



Green Biopolymer-Based Edible Coatings for Quality Retention and Extended Shelf Life of Table Eggs

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Abstract

This study aimed to evaluate the effectiveness of edible films and coatings formulated using Chitosan and whey protein in improving the internal quality, shelf life, and microbial safety of chicken eggs during storage. New eggs were divided into chitosan-coated, whey protein coated, combined Chitosan whey protein, and uncoated control groups, immersed in their respective solutions, dried, and stored at room temperature for 28 days. Egg quality parameters, including Haugh unit, albumen height, and weight loss, were recorded weekly, and microbial contamination was measured using standard plate counts. Egg-coated showed considerably greater Haugh units, lesser weight loss, and lower microbial loads compared to uncoated ones. Chitosan and combined chitosan/ whey protein coatings were found to have the best preservative properties because they have better properties regarding the barrier and inherent antimicrobial activity, which maintains albumen quality and prolongs the shelf life. The findings verify the fact that biodegradable chitosan and whey protein films provide an eco-friendly alternative to conventional egg preservation. Simultaneously, progress in the research on animal vocalizations has improved conservation efforts by ensuring accurate biodiversity tracking and preventing ecological disturbances at an early stage. Sound-related ecological measurements, further enriched by the acoustic recognition systems (AI-based), enable automated detection of species, evaluation of the quality of habitat, and the detection of degradation of ecosystem soundscapes introduced by pollution. These are technologies that are commonly applied in wildlife conservation that demonstrate the wider scope of applying biosensing and machine learning techniques to the study of food preservation, offering new avenues of real-time tracking of food spoilage, microbial activities, and environmental changes.

Keywords:

Chitosan, edible films and coatings, eggs, haugh unit, microbial contamination, whey protein.

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Introduction

Eggs are readily eaten as they are affordable and nutritious, containing all the necessary proteins, vitamins, antioxidants, carotenoids, and phospholipids. Nevertheless, eggs start to degrade as soon as they are laid, and environmental conditions, including temperature, humidity, air flow, and handling, impact the quality of eggs and their safety. Refrigeration is commonly practiced across cultures because it preserves nutrients and suppresses microbial growth. The gap in this level is that despite the knowledge of consumers and industries of the perishability of eggs, there is still a lack of efficient and sustainable preservation systems to avoid microbial contamination and preserve internal quality. Increasing environmental awareness has brought about the increasing substitution of petroleum-based plastics with cheaper, renewable, and biodegradable biomaterials. Policies obtained in the form of lignocellulosic biomass of residual wood, plant waste, and aquatic organisms are polysaccharides, proteins, and lipids and have proven applicable in areas such as food packaging, cosmetics, and healthcare (Kamarudin et al., 2023). High-performance packaging is currently under investigation with polysaccharides like cellulose, chitin, Chitosan, starch, and gums and proteins like whey protein isolates (Salih et al., 2024; Surendar, 2024). The gap here is that although these natural biopolymers are well studied, their comparative advantages and blended effects in egg preservation systems remain unexplored.

Edible coatings such as chitosan and whey protein isolates offer environmentally friendly alternatives to plastic packaging. Chitosan is antimicrobial, biodegradable, and derived from chitin, whereas whey protein isolates form films that improve barrier properties but are moisture-sensitive; therefore, plasticizers, surfactants, and sensory enhancers are often added to improve coating performance (Ziwei et al., 2023; Saniya et al., 2025; Pires et al., 2024). To prevent changes in viscosity and microbial degradation, chitosan solutions should be kept at specific temperatures (4–10 °C), in darkness, and at a pH of 4.5 - 6.0. The remaining loophole in this field is that although these biopolymers have been extensively used, little research has been done to analyze the coating under actual storage conditions using standardized formulation and application protocols. Eggshells are porous and susceptible to contamination by *Staphylococcus aureus*, *Salmonella spp.*, *Listeria monocytogenes*, *Streptococcus spp.*, *Bacillus spp.*, and *Escherichia coli* due to contact with feces, soil, nesting materials, pests, and poor handling (Hinson et al., 2025; Chousalkar et al., 2021; Desai & Joshi, 2023). Moisture and carbon dioxide can pass through shell pores, reducing internal egg density and accelerating quality loss. Edible coatings reduce these exchanges and maintain Haugh unit, albumen stability, and structural integrity (Caner et al., 2022; da Silva Piresl et al., 2020). The scientific gap in this case is that earlier research has mostly tested coatings singly, but has not studied the possibility of blended formulations of a specific kind, such as a chitosan blend with whey protein, that can have synergistic protective effects.

The objective of the present study is to evaluate the preservation efficacy of Chitosan, whey protein, and their combined formulation on internal egg quality, microbial safety, and shelf life during storage. The aim is to establish whether a biopolymer coating blend could perform better than the individual coating in terms of assuring quality indicators, with a contribution to environmental sustainability. The gap within current objectives is that existing research does not sufficiently integrate eco-friendly preservation, materials science, microbial safety, and technological innovation into a single comparative framework. The central hypothesis is that chitosan and whey protein coatings will each reduce microbial contamination and maintain egg quality better than uncoated eggs, and that their combination will have a synergistic effect, producing superior barrier properties and extended shelf life. A secondary hypothesis is that integrating emerging biotechnological

approaches such as acoustic communication, acoustic biosensors, and machine-learning-based ecological monitoring can enhance conservation efforts and also offer conceptual insights for developing real-time food safety monitoring systems. The knowledge gap that can be bridged with the help of the proposed hypothesis is that the relationship between the research on food preservation and sound-based environmental biotechnology has not been studied before.

The main contributions of the article are the first large-scale comparison of Chitosan, whey protein, and their mixture in terms of storing eggs under controlled conditions; the demonstration of the synergistic character of the combination of biopolymers; and the availability of an effective preservation method based on biodegradable polymers, without the use of petroleum-based packaging. In this study, there is also a connection between food safety and environmental conservation, whereby the edible coating technologies are related to acoustic monitoring tools that are currently prevalent in the assessment of biodiversity and ecosystem monitoring. This theoretical synthesis focuses on the role of biosensors and machine-learning sound interpretation in early warning of spoilage, microbial contamination, and environmental instability, and fills a last gap in the literature: the absence of interdisciplinary models that integrate sustainable materials, food security, and biotechnology-based ecosystem surveillance.

