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EFFECT OF ZINC OXIDE NANOPARTICLES ON SHELF LIFE OF COMMONLY USED VEGETABLES

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Abstract

Most of the harvested fruits and vegetables cannot be stored in natural conditions for satisfactory shelf life duration due to their perishable nature. Applications of nanotechnology to fruits and vegetables shelf life extension may overcome the shortcomings of traditional preservation technologies because nanotechnology has many unique properties. The aim of this study was to examine the effect of zinc oxide nanoparticles (ZnO NPs) synthesized from *Nigella sativa* extract on shelf life of cucumber and tomato. Nanoparticles were characterized by using UV-Vis spectrophotometry and particle size analyzer. Vegetables were soaked in tap water, distilled water and treated with different concentrations of ZnO NPs (0.05, 0.1, 0.2 and 0.5 g/100 mL). The effect of ZnO NPs on vegetables quality factor like external appearance, weight loss measurement, carotenoid content and total phenolic content was observed. The result showed that ZnO NPs reduced weight loss, delayed disease development and maintained firmness to extend shelf life of vegetables.

Key words: cucumber, *Nigella sativa*, nanoparticles, shelf life, tomato, UV-Vis spectrophotometry

Introduction. Fruits and vegetables are important source of nutrients in daily human diet. It has been stated that about 20–40% of fruits and vegetables are spoiled and deteriorated worldwide due to lack of proper shelf life extension

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methods [1]. In many agriculture based countries, the main problem is to keep the fruits and vegetables safe and fresh until they reach the consumer hands. Although fruits and vegetables have a natural waxy coating, it is not sufficient to offer protection against water loss, high respiration rate and microbial attacks [2]. Therefore prolongation of shelf life is an important issue in the food industry for increasing concerns related to food security worldwide when most of the world population is facing the problem of food security, particularly the low income countries [3].

The history of food preservation dates back to ancient civilization. Researchers have developed various shelf life extension technologies. Some of them are cold storage, heat treatment, chemical based preservations and irradiation. But these technologies have many disadvantages, they are not suitable for all types of fruits and there is always a space for innovation. Furthermore, these technologies are not available in all countries which have different average temperatures and different population to land ratio. These create problems with the applications of food storage technologies. To overcome the shortcomings of these technologies there is a need to develop technologies which are safe, easy to use and environmental friendly [4].

Recently, nanotechnology has been widely employed in agricultural production. Nanomaterials are used against various pathogenic microbes and food preservation due to their excellent physiochemical nature and antimicrobial potential [5]. Nanomaterials can be considered a viable solution to overcome the disadvantages of traditional preservation technologies [6]. Coating of nanomaterials maintains the quality and extends shelf life of fruits and vegetables. Coating provides moisture barrier to prevent gas exchange between vegetable and surroundings and to reduce loss of bioactive compounds. It is also helpful by reducing the action of microbes at the fruit surface, hence decreasing the fruit deterioration for a longer period of time [7].

Zinc oxide is an inorganic and biosafe material, non-toxic to humans and is also an antimicrobial agent [8]. It damages cell membrane of bacteria and enhances membrane permeability. ZnO NPs have good biocompatibility to human cells and have been generally recognized as safe by US Food and Drug Administration [9]. In the present research work, various concentrations of zinc oxide (phyto-mediated) were used to preserve cucumbers and tomatoes at two different storage temperatures i.e., room temperature (25 °C) and cold storage (4 °C). This study aims to find the effect of zinc oxide nanoparticles on increasing the storage capacity of tomato and cucumber.

Materials and methods. ***Plant material.*** *N. sativa* was purchased from local market for the bio-fabricated synthesis of zinc nanoparticles. *Solanum lycopersicum* (tomato) and *Cucumis sativus* (cucumber) were also purchased from local market as experimental organisms. These vegetables were of eating quality without any damage or blemishes on their surfaces.

