RESEARCH PAPER

Application of Carboxymethyl Cellulose Coating with Different Concentrations of Copper and Zinc Nanoparticles to Improve the Quality Parameters of Cucumber During Storage

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ABSTRACT

Cucumber is a highly perishable crop and it is very important to reduce the chemical and microbial changes that occur during storage. The use of new technologies such as edible coatings combined with metal nanoparticles, can improving cucumber storage and preserving their quality. In this study, the effects of nano coating materials such as Copper nanoparticles (Cu-NPs) and Zinc nanoparticles (Zn-NPs) with the addition of carboxymethyl cellulose (CMC) as an edible coating to increase the quality of cucumber during storage were investigated. The extract of Rosmarinus officinalis was applied to green synthesis of Cu-NPs and Zn-NPs. Characterizations of these nanoparticles were tested by scanning electron microscopy (SEM) and X-ray diffraction (XRD). The size of synthesized nanoparticles was at about 30-70 nm. Cucumber fruits were treated with different concentrations of Cu-NPs and Zn-NPs, ranging from 0% to 3% at 24 °C for 12 days. These treatments were conducted both with a coating of CMC (0.5 %) and without any coating. The results indicated that cucumbers coated with 3% Zn-NPs + CMC had a significant delay in weight loss, firmness, titratable acidity, total chlorophyll content, vitamin C and fungal growth counts compared to uncoated and control treatments. Also, higher concentrations of nanoparticles in coatings provide stronger antimicrobial and antioxidant effects, leading to better preservation of the cucumber's visual quality.

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INTRODUCTION

The United Nations has projected that the world's population will reach 9.1 billion by 2050, and this population growth has significant implications for food security and human livelihoods [1]. Minimizing postharvest losses is a crucial pathway to ensure food availability, rather than solely focusing on increasing food production. Postharvest losses refer to any changes in the quality and quantity of produce that occur after harvest, resulting in the prevention of future use or a reduction in its marketable value. These

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losses can occur due to various factors such as improper handling, inadequate storage facilities, transportation issues, and lack of postharvest management practices. Addressing postharvest losses is important for several reasons. Firstly, it helps to maximize the utilization of the existing food supply, reducing the need for producing additional food to meet growing demands [2]. Cucumber fruit (*Cucumis sativus* L.) belongs to the Cucurbitaceae family and can be cultivated in many countries of the world [3]. Cucumber is considered a highly perishable crop and several factors, including growing conditions, transportation, and storage, can significantly affect its quality and shelf life [4]. Postharvest storage conditions play a crucial role in maintaining the quality of cucumbers. Improper storage conditions can lead to various chemical and physical changes in the fruit, resulting in quality deterioration and increased postharvest losses. Some of these changes include moisture loss, yellowing, changes in firmness, physiological injuries, shriveling, and microbial growth [5]. Use of technologies such as edible coatings, like carboxymethyl cellulose (CMC) combined with metal nanoparticles, can help mitigate some of the chemical and microbial changes that occur during cucumber storage [6]. Carboxymethyl cellulose (CMC) is a derivative of cellulose, which is a renewable and biodegradable polymer. It is known for its film-forming properties due to its high molecular weight chain structure. CMC coatings are cost-effective, non-toxic, and environmentally friendly [7]. The coating acts as a protective barrier, slowing down moisture loss, reducing yellowing, maintaining firmness, and minimizing physiological injuries and microbial growth. By creating a barrier to oxygen and moisture, CMC coatings help preserve the quality of cucumbers and extend their shelf life [3,8]. Copper and Zinc are promising metals with antimicrobial activity, which enhanced the antimicrobial activity by transforming them into nanoparticles. The physical and chemical properties of these nanoparticles are significantly different from their conventional counterparts [9]. Overall, the combination of copper or zinc nanoparticles with CMC can further inhibit microbial growth on the surface of cucumbers during storage. This synergistic effect enhances the overall effectiveness of the coating in reducing the degradation rate of cucumbers [10].

It's important to note that more research and development are needed to fine-tune the formulations and methods of applying these coatings to different fruits and vegetables, including cucumbers. The aim of this study is to assess how a nano-coating material, consisting of Zinc and Copper nanoparticles combined with CMC, can improve the quality of cucumbers stored at a temperature of 23 °C.