Literature Survey

The consumption of eggs is promoted by their low cost, nutritional value, and variety in cooking. Nonetheless, the quality of eggs starts to decrease the moment they are laid because of natural aging factors that depend on the environmental component, including temperature, humidity, and handling conditions. Refrigeration is a common practice in most cultures in order to reduce the growth of microbes and maintain freshness, but eggshell contamination is a major health issue in the community. Eggs may get contaminated by feces, soil, nesting material, pests, rodents, and unsanitary storing conditions, exposing them to pathogens such as *Staphylococcus aureus*, *Salmonella spp.*, *Listeria monocytogenes*, *Bacillus spp.*, *Streptococcus spp.*, and *Escherichia coli* (Hinson et al., 2025; Chousalkar et al., 2021; Desai & Joshi, 2023). This contamination brings out the use of superior sustainable preservation methods, which can guarantee the integrity of microbes and nutritional value. Eggshells are porous in nature; thus, diffusion of gases like moisture and carbon dioxide can take place. This constant exchange increases the rate at which the internal quality is lost, causing weight loss, a decrease in density, and low permanence of alpha-lactate globules with time. The traditional packaging materials consist of simple physical protection, which does not stop the entry of microbes or the diffusion of gases. Moreover, the extensive usage of petroleum-based plastics causes environmental pollution in the long run, enhancing the necessity of using biodegradable and renewable materials in the world. Consequently, a significant interest has turned towards the biopolymers obtained through lignocellulosic biomass, such as agricultural residue, woody biomass, and aquatic resources that contain polysaccharides, proteins, and lipids that can be applied to the food, cosmetic, pharmaceutical, and energy industries (Kamarudin et al., 2023). In particular, polysaccharides such as cellulose, starch, chitin, Chitosan, and gums can be appreciated because of their abundance, biodegradability, and predisposition to the packaging and edible covers (Salih et al., 2024; Surendar, 2024).

Plant-based polymers, natural fibers, and agricultural waste biodegradable materials provide a solution to sustainable packaging because they help decrease fossil fuel consumption and plastic pollution. In this context, edible coatings are also an exciting new option in the extension of the shelf life of eggs and their safety. Chitosan is a crustacean chitin derivative, antimicrobial polysaccharide that forms a film, which has been widely used in reducing moisture, limiting gaseous exchange, and sealing eggshell holes and inhibiting microbial growth, and is a natural and environmentally friendly substitute of synthetic preservatives (Caner et

al., 2022). The secret of successful performance of chitosan coating is that control of temperature, pH, and the preserving environment should be done to make sure that the structure is not influenced and the antimicrobial effects of the coating are not destroyed.

Edible coatings. One more option is to use whey protein isolates, which are dairy products processed into an edible coating; such isolates have a great film-forming potential, but they cannot be used independently because of moisture sensitivity. Research indicates that with the addition of whey protein to Chitosan, the hybrid films prove to have enhanced mechanical strength, moisture barrier, and improvements in antimicrobial properties, as revealed in earlier studies on the preservation of fruit products (Pires et al., 2024). Plasticizers, fatty acids, surfactants, antioxidants, and antimicrobials are the average supplements that are employed to increase the flexibility and sensory acceptance and protection of the coating (Ziwei et al., 2023; Saniya et al., 2025). Additionally, traditional and emerging egg preservation techniques, including wax coating, mineral oil treatment, CO₂ flushing, and biopolymer-based edible coatings, have shown substantial improvements in controlling spoilage and extending egg freshness (da Silva Pires et al., 2020). Chitosan and whey protein formulations are some of these strategies that have the ability to be high-potential and environmentally-friendly solutions that are consistent with the global sustainability agenda. Acoustic communication and sound-based ecological surveillance have been central in conservation biology in recent years as novel means of evaluating environmental quality and biodiversity. Bioacoustics allows a researcher to identify the presence of the species, identify the behavior of the wildlife, and assess the ecological changes using the passive record of the sounds. Machine learning and artificial intelligence (AI) have extended these abilities to such an extent that species are automatically detected, noise is identified, and ecological disturbances are detected (Stowell et al., 2019; Gibb et al., 2023). These systems are currently common in the monitoring of bird populations, amphibian behavior, insect behavior, and mammal behavior to provide information about the health and condition of the ecosystem, habitat fragmentation, and biodiversity loss.

Moreover, biosensing technologies based on sound are coming out as effective in the management of environmental pollution. Acoustic sensors integrated with AI algorithms can detect anomalies in aquatic ecosystems, track industrial noise pollution, and assess changes in vegetation structure by analyzing soundscapes. They have been utilized to track the freshwater pollution, deforestation, and species communication changes caused by climate change (Farina & James, 2016; Kahl & Clink, 2020). The biotechnological opportunities of sound-based monitoring through machine learning show the power of using interdisciplinary conservation science. These acoustic technologies may be considered as methods of environmental biology; however, the way they work provides conceptual applicability to the food preservation systems. Combining the concept of smart biosensors, acoustic monitoring of microbial activity, and AI-based monitoring might offer real-time data on the process of spoilage of eggs, microbial contamination, and variations in the environment during storage. This interdisciplinary approach assists in the creation of new and environmentally friendly preservation systems, which integrate biopolymer-based coating, along with state-of-the-art sensors.

Materials and Working Methods

Preparation of Chitosan Coating Solution

Chitosan is a biopolymer that is mainly employed in the food industry because of the distinct physical, chemical, and biological characteristics of the material, which is based on oligosaccharides. This is due to its solubility in dilute acidic media, viscosity, and its ability to combine with electrolytic solutions to form stable edible coatings.

In order to obtain the chitosan coating, 3 g of Chitosan was dissolved in 100 mL of distilled water and stirred with a magnetic stirrer. This was followed by the addition of 1 percent acetic acid in small portions in order to facilitate dissolution. A plasticizer, 0.25 mL of glycerol per 21 g of Chitosan, was incorporated to improve film flexibility. The mixture was stirred at a low temperature for 60 minutes until a uniform coating solution was obtained.

Preparation of Whey Protein Coating Solution

In casein coagulation, a high amount of liquid waste is produced in the dairy industry. When cheese is produced, casein is the main milk protein that is coagulated, and the rest of the protein that is left behind and water-soluble is called dairy whey (DW), which is a yellowish-green byproduct. DW can be obtained from milk of different animals like cows, goat, sheep, and even camels. In Western countries, bovine whey is the most common type that is used. Whey protein (WP) is composed mostly of β -lactoglobulin (β -Lg), which represents almost 50% of the overall protein content. α -lactalbumin (α -La) follows with a contribution of about 20%, glycomacropeptide (GMP) comes next with 15%, immunoglobulin (Ig) has a share of 10%, and bovine serum albumin (BSA) holds the smallest share of 8%. WP still contains fractions with very low abundance, around 10%, such as lysozyme (LZ), osteopontin (OPN), proteose peptones (PP), lactoferrin (LF), and lactoperoxidase (LP) (El-Aidie & Khalifa, 2024; Cortés-Ramírez et al., 2024). The characteristics of the cheese made (type and method of production), the coagulation method (enzymatic or acidic), and the milk source (bovine, ovine, or caprine), animal feeding schedules, and lactation stage are all factors that can contribute to significant variations in whey protein yield. These changes dramatically influence the solubility and bioavailability of the proteins and other beneficial substances. Whey proteins have many different kinds of disulfide bonds. At the secondary, tertiary, or quaternary levels, they are made up of small globular molecules. They are vulnerable to several factors, including heat and dephosphorylation. β -lactoglobulin (β -Lg) is a tiny globular protein that has 162 amino acids. Its secondary and tertiary structures are unique. There are five cysteine residues in this protein, and four of them form two disulfide (S–S) links. Cys 121, the fifth cysteine, lies in the center of the natural β -Lg structure. It possesses a free thiol group, which is necessary for making films. β -Lg is the main portion of whey and is particularly significant for the gel formation and aggregation of whey protein compositions. The second most common protein in whey is α -lactalbumin (α -La). It is a small, acidic protein that is kept together by four disulfide linkages and calcium ions. These links retain their tertiary structure and make it stable against denaturation. Battling against denaturation. The main protein part of bovine serum albumin (BSA) has an alpha-helical structure and has one free thiol group and 17 disulfide bonds. Whey proteins provide a number of benefits for human health. Some people name them "fast" proteins because they go quickly from the stomach to the small intestine. This is because β -Lg has a lot of branched-chain amino acids (BCAAs), such as isoleucine, leucine, and valine. Whey protein offers several benefits that other protein sources, such as soy or eggs, don't have. Research with both animals and people has shown that whey proteins and their parts may help people on low-calorie diets keep their lean body mass and improve their heart health. They may also have antioxidant, antibacterial, antiviral, immune-modulating, and anticancer effects. We are making progress on new technologies that can turn a lot of DW into useful goods. One possible technique is to exploit its protein content to make biopolymers that the food industry may employ to make edible coatings and films. DW is a special raw ingredient for making edible biopolymers that are utilized in packaging. Lactose is the main organic part of whey. Lipids, proteins, minerals, and lactic acid come next. Whey proteins (WP) are the most important part of whey in terms of cost and technology. They make up around 15–20% of total milk proteins. The primary part of WP is β -lactoglobulin, which usually has five main parts.