Preparation of ZnO nanoparticles. Nanoparticles were prepared by using the slightly modified method of [10]. Powdered *N. sativa* was weighed (10 g) accurately by using electric balance. Plant material (10 g) was mixed with 50 mL of distilled water and stirred thoroughly. The mixture was exposed to microwave (700 W) for 150 seconds and filtered. For synthesis of nanoparticles the filtrate was used. 0.2 M salt solution was prepared by adding 49 mL water, 0.5 mL acetic acid and 1.61 g $ZnSO_4$. For nanoparticles formation 10 mL of 0.2 M $ZnSO_4$ solution was added to 20 mL of plant extract. In Eppendorf's 1.5 mL solution was taken and centrifuged at 10000 rpm for 15 min. In the form of pellets nanoparticles were separated, washed with distilled water and stored in refrigerator for further analysis.

Characterization of ZnO nanoparticles. Characterization of the ZnO nanoparticles was carried out using UV-Visible spectroscopy. The reduction of zinc ions to the nanoparticles was monitored by measuring the UV-Visible spectra of the solutions. Furthermore, particle size and size distribution of NPs were analyzed by particle size analyzer.

Preparation of concentration of ZnO nanoparticles. Different concentrations (0.05, 0.1, 0.2 and 0.5 g/100 mL) of ZnO nanoparticles were prepared in distilled water.

Sample preparation. Fresh vegetables were washed with tap water followed by a washing with distilled water. These vegetables were then air dried and soaked in tap water, distilled water, 0.05 g/100 mL, 0.1 g/100 mL, 0.2 g/ 100 mL and 0.5 g/100 mL of ZnO nanoparticles treatments for 10 minutes in bowls. After 10 min half of the vegetables from each bowl were taken and put on shelf and the other half were stored in fridge. After 3 weeks different parameters (colour, weight loss measurement, total phenolic content and carotenoid content) were observed.

Measurement and analysis. *Colour parameter.* The colour of vegetables in control and experimental group was observed after an interval of 5 days. Physical appearance of vegetables was also noted for its firmness, freshness and microbial growth as well.

Weight loss. The effect of ZnO NPs on weight loss (%) of vegetables was determined by measuring the weight of the fresh fruits on a digital balance and then comparing the difference with initial weight before storage (W_o) with final weight after storage in the indicated period (W_f). The weight loss percentage was calculated by the following equation

$$\text{Weight loss } (\%) = \frac{(W_o - W_f) \times 100}{W_o}.$$

Total phenolic content. One gram of vegetables was added to 5 mL of 70% (v/v) methanol and homogenized for 1 min. The solution was centrifuged at 15000 rpm for 15 min and supernatant was used for total phenolic content analysis. The total phenolic content was determined by Folin-Ciocalteu assay using UV-Vis

spectrophotometer. The fruit sample extract (250 µL) was mixed with 1250 µL of Folin-Ciocalteu reagent and 1000 µL of 7.5% Na_2CO_3. Mixture thus formed was incubated for 60 min at room temperature. Gallic acid was used as standard. The absorbance of the reaction mixture was measured at 765 nm.

Measurement of carotenoids. 0.55 g of vegetables were weighed and crushed with pestle and mortar one by one and 5 mL of 80% acetone (20 mL distilled water and 80 mL acetone) was added to it and filtered. Absorbance was measured at 470 nm, 645 nm and 663 nm (BMS, UV-2600). The value of chlorophyll *a*, chlorophyll *b*, total chlorophyll and carotenoid was measured by the formula of Arnon and Whatley [11].

Statistics. Results were statistically analyzed by applying one way analysis of variance. Means were compared then by applying Duncan's Multiple Range Test as post hoc test at 5% level of significance. SPSS software was used for this purpose.

Results and discussion. ***Synthesis and characterization of ZnO NPs.*** ZnO NPs were characterized by colour change, UV-Vis spectroscopy and particle size analysis. Figure 1 shows absorption peak at 370 nm. Previous studies indicated the successful synthesis of ZnO NPs if absorption peak is between 348–380 nm [12]. Sharp absorption peak of ZnO NPs also shows the monodisperse nature of formed NPs. By following the simplified mathematical formula of Talam et al. [13] the size of NPs was found to be 2.7 nm. Particle size analyzer showed an average particle size ranging from 2 to 28 nm.

