-chemical factors, like its refractive index, measure of viscosity, interfacial tension, chemical steadiness, polarity, density, water dissolving ability, and behavioral pattern of phase, are largely responsible for the stability qualities and generation of nanoemulsions (Mushtaq et al.,2023). Nanoemulsions can be produced far more viscous or gel-like than conventional emulsions with much lower droplet concentrations, and as a result, they can be used to make substances with novel textural characteristics and low-calorie content. Due to the low oil content, nanoemulsions with low fat content suffer difficulties during preparation and should be created using binders, emulsifiers or fillers to increase their viscosity. Emulsions have been shown to be useful for creating nanoemulsions with low-fat solids that are very stable and function as thickeners. According to Dasgupta et al. (2019), the improved characteristics of nanoemulsions can be used in inventive, suitable, and original industrial applications.

2.5 EDIBLE COATING APPLICATION IN FRUITS AND VEGETABLES

Due to extensive commercialization, nanotechnology has recently grown to be an industry with a worldwide turnover. Numerous studies, papers, and patent applications have demonstrated the significant impact that nanotechnology has already had on different facets of food and associated businesses. According to Yousuf et al. (2018), coatings that are edible and edible films are recognized as practical, GRAS, and economical ways to extend fruit quality and storage life. Nanoemulsions are suitable for a variety of applications due to their enormous surface area and small size, which also provide several physiological and physicochemical characteristics. When products are stored, there is a significant loss of fresh produce, primarily as a result of mechanical damage, senescence, moisture loss, biochemical and microbiological degradation, and shrinkage. To improve the storage of foods like cheese, fresh-cut vegetables, meat, fruits, and confections, edible nanocoating based nanoemulsions can be used. These coatings contain enzymes, antioxidants, Anti browning, and antimicrobial agents. Aldehydes, esters and ketones are functional groups that coloring and flavoring ingredients have, leaving them susceptible to photolytic and oxidative destruction in food.

Ingredients can be encapsulated within nanoemulsions to reduce negative effects and extend shelf life. For food-borne viruses and microorganisms, oils from plants including oregano oil, citrus, thyme oil of essential form, and clove essential oil and their components like carvacrol, limonene, thymol, cinnamon, and eugenol have a high antibacterial potential. However, because they are hydrophobic, their use in food products is limited. Essential oil nanoemulsions are used in food packaging and as natural food preservatives. According to Jo et al. (2015), a nanoemulsion based on cinnamaldehyde prevented bacteria from growing in the melon juice. Nanoemulsion coatings have the ability to prevent gaseous exchange, minimize moisture loss, and prevent food deterioration. Due to their increased Oil in water surface area, oil-in-water nanoemulsions have the benefit of being quickly digested in the gastrointestinal tract for a variety of applications.

The effects of covering pears with a sodium alginate based nanoemulsion made of lemongrass or citral essential oil were investigated in a study. In variance to coated pears, which showed their efficacy in delaying maturation process, uncoated fruit shows loss of firmness and significant electrolyte breakdown. A coated pear with *Cymbopogon citratus* nanoemulsions also displayed no surface symptoms of scalding and maintained sensory qualities for up to 7 days of storage (Gago et al., 2020). Pineapples pieces were stored at 4 °C in 90% relative humidity for 12 days, and Prakash et al. (2020) examined the effects of sodium alginate-based nano-emulsions with varying amount of citral (1.0, 0.1 and 0.5%) on the physico-chemical, sensory, and microbiological activity of the fruit. Pineapple pieces coated with 0.5% and 1.0% citral showed more color retention, reduce and small respiration rate, and diminishing microbial load over the course of a 12-day storage period, indicating that these coatings could be used commercially to extend the shelf life of fresh-cut pineapple. Strawberry postharvest losses during storage and transit are very likely. They have a high respiration activity, which is influenced by storage period, maturity level, handling, and temperature conditions. Strawberries are prized for their phytochemical components, flavour, and scent. Horison et al., (2019) investigated the impact of nutmeg essential oil enriched with chitosan-based nanoemulsions on fresh strawberries stored at 10 °C for five days. The findings showed that samples coated with nanoemulsion had less Mould and yeast, and other microbial development at the conclusion of the storage time than the controls, demonstrating that nanoemulsion helps preserve the quality of strawberries stored for 5 days.

The effectiveness of quinoa protein/chitosan nanoemulsion coating incorporating thymol on quality, sensory, and safety features of chilled strawberries kept in commercial settings was the subject of additional research by Robledo et al. (2018). The addition of Thymol demonstrated fungicidal effects in strawberries kept under commercial refrigeration conditions and for nearly 10 days to quinoa protein/chitosan coatings (Mushtaq et al.,2023). The sensory parameter of strawberries, their flavor and perfume, were preserved by adding thymol antibacterial agent to the coating up to twelve days of cold temperature. Additionally, compared to control, the coated strawberry's post-harvest storage time was extended by four days. Mangos are climacteric fruits that are especially prone to postharvest losses because they are perishable. According to Klangmuang and Sothornvit (2018), biopolymers such hydroxypropyl methylcellulose (HPMC) does not exhibit inherent fungicidal properties. The essential oil (EOs) was added to the HPMC incorporated coating to enhance its antifungal property, and its effects on mangoes were studied. According to the results, mangoes coated with HPMC nano-composite coating and enhanced with Thai oils were able to be stored for 18 days at 13 °C. In addition, fruits with coating scored higher overall than uncoated control in terms of acceptability. In order to test the efficiency of nanoemulsions and physico-chemical parameters and microbial safety of plum fruit preserved at 4 °C and 25 °C, Kim et al., (2013) created wax of carnauba nanoemulsions having essential oil of lemongrass. The findings showed that by preventing microbial development, a nano-emulsion enhanced with lemongrass oil prolonged the shelf-life of plums. The impact of an edible nanoemulsion coating made of sodium alginate and sweet orange essential oil on several tomato quality indicators was studied by Banerjee et al. in 2020. The findings showed that compared to uncoated tomatoes, coated tomatoes exhibited a considerable rise in firmness, a drop in total bacteria, and a weight loss that was three times lower. The use of edible coatings boosted the tomato's overall approval score, according to the sensory analysis.

The freshly cut bell-pepper (red) quality stored for up to 21 days at common refrigerated temperature (4°C) was the subject of Sathiyaseelan et al.'s (2021) investigation into the effects of calcium chloride (CaCl₂), chitosan of low molecular weight, and tea tree essential oil nano emulsified coatings. Compared to the control, the application of a nanoemulsion preserved and retained the sensory behavior, overall quality, and texture character of freshly cut red bell peppers for about 18 days. The effects of applied antibacterial nanoemulsion of carvacrol on

leaves of spinach were investigated by Zhang et al. in 2021. The outcomes showed that the plants survived while being sprayed with carvacrol nanoemulsions at low concentrations (0.005 to 0.5%) on spinach leaves. However, a 5% rise in carvacrol concentration lowered spinach biomass and chlorophyll content while increasing electrolyte leakage and malondialdehyde production (Mushtaq et al.,2023). During storage at 5°C for up to 7 days, the impact of coatings with nanoemulsion containing antioxidant character and antimicrobial agents on the physicochemical characteristics, and microbiological stability of fresh cut kiwi slices was investigated by Manzoor et al., (2021). Alginate and carboxymethylcellulose coating formulations with Tween 80, vanillin, and ascorbic acid were employed as nanoemulsions. Nanoemulsion coatings were applied to freshly cut kiwi slices, with a control sample being an untreated sample immersed in water. The findings showed that, when compared to the control, nanoemulsion coatings considerably postponed loss of weight (8–14%), ascorbic acid content (73–79 mg/1001 g), decay rate (3-59%), preserved softness (14.1–17.9 N), and antioxidant activity (53–59%). A combination of emulsion coatings and the dispersion of various antimicrobial agent concentrations substantially reduced bacterial, yeast, and Mould growth during storage, extending the shelf life of fresh, ready-to-eat kiwi fruit. Pectin-coated fresh-cut apples were studied by Naqash et al., (2021). It was discovered that pectin nanoemulsion was more successful at limiting the spread of both pathogenic and non-pathogenic microorganisms, hence improving its safety. It made sure to keep the microbiological load within reasonable bounds, permitting the extension of shelf life. All coating treatments beat the control in terms of quality parameters including color and texture. The freshly cut apple treated with nanoemulsion was preferred over an untreated apple in terms of sensory qualities as well. As a result, pectin coatings functionalized with clove essential oil served as a useful biopolymer-based packaging component. This study demonstrates that pectin can be used as a possible candidate in creating edible active packing.

2.6 PREPARATION OF NANOEMULSION

For a nanoemulsion to be successfully formulated, choosing the right ingredient is crucial. Due to their low toxicity, GRAS: Generally Recognized as Safe ingredients such as food biopolymers (polysaccharides, proteins) and surface-active agents (spans, Tweens) are now employed to create nanoemulsions. The structural components utilized in edible films and

coatings, such as lipids, proteins, polysaccharides, or composites, are typically used to classify them. According to Liu et al. (2019), there are typically three techniques used to increase the chemical stability of nanoemulsions: adding antioxidants or chelating agents, modifying the interfacial properties (such as charge, thickness, and chemical reactivity), and managing the environmental factors (such as temperature, oxygen content, pH, and light). Various substances are used to create nanoemulsions and their characteristics. The creation of kinetically stable nanoemulsions via the sonication or ultrasonic method of homogenization is widely used in research labs nowadays (Jasmina et al., 2017). They increase the sonication period, power level, and emulsifier concentration, which reduces the droplet size by creating pressure waves from electrical waves (Fig. 3). High frequency sound waves with a frequency of at least 20 kHz are required for the ultrasonication process. Inertial cavitation is used by the ultrasonic probe to provide vital energy for breaking the droplets. In processes of nano emulsification performed by using ultrasonication devices, variables including the concentration/amount of gas used, apparatus configuration, hydrostatic pressure, and temperature play a crucial role. Out of all the high energy procedures, ultrasonication utilizes the least energy. Commercial homogenizers based on sonication have been developed for large-scale applications; in these devices, the nanoemulsion is then passed through a specific column with the capacity to generate ultrasonic waves.

2.7 METHODS OF APPLICATION

For the purpose of putting coatings on the surface of meals, various techniques have been developed. Many frequently used methods, like spraying, depositing layer by layer, dipping, casting, spreading, and electrospinning are commonly used and have been employed for centuries. Spraying uses globule droplets that are set to be roughly 20 μ m in size, which makes the viscosity of solution very less, and is delivered at pressure (60–80 psi) to the surface of the products. For the purpose of evenly applying coatings to the whole surface of the product, material is turned upside down during spraying (Ju et al., 2019). According to Chaudhary et al. (2020), the application method, drying time, and temperature all affect the uniformity of coating applied. When coating solutions with a high viscosity are involved, spreading rather than spraying is typically used. Although this process is quick and easy, it has several drawbacks, such as uneven coating manufacturing.

The products to be used are completely submerged in the formed solution of coating typically for ten to thirty seconds, and after normal air drying in fan at room temperature, a homogeneous layer is left over the surface. This technique is known as dipping (Fig. 4). Dipping needs to be carefully supervised since it creates a thick coating on the products' surface, which could lead to anaerobic conditions. When using the panning technique, the products are coated with a coating mixture in a "pan," which can be a pot or vessel. For the coating to be applied to the product's surface uniformly and completely, the solution travels continually. This method of coating application is used for both food and pharmaceutical products. After that, forced air is used to dry the pan or the temperature is raised. The process is mostly used to give confectionary items a final touch. The fluidized bed coating is a recently developed coating application process that has not only been embraced by the food and beverage industries but is also being used by the pharmaceutical business to polish and give shining to tablets (Chaudhary et al., 2020).

CHAPTER 3

MATERIALS AND METHODS

3.1 Procurement of raw material

Pineapple was purchased from a local vendor and stored in a refrigerator for further work. The essential oil of clove and ajwain were brought from an online store.

3.1.1 Apparatus used

A. Magnetic stirrer: A magnetic stirrer, also called a magnetic mixer, is a piece of lab equipment that employs a rotating magnetic field to fast spin a stir bar in order to stir a liquid. A magnetic stirring system often includes a related heating device for warming the liquid.



Figure 3.1 magnetic stirrer

B. Weighing Balance: A weighing balance is a tool used to calculate the mass or weight of an object. They are crucial instruments in laboratories, commercial kitchens, and pharmacies and come in a variety of sizes and weighing capacities. The unit of the weighing balance can be changed as necessary. A scientific tool used to precisely determine an object's weight is an electronic balance. Electronic balances are used in bakeries, chemical labs, and research facilities for science and medicine. A digital balance has a liquid crystal display, a backdrop light, the ability to count and weigh moving things, and other characteristics. They have a limited volume and great density.



