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MICROBIOLOGICAL EVALUATION OF AN EDIBLE ANTIMICROBIAL COATINGS ON MANGOES FRUIT STORED UNDER EV APORATIVE COOLANT SYSTEM (ECS)

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ABSTRACT

Two different coatings were developed from the mucilage of Cactus and their effects were investigated on the microbial qualities of mango fruits. The two experimental coatings were: Pure mucilage extracts (ME) and Mucilage extract mixed with 5ml glycerol (MEG) which served as plasticizer. Samples of the mangoes were immersed into these coatings and they were stored for seven weeks at an average temperature of 27±2°C and relative humidity55-60% under Evaporative Coolant System(ECS) . The *Cactus* mucilage treatment of mangoes significantly attenuated the microbial growth on fruits compared to control. Concentration of yeast and mold was found to be decreased from 10.23log CFU g^{-1} to 0.34 log CFU g^{-1} (MEG) and 2.11log CFU g^{-1} (ME),also the concentration of aerobic Psychrotrophic bacteria was decreased from 23.56log CFU g^{-1} in control to 1.23 log CFU g^{-1} (MEG) and 5.30 log CFU g^{-1} (ME) mucilage treated fruits while the concentration of aerobic mesophilic bacteria was decreased from 20.84log CFU g^{-1} in control to 1.07 log CFU g^{-1} (MEG) and 3.12 log CFU g^{-1} (ME) mucilage treated fruits. The overall result showed that Cactus mucilage coating hindered the growth of microorganisms significantly ($P<0.05$) and extended the shelf-life of mango fruits when compared to untreated in the order, MEG>ME>Control.

Keywords: Antimicrobial coatings, Evaporative Coolant System, *Cactus* mucilage, microbial qualities.

INTRODUCTION

Fruits and vegetables are an extraordinary dietary source of nutrients, micronutrients, vitamins and fiber for humans and are thus vital for health and well being. Well balanced diets, rich in fruits and vegetables, are especially valuable for their ability to prevent vitamin C and vitamin A deficiencies and are also reported to reduce the risk of several diseases (Kalia and Gupta, 2006).

Fruits and vegetables are widely exposed to microbial contamination through contact with soil, dust and water and by handling at harvest or during postharvest processing. They therefore harbor a diverse range of microorganisms including plant and human pathogens (Nguyen-the and Carlin, 1994; Dunn et al., 1995; Carmo et al., 2004). Differences in microbial profiles of various fruits and vegetables result largely from unrelated factors such as resident microflora in the soil, application of nonresident microflora via animal manures, sewage or irrigation water, transportation and handling by individual retailers (Ray and Bhunia, 2007; Ofor et al., 2009). Thus despite their nutritional and health benefits, outbreaks of human infections associated with the consumption of fresh or minimally processed fruits and vegetables have increased in recent years (Hedberg, 1994; Altekruse and Swerdlow, 1996; Beuchat, 1996, Beuchat, 2002). Enteric pathogens such as *Escherichia coli* and *Salmonella* are among the greatest concerns during food-related outbreaks (Buck et al., 2003).

Even though pharmacological industries have produced a number of new antibiotics in the last three decades, resistance to these drugs by microorganisms has increased. In general, bacteria have the genetic ability to transmit and acquire resistance to drugs, which are utilized as therapeutic agents (Gislene et al., 2000). For a long period of time, plants have been a valuable source of natural products for maintaining human health. The use of plant extracts and phytochemicals, both with known antimicrobial properties, can be of great significance in therapeutic treatments (Seenivasan et al., 2006). Many plants have been used because of their antimicrobial traits, which are due to compounds synthesized in the

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secondary metabolism of the plant. These products are known by their active substances e.g. the phenolic compounds which are part of the essential oils, as well as tannin (Tyagi and Malik, 2010).

Numerous studies have been published on the antimicrobial activities of plant extracts and many of these studies have included plants used in ethno medicine. Some of the compounds responsible for the antimicrobial activity observed from the plants have been isolated by activity-guided fractionation and characterized using various spectroscopic methods. The antimicrobial agents from plants can, therefore, be classified into groups such as essential oils and terpenoids, tannins, phenols and alkaloids to name a few (Cowan, 1999).

