Synergistic Effect of Chitosan and Clove Oil on Raw Poultry Meat

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Abstract

Poultry raw meat is a perishable and nutritious food and considered as a prime source of protein. Its high moisture content favors the bacterial growth and leads to deteriorative changes. Bacterial contamination and lipid oxidation reduce the quality and stability of chicken meat and ultimately result in social health concern. For the purpose of extending shelf life of meat and to prevent further bacterial contamination, antimicrobial based edible coatings (food grade material) are being widely used now days which also maintain the quality of meat. In present study, antimicrobial potential of chitosan and clove oil as coating material was checked on quality attributes of fresh poultry meat. The study was divided in to three phases; first phase deals with preparation of edible coating for its application on meat. In second phase, proximate composition of fresh raw chicken meat was checked prior to application of coating material followed by storage at refrigeration temperature (24 days). In last phase, chitosan based coated and uncoated (control) samples were studied periodically for microbial analysis (TPC) and sensory characteristics. The obtained results were subjected to statistical analysis and a significant difference \( p \leq 0.05 \) was observed among treatments. On over all basis, it was concluded that treatment, \( T_c \), containing 1% chitosan solution +2% starch solution +1.5 mL clove oil performed best due to their synergistic barrier properties against aerobic microbes and increased the shelf stability of raw meat in comparison to uncoated meat samples.

Keywords: Bacterial contamination; Chicken meat; Edible coating; Overall acceptability; Shelf life

Introduction

According to the Center for Disease Control and Prevention (CDC), 48 million people get sick, 128,000 are hospitalized, and 3000 die every year due to food borne diseases. Maintaining nutritional and organoleptic properties of food commodities as well as ensuring their microbiological safety is still the priority of present era [1]. Use of antimicrobial packaging is much feasible to provide safety and quality to refrigerated products [2]. Antimicrobial based package is considered as one of the types of active packaging [3,4]. The incorporation of active agents into packaging tends to create an environment inside the package which could delay/prevent the growth of microorganisms on product's surface and, hence, lead to extension in its shelf life [5].

An edible coating is responsible for better oxygen permeability, solute movement, provide good barrier properties to moisture, UV while ease in incorporation of nanoparticles, nanosensors, synthetic and natural antimicrobial agents [6,7]. These coatings also act as a host of various additives which impart a variety of functional properties in minimally processed foods [8]. Recently, edible coating and films are being used to decrease the oil uptake in fried foodstuffs. These are slight layers of edible stuffs, formed formerly, coated on foods which can prevent the growth of microorganism and can be used in replacement of synthetic plastics for food applications [9,10].

The major advantages of edible film and coating are; it can be eaten with food product, prevents moisture losses during frozen storage, retains color of fresh meat, improves flavor and texture, resists lipid oxidation, reduces spoilage, and reduces environmental pollution (such as in poultry, seafood, frozen and processed meat) [11,12].

Due to awareness in scientific community, natural antimicrobial agents (chitosan (CTS) and cloves) as a coating material are preferably used as an alternative to synthetic active packaging [13]. Chitosan is an abundant polysaccharides in nature (ranked second after cellulose) and has high molecular weight, usually attained by alkaline deacetylation of chitosan, present in exoskeleton of crustaceans, cell wall of fungus and other organisms [14,15]. It is demonstrated as a non-toxic, bio-functional, bio-degradable, bio-compatible and exerts antifungal as well as antibacterial effects in food matrices [16,17], although, it's antifungal activity is less as compared to antibacterial [18,19]. This food grade material comprises of three types of reactive functional groups, an amino group on C-6 position and two hydroxyl groups at C-6, C-3 positions which impart the antimicrobial potential to this substance [20,21]. Antimicrobial potential of chitosan has been reported against a wide variety of fungi, yeasts and bacteria [22-24].

Likewise, clove oil is also very useful preservative, flavor enhancer and exhibits antimicrobial activity [25]. This oil is obtained by the process of distillation of leaves, stems and flower of clove tree (Eugenia aromatic) and satisfies the criteria for being natural, safe, and healthy “preservatives” [16,26]. The Food Drug Administration (FDA) described its recommended level for food application (normally not>1500 ppm) and its daily intake level should be 2.5 mg/kg of body weight on the prescriptions by World Health Organization (WHO) [27].

