Antibacterial activity of zinc oxide nanoparticles against *Staphylococcus aureus* in raw chicken breast fillets

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**ABSTRACT**

Zinc oxide nanoparticles (ZnO NP) were compared with bulk ZnO powder at various concentrations (4, 6, and 8 mM) for their antibacterial activity against *Staphylococcus aureus* inoculated into raw chicken breast fillets during refrigerated storage at 4°C. ZnO NP was more efficient as antibacterial agent against *Staphylococcus aureus* than bulk ZnO powder. 8 mM suspension of ZnO NP were the most effective since it reduced the initial load of inoculated *Staphylococcus aureus* from 9.63 to 3.97 log cfu/g (58.77%) within 12 days of refrigerated storage at 4°C. These data suggested that the antibacterial efficiency was indirectly proportional to size and directly proportional to concentration. The promising results of ZnO NP antibacterial activity suggest its usage in food systems as preservative agent in the food industry after further required investigations and risk assessments.

**Key words:** Zinc Oxide nanoparticles, *Staphylococcus aureus*, antibacterial activity.

**INTRODUCTION**

The use of chicken products is increasing due to its convenience for consumer. The increasing demand for meat products is posing challenges for food safety and quality (Jutaporn et al., 2011). Food safety is the major global concern of the consumers and food industries.

Fresh chicken meat is a highly perishable commodity for growth and multiplication of spoilage and pathogenic microorganisms, resulting in foodborne infections and intoxications (Cardenas et al., 2008). One of the major causes of several outbreaks is believed to be lack of or insufficient cleaning and disinfection of equipment, hands and surfaces in food-related environments (Awny et al., 2010). Toxin production by food borne bacteria has raised public concern because of direct and associated food poisoning. Undesirable microbial growth is responsible for most spoilage in meat products, whereas biochemical and enzymatic deteriorations also occur (Devlieghere et al., 2004). Foodborne pathogens cause significant economic losses for food industry.

For many decades, foodborne diseases have been observed as serious threats to public health all over the world. Despite the stringent regulatory systems on food processing and preservation, foodborne diseases incidence are increasing in developing countries. Each year in United States of America, there is an estimated 25 - 81 million foodborne illness cases resulting, approximately 9000 deaths associated with contaminated food consumption (Jevsnik et al., 2013).

The contamination of chicken meat with *Staphylococcus aureus* has been repeatedly reported and is considered as a potentially hazardous food worldwide (Akbar and Anal, 2013). *S. aureus* is a leading cause of food poisoning resulting from the consumption of food contaminated with staphylococcal enterotoxins. Different food can act as a good medium for *S. aureus*, such as raw meat and meat products (Guven et al., 2010). Enterotoxins are highly thermostable; normal cooking and pasteurization cannot totally inactivate them, so they cause food poisoning (Nagarajappa et al., 2012) . The onset of symptoms depends on the amount of enterotoxin ingested. Classic
SEs antigens have been identified as SEA, SEB, SEC1, SEC2, SEC3, SED and SEE (Bergdoll and Robbins, 1973). The enterotoxin genes however, are not uniformly distributed among \textit{S. aureus}.

SEA is the most common enterotoxin recovered from food poisoning outbreaks (Balaban and Rasooly, 2000) and it is known that 59% of staphylococcal food poisoning outbreaks are caused by SEA to SEE (Bergdoll, 1989). \textit{S. aureus} may be life threatening in immunodeficient conditions (Bhunia, 2008).

Reducing the chances of pathogens entry and safeguard of meat products are the primary concern to ensure food safety. The increasing prevalence of antibiotic resistant strains and reemerging of infectious diseases at an alarming rate is a continuous threat to public health in the modern world. Moreover, the accumulation of antibiotics both in the environment and in food is potentially risky to consumers and the environment (Alderman and Hastings, 1998). One way to control microbial growth in these food products, thereby improving safety and delaying spoilage, is the application of antimicrobial dips or sprays on the surface of the product (Kerry et al., 2006). However, in these applications, the efficiency of the antimicrobial substances is restricted due to uncontrolled migration into the food and partial inactivation of the active compounds following interaction(s) with food components (Quintavalla and Vincini, 2002). Such challenges and adverse effects have led to a pressing demand for using alternative effective means and novel technologies to control the spread of foodborne pathogens (Li et al., 2013).