MATERIALS AND METHODS

Biosynthesis of Copper and Zinc nanoparticles

According in our previous study, we synthesized Copper and Zinc nanoparticles using rosemary leaf extract. Rosemary leaf extract was utilized as a natural and eco-friendly reducing and stabilizing agent to produce these nanoparticles. This approach offers a sustainable and green method for synthesizing nanoparticles, avoiding the use of harsh chemicals and reducing the environmental impact [9].

Characterization of synthesized nanoparticles

The morphology and size of the synthesized Copper and Zinc nanoparticles were characterized using Scanning Electron Microscopy (VEGA TESCAN-JEOL/EO JSM-5600) (TEM - JEOL, Tokyo, Japan). Also, the formation of these nanoparticles using X-ray diffraction (XRD) with a Ni filter and Cu Kα radiation (λ =0.15406 nm), on the PANalytical X'pert PRO powder X-Ray diffractometer.

Preparation of Coating Solutions

An edible coating solution was prepared according to the method described by Yulian et al. 2019. Carboxymethyl cellulose (CMC; 0.5 %) was prepared by dissolving 5 g of CMC powder (Sigma-Aldrich, Merck, USA) in 1000 mL of distilled water mixture at 75 °C by stirring at 800 pm for 30 min. Next, 0.25 % glycerol monostearate was added to the mixture, and the solution was stirred under the same conditions for 15 min. The addition of glycerol as a plasticizer helps improve the flexibility and film-forming properties of the CMC coating, contributing to its effectiveness in providing a protective barrier for cucumbers during storage.

Preparation of Nano-coating Solutions

Nano-coating solution was prepared by dispersing 0, 0.5%, 1.5%, 3% of Copper and Zinc nanoparticles, synthesized using rosemary leaf extract in the prepared Carboxymethyl cellulose (CMC) solution. The dispersion process involved adding the desired amount of Copper and Zinc nanoparticles to the CMC solution while continuously stirring at a specific speed. The control coating consisted of 100% distilled water.

Coating Processes

Cucumber samples were sorted for uniform size and no sign of any physical or mechanical damage or even fungus infection. Cucumber samples were washed with tap water to remove any loose soil, then with sodium hypochlorite (1%) for 5 min, and fan-dried at room temperature. Then, the fruits were immersed into the treatment solutions for 5 min. After the coating application, coated cucumbers were placed on a metal wire mesh and allowed to be completely dried. The coated and surface-dried cucumbers were placed in plastic bags, and storage for 12 days at 24 °C (Fig. 1).

RESULTS AND DISCUSSION

SEM images of synthesized copper and zinc nanoparticles

SEM images of copper and zinc nanoparticles synthesized with rosemary extract were showed in Fig. 2. As may be seen, the mean particle size of copper and zinc nanoparticles is about 30-70 nm and the nanoparticles have good homogeneity and spherical shape.