Figure 3.2 Weighing balance

C. Ultrasonicator: A sonicator is an electronic device that performs sonication or applying sound energy to the target sample. The apparatus converts electrical energy into ultrasonic sound energy in the form of an ultrasonic bath or an ultrasonic probe (or probe sonicator). Dirt is frequently cleaned and removed using ultrasound. Using cavitation and ultrasonic wave bombardment, this technique can be utilized to remove solid deposits. Cavitation produces heat and energy, which raises the temperature and makes it easier to remove deposited materials.



Figure 3.3 Ultrasonicator

D. Refrigerator: A refrigerator, also known as a fridge, is a commercial and residential appliance made up of a compartment that is thermally insulated and a heat pump that transports heat from the inside to the exterior to cool the inside portion to below the room temperature. Globally, refrigeration is a vital method of food preservation.



Figure 3.4 Refrigerator

E. Hot air oven: The item is sterilized in a hot air oven for a predetermined amount of time under conditions that include humidity, pressure, and other environmental factors. Hot air ovens have the ability to control the humidity level by drying out the items and combining heat and airflow.



Figure 3.5 Hot air oven

F. Hot plate: A hot plate is a laboratory tool used to evenly heat materials, liquids, and samples at controlled temperatures without the risk of an open flame. Heat sources are located beneath a flat surface on hot plates.



Figure 3.6 Hot plate

G. Autoclave: In the healthcare or industrial sectors, steam sterilizers, often known as autoclaves, are frequently used. To eliminate harmful bacteria, viruses, fungi, and spores on things that are placed inside a pressure vessel, a device called an autoclave uses steam under pressure.



Figure 3.7 Autoclave

H. pH meter: A pH meter finds the hydrogen-ion activity in water-based solutions and uses that information to determine whether a solution is acidic or alkaline and expresses that information as pH. Since it measures the difference in electrical potential between a pH

electrode and a reference electrode, the pH meter is frequently referred to as a "potentiometric pH metre".



Figure 3.8 pH meter

I. Shaking incubator: Cell aeration, solubility tests, and cell culture are frequently performed in shaking incubators. The instruments equally distribute nutrients and introduce oxygen into the culture media without the need for a separate shaker within an incubator. It is used in microbiology mostly where optimum breeding conditions are required.



Figure 3.9 Shaking incubator

J. Spectrophotometer: A spectrophotometer is a device used to calculate how much amount of light a substance can absorb. It measures the intensity of light of a sample by passing a beam of light through it.



Figure 3.10 Spectrophotometer

K. Burette (acidometer): A burette is a graded glass tube with a tap at one end that is used to dispense known volumes of liquid, especially in titrations. The long, graded glass tube has a stopcock at the bottom, and a tapered capillary tube is situated at the stopcock's output. The stopcock valve controls how much liquid travels down the tube **to the burette tip**.



Figure 3.11 Burette

L. Refractometer: A refractometer is an easy-to-use instrument that is used to gauge the concentrations of aqueous solutions, including gases, liquids, and translucent solids. In accordance with the application, various refractometer kinds are offered.



Figure 3.12 Refractometer

M. Laminar air flow: Laminar flow is generally used as airflow within which the direction and velocity of all the air inside the given area are uniform. A laminar flow is a meticulously and carefully constructed table that is intended to keep biological and microbiological samples, semiconductors, and other very sensitive particle objects free of contamination.



Figure 3.13 Laminar air flow

3.1.2 Chemicals used

Pectin, clove essential oil and ajwain essential oil were used for the preparation of coating solution along with glycerol and polysorbate. Different types of glassware like conical flask, beakers, measuring cylinder, and storage container were used.

3.2 Preliminary trials

Preliminary trials refer to the earliest phases of testing or experimenting that are carried out before to a larger study or project. Before moving forward with further study or implementation, these trials are designed to collect preliminary data, evaluate feasibility, spot any problems, and make any necessary adjustments. In a variety of industries, including scientific research, medical investigations, product development, and engineering projects, preliminary trials are frequently used. In preliminary trials, scientists or project teams frequently use a smaller sample size or carry out tests on a smaller scale to get a better idea of how a certain concept or intervention would function in actual situations. The outcomes of early trials aid in decision-making and assist in determining whether additional funding, study, or development is necessary.

The different composition of coating material and essential oils were used to prepare an effective final coating solution. A total of four trials were performed to formulate coating solution with changing composition of pectin and essential oil. Table 3.2.1 shows the four preliminary trials used in this study.

Table 3.1 Preliminary Trials of samples

	Trial 1	Trial 2	Trial 3	Trial 4
Pectin	1.5	2.5	2.0	3.0
Clove essential oil	0.5	1.0	2.5	2.0
Ajwain essential oil	0.5	1.0	2.5	1.5
Clove + ajwain essential oil	1.5	2.0	3.0	2.5

3.2.1 Control parameters

Control variables are maintained constant so that their impact on other interesting variables can be observed. Control parameters in scientific studies assist in ensuring that any variations in the result may be attributed to the variables being researched rather than to unrelated external

variables. Researchers can isolate the effects of the variables they are examining by changing or controlling specific parameters. The integrity and validity of studies must be maintained by using control parameters, which serve to account for or eliminate potential confounding factors that could otherwise alter the results. The volume of coating solution and nanoemulsion solution are kept constant and it is not changed throughout the study to avoid any changes in results. Control parameters in this study is shown in table 3.2.2

Table 3.2 Control parameters

Weight of sample	
Coating solution	100 ml
Nanoemulsion solution	50 ml

3.2.2 Selection of independent variables

In an experiment, independent variables are the variables that are altered or changed to see how they affect the other variables. They are the presumptive causes or variables that might affect the dependent variable, which is the result or response being assessed. In order to analyze how the independent variable affects the dependent variable, the researcher must control or manipulate the independent variable. Independent variables are selected by changing or modifying the composition of coating material and essential oils. The composition of pectin, clove essential oil and ajwain essential oil was modified to check the effect of coating on fresh cut pineapple.

Table 3.3 Independent variables

Independent variable
Pectin
Clove essential oil
Ajwain essential oil

3.2.3 selection of dependent variables

The variable that is examined or observed in an experiment or study to determine the impact of changes in the independent variable is known as the dependent variable. Experimental research is centered on the relationship between the independent and dependent variables. To see how the independent variable affects or causes changes in the dependent variable, researchers modified the independent variable. The values of the dependent variable depend on the modifications made to the independent variable. Fruits and vegetables during storage change physically and chemically. In this study fresh cut pineapple is coated with pectin based nanoemulsion coating. Fruits have their own moisture content, pH range, total soluble solids, titratable acidity, color, phenolic activity, antioxidant activity and pineapple is also rich in vitamin C. The above-mentioned physicochemical properties along with antimicrobial effect of nanoemulsion coating are set as dependent variables for this study.

Table 3.4 Dependent variables (responses)

1. Responses for fresh cut pineapples	2. Response for nanoemulsion coating
a. Moisture	a. Antimicrobial activity
b. pH	
c. Color	
d. Total soluble solids	
e. Titratable acidity	
f. Vitamin C	
g. Total phenol	
h. antioxidant content	

3.3 sample preparation

Pineapple was washed to remove dirt and dust. The fruit crown was cut first, then the fruit was cleaned with distilled water and was dried with the help of a paper towel. The fruit was then peeled and cut into fan shapes of even length and width. The eye of the pineapple was also removed. All utensils included in handling and cutting of fruit were also cleaned and washed.



Figure 3.14 flow chart of Sample preparation

3.3.1 Preparation of pectin coating solution

Pectin coating solution of 100 ml was prepared by dissolving pectin powder with glycerol in distilled water and was continuously mixed with the help of magnetic stirrer at 70-100 rpm until it gets homogenized. The solution was then allowed to cool down and was mixed with nanoemulsion to form a final coating.

Pectin (2gm) + glycerol (0.6gm) + distilled water (98 ml)

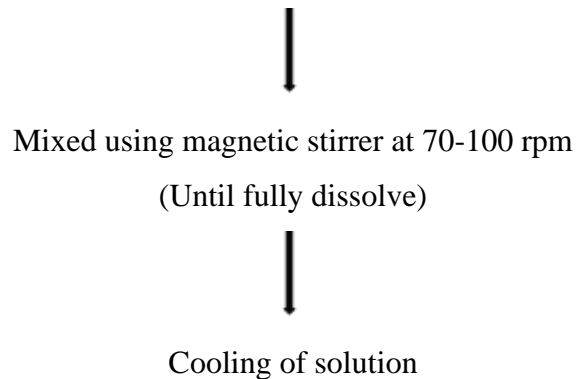


Figure 3.15 flow chart of Pectin coating solution

3.3.2 Preparation of nanoemulsion

45 ml distilled water with 5% clove and ajwain essential oil each and 6% tween 80 were mixed together and kept on magnetic stirrer was uniform solution to form a nanoemulsion of 50 ml. The solution was then kept in ultrasonicator for particle size reduction. This results in white milky and opaque nanoemulsion solution.

Distilled water + clove essential oil + ajwain essential oil + Tween 80

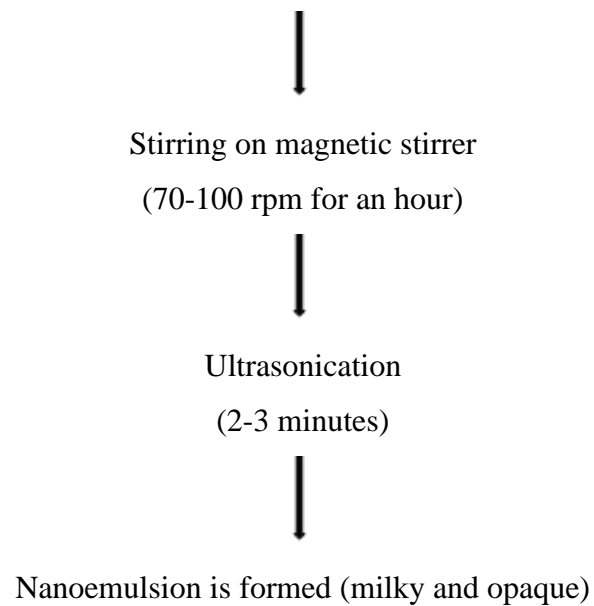


Figure 3.16 flowchart of nanoemulsion preparation

3.3.3 Formation of edible coating

The final coating was prepared by mixing all the ingredients of pectin coating solution and nanoemulsion of ajwain and clove essential oil together to form the final edible coating for fresh cut pineapple pieces.

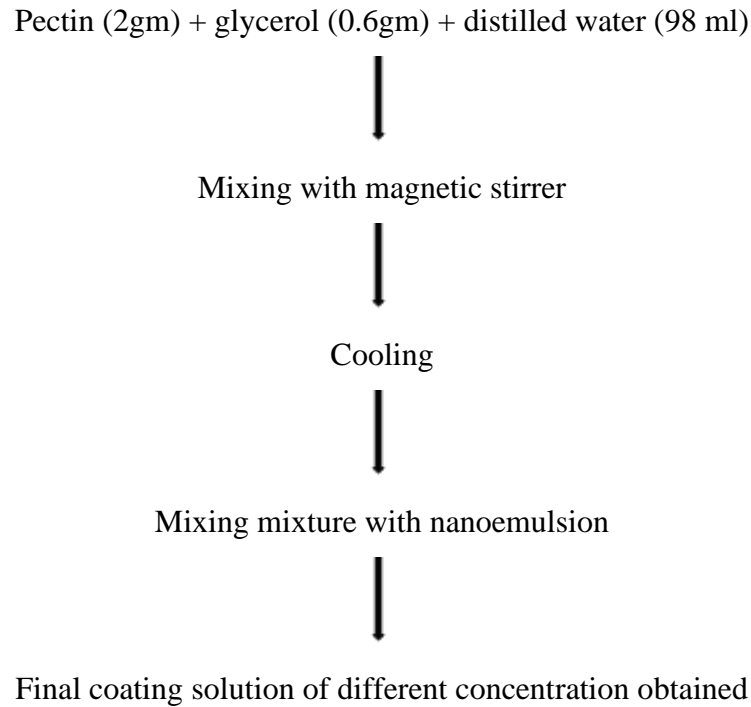


Figure 3.17 flow chart of final edible coating

Table 3.5 composition of edible coating solution formed.