Whey Proteins as Coating Materials

The Improvements in membrane and ion exchange technologies have led to a big increase in the amount of whey protein (WP) made across the world. This is because WP may be extracted with important functional properties, such as acid stability, gelation, film-forming effectiveness, aeration, and emulsification (Kandasamy et al., 2021). There are several kinds of whey that you may buy. There are fresh pasteurized liquids and concentrated variants, such as lactose-reduced whey, demineralized whey, whey protein hydrolyzates, whey protein isolates (WPI), which contain more than 90% protein, and whey protein concentrates (WPC), which have 35–80% protein. The composition and purity levels of protein fractions might differ depending on the methods employed for pretreatment and filtering. Ultrafiltration, diafiltration, electrodialysis, gel filtration, ion-exchange chromatography, and reverse osmosis are all common methods for getting clean water back. Ultrafiltration is used to selectively extract high-molecular-weight components from whey permeate in order to generate WPC. This whey recipe has less cholesterol and fat and more healthy stuff than other whey formulations. WPI is made from WPC using a different ion-exchange chromatography method that gets rid of lactose and minerals. WPI has a lot of protein; however, it doesn't have as many bioactive compounds (Charcosset, 2021). You can dry both WPC and WPI by spraying them with water. These fractions have a lot of pleasant things in their way when it comes to the use of food. They contain few and minimal calories, fats, and salt, are high in protein and amino acids, do not have any infections or harmful metabolites, are biocompatible, safe, and readily available, becoming affordable as an example. Whey Protein Concentrate (WPC) and Whey Protein Isolate (WPI) are the two most used coating agents for concentrated whey products. Whichever the process of producing the film, the resultant films are gel-forming and rearranged in space, containing a large number of film-forming components. The casting procedure is widely used to make WP-based films. This entails putting the film solution on flat surfaces to make a dried gel that may subsequently be used to cover food (Chaudhary et al., 2022). Food items are commonly soaked in the film-forming solution for a certain period of time (30 seconds to 1 minute) before being left to air dry. This ensures that the food surface is completely covered and that the film sticks better and stays intact. Coatings and films made by WP are transparent, colorless, odorless, and flexible. WP is different from other film-forming polymers because it possesses amphiphilic properties, a specific shape, the ability to denature, and electrostatic charges. Charge density and the ratio of hydrophilic to hydrophobic properties can affect the conformation of WP, which can therefore change the physical and mechanical properties of coatings and films (Gerna et al., 2023). Native WP is seen as a bad sticky polymer because it has a tight globular structure and very small molecules. Some circumstances allow this spherical form to be changed to more linear forms, and eventually, clumps that cannot be returned to the original one are formed by the thiol-disulfide exchange. Cohesive films can be formed by the hydrogen bonding, van der Waals forces, or even forced by electrostatic forces between native WP molecules. To prepare films and coatings with the aid of WP, the dissolution of WPC or WPI (5-12%) in distilled water, raising the pH to 7 - 8, or simply leaving it alone, and heating the liquid at 80-90 °C for 10-30 minutes is often required to denature the proteins. Before and after heating, you can add more ingredients, which depend on their compatibility. After heating, heat-sensitive chemicals like probiotics, antioxidants, and antimicrobials are added. Components that are heat-sensitive, including prebiotics, starches, and blends, are placed at the start (Kowalczyk et al., 2022). The thiol group reactivity occurs when the group breaks apart under alkaline conditions. Ultimately, gels are dried to convert them into WP films. This can happen with either heat-set or cold-set gels. The unusual way to dehydrate is to dry things in their natural state, usually at temperatures between 21 - 50% relative humidity (RH) and 23°C. But to ensure the quality, handling management throughout this process needs to be done carefully. Applying edible coatings to food needs to be done in a way that enables the food to dry slowly. If the food dries too soon, the films may become less flexible, thinner, and stiffer. According to Di Piero et al. (2018), adaptable. The rate of drying, the temperature, and the

humidity are of immense effect on the way the film dries and what the film will subsequently be. As an example, the drying temperature was reduced to 5°C instead of 25°C, which resulted in increased solubility and reduced permeability of whey protein emulsion films made with beeswax. Dried films (95°C, 30 % relative humidity) were thinner, stiffer, more long-lived, and less extensible than dried films (21°C, 50 % relative humidity). WP films that were dried in a microwave for 5 minutes showed significant gains in elongation and tensile strength compared to shorter drying times. But the air-dried films were also equal to the WVP films. Things can now be done in a number of ways. Some of the techniques include casting, dipping, extrusion, enrobing, foaming, spraying, and UV polymerization of edible WP films and coatings. The manner in which things are being handled should also be observed, as a change in processing environment would change the speed and process of filming. The development of a WP movie should be designed to work with food, so the movie will be deemed safe to eat. This involves differences in temperature, pH, salt, speed of enzyme, dehydration, and the use of chemicals and solvents. Also, the biopolymer needs to work with plasticizers and other additives.

Technologies for WP Coatings and Film Processing

Adhesion (the contact between the polymer and the substrate) and cohesion (the link between polymers) are the two main interactions that impact film formation and its characteristics. Polarity, chain structure, and molar mass are some of the polymer features that are very important in determining how well they stick together. Wet processing and dry processing are the two main ways to make protein films that you can eat. Solvent casting, which is a wet processing method, is the most common and commonly used of them. This approach works especially well for putting liquid coatings on food by brushing, dipping, or spraying them on. On the other hand, dry processing has become more popular in recent years. This method takes advantage of the thermoplastic properties of polymers to produce films by extruding and compressing the same when it is drier.