During the past few decades, nanotechnology has emerged and elicited much interest as an innovative promising technology for the synthesis of new types of nanomaterials. These nanomaterials are particles in the nanometer size, with the potential to improve food quality and safety and exhibit antimicrobial effects owing to their small size, high surface area-to-volume ratio enabling intimate interactions with microbial membranes (Allaker, 2010), high fraction of surface atoms and unique, unusual and interesting physical, chemical, and biological properties (Kim et al., 2007), such as catalytic, optical, electronic, magnetic, antimicrobial (Ingle et al., 2008) wound healing and anti-inflammatory properties (Taylor et al., 2005). Metal nanoparticles have various functions that are not observed in bulk phase (Sosa et al., 2003). Recent advances in the field of nanotechnology have led to the development of new biocidal agents. Nano-materials are called “a wonder of modern medicine”. It is stated that antibiotics kill perhaps a half dozen different disease-causing organisms, but nanomaterials can kill some 650 cells (Sungkaworn et al., 2007).

In recent years, inorganic metal oxides such as ZnO, have been explored as antimicrobial agents in food preservation (Mirhosseini and Firouzabadi, 2013), and feasible to incorporate in active food packaging for such properties as ethylene oxidation or oxygen scavenging, which can extend food shelf life (Llorens et al., 2012). Besides, they are not only stable under high temperatures and pressures that may occur in harsh food-processing conditions (Sondi and Sondi, 2004), but they are also GRAS by FDA relative to organic substances. ZnONP are nontoxic, biosafe and biocompatible as it degrades into mineral ions and absorbed by the body within few hours (Zhou et al., 2006).

Zinc is a source of micronutrient and plays an important role in body growth and development (Shi et al., 2008). Many studies have shown that ZnO NP have selective toxicity to bacteria and only exhibit minimal effect on human cells, which recommend their prospective uses in agricultural and food industries (Zhang et al., 2007), providing solutions to the food safety challenges related to extending shelf life, improving food safety, monitoring food spoilage (Ozcalik and Tihminlioglu, 2013) and packaging with economic and environmental friendly ways (Silvestre et al., 2011).

ZnO NP, due to their antimicrobial property and strong adsorption ability, are being incorporated into a variety of successful applications in numerous technological fields, such as optoelectronics, nanodevices, nanoelectronics, nanosensors, optical, piezoelectric, magnetic storage media, gas sensing, catalysis, solar energy transformation, (jiang et al.,1998), electrical engineering, chemistry, material sciences, cosmetics (Kumar, 2006), medical instrument and devices, dermatological applications in creams, lotions and ointments (Martínez Flores et al., 2003), treatment of bacterial infections in skin and burn wounds (Rai et al., 2009), as a mild topical astringent in eczema, slight excoriations, wounds and hemorrhoids (Sweetman, 2005), drug carriers and medical filling materials, that is, materials used to fill a space such as a tooth cavity or root canals, (Zhou et al., 2006; Jiang et al., 2007), pharmaceuticals (Duncan, 2011), diagnosis of chronic diseases (Andersen et al. 2009) and (Hong et al., 2008), wallpapers in hospitals (Richards, 2008), manufacture of sunscreens(Wang et al., 2008), removal of metabolites and contaminants from polluted water (Celebi et al., 2007), linings of food cans in packages for meat, fish, corn and peas to preserve colors and to prevent spoilage (Huang et al., 2008). Antimicrobial compounds can be attached to the cores of nanoparticles and delivered into bacterial cells (Gaysinsky et al., 2004). For instance, ZnO inhibits the adhesion and internalization of enterotoxicogenic \textit{E. coli} (ETEC) into enterocytes (Roselli et al., 2003). In addition, ZnONPs can reduce the attachment and viability of microbes on biomedical surfaces (Braeyner et al., 2006).

Therefore, the main objective of this study was to evaluate the antibacterial activity of ZnO nanoparticles (ZnONP) in comparison with bulk ZnO powder against a foodborne pathogen and a representative microorganism of public concern in food-related environments as \textit{S. aureus} inoculated into raw chicken breast fillets \textit{(in vivo)}, during refrigerated storage (4°C). The information from this study may be of use to researchers and industry personnel who are interested in finding novel ways to...
control pathogenic microorganisms in meat and poultry products.