Effect of ZnO NPs on appearance of cucumbers and tomatoes stored at room temperature and in refrigerator. Quality of food can be determined

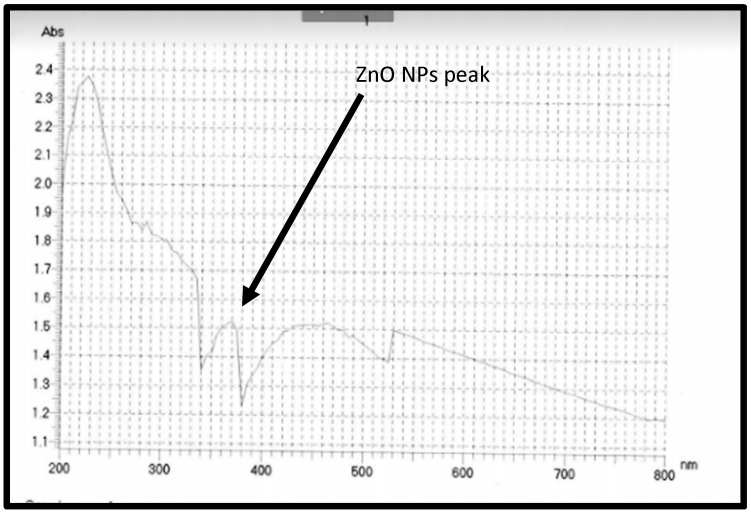

Fig. 1. UV-visible spectra of plant extract and zinc salt mixture after 6 hours

by firmness, colour and spoilage of vegetables by fungus and bacteria. Figures 2 and 3 show the shelf life of cucumbers and tomatoes stored at room temperature, respectively. The shelf life of cucumbers and tomatoes was found to be significantly better when treated with 0.5 g/100 mL of ZnO NPs. It was observed that they maintained their firmness, shape and remained unaffected by microorganisms. Furthermore, it was observed that the cucumbers and tomatoes stored in refrigerator at 4 °C also showed that the storage capacity increased at 0.2 g/100 mL and 0.5 g/100 mL of ZnO NPs. They remained firm and fresh. Nanoparticles can extend the shelf life of fruits and vegetables by inhibiting the reproduction of microorganisms and killing individual microorganisms [14]. Nanoparticles are known to reduce the peroxidation rate of cells, thus increasing the shelf life of fruits and vegetables [15]. Post-harvest treatment of fruits and vegetables with