The greatest losses in food are due to microbiological alterations. Many chemical and physical processes have been developed to preserve food quality. Among such processes, adequate packaging is a fundamental factor in the conservation and marketing phases. Thus, packaging plays a prominent role in maintaining food quality (Debeaufort et al ., 1998). Antimicrobial films and coatings have innovated the concept of active packaging and have been developed to reduce, inhibit or delay the growth of microorganisms on the surface of foods in contact with the packaged product (Appendini & Hotchkiss, 2002). In most fresh or processed foods, microbial contamination occurs at a higher intensity on the food surface, thus requiring an effective microbial growth control (Padgett et al., 1998). Traditionally, antimicrobial agents are added directly to the foods, but their activity may be inhibited by many substances in the food itself, diminishing their efficiency. In such cases, the use of antimicrobial films or coatings can be more efficient than adding antimicrobial agents directly to the food since these may selectively and gradually migrate from the package onto the surface of the food, thereby high concentrations being maintained when most necessary(Ouattara et al., 2000).

Edible films and coatings can be used to help in the preservation of fruit and vegetables because they provide a partial barrier to moisture, O_2 and CO_2, also improving mechanical handling properties, carrying additives, avoiding volatiles loss and even contributing to the production of aroma volatiles (Olivas & Barbosa-Ca´ novas, 2005).

Edible coatings may be composed of polysaccharides, proteins, lipids or a blend of these compounds (Mahmoud and Savello, 1992; Park et al., 1994a, b; Guilbert et al., 1996; Li and Barth, 1998; Arvanitoyannis and Gorris, 1999). Their presence and abundance determine the barrier properties of material with regard to water vapor, oxygen, carbon dioxide and lipid transfer in food systems (*Guilbert et al.*, 1996). However, none of the three constituents can provide the needed protection by themselves and so are usually used in a combination for best results (McHugh and Krochta, 1994a, b; Guilbert et al., 1996).

Cactus mucilage is an example of edible coating containing polysaccharides that is commonly used in food, cosmetics, pharmaceutical and other industries. The complex polysaccharide is part of dietary fibre and has the capacity to absorb large amounts of water, dissolving and dispersing itself and forming viscous or gelatinous colloids (Dominguez-Lo´pez, 1995).

Mango (*Mangifera indica* L) is a very delicious tropical fruit which belongs to family *Anacardiaceae*. It is an abundant source of vitamins, minerals and is famous for its excellent flavor, attractive fragrance and nutritional value. It is as an emerging tropical export crop and is produced in about 90 countries in the world with a production of over 820,877MT. The magnitude of post harvest losses in fresh fruits and vegetables is an estimated 5-25% in developed countries and 20-40% in developing countries, depending upon the commodity (FAO, 2001).

The present study is aimed to evaluate the effect of prickly pear cactus (*O. ficus indica*) mucilage as an edible coating on the microbiological properties of mango fruits during storage in ECS.

MATERIALS AND METHODS

Source of fruits and Coating materials

Ogbomoso mangoes, common consumer varieties, were purchased from a local market on the day after harvest and were immediately placed in ambient storage ($27^oC \pm 2$).Uniform sized, defect-free fruits were selected. Cactus stems were obtained from a local farmer and were stored at $23^oC \pm 1$ prior to formation of the coating solution. Glycerol (99.5%) was purchased from Sigma Chemical Co.

Preparation of Coating solution

Cactus stems were peeled and cubed (1 cm^3). Samples were homogenized (20% w/v) in distilled water. The slurry was centrifuged for 10 min at 4500 × g and the supernatant obtained was used to prepare the edible coating (Sa´enz, Va´squez, Trumper, & Fluxa´, 1992).
It was then pasteurized to form a pure mucilage extract(ME) .Mangoes were dipped in coating solution for 30secs, the excess coating was drained and the coated mangoes were dried in a forced-air dryer (20 ^oC) for 30 min. Mangoes dipped in distilled water were used as a blank.

Treatments

T_o (control):-Mangoes that wasn't coated with cactus mucilage ; $T_{1:-}$ Mangoes coated with Pure mucilage extract (ME); T2:-Mangoes coated with mucilage extract(ME) as described above was mixed with 5ml glycerol (MEG) serving as plasticizer for the antimicrobial compounds present on the pure mucilage extract to be directly attached to mango fruits.
The treated and untreated fruits were packed in small plastic baskets and each basket contained 20 mango fruits. The baskets were stored at ECS temperature and relative humidity (27±2°C and 55-60%).