MATERIALS AND METHODS

Bulk ZnO powder and ZnONPs

The bulk ZnO powder (~5 mm) was purchased from El Gomohoria Company, Egypt. While, ZnO nanoparticles (size of 20 nm; purity 99.98%) (Figure 1) were purchased from NanoTech Egypt for Photo-Elecronics according to NT-ZONP brand with certificate of analysis. The bulk ZnO powder and ZnO NPs suspensions at different concentrations (4, 6 and 8 mM) were prepared analogously and resuspended in 150 ml sterile distilled water and sonicated for 30 min for uniform dispersion and formed a colloidal suspension. Thereafter, the resulting homogenous suspensions were autoclaved for 30 min for sterilization (Mottaki et al., 2014).

Physical properties:
Appearance (Color): White.
Appearance (Form): Powder.
Solubility: Stable colloid in mixture of Methanol and chloroform and water.
Optical Prop. (Abs.): $\lambda_{\text{max}} = 301$ nm, and $380$ nm
Avg. Size (TEM): $20 \pm 5$ nm
Shape (TEM): Spherical shape.

Preparation of bacterial strain

*S. aureus* PTCC1431 strain was obtained from culture collection of Cairo MIRCEN, Agricultural Studies and Consultation Center, Faculty of Agricultural, Ain Shams University, Egypt for use in this study. It was maintained on Tryptic Soy Agar (TSA; Merck, Darmstadt, Germany) at 37°C for 24 h and stored at 4°C. The strain was propagated
twice in Tryptic Soy Broth (TSB; Merck, Darmstadt, Germany) and incubated at 37°C for 16 h before use in subsequent experiment.

Challenge study

A grand total of 1400 g of skinless deboned fresh raw chicken breast fillets were purchased from local markets in Tanta, Gharbia governorate, Egypt. The fillets were free of visible blood splash or bruising. The samples were transferred directly to the laboratory within 15 min under complete aseptic conditions, packed in separate insulated polystyrene boxes with ice. Any remaining surface fat was physically removed after visual inspection. In the laboratory, the chicken meat samples were divided into seven equal groups (200 g each), washed with distilled water and stored at 4°C for 2 h before use. Thereafter, the chicken meat samples were aseptically cut into sections (5 cm × 5 cm) and dipped in 250 ml of peptone water containing S. aureus at approximately 7 log cfu/ml for 15 min at room temperature (25°C). After the inoculation, the chicken fillet samples were kept at room temperature for 20 min to allow for bacterial cell attachment. S. aureus in the inoculated samples was enumerated to get the initial load before dipping treatments were performed.

The inoculated chicken meat samples with known S. aureus load were dipped separately for 2 to 4 min in (i) sterile double-distilled water (control), (ii) 4 mM ZnO, (iii) 6 mM ZnO, (iv) 8 mM ZnO, (v) 4 mM ZnO NP suspension, (vi) 6 mM ZnO NP suspension, and (vii) 8 mM ZnONP suspension. After dipping, the inoculated chicken meat samples were separately packed in multiple sterile labeled low density polyethylene bags, heat sealed, and stored at refrigeration temperature of 2 - 4°C (Dubal et al., 2004). Sensory analysis (overall acceptability) was conducted every 24 h during storage to estimate the shelf life. While St. aureus count was carried out on days 0, 3, 6, 9, and 12 of refrigerated storage, using the serial dilutions and the standard spread plate technique on Baird Parker agar. Tests were performed in triplicate.

Sensory analysis

Overall acceptability of all samples was carried out using a ten - point standardized numerical scale, where ten corresponded to 'components characteristic of the highest quality'. The panel consisted of 10 members of the staff who were familiar with meat characteristics (Kanatt et al., 2010).

Bacteriological analysis

At each sampling interval, 10 g of each sample were aseptically homogenized in 90 of sterile peptone water (0.1%) for 2 min using a stomacher (Stomacher 400 Circulator; Seward Medical Ltd., London, UK). Then, 10-fold decimal serial dilutions (using 0.1% sterile peptone water) were prepared. Subsequently, 1 ml from each of the previously prepared serial dilutions was separately spread plated in duplicate onto Baird Parker agar (BP, Difco) for S. aureus to determine the number of remaining cells. The inoculated plates were incubated at 37°C for 48 h. After incubation, the resulting black shiny colonies with narrow white margins surrounded by a clear halo zone extending into the opaque medium were enumerated, converted to log and the remaining populations were expressed as log cfu/g of sample (Trinetta et al., 2010).

Statistical analysis

The obtained data were statistically analyzed using one-way ANOVA under significance level of P < 0.05 for the obtained results using SPSS package (SPSS 19.0, Chicago, IL, USA). Duncan's post hoc test was used to determine the significance of the differences between mean values. All experiments were performed in triplicate. The results were presented as means ± SD.

