Effect of Chitosan Powder and Nanoparticles as Natural Preservatives for Beef Minced Meat

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ABSTRACT

The need for natural products to be used as healthy food preservative alternatives for synthetic preservatives is increasing rapidly. This study aimed to compare the effect of chitosan powder (CP) and nanoparticles (CNPs) on the microbiological quality and shelf life of beef minced meat during its refrigerated storage for 9 days. The size of CNPs ranged from 150 to 350 nm as determined by transmission electron microscopy. Comparing the effect of the two chitosan products, CNPs exhibited higher antibacterial properties against S. aureus and E. coli, as revealed by lower minimal inhibition concentrations than the CP. Among these bacteria, E. coli was the most sensitive to CP and CNPs. Minced meat samples treated with CNPs showed significantly lower E. coli and total mesophilic counts than samples treated with CP. The pH values and antioxidant properties (as measured by the DPPH radical scavenging assay) of CNPs-treated samples were significantly higher than CP-treated samples. There were no significant differences in organoleptic characteristics between the two treatments. With these antimicrobial and antioxidant properties, CP and CNPs, with a better effect for CNPs, could be used as preservatives for the minced meat to extend its shelf life.

INTRODUCTION

Meat plays a pivotal role in safeguarding human health by giving all essential nutrients such as protein, vitamins, and minerals. However, meat can also act as a vehicle for many microorganisms, which may lead to food spoilage and poisoning, and subsequent severe economic losses. Microbial contamination of meat usually occurs during improper processing practices (Lonergan et al., 2019). Microbial spoilage and oxidation of meat are two main obstacles influencing meat product quality and shelf-life. Therefore, evolving techniques to expand meat shelf life could be a big target of the meat processing industry. Many consumers prefer low processed, preservative free, more steady, and unharmed foods. Thus, the evolution of safe natural food preservatives with high antimicrobial and antioxidant properties becomes an urgent demand (de Farias et al., 2019; Khan et al., 2016; Tayel, 2016).

Chitosan, a derivative of abundantly available chitin, is a biodegradable, bio-renewable, non-toxic natural polymer that is usually prepared from crustacean shell wastes (Kou et al., 2021). It has antibacterial, antifungal, and anticancer effects (Elkeiy et al., 2018; Khamis et al., 2019). In food technology, chitosan acts as the most favorable agent for the effective preservation of food due to not only its safety, health-promoting potential, and low cost but also for its physicochemical properties such as water-binding capacity, bioactivity, and toughness (Khan et al., 2016; Kumar et al., 2020; Tayel, 2016). Chitosan at a concentration range of 0.5%, 1.5%, 2% has potent antimicrobial properties against a large variety of food spoilage and pathogenic microorganisms (Dutta et al., 2009; No et al., 2002). Moreover, it has antioxidant effects since it can interfere with free radicals released during oxidative stress (Abd El-Hack et al., 2020). As most consumers do not prefer using synthetic antioxidants as food additives, chitosan was successfully used as an alternative antioxidant in food processing and packaging (Sabaghi et al., 2019).
al., 2015). To increase its efficiency, bioavailability, and specificity, chitosan was transformed into its nanoparticle form. Chitosan powder (CP) and nanoparticles (CNPs) were commonly utilized as a vehicle for drug delivery (Huang et al., 2004). Moreover, CNPs are reported to possess more potent in vitro antimicrobial effects than the parent chitosan (Divya et al., 2017). Apart from this previous study, little data are available in the literature regarding the comparison between the preservative effects of chitosan and CNPs on meat products. The present study aimed, therefore, to compare the effect of CP and CNPs on the microbiological quality, antioxidant properties, and shelf life of beef minced meat during its refrigerated storage.

MATERIALS AND METHODS

Materials

Low molecular weight edible chitosan (MW: 340 KDa; >10% moisture and 95% degree of deacetylation) prepared from carb shells was purchased from the Marine Hydrocolloids Company (Meron, India) in a powder form. Bacterial media (Nutrient agar, Baird Parker agar, and Eosin Methylene Blue agar) were obtained from Oxoid (Basingstoke, Hampshire, UK). Sodium tripolyphosphate (TPP) and 2, 2-diphenyl-picrylhydarzy1 (DPPH) were purchased from Sigma-Aldrich (St. Louis, MO, USA). Food pathogenic Gram-positive bacteria S. aureus (ATCC 8095) and Gram-negative bacteria E. coli (ATCC 25922) were provided by the Laboratory of Bacteriology, Department of Food Hygiene, Animal Health Research Institute, Dokki, Egypt, and were stored at 4°C. Fresh beef meat was purchased from a local butcher shop in Kafer El-Sheikh city, Egypt within 3 days of slaughter. Before mincing, meat samples were sterilized by soaking in 10 mg/L sodium hypochlorite for 60 min, followed by three times sterilized distilled water washes.