Raw material	Control 1 (%)	Control 2 (%)	Sample 3 (%)	Sample 4 (%)	Sample 5 (%)
Pectin	0	2	2	2	2
Clove essential oil	0	0	2.5	0	1.5
Ajwain essential oil	0	0	0	2.5	1.5

3.4 Method of coating

Clean fresh cut pineapple pieces were coated with edible coating using dipping method to ensure uniform coating on whole piece. Pineapple pieces were dipped in solution one by one for approximately two minutes then it was taken out so that extra coating can be drip off from the piece. The fruit piece was properly air dried and kept in container, similarly all the pieces were coated and kept in different containers labelled as C1, C2, S3, S4 and S5, where C1 is an uncoated control sample. These samples were stored in refrigerator and different sets of experiments were designed to test the effect of different concentration of nanoemulsion on quality of fresh cut pineapples. Analysis like moisture, pH, titratable acidity, total soluble solids, antioxidant content, total phenolic content, vitamin c, and antimicrobial activity were performed.



Fig. 3.18 Selection of raw material



Fig. 3.19 preparation of pectin coating solution



Fig. 3.20 Preparation of nanoemulsion



Fig. 3.21 Preparation and application of final nanoemulsion coating on fresh cut pineapples

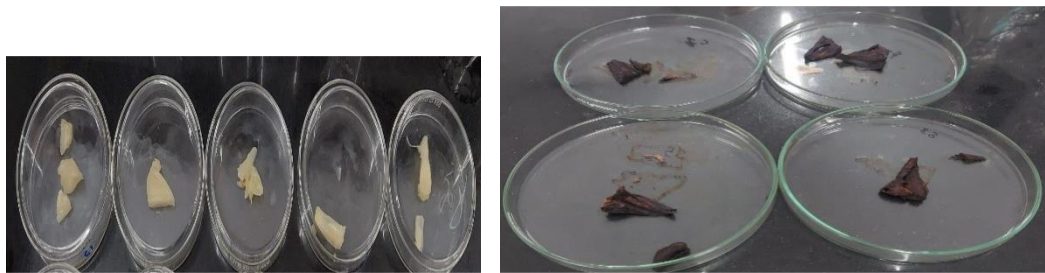


Fig. 3.22 Moisture content



Fig. 3.23 pH determination analysis



Fig. 3.24 TSS analysis



Fig. 3.25 TA

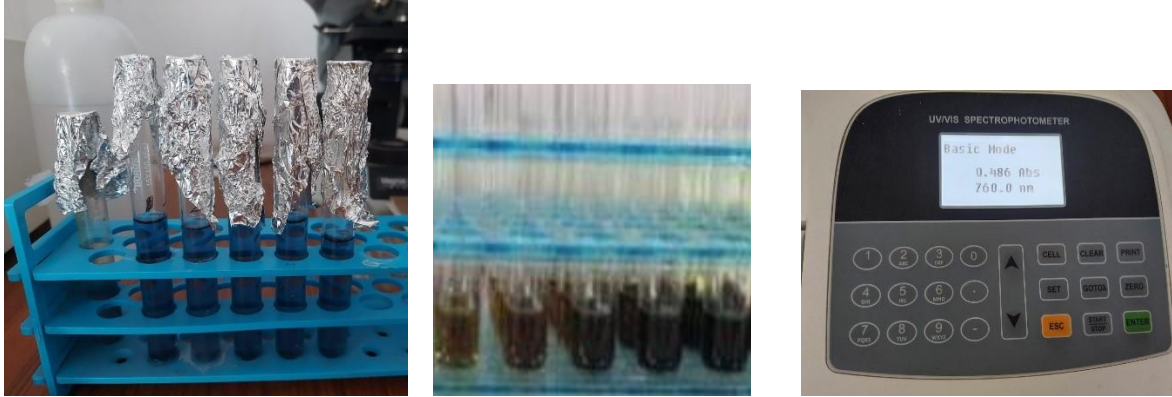


Fig. 3.26 Total phenolic content and Antioxidant analysis

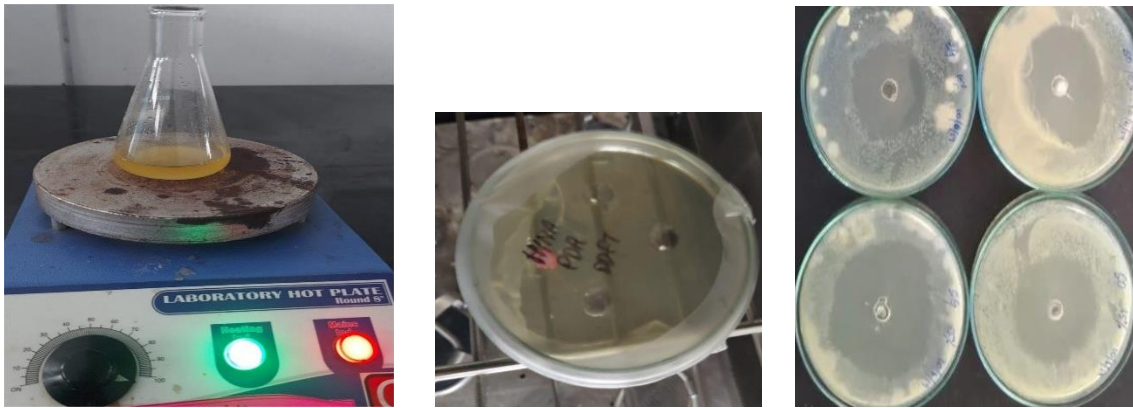


Fig. 3.27 10 Antimicrobial analysis

3.5 Physicochemical analysis

3.5.1 Moisture content

The moisture content of sample was determined using a hot air oven. A small amount of sample was taken and weighed between the range of 3-5 grams by using electrical weighing balance. The weighed sample was then placed on clean and sanitized petri-dishes/plates and kept in hot air oven at 135°C for 2 hrs. The determination of moisture content is shown in plate 5. Moisture content of each sample was calculated using the formula given below:

$$\text{Moisture content (\%)} = \frac{W_1 - W_2}{W_1} \cdot 100$$

Where,

W1 = weight of sample, and

W2= weight of sample after drying.

3.5.2 Determination of pH

For the determination of pH a total of 5 grams sample of pineapple was crushed to form pulp and added to 50 ml distilled water at room temperature. The mixture was homogenized with the help of a homogenizer and kept resting for 10 minutes in a beaker. The electrodes were dipped in each beaker to measure the pH of each sample. The electrode was cleaned by distilled water each time before dipping in beakers. The pH determination of samples was shown in plate 6.

3.5.3 Titratable acidity

The titratable acidity of all five samples were calculated using 0.1 sodium hydroxide solution and phenolphthalein indicator. A cut slice of sample was crushed to form juice in titration flask, then add distilled water and few drops of phenolphthalein indicator to it. Titrate the sample with NaOH to faint pink color using burette or pipette. Note the titre value and calculate the percentage titratable acidity with the formula given below. Plate 8 shows the titratable acidity analysis.

% Acidity =

$$\frac{\text{titre} \cdot \text{normality of alkali} \cdot \text{volume made} \cdot \text{equivalent weight of acid} \cdot 100}{\text{wt. of sample} \cdot \text{volume of aliquot} \cdot 1000}$$

3.5.4 Total soluble solids (TSS)

Total soluble solid is measured with the help of refractometer. The analysis of TSS is shown in plate 7. 5 grams of sample is diluted with distilled water. A small drop of sample was taken with the help of dropper and kept on the meter. The meter was gently closed to avoid air bubbles. The reading is shown on the meter by itself.

3.5.5 Color analysis

The color of pineapple at each interval of days were observed using CIE Lab system with the help of digital colorimeter. It expresses color as three values: L* for perceptual lightness that ranges from 0 to 100, a* ranges from green (-) to red (+) and b* ranges from blue (-) to yellow (+). The color analysis was carried out at different intervals of days, and it is shown in plate 11.

3.5.6 Vitamin c content

The vitamin C concentration was determined using the AOAC standard method for Vitamin C-Ascorbic Acid in vitamin supplements and juices (AOAC, 2005). Approximately 20 g of pineapple juice were used to titrate the iodine standard solution for this test. By using the volume titrated (mg of ascorbic acid divided by sample in g in wet basis), the vitamin C content of the sample was calculated.

0.5g starch was added to 50 ml distilled water to prepare starch solution of 1%. 5 gm potassium iodide and 0.268 gm potassium iodate were added to 200 ml distilled water. 30 ml 3M sulfuric acid was added in beaker and then added to distill water until 500 ml solution was prepared. 0.250 g of ascorbic acid and 100 ml of demineralized-distilled water were dissolved in the beaker to form the vitamin C standard solution. The mixture was poured into a 250 ml volumetric flask and diluted with distilled water to make 250 ml. By pipetting 25 ml of vitamin C solution into a 125 ml flask, the iodine solution was standardized with the vitamin C standard

solution. When blue-black color was seen, 10 drops of the 1% starch solution were added and titrated against the iodine solution. The titrations were carried out three times. Each fruit sample's volume was measured, and the ascorbic acid content per 100 ml of fruit was estimated using formula given below:

Concentration of ascorbic acid used in mg/100ml =

$$\frac{\text{concentration}(mg/ml)}{\text{standard weight of sample in gram}} \times 1000$$

3.5.7 Total phenolic content

The total phenolic content of all samples were calculated quantitatively by using the Folin Ciocalteu reagent, with Gallic acid as the acid standard. The TPC determined as the gallic acid equivalent is directly proportional to the absorbance. Phenolic components are very important plant constituents that have redox properties which are responsible for antioxidant character. 1mg/ml methanol sample is prepared. Folin Ciocalteu reagent and sodium bicarbonate is added to the sample kept in test tube this results in blue color complex which is incubated for 2 hours in dark. The absorbance is then noted at 765nm. Plate 9 shows the total phenolic content determination.

3.5.8 Antioxidant content

The antioxidant content of five samples were determined by DPPH scavenging method. The DPPH is frequently used to refer is an organic substance with formula 2,2-diphenyl-1-picrylhydrazyl. It is in the form of a powder in crystalline form with a black color and made of free radical particles that are stable. It is employed to assess the capacity of plant components to scavenge and destroy free radicals. This method is completely based on spectrophotometer analysis of the change and alteration in DPPH concentration that are brought due to the interaction with an antioxidant. 1 mg/ml methanolic extract is prepared further DPPH and methanol are added to extract, the sample is kept in dark for 30 mins and absorbance is checked at 540nm.

3.5.9 Antimicrobial activity

The antibacterial and antifungal activity was analyzed using agar well diffusion method. The bacterial and fungal strains were revived using nutrient broth and potato dextrose broth for bacteria and fungi respectively. Then it was kept in shaking incubator at 37°C for 24 hrs. Afterwards, nutrient agar media and potato dextrose agar media was formed and solidified on petri dishes. Bacterial and fungal suspensions were spread evenly on each plate. The wells of about 8.00mm (about 0.31 in) were made and were filled with nanoemulsion sample, an antibiotic/antifungal, and DMSO and were kept in incubator for 24 hrs. at 27°C. The Zone of inhibition (ZoI) was measured using electronic digital caliper.

3.5.10 Sensory evaluation

sensory evaluation was carried out on first, fourth, eighth and twelfth day of the storage period, a panel of 25 members were chosen in the sensory evaluation of pineapple fruit, they evaluated samples of freshly cut pineapple on a nine-point hedonic scale. The panelists were asked to rate the fresh cut fruit samples on the basis of color, texture, taste, aroma and general appeal. the order for hedonic scale was 1: strongly dislike, 5: neither like nor dislike, and 9: strongly like.

3.5.11 Fourier transform infrared (FTIR)

The investigation of coating systems can easily be done using the potent analytical method known as FTIR. The spectrum of FTIR offers a plethora of data about the fundamental functional chemical groups in a sample that can be utilized for identification and evaluation of chemical changes brought on by exposure to the environment. FTIR spectroscopy is an analytical method that stimulates molecular movements using infrared radiation from the electromagnetic spectrum. The energy needed to start intramolecular oscillations correlates with the wavelength of absorption. Stretching, rocking, wagging, twisting, and bending are common oscillations in molecules, and the FTIR spectrometer records these motions as "peaks" or "absorbances." These movements are specifically related to the oscillations of the functional group and are found in the same location of infrared spectrum.