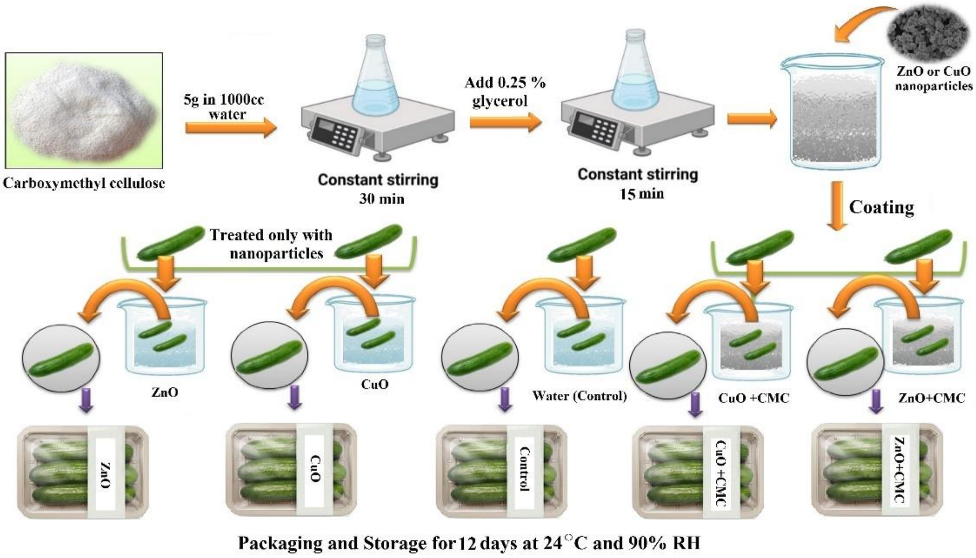

Fig. 1. Schematic presentation of nano-coating synthesis and its use in cucumber samples.

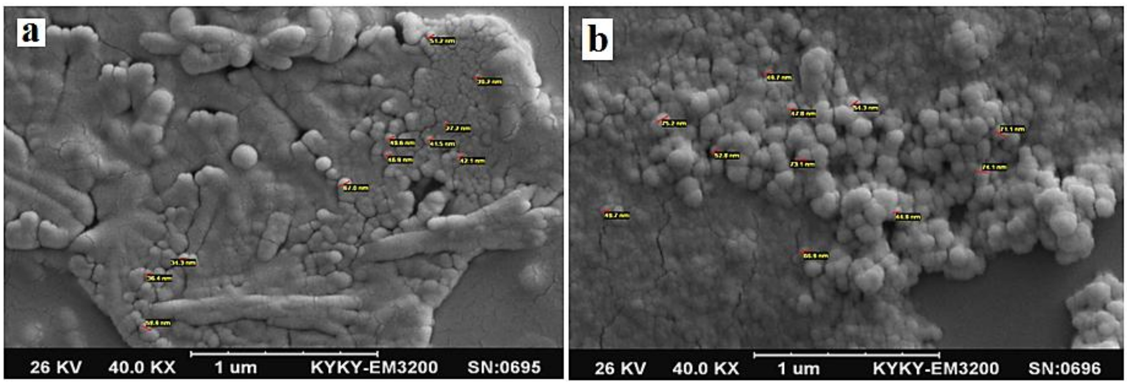

Fig. 2. SEM image of copper nanoparticles (a) and zinc nanoparticles (b).

XRD pattern of synthesized nanoparticles

The crystal structure of the copper and zinc nanoparticles were characterized by X-raydiffraction (XRD) analysis (Fig. 3). The XRD patterns show that all of the diffraction peaks are in good agreement with the standard diffraction data. The diffraction peaks of zinc oxide correctly matched the hexagonal wurtzite structure of zinc oxide [11].

Weight loss

The results of weight loss in Fig. 4 shows that the maximum weight loss was detected for control (13.45%) and minimum value was established for Zn-NPs+CMC (3.88%). In generally, moisture loss during storage leads to the weight loss of the crops. It is plausible that the metal nanoparticles and CMC played a role in preserving moisture, rate of respiration and regulating enzyme functions, therefore mitigating weight loss in the treated cucumber fruits [12].

Firmness

According to Fig. 5, all treatments exhibited a gradual decline in firmness and the control without CMC coating reached a firmness value of 4.3 N. In contrast, the Zn-NPs+CMC treatment had firmness values of 10.7 N. This indicates that the

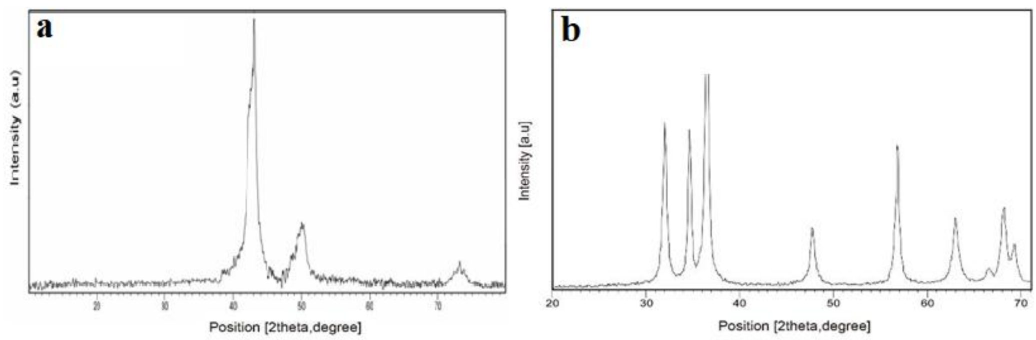

Fig. 3. XRD patterns of copper nanoparticles (a) and zinc nanoparticles (b).