RESULTS AND DISCUSSION

Contaminated food is a real threat to human welfare. Foodborne diseases are mainly caused by pathogenic bacteria which are either transmitted to humans from the animal reservoir or which contaminate the food process line. S. aureus currently has gained increasing attention due to its capability of producing a range of enterotoxins and tissue degrading enzymes (Schoeni and Wong, 2005).

When evaluating hurdle approaches to food processing, sensory quality also must be considered to determine the appropriate microbial interventions (Leistner, 2000). Some hurdles influence sensory qualities of products, such as color, flavor, and texture.

The overall acceptability sensory scores of chicken breast fillet samples were assessed every 3 days during refrigerated storage at 4°C. Slight discoloration and slight off-odor indicating initiation of spoilage were evident on 3rd day of storage for chicken meat samples inoculated with S. aureus. Complete spoilage with greenish discoloration and putrid odor was evident on 12th day with the corresponding increases in populations of S. aureus (Table 1).

The results of the present are consistent with recent reports on the strong antibacterial activities of ZnO NPs against major food borne pathogens, such as E. coli O157:H7, B. subtilis, Ps. fluorescens, L. monocytogenes, S. enteritidis, S. typhimurium, S. aureus, S. epidermidis, Str. pyogenes and E. faecalis, in-vitro (Yadav et al., 2006; Qi et al., 2007; Jones et al., 2008; Jiang et al., 2009; Wei et al., 2010; Dutta et al., 2012; Priyanka and Arun, 2013; Morsy et al., 2014). Also, It
has been found to have bactericidal property against Streptococcus, Staphylococcus (Huang et al., 2008), E. coli (Padmavathy and Vijayaraghaven, 2008) and Salmonella (Jin et al., 2009). ZnO quantum dots used as antimicrobial agents significantly inhibited L. monocytogenes and S. enteritidis in liquid egg whites. Similar inhibitory effects of ZnO NPs on S. aureus and E. coli have been observed in milk samples (Mirhosseini and Firouzabadi, 2013).

Meanwhile, according to Adams et al. (2006), ZnO NPs inhibited the growth of Gram-positive bacteria by 90%, but Gram-negative bacteria were much more resistant. It has been reported that S. aureus is more sensitive to ZnO NPs as compared with E. coli and Ps. aeruginosa (Premanaganath et al., 2011).

Although, ZnO NPs have been known to act as antimicrobials, the exact mode of action remains a matter of active research. The antibacterial activity of ZnO NPs is believed to be explained by the following hypothesized mechanisms:

1) The contact between ZnO NPs and bacterial cell is initiated by surface charges on the particle (Neal, 2008) and the electrostatic attraction between negatively charged bacterial cell membrane and positively charged ZnO NPs (Saadati et al., 2011). ZnO NPs were suggested to embed themselves in the cell membrane (Jang et al., 2003). The particle size plays a vital role in the antibacterial activity. The bacterial cell membranes contain pores in nanometer range. ZnO NPs, which have a size less than that of pore size in the bacteria, have a unique property of crossing the cell membrane without any hindrance (Sunita et al., 2011), resulting in the production of toxic oxygen radicals, which damage DNA, cell membranes or cell proteins, and may finally lead to the inhibition of bacterial growth and eventually to bacterial death (Tankhiwale and Bajpai, 2012).

2) After the contact of ZnO NPs with bacterial cell membrane, high rate of intercellular reactive oxygen species (ROS), such as hydroxyl radicals, super-oxides and \( \text{H}_2\text{O}_2 \) are released in the presence of moisture (Tankhiwale and Bajpai, 2012). Because the hydroxyl radicals and super-oxides are negatively charged particles, they cannot penetrate into the cell membrane and must remain in direct contact with the outer surface of bacteria, affect its integrity and causing severe damage to proteins, lipids, and DNA. Whereas, the generated \( \text{H}_2\text{O}_2 \) can penetrate the cell membrane and interact with membrane proteins and kill the bacterial (Bajpai et al., 2012). The cell membrane architecture could be impaired through lipid peroxidation by ROS (Bryan et al., 2006).

3) The release of \( \text{Zn}^{2+} \) ions can damage cell membrane and interact with intracellular contents (Espitia et al., 2012).