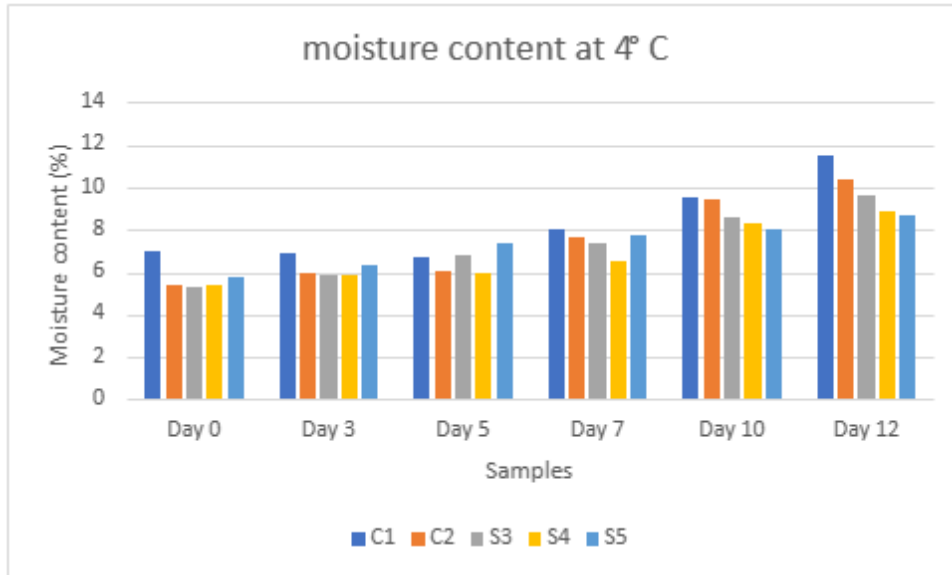
CHAPTER 4

RESULT AND DISCUSSION

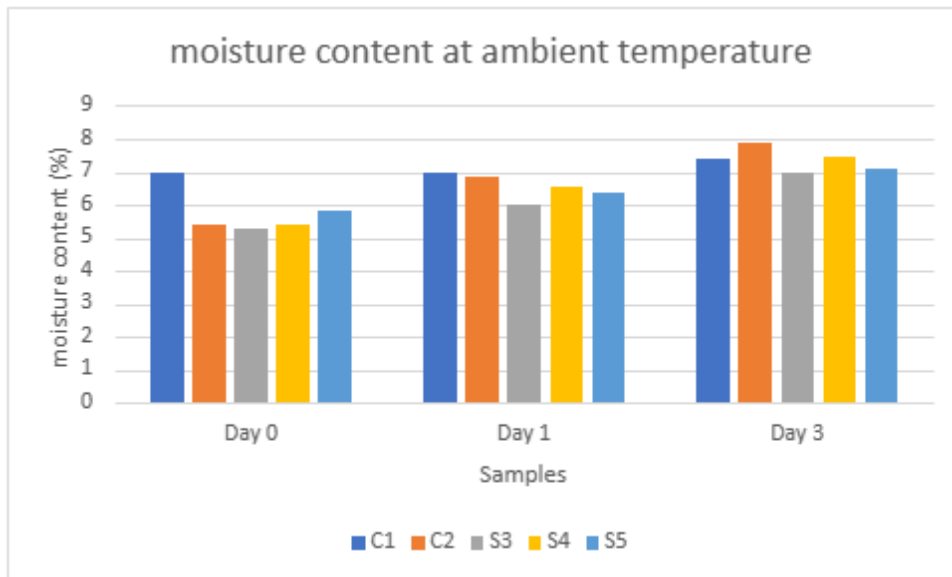
Fresh cut pineapple is a rich source of ascorbic acid, phenols, and antioxidants. They also contain plenty of water. Extending the shelf life of pineapple using pectin as a base coating material because pectin is a natural vegetarian polysaccharide occurring in fruits. Clove and ajwain essential oil are also rich in antioxidants and show very good antimicrobial activity. Hence, they are also used as therapeutic drugs for treatment of many diseases. A total of five samples were studied in this research work on the basis of pectin and nanoemulsion concentration of clove and ajwain essential oil and their combination. The shelf life and physicochemical analysis of these five samples was conducted to evaluate the optimized sample. The physicochemical analysis performed were moisture content, determination of pH, titratable acidity, total soluble solids, color analysis, vitamin c content, total phenolic content, antioxidant activity and antimicrobial activity. The characterization of clove essential oil with tween80 nanoemulsion of different concentration was also performed by means of Fourier transform infrared (FTIR).

4.1 Moisture content

The weight of the water contained in a particular object or material is referred to as the moisture content (or water content). The most common way to express it is as a percentage of weight. It is determined as weight loss on drying of food product. The moisture content of all samples prepared in the laboratory are given below in figure 4.1. The moisture content of control sample was initially 7.00% while at the end of storage, the loss moisture increases to 11.52%. sample 5 expect all other sample shows least moisture loss by the end of storage at 4° C that is 8.7% due to presence of clove and ajwain essential oil and pectin as a coating material. Moisture loss of coated samples were less than uncoated sample, though there was an increase in moisture loss in all samples, but the control sample shows changes at higher level. Similar results were obtained in study by Liao et al (2023) where fresh sliced pineapples were treated with edible coating and modified atmospheric packaging.



a)

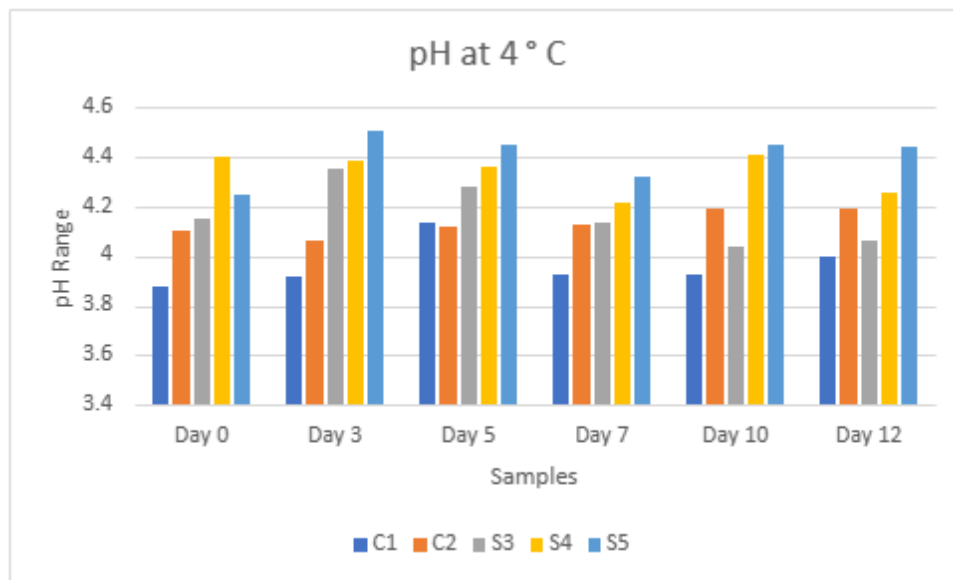


b)

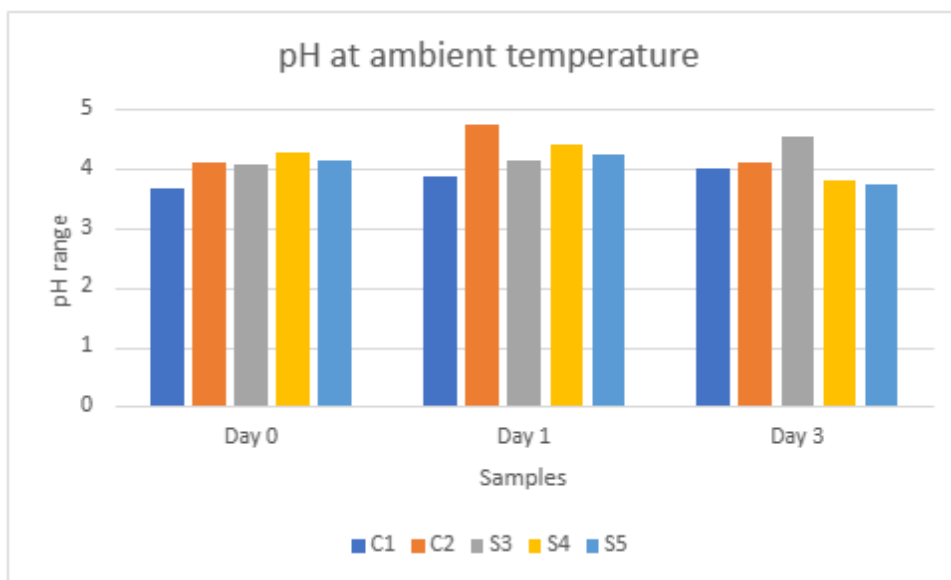
Figure 4.1 a) & b) moisture content in fresh cut pineapple sample at 4°C and at ambient temperature

4.2 pH content

pH is a scale used to describe how basic or how acidic an aqueous solution is. Lower pH values are determined for acidic solutions than for basic or alkaline solutions (solutions with larger quantities of hydrogen (H⁺) ions). It has a range of 0 to 14, with 0 to 6 being acidic, 8 to 14 representing basic, and 7 indicating neutral. Because pineapples are so acidic, this is the case they normally fall between the range of 3 to 4.5 on pH scale. During study of coated and uncoated fresh cut pineapple, the pH range was between 3-4.5. The non-coated sample were having low pH during storage compared to coated sample. Control 1 sample had pH of 3.88 at the beginning which increased to 4.00 at 12th day of storage at 4°C. Coated sample shows more pH because of addition of pectin and nanoemulsion of clove and ajwain essential oil. During the whole study it has been seen that pH slightly increases but by the end of storage it declines. There is least effect of pH on both coated and uncoated samples. Prakash et al.,(2020) who studied the effect of citral nanoemulsion on fresh cut pineapple also found similar results.



a)

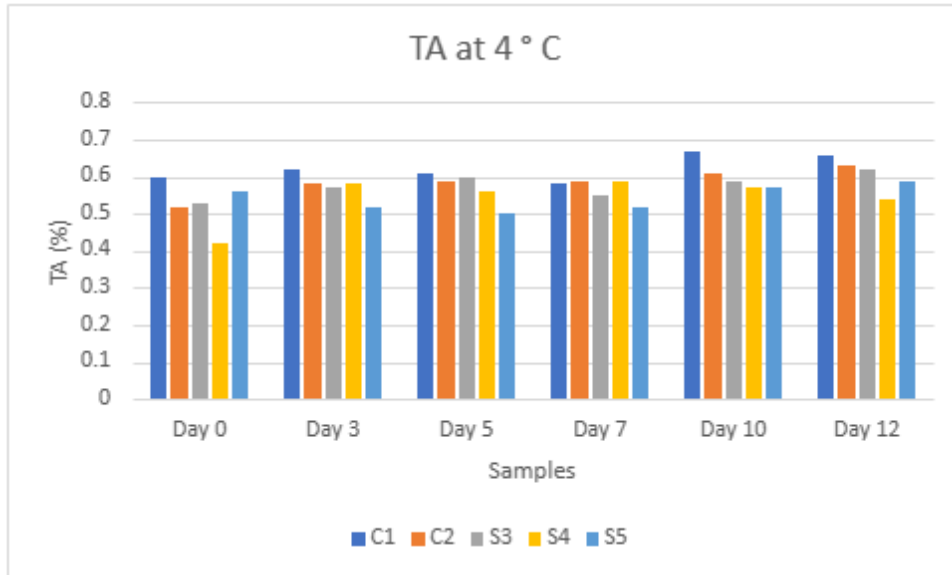


b)

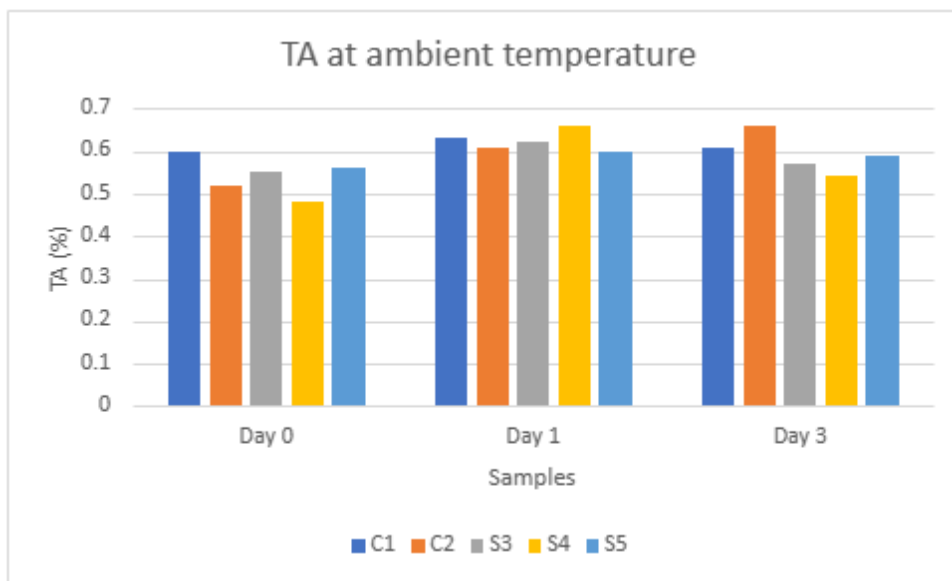
Figure 4.2 a) & b) pH values of fresh cut pineapples at 4°C and at ambient temperature

