



Evaluation of nanocomposite packaging containing Ag and ZnO on shelf life of fresh orange juice

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ABSTRACT

Nanocomposite LDPE films containing Ag and ZnO nanoparticles were prepared by melt mixing in a twin-screw extruder. Packages prepared from the films were then filled with fresh orange juice and stored at 4 °C. Microbial stability, ascorbic acid (AA) content, browning index, color value, and sensory attributes of them were evaluated after 7, 28, and 56 days of storage. Packages containing the nanomaterials, except 1% nano-ZnO, kept the microbial load of fresh juice below the limit of microbial shelf life (6 log cfu/ml) up to 28 days. The least degradation of AA (80.50 mg/100 g), development of brown pigments (OD = 0.23) and losing of color ($\Delta E = 6.0$) were observed in pouches containing 0.25% nano-ZnO, after the same time. Sensory attributes were also ranked highest for the juice thus packed in the recent packages after 28 days ($p < 0.05$). Packages containing nanosilver increased shelf life of fresh juice although part of its sensory attributes were lost.

Industrial relevance: Compared with pure packaging materials, antimicrobial nanocomposite packages containing Ag and ZnO as an alternative non-thermal technology can extend the shelf life of fresh orange juice up to 28 days. However, a certain concentration of nano-ZnO in the packages showed less adverse effects on sensory characteristics.

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1. Introduction

Orange juice is one of the most globally accepted fruit products (Meléndez-Martínez, Vicario, & Heredia, 2007). Demand for natural orange juice with high quality in terms of nutritional value, physico-chemical properties and sensory characteristics with minimal or no heat treatment has increased considerably (Bull et al., 2004; Souza, Benassi, Meneghel, & Silva, 2004). Natural orange juice, even kept under refrigeration, has a short shelf life due to increasing microbial spoilage (Souza et al., 2004). Recently, extensive studies have been conducted to develop non-thermal processing techniques (PEF, HHP, IR, UV, and US) as replacements for thermal processing in order to keep the freshness of the juice along with extending its shelf life (Baxter, Easton, Schneebeli, & Whitfield, 2005; Elez-Martínez, Soliva-Fortuny, & Martín-Belloso, 2006; Foley et al., 2002; Tran & Farid, 2004; Valero et al., 2007). Although some of these technologies are capable of decontaminating orange juice, they are energy-intensive and require costly equipment; hence, their yet relatively limited commercial applications (Han, 2007). Nanotechnology recently introduced in the food packaging industry can potentially provide solutions to food packaging challenges such as short shelf life (Chaudhry et al., 2008; Joseph & Morrison, 2006). Antimicrobially active

packaging is a new generation of nano food packaging based on metal nanocomposites which are made by incorporating metal nanoparticles into polymer films (Chaudhry et al., 2008). The high performance of nanoparticles is due to their high surface area/volume ratio, which is the main reason for increasing antimicrobial activity of metal nanoparticles (Damm, Neumann, & Münstedt, 2006).

Nanoparticles (NP) of Ag and ZnO are being used industrially for several purposes (Gajjar et al., 2009). ZnO has found many applications in daily life such as in drug delivery, cosmetics, and medical devices (Yan et al., 2009) due to its strong antimicrobial effect on a broad spectrum of microorganisms (Jones, Ray, Ranjit, & Manna, 2008). Moreover, it is currently listed by FDA as a generally recognized as safe (GRAS) material (Jin, Sun, Su, Zhang, & Sue, 2009). Silver has also been long known to have microbial inhibition (Lok et al., 2006). The antimicrobial activity of these nanoparticles may be related to several mechanisms including, induction of oxidative stress due to generation of reactive oxygen species (ROS) which may cause the degradation of the membrane structure of the cell (Sawai, 2003; Sawai & Yoshikawa, 2004; Sawai et al., 1998), release of ions from the surface of nanoparticles that has been reported to cause bacterial death due to binding to cell membrane (Feng et al., 2000; Sondi & Salopek-Sondi, 2004). However, the mechanism of toxicity is still only partially understood (Li et al., 2008).

Several methods are generally used to produce antimicrobial polymer nanocomposites. Because of the thermal stability of metal nanoparticles and the thermal processing method used for producing the LDPE film as a contacting juice layer in the package, melt mixing is

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a good approach for this nanocomposite (Appendini & Hotchkiss, 2002; Damm et al., 2006).

The main objectives of this study is to evaluate the capabilities of ZnO and Ag nanoparticles filled LDPE nanocomposite packaging as a new approach to preservation and prolonging shelf life of orange juice.