It is a suitable material for designing food grade coatings and packaging structures for stored products in order to extend shelf life [28-30]. Keeping in view the excellent film and gas retaining capacities as well as good barrier properties, present study was designed to evaluate the antimicrobial effect of chitosan and clove oil on raw poultry meat and its effect on quality parameters in order to extend the shelf life during refrigeration storage.

Materials and Methods

Procurement of raw materials

Fresh raw poultry meat of uniform cuts, chemicals and other coating formulations were purchased from local market. The analysis
was performed in Food Microbiology and Biotechnology Laboratory, National Institute of Food Science and Technology, University of Agriculture Faisalabad.

Preliminary cleaning and proximate analysis of raw poultry meat

Meat was cleaned to remove the blood residues and any dirt and then subjected to proximate analysis. Moisture, crude protein, fat, and ash percentages were estimated from minced chicken by the published methods of AOAC [31].

Preparation of coating solution and treatment of boneless chicken meat

Antimicrobials based coating solutions were prepared according to modified procedure of Ojagha et al. [32]. To formulate the coating, following treatment plan was adopted (Table 1).

Application of antimicrobial coating

The prepared antimicrobial based coating solutions were applied on the boneless chicken meat. Prior to its application, meat was divided into five lots of uniform weight. One lot without coating application was considered as control while other four lots were dipped into coating solutions for 2 min and placed for drying for 15-20 min.

Storage of the treated chicken meat

Both coated and uncoated chicken meat samples were stored at refrigeration temperature (4 ± 1 °C). Samples from each lot were evaluated (microbial analysis and sensorial evaluation after frying) at 6 days interval starting from day 0 up to 24 days.

Microbial analysis of chicken meat

For microbial count the method as described by Yetunde et al. [33] was followed. Nutrient agar was obtained as commercially dehydrated powder from local supplier. This was prepared according to manufacturer’s instruction and was sterilized at 121°C and 15 psi for 20 min. The media was cooled slowly followed by pouring ~20 mL into petri dishes separately and allowed to solidify. After that, the serially diluted sample (0.01 mL) was spread on respective plates. The petri plates with inoculated samples were then inverted and incubated at 37°C for 24 h. The number of organisms was calculated by multiplying the count obtained from the selected dilution by reciprocal of the respective dilution factor, dividing the resultant by volume plated and expressed as Log CFU/g. The analysis was performed in triplicate to get precision.

Sensory evaluation

Sensory evaluation based on flavor, taste, texture, and overall acceptability was conducted by the panel of twenty trained and expert judges of National Institute of Food Science and Technology and scores were given by using 9 point hedonic scale (9=like extremely; 1=dislike extremely) method as described by Meilgaard et al. [34].

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Concentration</th>
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<tr>
<td>To</td>
<td>Control</td>
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<tr>
<td>T1</td>
<td>0.5% chitosan solution + 2% starch solution+1 mL clove oil</td>
</tr>
<tr>
<td>T2</td>
<td>1% chitosan solution+2% starch solution+1 mL clove oil</td>
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<tr>
<td>T3</td>
<td>0.5% chitosan solution+2% starch solution+1.5 mL clove oil</td>
</tr>
<tr>
<td>T4</td>
<td>0.1% chitosan solution+2% starch solution+1.5 mL clove oil</td>
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Table 1: Treatment plan of study.

Statistical analysis

The numerical data obtained for each parameter was subjected to statistical analysis to determine the level of significance and comparison of means was carried out according to the method as described by Montgomery [35].

Results and Discussion

The mean values for moisture, crude protein, crude fat and ash percentages of fresh boneless chicken meat are presented in the Figure 1. It was demonstrated that the moisture content, protein content and ash varied from 73.7 to 75.8%, 22.0 to 22.9%, and 1.04 to 1.74% respectively, in boneless chicken meat samples. The findings of present investigations are in accordance with the results given by previous researchers [36-38] who determined moisture, protein and fat contents which range between 74.2 to 75.38%, 19.51 to 20.62%, 2.14 to 4.48% accordingly. These researchers also noted non-significant difference in proximate composition of raw meat which supports the findings of the present study.