4.3 Titratable acidity

The overall titratable acidity of the sliced pineapples, which varied amongst the treatments during the storage period and ranged from 0.42 to 0.66, was also barely impacted by the different nanoemulsions used in the edible coatings. Figure 4.3 shows the value of titratable acidity (TA) at first there was a slight change, TA shows significant increase at 12th day of storage. Uncoated and control samples show higher increase in TA at the end of storage (0.66%) than coated sample (0.59%). The TA decreases because of the transformation of acids during the fruit ripening process. This study shows no significant effect on the TA and same observations were made in other edible coating research (Prakash et al.,2020).



a)

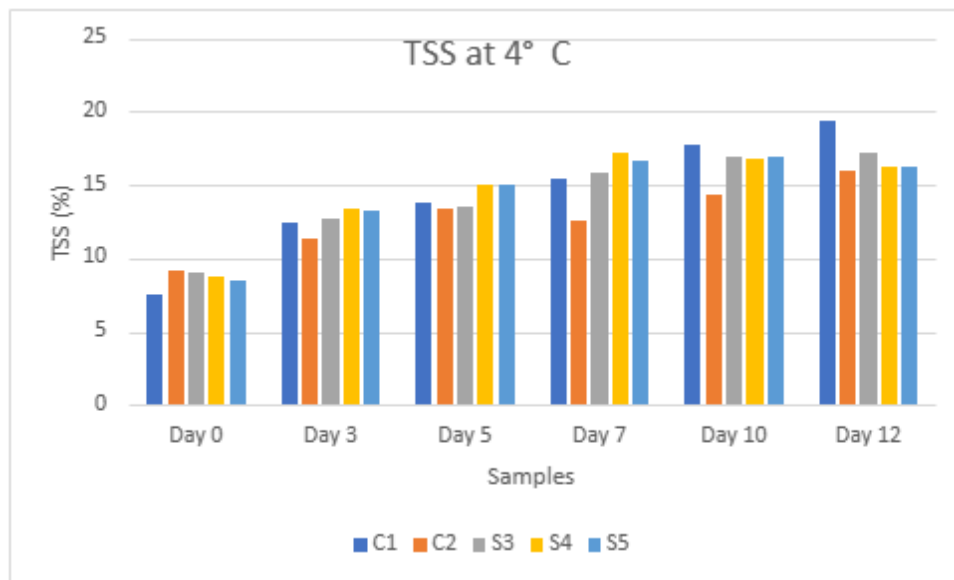


b)

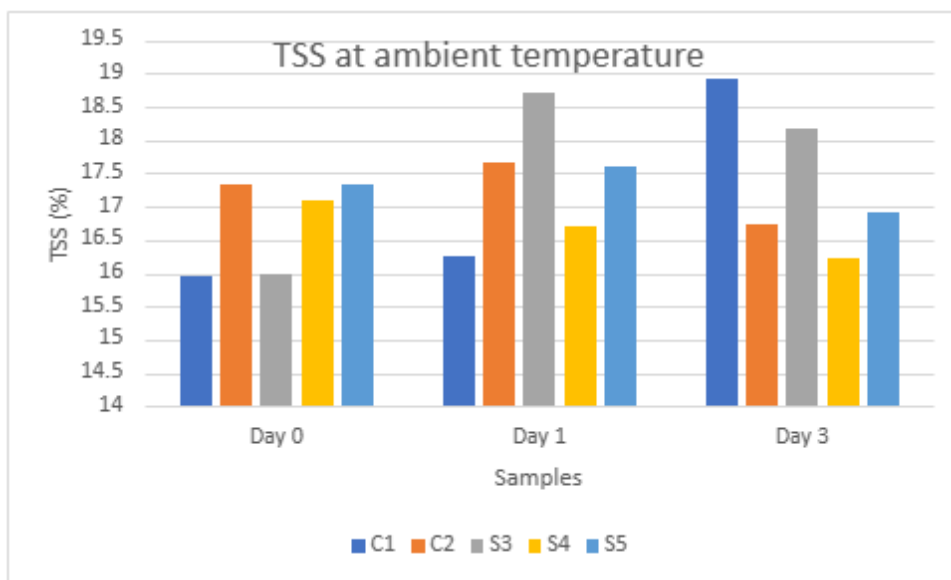
Figure 4.3 a) & b) Titratable acidity of fresh cut pineapples at 4°C and ambient temperature

4.4 Total soluble solids (TSS)

The carbohydrates, proteins, lipids, minerals, organic acids in fruits are all measured by the TSS, or sugar content. It makes up between 10-20 percent of the fresh weight of fruits and it rises as the fruit ripens to produce a sweeter and less acidic fruit. In the case of pure sucrose solutions, brix is a measurement of total soluble solids (TSS). Brix is a valuable indicator of TSS or sugar concentration since fruit juices often contain more sugar than other soluble elements. In this study all five samples show the TSS between 7.75-20% (figure 4.4). The TSS of all samples (coated and uncoated) increases during storage and makes the fruit unacceptable for consumption. The uncoated sample shows a higher value of TSS during storage but there was no big difference in all samples. Thus, the application of nanoemulsion does not control the TSS of pineapple. The TSS of sample generally increases as fruit undergoes ripening. Hence, the following data is considered to be similar with the study of citral nanoemulsion on fresh cut pineapple (Prakash et al., 2020).



a)



b)

Figure 4.4 a) & b) TSS of fresh cut pineapple during storage at 4°C and ambient temperature

4.5 Color analysis

The main notable change in fresh-cut fruit that occurs during storage and has a direct impact on how buyers perceive its quality is color change. Table 4.5 illustrates how the L* a* and b* values of the color attributes of freshly cut pineapple fruit were impacted by nanoemulsion edible coatings. The results showed that after 12 days of storage, the L* and b* values in all five samples began to significantly decline. The L* and b* values decline significantly at a higher rate in all samples except sample 5. This shows that sample 5 having both ajwain and clove essential oil nanoemulsion with pectin as well is successful in retaining the color quality of fresh cut pineapples during storage as it shows the lowest reduction in L* and b* value that is 60.54 and 50.78 respectively because of its antioxidant properties. These trends were also found in a study by Rasool et al., (2023) where fresh cut pineapple was treated with natural almond gum edible coating.