The evaporative cooling system

The evaporative cooler used for the study consisted of a double-walled rectangular brick construction with the interspaces filled with riverbed sand saturated with water. The clay brick used in the wall construction was factory baked (at 600 ^oC) and was of dimensions 25.5 cm x 12.0 cm x 6 cm thick. The sand was previous to water. The water used was clean and free of foreign matter so as to maintain previousness and avoid clogging of the sand.
The interior surfaces of the cooling chamber walls were given a smooth finish with 1.2 cm thick cement plaster, while a heat insulating cover of 1.9 cm thick particleboard closed the top. The walls were built on a short plinth of concrete to prevent water seepage into the soil. Two framed doors of sawn wood were fixed one to each of the adjacent walls on a side to provide access to the 1.38 m^3 capacity chamber. These two doors were considered adequate for the needed thermal insulation against heat flux but an additional polystyrene foam board could be installed for more insulation. The permanent structure was erected in open space exposed to free air but shaded from direct solar radiation with an open sided shed of thatch

Microbial analysis

Thirty grams of mango fruit pulps were removed aseptically from each treatment. The sample was then homogenized in peptone saline solution (8.5 g l^{-1} NaCl+1 g l^{-1} peptone (Oxoid, L34)) for 1 min in a stomacher (S400, Shanghai Scientific Instrument Co., Ltd., Shanghai, China). After making serial dilutions in peptone water, the samples were plated on different media as follows: (1) plate count agar (PCA, OxoidCM325) for isolating total aerobic psychrotrophic micro- organisms was incubated at 30^oC for 72 h; (2) Sabouraud media (Oxoid CM41) for isolating yeasts and molds was incubated at 25 ^oC for 120 h. (3) Mesophilic aerobic counts were enumerated using Plate Count Agar (Oxoid) and incubation at 30^oC for 3 days. Colonies were counted and the results expressed as $CFUg^{-1}$ of mangoes. Analyses were carried out periodically in randomly sampled pairs of trays within 7 weeks. Two replicate counts were performed for each tray.

Statistical analysis

Statistical analysis of the data was carried out employing analysis of variance (ANOVA) package included in StatSoft Statistica 8.0 (Hill and Lewicki, 2007).

RESULTS AND DISCUSSION

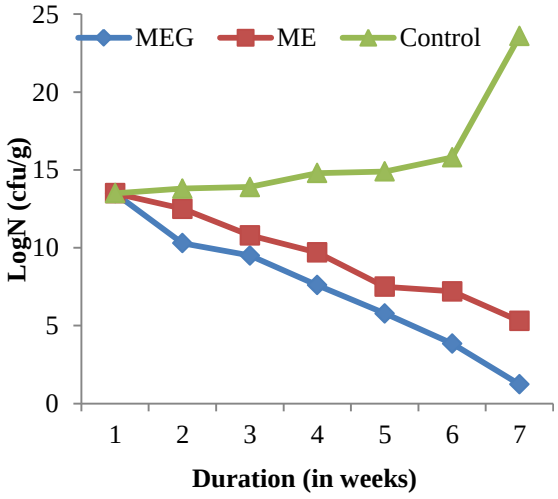

Fig 1: Effect of mucilage coatings on Total aerobic psychrotrophic count of mango fruit during storage in ECS

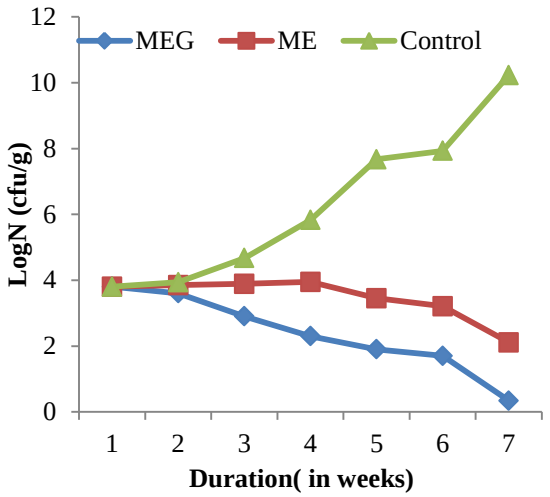

Fig 2: Effect of mucilage coatings on Moulds and yeast count of mango fruit during storage in ECS