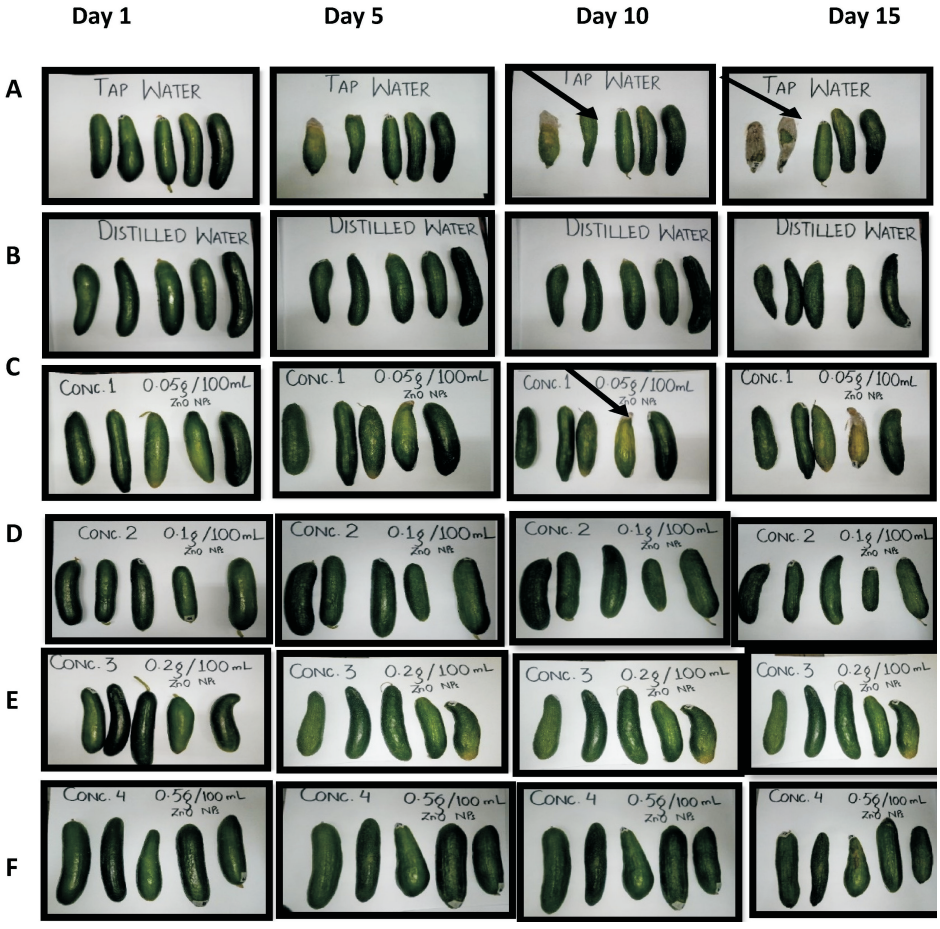

Fig. 2. Appearance of cucumber treated with A: Tap water; B: Distilled water; C: 0.05 g/100 mL ZnONPs; D: 0.1 g/100 mL ZnONPs; E: 0.2 g/100 mL ZnONPs; F: 0.5 g/100 mL ZnONPs and stored at room temperature