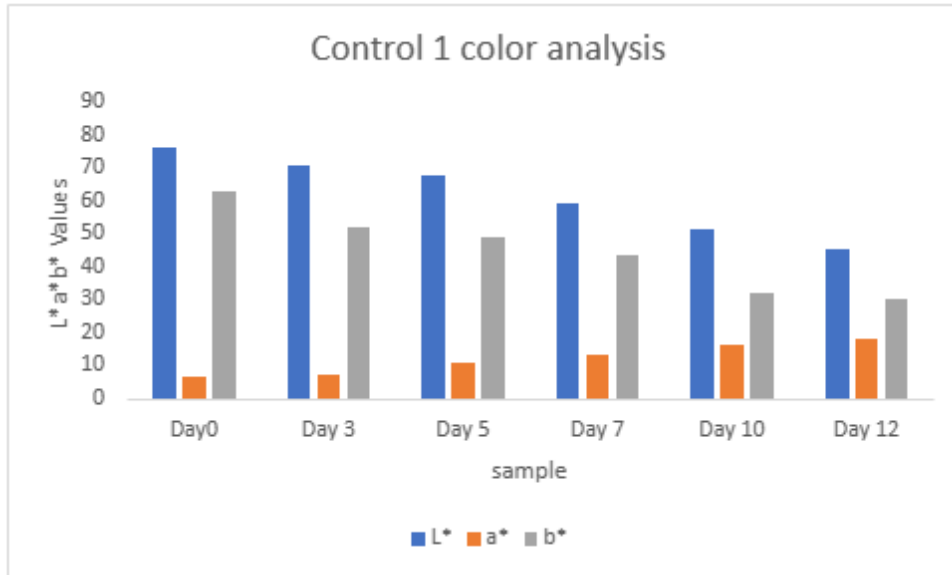


Figure 4.5 a color analysis of control 1

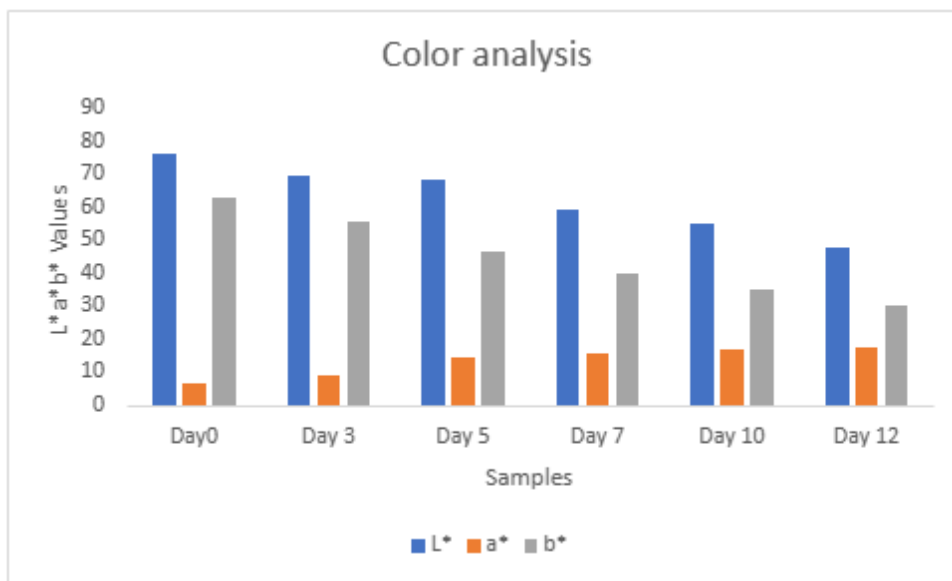


Figure 4.5 b color analysis of control 2

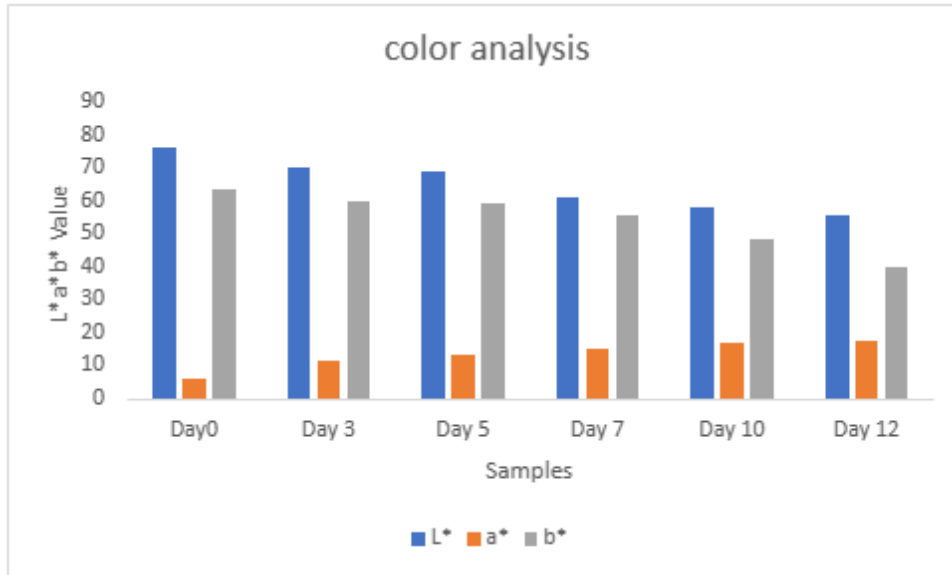


Figure 4.5 c color analysis of Sample 3

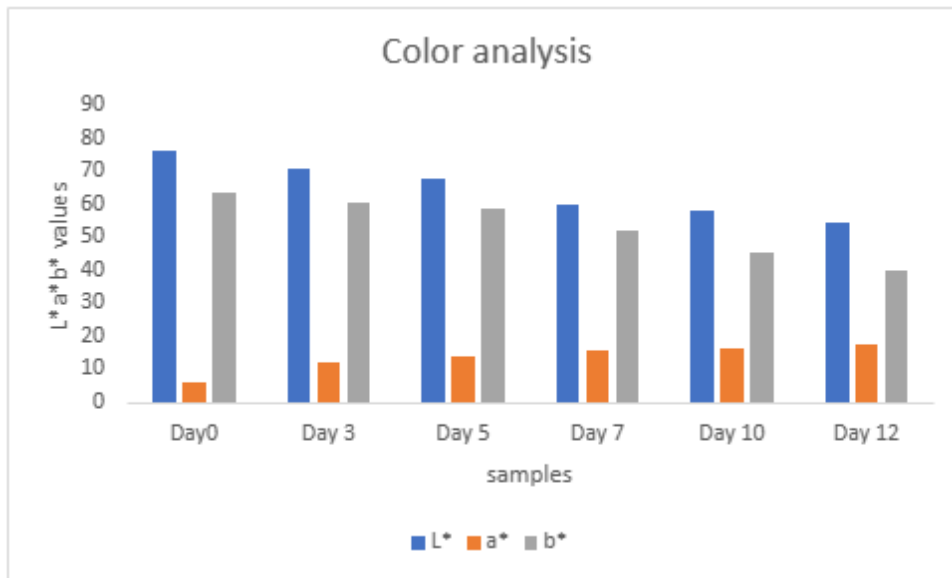


Figure 4.5 d color analysis of sample 4

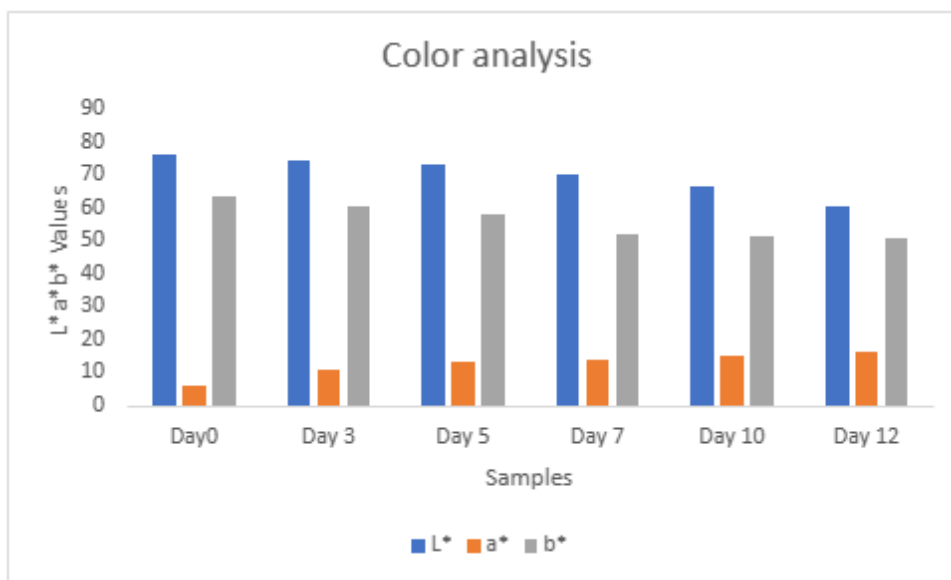
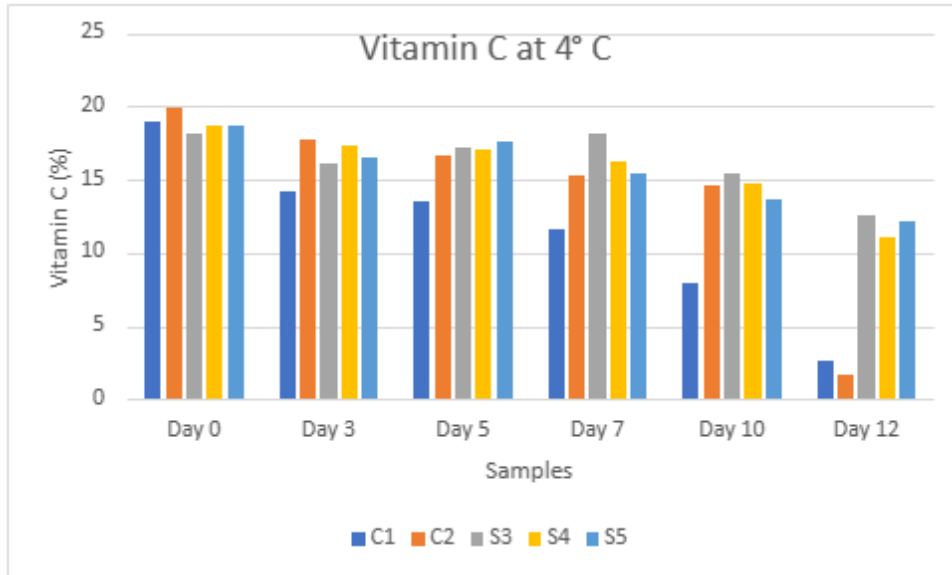


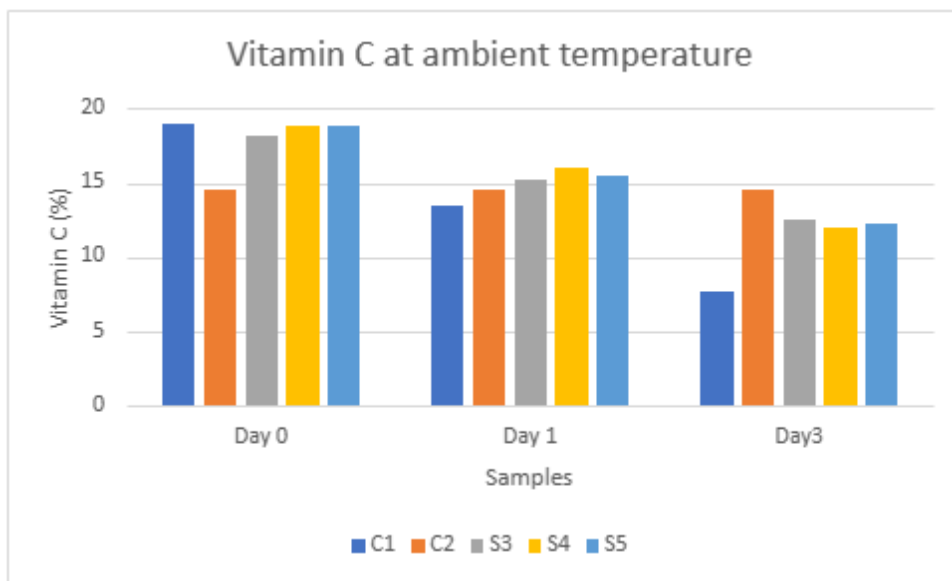
Figure 4.5 e color analysis of sample 5

4.6 vitamin C (Ascorbic acid)

Changes in vitamin C is shown in Figure 4.6 shows the ascorbic acid level of fresh-cut pineapple after 12 days storage at 4 °C. Vitamin C content reduces throughout the storage period; the drop in vitamin C of control sample was more than that of coated sample, and the lowest drop value was discovered in the sample 5 (Pectin+ CEO + AEO) which were followed by the sample 4 (Pectin + CEO) and sample 3 (Pectin+ AEO). This might be the result of coatings having low permeability, which might stop the disintegration of ascorbic acid. Similar observations were made in research on citral nanoemulsion coating on fresh cut pineapple (Prakash et al.,2020).Operations used in processing can result in the loss of vitamin C, which over time may cause a greater loss of weight and contribute to the occurrence of oxidative reactions, which may darken the product, cause endogenous pigments to become discolored, lose flavour, aroma, or smell, change the texture, or result in nutritional loss.



a)

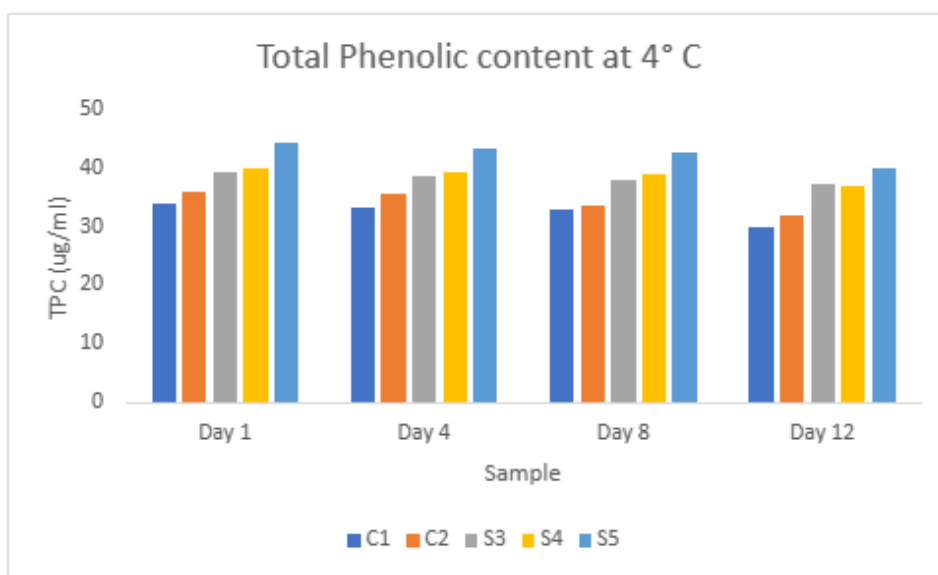


b)

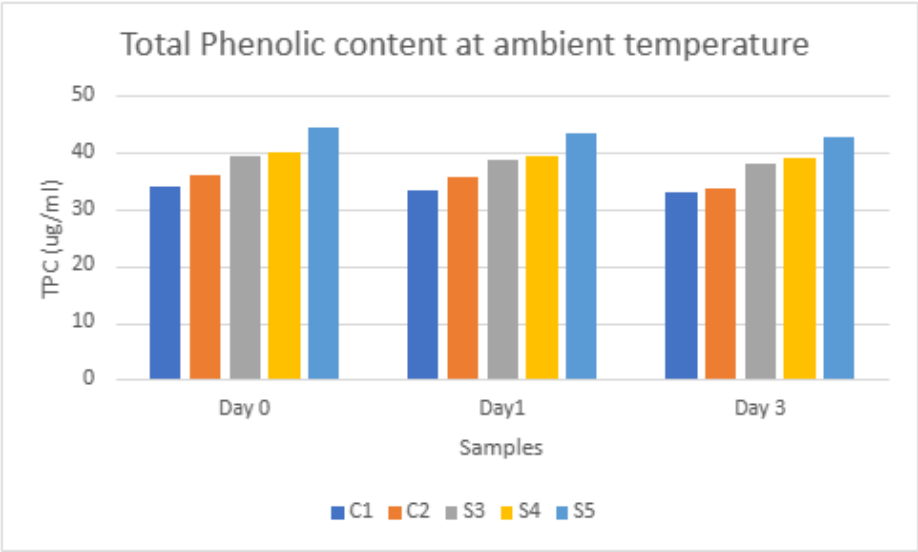
Figure 4.6 a) & b) vitamin c content of fresh cut pineapple at 4°C and at ambient temperature

4.7 Total phenolic content

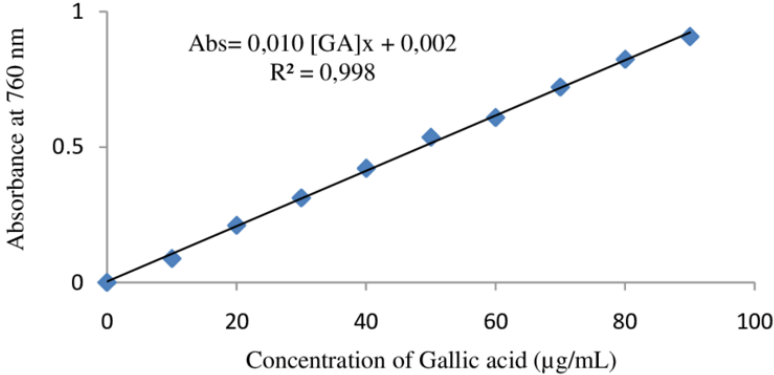
Phenolic compounds have antioxidant characteristics, they can assist the body in scavenging dangerous free radicals that can cause oxidative stress and a number of chronic illnesses, including cancer and cardiovascular ailments. To ascertain a sample's total phenolic content, many techniques are used. The Folin-Ciocalteu assay is one of the frequently employed techniques. In this procedure, the sample is combined with a substance known as the Folin-Ciocalteu reagent. A blue color complex is created when the phenolic chemicals in the sample and the reagent react, and this complex can be detected spectrophotometrically. The amount of phenolic chemicals present in the sample directly correlates with the color's intensity. The sample absorbance was taken at 760nm. All five samples were analyzed for TPC, and total phenolic activity ranged between 33.4 - 43.5 mg/ml where, sample 5 have the highest TPC value due to the presence of both CEO and AEO as they contain antioxidant resulting in high total phenolics, sample 3 and sample 4 have barely minimum difference in TPC value and control sample 1 have lowest TPC value. Plate 9 a) shows the TPC of samples. The TPC of all sample degrade at end of storage due to enhance oxidative stress .it was seen earlier in a study done by Liao et al (2023) to check the effect of modified storage packaging and edible coating on pineapple slices.



a)



b)