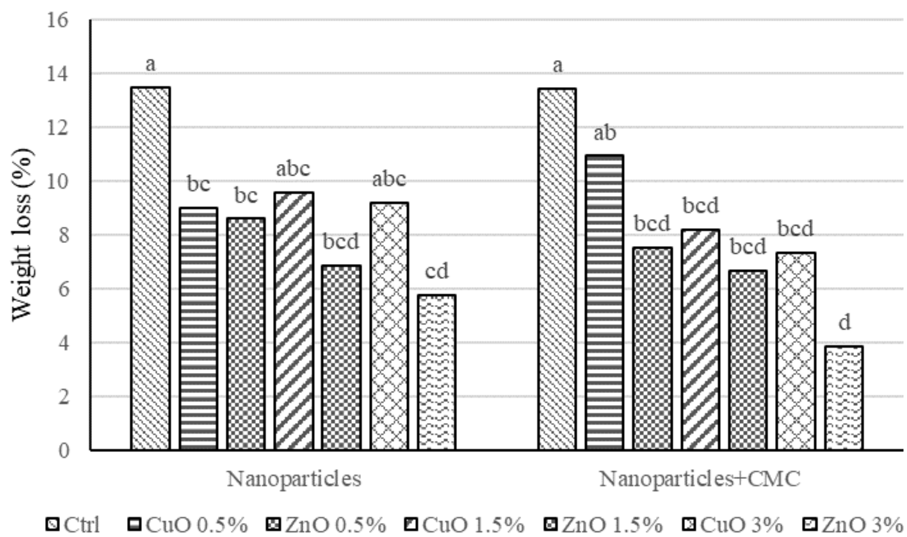

Fig. 4. Effect of nanoparticles and nanoparticles + CMC coated treatments on weight loss in cucumbers stored for 12 days at 24°C.

presence of Zn-NPs, particularly when combined with CMC coating, contributed to a higher level of firmness in the cucumber fruits compared to the control without CMC. The decrease in firmness observed after harvesting can be attributed to various factors, including cell growth, water migration, and the absence of a protective surface cuticle [13]. Pectin depolymerization and cellulase activity are some of the leading causes of firmness loss and softening in fruits and vegetables [14]. CMC coating can create a modified atmosphere around the cucumber fruits, reducing the oxygen

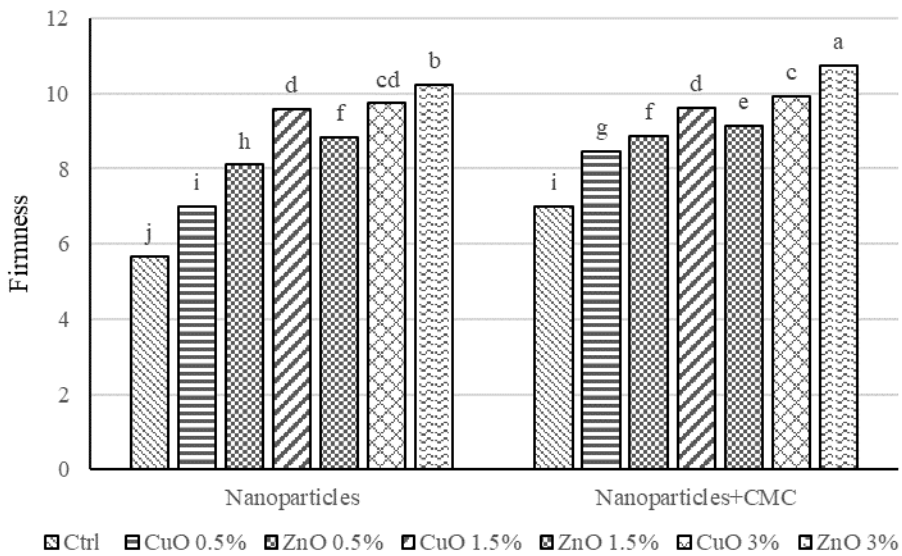

Fig.5. Effect of nanoparticles and nanoparticles + CMC coated treatments on firmness in cucumbers stored for 12 days at 24°C.

