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**Durability and feasibility test of a new organic
food packaging**

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Abstract:

The research tested new organic food packaging made from gelatin, glycerol, and sorbitol, with gelatin extracted from chicken feet using acetic acid. Different biopolymer combinations with essential oil were evaluated.

Physical analyses included opacity, permeability, thickness, and solubility. A microscopic study examined the biofilm surface prepared from gelatin on chicken feet.

The gelatin yield from 100 g of chicken feet was 11.22%, producing a light yellow, odorless crystalline powder. Sorbitol-based films were more transparent and flexible, while glycerol-based films were slightly yellow with normal flexibility. Both types provided reasonable food protection and reduced environmental impact.

Biofilm solubility varied with composition, with Sample 4 showing the highest solubility (48.78). Sample 8 had the highest opacity, and Sample 5 had the lowest water vapor permeability.

The biofilms were ecologically harmless and biodegradable. Cherry tomatoes remained fresh for 10 days using a film of sample 4.

Keywords: chicken feet, biodegradable film packaging, gelatin, microscopic study, conservability.

ملخص

اختبر البحث عبوات أغذية عضوية جديدة مصنوعة من الجيلاتين والجلسرين والسوربيتول، مع الجيلاتين المستخرج من أقدام الدجاج باستخدام حمض الأسيتيك. تم تقييم مجموعات مختلفة من البوليمر الحيوي مع الزيوت العطرية. وشملت التحليلات الفيزيائية العتامة والنفاذية والسكك والذوبان. دراسة مجهرية فحصت سطح الأغشية الحيوية المحضرة من جيلاتين أقدام الدجاج.

كان إنتاج الجيلاتين من 100 جرام من أقدام الدجاج 11.22%، وينتج مسحوق بلوري أصفر فاتح عديم الرائحة. كانت الأغشية المرتكزة على السوربيتول أكثر شفافية ومرونة، في حين كانت الأغشية المرتكزة على الجلسرين صفراء قليلاً مع مرونة عادية. يوفر كلا النوعين حماية غذائية معقولة وتقليل التأثير البيئي.

تباينت قابلية ذوبان الأغشية الحيوية حسب التركيب، حيث أظهرت العينة 4 أعلى قابلية للذوبان (48.78%). كانت العينة 8 هي الأعلى عتامة قرنفل. العينة 5، كانت لها أدنى نفاذية لبخار الماء.

كانت الأغشية الحيوية غير ضارة بيئياً وقابلة للتحلل. ظلت الطماطم الكرزية طازجة لمدة 10 أيام باستخدام طبقة من عينة .4

الكلمات المفتاحية: أقدام الدجاج، التغليف بالأغشية القابلة للتحلل، الجيلاتين، دراسة مجهرية، قابلية الحفظ.

Résumé:

La recherche a testé de nouveaux emballages alimentaires biologiques à base de gélatine, de glycérol et de sorbitol, avec de la gélatine extraite de pattes de poulet à l'aide d'acide acétique. Différentes combinaisons de biopolymères avec de l'huile essentielle ont été évaluées.

Les analyses physiques comprenaient l'opacité, la perméabilité, l'épaisseur et la solubilité. Une étude microscopique a examiné la surface du biofilm préparé à partir de gélatine de pattes de poulet.

Le rendement en gélatine de 100 g de pattes de poulet était de 11,22 %, produisant une poudre cristalline jaune clair et inodore. Les films à base de sorbitol étaient plus transparents et flexibles, tandis que les films à base de glycérol étaient légèrement jaunes avec une flexibilité normale. Les deux types offraient une protection alimentaire raisonnable et un impact environnemental réduit.

La solubilité du biofilm variait selon la composition, l'échantillon 4 présentant la solubilité la plus élevée (48,78 %). L'échantillon 8 avait l'opacité la plus élevée. L'échantillon 5, présentait la perméabilité à la vapeur d'eau la plus faible.

Les biofilms étaient écologiquement inoffensifs et biodégradables. Les tomates cerises sont restées fraîches pendant 10 jours grâce à un film d'échantillon 4.

Mots clés : pattes de poulet, emballage biodégradable, gélatine, étude microscopique, conservabilité.



Knowledge

First, I would like to thank Allah for the will, ability, and patience he has given me to accomplish this work.

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أهدي هذا البحث اولا .. إلى سند قلبي وضلعي الثابت ، وإلى الذي حرس لي بظله الطريق الشاق الى من كلله الله بالهبة والوقار إلى من أحمل اسمه بكل فخر ومن وضع كل فخره بي ستبقى كلماتك نجوم اهتدي بها الى حبيب روعي والدي الحبيب وقرّة عيني وطمأنينة فؤادي ابراهيم حفظه الله .

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Abbreviation list	
AFNOR	The French Association of Standardisation
ECHA	European Chemicals Agency
ANSM	According to the National Agency for the Safety of Medicines and Health Products,
SWOT	strengths, weaknesses, opportunities, threats
DMT	maximum tolerated dose
LD	lethal dose
%	Percentage
BSE	bovine spongiform encephalopathy
ALA	Alanine
PRO	Proline
GLY	Glycine
ARG	Arginine
HYP	L'hydroxyproline
PH	Potential hydrogen
BC	Before Christ
KG	Kilo grams

Introduction

Introduction

The packaging significantly contributes to creating an environment that maintains the food's taste, nutritional value, and health benefits. Consequently, the food is maintained in a contaminant-free environment, devoid of dust, microorganisms, and chemicals. Additionally, it is protected from external agents such as oxygen, which can rapidly deteriorate its quality and safety (Guillard & Gontard, 2017).

Extensive research has been conducted on biodegradable films due to their capacity for material protection. Food products and their ability to act as a barrier against moisture, gases, and odors. The solute has benefits such as the use of non-toxic ingredients and affordable. Contemporary patterns in using biopolymers for packaging expanded the variety of materials, encompassing natural substances and extracts. Regarding the subject of plants and nanomaterials. *Advanced Technologies: Active, Intelligent, and Nanotechnology* (Said & Sarbon, 2022).

Gelatin packaging films have been previously suggested to safeguard, preserve, or extend the shelf life of food items due to their excellent ability to form films and serve as exterior barriers, shielding the food from light and oxygen. Nevertheless, gelatin films have become less common because of the rise of cutting-edge technologies and evolving consumer demands for food safety. An idea has been proposed to expand the application of gelatin films as active and intelligent biodegradable packaging to ensure the safety of food products (Said & Sarbon, 2022).

Essential oils are composed of aromatic molecules of plant origin with diverse structures. However, these essential oils are obtained with meagre yields (about 1%), making them fragile substances that are rare and valuable. The different extraction techniques of essential oils or aromatic extracts must, on the one hand, consider these characteristics and, on the other hand, provide quantitative performances satisfying an increasingly demanding demand. Based on different phenomena (Lucchesi, 2005).

The objective is to extract gelatin from chicken feet and use it to create active biodegradable packaging. This packaging will be tested for durability and feasibility for a new organic food packaging.

This study comprises five parts: the first is a general introduction, the second is a bibliography devised into 3 chapters, the biodegradable packaging, the second is a part of gelatin, and the third is essential oil. The fourth part presents the material and methods,

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containing a presentation of place and raw material, gelatin extraction, film creation, physical analysis, the fourth part results, and a discussion of our results.

I. Bibliography

1. Definition of packaging

Packaging serves as a means of communication with consumers, functioning as a marketing tool while safeguarding the product from potential harm caused by the external environment (Otles & Yalcin, 2008).

Packaging typically comprises diverse components with varying shapes, functions, and materials, irrespective of the composition of the packaging itself. Food packaging, particularly for delicate and perishable items, must not cause any health hazards as its primary purpose is to store and safeguard goods. The product's compatibility should align with its inherent characteristics, including its physical shape, protective measures, and susceptibility to deterioration from biological or chemical sources (ŠČETA, 2022).

2. Packaging History

The history of packaging is inseparable from human exchanges and their movements. When people needed to venture away from their tribes and carry provisions, they had to invent packaging to gather, transport, protect, and preserve goods. The earliest forms of packaging date back to prehistoric times, using animal skins (such as the hunter's pouch or gourd) or leaves. Around 6000 BC, ceramics and baskets emerged. By 1500 BC, the Egyptians were crafting glass containers (Dumeni, 2006).

In 1746, the first branded product appeared in England: a box of fever powder. The country further distinguished itself with branded soaps, oil, and mustard packaging. Until the late 19th century, people made the best use of materials provided by nature for packaging, including wood, cork, leather, clay, and fibers (such as hemp, jute, raffia, and willow), as well as transformed materials like glass, metals, and paper (Dumeni., 2006).

In 1850, the introduction of the flexible paint tube allowed artists to travel through the countryside and paint outdoors, liberating them from the confines of the studio or the window of a dwelling. It was this packaging innovation that facilitated the rise of Impressionism (Dumeni, 2006).

The history of packaging reflects the evolution of human society through the centuries. Since the initial use of wood, humans have constantly innovated how to condition objects, exploring new materials over time. First, wood, then pottery, glass, and some metals were used to pack various products (Carré, 2016).

In the early 1900s, the introduction of tin represented notable progress, which was soon followed by the emergence of plastics in the aftermath of World War (Carré, 2016).

3. Development of packaging

Packaging is a crucial element of a continuous and forward-thinking process that aims to minimize losses. This process involves using various techniques relevant to each step of the packaging process (Ols mats & Wallteg, 2009).

The food packaging sector worldwide has the potential to greatly decrease losses, guarantee food safety, and improve international food trade, all of which are crucial for the growth of diverse economies (World Economic Forum, 2009).

Furthermore, the state and food packaging trends in developing countries are examined, followed by an analysis of the food sector's strengths, weaknesses, opportunities, and threats (SWOT). This information is necessary to propose solutions for developing countries to overcome current challenges in the agri-food sector to meet their packaging needs (FAO, 2014).

The packaging sector in emerging countries has experienced general expansion due to the rising demand from the food and beverage market (Global, 2010).

4. Active biodegradable packaging

4.1. Definition of a biodegradable film

Biodegradable films refer to a material designed to decompose in the environment naturally. These films are typically made from plant-based materials like cellulose or proteins and are often utilized as an alternative to traditional plastic films (Pirsa & Aghbolagh, 2020).

Bacteria, yeast, and fungi can break down biodegradable materials through enzymatic action. The food packaging industry has focused on developing biopolymers (bioplastics) derived from renewable sources. These bioplastic materials should possess sufficient durability to maintain their mechanical and physical properties for product quality while rapidly biodegrading upon disposal (Yahyaoui, 2020).

Biodegradable films are designed as substitutes for polyethylene films used for various purposes, from industrial films to packaging products and bags for organic waste collection. Such materials exhibit superior properties compared to traditional non-degradable plastics. They resist moisture, maintaining the warmth of organic matter for several weeks or even

months without altering their physical properties. This allows for greater flexibility and makes them excellent replacements for current films used in product storage, transportation, and packaging, as they are fully biodegradable (Ivonkovic & al, 2017).

4.2. Definition of Active Packaging

Active packaging is designed to extend packaged food products' shelf life or maintain or improve their condition. These packages are intentionally composed of components that release or absorb substances into the packaged food products or their environment. In 1995, the Definition focused more on the preservation role, considering packaging to have a role other than simply serving as a passive barrier against the external environment (Gimenez & Aoussat, 2011).

The condition of food, within the context of active packaging, encompasses various factors influencing its shelf life, such as physiological processes (like the respiration of fresh fruits and vegetables), chemical processes (like lipid oxidation), physical processes (like bread staling and dehydration), microbiological aspects (like spoilage by microorganisms), and infestation (for example, by insects) (Altaf & al, 2018).

4.3. Active biodegradable packaging property:

Active biodegradable packaging is both biodegradable packaging and has additional qualities that can assist in maintaining the quality and shelf life of food products. Possible characteristics of active biodegradable packaging may encompass:

- Ensure consumer safety.
- Preserve the organoleptic properties of products for more extended periods.
- Help prevent bacterial growth in food (Shaikh & al, 2021).
- And prolong the duration that things can be stored without spoiling.
- Water vapor and aromatic molecules are crucial for enhancing product shelf life and improving the performance of oxygen and water vapor barriers (Sabry, 2022).
- Preserving, safeguarding, and containing food: Packaging serves as a barrier to prevent moisture, light, gases, and microbes from affecting food, reducing food waste and preventing contamination that could harm people's health (Muszynski & al, 2021).

4.4. Materials are involved in the synthesis of a biodegradable film

4.4.1. Polysaccharide-based Packaging

Carbohydrate polymers are composed of elongated chains in which monosaccharide units are connected by glycosidic linkages (Baghi, 2022).

Biodegradable films are often produced using polysaccharides such as starch, cellulose, chitosan, and hydrocolloid gums. These polysaccharides' unique molecular features directly impact the physical, chemical, and functional attributes of the packaging materials produced from them (Sani, 2021).