4) ZnO NPs bind with thiol (-SH), sulphydryl or disulphide functional groups of protein present in the cell wall, inhibiting enzyme functions and interfering with vital cellular processes (Duncan, 2011).

5) ZnO NPs interact with bacterial cell membrane lipids directly leading to disorganization of the membrane structure, loss of membrane integrity, mitochondrial malfunction, abnormal cell morphology, damage to the cell membrane, decrease in the cell permeability (Krishnamoorthy et al., 2012), and leakage of cytoplasmic contents (Sharma et al., 2010), minerals, proteins and genetic materials, causing cell death (Sangilidayandi, 2014). Therefore, most of the nanoparticle treated cells were ghost cells from which intracellular material was released into the cell suspension. This stress in the cell wall produces more lactate dehydrogenase enzymes, leading to damage of the cell membrane, and the severity depends upon the exposure time (Weihseng et al., 2006).

6) ZnO NPs interact with phosphorus moieties in DNA, resulting in inactivation of DNA replication (Jiang et al., 2009). Bacterial cell nutrients adsorb to the large surface area of ZnO NPs, which starves the bacterial cell.

Mirhosseini and Arjmand (2014) suggested that the antibacterial activity of ZnO NPs was concentration dependent. This fact may be attributed to the difference in generation of \( \text{H}_2\text{O}_2 \) in ZnO and this generation was linearly proportional to their ZnO NPs concentration (Yamamoto et al., 2000). Also, the amount of protein released from the

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**Table 1:** Effect of ZnO NP and bulk ZnO powder on overall acceptability of raw chicken breast fillet samples during refrigerated storage.

<table>
<thead>
<tr>
<th>Groups</th>
<th>Zero day</th>
<th>3rd day</th>
<th>6th day</th>
<th>9th day</th>
<th>12th day</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>8.00 ± 1.00</td>
<td>Spoiled</td>
<td>Spoiled</td>
<td>Spoiled</td>
<td>Spoiled</td>
</tr>
<tr>
<td>4 mM ZnO</td>
<td>7.00 ± 1.00</td>
<td>5.00 ± 1.00c</td>
<td>6.00 ± 1.00b</td>
<td>5.00 ± 1.00c</td>
<td>5.00 ± 1.00c</td>
</tr>
<tr>
<td>6 mM ZnO</td>
<td>7.33 ± 1.53</td>
<td>6.33 ± 0.58b</td>
<td>6.00 ± 1.00c</td>
<td>5.00 ± 1.00c</td>
<td>5.00 ± 1.00c</td>
</tr>
<tr>
<td>8 mM ZnO</td>
<td>7.00 ± 1.00</td>
<td>6.00 ± 1.00c</td>
<td>6.00 ± 1.00b</td>
<td>5.00 ± 1.00c</td>
<td>5.00 ± 1.00c</td>
</tr>
<tr>
<td>4 mM ZnO NP</td>
<td>6.33 ± 0.5b</td>
<td>8.00 ± 1.00a</td>
<td>7.00 ± 1.00a</td>
<td>4.67 ± 0.58b</td>
<td>7.00 ± 1.00a</td>
</tr>
<tr>
<td>6 mM ZnO NP</td>
<td>8.33 ± 0.58a</td>
<td>8.33 ± 0.58a</td>
<td>7.00 ± 1.00a</td>
<td>7.67 ± 0.58b</td>
<td>7.00 ± 0.00</td>
</tr>
<tr>
<td>8 mM ZnO NP</td>
<td>6.67 ± 0.58a</td>
<td>7.67 ± 0.58a</td>
<td>6.67 ± 0.58a</td>
<td>7.00 ± 1.00a</td>
<td>6.67 ± 0.58a</td>
</tr>
</tbody>
</table>

The values represent Mean ± SD of three experiments.

Means within a column followed by different letters are significantly different (\( P < 0.05 \)).

Score System for Sensory Evaluation (Kanatt et al., 2010):

9: Excellent; 6: Good; 3: Poor.

8: Very very good; 5: Medium; 2: Very poor.

7: Very good; 4: Fair; 1: Very very poor.
bacterial cells increased along with increasing concentration and contact period of ZnO NPs (Zhang et al., 2008).

Generally, in this study, the antibacterial activity of ZnO NP (20 nm) was much stronger and effective than that of bulk ZnO powder (~5 mm) (Table 2 and Figure 2). This is in agreement with the findings of (Pal et al., 2007; Padmavathy and Vijayaraghavan, 2008; Tayel et al., 2011; Mostafa-Azza, 2015).