Solvent processing, also referred to as solvent casting, is the most appropriate method of creating thin-layer coatings as it is simple to operate and cost-effective. To determine the efficacy of the formation of the films, this casting method is frequently employed in scientific research. Research facilities have the technology that is capable of pilot-scale application of solvent casting of whey protein (WP) films and coatings. Solvents are used to make films by dissolving the proteins in one of the solvents. This might be water, alcohol, or a mix of solvents. This process also includes mixing and homogenizing plasticizers, polysaccharides, lipids, or emulsifiers. Plasticizers are necessary to make the protein film less fragile, which happens when there is too much crosslinking in the protein matrix. Nevertheless, one should verify the compatibility of each element of the movie with the other as they will alter the characteristics of the final film (Coltelli et al., 2015). Once this has been done, PH modifications may be effected as required, or certain crosslinking between proteins may commence, particularly through heating, to enable the film to form. The heating of the surface can be done prior to, concurrently, or subsequent to the coating process. In order to form the protein film, you apply the film solution manually on a Petri dish or Teflon plate and allow the solvent to dry. The coating is applied to the food surface before drying the solvent through dipping, spraying, or wrapping the film solution around the food surface. According to (Azeredo & Waldron, 2016), constant thickness is a film type that is produced mechanically using batch methods or continuous methods through the use of coating. The qualities of the movie are decided according to the composition and techniques employed at the production levels. Due to this fact, wet-processed materials possess a vast variety of functional characteristics, including protein concentrations, solution pH, type of solvents and additives used, and drying conditions, including temperature and speed. The type of drying technique to be used, such as ambient conditions, conventional hot-air drying, infrared, or microwave drying, will also influence the final properties of the film (de Castro et al., 2017; Di Pierro et al., 2018).

Covering and Keeping

Each egg, as well as control samples and eggs covered with wheat protein and chitosan membrane, was weighed using an accurate electronic balance. The goods were dehydrated at a storage temperature of 25 °C throughout the entire night. There were three sets of eggs, namely uncoated control samples, chitosan-coated samples, and wheat protein-coated samples. All the samples were incubated at room temperature (25°C and 60 % humidity) on cardboard egg trays. The sample names are the following: T0 control samples (uncoated eggs), T1 2 grams of chitosan coating, T2 3 grams of chitosan coating, T3 5 grams of chitosan coating, T4 2 grams of wheat protein coating, T5 3grams of wheat protein coating, and T6 5grams of wheat protein coating.

Assessment of Weight Reduction During Storage

The method outlined by (Ali, 2024). The procedure was carried out to determine the extent to which the weight of the egg samples dropped as they were being preserved. The eggs were first weighed in a saturated sodium chloride solution. They were then put in a glass dryer, which was 50% humid. The decrease in weight was seen after four weeks. Was computed by using the equation below:

$$\text{Weight loss(\%)} = \left(\frac{W_0 - W_t}{W_0} \right) \times 100 \quad (1)$$

In this equation (1), W₀ is the initial weight of the egg when the storage is going to be done, and W_t is the weight of the egg at the end of the storage period. This weight loss is then calculated as a percentage of the original weight, which shows the weight that has been lost by the egg over time. It is used to determine the ability of various treatments or storage conditions to inhibit the loss of weight in eggs during preservation.

Checking the Haugh Unit and Albumen Ph

Egg quality was evaluated using a digital egg tester (Model DET6500, NABEL Co., Japan) for Haugh Unit (HU), and a pH meter (Eutech Instruments pH-510, Singapore) for albumen pH. Five eggs per treatment were analyzed weekly.

HU was calculated as:

$$HU = 100 \log(H - 1.7W^{0.37} + 7.6)$$

where H is albumen height (mm) and W is egg weight (g).

Bacterial Enumeration

We used the approach described by (Humphrey, 1994; Jahangiri et al., 2024) to look at the microbiological contamination of the outside of the egg shell and the inside (albumen and yolk). Microbial specimens were obtained from the eggshell surface at 0,7, and 14days post-application using a sterile swab moistened with one milliliter of distilled sterile water. Next, the swab was mixed for 30 seconds in a glass tube with sterile distilled water. The albumen and yolk mixing was done in a sterile plastic bag. In sterile distilled water, serial dilutions of the samples were done for both exterior and interior specimens. We inoculated 0.1 mL of samples of each of the treatment groups and the control on MacConkey agar plates. The count of bacterial colony-forming units (CFU/mL) was determined after the plates had incubated at 37°C for 48 hours.

Eggs' Nutritional Value

The protein, fat, moisture, and energy of the eggs were studied by the AOAC method through tests on ash and carbohydrates. This study involved mixing the albumen and egg yolk at 25°C. Five samples were examined for each condition.

Integration of Biotechnology and Environmental Monitoring Tools

In order to make the analysis of food quality more comprehensive, this work does not overlook the current developments in AI-based sound recognition systems that are utilized in environmental control and wildlife protection. Such technologies that frequently use bioacoustic sensors demonstrate how machine learning models can be used to analyze natural soundscapes and detect ecological shifts or species migration to support sustainable food systems and biodiversity conservation.

Besides, there is a growing trend in biodegradable sensors, bio-polymeric films, and eco-engineered materials, which underscores the increasing involvement of biotechnology in food preservation, sustainability of the environment, and smart packaging. These tools can be used in line with the overall goal of the present study, namely the promotion of sustainable and biodegradable coating materials, lessening waste and environmental footprint.

Statistical Analysis

Analysis of variance (ANOVA) and New Multiple Range Test (DNMRT) by Duncan were used to statistically examine the data at 5 % significance level. The findings were made using the data.

Results

The Effects of Different Coatings on the Reduction of Egg Weight

The mean of the results indicates that the weight reduces as it is stored. According to the FAO, a 2 -3 percentage loss of weight in eggs during marketing is acceptable. Refrigeration maintained this acceptable range of the weight loss of uncoated eggs two weeks longer than when they were maintained at room temperature, as illustrated in Figure 1. The longer the food is stored, the more it tends to lose its weight. Eggs impregnated with chitosan and cuttlefish protein did not lose as much weight during storage as compared to control eggs. The greatest weight loss occurred in uncoated egg samples, and this was between 1.84 % in the first week to 6.78 % in the fourth week. Eggs covered with Chitosan, however, did not lose the most. The weight loss in therapy T2 was of a complexity of 1.65-1.97 %. The findings of the study conducted by (Bhale et al., 2003) showed that the weight reduction was slightly different based on the applied coating, storage time, temperature, size, and porosity of the shell. Chitosan coating was more efficient compared to cuttlefish protein coating in relation to weight loss prevention. Research has shown that the weight loss occurring during storage could be reduced by coating eggs with chitosan and cuttlefish proteins, which provide a protective layer against the loss of moisture through the eggshell. However, the hydrophilicity of whey protein films could result in a lack of moisture barrier properties. The weight loss values of this study were likely to be low since the WPC coating was likely to prevent the migration of moisture as compared to the chitosan coating.