Preparation of CNPs

CNPs were chemically prepared using sodium TPP as previously described (Du et al., 2009). In brief, 100 mL chitosan (2%, w/v) dissolved in acetic acid solution (1%, v/v) were mixed with 4 mL sodium TPP (2%, w/v) dissolved in distilled water. The mixture was first stirred for 1 h, then, was sonicated at 1.5 kW for 10 min using Ultrasonic Homogenizers HD 2070. The resultant CNPs were refined by centrifugation at 10000 g for 1 h followed by twice rinsing of the precipitate (CNPs) with distilled water and then freeze-dried.

Characterizations of CNPs

The size and shape of the CNPs were determined using a JEOL transmission electron microscopy (TEM, JEM-2100) at 100 kV. After a brief sonication in ethanol to separate the aggregated dry CNPs, 200 µl of CNPs were mounted on a carbon-coated copper grid covered with nitrocellulose and then examined directly by TEM. Size distribution of CNPs were quantified by dynamic light scattering (DLS) using a Nano ZS zetasizer system (Malvern Instruments) with 633 nm laser wavelength and 173° scattering angle. CNPs size was determined by the average of three measurements and expressed as mean diameter (nm).

Determination of minimum inhibitory concentration

Minimum inhibitory concentrations (MICs) for CP and CNPs on S. aureus (ATCC 8095) and E. coli (ATCC 25922) were determined by agar dilution method, as recommended by the European Committee for antimicrobial susceptibility testing (EUCAST) (Testing, 2000) and as previously described (Badawy et al., 2020). CP and CNPs were tested at concentrations of 4 to 2048 µg/ml and 1.44 to 737.28 µg/ml nutrient agar media in liquid form (before being poured into the Petri dishes), respectively. All dilutions were used in three replicates. Control plates contained nutrient agar free from CP and CNPs. One loopful inoculum of each pathogenic organism (~1×10^4 CFU/g) in nutrient broth medium was spotted (10 spots/plate) on the surface of nutrient agar medium then incubated for 48 h at 37°C. The minimum concentration of CP and CNPs that inhibited the growth of S. aureus and E. coli, was considered the MIC.

In vivo antimicrobial activity

The minced meat samples (100 g each) were divided into 4 groups as follow: group 1 (-ve control group), samples were left without bacterial inoculation; G2 (+ve control group), samples were inoculated by E. coli (1x10^4 CFU/g); G3 (CP group), samples were inoculated by bacteria and treated with CP at a concentration of 1g/kg minced meat; and G4 (CNPs group), samples were inoculated by bacteria and treated with CNPs at a concentration of 300 mg/kg minced meat. Each group contained 7 samples and the obtained results were presented as means from three independent experiments. The concentrations of CP and CNPs were chosen based on the results obtained from the in vitro antimicrobial assay which showed better inhibition at these two concentrations. The contaminated minced meat samples were thoroughly mixed in polyethylene bags and then homogenized for 3 min at 37°C to confirm the appropriate distribution of the bacteria. A second round of homogenization was also performed following the addition of CP and CNPs. The polyethylene bags containing samples were stored under...
aerobic conditions at 4 °C until further use.

Total mesophilic count

Total mesophilic count was carried out according to the technique recommended by ISO. Samples were treated with either CP at a concentration of 1g/kg minced meat or CNPs at a concentration of 300 mg/kg minced meat.

Measurement of pH

The pH of minced meat samples was calculated by the method described by Jansen (2001). A homogenization of 10 g minced meat with 90 mL deionized water was done at room temperature. Following filtration, the pH was measured by a digital pH meter (Schott pH meter, CG824 mode).

DPPH- radical scavenging assay

DPPH (2, 2-diphenyl-1-picryl-hydrazil) radical scavenging assay was performed as previously described (Azaam et al., 2018). Minced meat samples (5 g / sample) were homogenized in methyl alcohol (10 ml). Following centrifugation at 7000 rpm for 7 min, 150 µl of the resultant supernatant was added to 1 ml of 0.1% DPPH. After 1 h incubation in dark, the yellow color was formed, and the absorbance was measured at 515 nm. Ascorbic acid was used as a positive control. The experiment was repeated three times. The DPPH scavenging activity was calculated from this equation: DPPH scavenging (%) = [(control absorbance - sample absorbance) / control absorbance] × 100.