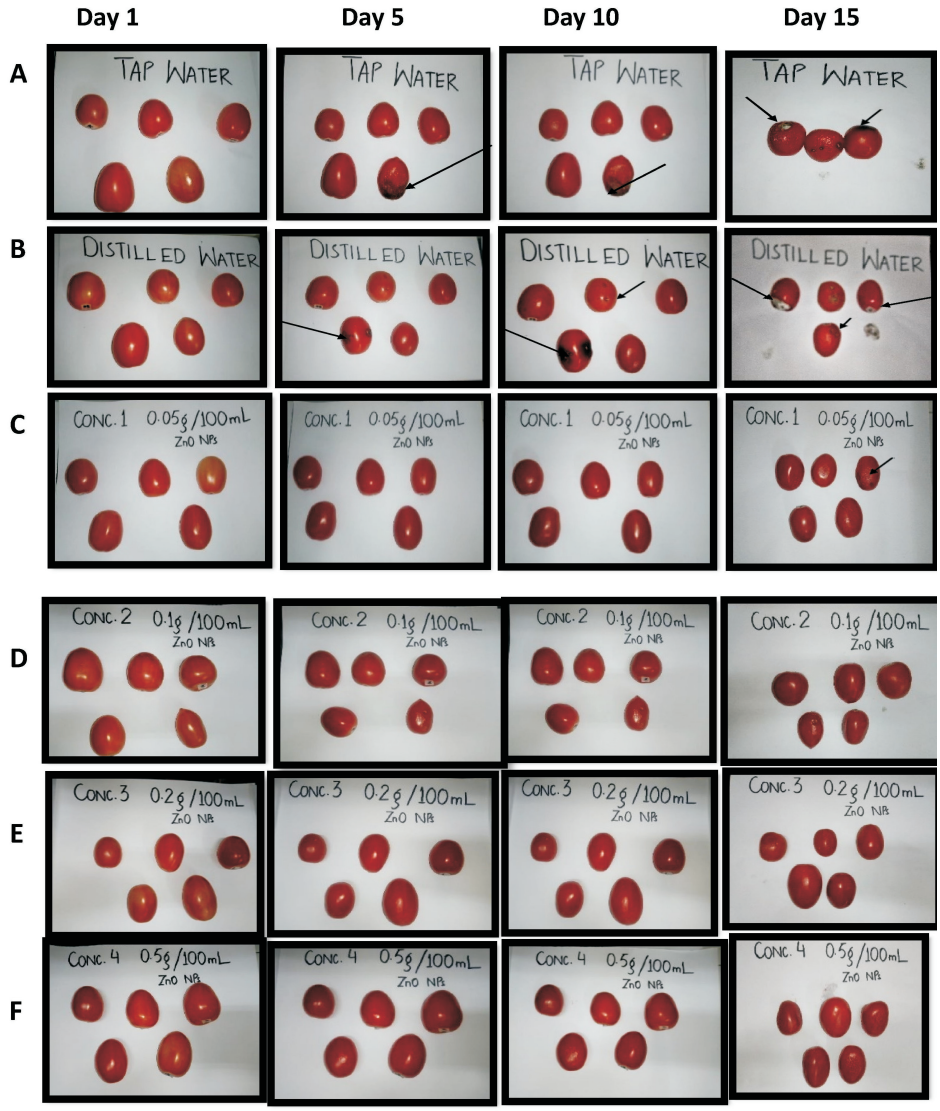

Fig. 3. Appearance of tomatoes treated with A: Tap water; B: Distilled water; C: 0.05 g/100 mL ZnONPs; D: 0.1 g/100 mL ZnONPs; E: 0.2 g/100 mL ZnONPs; F: 0.5 g/100 mL ZnONPs and stored at room temperature

nanoparticles has been reported earlier to enhance their shelf life.

Effect of ZnO NPs on percentage fresh weight of cucumbers and tomatoes. The weight loss percentage of cucumbers and tomatoes stored at room temperature reduced when treated with 0.5 g/100 mL of ZnO NPs compared with cucumbers and tomatoes present in control group. The highest weight loss

was noticed in control treatment of cucumbers and tomato fruit i.e., 58.98% and 19.78%, respectively. The weight loss of nanoparticles coated cucumbers and tomato was noticed to be minimum in 0.5 g/100 mL treated vegetable (1% and 5%, respectively) as shown in Fig. 4a. ZnO NPs reduced weight loss percentage in tomatoes and persimmon fruits which might be due to coating created modified atmosphere around fruit surface and surrounding [16].

The weight loss percentage of cucumber and tomatoes stored in refrigerator at 4 °C also decreased when treated with ZnO NPs and it was also lesser than vegetables stored at room temperature. The highest weight loss in cucumbers and tomatoes in control group in CS (cold storage) was 41.26% and 11.59%. The weight loss percentage in cucumber and tomatoes treated with 0.5 g/100 mL of ZnO NPs was 9.68% and 4.29% only as shown in Fig. 4a. It has also been reported earlier that tomato stored at the chilling temperature (4 °C) showed the lowest weight loss compared to the fruits stored at non-chilling temperature (20 and 30 °C) [17]. The decrease in weight loss due to nanoparticles coating may be due to different diffusion rates of water environment around the fruits or vegetables. It is also attributed to the reduced postharvest microbial diseases. Postharvest diseases of tomato include grey mold and soft rot [18], whereas fungal rots and mosaics are the common problems associated with cucumber storage. When these pathogens are grown on the surface of fruits, they deteriorate the quality of fruits. Whereas nanoparticles prevent the development of microbial growth on the surface of fruits which is also evident in Fig. 2, 3.

Effect of ZnO NPs on total phenolic content of cucumbers and tomatoes stored at room temperature and in refrigerator. Figure 4b shows that the phenolic content of tomatoes stored at room temperature and in refrigerator was reduced significantly at 0.5 g/100 mL ZnO NPs treatment compared to the tomatoes in control group. Similarly, cucumbers at room temperature also had decreased phenolic content as compared to the same control group. But cucumbers stored in refrigerator showed almost the same phenolic content for control treatment as well as nanoparticle treatment. Earlier it was reported that the nano-$CaCO_3$-LDPE packaging significantly inhibited the increase of total phenolic and malondialdehyde content in yam [19]. Nanoparticles have been reported to increase the antioxidant chemicals inside the cell but it is also reported that a dose can be optimized for nanoparticles (depending upon their type and vegetable under study) which can have positive effect on stress tolerance of preserved fruits. This decrease in total phenolic content may be due to reduced stress on plants. Further study can be suggested to find the individual phenolics in fruits.