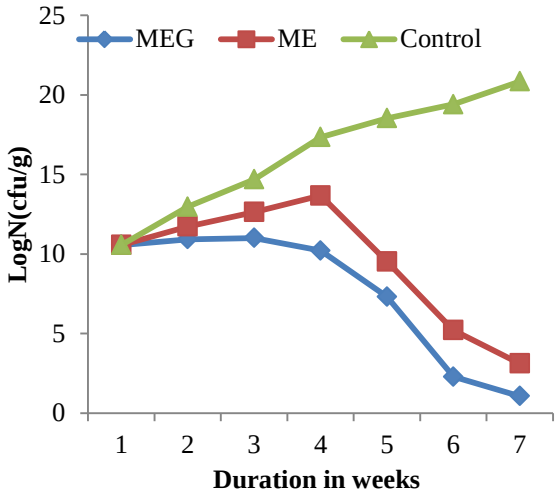

Fig 3: Effect of mucilage coatings on Total aerobic psychrotrophic count of mango fruit during storage in ECS

The predominant microflora which influences the shelf life of fruits and vegetables are psychrotrophic bacteria (Garcia- Gimeno & Zurera-Cosano, 1997; Hotchkiss & Banco, 1992). Changes in the total aerobic Psychrotrophic count, total number of yeasts and moulds in mangoes stored for seven weeks at an average temperature of 27±2°C and relative humidity55-60% are shown in Figs. 1 and 2. During the period of storage coating significantly hindered the increase in total aerobic psychrotrophic count compared with the control samples (Fig.1). Similar effect of coating was observed in reducing the growth of yeasts and moulds during the storage (Fig. 2). At the end of 7 weeks of storage, virtually apparent differences were observed between coated MEG and ME and the control samples ($p<0.05$) at both temperatures.

At harvest, mangoes had 13.5 and 3.8 log CFU g^{-1} for total aerobic psychrotrophic and yeast and mold counts, respectively. Following 7 weeks of ECS storage at temperature of 27±2°C and relative humidity55-60%, the microbial populations of *Cactus* mucilage-treated mangoes were significantly reduced, the reduction being slightly more effective for yeast and mold counts from coated mangoes MEG and ME which were (0.34 log CFU g-1) and (2.11log CFU g^{-1}) respectively than for aerobic Psychrotrophic counts from coated mangoes MEG and ME which were (1.23 log CFU g^{-1}) and (5.30 log CFU g^{-1}) respectively. Moreover, the yeast and mold from uncoated mangoes fruit significantly increases to (10.23log CFU g^{-1}) while the aerobic Psychrotrophic in the uncoated mangoes fruit significantly increases to (23.56log CFU g^{-1}) , but which were significantly reduced in *Cactus* mucilage-coated mangoes.

Fig 3. shows that the Cactus mucilage reduces the microbial load of mesophilic population on the coated mangoes MEG and ME which were (1.07 log CFU g^{-1}) and (3.12 log CFU g^{-1}) compared to the uncoated mango which had (20.84 log CFU g^{-1}) at the end of storage.

The microorganisms present in fruits and vegetables are a direct reflection of the sanitary quality of the cultivation water, harvesting, transportation, storage, and processing of the produce (Beuchat, 1996; Ray and Bhunia, 2007).

The number of mesophilic aerobic microflora present was very similar to that of psychrotrophic microflora and therefore most of the microorganisms were able to grow at storage temperatures. Garg et al. (1990) pointed out that mesophilic and psychrotrophic counts may be of similar magnitude at the time of processing. Storage at refrigeration temperatures generally selects in favour of the growth of psychrotrophic microorganisms, including the pectinolitic pseudomonads (Nguyen-the and Carlin, 1994). Nevertheless, mesophilic microorganisms may continue to grow at low temperature, albeit at reduced growth rates (Vescovo et al., 1996; Carlin et al., 1990).

Observed numbers of yeasts and moulds were lower than bacteria, during storage in this study. Tournas (2005) obtained similar results with samples of fresh and minimally-processed

vegetables, and sprouts. The role of yeasts in the spoilage of fresh vegetables is not well studied, although they have been implicated in the spoilage of fermented vegetable products and the development of soft rot (Fleet, 1992).

However, some authors (Tournas, 2005; Tournas and Katsoudas, 2005) have pointed out the possible health problems associated with the presence of moulds in fruit and vegetables, as some may produce mycotoxins and others are known to cause allergies when they are able to produce large numbers of conidia.