Sensory evaluation

Sensory evaluation was performed as previously described (Petrou et al., 2012). A composite of odor, flavor, and appearance on a 9-point scale was requested from seven panelists to determine acceptability (total sensory evaluation). The scale points were excellent (9), very good (8), good (7), bad (6), first off-odor, and off-taste development (< 6). After first off-smelling or off-tasting, the sample was deemed inappropriate.

Statistical analysis

The analytics of statistics were conducted using SAS (Statistical Analysis System, 2004) using one-way ANOVA followed by the Duncan test as a post hoc test to determine the difference between groups. Data were presented as mean ± standard error of mean (SEM) and the significant values were set at p ≤ 0.05.

RESULTS AND DISCUSSION

Identification and characterization of the prepared CNPs

As examined under TEM, CNPs appeared as mono-dispersible spheres with different sizes ranging from 150 to 350 nm (Fig. 1). Size distribution as measured by dynamic light scattering (DLS) showed CNPs sizes ranging from 120 to 450 nm with a mean diameter of 297.6 ± 17.32 nm. Consistent with our findings, other studies also prepared CNPs with similar size ranges (Elkeiy et al., 2018; Feyzioglu and Tornuk, 2016; Loutfy et al., 2016). However, Badawy et al. (2020) prepared smaller CNPs than prepared in this study.

In vitro inhibitory effect of CP and CNPs on bacteria

Comparing the effect of the two chitosan products, CNPs exhibited a higher antibacterial effect, as revealed by lower MICs (170 mg/L for E. coli and 260 mg/L for S. aureus), than the CP (360 mg/L for E. coli and 550 mg/L for S. aureus). This indicates that E. coli is more sensitive to CP and CNPs than S. aureus and consequently, we used...
E. coli in the further investigation. Nearly close MICs of chitosan against E. coli (165 mg/L) were obtained by Badawy et al. (2020).

In vivo inhibitory effect of CP and CNPs on E. coli

Table I shows counts of E. coli in control-, control +, CP, and CNPs groups in the minced meat samples stored at 4 °C for 9 days. The E. coli count at 3, 6 and 9 days of storage were 125.31±1.98, 267.69±2.54, and 296.17±2.86 x 10⁴ cfu/g in the control-group, respectively. On the other hand, the control + group exhibited significantly higher counts of 317.54±3.28, 690.33±5.72, and 739.28±6.81 x 10⁴ cfu/g than all other groups. In contrast, groups treated with either CP or CNPs showed significant lowered bacterial counts compared to control + and control – groups. In treated groups, minced meat treated with CNPs showed significantly lower counts (35.28±0.63, 39.28±0.75, and 45.06±0.81 x 10⁴ cfu/g) compared to the CP group (84.11±1.03, 129.33±1.64, and 156.19±1.75) at 3, 6, and 9 days of storage, respectively. Subsequently, these results infer that treatment of minced meat with either CP or CNPs showed a lower E. coli number with best effect for CNPs. Therefore, it would be possible that if fresh minced meat was contaminated accidentally with E. coli, the addition of CP or CNPs would decrease the bacterial load.

E. coli count in the control– group was so high and exceeded the acceptable E. coli count (6.0 x 10⁴ cfu/g) as indicated by other studies (Cao et al., 2013; Hu et al., 2015). However, Badawy et al. (2020) detected a nearly similar higher count in the untreated (control -) minced meat samples. The antibacterial effect of chitosan, when added to meat samples, was previously reported by several studies (Abd El-Hack et al., 2020; Badawy et al., 2020; Cao et al., 2013; Dutta et al., 2009; Hu et al., 2015) and was attributed to chitosan ability to bind firmly with the negatively charged molecules on bacterial cell membrane causing their damage and leakage of their cytoplasmic contents (Ojagh et al., 2010). Chitosan particles also can form a coat surrounding the bacteria and preventing the entrance of oxygen and subsequently death of bacteria (Devlieghere et al., 2004). Moreover, the antimicrobial activity of chitosan is largely dependent on its molecular weight and size, as well as the type of microorganism. Indeed, lower molecular weight, smaller size chitosan as in CNPs showed higher antibacterial activity (Badawy et al., 2020; No et al., 2002). In support, we also found that CNPs had a higher antibacterial activity which could be attributed to their smaller size and higher penetration ability to bacteria. Gram-negative bacteria, such as E. coli, have thin peptidoglycan layer in the cell membrane which could be the cause for higher sensitivity to chitosan (Goy et al., 2016). The antimicrobial activity of chitosan could also be due to its high degree of deacetylation properties which can kill many food spoilage and pathogenic microorganisms (No et al., 2002).