Effect of ZnO NPs on carotenoid content of cucumbers and tomatoes stored at room temperature and in refrigerator at 4 °C. The carotenoid content in cucumbers stored at room temperature and in refrigerator at 4 °C was highest when treated with 0.5 g/100 mL of ZnO NPs which means that the stress on plants was reduced as shown in Fig. 4c. The carotenoid content of tomatoes

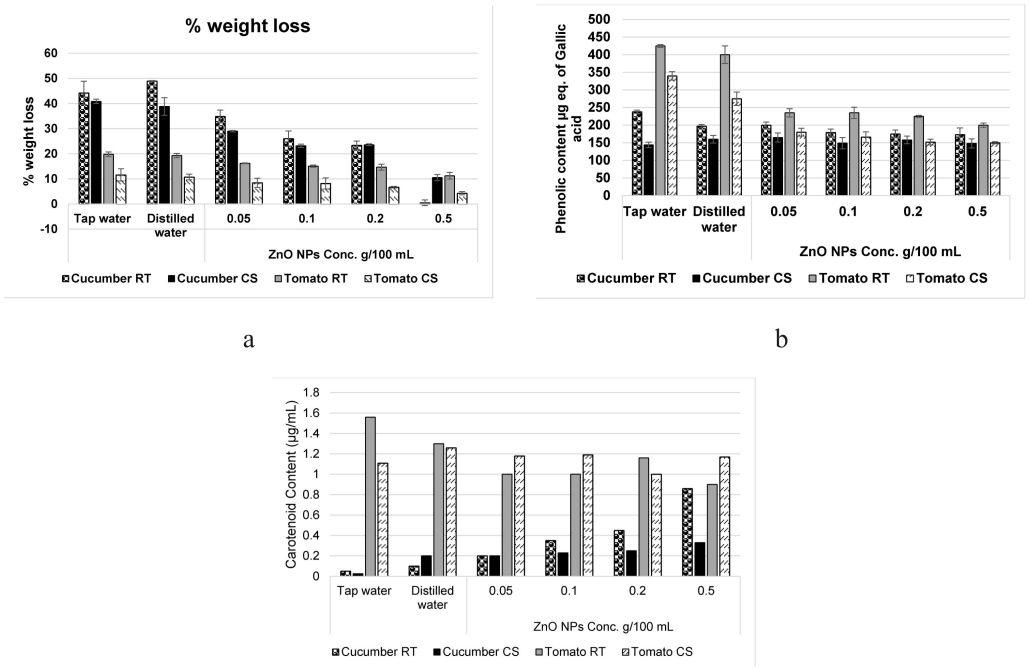

Fig. 4. Effect of ZnO NPs on: a) percentage fresh weight; b) phenolic content of cucumbers and tomatoes at room temperature (RT) and cold storage (CS); c) carotenoid content of cucumbers and tomatoes at room temperature (RT) and cold storage (CS)

decreased when treated with 0.5 g/100 mL of ZnO NPs and stored at room temperature. The tomatoes stored in refrigerator at 4 °C also showed decrease in carotenoid content at 0.2 g/100 mL of ZnO NPs as shown in Fig. 4c. In tomatoes, treated with 0.5 g/100 mL ZnO NPs and stored at refrigerated conditions, was observed, although a small increment in the content of carotenoids, compared to the same, treated with tap water. One of the most noticeable characteristics of ripening is the dramatic increase in the carotenoid content of the fruit. The carotenoid synthesis in tomatoes stored at 4 °C was slowed down due to chilling stress [20].

Conclusions. On the basis of the above finding, the present study concludes that ZnO NPs extended the shelf life of vegetables by decreasing weight loss and phenolic content. Nanoparticles also have antimicrobial potential that aid in reducing the chances of infection on the surface of fruits and vegetables, which in turn enhances their shelf life which is clear from the study of appearance of vegetables. Nanotechnology related preservation technologies can efficiently extend the shelf life of vegetables and ensure the better quality of products during storage.

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