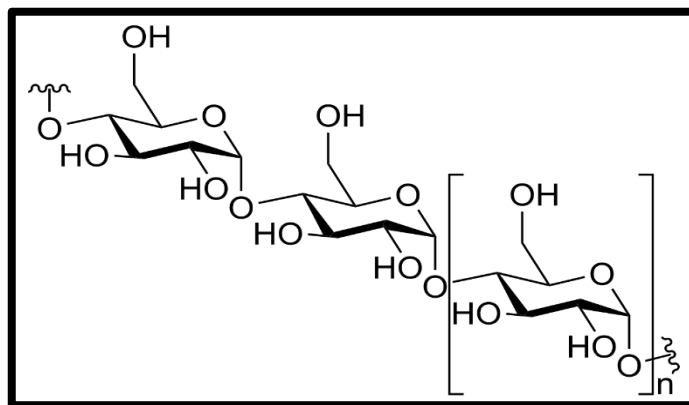


Figure 01: Structure of polysaccharides **anonym 01 (electronic document)**

4.4.2. Lipid-based packaging

Lipids are compounds that originate in natural sources such as animals, insects, and plants. The diversity of the lipid functional groups comprises phospholipids, phosphatides, mono-, di-, triglycerides, terpenes, cerebrosides, fatty alcohols, and fatty acids (Mohamed & al,2020).

They are derived from diverse natural sources, such as animals, plants, and insects. These lipids, glycerides, and waxes are predominantly employed in the production of films. These chemicals are predominantly nonpolar and highly hydrophobic, rendering them unable to dissolve in water-based environments yet capable of dissolving in organic solvents. Because of their exceptional ability to prevent moisture from passing through, they are utilized to produce coatings and edible films using a range of biodegradable substances, including polysaccharides and proteins (Baghi & al, 2022).

4.4.3. Protein-based packaging

Proteins, derived primarily from plants and animals, are vital biobased polymers. They consist of different α -amino acids, regardless of their polarity. Plant-based protein sources include wheat gluten, corn zein, soy, peanuts, rice bran, cottonseed, barley, and sunflower. Animal-derived alternatives include Gelatin, collagen, casein, whey protein, and fish myofibrillar protein. Proteins are widely used in packaging materials due to their exceptional mechanical qualities, easy availability, and high nutritional value (Baghi & al, 2022).

Ils peuvent provenir de films et de revêtements hydrophiles et de bonnes barrières à l'oxygène mais une faible résistance mécanique (Barbosa & al, 2021).

Moreover, their non-toxic and biodegradable properties make them highly intriguing contenders for active packaging applications, presenting prospects for innovation in this domain (Baghi & al, 2022).

1. Definition of Gelatin

Gelatin, a protein-rich food substance, is primarily produced through the thermal transformation of collagen, a fundamental protein widely present in animal tissues and playing a significant structural role (Mariod & Fadul, 2013).

En général, le collagène utilisé dans la fabrication de gélatine est extrait principalement des os de bovins et des peaux de porc. Cependant, il peut également être occasionnellement obtenu à partir de sources alternatives telles que la peau de poisson, les peaux ou le poulet, selon (Amirrah & al, 2022).

Gelatin is a protein material derived from collagen, a natural protein found in animals' tendons, ligaments, and tissues. It is derived by simmering animal bones and skin tissues, often from cows and pigs (Kucińska & al, 2014).

2. Gelatin structure

The gelatin molecule can contain between 300 and 4000 units of amino acids. The sequence of these is as follows -Ala-Gly-Pro-arg-Gly-Glu-Hyp-Gly-Pro- as the following schema represents.

The hydrolysis in the acid medium of collagen allows the dissociation of the three chains of this Protein, called tropocollagen, responsible for the texture of Gelatin, which are suspended particles (Krimm & Bandekar, 1986; Ibrahim & Chem., 2000). This gelatin molecule is characterized by the crude chemical formula $C_{102}H_{151039}N_{31}$ (Meudre, 2015).

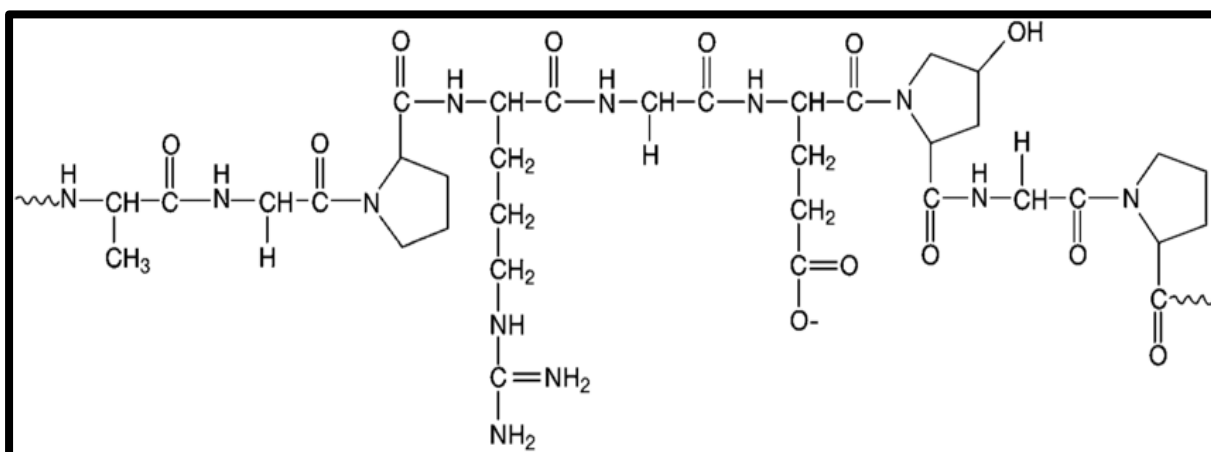


Figure 02 : structure chimique de gélatine Anonyme 2 (document électronique).

3. Nutritional quality of Gelatin

Gelatin is rich in proteins and antioxidants, which can safeguard biological cells. This, in turn, promotes the well-being of the digestive system, bones, skin, joints, and other bodily functions (Dany & al, 2023).

Additionally, it serves as a superb reservoir of:

- Calcium;
- Magnesium;
- Folate;
- Choline;
- Sodium;
- Selenium;
- Nutrients per serving (Dany et al, 2023).

A tablespoon (7 grams) of Gelatin contains (Dany et al, 2023):

- Calories: 10.
- Protein: 6 grams.
- Fats: 0 g.
- Carbohydrates: 2 grams.
- Fibre: 0 grams.
- Sugar: 2 grams.

4. Source of gelatin

4.1. mammalian gelatin

sources primarily consist of beef, pork, and bone. 41 grams per 100 grams of gelatine worldwide is derived from hog skin, while 28.5 grams per 100 grams comes from bovine skin and 29.5 grams per 100 grams from bovine bone. The use of breast gelatin is limited in many countries due to concerns related to bovine spongiform encephalopathy (BSE) and religious beliefs (Shakila, 2022).

4.2. Insect gelatin

Insect gelatin can serve as a suitable substitute for Muslim items. In Sudan, edible insects are commonly consumed, with the pilgrim cricket being particularly renowned in various areas (Mariod & al, 2013).

4.3. Poultry gelatin

Poultry gelatin derived from poultry skin, feet, and bones has garnered interest as a potential alternative to gelatin derived from mammals. The avian species comprise duck, chicken, and turkey (Said & al, 2023).

Avian gelatin has been found to possess amino acids, a secondary structure, and a molecular weight that is nearly identical to mammalian gelatin (Said & al, 2023)

5. Type of gelatine:

Gelatin can be classified into two categories based on its specific treatment during manufacturing. Type A gelatin is derived from collagen treated with acid and has an isoionic point ranging from 6 to 9. Conversely, type B gelatin, which has an isoionic point of 5, is obtained from a precursor treated with alkali. Historically, Gelatin sourced from pigskin is categorized as type A, whereas Gelatin generated from cattle skin is classed as type B (Jones, 1987).

6. Properties associated with Gelatin:

6.1. Viscosity of Gelatin

Viscosity is an essential property of Gelatin-Gelatin that is vital in preserving the smooth texture of products made with Gelatin-Gelatin. It is impacted by multiple aspects, including extraction conditions (pH and temperature), gel strength, molecular weight distribution, and characteristics of input materials. Viscosity is influenced by factors such as extraction time, temperature, pH, polydispersity, molecular size, and weight distribution (Duan & al, 2018).

6.2. Gelification of Gelatin

Gelatin is extensively utilized in the food business because of its ability to form a gel and its sensory qualities. The gelation properties of Gelatin are essential for determining its gel strength, gelation time, Viscosity, texturing, thickening, setting, melting temperature, and water binding capacity. The distinctive attributes of Gelatin are influenced by various aspects, including the origin of the raw materials, the makeup of amino acids, the method used for

gelatin extraction, and the general patterns of amino acid cross-linking (Ranasinghe & al, 2022).

6.3. pH of Gelatin

The pH of Gelatin is mostly determined by the chemicals used in the extraction process of its raw materials and the distribution of charge of the amino acids found in the Gelatin (Al-Hassan, 2020).

7. Applications of Gelatin

Gelatin has historically been employed in three primary sectors: the food, medicinal, and photography industries. The most prevalent application of gelatin in the culinary business is typically in gelatin desserts and confectionery products. Furthermore, it is a binding agent and coating substance in meat products. Gelatin is utilized in the pharmaceutical industry to manufacture intricate and pliable capsules for pharmaceuticals, health supplements, syrups, and other related products. It has a high level of digestibility and functions as a natural protective layer for medications (Anonyme, 2001).

- **In the food industry:**

Gelatin is widely utilized in modern food production for its desirable texture, making it an essential element in business. It is frequently added to margarine, low-fat butter, spreads, and reduced-fat cheeses. Gelatin is essential in many confections, such as marshmallows, due to its neutral taste, high water content, gel-forming ability, and pleasant mouthfeel (Anonyme, 2001).



Figure 03 : gummies (Anonyme, 2001).

- **In the pharmaceutical industry**

Gelatin produces capsules and coatings that safeguard pharmaceuticals from light and oxygen's detrimental impact. Capsules are predominantly for liquid pharmaceuticals, while coatings are employed for powders. Gelatin acts as a binder for the prescription's active ingredients, thereby prolonging their shelf life by carefully selecting and measuring their dosage. Gelatin can modulate the release rate of active substances, either accelerating or decelerating it (resulting in a delayed effect) (Anonym, 2001).



Figure 04: Gelatin in pharmaceutical production (Anonyme, 2001).

1. Definition of essential oil

Essential oils are natural secretions of aromatic plants' various organs (leaves, fruits, roots, petals, etc). There are several definitions to characterize them. According to the National Agency for the Safety of Medicines and Health Products (**ANSM**), " the essential oil is a fragrant product, usually of a complex composition, obtained from a botanically defined plant raw material, either by steam entrainment, dry distillation, or by a suitable mechanical process without heating. The essential oil is most often separated from the aqueous phase by a physical process that does not cause a significant change in its composition " (Fourmentin & Kfoury, 2024)

The French Association of Standardization (**AFNOR**) gives a precise and official definition of essential oil: a product obtained from a natural raw material of vegetable origin, either by steam drive or by mechanical processes from the epicarp of citrus fruits (citrus) or by dry distillation, after separation of the possible aqueous phase by physical processes that do not result in a significant change in its composition " (Fourmentin & Kfoury, 2024).

Finally, according to the European Chemicals Agency (**ECHA**): «An essential oil is defined as a volatile part of a natural product, which can be obtained by distillation, steam distillation or expression in the case of citrus fruits. It contains mainly volatile hydrocarbons. Essential oils are derived from various plant sections. Oil is «essential» in the sense that it has a distinctive smell, or essence of the plant» (Fourmentin & Kfoury, 2024).

2. Toxicity of essential oil

Despite the beneficial effects of essential oils, they can become toxic either in situ (irritation, allergy, phototoxic reactions) or at the level of the organs (neurotoxicity, hepatotoxicity, nephrotoxicity, etc.). To avoid all risks, it is necessary to assess the dangers a risk may present at a given level of exposure (Charik & Kadri, 2020)

Essential oils are intricate combinations of molecules with properties like lipophilicity and low molecular weight, facilitating their effective diffusion in the body. Despite limited research, it is crucial to recognize their potential toxicity, including hepatotoxicity, neurotoxicity, and recently identified dermal toxicity. The broad spectrum of their usage poses a significant risk of poisoning, whether due to accidental exposure or misuse (Lucette, 2001)

Clinical cases have revealed only a few specific signs of intoxication, making differential diagnosis challenging for clinicians. Treatment typically focuses on managing symptoms, though there is optimism for more targeted and, thus, more efficacious therapies in the future. It is essential to remain vigilant and dispel the misconception that all "natural" products are inherently safe, as they can still carry toxic risks under certain circumstances (Lucette, 2001).