This could explain the fact that the antimicrobial activity of the nanoparticles is known to be a function of the size of nanoparticles and the surface area in contact with the microorganisms. The generation of $\text{H}_2\text{O}_2$ depends strongly on the surface area of ZnO. As such, with decreasing particle size, the number of ZnO powder particles per unit volume of powder slurry increases, resulting in increased surface area, increased generation of $\text{H}_2\text{O}_2$, more production of ROS on the surface and higher antibacterial activity of the smaller nanoparticles with microbial membranes and higher cytotoxicity (Yamamoto et al., 2008).

Taken together with previous studies, nanoparticles are more abrasive in nature than bulk ZnO, thus contributing to the greater mechanical damage to the cell membrane which result in enhanced bactericidal effect (Padmavathy and Vijayaraghavan, 2008).

In conclusion, ZnO NPs exhibited higher remarkable antibacterial activity as compared with bulk ZnO powder against $S.\text{ aureus}$ in meat. Their antibacterial efficiency was indirectly proportional to size and directly proportional to concentration. The above findings suggest that ZnO NPs constitute an effective, inexpensive and alternative antimicrobial agent against $S.\text{ aureus}$ over conventional chemical antimicrobial agents, and provide a novel way to enhance the safety and shelf-life in food systems. Therefore, they are promising and have good potential in many food applications.

However, more research is needed to determine the

Table 2: Effect of different concentrations of ZnO NP and bulk ZnO powder on Staphylococcus aureus count (log cfu/g) artificially inoculated into raw chicken breast fillet samples during refrigerated storage.

<table>
<thead>
<tr>
<th>Groups</th>
<th>Zero day</th>
<th>3rd day</th>
<th>6th day</th>
<th>9th day</th>
<th>12th day</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>7.66 ± 0.39 a</td>
<td>Spoiled</td>
<td>Spoiled</td>
<td>Spoiled</td>
<td>Spoiled</td>
</tr>
<tr>
<td>4 mM ZnO</td>
<td>7.64 ± 0.38 a</td>
<td>6.42 ± 1.14 a</td>
<td>Spoiled</td>
<td>Spoiled</td>
<td>Spoiled</td>
</tr>
<tr>
<td>6 mM ZnO</td>
<td>7.61 ± 0.32 a</td>
<td>6.15 ± 0.36 a</td>
<td>5.85 ± 0.26 a</td>
<td>5.82 ± 1.22 a</td>
<td>Spoiled</td>
</tr>
<tr>
<td>8 mM ZnO NP</td>
<td>7.60 ± 0.34 a</td>
<td>6.11 ± 0.78 a</td>
<td>5.83 ± 0.21 a</td>
<td>5.70 ± 0.35 a</td>
<td>Spoiled</td>
</tr>
<tr>
<td>4 mM ZnO NP</td>
<td>7.58 ± 0.34 a</td>
<td>5.91 ± 0.38 a</td>
<td>5.75 ± 0.55 a</td>
<td>5.57 ± 0.06 a</td>
<td>5.27 ± 0.27</td>
</tr>
<tr>
<td>6 mM ZnO NP</td>
<td>7.55 ± 0.34 a</td>
<td>5.87 ± 0.65 b</td>
<td>5.74 ± 0.51 a</td>
<td>5.57 ± 0.06 a</td>
<td>5.27 ± 0.27</td>
</tr>
<tr>
<td>8 mM ZnO NP</td>
<td>7.52 ± 0.34 a</td>
<td>5.77 ± 0.48 ab</td>
<td>5.50 ± 0.25 b</td>
<td>4.86 ± 0.18 b</td>
<td>3.97 ± 3.45</td>
</tr>
</tbody>
</table>

Initial load of $S.\text{ aureus}$ = 9.63 ± 0.35 log cfu/g

The values represent Mean ± SD of three experiments.

Means within a column followed by different letters are significantly different ($P < 0.05$).

Figure 2: Reduction % of Staphylococcus aureus count (log cfu/g) artificially inoculated into raw chicken breast fillet samples treated with different concentrations of ZnO NP and bulk ZnO powder.
stability of ZnO NPs under various conditions (temperature, humidity, light, and so on), mechanical properties (strength, elasticity, and so on) and sensory properties. Further long-term toxicity, mutagenicity and carcinogenicity studies are required to clarify any adverse effects, and are necessary to support the safe use of ZnO NPs.

REFERENCES