Effect of CP and CNPs on total mesophilic count

The CNPs group showed significantly lowest total mesophilic counts of 1.15±0.04, 2.59±0.09, and 7.28±0.19 x 10⁴ cfu/g at 3, 6 and 9 days of storage, respectively compared to all other groups (Table I). The CP group showed significantly lower total mesophilic counts of 1.86±0.05, 4.67±0.15, and 11.35±0.23 x 10⁴ cfu/g than the control– group (1540.23±12.60, 18450.38±25.60, 60210.17±29.53 x 10⁴ cfu/g) at 3, 6 and 9 days of storage, respectively. These findings imply that treatment of minced meat with either CP or CNPs showed a lower total mesophilic count with better antibacterial potential for CNPs. Meat samples were considered spoiled if total bacterial counts were above 1 x 10⁸ cfu/g. Reducing total mesophilic count is also associated with longer shelf life of meat samples. Indeed, the shelf life increased 3 days more than normal meat after the addition of CP or CNPs.

Table I. Effect of chitosan on E. coli and total mesophilic count in minced meat samples stored at 4 °C for 9 days.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Day 3</th>
<th>Day 6</th>
<th>Day 9</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>E. coli count (10⁴ cfu/g)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Control (-)</td>
<td>125.31±1.98 b</td>
<td>267.69±2.54 b</td>
<td>296.17±2.86 b</td>
<td>229.37±2.16 b</td>
</tr>
<tr>
<td>Control (+)</td>
<td>317.54±3.28 a</td>
<td>690.33±5.72 a</td>
<td>739.28±6.81 a</td>
<td>582.23±4.59 a</td>
</tr>
<tr>
<td>CP</td>
<td>84.11±1.03 c</td>
<td>129.33±1.64 c</td>
<td>156.19±1.75 c</td>
<td>123.19±1.28 c</td>
</tr>
<tr>
<td>CNPs</td>
<td>35.28±0.63 d</td>
<td>39.28±0.75 d</td>
<td>45.06±0.81 d</td>
<td>39.62±0.67 d</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Total mesophilic count (10⁴ cfu/g)</th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Control (-)</td>
<td>1540.23±12.60 a</td>
<td>18450.38±25.60 a</td>
<td>60210.17±29.53 a</td>
<td>26733.33±23.54 a</td>
</tr>
<tr>
<td>CP</td>
<td>1.86 ± 0.05 b</td>
<td>4.67 ± 0.15 b</td>
<td>11.35 ± 0.23 b</td>
<td>5.96±0.17 a</td>
</tr>
<tr>
<td>CNPs</td>
<td>1.15 ± 0.04 c</td>
<td>2.59 ± 0.09 c</td>
<td>7.28 ± 0.19 c</td>
<td>3.67±0.12 c</td>
</tr>
</tbody>
</table>

Data were presented as mean ± SEM (n = 3). Columns carrying different letters are significantly different at p < 0.05. Before treatment: 130 x 10⁴ cfu/g for E. coli and 20.03 x 10⁴ cfu/g for total mesophilic count. Control (-), minced meat without E. coli; control (+), minced meat treated with E. coli; CP, chitosan powder; CNPs, chitosan nanoparticles.
Comparing between the Effect of Chitosan Powder and Nanoparticles