The toxicity of essential oils is very variable, depending on the mode of administration, the dose taken, the profile of the user, its tolerance threshold, and each essential oil. Theoretically, there is acute and chronic toxicity even if, in practice, there is not much data available. Acute toxicity is calculated by the lethal dose that kills 50% of animals (LD50) and is expressed in mg/kg. Chronic toxicity, meanwhile, considers repeated use of therapeutic doses. It is determined by the maximum tolerated dose (DMT) in g/kg day over eight weeks. This maximum tolerated dose is the dose that can be administered without risk of toxicity (Poudevigne, 2024).

Although approved as food additives, certain essential oils such as lavender, jasmine, rosewood, laurel, eucalyptus, ylang-ylang, lemongrass, clove, and pomegranate can cause allergic reactions (Yahyaoui, 2020).

3. Application of essential oil:

Essential oils have numerous applications in daily life, particularly in cosmetics, health, well-being, and aromatherapy. Here are some common examples of how essential oils are used:

- ❖ **Cosmetics:** Essential oils are widely used in the cosmetic industry for their beneficial properties for the skin and hair. They can be added to creams, lotions, soaps, shampoos, and other beauty products for their purifying, soothing, decongestant, firming, and antioxidant effects (Fourmentin & Kfoury, 2024).
- ❖ **Aromatherapy:** Essential oils are at the core of aromatherapy, a natural therapeutic approach that uses aromatic plant extracts to promote physical and mental well-being. They can be inhaled, applied to the skin, or ingested to treat various psychological disorders, calm the nervous system, relieve stress and anxiety, and improve sleep quality (Fourmentin & Kfoury, 2024).

- ❖ **Perfumery:** Essential oils are extensively used in the perfume industry to create natural perfumes and unique fragrances. Their use dates to antiquity and is valued for their subtle and varied aromas (Fourmentin & Kfoury, 2024).

II. Material and Methods

II. Material and Methods

1. Objective:

The most important thing is to provide biodegradable packaging materials that are comparable in characteristics and advantages to the conditioning materials because we intend to use poultry feet instead of destroying them.

This study aims to extract gelatin from chicken feet and use it to create active biodegradable packaging. This packaging will test the durability and feasibility of new organic food packaging.

2. Study Location:

Our study was conducted at laboratory No. 01 to Microbiology in the teaching block B Department of Applied Biology, Faculty of Natural Sciences and Life Sciences, Echahide Chikh Larbi University Tebessa –Tebessa.

3. Animal Material:

The chicken feet used in this study were brought from the poultry slaughterhouse Boutarfa - Tébessa-, more precisely, in the Industrial area. The feet have four fingers and are covered with scales. They end up with big claws and an unpleasant smell. They are transported to the Lab at 1°C. We chose fresh feet, which are not bleeding or moldy yellow.



Figure 05: Chicken paw after cutting nails and removing crust external (personal photo).

II. Material and Methods

For the extraction of gelatin, the products and materials are listed in the following table:

Table 01: Material and products used in gelatin extraction.

Material and Glassware	Products
<ul style="list-style-type: none">• cooler.• Refrigerator.• ADE Scout Pro analytical scale.• Heated agitator by LAB TECH.• Etuve at 45C°• Benzene beak• Beakers (500ml).• Erlenmeyer (1l).• Graduated pipette.• Petri.• Micropipettes.• Aluminum foil.• Parafilm.• Muslin cloth• Spatulas.• Scratch Limes.• Knife.	<ul style="list-style-type: none">• Chicken feet.• NaOH SIGMA ALDRICH.• acetic acid Ridel-de Haen.• Distilled water.• Tween 20.• Glycerol.• Ethanol.• Agar PCA CONDALAB.• GN.• MacConkey.

4. Sample preparation:

- Wash chicken feet with tap water at least twice to remove dust and all pollutants and impurities;



Figure 06: Washing chicken feet (personal photo).

-Remove nails by nail clipping and pliers, remove crust from the skin of the feet of chicken with a knife, and wash for another time with water;

- Cut chicken feet into small pieces (5cm), then ground them with a meat grinder.

II. Material and Methods



Figure 07: Cutting the feet
(personal photo).



Figure 08: Cut the nails and decouples the skin
(personal photo).

-Weigh each 100g in a plastic bag and store them in the freezer at 4°C until the chicken feet are useful.



Figure 09: Grinding chicken feet by meat grinder (Personal photo).



Figure 10: Weigh each 100g in a plastic bags (personal photo).

II. Material and Methods

5. Preparation of solutions

5.1. Preparation of sodium hydroxide (NaOH) solution 0.5 M:

-Introduce a mass of 20 g NaOH into a 1L volumetric flask, half filled with distilled water, using a funnel;

-Homogenize the solution by turning and shaking the blocked vial, then complete to the gauge line.

5.2. Preparation of 5% acetic acid solution from 100% concentration stock solution:

-Pour 50 ml of acetic acid into a 1L volumetric flask containing little distilled water, plug the vial, and shake the solution;

-Complete with distilled water to gauge line, shake to make homogeneous;

-Acetic acid solution should be prepared when used.

Note! During the preparation of acetic acid solution, wear glasses, protective gloves, and lab coats. Work under a hood or in a ventilated area.

5.3. Gelatin extraction

Extraction was carried out according to (Suparno & Prasetyo, 2019), (Chakka al. 2017), and (Fatima & al, 2022) with modification, and after several tests, we followed the present protocol with (Dr. Ferhi Selma).

The first step is the pretreatment process with a NaOH solution to eliminate non-collagen proteins and other impurities such as fats, minerals, pigments, and odors (Suparno & Prasetyo, 2019). 100g of ground chicken feet were mineralized with sodium hydroxide for 20 h at refrigeration, then filtered by muslin cloth three times (Chakka & al, 2017). The residue was washed repeatedly with distilled water until pH became neutral.

Excess water was removed by filtration using a muslin cloth (Fatima& al, 2022).

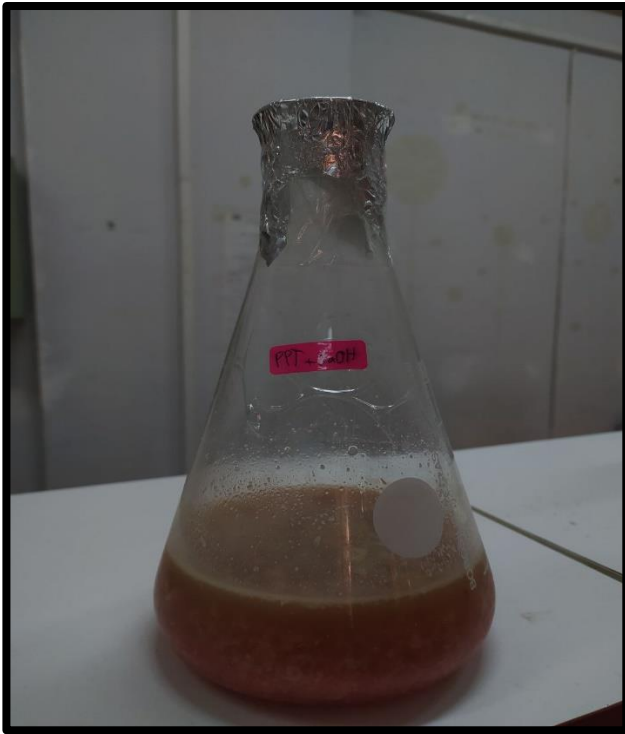


Figure 11: Feet after treatment
By NaOH (personal photo).



Figure 12: Muslin filtration
(personal photo).

5.4. Hydrolysis with a solution of acetic acid

The second step was hydrolysis with an acetic acid solution (CH_3COOH) to modify the structure of collagen fibers to facilitate the extraction process (Suparno & Prasetyo, 2019). Chicken feet are soluble in acetic acid for at least 3.5 hours under heating agitation at $66\text{ }^\circ\text{C}$.

Acetic acid is evaporated and filtered by muslin cloth at least two times.

Small holes were realized on the surface after putting the gelatin in Petri dishes. Keep the boxes in the freezer at $-4\text{ }^\circ\text{C}$ one night.



Figure 13\ 14: Acetic acid hydrolyzation (personal photo).

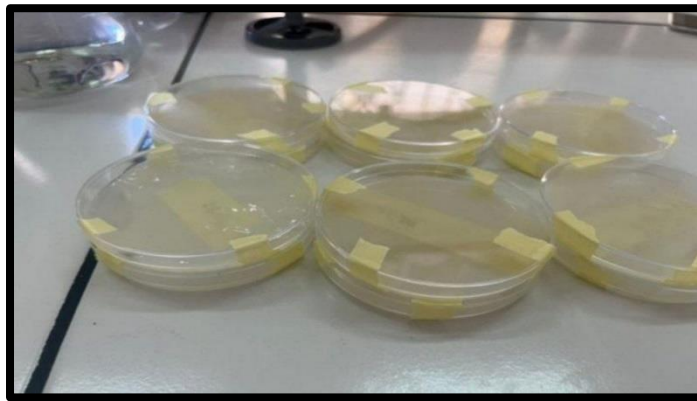


Figure 15: Put the gelatin in the kneaded boxes (personal photo).

5.5. Drying

The third step is drying the gelatin solution using a steam room (steamer) at 45°C for at least 48 hours until total water removal; then, the gelatin sheet was ground manually using mortar to obtain gelatin powder.

II. Material and Methods



Figure 16: Put the boxes in the etuve (personal photo).



Figure 17: Gelatin (personal photo).

II. Material and Methods

6. Determination of extraction yield:

The yield of gelatin extraction was expressed as a percentage and calculated as follows:

$$R = (PG/PP) \times 100$$

R: extraction yield in %.

P.G.: Weight of gelatin powder.

PP.: Weight of chicken feet.

7. Preparation of gelatin film

7.1. Material used in film preparation.

7.1.1. Gelatin: Extracted from poultry feet:

Gelatin's gelling ability is undoubtedly one of its most essential properties. When placed in cold water, gelatin swells, absorbing 5 to 10 times its volume of water. When heated to temperatures above its melting point, swollen gelatin dissolves, forming a gel on cooling. This sol-gel conversion is reversible and can be repeated, which is advantageous in many food applications.

Also, gelatin gels begin to melt between 27 and 34°C and tend to melt in the mouth. This is a desirable property in many foods (Imeson, 1992).

8. Essential oil of organic clove:

The essential oil used in this study is an oil clove of Girofle obtained by steam distillation. The part of the plant used for extracting the oil is Nails.

Quality: Botanically and Biochemically Defined Essential Oil (HEBBD).

- 100% pure (free from other similar essential oils).
- 100% natural (not denatured with synthetic molecules).
- 100% integral (non-discolored, non-terpenated, non-rectified, etc.).

Food quality

Country of origin: Madagascar.

Culture: Organic, certified by Ecocert FR-BIO-01.

Presentation: Amber glass bottles with codigoutte.

II. Material and Methods

Conservation: Essential oils are sensitive to UV radiation and the gradual evaporation of their constituents. Therefore, they must be stored in a colored bottle at 5°C.

For the creation of films, the products and materials are listed in the following table:

Table 02: Materials and products used in the creation of the film.

Material and Glassware	Products
<ul style="list-style-type: none">• Refrigerator.• Scout Pro analytical scale.• Precision scale KERN ALS220-4N.• heating agitator LAB TECH.• Etuve at 45°C.• Benzene beak<ul style="list-style-type: none">- Beakers (500ml).- Beakers (50 and 20 ml)• Erlenmeyer (1l).• Graduated pipette.• Petri.• Micropipettes.• Aluminum foil.• Parafilm.• Muslin cloth.• Spatula.	<ul style="list-style-type: none">• Gelatin extracted from chicken feet.• Tween 20.• Glycerol.• Essential oil of clove.• Distilled water.• Sorbitol.

9. Film creation:

Gelatin-based film preparation was performed according to (Lee & al, 2015) and (Nazim & Sarbon, 2019) with modification, and after several tests, we followed this protocol with (Dr. Ferhi Selma).

To prepare a gelatin powder film-forming solution, gelatin was mixed with 100 ml of bidistilled cold water (Lee & al, 2015), ensuring that all particles were moistened evenly and allowed to swell sufficiently for at least 5 min; the mixture was stirred continuously for 10 min at 60 °C. Then, glycerol (plasticizer), tween 20 (emulsifier), and sorbitol (Lee & al, 2015) were added, stirring for 10 min at 40°C.

Add frankincense essential oil, stirring for 2 min after lowering the solution's temperature to preserve the oil's volatile compounds. After filtration of the film-forming solution with a muslin cloth (the filtrate 80 ml), each 20 ml was poured onto the Petri dish and left at room temperature for 18h and oven-dried to 45°C (Nazmi & Sarbon, 2019).

