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Influence of chitosan edible coating on postharvest qualities of *Capsicum annum* L. during storage in evaporative cooling system

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ABSTRACT

Chitosan is a biopolymer edible coating which can act as physical barrier when coated on fruits and efficiently alter their internal atmosphere and delay the ripening process. This study evaluates the usage of chitosan (CH) as edible coating to extend the shelf life of green bell pepper. Physicochemical and microbial analysis of the green bell pepper was conducted during evaporative coolant structure storage (ECS). The effect of chitosan coating on green bell pepper significantly delayed a loss in firmness, weight loss, and vitamin C content and inhibited the growth of heterotrophic bacteria, mesophilic bacteria, yeast and mould during the five weeks of storage. On the whole, this study established that edible coating from chitosan could form a natural and permanent replacement to the chemically preservatives used for postharvest management of green bell pepper.

Introduction

It has been stipulated that the world population will increase drastically which will surpass 9 billion people by mid-century (FAO, 2017). This implies that there will be a greater request for healthy food, potable water, adequate land and environmental impacts (Ricardo et al., 2018). Moreover, there is a need to critically look into so many challenges mitigating against the achievement of sustainable agriculture and food security like food safety, nutrition deficits, pest and diseases, post-harvest losses, food safety issue (Jenny et al., 2016). Therefore, the field of food science and technology has a great responsibility to play in order to mitigate and achieve a sustainable food production in order to feed the ever-increasing world population (Bartkiene et al., 2018; Eriksson et al., 2018).

The eating of fruit and vegetable-based diets has been documented to minimize the consequence of diet-related chronic diseases which have led to the high rate in the number of death recorded worldwide (Ahmed et al., 2018). Fruits and vegetables contain necessary micronutrients required for healthier diets (Schreinemachers et al., 2018). The World Health Organization has recommended a minimum intake of 400 g per day to avoid chronic diseases and provide necessary micronutrients (WHO, 2015).

Bell peppers are crucial crop that has definite postharvest treatment requirements due to a high vulnerability to quality deterioration from biotic and abiotic stress (Samira et al., 2013). Most of the horticultural crops require low temperature and high relative humidity during storage and transportation. Most of the farmers, rural dwellers, transporters, and consumers cannot afford the cost to maintain controlled atmospheric storage facilities (FAO, 2017).

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Therefore, there is a need to develop a cheap and environmentally friendly technology that will maintain the post-harvest attributes of fruits and vegetables. This will also help to minimize the large quantities of crustacean wastes causing serious environmental problems and their conversion into value-added products (O'Donoghue et al., 2017, Adetunji et al., 2018).

The utilization of ECS could be a sustainable technology that could help farmers that cannot afford the high price of control and modified temperature for the postharvest management of fruits and vegetables. ECS has so many advantages because it requires zero energy cooling system and it lowers the storage temperature as well as increases the relative humidity so as to maintain the freshness of the horticultural crops (Ial Basediya et al., 2013).

The application of chitosan has been documented in various fields which include manufacturing industries (Lang and Clausen, 1989), nutrition (Shahidi et al., 2001), biotechnology, pharmacology and medicine (Liu et al., 2001) and agriculture (Bautista-Baños et al., 2006). It has been observed that whenever chitosan is applied as a coating on fruits and vegetables, it forms a semipermeable film that controls the gas interchange, decreases transpiration losses, reduces fruit ripening, eventually reduction in gaseous exchange and water loss thereby improving the shelf lives of fruits and vegetables (Bautista-Baños et al., 2006). Moreover, chitosan has been applied as a foliar spray in order to improve the growth, yield, and value of vegetable crops (Fawzy et al., 2012).

The present study was intended to evaluate the effect of 1.5 % chitosan and to improve the postharvest management of green bell pepper during storage at room temperature.

Materials and methods

Preparation of chitosan

Matured brownish crabs were collected from the brackish waters of Warri in the Delta State of Nigeria. The animals were sacrificed and the viscera and muscles were carefully removed. The shells were dehydrated at 60 °C overnight and milled with a grinder that was fabricated locally at Postharvest Engineering Research Laboratory, NSPRI in Ilorin. The methods established by Pandharipande and Prakash (2016) were used to obtain chitin from crab shell waste and the method described by Adetunji et al. (2014) was used for further treatment of chitin to produce the chitosan used for this study. Subsequent washing and drying steps yielded chitosan.