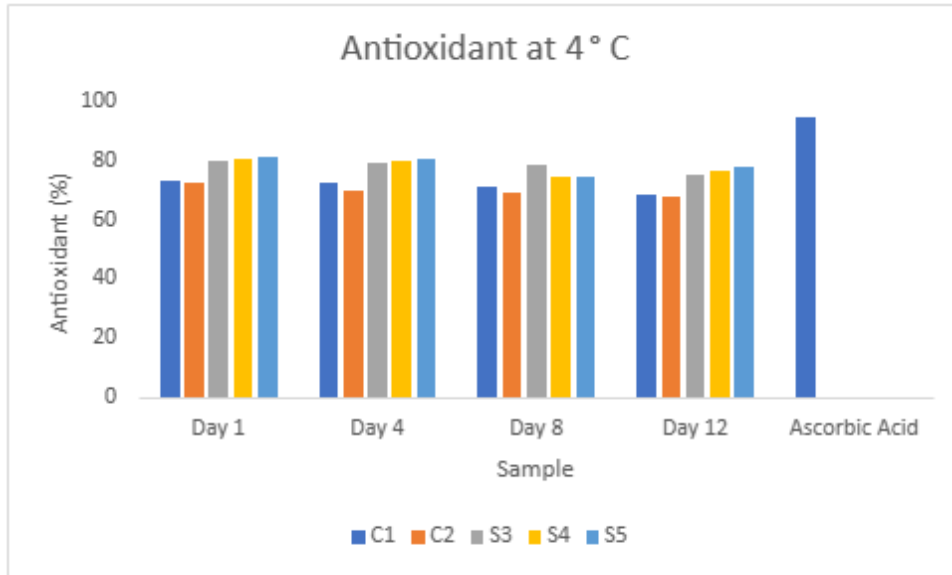
c)

Figure 4.7 a), b) & c) Total phenolic content

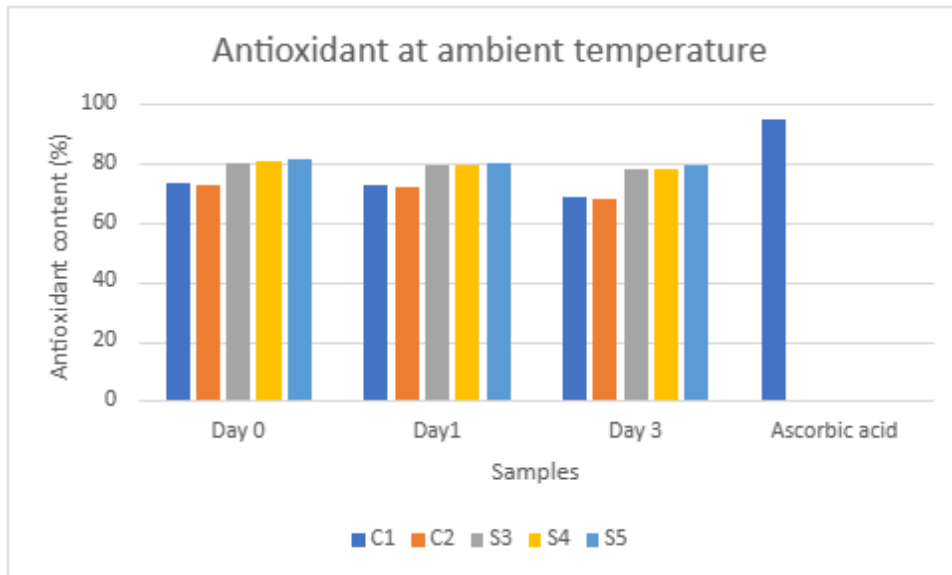
4.8 Antioxidant activity

Freshly cut fruits and vegetables lose some of their overall antioxidant activity due to oxidative stress brought on by cutting, which also damages membranes and affects the content and concentration of bioactive chemicals, primarily phenolics and vitamin C.

The DPPH (2,2-diphenyl-1-picrylhydrazyl) assay, a popular technique for evaluating a substance's capacity to scavenge free radicals and counteract oxidative stress, can be used to measure antioxidant activity. The DPPH assay is based on the idea that when a free radical combines with an antioxidant, it loses some of its stability and turns a deep purple color. A change from purple to yellow as a result of this reduction can be seen using spectrophotometry. The antioxidant activity of coated and uncoated sample was analyzed, coated sample exhibit better antioxidant activity than uncoated sample due to the presence of CEO and AEO in the form of nanoemulsion. The antioxidant activity ranges from 70 – 80% in samples. Sample 4 and sample 5 show 80.31% and 80.84% antioxidant activity due to the presence of essential oils which have good antioxidant property, while sample 2 shows lowest antioxidant activity of 70.35%. Cutting causes oxidative stress, which damages membranes and affects the concentration and composition of bioactive chemicals, primarily phenolics and vitamin C, changing the level of the overall antioxidant. in this study on freshly cut pineapple fruit, antioxidant activity increased during the first few days of storage but gradually decreases over time. The higher stress level in the tissue caused by fruit cutting is shown by the bigger increase in antioxidant activity. In order to counteract the effects of minimal processing, effective antioxidants may be lost as storage duration increases as a result of their interactions with free radicals (Sharma & rao.,2018).



a)



b)

Figure 4.8 a) & b) Antioxidant content

4.9 Antimicrobial activity

Table 4.9 displays the outcomes of the CEO+AEO nanoemulsion antibacterial activity. It is obvious that the prepared nanoemulsion (NE) was effective against the investigated bacteria and fungus. Gram negative *A. faecalis* and *E. coli* had inhibition zones with diameters of 28.47 mm and 27.56 mm, respectively, while Gram positive *B. subtilis* and *S. aureus* had inhibition zones with diameters of 17.92 mm and 15.01 mm, respectively. The dissolution of the outer membrane by NE in Gram negative bacteria, which allows for easy penetration of the peptidoglycan layer that is much thicker in Gram positive bacteria, is the reason why our results show higher antimicrobial activity against Gram negative bacteria than gram positive bacteria for the same concentrations. The NE prepared demonstrated effective antifungal action against strains of fungi, with inhibition zone dimensions on *Aspergillus niger* and *Aspergillus flavus* was 29 mm and 28.7 mm, respectively. The results were similar to the antimicrobial effect analyzed by Basumatary et al., (2022) who find an effective chitosan coating for preservation of pineapple.

Table 4.1 Antibacterial and antifungal analysis of NE

Bacterial strain	Inhibition zone (mm)
Staphylococcus aureus	15.01
Bacillus subtilis	17.92
Escherichia coli	28.47
Alcaligenes faecalis	27.56
Fungal strain	Inhibition zone (mm)
Aspergillus niger	29
Aspergillus flavus	28.7

4.10 Sensory evaluation

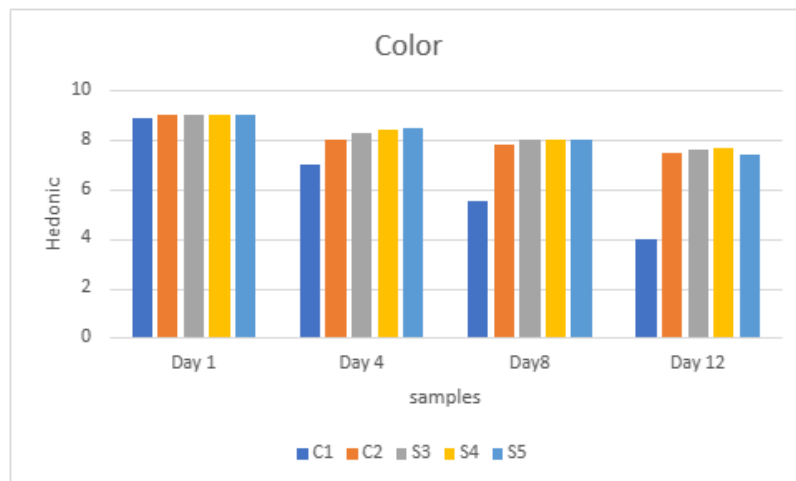
The consumption of fresh cut fruits with edible coating has increased and it is important to evaluate the sensory aspect of these coated fruits as they contain different components which affect the aroma and taste of the fruit. The sensory analysis was done on the basis of color,

texture, taste, aroma and overall acceptance on the nine-point hedonic scale within 12 days of storage at difference of 4 days i.e., on first, fourth, eight and twelfth day.

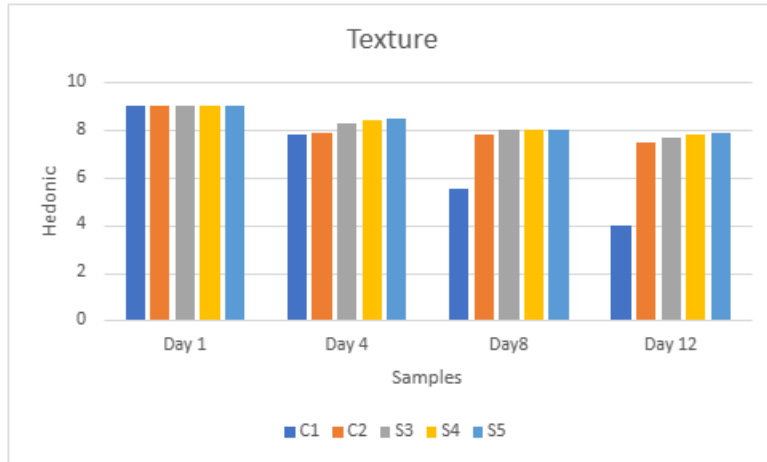
The rating for color of C1 decreases from 8.5 to 4 at end 12th day, while S5 ranged between 8.5 to 7.8 until the end of storage. Similarly, the texture score of C1 was seen decreasing from 8.6 to 3.9 due to loss of moisture and during storage, while other coated sample ranges between 8.5 to 7.6. Hence S5 shows best range from 8.5 on the 1st day to 7.9 at 12th day of storage.

The taste score of samples were evaluated and due to loss of nutrients and water the C1 sample shows lowest score at the end of storage of 3.9 while S5 due to presence of CEO and AEO shows good taste as it acts as barrier to moisture losses. The aroma of all coated samples was more than uncoated sample due to presence of aromatic compound in edible coating. Clove and ajwain have strong aroma. C1 on the 1st day was 8.7 and goes down to 3.5 on the 12th day. While other coated samples range between 7-7.9 at the end of storage period.

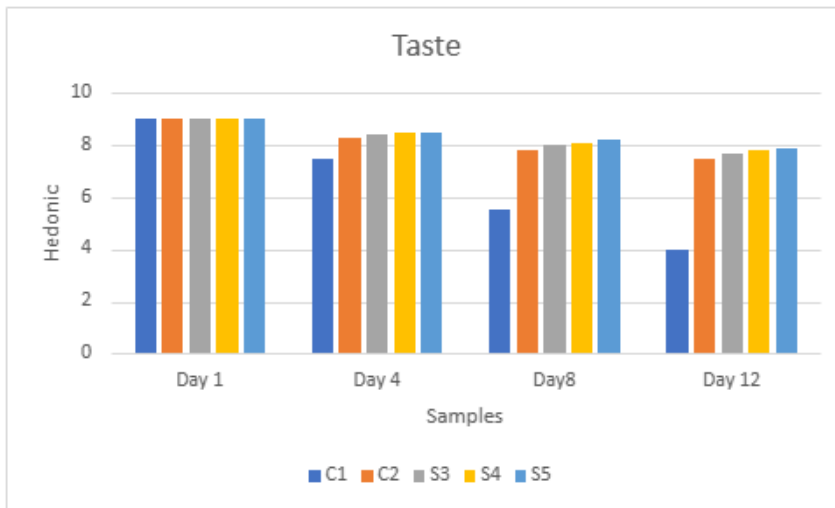
The overall acceptability of all samples was the same on day 1 but it decreases till day 12. C1 shows more difference in overall acceptance than other samples. The overall acceptability of S5 was highest i.e 8 than other samples (Prakash et al.,2020).