II. Material and Methods

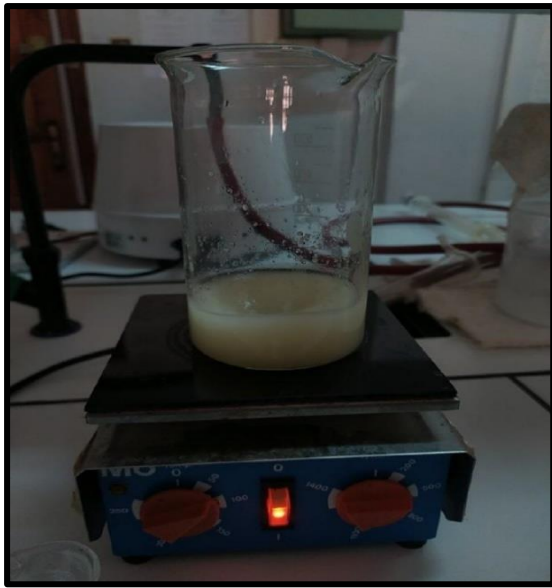


Figure 18\19: Preparation of the filmogenic solution (personal photo).



Figure 20\21: Filtration of the filmogenic solution (personal photo).

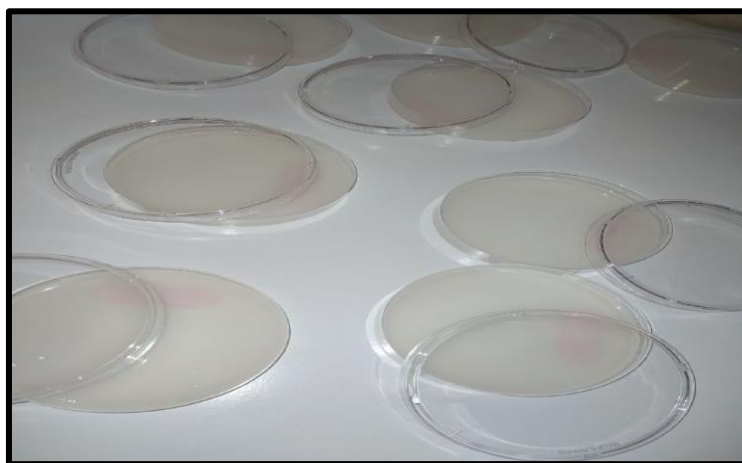


Figure 22: Injection of the filmogenic solution in the boxes Petries (personal photo).

II. Material and Methods

10. Microscopic biofilm analysis:

10.1. Polarizing light microscope

The microscope used in our LEICA DMLP brand study is a high-quality polarizing light microscope for research. This microscope is configured for essential polarization for hair and fibre analysis purposes.

10.2. Method of work

We collected biofilm samples containing gelatin, frankincense oil, glycerol, and sorbitol, respectively. The samples were placed directly on the microscope slide, and observations were made using magnifications X10 and X20.

The samples analyzed: sample 1, sample 2, sample 3, sample 4, sample 5, sample 6, sample 7, sample 8.



Figure 23: Microscope Leica Microsystems Wetzlar GmbH Type DM LM/P 11888500.

11. Sensory analysis

Sensory analysis is a multidisciplinary science involving tasters and their senses of sight, smell, taste, touch, and hearing to measure, analyze, and interpret reactions to organoleptic characteristics and product acceptability of food and many other products (Watts & al, 1991; Jain & Upta, 2005).

II. Material and Methods

Therefore, it is a question of using the human being as an instrument of measurement, using olfactory, gustatory, visual, auditory, and tactile abilities to characterize and evaluate a product's sensory qualities (ISO 5492, 1992; Claustrioux, 2001; Chen-yen-Su, 2016).

- The sensory analysis of extracted gelatin and the different biofilms was conducted in our laboratory.
- The sensory differentiation between the samples of the bios films developed was conducted to compare color, texture, smell, and appearance; eight samples of biofilms are put in comparison:

12. Physical Analysis

12.1. Film thickness

The thickness of the biodegradable gelatin-based films developed was determined using a manual micrometre (Mitutoyo, model 102 - 707, Japan, accuracy: 0.001 mm) from the average of at least three random measurements made on each film (NurHanani & al, 2013).



Figure 24: Micrometer TESAMASTER (personal photo).

12.2. Opacity

The opacity of the films was determined by measuring the optical density of the films cut into rectangular pieces from a size of 1/2cm to a wavelength of 500 nm. Subsequently, the opacity was calculated according to the following equation described by (Gontard & al, 1994).

$$\text{Opacity} = \text{absorbance at 500 nm/film thickness}$$

II. Material and Methods

12.3. Solubility of films in water

The solubility of prepared films in water was determined using the method described by (Rhim & al, 2005).

Samples randomly selected from each film were dried in an oven for six hours to determine the initial dry film mass (DM).

Then, each sample is individually placed in beakers containing 40 ml of distilled water.

The beakers are then covered with parafilm and kept at room temperature for 24 hours.

The undissolved film mass was determined by removing the film pieces from the beakers, rinsing them with distilled water, and drying them in an oven (6 h).

The water-soluble film mass (Mh) was calculated by subtracting the undissolved dry matter mass from the initial dry film mass.

12.4. Permeability of films to water vapor

Calcium chloride (CaCl₂) was added to the beakers to measure the permeability of films prepared with water vapor.

These beakers are then covered by films, without holes or defects, and sealed with parafilm.

The beakers are placed inside a desiccator containing distilled water to maintain a relative humidity gradient of around 100% through the film.

The relative humidity inside the beaker is always lower than outside. The water vapor transferred through the film and adsorbed by the desiccant is determined from the mass increase of calcium chloride recorded at different times.

The beakers are weighed at the start and every 12 hours within 24 hours.

13. Solid-medium (soil) biodegradation tests:

Soil biodegradation tests are most often performed in natural conditions. In this case, 08 samples were used:

Samp 1, samp 2, samp 3, samp 4, samp 5, samp 6, samp 7, samp 8.

The biodegradability of the films was achieved through the laboratory-wide soil degradation experiment described by Sanhawong & al, (2017).

The duration of the tests varies from 06 to 10 days.

II. Material and Methods

We used 08 Becher and put soil in both, not forgetting to keep the depth of the outer surface at 02 cm, then we put the sample into both; after we finished putting the soil to get up to the surface, continuing to swallow the ground every day.

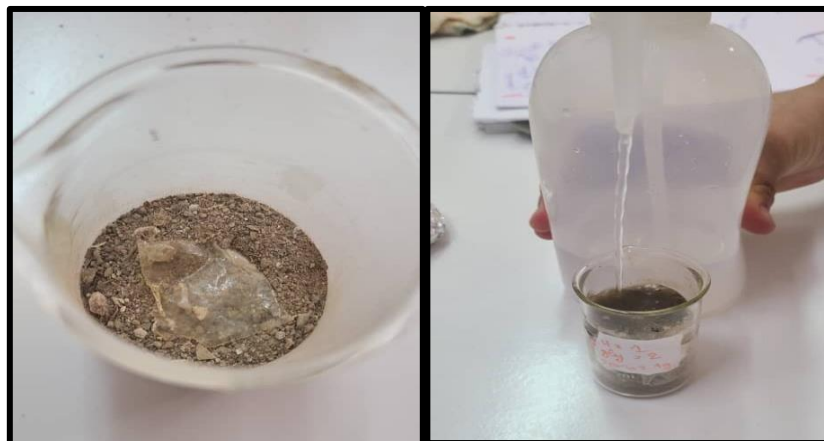


Figure 25: Add the films to the soil (personal photo).

14. Application of filmogenic solutions as cherry tomato coating (enrobing)

Fresh cherry tomatoes imported from the local market of the same caliber and without deterioration or injury were soaked in film-based coating solutions for 60 seconds before being drained from the excess coating.

The test group was tricked into distilled water after drying it with compresses. Subsequently, groups of cherry tomatoes were kept separately at room temperature at 20 °C for 10 days.

Table 03: Shelf life of the eight groups of cherry tomatoes.

Solutions	Number of pieces	Retention period
Solution 1	10 pieces of cherry tomatoes.	10 days
Solution 2	10 pieces of cherry tomatoes.	10 days
Solution 3	10 pieces of cherry tomatoes.	10 days
Solution 4	10 pieces of cherry tomatoes.	10 days

III. Results and Discussion

III. Results and Discussion

1. Gelatin Extraction

The extraction yield of gelatin from the chicken feet was calculated based on the weight ratio of 3 samples of gelatin powder. As indicated (**Diagram 02**), the extraction yield was approximately **11.22%**. This value is higher than that found by Fatima & al,2022 (**Between 3.50 % and 7.65 %**) and too high compared to the yield of (Suparno & Prasetyo, 2019) (**0.14 %**). In addition, the value is also higher than the gelatin yield of (Chakka & al, 2017) (**10.16%**) on both sides and higher than the 2 values (**6.59% and 8.51%**) presented by the same reference.

The acetic acid extraction method has the advantage of producing collagen relatively fast, requiring little equipment, producing collagen continuously, producing little waste, and reducing production costs. The difference in yield can be caused by the difference between extraction methods, solution concentration, material type, and extraction temperature and time (Zaelani & al, 2019).

Table 04: % gelatin yield extracted from chicken feet by various acid treatments.

Source and extraction method	The yield in %
Our result	11,22%
Extracting from chicken feet by acetic acid (Chakka & al, 2017).	6.59 %
	8,51%
	10,16%
Extracting from chicken feet by acetic acid (Fatima & al, 2022).	0,14 %
Hydro-extraction from chicken feet (Suparno & Prasetyo, 2019).	Between 3.50 % et 7.65 %

2. Organoleptic characterization of essential oil

Essential oils are usually liquid and volatile at room temperature, with aromatic odors distinguishing them from fixed oils. They are colored (AFNOR, 2000).

The results of the organoleptic properties of Girofle oil are presented in the following table:

III. Results and Discussion

Table 05: Organoleptic characteristics of oils.

Essentials oils	Organoleptic characteristics of oil		
	Appearance	Color	Odor
Girolfe oil	Liquide	Yellow	aromatic, spicy, and powerful, with a rising aldehyde odor.

3. Sensory analysis of gelatin

The sensory test results (color, taste, aroma, appearance) for gelatin extracted from chicken feet are presented in the following table:

Table 06: Sensory characteristics of gelatin extracted from chicken feet.

Variables	Criteria
Color	Light yellow
Aspect	Solid Powder (Cristaux)
Smell	No smell

The gelatin extracted from the chicken feet was an excellent crystal powder, light yellow and without smell.

We note that the properties of the gelatin extracted from chicken feet are entirely identical to the gelatine sold on the market.

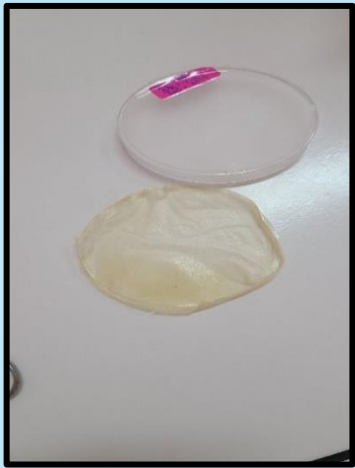


Note: Our result is the best that was found last year.

4. Sensory characteristics of biofilm

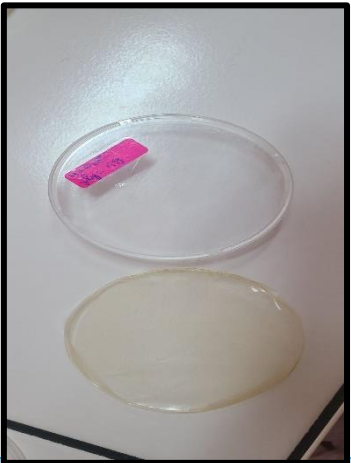


The sensory characteristics of different developed biofilms are presented in Table 03 below:

III. Results and Discussion


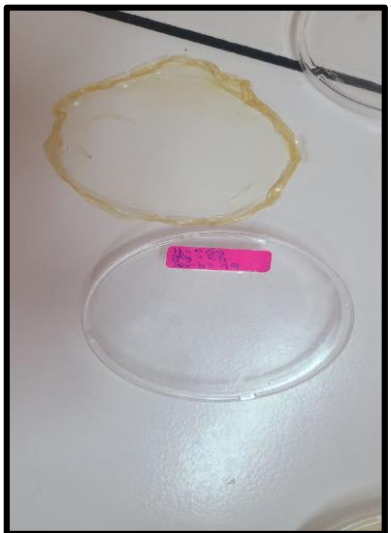
Table 07: Sensory characteristics of gelatin-based biofilm.