Source of fruits

A local cultivar green bell namely 'tatase' which is fully matured, green coloured and thick, wax covered green bell pepper was harvested from a commercial farm located in the Ogbomosho town (8° 8' 0" North, 4° 16' 0" East), Oyo State of Nigeria and immediately transported to the postharvest laboratory of Nigerian Stored Product Research Institute, Ilorin (8° 30' 0" North, 4° 33' 0" East), Kwara State, Nigeria. Some of the factors that were considered are uniformity in size, colour, shape, greenness and absence of injuries before selecting green bell pepper for the experiment, while blemished and diseased fruits were discarded. The green bell pepper was later washed under running tap water, immersed in a chlorine solution (0.0003 %) for 30 s and then washed with sterilized water.

Treatments

T₀ (control):- T₀ was selected as the control (untreated green bell pepper); T₁ green bell pepper was coated with 1.5% chitosan. The treated and untreated samples were arranged in a blue coloured plastic basket with green bell peppers. The basket was stored in ECS at a temperature of 13 ± 2 °C and relative humidity of 85–90 %. The physiochemical analysis was carried out in the period of five weeks after the coating from week 1-5 of after coating.

Features of the sprayer

The spraying bottle is made of heavy duty high-density polyurethane (HDPE) with a cushion grip and a pebble textured neck. It has a capacity of 32 ounces (1 quart). It is made of an opaque bottle with 3 3/8" diameter x 10" high with adjustable trigger nozzle necessary for adequate spraying of the edible coating solution. It contains a 10" pickup tube with decontamination filter to trap debris before enters nozzle.

The Evaporative Cooling System

The ECS used for this study was constructed with a clay brick having a dimension of 25.5 cm × 12.0 cm × 6 cm. It contains a double-walled rectangular brick construction with the inter-space filled with riverbed sand saturated with water. The walls were constructed on a short platform of concrete to avert water leakage into the soil. The ECS has a capacity chamber of 1.38 m³. The two doors were considered adequate for thermal insulation against heat flux. This structure was placed in an open space exposed to fresh air. A shed was built with thatch to prevent the ECS from the radiation effect of the sunlight.

Weight loss: To determine weight loss, separate samples in 3 replicates from the coated green bell pepper using the following formula: $\text{Weight loss (\%)} = [(A-B)/A] \times 100$. Where A denotes green bell pepper weight at the time of harvest and B denotes the weight of coated green bell pepper after storage intervals (A.O.A.C., 1994).

Firmness:- The firmness of the green bell pepper was determined using a penetrometer, which has a probe 8 mm in diameter that was pressed gradually onto the surface of each green bell pepper.

Vitamin C content:- The amount of vitamin C content from green bell pepper was determined using 2,5-6 dichlorophenol indophenols' method defined by AOAC (1994).

Microbial analysis

The total microbial count was carried out by grinding 1 g of bell pepper, which was then mixed thoroughly with 10 mL water and a 1:10 dilution was further serially diluted to 10^{-7} . Total aerobic mesophilic bacteria and heterotrophic bacteria present in the samples were determined by plating 0.1 mL of the dilutions on count agar plates using a sterilized L shaped glass spreader. Plates were incubated at 37 °C for 48 h. Approximately 0.1 mL of each serial dilution of the bell pepper homogenate was plated on potato dextrose agar containing 100 µg/g streptomycin and incubated at 28 °C for 5 d to evaluate yeast and mold counts. Colony forming units (CFU) per gram of bell pepper were evaluated in triplicate and designated as log CFU/g.

Statistical analysis

All data were obtained in triplicate and analyzed by analysis of variance (ANOVA) using SPSS v 16.0 and significant difference ($P = 0.05$) among means of significant were separated using Duncan's multiple range test.