The Cactus mucilage treatment of mangoes significantly attenuated the microbial growth on fruits compared to control. Concentration of yeast and mold was found to be decreased from 10.23log CFU g-1 to 0.34 log CFU g^{-1} (MEG) and 2.11log CFU g^{-1} (ME),also the concentration of aerobic Psychrotrophic bacteria was decreased from 23.56log CFU g^{-1} in control to 1.23 log CFU g^{-1} (MEG) and 5.30 log CFU g^{-1} (ME) mucilage treated fruits while the concentration of aerobic mesophilic bacteria was decreased from 20.84log CFU g^{-1} in control to 1.07 log CFU g^{-1} (MEG) and 3.12 log CFU g^{-1} (ME) mucilage treated fruits. Rojas-Gra¨u et al. (2007) found that lemongrass, oregano, and vanillin essential oils in alginate coatings reduced the growth of psychrophilic aerobes, yeast, and molds on apple cuts by more than 2 log cfu/g. Natamycin in a bilayer coating of chitosan significantly decreased fresh melon decay caused by two strains of spoilage fungi (Cong et al. 2007). Clove, cinnamon, and oregano essential oils totally inhibited the growth of *Candida albicans*, *Aspergillus flavus*, and *Eurotium repens* in vitro when they were used in paraffin coating of paper packaging materials and completely protected strawberry from visible fungal growth during storage for 7 days at 4^{o}C (Rodriguez et al. 2007). Chitosan is a natural antimicrobial compound and is capable of forming stable coatings on fresh-cut papaya that suppress microbial growth (Gonz´alez-Aguilar et al. 2009).

Addition of glycerol to the cactus mucilage extract greatly improves the increase the shelf life of mangoes during storage compared to the mangoes without plastizicer.

Edible films and coatings need to have good elasticity and flexibility, a low brittleness, a high toughness and to prevent cracking during handling and storage (Barreto et al. 2003). Therefore, plasticizers of low molecular weight (non volatile) are typically added to hydrocolloid film forming solutions to modify the flexibility of edible films. Plasticizers with characteristics such as small size, high polarity, more polar groups per molecule, and greater distance between polar groups within a molecule generally impart greater plasticizing effects on a polymeric system. Indeed, they act by increasing the free volume or in other word by decreasing intermolecular attractions between adjacent polymeric chains by reducing hydrogen bonding between polymers chains (Myllarinen et al. 2002).

Several studies on plasticization of chitosan films revealed that poly (ethylene glycol) (PEG) could improve the elastic properties of chitosan. Caner et al. (1998) observed that chitosan plasticization using PEG was stable until 9 weeks of storage. On the contrary, Butler *et al*. (1996) found the water barrier and mechanical properties of plasticized chitosan films with glycerol changed during storage. Other authors used plasticizers in chitosan blends. Hosokawa et al. (1990) used glycerol to plasticize chitosan/cellulose composites, whereas Arvanitoyannis et al. (1998) used sorbitol and sucrose to plasticize chitosan/poly (vinyl alcohol) blends. They stated that the elongation of blended films increased with increasing plasticizer contents, but at high plasticizer contents there were decreases in both tensile strength and modulus.

CONCLUSIONS

The results of this experiment showed that the use of an antimicrobial coating consisting Pure mucilage extract (ME) and mucilage extract mixed with 5ml glycerol (MEG) , is a viable alternative in controlling the microbiota present in minimally processed mango, since the growth of psychrotroph sand yeasts and molds, was substantially inhibited by the application of MEG and ME .

Based on the concept of protection barrier technology, the use of such coating may contribute to improve safety in mango fruit thereby prolonging its shelf life. Coating may be applied on fruits and vegetables, combined to other types of controls, such as quality raw material, hygienic processing conditions and storage temperatures. The combination of these treatments as barrier offers a greater potential for shelf-life extension of fruits and vegetables.

REFERENCES

Appending P and Hotchkiss JH, 2002. "Review of antimicrobial food packaging". Innovative Food Science and Emerging Technologies. 3:113–126.

Arvanitoyannis I and Gorris LGM, 1999. "Edible and biodegradable polymeric materials for food packaging or coating in processing foods: Quality optimization and process assessment". CRC Press, Boca Raton, FL., 357-371.

Arvanitoyannis I, Nakayama A and Aiba S, 1998. Edible films made from hydroxypropyl starch and gelatin and plasticized by polyols and water. Carbohydrate Polymers. 36:105-119.

Altekruse SF and Swerdlow DL, 1996. The changing epidemiology of food borne diseases. American Journal of Medical Science. 311:23-29.