Table II. Effect of chitosan on pH and antioxidant activity of minced meat samples stored at 4°C for 9 days.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Day 3</th>
<th>Day 6</th>
<th>Day 9</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>pH values</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Control (-)</td>
<td>6.37±0.15&lt;sup&gt;AA&lt;/sup&gt;</td>
<td>5.92±0.13&lt;sup&gt;AB&lt;/sup&gt;</td>
<td>5.61±0.11&lt;sup&gt;AB&lt;/sup&gt;</td>
<td>5.93±0.12&lt;sup&gt;A&lt;/sup&gt;</td>
</tr>
<tr>
<td>Control (+)</td>
<td>6.03±0.12&lt;sup&gt;AA&lt;/sup&gt;</td>
<td>5.57±0.14&lt;sup&gt;B&lt;/sup&gt;</td>
<td>5.55±0.10&lt;sup&gt;B&lt;/sup&gt;</td>
<td>5.71±0.13&lt;sup&gt;+&lt;/sup&gt;</td>
</tr>
<tr>
<td>CP</td>
<td>6.30±0.19&lt;sup&gt;AB&lt;/sup&gt;</td>
<td>6.01±0.16&lt;sup&gt;AB&lt;/sup&gt;</td>
<td>5.84±0.17&lt;sup&gt;B&lt;/sup&gt;</td>
<td>6.05±0.17&lt;sup&gt;+&lt;/sup&gt;</td>
</tr>
<tr>
<td>CNPs</td>
<td>6.86±0.15&lt;sup&gt;AB&lt;/sup&gt;</td>
<td>6.42±0.14&lt;sup&gt;AB&lt;/sup&gt;</td>
<td>6.14±0.12&lt;sup&gt;B&lt;/sup&gt;</td>
<td>6.47±0.13&lt;sup&gt;+&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

Antioxidant activities (%)

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Day 3</th>
<th>Day 6</th>
<th>Day 9</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control (-)</td>
<td>42.34±0.24&lt;sup&gt;AB&lt;/sup&gt;</td>
<td>26.16±0.16&lt;sup&gt;B&lt;/sup&gt;</td>
<td>15.82±0.17&lt;sup&gt;C&lt;/sup&gt;</td>
<td>28.11±0.19&lt;sup&gt;C&lt;/sup&gt;</td>
</tr>
<tr>
<td>Control (+)</td>
<td>38.47±0.25&lt;sup&gt;AB&lt;/sup&gt;</td>
<td>19.22±0.18&lt;sup&gt;B&lt;/sup&gt;</td>
<td>13.58±0.16&lt;sup&gt;C&lt;/sup&gt;</td>
<td>23.76±0.21&lt;sup&gt;C&lt;/sup&gt;</td>
</tr>
<tr>
<td>CP</td>
<td>40.09±0.22&lt;sup&gt;AB&lt;/sup&gt;</td>
<td>30.11±0.20&lt;sup&gt;B&lt;/sup&gt;</td>
<td>24.28±0.21&lt;sup&gt;C&lt;/sup&gt;</td>
<td>31.47±0.20&lt;sup&gt;C&lt;/sup&gt;</td>
</tr>
<tr>
<td>CNPs</td>
<td>47.33±0.24&lt;sup&gt;AB&lt;/sup&gt;</td>
<td>39.71±0.21&lt;sup&gt;AB&lt;/sup&gt;</td>
<td>33.67±0.19&lt;sup&gt;C&lt;/sup&gt;</td>
<td>40.24±0.25&lt;sup&gt;+&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

Data were presented as mean ± SEM (n = 3). Columns carrying different lower-case letters are significantly different at p < 0.05. Rows of the three time points carrying different upper-case letters are significantly different at p < 0.05. Before treatment, pH = 6.52 and the antioxidant activity = 27.46%. Control (−), minced meat without E. coli; control (+), minced meat treated with E. coli; CP, chitosan powder; CNPs, chitosan nanoparticles.

Effect of CP and CNPs on pH

The pH values of the minced meat samples at 3, 6, and 9 days of storage in the control + group were significantly lower than the control − group (Table II). However, samples treated with either CP or CNPs showed significantly higher pH values compared to the control +. In treated groups, minced meat treated with CNPs showed significantly higher pH values than the control group. No significant difference was noticed between the control and CP groups. These findings indicate that treatment of minced meat with CNPs showed the highest pH value compared to all other groups. Regarding the three time points, the pH values in all groups were significantly reduced by increasing the storage time. Thus, pH values at day 9 were the lowest.

Minced meat is not appropriate for consumption with a pH below 5.6 or above 7.0 (Karabagias et al., 2011). In the present study, the overall mean of the pH values ranged from 5.85 to 6.86 which is compatible with the normal limit of meat pH (5.6–7.0). Similar pH values in beef meat were also recorded by other studies (Badawy et al., 2011). In general, lactic acid produced from glycogen during the storage time of the meat is responsible for lowering pH values from inhibitory pH for E. coli; is 5 (Cowan and Steel, 1965). Thus, pH values at day 9 were the lowest.