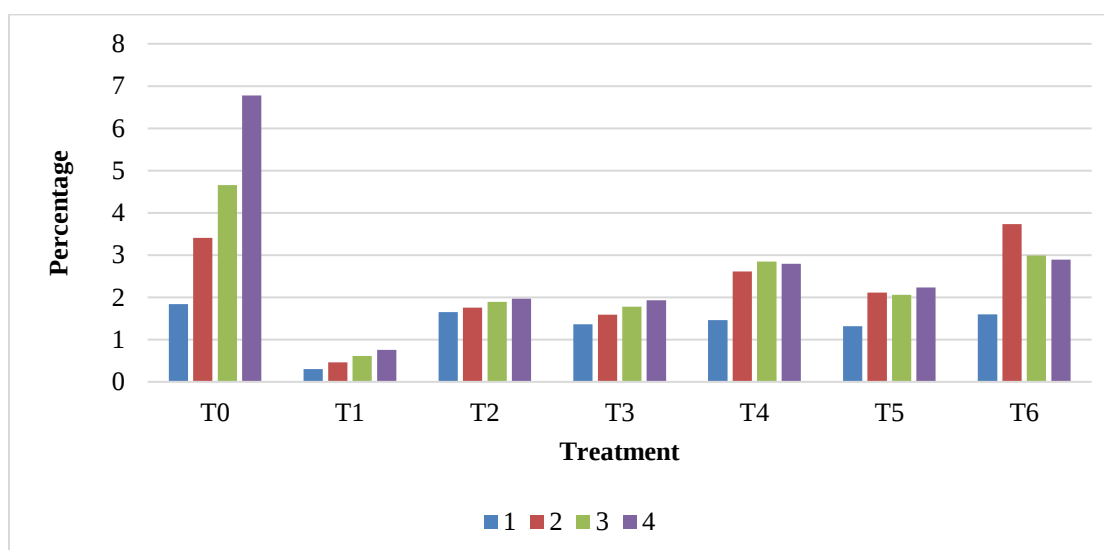


Figure 1. Comparative weight loss of eggs treated with different coating formulations

In this bar chart, the percentage value of five groups (1–4) has been represented in seven treatments (T0–T6). Each treatment includes five colored bars, allowing comparison of how the percentage of each group changes with different treatments.

Different Coatings' Effects on the Haugh Unit (HU)

The albumen quality is correlated with the Haugh Unit (HU), which is the commonly used measure to estimate the weight of the egg and the thickness of the albumen in the egg. Fresh eggs of superior quality have an HU value of 70 or above, while eggs that have been stored for longer periods of time have lower HU values. When compared to eggs covered with edible films, the control samples showed considerably lower HU values. Eggs can be graded according to their HU values: AA (above 72), A (60-72), B (31-59), C (less than 30) (Waimaleongora, EK, et al., 2009).

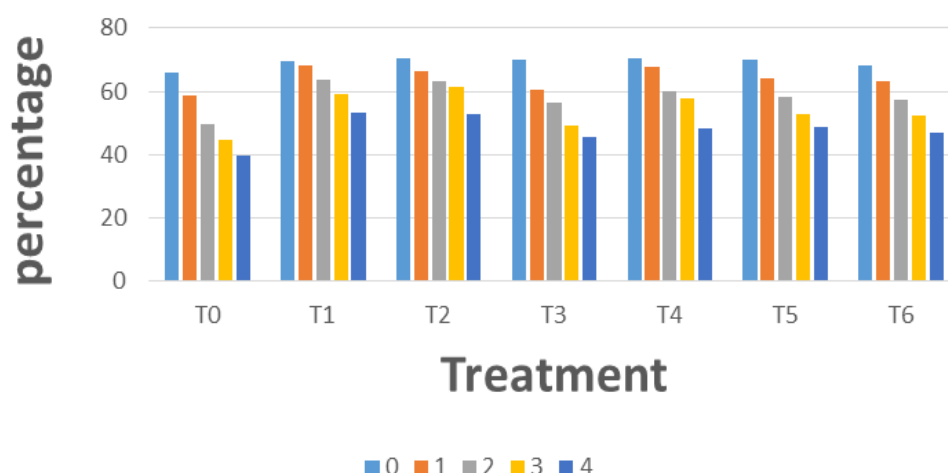


Figure 2. Comparison of percentage responses under different treatments

The bar graph indicates percentage values in the various treatments (T0-T6) in five groups (0-4). In both treatments, there are five colored bars that depict the groups to enable a comparison of the proportion of changes in percentages within the treatments.

Figure 2 presents the mean values associated with weight loss during storage. The largest values of HU had been registered in the uncoated egg samples that ranged between 66.07 and 39.63. However, the HU values of the chitosan-coated eggs did not change significantly during the storage period. The range was 70.65 to 52.75 in T5 therapy, whereas in treatment T2, it was 69.40 to 53.10. Uncoated eggs had their quality grade of AA reduced to B and C in the third and fourth weeks. Conversely, in the fourth week, chitosan-coated eggs received a grade of A, whereas cuttlefish protein-coated eggs received a grade of B. On the other hand, it was noted that uncoated eggs maintained their initial AA grade for five weeks at 4°C and for four weeks at 7°C (Biladeau & Keener, 2009). Mineral oil-coated eggs, on the other hand, kept their AA grade for 12 weeks at 7°C and for the whole 15 weeks at 4°C. Overall, even though storage time has a major detrimental influence on the albumen quality of eggs at 25°C, this effect may be successfully lessened by using refrigeration and an inexpensive oil surface coating.

Table 1 presents the comparative effects of various edible coating treatments (T0–T6) on egg quality indicators over four weeks of storage at 25°C. Weight loss (%) increased in all treatments over time, with the control group (T0) showing the highest loss (6.78%) and the Chitosan–Whey combination (T6) showing the lowest (2.4%). The values of Haugh Unit decreased in both treatments, signifying that albumen quality decreased with a lengthy storing period; nevertheless, coated eggs, particularly T2 and T3, recorded significantly the highest HU values than the uncoated ones. The microbial load (CFU/mL) accumulated at zero to different degrees by Week 4, and the lowest contamination was registered in the chitosan-based coating (T1–T3) and the combination coating (T6), indicating that they offered the best antimicrobial shield.

Table 1. Summary of weight loss, haugh unit, and microbial load of eggs under different coating treatments at week 0 and week 4

Treatment	Week 0 Weight Loss (%)	Week 4 Weight Loss (%)	Week 0 Haugh Unit (HU)	Week 4 Haugh Unit (HU)	Week 0 Microbial Load (CFU/mL)	Week 4 Microbial Load (CFU/mL)
T0 (Control)	0.0	6.78	72	39.63	0	2000
T1 (Chitosan)	0.0	3.5	72	60	0	400
T2 (Chitosan)	0.0	2.8	72	62	0	450
T3 (Chitosan)	0.0	3.1	72	64	0	500
T4 (Whey)	0.0	3.0	72	62	0	500
T5 (Whey)	0.0	3.5	72	60	0	600
T6 (Chitosan- Whey)	0.0	2.4	72	63	0	400

Impact of Various Coatings on Egg PH

An elevation in pH readings signifies a definitive indicator of CO₂ loss to the external environment via the shell pores as eggs mature. Mineral oils and Chitosan are two examples of coating materials that work well as barriers to stop carbon dioxide and other gases from escaping through the eggshell. This helps maintain a lower pH in the egg white and preserves its quality. Figure 3 presents the average pH values for the egg samples. The highest pH values were recorded in uncoated egg samples. As an example, the pH of T1 samples was 7.30 on the first day of the treatment, and it increased to 9.12 at the fourth week of storage. Conversely, eggs enveloped in Chitosan exhibited reduced pH values. In treatment T2, the pH level was 7.07. This is consistent with what he mentioned, that compared to eggs that were not coated, coating with mineral or soybean oil (which is more economical) may significantly minimize weight loss and retain the albumen quality of eggs for at least four extra weeks throughout the five weeks that they were stored at 25°C.