Samples	Color	Smell	Texture	Surface	Figure of samples
Sam 1:	Light yellow	Smell of Oil of clove	Elastique	Smooth	
Samp 2:	Light yellow	Smell of Oil of clove	Elastique	Smooth	
Sam 3:	Transparent	Smell of Oil of clove	Very Elastique	Smooth	

III. Results and Discussion

Sam 4:	Light Yellow	Smell of Oil of clove	Elastique	Smooth	
Sam 5:	Transparent	Smell of Oil of clove	Very Elastique	Smooth	
Sam 6:	Transparent	Smell of Oil of clove	Very Elastique	Smooth	

III. Results and Discussion

Sam 7:	Light yellow	Smell of Oil of clove	Elastique	Smooth	 A photograph showing a petri dish with a light yellow, smooth film spread across its surface. A pink label is visible on the lid of the dish.
Sam 8:	Transparent	Smell of Oil of clove	Very Elastique	Smooth	 A photograph showing a petri dish with a transparent, smooth film spread across its surface. A pink label is visible on the lid of the dish.

The presence or absence of incense essential oil only affects the smell. In its presence, the film smells of Clove of Girofle oil (*Eugenia caryophyllus*). Changing the concentration of oil had no effect on the texture and surface.

III. Results and Discussion

Table 08: physical characteristics of samples.

Samples	Flexibility	Resistance	Transparency	Smell
Samp 1	++	+++	++	+++++
Samp 2	+++++	+++++	+++++	+++
Samp 3	+++++	+++++	++	+++
Samp 4	+++++	+++++	+++++	+++
Samp 5	++	+++	++	+++++
Samp 6	+++++	+++++	+++++	+++++
Samp 7	++	+++	++	+++++
Samp 8	+++++	+++++	+++++	+++

Normal character: ++

Good character: +++

Perfect character: +++++

The film created from sorbitol (samples 3,5,6,8) is clear and transparent, making it ideal for packaging food while allowing for easy visibility. The film is soft and flexible, enabling it to conform to different shapes and sizes of food. It is tear and puncture-resistant, protecting the packaged food.

The food film is made from glycerol (sample 1,2,4,7). It is supple, flexible, and tear-resistant, protecting packaged food well. The film is clear and light yellow, allowing for easy visibility of the packaged food.

Both types of biofilms have similar characteristics, with some notable differences. Sorbitol-based films appear more transparent and flexible, while glycerol-based films are lighter yellow and have normal flexibility. Both types of biofilms provide reasonable protection for packaged foods and contribute to reducing the environmental impact of food packaging.

III. Results and Discussion

5. Physical properties of gelatin film

The physical properties of the developed biofilms are shown in the following table:

Table 09: Physical properties of biofilms developed.

Samples	Solubility %	Opacity g ⁻¹	Thickness μm	Water vapor permeability
Sam 1:	43,72%	8,71.10 ⁻³	14	1,5028
Samp 2:	41,32%	5,91.10 ⁻³	17,25	1,607
Sam 3:	37,67%	8,41.10 ⁻³	14,5	\
Sam 4:	48,78%	9,2.10 ⁻³	12,5	\
Sam 5:	43,70%	7,63.10 ⁻³	15,2	1.283

III. Results and Discussion

Sam 6:	41,19%	$5,26.10^{-3}$	23,75	\
Sam 7:	40,30%	$8,38.10^{-3}$	13	\
Sam 8:	43,72%	$9,02.10^{-3}$	11,75	1,5364

6. Film solubility in water

Water solubility is an essential property of edible films because some food applications may require good insolubility in water to improve product integrity and water resistance (Perez-Gago & Krochta, 1999). As a rule, a higher solubility would indicate a lower water resistance (Bourtoom & Chinnan, 2008).

In the study comparing the solubility of films made from different formulations, eight samples were analyzed for their solubility characteristics. The compositions of the samples varied in terms of gelatin, sorbitol, glycerol, and clove oil content. Results indicated that Sample 4, with gelatin, sorbitol, glycerol, and clove oil, exhibited the highest solubility at 48.78%. This finding suggests that the specific composition of Sample 4 may contribute to enhanced solubility compared to other formulations tested in the study. Samples 1, 2, 3, 4, 5, 6, 7, and 8 showed solubility percentages ranging from 37.67% to 43.72%. These results provide insights into the impact of varying formulation components on film solubility properties.

The solubility of biofilm formulations can be significantly influenced by their compositional components, as demonstrated in the study, which tested different compositions of gelatin, sorbitol, glycerol, and clove oil. Sample 4, which had the highest solubility at 48.78%,

III. Results and Discussion

contained gelatin, sorbitol, glycerol, and clove oil. This result can be analyzed in the context of existing literature to understand the roles of these components in enhancing solubility.

Gelatin is a well-known biopolymer used in film formation due to its excellent film-forming properties and biodegradability. Studies have shown that the protein network formed by gelatin can significantly influence the solubility of the films. For example, research by Arvanitoyannis and Biliaderis (1999) highlighted that gelatin's solubility increases with gelatin's content due to its hydrophilic nature, facilitating water absorption and film dissolution.

Plasticizers like glycerol are crucial in modifying biopolymer films' mechanical properties and solubility. Glycerol, a small molecular weight plasticizer, increases the flexibility and solubility of films by reducing intermolecular forces within the polymer matrix (Sothornvit & Krochta, 2000). The presence of 1g glycerol in Sample 4 likely contributed to its higher solubility, as glycerol can enhance the water permeability of films, making them more soluble.

Sorbitol is another plasticizer often used in biofilm formulations. However, your study found that the absence of sorbitol in Sample 4 resulted in the highest solubility. This is supported by the findings of Matta Fakhouri & al, (2012), which suggested that while sorbitol improves the mechanical properties of films, its impact on solubility is less pronounced than glycerol. Therefore, the absence of sorbitol in Sample 4 could have prevented the reduction in solubility often associated with its presence.

The clove of Girofle oil (*Eugenia caryophyllus*), included at 0.5g in Sample 4, is known for its antimicrobial properties and potential impact on film solubility. While essential oils can sometimes decrease the solubility of films due to their hydrophobic nature, the specific combination and proportion in Sample 4 might have worked synergistically with gelatin and glycerol to enhance solubility. A study by Azeredo & al, (2016) on incorporating essential oils into biopolymer films indicated that the interaction between essential oils and other components is complex and can vary widely.

Our findings highlighted the importance of the specific formulation components in determining the solubility of biofilms. The high solubility of Sample 4 suggests a favorable interaction between gelatin and glycerol, which is supported by the literature on biopolymer film formulations.

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7. Opacity:

Various research has been conducted to improve film and edible coatings' mechanical properties and barriers. However, few studies have focused on optical properties such as color, brightness, and transparency. Optical properties are surface characteristics generally detected by human vision, thereby affecting some crucial aspects of the organoleptic quality of food (Brindle & Krochta, 2008). The opacity values of the biofilms developed are presented in table 11. The film microstructure, both internal and surface, plays a vital role in the optical properties of the film. The study of film opacity is critical and sometimes determines the dietary application of mixed films. In addition, it can also be seen as an effective means of indicating the structural aspect of a mixture. Indeed, a translucent aspect has been attributed to a less viscous phase forming a continuous matrix (Brindle & Krochta, 2008). Yoo & al, (2011) showed that whey protein films are more transparent than polysaccharide or protein-polysaccharide films. The films obtained from both formulations are homogeneous and transparent.

In this study comparing the opacity of films made from different formulations, eight samples were analyzed for their opacity characteristics. The compositions of the samples varied in terms of gelatin, sorbitol, glycerol, and clove oil content. Results indicated that Sample 8,4, had the highest results at $9,02.10^3$

8. Water vapor permeability

Water vapor permeability (WVP) measures how easily water vapor enters a material. It also measures the ability of materials to transfer water between the product and its environment. Our film Sample 5, formed at 1.283, has very low WVP values. The other three samples, with different compositions, resulted in between (1,5028 and 1,607).

Water vapor permeability measures how easily water vapor enters a material. Water vapor permeability measurement (WVP) measures the ability of materials to transfer water between the product and its environment. The main function of food packaging is often to avoid or at least reduce the transfer of moisture between the food and the surrounding atmosphere or between two components of a heterogeneous food product; the water vapor permeability of the films should be as low as possible (Gontard & al, 1992).

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Water vapor permeability (WVP) control is essential for the maximum safety and stability of the packaged product throughout the storage and distribution process to avoid moisture transfers that may affect food quality. The surface dehydration of some fresh or frozen foods is curbed by using hydrophilic polysaccharide-based coatings (aqueous gel) that dehydrate before the product and form a protective film (Zuo & al, 2009).

Our biofilm has shown remarkable resistance to water permeability. The presence of essential and sorbitol oil has blocked water vapor from passing, and pure gelatin, which has a compact character, also stops water vapor.

9. Microscopic biofilm analysis

Sample 1: enlargement X 10

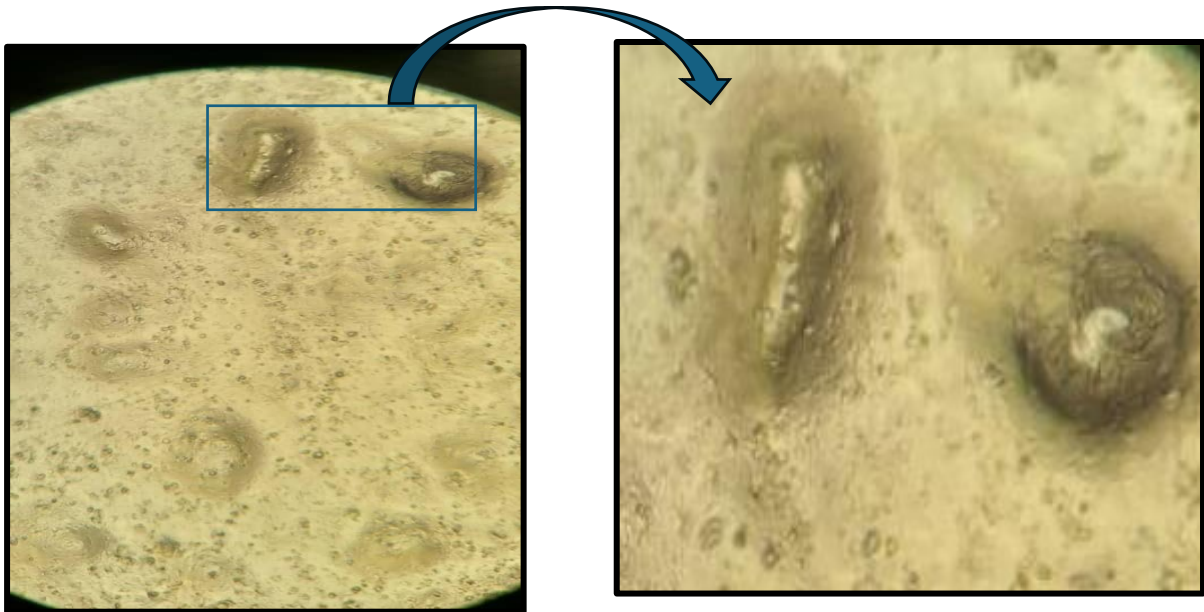


Figure 26: The sample 1 enlargement X 10.

- In the cut, we observe that the sample's surface has an enlarged X10, brown spots indicating the presence of essential oil, and a rough, non-homogeneous surface.

III. Results and Discussion

Sample 1: enlargement X 20.

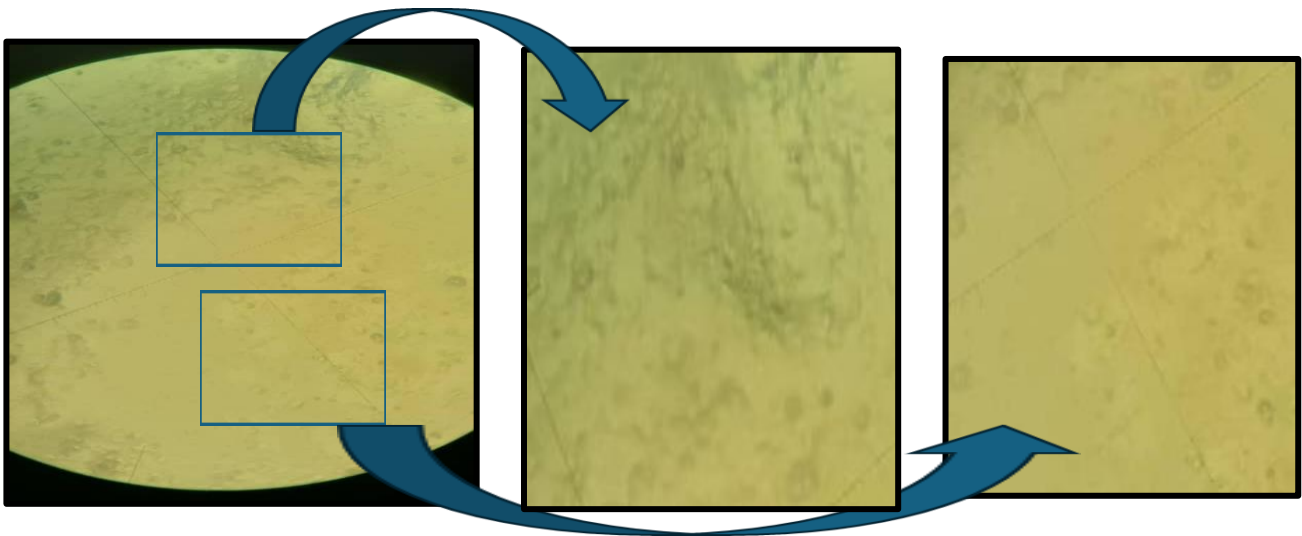


Figure 27: sample 1 enlargement X 20.