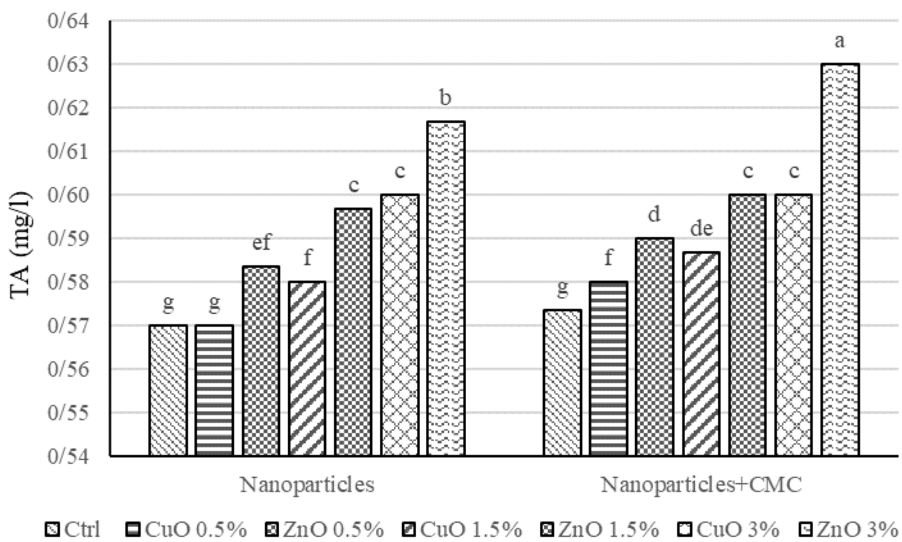

Fig.6. Effect of nanoparticles and nanoparticles + CMC coated treatments on titratable acidity in cucumbers stored for 12 days at 24°C.

levels and increasing the carbon dioxide levels. This altered atmosphere can help slow down respiration and metabolic processes, thereby delaying the softening and degradation of the fruits [15]. Also, nano-coating materials has been reported to have inhibitory effects on cellulase enzymes. By reducing cellulase activity, the degradation of cellulose and subsequent loss of firmness can be slowed down [16,17].

Titratable Acidity (TA)

Titratable acidity (TA) significantly decreased

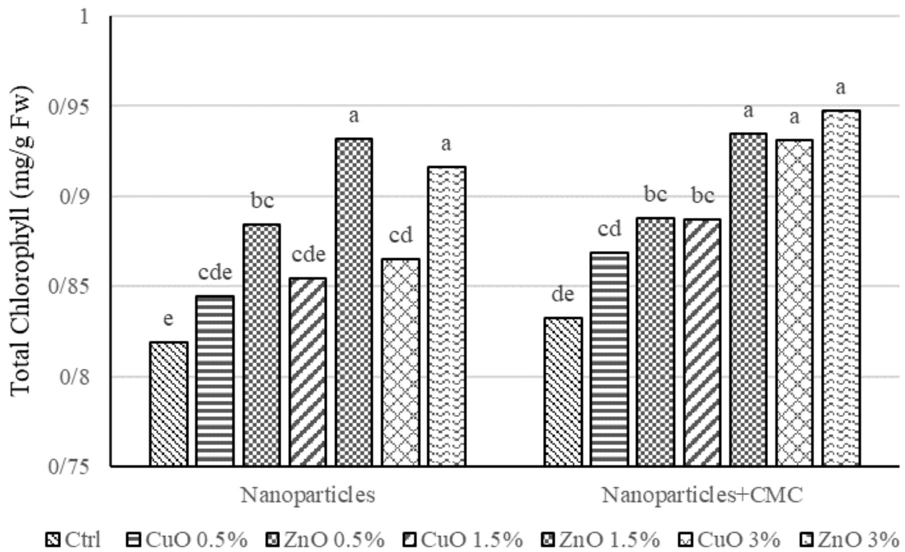

Fig.7. Effect of nanoparticles and nanoparticles + CMC coated treatments on total chlorophyll contentsin cucumbers stored for 12 days at 24°C.