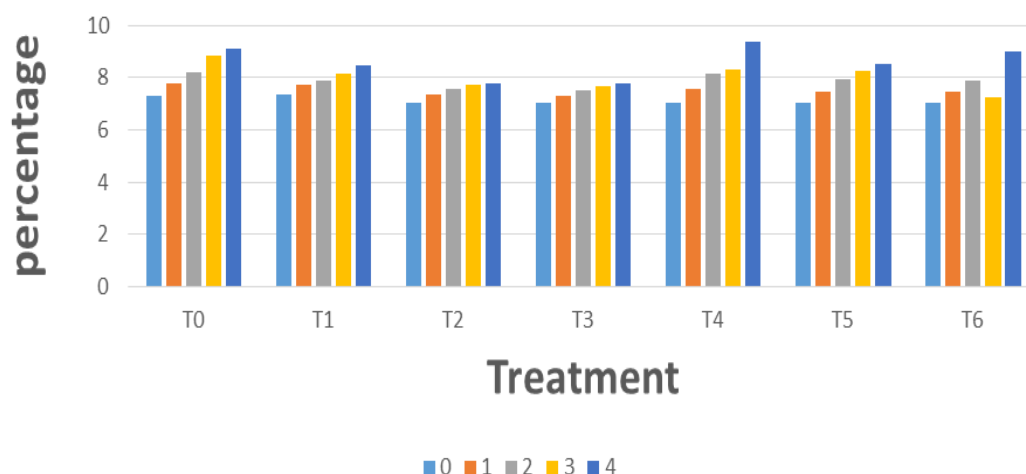


Figure 3. Variation of percentage values under different treatments

This bar chart represents the percentage of five groups (04) under seven treatments (T0- T6). Each treatment includes five colored bars, showing how the percentages vary slightly between groups and across different treatment levels.

Impact of Various Coatings on Microbial Proliferation on Eggs

Figure 4 reveals that eggs that were coated with Chitosan had a lower number of Coliform colonies as compared to the control. The findings showed the antibacterial effect of the chitosan coating, which led to the reduction of bacterial contamination on the eggshell. It has been shown that Chitosan is very effective when used against Gram-positive bacteria (Atakan et al., 2025). Differences in microbial communities can be associated with interactions of both Gram-positive and Gram-negative microorganisms and coliform bacteria. The eggshell was not observed to be contaminated with any bacterium within the first week following the coating application with the chitosan and cuttlefish protein, which is probably attributed to the antibacterial properties of the chitosan coating. The accumulation of wastes led to the proliferation of microbial communities in the second week. This corresponds to the results of who suggested that microorganisms should be tested. The outcomes of the Petri dish showed that none of the microbes could enter the eggshell of all the coated eggs because the yolk and the white samples did not grow. However, the *Aspergillus niger* proliferated in the white samples of the control sample. The washing could have caused the development of *Aspergillus* infection in the egg by destroying the cuticle and exposing the shell pores to the entrance of microbes. Germs were also present on the shell surface in large amounts. Several studies indicate that the removal of this outer layer promotes fungal growth, particularly in species within the *Cladosporium* genus. Some polymers, such as Chitosan, have antibacterial properties that have been used to stop germs from growing in table eggs. Other studies have aimed at applying coatings to minimize the number of microorganisms that contaminate food.

It is a graph depicting the distribution of the different components (Ash, Carbohydrates, Fat, Moisture, Protein) at two time points (Week 0 and Week 4) under each of the treatments (T0 -T6). Each treatment is designated by a definite color, indicating the manner in which the percentage of the components varies over time.

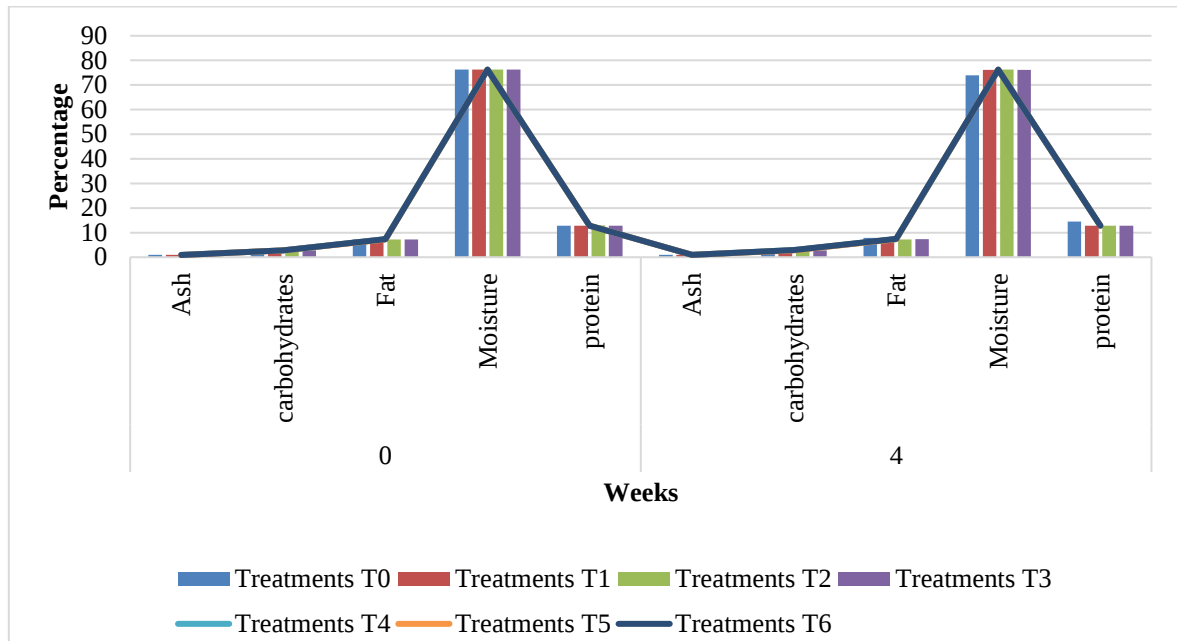


Figure 4. Nutritional composition of eggs under different coating treatments at week 0 and week 4 of storage

Impact of Various Coatings on Microbial Presence in the Internal Contents of Eggs

As seen in Figure 5, eggs coated with Chitosan had a lot fewer coliform bacteria colonies than the control samples. Treatment T2 was the most effective in lowering pollution levels on the shells. Eggs have a natural way of protecting themselves against bacteria getting into the vitelline and albumen membranes. After bacteria get inside the albumen and vitelline membranes, they can quickly multiply in the egg yolk while the egg is being stored. The chitosan covering keeps microorganisms from getting into the inside contents. There was no microbial contamination on the eggshell in treatment T2; however, there was microbial contamination inside the eggs. This may result from the presence of natural air within the eggs, which serves as a substrate for bacterial proliferation during egg packing. Elevated microbiological contamination may signify issues with hygiene and sanitation on the farm.