2. Experimental

2.1. Preparation of antimicrobial nanocomposite films

Film grade LDPE resin pellets (LF0200, MFI 2 g/10 min, density 0.92 g/ml, softening point 94 °C) and antimicrobial agents including P105 powder (a combination of 95% TiO₂ powder which provided a base for doping of nanosilver, plus 5% metal nanosilver with particle diameters of about 10 nm) and ZnO nanoparticle powder with an average particle diameter of about 70 nm (Fig. 1a, b) were obtained from Pars Nanonassb Tehran, Iran. Film grade LDPE resin pellets (0.9 kg) were directly mixed with each of the antimicrobial agents (P105 and nano-ZnO particles) (0.1 kg) separately and the mixture was fed into a twin-screw extruder machine (Cincinnati Milacron, Batavia, OH) with a screw diameter of 55 mm and a screw length/diameter ratio of 30 mm to be cut into masterbatch nano-granules. The mass fraction of the filler for each antimicrobial agent was 10%. The heating profile was set to six heating zones of the twin-screw extruder including 160 °C, 160 °C, 175 °C, 150 °C, 150 °C, and 140 °C. Proper amounts of masterbatch resins were then added to pure LDPE resin pellets into a single-screw blowing machine with a screw diameter of 45 mm and a length/diameter ratio of 28 mm (Venus Plastic Machinery, Taiwan) to fabricate the final nanocomposite film (50 µm thick) with the desired nanomaterial concentrations (0.25 and 1% for nano-ZnO and 1.5 and 5% for P105). The temperature profile for the single extruder was maintained at 190 °C in the two barrel zones.

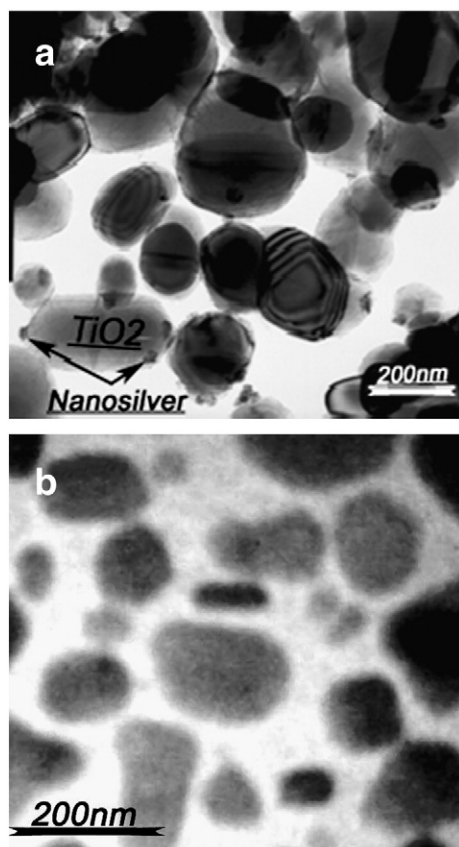


Fig. 1. TEM micrograph of a: P105 and b: nano-ZnO.

Film thickness was measured using a micrometer (Mitutoyo, Japan) and reported as the average of five readings taken at five different points on the film sample.

2.2. Transmission electron microscopy analysis

Dispersion quality of nanomaterials into the polymer matrix film was monitored using the Transmission Electron Microscope (PHILIPS CM 200 kV, The Netherlands).

2.3. Orange juice production

To prepare natural orange juice, 30 kg of oranges (*Citrus sinensis* cv. Khaf) were purchased from the local market in Isfahan, Iran. They were juiced using a semi-industrial juice extractor (M2000A-1, CMEC food machinery, China) equipped with a central fruit halving knife and a pair of holding cups, 90 mm in diameter, thoroughly washed with detergent and hot water. The juice (with an efficiency of 25.8%) was passed through a 1 mm mesh filter and immediately transferred into a sterile glass container under sanitized conditions. Packages were prepared by a hand heat sealer using antimicrobial nanocomposite and pure LDPE films 15×10 cm in size, similar to Doypack packaging commonly used for packaging fruit juice. The packages were immediately wrapped in aluminum foil and sanitized at 95 °C for 2 min. After cooling and under a sterile laboratory hood, 175 ml of fresh orange juice was poured into each package and sealed by the heat sealer.

2.4. Storage

Packages containing orange juice were stored in dark and cool conditions (4 °C). The samples were evaluated in duplicate for their microbiological, physicochemical, and sensory characteristics immediately after packaging and after 7, 28, and 56 days of storage.

2.5. Microbiological evaluations

Decimal dilutions were prepared from orange juice samples with sterile peptone water (0.1%). Volumes of dilution samples (0.1 ml) were then used. Total aerobic plate counts were enumerated using the pour plate method on the plate count agar (PCA, Scharlau Chemie, S.A., Barcelona, Spain). Incubation was performed at 30 °C for 3 days. Total yeast and moulds were enumerated using the surface plate method on the potato dextrose agar (PDA, Scharlau Chemie, S.A., Barcelona, Spain) + 10% tartaric acid. Incubation for total yeast and mould counts was performed at 25 °C for 5 days. Each test was performed in duplicate and results were expressed as colony-forming units (CFU) per milliliter.

2.6. Ascorbic acid degradation

Most chemical analyses are based on the fact that ascorbic acid is easily oxidized. The most common method relies on the reduction of 2,6 dichlorophenolindophenol reagent. Ascorbic acid degradation was determined using the titrimetric method (A.O.A.C., 1967.21, 2002a).

2.7. Color measurement

Color was measured using a digital imaging method that used a combination of a digital camera (Panasonic, Japan), a computer, and a graphics software. A Petri dish containing 25 ml of orange juice was placed into the lighting system that consisted of two CIE source D65 lamps 45.0 cm long, mounted on the two sides of a frame installed on either side of the Petri dish, 30.5 cm above and at an angle of 45° to the orange juice sample plane. Images of the bottom surface of the orange juice were taken and saved using the digital camera that was placed 30.5 cm above the sample with its lens facing downwards towards the

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