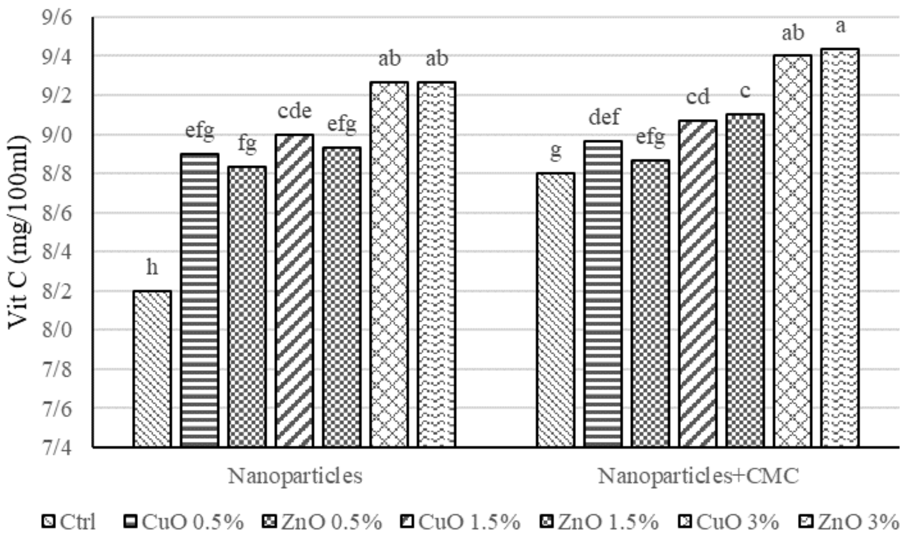

Fig. 8. Effect of nanoparticles and nanoparticles + CMC coated treatments on vitamin C in cucumbers stored for 12 days at 24°C.

during storage, but this decrease was less in nano-coated fruits (Fig. 6). The decrease in fruit acidity during storage is primarily attributed to the reduction in organic acids, such as citric or malic acid, which serve as the primary substrates for the respiration process in fruits. Consequently, a decrease in acidity is expected in highly respiratory fruits [18]. As previously mentioned, the application of the nanoparticles+CMC coating slows down the rate of respiration and

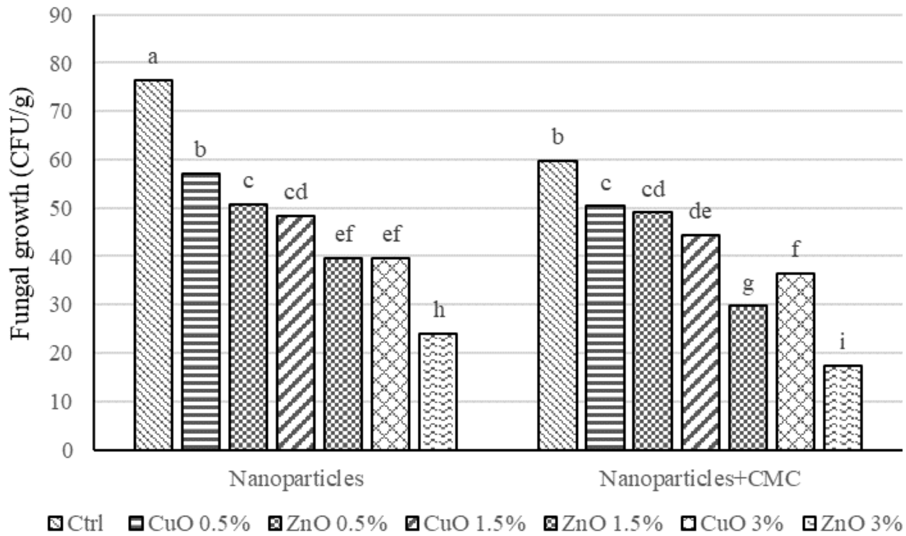

Fig. 9. Effect of nanoparticles and nanoparticles + CMC coated treatments on fungal growth in cucumbers stored for 12 days at 24°C.