- Sample 1 in X20 enlargement in the first selected section. The film appears smooth and a little homogeneous, and in the second section, the surface is rough and heterogeneous. Although the samples are taken from the same section, they differ because of their position inside the etuve.

Sample 02 : enlargement X 10

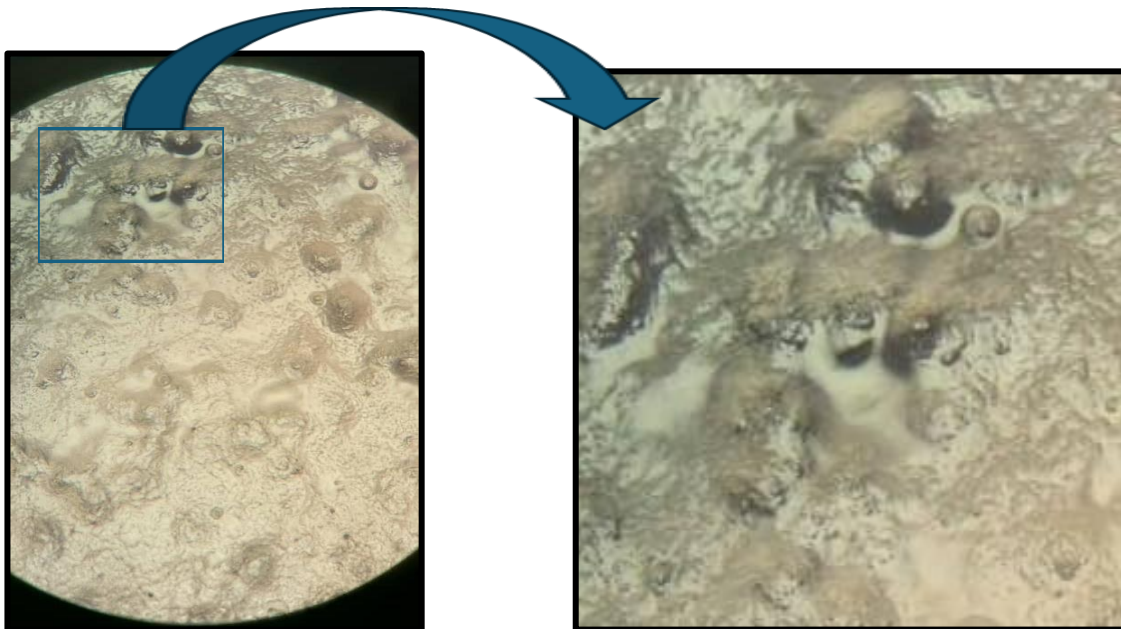


Figure 28 : sample 2 enlargement X 10.

- We observe that in the cut, the surface of the sample has an enlargement of X10 of brown spots, indicating the presence or excess essential oil, as well as a rough and No-homogeneous surface.

III. Results and Discussion

Sample 02: enlargement X 20

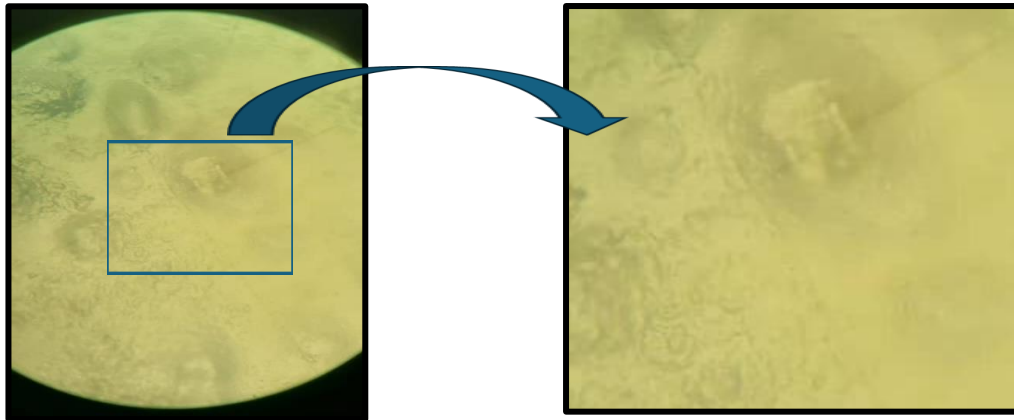


Figure 29: Sample 2 in enlargement X20.

- Sample 2 was analysed at X20 enlargement, and its surface appears rough and non-homogeneous.

Sample 03: enlargement X 10

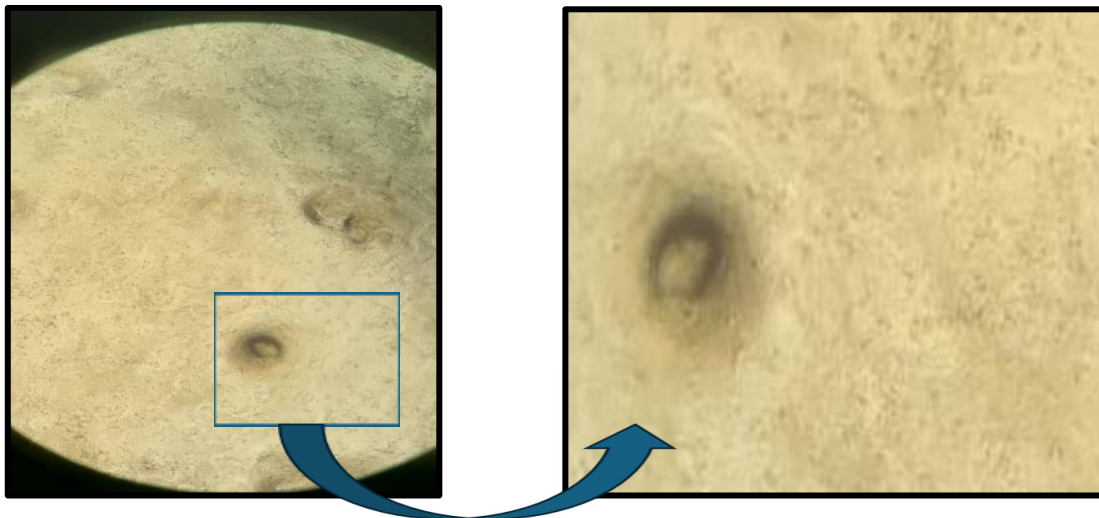


Figure 30: Sample 2 enlargement X10.

- In the cut, we observe that the sample's surface has an enlarged X10, brown spots indicating the presence of essential oil, and a rough, non-homogeneous surface.

III. Results and Discussion

Sample 03: enlargement X 20.

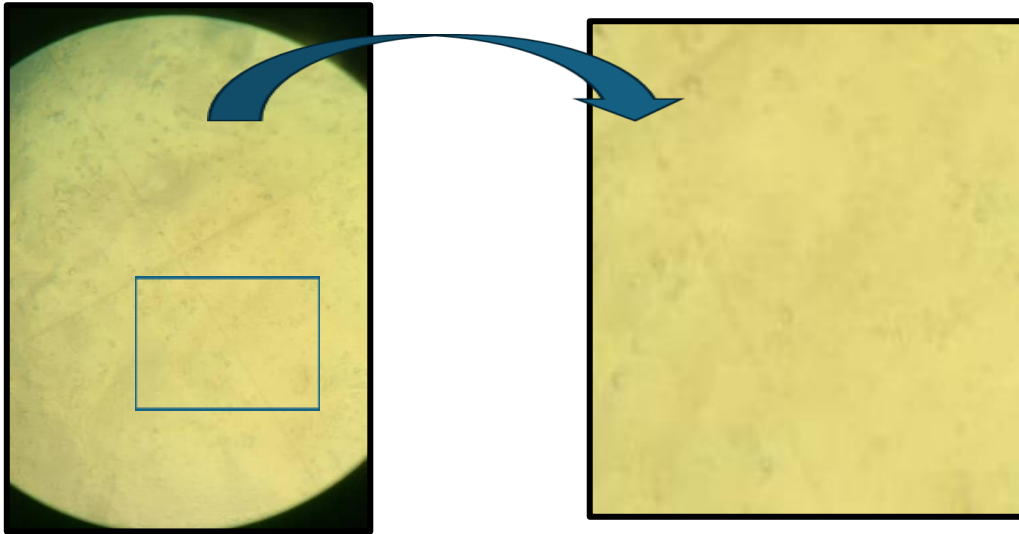


Figure 31: Sample 3 in enlargement X20.

- We notice a homogeneous and smooth surface in this cut that appears in the x20 magnification.

Sample 04: enlargement X10.

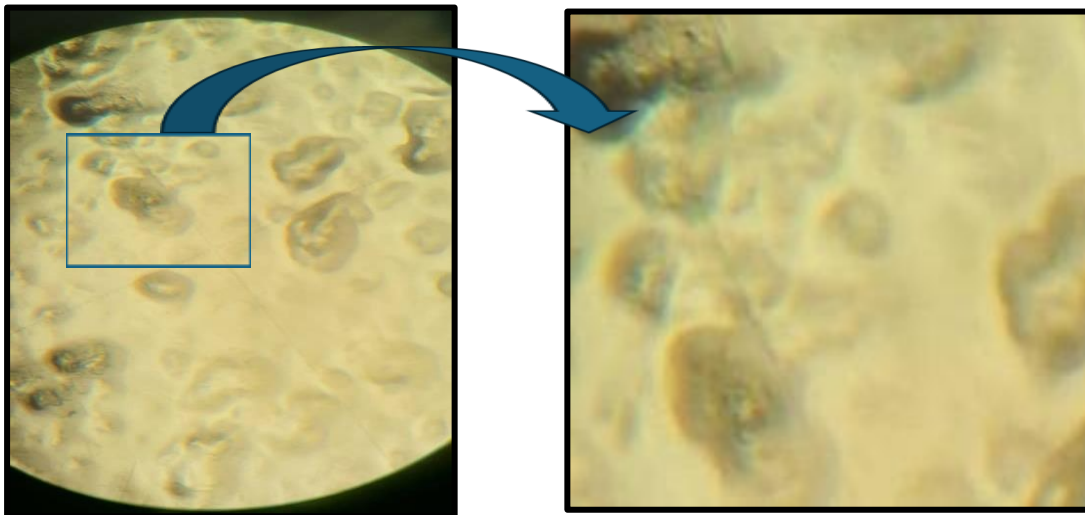


Figure 32: Sample 4 in enlargement X10.

- In this cut, which appears in the figure with an enlarged X10, we notice stains, indicating the existence of essential oil and sorbitol on a non-homogeneous surface.

III. Results and Discussion

Sample 04: enlargement X 20.

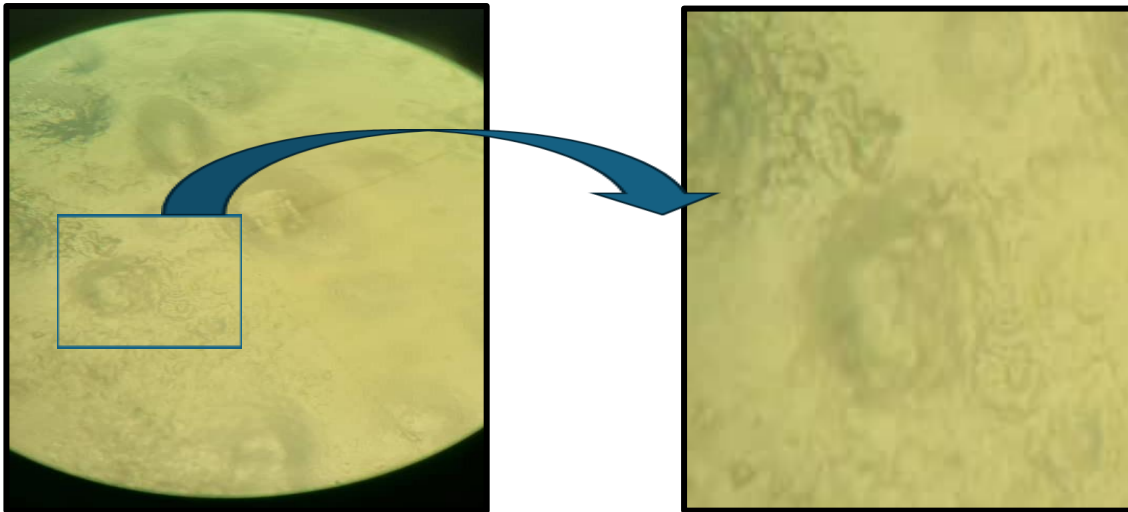


Figure 33: Sample 4 in enlargement X 20.