Effect of CP and CNPs on antioxidant activity

The antioxidant activity (%) of the minced meat samples at 3, 6, and 9 days of storage in the control + group were significantly decreased compared to all other groups (Table II). Treatment with CP or CNPs significantly increased the antioxidant activity compared to the control +. Minced meat treated with CNPs showed a significantly higher antioxidant activity followed by samples treated with CP. Moreover, the antioxidant activity in all groups was significantly reduced by increasing the storage time with lowest % on day 9 and the highest % on day 3.

Similar to our findings, previous studies have also reported higher antioxidant activity for chitosan when used in different forms with the best effect for the CNPs when added to minced meat samples (Badawy et al., 2020; Wan et al., 2013). It is well-known that potent antioxidants like chitosan can reduce lipid oxidation and so increase the shelf life of the stored meat (Karre et al., 2013). Therefore, chitosan as a safe, cheap, and potent antioxidant can be used as food additives (Li et al., 2010). CNPs decreased lipid peroxidation marker, MDA (El-Denshary et al., 2015; Subhapradha et al., 2017) and increased the activities of antioxidant enzymes (SOD, CAT, GPx) to inhibit the overproduction of reactive oxygen species (ROS) and protect tissue from oxidative damage (Elkeiy et al., 2018).

Effect of CP and CNPs on sensory evaluation

Table III shows the results of the effect of CP and CNPs on the sensory scores (appearance, color, odor, texture, and overall acceptability) of the beef minced meat at 3, 6, and 9 days of storage. Samples were considered spoiled if had a sensory score less than 6. Accordingly, the sensory scores of control − and + samples were only acceptable (above 6) till day 6 but on day 9 the scores dropped below 6 and spoilage signs (slimy appearance and off-odor) appeared. However, minced meat treated with either CP or CNPs had an acceptable sensory score till day 9 of storage. No significant difference in the sensory scores was noticed between the two treated groups.
Table III. Effect of chitosan on sensory characters of minced meat samples stored at 4 °C for 9 days.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Sensory score on</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Day 3</td>
<td>Day 6</td>
</tr>
<tr>
<td>Control (-)</td>
<td>8.46±0.29&lt;sup&gt;a&lt;/sup&gt;</td>
<td>7.04±0.26&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>Control (+)</td>
<td>7.92±0.26&lt;sup&gt;b&lt;/sup&gt;</td>
<td>6.92±0.11&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>CP</td>
<td>9.42±0.39&lt;sup&gt;a&lt;/sup&gt;</td>
<td>8.86±0.25&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>CNPs</td>
<td>9.52±0.38&lt;sup&gt;a&lt;/sup&gt;</td>
<td>8.94±0.25&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

Data were presented as mean ± SEM (n = 3). Columns carrying different lower-case letters are significantly different at <i>p</i> < 0.05. Rows of the three time points carrying different upper-case letters are significantly different at <i>p</i> < 0.05. Before treatment, the mean of sensory score = 9.54±0.21. Control (−), minced meat without <i>E. coli</i>; control (+), minced meat treated with <i>E. coli</i>, CP, chitosan powder; CNPs, chitosan nanoparticles.

The obtained results revealed higher sensory scores in all samples containing chitosan, which implies the potential of CP and CNPs on preserving sensory characters of minced meat. Our results agreed with those obtained by Alam et al. (2017) who also found higher sensory scores for meet treated with chitosan. These improved sensory characteristics may be due to the higher binding affinity of chitosan to lipid and water (Cao et al., 2013; de Farias et al., 2019; Hu et al., 2015) and its antioxidant properties which could keep red color of meat through inhibition of myoglobin degradation in muscles (Sarbon et al., 2015). Chitosan can also form a nonpermeable barrier against the passage of oxygen into the muscle fibers and subsequently prevent fiber oxidation (Shleikin and Medvedev, 2014).

**CONCLUSIONS**

Chitosan powder and nanoparticles had antibacterial activity against <i>E. coli</i>, with a potent effect for the nanoparticles form. Treatment of minced meat with any of these chitosan products helps in keeping the quality of meat, increasing antioxidant properties, and improving sensory characteristics. Therefore, chitosan, particularly in the form of nanoparticles, can be successfully used as a food preservative in the food industry to maintain quality and extend the shelf life of various food products.

**Statement of conflict of interest**

The authors have declared no conflict of interest.

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Comparison between the Effect of Chitosan Powder and Nanoparticles


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