Barreto PLM, Pires ATN and Soldi V, 2003. Polymer Degradation and Stability. 79:147–152.

Beuchat LR, 1996. Pathogenic microorganisms associated with fresh produce. Journal of food Protection. 59:204-216.

Butler BL, Vergaro PJ, Testin RF, Bunn JM, and Wiles JL, 1996. Mechanical and barrier properties of edible chitosan films as affected by composition and storage. Journal of Food Science. 61: 953-961.

Beuchat LR, 2002. Ecological factors influencing survival and growth of human pathogens on raw fruits and vegetables. Microbes Infection. 4:413-423.

Buck JW, Walcott RR and Beuchat LR, 2003. Recent trends in microbiological safety of fruits and vegetables. Plant Health Progress. 10:1094.

Cowan MM, 1999. Plant products as antimicrobial agents. Clinical Microbiology Review. 12:564-582.

Carmo LS, Cummings C, Linardi VR, Dias RS, Souza JM, Sena MJ, Santos DA, Shupp JW, Pereira RK and Jett M, 2004. A case study of a massive *Staphylococcal* food poisoning incident. Foodborne Pathogen Diseases. 1:241-246.

Caner C, Vergano PJ and Wiles J.L, 1998. Chitosan film mechanical and permeation properties as affected by acid, plasticizer and storage. Journal of Food Science. 63(6):1049-1053.

Carlin F, Nguyen-the C, Chambroy Y, Reich M, 1990. Effects of controlled atmospheres on microbial spoilage, electrolyte leakage and sugar content of fresh 'ready to use' grated carrots. International Journal of Food Science and Technology. 25:110–119.

Cong F, Zhang Y, and Dong W, 2007 .Use of surface coatings with natamycin to improve the storability of Hami melon at ambient temperature. Postharvest Biology and Technology. 46:71–75.

Dunn RA, Hall WN, Altamirano JV, Dietrich SE, Robinson-Dunn B and Johnson DR, 1995. Outbreak of *Shiegella flexneri* linked to salad prepared at a central commissary in Michigan. Public Health Reports. 110(5):580-586.

Debeaufort F, Quezada-Gallo JA, and Voilley A, 1998. "Edible films and coatings: tomorrow packaging: a review". Critical Reviews in Food Science. 38(4):299–313.

Dominguez-Lo´pez, A, 1995. "Review: use of the fruit and stems of the prickly pear cactus (*Opuntia spp*.) into human food". Food Science and Technology International. 1: 65–74.

FAO, 2001. Food Balance Sheets: A Hand Book. Economic and Social Development Department, Food and Agriculture Organization of United Nations, Viale delle Terme di Caracalla, 00153 Rome, Italy, pp: 14.

Fleet GH, 1992. Spoilage yeasts. Critical Reviews in Biotechnology. 12: 1–44.

Guilbert S. Gontard N. and Gorris LGM, 1996. Prolongation of the shelf-life of perishable food products using biodegradable films and coatings. Lebensmittel-Wissenschaft und-Technologie., 29:10-17.

Gonz´alez-Aguilar GA, 2009. Effect of chitosan coating in preventing deterioration and preserving the quality of fresh-cut papaya "Maradol". Journal of the Science Food Agriculture. 89:15–23.

Garg N, Churey JJ, Splittsotesser DF. 1990 Effect of processing conditions on the microflora of fresh-cut vegetables. Journal of Food Protection. 53:701–703.

Garcia-Gimeno RM, and Zurera-Cosano G, 1997. "Determination of ready-to-eat vegetable salad shelf-life". International Journal of Food Microbiology. 36:31–38.

Gislene GF, Locatelli N.J, Paulo CF and Giuliana LS, 2000. Antibacterial activity of plant extracts and phytochemicals on antibiotic resistant bacteria. Brazilian Journal of Microbiology.,31:247-256.

Herdberg CW, McDonald KL and Osterholm MT,1994. Changing epidemiology of foodborne disease: a Minneasota perspective. Clinical Infectious Diseases., 8:671-682.

Hill T. and Lewicki P. (2007). Statistics Methods and Applications. Tulsa, OK. StatSoft, USA.

Hotchkiss J and Banco M,1992. "Influence of packaging technologies on the growth of microorganisms in produce". Journal of Food Protection.,55:815–820.

Hosokawa J, Nishiyama M, Yoshihara K, Kubo T. 990 Biodegradable film derived from chitosan and homogenized cellulose. Industrial and Engineering Chemistry Research .,29(1):800-805.