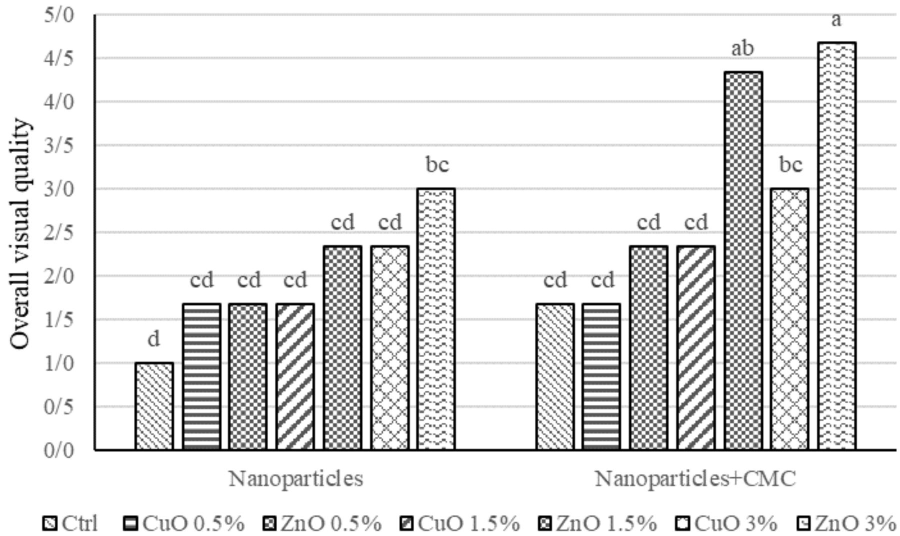

Fig. 10. Effect of nanoparticles and nanoparticles + CMC coated treatments on vitamin C in cucumbers stored for 12 days at 24°C.

metabolic processes that convert organic acids in respiration reactions [19]. Organic acids, which are the substrates for the respiration process, are consumed during the senescence of the produce in the storage. Therefore, with the progression of the storage period, titratable acidity of the fresh produce decreases [5,20].

Total chlorophyll contents

The statistical analysis revealed significant differences in total chlorophyll contents among different concentrations of nanoparticles (Fig. 7). Nanoparticles, such as Cu-NPs and Zn-NPs, have the potential to interact with plant cells and affect various cellular processes. When applied at different concentrations, nanoparticles can induce changes in cellular signaling, gene expression, enzyme activity, and nutrient uptake, among other factors. These changes can directly or indirectly impact chlorophyll synthesis and degradation pathways [10]. Excessive water loss in untreated cucumbers with nano-coating materials, can lead to water stress, which in turn can accelerate ethylene production and result in yellowing of the cucumbers [3]. The modified atmosphere created by the coatings, such as CMC coating, can help delay the factors that trigger chlorophyll degradation by reducing the availability of oxygen and regulating gas exchange. This delay in chlorophyll degradation helps to maintain the green color of the cucumbers for a longer period. The high values of total chlorophyll observed in the nano-coated fruits can be attributed to the reduced oxidative and enzymatic browning of the skin [3].

Vitamin C contents

The vitamin C content of the treated samples was the highest in Zn-NPs+CMC treatment that possessed 9.43 mg/100 ml when compared to Control samples with 8.20 mg/100 ml (Fig. 8). The decrease in vitamin C during the storage may be due to the conversion of ascorbic acid to dehydroascorbic acid because of the action of ascorbic acid oxidase [21]. Nano-coating combination films may protect the ascorbic acid contents by limiting gas exchange and respiration rates with the environment, inhibiting the ascorbic acid exposure to O_2 and concentrating it in the fruit [22].