- Sample 4 in enlargement X20. We observe that the film appears smooth and has little homogeneity.

Sample 05: enlargement X10.

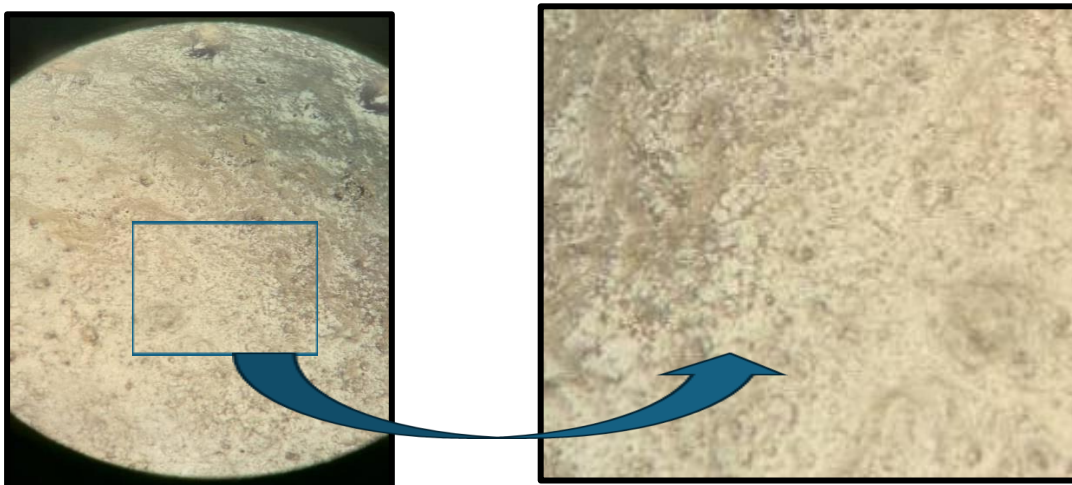


Figure 34: sample 5 in enlargement X10.

- Sample 5 was analysed at X10. En enlargement appears to show a heterogeneous and non-homogenous surface.

III. Results and Discussion

Sample 05: enlargement X20.

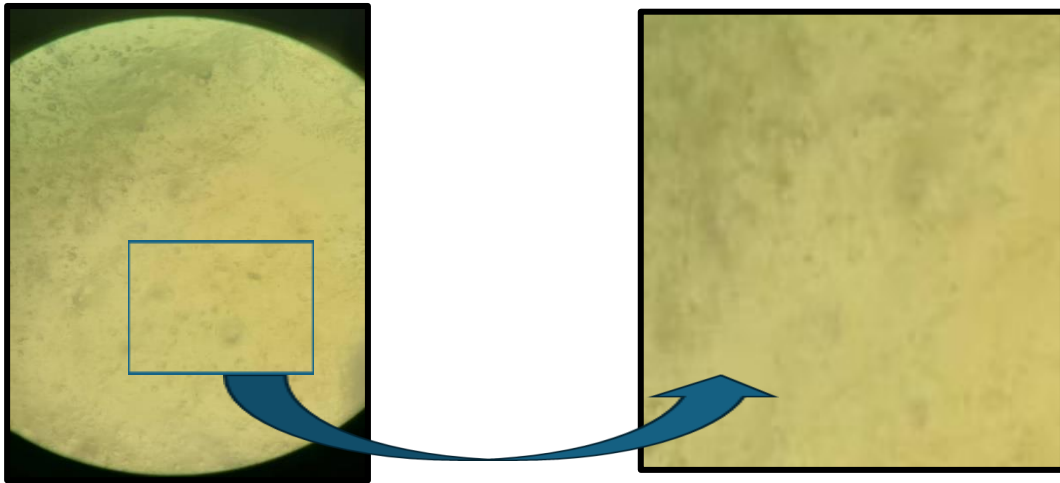


Figure 35: Sample 5 in enlargement X20.

- Sample 5 for enlargement X20. We observe that the film appears smooth and has little homogeneity,

Sample 06: enlargement X10.

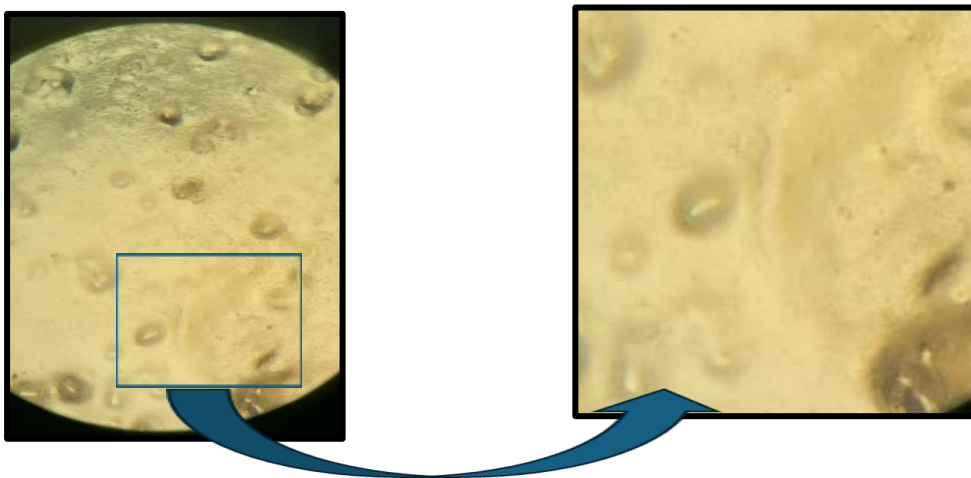


Figure 36: Sample 6 in enlargement X10.

- In this cut, which appears in the figure with an enlarged X10, we notice stains, indicating the existence of essential oil and sorbitol on a non-homogeneous surface.

III. Results and Discussion

Sample 06: enlargement X20.

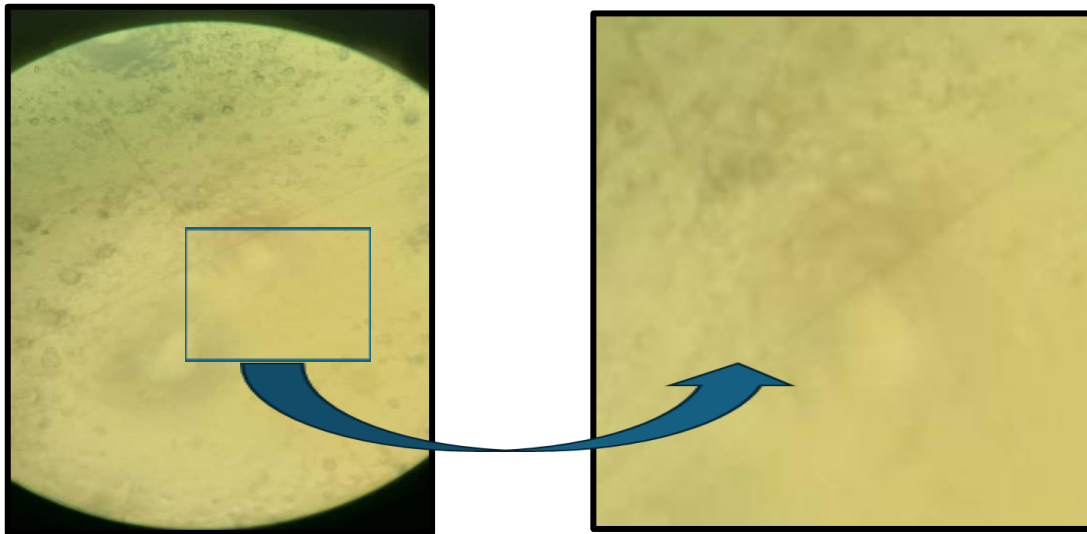


Figure 37: Sample 6 in enlargement X20.

- Sample 6 in enlargement X20. We noticed that the film appears smooth and has little homogeneity.

Sample 07: enlargement X10.

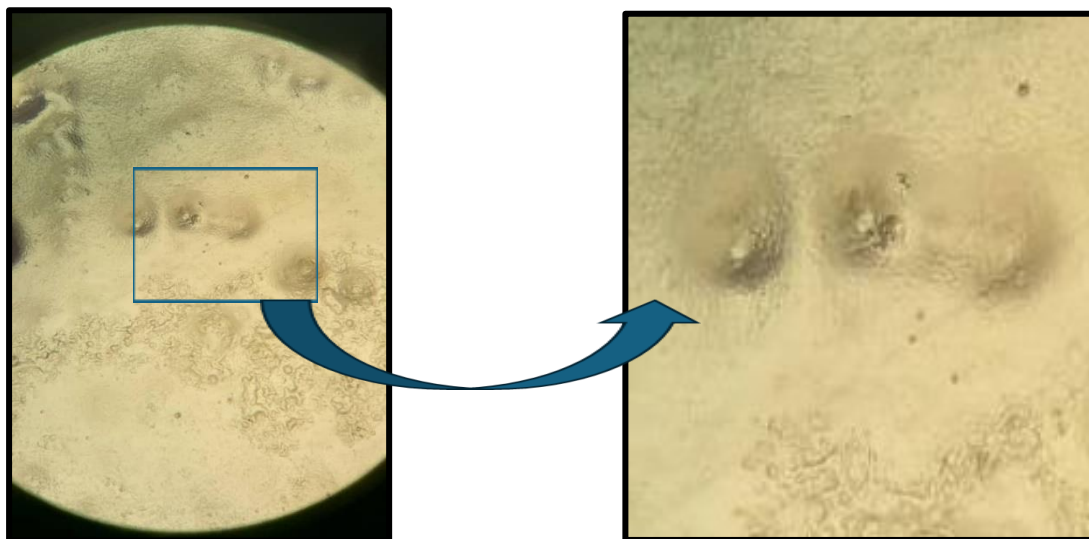


Figure 38: Sample 7 in enlargement X10.

- In the cut, we observe that the sample's surface has an enlarged X10, brown spots indicating the presence of essential oil, and a rough, non-homogeneous surface.

III. Results and Discussion

Sample 07: enlargement X20.

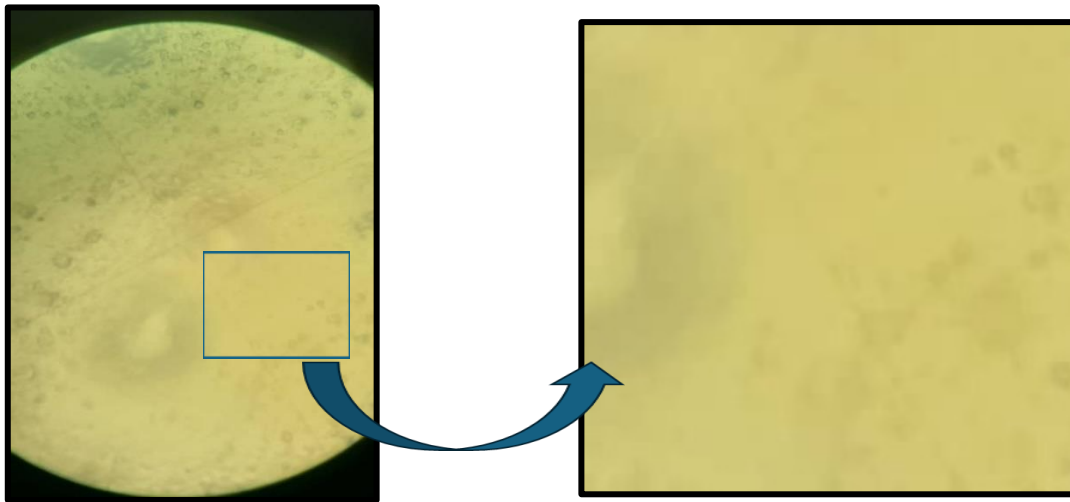


Figure 39: Sample 7 in enlargement X10.

- Sample 7 in enlargement X20. We noticed that the film appears smooth and has little homogeneity.

Sample 08: enlargement X10.

