Benha University
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"Improvement of Meat Quality Using nanotechnology"

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SUMMARY

Antibacterial nanoparticles are introduced as a new method to prevent microbial food spoilage as TiO2, ZnO and mixed TiO2–ZnO nanoparticle represented great antibacterial action.

In our study there are two experimental parts

First paper

Antibacterial Efficacy of Zinc Oxide and Titanium Dioxide Nanoparticles against *Escherichia coli* in Minced Meat

The *in-vitro* examination for the antibacterial effect of different concentrations of different types of nanoparticles as ZnO and TiO2 revealed that ZnO NPS have no inhibition zone but the inhibition zone start to appear with concentration of 3mM and the maximum inhibition zone appeared at concentration of 12mM, also TiO2 NPS have no inhibition zone at 3mM, while the maximum inhibition zone diameter was at concentration of 12mM.

After that Fresh minced beef was purchased and immediately transported to the laboratory in an icebox and stored at 4 °C until use. Thin sheets of minced beef were treated with ultraviolet light (wavelength 385 nm) for 30 min, 15 min to each side to eliminate background microflora, and inoculated by ~ 6 logs CFU/g of minced meat. Then, they were mixed thoroughly by gently squeezing the bags by hand till even distribution of microbe occurred, and left for 30 min for complete attachment between inoculated *E. coli* and minced meat. Minced meat sample was divided into six groups (200 g each); Group 1 (PBS + *E. coli*), Group 2 (6mM ZnO + *E. coli*), Group 3 (6mM TiO2 + *E. coli*), Group 4 (12mM ZnO + *E. coli)*, Group 5 (12mM TiO2 + *E. coli*), and Group 6 (6mM ZnO + 6mM TiO2 + *E. coli*). Counting of *E. coli* and

sensory evaluation were performed on days 0, 3, 6, 9, 12, 15, and 17of chilling storage. The experiment was repeated in triplicate for each group and mean values were calculated.

In minced meat, 12 mM (ZnO) were identified as the best case to improve shelf life and prevent *E. coli* growth in minced meat as it decreased the count by ~ 5 log (CFU/g) that become 1.31 log (CFU/g).Also the results obtained by mixture of ZnO +TiO2 (6 mM) 2.02 log (CFU/g) and TiO2 (12 mM) 2.92 log (CFU/g) which decreased the count by ~ 4 log (CFU/g). Also, ZnO (6 mM) 4.30 log (CFU/g) and TiO2 (6 mM) 4.27 log (CFU/g) that decreased the count by ~ 2log (CFU/g).

By observation to the sensory parameters (color, odor and texture) throughout the storage period of minced meat the overall acceptability showed that, the concentrations of 12 mM (ZnO), 12mM (TiO2) and mixture of (6mm) (ZnO + TiO2) enhanced shelf life time of minced meat and delayed its spoilage until 17^{th} day, while minced meat treated with concentrations of 6mM ZnO and 6mM TiO2 spoiled on 15^{th} day and control spoiled before 6^{th} day of storage.

Also the present study used transmission electron microscope for examination of the damage effect of these NPS (ZnO, TiO2 and mixture of (ZnO+ TiO2)) on *E. coli* and differentiate it from normal structure. ZnO NPS caused severe damage for cell as made pores in cell wall that cause elongation and swelling end with rupture for bacterial cell. Mixture of (ZnO+TiO2) NPS also have anti-bacterial effect as they accumulated around bacterial cell and caused elongation of bacterial cell and disruption of cytoplasm. TiO2 caused elongation of bacterial cell, thinning of cell wall and pores in plasma membrane**.**

Second paper

Nanotechnological enhancement of meat balls quality

Homogeneous dough were obtained from mixing of minced meat and all ingredients of spices, and then divided into 4 groups. 1st group Zn O (12mM) was added, 2nd group TiO2 (12mM), 3rd group Mix (6mM Zn O+6mM Ti O2 50%:50%) and 4th group control one without nanoparticles. Meatballs (50 ± 2 g) were formed by hand, placed into plastic trays, sealed with one layer of a wrapping film, and stored at 4 ± 1 °C for 20 days. The samples were taken for microbiological, physicochemical and sensory analysis at 0, 3, 6, 9, 12, 15, 17 and 20 days of chilling storage. This study was repeated in triplicate for three different sources of meat.

In meat balls, APC has been decreased ~3log CFU/gm in samples treated with (12mM (ZnO) and 12mM(TiO2)), decreased ~2 log CFU/gm in samples treated with a mixture of 6mM (ZnO + TiO2),While control one increase ~3log CFU/gm. Also Psychrotrophic bacterial count (PTC) has been decreased ~3log CFU in samples treated with ZnO 12mM , decreased ~2 log CFU in samples treated with12mM (TiO2), mixture 6mM (ZnO + TiO2), while control one increase ~2log CFU (6.79 log CFU) which accompanied by slime formation and putrefactive odor.

Moreover, Staphylococci bacterial count becomes 1.41, 1.69 and 1.83 (log CFU/gm.) for 12mM (ZnO), 12mM (TiO2) and mixture 6mM (ZnO and TiO2) respectively, while the count for control one become 5.40 log CFU/gm. The result proved that Coliforms count decrease to 1.43, 1.92 and 2.14 (log CFU/gm.) for 12 mM (ZnO), 12 mM (TiO2) and mixture 6mM (Zn O + TiO2) respectively, while the count for control one become 6.86 log CFU/gm.

The physicochemical properties of meat balls also improved due to the usage of ZnO, TiO2 and their mixture. The pH values were 7.2, 7.4 and 7.6 for ZnO 12mM, TiO2 (12mM) and a mixture (ZnO and TiO_2 (6mM)), respectively and 8.05 for control one at the end of challenge study. TVB-N values were 22.2, 30.4 and 32.9, (mg/100gm) for ZnO (12mM), Ti O2 (12mM) and a mixture (ZnO and TiO_2 (6mM)) respectively and 45.4 mg/100mg for control one at the end of challenge study. TBA values were 0.92, 1.1 and 1.1 (mg malonaldehyde /kg) for Zn O (12mM), TiO2 (12mM) and mixture of 6mM (ZnO and TiO2) respectively, and 1.90 mg malonaldehyde /kg for control one at the end of challenge study.

Moreover, the acceptability (odor, texture and color) of meat balls during storage at 4^{o}c also affected by using nanoparticles of ZnO, TiO2 and their mixture. The data also reveals that the shelf life of meat balls extended till17^{th} day and spoiled at 20^{th} day of chiller storage. When treated with ZnO (12mM) become 4.1±0.2, then samples treated with Ti O2 (12mM) become 4.1±0.2 at 17^{th} day and mixture of 6mM (ZnO and TiO2) become 3.6±0.1at 17^{th} day of chiller storage. The control one spoiled at 9^{th} day (4±0.01).

The benefits of nanotechnology use are numerous and this technology offer lot of possibilities in different fields especially meat industry. The use of nanoparticles as ZnO and TiO2 could be a fully effective method to improve meat safety and shelf life, but good governance and regulatory framework of the application of nanotechnology within food should be implemented. This clearly needs close collaborations between nanoparticles (and products) developers, risk assessors, regulators and researchers.