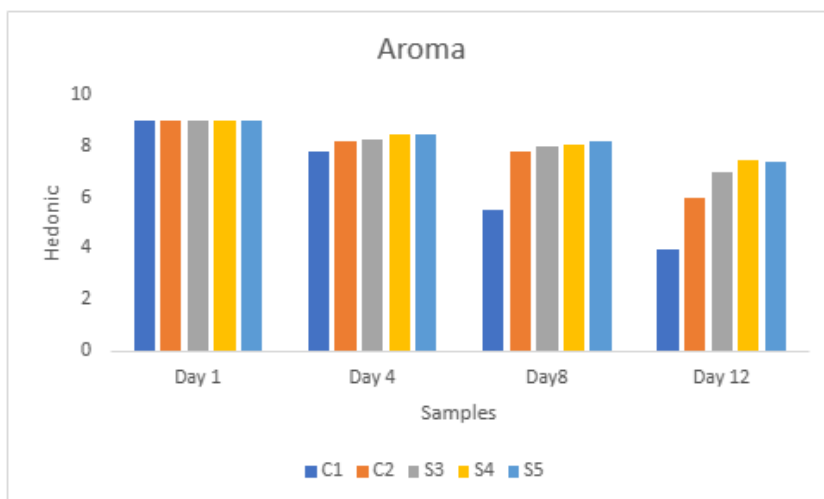
a)



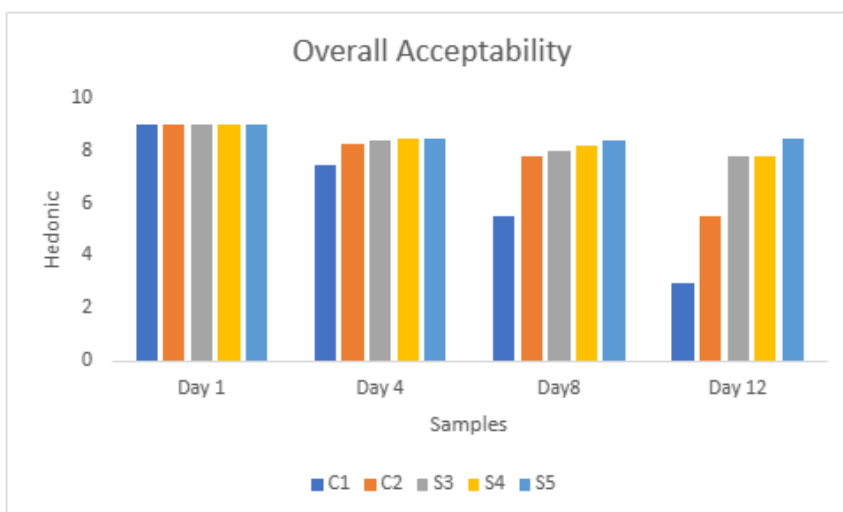
b)



c)



d)



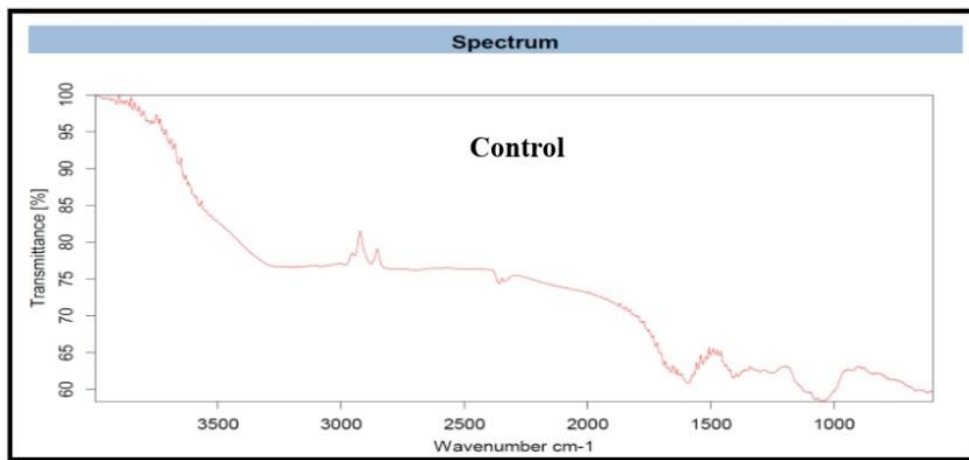
e)

Figure 4.9 a) b) c) d) & e) Sensory evaluation

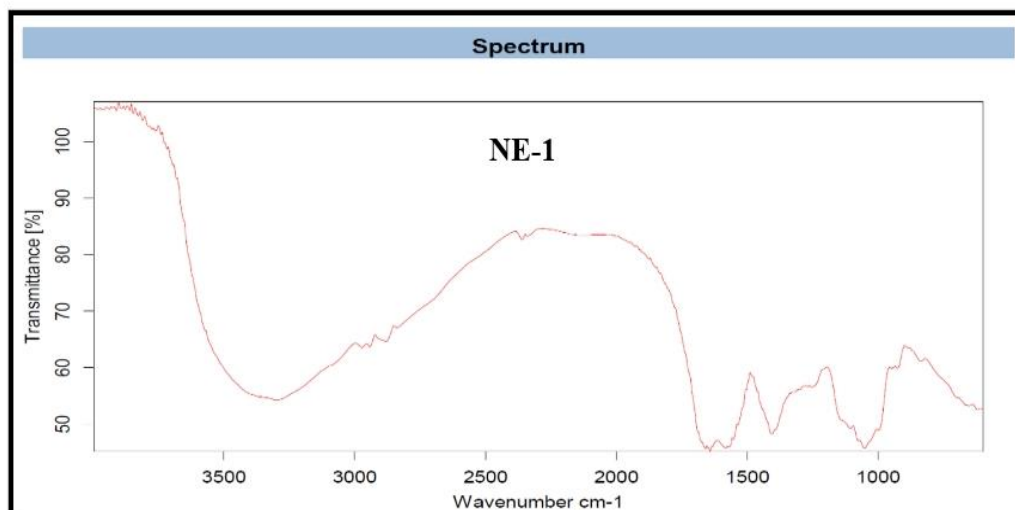
4.11 FTIR

Inorganic and organic chemical components are mixed in complex ways in coating formulations. A uniform dispersion of resins, pigments, and binders is often left behind after the carrier solvent in most coatings evaporates. Each element is further separated into configurations of atoms linked together in certain combinations from a basic chemical standpoint. These atomic arrangements, known as functional groups, serve as the fundamental building blocks for specifying a molecule's chemistry. For instance, the term "acrylic," which

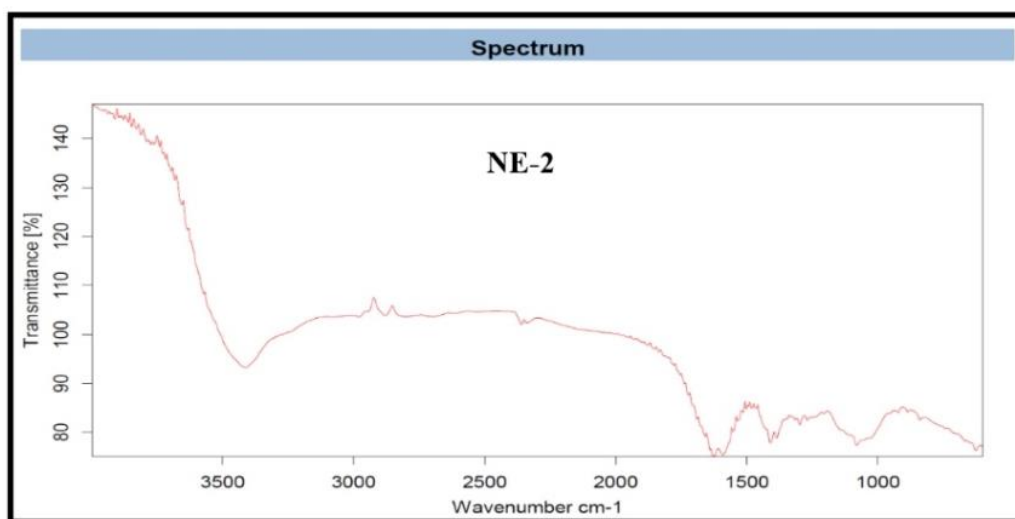
is frequently used and is chemically defined as "COO-R," mention to the functional groups present in the polymeric binders used in the coating. In the control sample a strong broad peak at around 3204.40 cm⁻¹ shows C-H stretching, sharp peak at 1591.25 cm⁻¹ representing C=O stretching and broad peak at 1048.86 cm⁻¹ shows elongation of C-O group. In NE1 sample broad peak at wavenumber 3294.6 cm⁻¹ shows C-H bonding with cumulative O-H stretching, small peak at wavenumber 1642.17 cm⁻¹ represents better C=O bonding and sharp peak was observed at wavenumber 1052.08 cm⁻¹ C-O. In NE2, a broad peak at wavenumber 3414.25 cm⁻¹ shows strong O-H bonding with cumulative C-H stretching. small peak at wavenumber 1629.53 cm⁻¹ represents better C=O bonding, and A small peak at wavenumber 1080 cm⁻¹ was noticeable that shows presence of weak C-O bonds in the sample. In NE3 sample a broad peak at wavenumber 3369.1 cm⁻¹ shows strong presence of O-H bonding. A small peak at wavenumber 1642.21 cm⁻¹ represents better C=O bonding, small sharp peak at wavenumber 1408.11 cm⁻¹ was present that shows same C=O stretching as the previous sample, and small peak was observed at wavenumber 1022.36 cm⁻¹ C-O stretching.



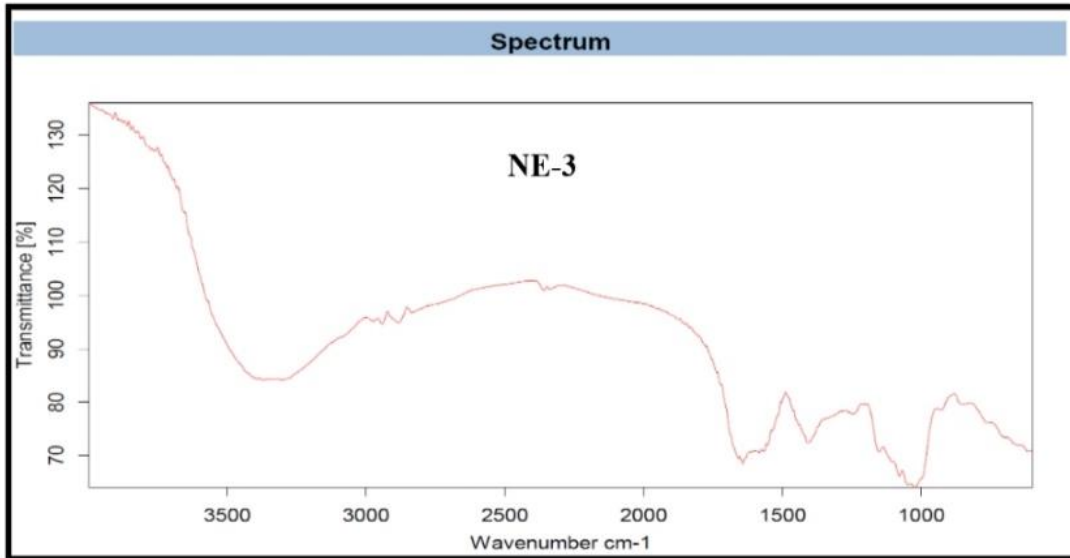
a)



b)



c)



d)

Figure 4.10 a), b), c) & d) shows FTIR analysis of clove essential oil at different concentration.

CHAPTER 5

SUMMARY AND CONCLUSION

5.1 Summary

The demand and supply for fresh cut fruits and vegetables is growing day by day constantly due to a high trend of eating healthy and fresh. Fresh cut fruits like pineapples are available in markets, and they are convenient to eat. People are all inclined towards fresh cut fruits because they are already processed and healthy. There are a number of techniques to improve and prolong the storage and keeping life of fresh cut fruits without damaging its nutritional quality. Fresh cut fruits are very perishable and prone to spoilage. Hence, sensory, nutritional and microbiological damage occurs very rapidly in fresh sliced fruits and vegetables. After peeling and cutting of fruits it tends to increase the metabolic and enzymatic processes due to presence of enzymes and causes quality deterioration like changes in color, pH, moisture, TSS, and microbial activity. The awareness to preserve and increase the storage life of freshly cut fruits and vegetables in natural ways is increasing. Use of edible coating with nanoemulsion is a natural technique to increase the period of storage of fruits and vegetables and is a cost effective, convenient and safe method for fruits and vegetables. There are different kinds and forms of edible coating material like protein, polysaccharide, lipids, gums, etc. In this thin layer of coating is applied which acts as a moisture and gases barrier. Pectin was used as coating material because it acts as a gelling and emulsifying agent. Clove and ajwain essential oil show good antimicrobial activity when used in combination with pectin.

5.2 Conclusion

The effect of pectin coating solution in combination with clove and ajwain essential oil on fresh cut pineapple was evaluated. The pectin coating solution alone does not show big changes in physicochemical properties. Thus, pectin alone was not effective on prolonging and extending the shelf life of freshly sliced pineapple. There was change in color and growth of microbes were also observed.

Coating of clove essential oil with pectin was done on freshly sliced pineapples, the pineapple retains color and appearance for about seven days of storage period. Coating

having ajwain essential oil was used to coat freshly cut pineapple and it does not show proper color retention.

This study concludes that pectin coating solution along with both clove and ajwain essential oil nanoemulsion exhibit good antimicrobial activity and antioxidant activity. Thus sample 5 coating gives the best coating solution result among all other coated samples.

The utilization of edible coating incorporated with nanoemulsion has coating and potential effect of extending the shelf life of freshly sliced pineapple. In general, the coated samples had more shelf life compared to uncoated samples. Hence it provides to be a good choice for the formation of edible coating in fresh cut fruits.

CHAPTER 6

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