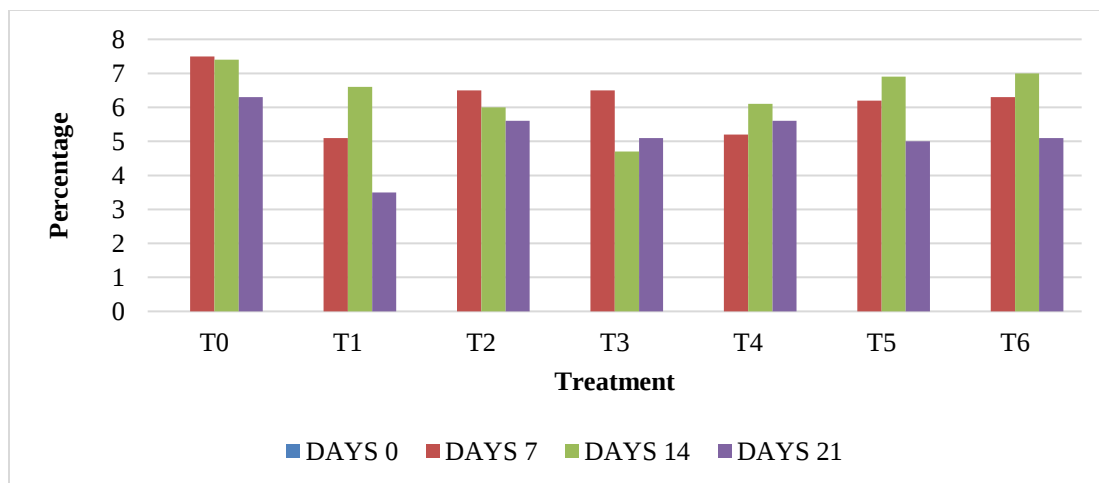


Figure 5. Effect of different edible coating treatments on microbial load (CFU/ml-log) 10) in egg internal contents during 21 days of storage

Nutritional Value of Various Egg Treatments During Storage

These substances in nutrition are protein, carbohydrates, and lipids. The level of protein and moisture of the eggs was determined after the synthesis of the albumen and egg yolk. The interior contents of the fresh eggs were also studied, as well as those of uncoated, chitosan-coated, and cuttlefish protein-coated eggs, as observed in Figure 6. The results indicated that, after four weeks at 25°C, the nutritional content of the coated and the uncoated eggs was the same as that of fresh eggs. These results are consistent with the assertions made by

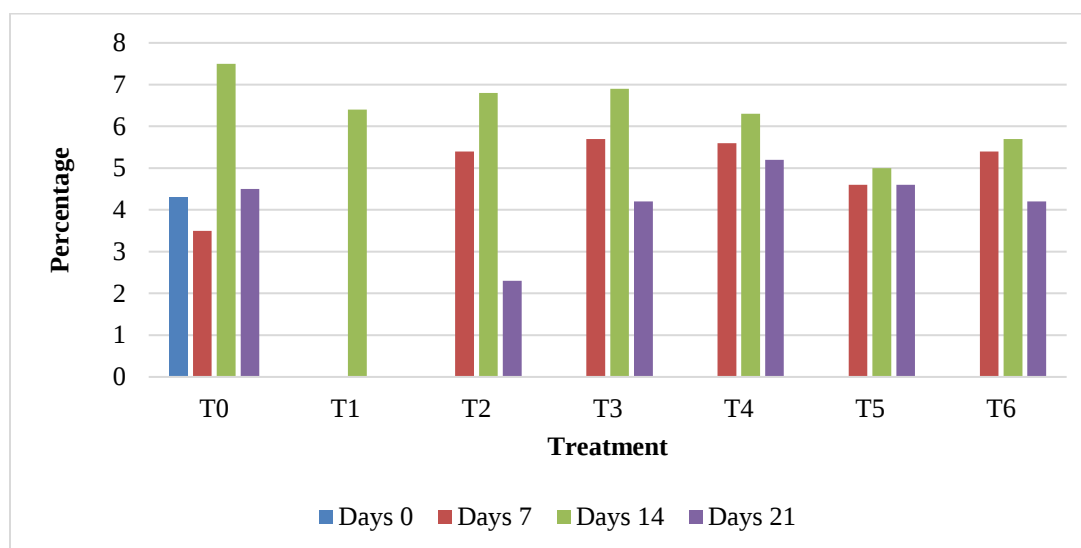


Figure 6. Changes in nutritional composition of eggs under different coating treatments during 21 Days of storage at 25 °C

This bar graph displays the percentage values across different treatments (T0–T6) at four time points (Days 0, 7, 14, and 21). Bars of each treatment are represented with varying colors and indicate the percentage changes of each type of treatment through time.

Integration of Biotechnology and Environmental Monitoring Tools

The incorporation of biotechnology-driven environmental monitoring tools in this study provides additional insight into how sustainable food systems can be strengthened beyond the immediate effects of edible coatings on egg quality. Although the primary focus of the experiment was to evaluate the influence of chitosan, whey protein, and chitosan–whey biopolymer combinations on egg preservation parameters, the extended framework highlights how machine learning, biodegradable sensors, and eco-engineered materials can complement food preservation research. The recent implementations of AI-based sound recognition models, especially bioacoustic sensors, depict the ability of machine learning algorithms to track the environmental conditions with extreme accuracy. Such systems identify the change in the vocalization patterns of the species, disturbances in the habitat, and the stressors of the environment. Such tools are relevant in the field of conservation science as they assist in monitoring biodiversity, recognizing indicators of species health, and notifying scientists about the initial ecological disturbances. These study findings are in line with these technological developments in two aspects. To start with, the efficacy of chitosan and chitosan whey coating in minimizing microbial contamination manifests how the biopolymeric material can serve as a bioactive layer similar to the biodegradable sensors in ecological surveillance. Both applications make use of natural polymers, which are environmentally friendly, low-cost, and react to biological stimuli. Second, environmental listening with machine learning technologies compares to the rise of food-monitoring technologies, smart biofilm, and

eco-engineered packaging to sense food spoilage, pH changes, or microbial growth. The findings of the reduced microbial load of coated eggs (mainly T1-T3 and T6) may indicate that a combination of such natural coatings with biodegradable sensor technologies would allow developing intelligent packaging that can deliver real-time quality information. Using the insights of the theory of AI-aided conservation literature, this study highlights the opportunity of data-driven, environmentally friendly monitoring systems in the food supply chain. These systems have the ability to reduce wastage, increase traceability, and reinforce sustainability. Machine learning can be applied in both agricultural and wildlife settings to improve the accuracy and responsiveness of the monitoring process, and eventually contribute to the resilience of the environment, management of biodiversity, and conservation of resources. In general, the combination of biotechnology and environmental monitoring devices focuses on the increased applicability of the proposed research: biodegradable materials not only safeguard food products but also support global efforts to promote sustainable ecosystems, conservation technology, and environmentally friendly innovation.