Kalia A and Gupta RP. Fruit Microbiology, in Hui Y.H, J., Cano, M.P., Gusek, W., Sidhu, J.W., Sinha, N.K,2006. Handbook of Fruit and Fruit processing. 1st Edition, Blackwell publishing., pp 3-28.

Li P and Barth MM, 1998. "Impact of edible coatings on nutritional and physiological changes in lightly processed carrots". Postharvest Biology and Technology., 14:51-60.

McHugh TH and Krochta JM, 1994a. "Milk protein based edible films and coatings". Food Technology., 48:97-103.

McHugh TH and Krochta JM,1994b. "Sorbitol vs glycerol plasticized whey protein edible films: Integrated oxygen permeability and tensile property evaluation". Journal of Agricultural and Food Chemistry., 42: 41-45.

Mahmoud R and Savello PA ,1992. "Mechanical properties of and water vapor transferability through whey protein films". Journal of Dairy Science., 75: 942-946.

Ouattara B, Simard R, Piette G, Be´gin A, and Holley RA. 2000 "Inhibition of surface spoilage bacteria in processed meats by application of antimicrobial films prepared with chitosan". International Journal of Food Microbiology.,62:139–148.

Olivas GI and Barbosa-Canovas GV, 2005. "Edible coatings for fresh-cut fruits". Critical Reviews in Food Science and Nutrition., 45:657-670.

Ofor MO, Okorie VC, Ibeawuchi II, Ihejirika GO, Obilo OP and Dialoke SA. 2009 Microbial Contaminants in Fresh Tomato Wash Water and Food Safety Considerations in South-Eastern Nigeria. Life Science Journal., 1:80-82.

Padgett T, Han LY and Dawson, P. L, 1998. "Incorporation of food-grade antimicrobial compounds into biodegradable packaging films". Journal of Food Protection., 61(10):1330–1335.

Park HJ, Chinnan MS and Shewfelt RL, 1994a. "Edible coating effects on storage life and quality of tomatoes". Journal of Food Science., 59:568-570.

Park HJ, Chinnan MS and Shewfelt RL, 1994b. "Edible corn-zein film coatings to extend storage life of tomatoe"s. Journal of Food Processing and Preservation., 18:317-331.

Ray B and Bhunia AK. 2007 Fundamental Food Microbiology. 4th Edn., CRC Press, USA. p. 492.

Myllarinen P, Partanen R, Sepalla J and Forsell P, 2002. Carbohydrate. Polymer., 50:355–61.

Rodriguez A, Batlle R and Ner´ın C, 2007. The use of natural essential oils as antimicrobial solutions in paper packaging. Part II. Progress in Organic Coatings., 60:33–38.

Rojas-Grau, M., Grasa-Guillem, R., Martin-Belloso, O., 2007. "Quality changes in fres-cut Fuji apple as
effected by ripeness stage, antibrowning agents and storage atmosphere. Journal of Food Science., 72(1):36-42.

Sa´enz, C, Va´squez, M, Trumper S, and Fluxa´ , C, 1992. "Extractio´n y Composicio´n Quı´mica del Mucı´lago de Tuna (Opuntia ficus indica). *In Proceedings II Congreso Internacional de Tuna y Cochinilla"*.pp 93–96.

Seenivasan P, Manickkam J and Savarimuthu I, 2006. *In vitro* antibacterial activity of some plant essential oils. Complementary alternative medicine ., 6: 39.

Tyagi AK and Malik A, 2010. Liquid and vapour-phase antifungal activities of selected essential oils against *Candida albicans*. Complementary alternative medicine.,10:65.

Tournas VH, 2005. Moulds and yeasts in fresh and minimally processed vegetables, and sprouts. International Journal of Food Microbiology., 99:71–77.

Tournas VH and Katsoudas E. 2005 Mould and yeast flora in fresh berries, grapes and citrus fruits. International Journal of Food Microbiology. 105:11–17.

Nguyen-the C and Carlin F. 1994 The microbiology of minimally processed fresh fruits and vegetables. Critical Reviews in Food Science and Nutrition. 34:371–401.

Vescovo M, Torriano S, Orsi C, Macchiarolo F, and Scolari G, 1996. Application of antimicrobial-producing lactic acid bacteria to control pathogens in ready-touse vegetables. Journal of Applied Bacteriology. 81:113–119.