Result and Discussion

Weight Loss

The weight loss of green bell pepper fruits during the ECS storage was significantly lower ($P < 0.05$) in coated fruits containing chitosan when compared to the uncoated control group. The result obtained in Figure 1. showed that CH treated fruits had a weight loss of $22.83 \pm 3.87\%$ while the uncoated control group had a weight loss of $30.67 \pm 1.43\%$. There was a decrease in weight loss of the coated green bell pepper fruits when compared with uncoated control group. The result obtained is in line with the observation of different researchers who observed a drastic reduction in weight loss of treated fruits when compared to the control fruits i.e pears (Zhou et al., 2008); papaya cubes (González-Aguilar et al., 2010), bell peppers and cucumbers (El-Ghaouth et al., 1991), grapefruit (Meng et al., 2006). Weight loss is an important factor that determines the postharvest shelf life of fruit during storage (Bai et al., 2002).

Firmness

The utilization of coating from chitosan significantly prevents ($P < 0.05$) the loss of firmness of the stored green bell pepper (Figure 2). The results showed that edible coatings from chitosan had a firmness of 2224.17 ± 91.09 N while the uncoated had a firmness of 1536.67 ± 310.27 N. As chitosan coatings applied a useful effect on fruit firmness, after storage, all the treatments showed higher flesh firmness values than control ($P < 0.05$). It has been established by several researchers that chitosan played a positive impact in extending firmness of fruits including tomato (El Ghaouth et al., 1992), kiwifruit (Du et al., 1997), mango (Zhu et al., 2008), papayas (Ali et al., 2011), and 'Murcott' tangor (Chien et al., 2007).

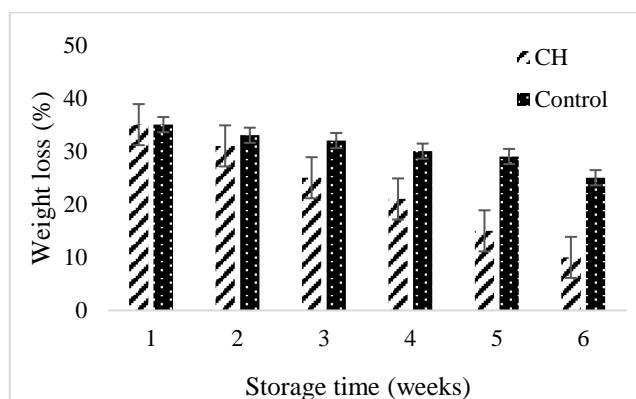


Fig. 1. Effect of chitosan coatings on the weight loss of green bell pepper

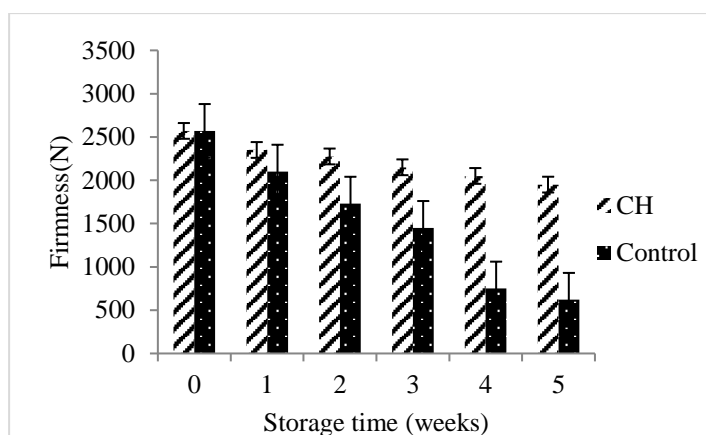


Fig. 2. Effect of chitosan edible coatings on the firmness of green bell pepper

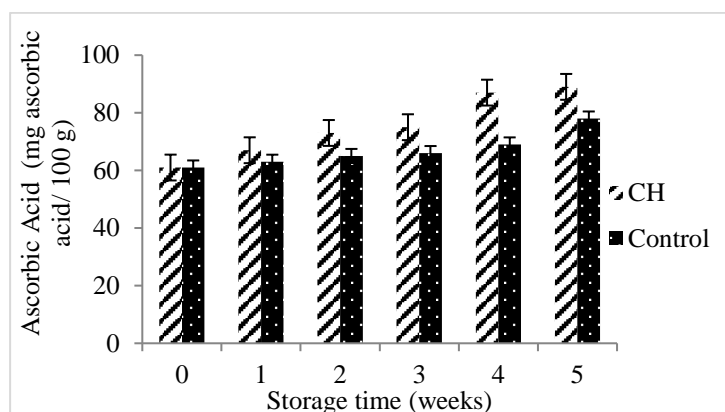


Fig. 3. Effect of chitosan edible coatings on the vitamin C content of green bell pepper