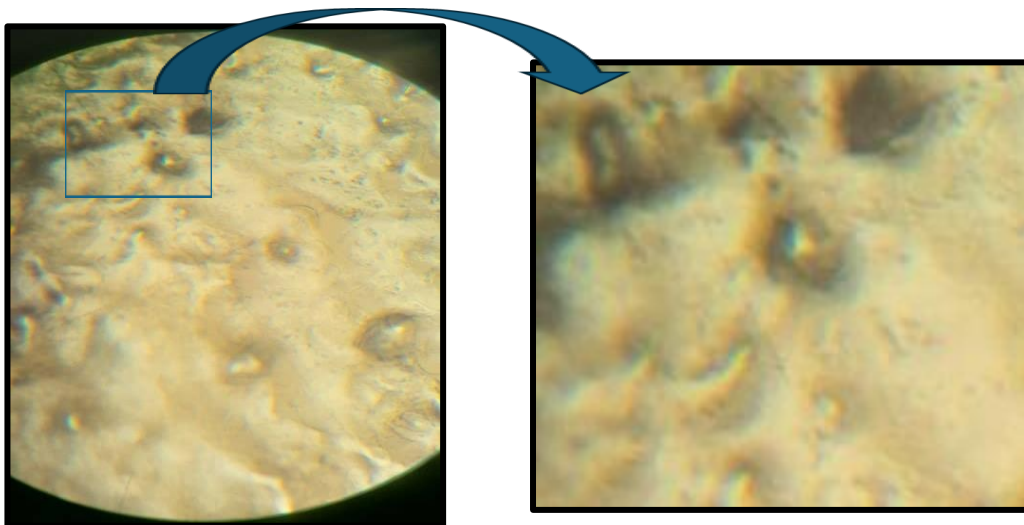


Figure 40: Sample of 8 enlargement X10.

- In this cut, which appears in the figure with an enlarged X10, we notice stains, indicating the existence of essential oil and sorbitol on a non-homogeneous surface.

Sample 08: enlargement X20.

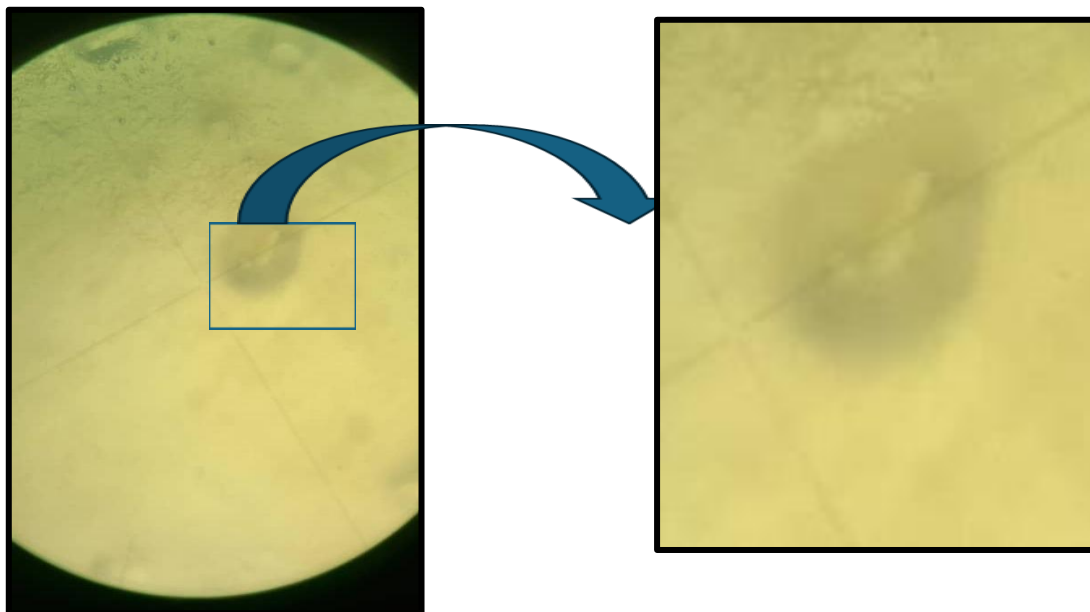


Figure 41: Sample 8 in enlargement X20.

- In the cut, we observe that the sample's surface has an enlarged X20, brown spots indicating the presence of essential oil and sorbitol, and a rough, non-homogeneous surface.

10. After the microscopic study:

Our films were prepared with gelatin, essential oil, sorbitol, and glycerol. Their surface was not homogeneous and smooth, and brown spots under the microscope.

The study by Ahmad & al, (2016) shows that collagen solubilized in acid presented a compact, smooth surface without stratification and cracking, which indicates an orderly film matrix. The film's better mechanical properties accompany this. However, the pepsin-solubilized collagen film had a slightly irregular, rougher surface and a micro-fibrous structure (characteristic of collagen fibrils).

11. Biodegradability of films

The biodegradability of biomaterials may be affected by weather conditions, moisture content, the type of microorganisms in the soil, and the characteristics of the biofilm (such as humidity, density, and the presence of plant bioactive substances) (Nguyen & al, 2016). According to figure (43), the film formulated in this study is ecologically harmless and biodegrades easily and quickly.

III. Results and Discussion

Gelatin-based biofilms are renowned for their excellent biodegradability, a key attribute that makes them suitable for various environmental and biomedical applications. Gelatin, derived from collagen, is a natural polymer that undergoes biodegradation through enzymatic activity and microbial processes in soil and aquatic environments (Song & al, 2018). The biodegradability of gelatin films is largely influenced by their protein structure, which is susceptible to enzymatic hydrolysis by proteases. Studies have demonstrated that gelatin films can degrade completely within a few weeks when exposed to composting conditions or soil, releasing non-toxic byproducts that can be assimilated into the ecosystem (Jongjareonrak & al, 2006). This rapid degradation is advantageous over synthetic polymers, which persist in the environment for extended periods and contribute to pollution. Furthermore, the biodegradability of gelatin can be tailored by modifying its formulation, such as by incorporating other biopolymers or additives, without significantly compromising its environmental benefits (Gómez-Guillén & al, 2009). Therefore, the inherent biodegradability of gelatin-based biofilms underscores their potential as sustainable alternatives in packaging, agriculture, and medical fields. Both types of biofilms have similar characteristics; they can be designed to be biodegradable, thus helping to reduce the environmental impact of food packaging.



Figure 42: Total Biodegradation of Films (personal photo).

12. Sensory analyses:

The following table shows the sensory analysis of packed cherries unpacked (test) of storage outdoors at room temperature (17-20 °C). Observed appearance, texture, brightness, and color changes are considered evaluation criteria.

III. Results and Discussion

Table 10: Sensory criteria of sample Negative test or fresh cherry.

visual spectrum: (fresh fruit before the packaging application) (figure....)	
Color	Red
State	Fresh
Smooth	Shiny
Texture	Hard
Shape	Irregular sphere
Olfactive	
Smell	No smell
Gustative	
Flavor	No flavor
Taste	sweet and fruity



Figure 43: Fresh cherry tomato (personal photo).

III. Results and Discussion

13. The results of the biofilm application test are presented in the figures.

The sample (test) without packaging showed visual deterioration after five days with complete rot, bad smell, and a radical change of color (red to dark red), but the sample packed with sample 4 and sample 3 , are the same change, but the samples 1 and sampling them with a Film of sample 2(preserved a structural appearance and no visual degradation). This test shows that the biofilm developed from the presence of sorbitol is more effective in preserving cherry tomatoes.

Sample 01:



Figure 44: cherry tomato 1st day of application biofilm. (personal photo).

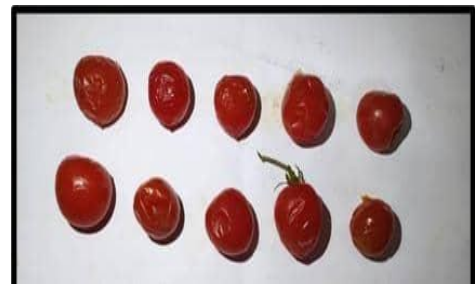


Figure 45: cherry tomato after the 6th day of biofilm application.

III. Results and Discussion

Sample 2:

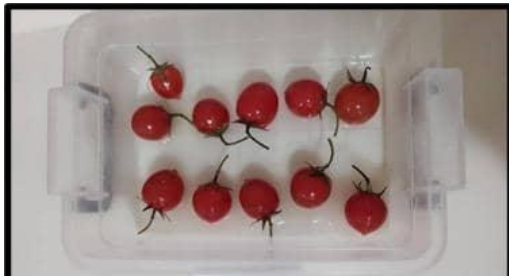


Figure 46: Cherry tomato 1st day of application of biofilm(personal photo).

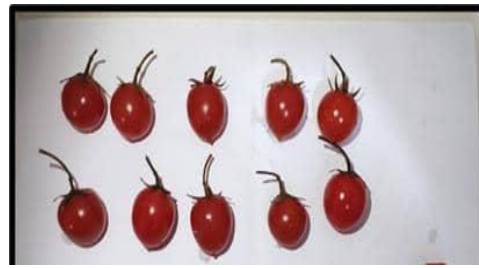


Figure 47: Cherry tomato after the 6th day of biofilm application.

Sample 3:



Figure 48: Cherry tomato 1st day of application of biofilm (personal photo).



Figure 49: Cherry tomato after the 6th day of biofilm application.

III. Results and Discussion

Sample 4:

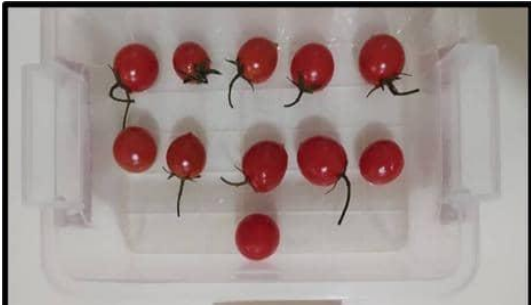


Figure 50: Cherry tomato 1st day of application of biofilm (personal photo).



Figure 51: Cherry tomato after the 6th day of biofilm application.



Figure 52: Cherry tomato after the 10th day of application of biofilm (personal photo).

The following table shows the sensory analysis of packed cherry tomatoes unpacked (test) after 10 days of storage outdoors at room temperature (17-20 °C). Observed appearance, texture, brightness, and color changes are considered evaluation criteria.

III. Results and Discussion

Table 11: The sensory analysis of packed cherry tomatoes unpacked after 10 days.

Samples	appearance	Color	Smell	Texture	Conservability
Sample1	+++	+++	++++	+++	+++
Sample 2	+++++	+++++	+++++	+++++	+++++
Sample 3	++	++	++	++	++
Sample 4	+	+	+	+	+

Bad character: +

Normal character: ++

Good character: +++

Perfect character: +++++

Conclusion

Conclusion

In conclusion, biodegradable packaging is a promising alternative to conventional food storage materials. Although few studies are available on using biopolymers for food packaging applications, continued growth in the bioplastics market is expected.

First, our work focused on the durability and feasibility tests of a new organic food packaging based on gelatin, glycerol, and sorbitol. The gelatin was extracted from chicken feet using acetic acid. Different combinations of these biopolymers and essential oils have been tested.

Secondly, our study provides physical analyses of biodegradable packaging materials, such as opacity, permeability, thickness and solubility.

Finally, our work involved the microscopic study of the surface of the active biofilm prepared by the gelatin extracted from chicken feet.

The yield of gelatin extracted from 100 g of chicken legs was estimated at 11.22%. The gelatin extracted from the chicken feet was an excellent crystal powder, light yellow and without smell.

Both types of biofilms have similar characteristics, with some notable differences. Sorbitol-based films appear more transparent and flexible, while glycerol-based films are lighter yellow and have normal flexibility. Both types of biofilms provide reasonable protection for packaged foods and contribute to reducing the environmental impact of food packaging.

The solubility of biofilm formulations can be significantly influenced by their compositional components. Sample 4, which had the highest solubility at 48.78%,.

The opacity of films made from different formulations; eight samples were analyzed for their opacity characteristics. The compositions of the samples varied in terms of gelatin, sorbitol, glycerol, and clove oil content. Results indicated that Sample 8,4 had the highest results ($9,02 \cdot 10^{-3}$).

Our film of Sample 5, had fewer results of Water vapor permeability (1,283).

The biodegradability of biomaterials in the film formulated in this study is ecologically harmless and biodegrades easily and quickly.

Our work involved the microscopic study of the surface of the active biofilm prepared by the gelatin extracted from chicken legs, which shows the heterogeneous biofilm surface.

Conclusion

After the conservation test, the tomato cherries remained in good condition for 10 days. The presence of sorbitol is more effective in preserving cherry tomatoes.

In perspective, it will be interesting to study the physicochemical properties and the microstructure, study the microscopy of the films obtained, and perform more application tests on the different foods to improve them.

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