Expanded Conservation-Relevant Interpretation

This study also has an implication on large-scale conservation efforts because its findings have shown that biodegradable biomaterials and biotechnology-based devices can be used to improve environmental monitoring. Just as environmental protection begins to be provided by edible coatings as protective layers that reduce microbial activity, biodegradable sensor systems applied in conservation can also monitor environmental stressors without adding to pollution. Machine-learning-driven bioacoustic monitoring has become one of the primary conservation instruments that can be used to determine the presence of species, changes in vocalization patterns, and ecological instability with high accuracy. These technologies are comparable to smart food-monitoring technologies, where real-time information on spoilage, microbial proliferation, or pH change can enhance the safety and sustainability of food storage. This study identifies a common objective when the principles of the natural polymer use are aligned with the principles of AI-supported acoustic surveillance, which empowers food security alongside the conservation of biodiversity. Finally, machine learning and biodegradable materials constitute a solution that can be used in unison and results in limiting environmental harm and enhancing the quality of monitoring in agricultural and ecological systems.

Discussion

The results of this paper indicate that the chitosan and whey protein concentrate developed edible coating can be used in preserving the internal and external qualities of table eggs stored at 25°C. There was a steady weight loss in all treatment conditions, which is consistent with prior reports that indicated that continued storage results in movement of moisture through the eggshell. Nonetheless, coated eggs, especially those coated with Chitosan and the blend of chitosan-whey, had a grossly lower weight loss compared to uncoated eggs. It means that such coatings developed efficient semi-permeable layers that reduced the process of vapor exchange. The excellent performance of the combination treatment indicates that chitosan and whey protein interactions could enhance film density and mass transfer reduction as compared to single-component coatings. The quality of albumen in terms of the Haugh Unit exhibited a distinct tendency of decline over time, whereby uncoated eggs became weaker in terms of grade AA up to lower grades in just a span of four weeks. Conversely, the coated egg had much higher HU values during storage, and it portrays better preservation of albumen thickness and internal CO₂ balance. This effect of protection is explained by the ability of the gas barriers of the surfaces that contain the eggs to preserve internal pressure and slow the alkalization rate that is typically observed in stored eggs. The findings justify the previous results that edible films can slow the process of albumen degradation by restricting the diffusion of carbon dioxide. Alterations in pH were also anticipated, with the

uncoated eggs having maximum pH changes as the eggs continued to be stored. Eggs coated with Chitosan had a reduced increase in pH, which once again validated the chitosan-based film to contain CO₂. This action is similar to that of the mineral oil coatings, which have been reported previously, only that it is biodegradable and non-synthetic. Constancy of pH in coated eggs helps in preserving albumin gel and general freshness. Microbial analysis indicated that chitosan shells possessed an evident antimicrobial benefit, which led to a significant decrease in contamination of the surface of the shell and the penetration of bacteria into the egg content. The strong antimicrobial activity of Chitosan, which has been well documented in the literature, explains the reduced microbial load observed in treatments T1–T3 and T6. In contrast, uncoated eggs showed increasing microbial contamination over time, demonstrating that the absence of a protective layer, especially after washing, facilitates microbial infiltration through shell pores. The results establish that not only can edible coatings retard the growth of microbes, but they can also substitute the natural cuticle removed during the washing process. The nutritional composition did not differ significantly among treatments, which suggests that none of the treatments, storage at 25 °C with and without coatings, had any significant effect on protein, fat, moisture, or carbohydrates of the eggs. This is consistent with previous studies that revealed that no significant changes in major nutrient components occur during moderate storage, provided that microbial spoilage and moisture loss are maintained. Viewed in a more general context, the findings of the current research are consistent with current developments in biotechnology and environmentally friendly monitoring systems. Chitosan and composite biopolymer coatings serve a similar purpose to biodegradable sensory materials employed in conservation technologies, with natural polymers being able to generate a response to an environmental or biological input without adding to pollution. Simultaneously, machine-learning-based bioacoustic surveillance systems applied in ecological research can be seen as the reflection of the new vision of smart food packaging, which would be able to sense the signs of spoilage in real-time. Through incorporating coating technology and such eco-engineered sensing systems, it is possible that future packaging solutions are able to provide preservation advantages and intelligent monitoring functions to improve food safety and sustainability. On the whole, the research confirms that biopolymer-based coating, in particular, Chitosan and chitosan-whey preparations, can significantly improve the egg quality preservation during storage. They help to minimize moisture loss, preserve albumen, stabilize pH, limit microbial growth, and preserve nutritive quality. At the same time, they align with global efforts to promote sustainable, biodegradable materials and data-driven monitoring tools, supporting both food security and environmental conservation.

Conclusion

This study establishes that edible coatings, particularly a 3% chitosan formulation, have a substantial effect on preserving and enhancing the overall quality of table eggs during storage. Chitosan-coated eggs exhibited significantly reduced weight reduction, increased Haugh Unit values, and slower pH alteration, which means that they retain freshness and albumen integrity better. Its high antimicrobial property was effective in preventing the pathogenic microorganisms on the shell surface and preventing the penetration of bacteria into the internal contents, thus enhancing microbial safety. The nutritional testing also ensured that the food sample coated with eggs did not change moisture, protein, fat, ash, and carbohydrate contents during the storage period, which indicated the preservation ability of chitosan-based films to maintain internal structure. Chitosan, a very efficient moisture and gas barrier, reduced the diffusion of CO₂ and also slowed down the degradation of albumen, compared to uncoated eggs. Chitosan is a natural, safe, biodegradable, and environmentally friendly material, and as such, would provide a more sustainable substitute to synthetic preservatives. The fact that it increases the shelf life of eggs, reduces post-harvest losses, and preserves the nutritional and functional properties of eggs makes it commercially viable and applicable to farmers and consumers seeking safer, fresher, and long-lasting table eggs. In addition to food preservation, the implications of this work in general

are related to environmental monitoring, sustainability, and conservation of biodiversity. The biodegradable sensors and eco-engineered materials based on natural biopolymers like Chitosan are becoming more popular in decreasing pollution and improving the health of the environment. New biotechnological devices, such as biosensors in natural polymer matrices, can detect pollutants, locate the quality of the habitat, and examine ecological alterations without depositing toxic residues in the ecosystems. Moreover, current developments on AI-based monitoring platforms, including machine-learning-based bioacoustic applications, can provide extremely specific ways of monitoring wildlife behavior, biodiversity changes, and even forecasting the initial signs of ecosystem distress. The concepts that were revealed in the observation of this study, barriers, microbial prevention, biocompatibility, and sustainability, are the same principles that inform the development of intelligent environmental monitoring systems. Thus, the usefulness of chitosan coats in preserving biological products promotes the broader view of the introduction of biodegradable technologies and AI-based environmental monitoring to protect the quality of food and nature. One of the possible areas of work in the future would be the development of new biosensors and AI-based systems that would be applied in the monitoring of wildlife, the protection of nature, and the control of pollution. With the incorporation of natural polymers, such as Chitosan, with intelligent sensing systems, sustainable technologies could be developed that are able to sense environmental pollution, monitor species behavior, or the real-time health of habitats. These types of innovations would not only enhance the environmental conservation initiatives but also encourage environmentally friendly monitoring techniques that are in tandem with the global sustainability objectives.

Author Contributions

All Authors contributed equally.

Conflict of Interest

The authors declared that no conflict of interest.

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