Texture is a critical quality used by the consumers before they can buy fresh fruit and vegetables. The process of ripening has a greater effect on fruit texture (Saladié et al., 2007). The two major enzymes that trigger the process of fruit ripening are Pectinmethylesterase and polygalacturonase. They induce fruit softness that normally occurs during ripening (Kays, 1991).

Ascorbic Acid content

Application of edible fruit coatings containing 1.5 % of chitosan, is significantly ($p < 0.05$) higher in ascorbic acid content when compared to the uncoated control groups (Figure 3). The utilization of edible coatings of chitosan can cause a decrease in oxygen diffusion which could inhibit the rate of ripening in fruits. This normally preserves the vitamin C contents and delays senescence in fruits (Yage et al., 2011). Some previous researchers have

demonstrated that edible coatings could increase carbon dioxide levels and reduce the level of oxygen surrounding the fruit which normally helps in the prevention of vitamin oxidation (Atres et al., 2010).

Yeast and mould

The result obtained from the treated fruits with CH edible coatings cause a reduction in the number of yeast and mould count from 9.3 log CFU/g during the first week of storage to 1.3 log CFU/g after 5 weeks of storage count when compared to the uncoated control group that had 22.3 log CFU/g. (Figure 4). Howard and Dewi (1995) discovered that edible coating made of cellulose material maintained the microbial quality of mini-peeled carrots during storage. Also, Olivas and Barbosa-Cánovas (2005) observed that edible coatings prevent the growth of fruit and vegetable spoilage microorganisms during storage.

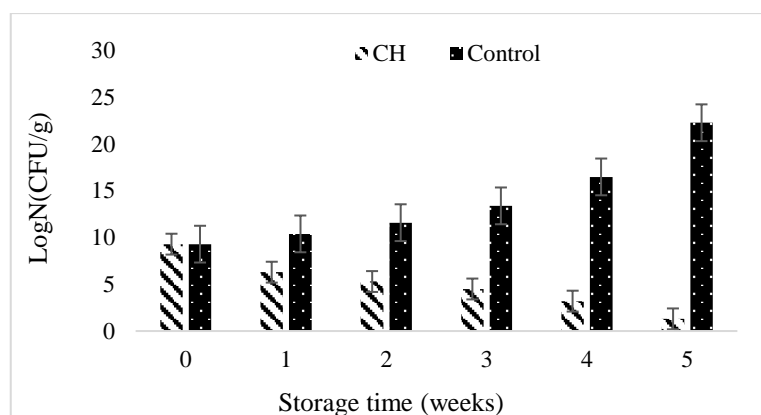


Fig. 4. Effect of chitosan edible coatings on the yeast and mould count (Log N (CFU/g)) of green bell pepper

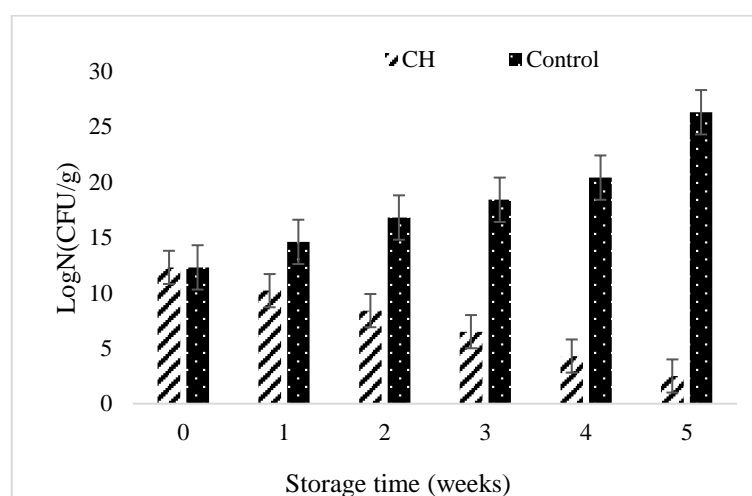


Fig. 5. Effect of the edible coatings from chitosan on the mesophilic count (Log N (CFU/g)) of green bell pepper