Microbial analysis of chicken meat

The effect of treatments, storage and their interaction (treatments × storage days) was found to be highly significant (p<0.01) on the total plate count (TPC) of refrigerated raw chicken meat. The main effects of storage days and treatment were significant (p<0.05) indicative of the fact that there are differences in mean values of the dependent variable (TPC) for both the various storage periods and various treatments. The mean values of TPC for all five different treatments (T0, T1, T2, T3, and T4) of raw chicken meat are given in Figure 2. The initial (day 0) TPC value for chicken meat (control and treated) was approximately 4.1 Log CFU/g which indicates the good quality of chicken meat, same observations were reported by Dawson et al. [39] and Latou et al. [40]. It is demonstrated from the Figure 2 that all treatments (T0, T1, T2, T3, and T4) on zero day and T4 on 12th day of storage differ non-significantly (3.42-3.08 Log CFU/g) effect on 6th day of refrigerated storage. It was also noted that TPC did not reach a value of 7 Log CFU/g, considered as the upper limit of TPC for fresh meat as defined by the ICMSF [41], during the whole period of storage (24 days). It was noticed from findings of present study that reductions in microbial counts (TPC) of treatments by dipping in chitosan solution were significantly lower

**Figure 1:** Proximate analysis of fresh raw chicken meat.
The scores for color reveal that T4 was preferred by the judges because it gave excellent color to meat sample, followed by T1, T3, T2 and To. The lowest color score was gained by uncoated treatment (To) (4.33) on the same day. The results of current study are in line with findings reported by Wang et al. [17], who conducted study with aim to investigate the effect of chitosan-carvacrol coating with or without caprylic acid (CAP) on the quality of Pacific white shrimp during 10 days of iced storage. The result showed that chitosan-carvacrol coating significantly inhibited the increase in total aerobic plate count (TPC) of shrimp in comparison to control which definitely can improve the overall acceptability. It was deduced from their study that chitosan-carvacrol coating may be promising to be used as active packaging for extending shell life, and similarly, incorporation of clove oil may also enhance the efficacy of the coating as observed in present investigation.

Sensory evaluation

Sensory evaluation is an important tool in product development. Acceptance of a food product depends upon the consumers perception about the quality attributes of a particular food. The results regarding analysis of variance (ANOVA) for color, flavour, taste and overall acceptability of coated vs control meat samples demonstrated the highly significant effect (p<0.01). The results pertaining to mean score of sensory attributes (color, flavour, taste and overall acceptability) are depicted in Figure 3. The scores for color reveal that T4 was preferred by the judges because it gave excellent color to meat sample, followed by T1, T3, T2 and To. The lowest color score was gained by uncoated treatment (To) (4.33) on the same day. The results of current study are in line with findings reported by Wang et al. [17], who conducted study with aim to investigate the effect of chitosan-carvacrol coating with or without caprylic acid (CAP) on the quality of Pacific white shrimp during 10 days of iced storage. The result showed that chitosan-carvacrol coating significantly inhibited the increase in total aerobic plate count (TPC) of shrimp in comparison to control which definitely can improve the overall acceptability. It was deduced from their study that chitosan-carvacrol coating may be promising to be used as active packaging for extending shelf life, and similarly, incorporation of clove oil may also enhance the efficacy of the coating as observed in present investigation.

Conclusion and Recommendation

The present study results suggested that chitosan and clove oil based coating were effective in inhibition of microbial growth in stored raw chicken meat and can be considered effective in maintaining the sensory quality of meat and meat products. The use of antimicrobial based coating for preservation of food item is a promising technology that can improve the quality of fresh meat products as well as increase their shelf life. Likewise, quality changes can be studied on meat and meat products by using other coating materials along with varying amounts and sources of essential oils other than clove oil.
Conflict of Interest

The authors declare that they have no conflict of interest.

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