Fungal growth

Fig. 9 demonstrates that the control treatment, without any nanoparticle or CMC coating, exhibited the highest fungal growth with a count of 76.3 CFU/g. This indicates that the absence of any protective treatment resulted in significant fungal growth. On the other hand, the Zinc

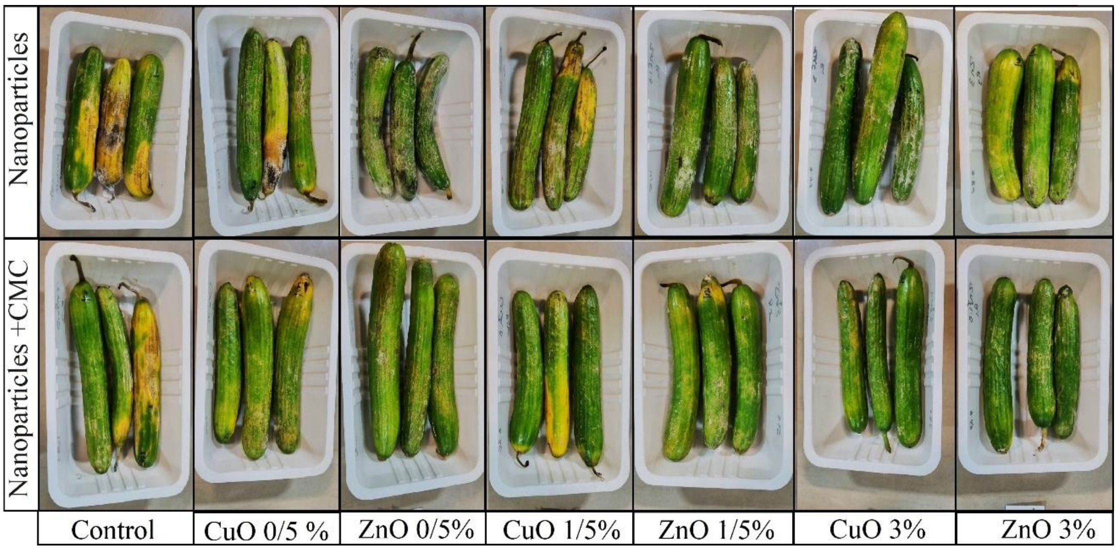

Fig. 11. The effect of different concentrations of copper and zinc nanoparticles with CMC coating and un-coating on the visual quality of cucumber samples.

nanoparticles + CMC treatment showed a more pronounced fungicide effect, with a fungal growth count of 17.4 CFU/g. This suggests that the combined application of Zinc nanoparticles and CMC coating had a substantial inhibitory effect on fungal growth, significantly reducing the microbial load on the cucumbers. Various studies have been shown to Zinc and Copper nanoparticles have the antifungal activity against fungi that infect fruits and increasing concentration of these nanoparticles can be enhanced antifungal effectiveness [9,23]. The CMC coating restricts the entry of oxygen, which is necessary for fungal respiration and growth. As a result, fungal growth is suppressed to some extent [24].

Overall visual quality

The treatments of 1.5% and 3% Zn-NPs combined with CMC coating can indeed have a significant effect on the overall visual quality of cucumbers during storage (Fig. 10). Visual quality score of cucumbers showed a gradual decrease with decreasing concentration of nanoparticles (Fig. 11). Higher concentrations of nanoparticles in coatings may provide stronger antimicrobial and antioxidant effects. As the concentration of nanoparticles decreases, their effectiveness in inhibiting microbial growth, reducing enzymatic reactions, and preventing oxidative processes may diminish, resulting in a decline in visual quality of the cucumbers. Also, CMC coatings create a protective barrier that limits gas exchange, moisture loss, and microbial contamination, thereby preserving the visual quality of the cucumbers [25].

CONCLUSION

The application of copper nanoparticles (Cu-NPs) and zinc nanoparticles (Zn-NPs) in combination with a carboxymethyl cellulose (CMC) coating has been found to effectively reduce the loss of quality in cucumbers compared to the control during storage. By increasing the concentration of Cu-NPs and Zn-NPs, their effectiveness in inhibiting microbial growth, reducing enzymatic reactions and preventing oxidative processes increased, and as a result, the storage quality of cucumbers improved.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interests regarding the publication of this manuscript.

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