Mesophilic bacteria

The coating treatment containing chitosan significantly reduced ($p < 0.05$) mesophilic bacteria population in the green bell pepper during storage. The result obtained shows that the treated fruits showed a reduction in the number of mesophilic bacteria from 12.3 log CFU/g during the first week of storage to 2.5 log CFU/g after 5 weeks of storage count when compared to the uncoated control group that had 26.3 log CFU/g. (Figure 5). Previous researchers have discovered that edible coatings containing antimicrobial agent tend to preserve the shelf life of fruits and vegetables when compared to fruit coated with an antimicrobial agent (Sangsuwan et al., 2008). The high reduction in the mesophilic bacteria count observed during this study might be due to the inhibitory effect from chitosan (Geraldine et al. 2008). The microbial count obtained from the chitosan applied as an edible coating during this study fall

below the limits set by Spanish regulations (BOE, 2001), which stated that the maximum count of aerobic mesophilic microorganisms in food samples should not exceed a level of 7 log CFU/g.

Heterotrophic bacteria

The result shows that the population of heterotrophic bacteria reduced from 18.4 log CFU/g during the first week of storage to 3.6 log CFU/g after 5 weeks of storage while the control had 34.5 log CFU/g (Figure 6). Several researchers have discovered that the major microflora which commonly shelf life of fruits and vegetables are psychotropic bacteria (Hotchkiss and Banco, 1992). Moreover, the application of edible coatings containing antimicrobes could minimize their number thereby, leading to the postharvest extension of fruits and vegetables (Garcia-Gimeno and Zurera-Cosano, 1997).

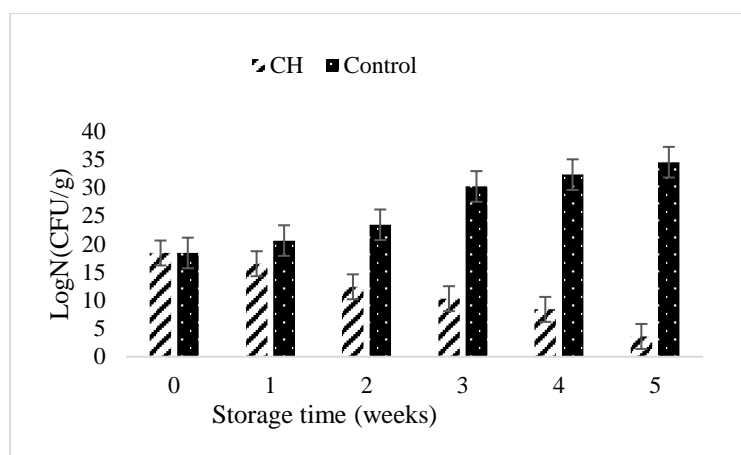


Fig. 6. Effect of chitosan edible coating on the heterotrophic count (Log N (CFU/g)) of green bell pepper

Conclusion

On the whole, our study showed that the method of chitosan application as a protective coating which is simultaneously edible is an innovatory and eco-friendly method of storage and increasing of the shelf-life of pepper. In the light of the obtained experiment results, the application of chitosan as an edible coating demonstrates that this preservative method could be applied to other fruits and vegetables. Moreover, the chitosan used during this study exhibits a high antimicrobial efficacy against some spoilage microorganism, but there is a need to carry out the mode of action through which the edible coating exhibits its antimicrobial activity at a molecular level.

Recommendation

This study has established that the edible coating containing chitosan could serve as an alternative to the chemical-based coatings used for postharvest loss prevention. Further investigation is needed to understand the mechanisms and mode of action of chitosan in preventing/ or inhibiting spoilage microorganisms at molecular levels. Therefore, this study could lead to the development of indigenous wax emulsions/ coatings for prolonging the shelflife of fruits, thereby ensuring food security in alignment with the objectives of Sustainable Development Goals (SDGs).

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