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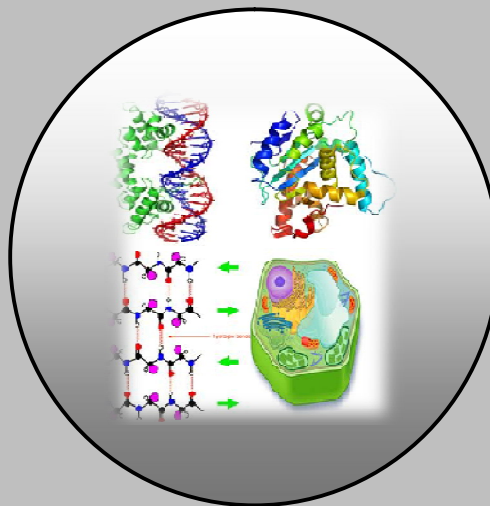
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RESEARCH PAPER

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Effect of ZnO Nano Particle Containing Packaging on Shelf Life of Fresh Nagpur Mandarin (*Citrus reticulata* Blanco) Segments

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ABSTRACT

*The ZnO Nanocomposites film used for the study was characterized by 40 nm ZnO nano particle embedded in 60 µm thickness poly film with tensile strength of 3.35±0.50 kg longitudinally and 1.67±0.27 kg transversely average breaking load which was provided by CIRCOT, Mumbai. Microbial stability, total soluble solid, acidity, ascorbic acid (AA) content, browning index, color value, limonin, carotenoid and sugars of Nagpur mandarin segments (*Citrus reticulata* Blanco) packed in ZnO nano particle containing film and LDPE film were studied at monthly interval in refrigerated storage. Packaging containing ZnO nano particle was suitable to maintain the microbial load below the threshold population limit (6 log CFU/ml) till 30 days of storage. The segments had better color in ZnO nano packaging after 30 days of storage. The total soluble solids (TSS), vitamin-C and carotenoid contents were found better in ZnO nano packaging than that of LDPE packaging. Relatively lesser value of browning was recorded in ZnO nanoparticle containing film (0.62mg/l). The limonin content was recorded to be higher than the threshold limit in both the packaging. The changes in segments juice reducing sugar; non-reducing sugar and total sugar were found to be at higher side in LDPE packaging than that of nano packaging. Study revealed that segments in nano packaging had 30 days of shelf life with better quality under refrigerated storage (4°C) temperature.*

Keywords – ZnO Nano Particle Containing Packaging, LDPE Packaging, Zinc Oxide Nano Particles, Nagpur Mandarin Segments, Refrigerated Storage, Microbial Stability.

INTRODUCTION

Citrus juice is one of the most globally accepted fruit products (Meléndez-Martínez, *et. al.*, 2007). Demand for natural loose jacketed orange juice having high nutritional, physico-chemical properties and sensory value with minimal or without heat treatment has increased considerably (Bull *et al.*, 2004; Souza, *et.al.* 2004). Natural orange juice, even kept under refrigeration, has a short shelf life due to increasing microbial spoilage (Souza *et. al.*, 2004). Recently Nanotechnology introduced in the food packaging industry can potentially provide solutions to challenges of short shelf life (Chaudhry *et al.*, 2008; Joseph and Morrison, 2006), anti-microbial active packaging is a new development of nano food packaging based on metal Nano composites which are made by incorporating metal nano particles into polymer films. 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, *et.al.*, 2008). Moreover, it is currently listed by FDA as a generally recognized as safe (GRAS) material (Jin, *et.al.*, 2009). Various methods have generally been used to produce antimicrobial polymer nano composites. The main objectives of this study is to evaluate the applicability of ZnO nano composite packaging as a new innovative approach for safe preservation with enhanced shelf life of Nagpur mandarin segments.

MATERIAL AND METHODS

The ZnO nanocomposites film of 50 μ m thicknesses was prepared by melt-blow method using 1:1 ratio of HDPE: LDPE and 1% concentration of ZnO nanocomposites provided by CIRCOT, Mumbai. The ability of the Nanocomposite to block UV light is measured in Specord 50 ANALYTIKJENA[®] UV- Visible spectrophotometer using an Integrating Sphere loaded with sample from 280 nm at an interval of 2 Nagpur mandarin reported by Vigneshwaran *et al.*, (2008).

Mature *Ambia* Nagpur mandarin fruits were collected from orchard and brought to processing laboratory for washing and cleaning with distilled water to remove surface residues. After that, segments were separated from the fruits and packed in two different types of packets i.e. nano zinc oxide composed packaging (50nm) and LDPE packaging (50 μ m) and stored in refrigerated condition at 4^oC temperature and observations were recorded periodically on color and TSS using digital colorimeter and digital refractometer, respectively. Acidity, ascorbic acid, browning index were determined by the method given by AOAC (1990). Limonin, carotenoids and sugars were estimated by method given by Wilson and Cruthchfield (1968), Ting, *et.al.*, (1986) and dinitro-Salicylic acid (DNSA) Miller (1972) respectively.

The microbiological evaluation of Nagpur mandarin segments stored in different packaging was done by serial dilutions which were made from segment juice samples using sterile distilled water. 1ml of segment juice sample was used. Total aerobic plate count was enumerated using 'spread plate technique' on sterile Nutrient agar and Potato Dextrose agar total aerobic count was taken and expressed in log CFU/ml (Aryou Emamifar, *et.al.*, 2011). The experimental data was statistically analyzed by using the method of Panse and Sukhatme (1967).



Figure 1



Figure 2

Figure. 1 and 2 Nagpur mandarin segments separation and nano packaging under aseptic condition.

RESULTS AND DISCUSSIONS

Study revealed that orange colour (a/b ratio) were found to be increased to 0.48 in ZnO nano packaging, while it was decreased to 0.08 in LDPE packaging than that of 0.32 in fresh juice colour indicating the attractive colour of segments maintained in nano packaging than that of LDPE packaging of Nagpur mandarin fruits. However, weight of test packets were found to be reduced both in ZnO nano packaging and LDPE packaging after 30 days in refrigerated storage (Table 1). The total soluble solids (TSS), acidity, vitamin-C and carotenoid contents were recorded to be decreased comparatively in nano packaging after 30 days in refrigerated storage compared to initial fresh Juice, but the decrease in value was higher in LDPE packaging than the nano packaging. While reverse trend were found in respect of browning of Nagpur mandarin segments in different packaging, which showed the relatively lesser value of browning as 0.60 mg/l in ZnO nano packaging and 0.64mg/l in LDPE packaging. The segment juice was bitter in both the packaging than that of fresh juice (6.00 ppm). The limonin content was recorded to be on par as 10.72 ppm in ZnO nano packaging segments and 10.78 ppm in LDPE packaging after 30 days of refrigerated storage indicating higher than that of threshold value. The slight changes in reducing sugar, (1.10 mg/ml), total sugar (4.41 mg/ml) and non reducing sugar (2.30 mg/ml) of LDPE packaging were recorded in comparison to initial fresh segment juice as 1.40, 5.23 and 3.64 mg/ml, respectively. The changes in juice reducing, non-reducing and total sugar were found to be at higher side in LDPE packaging than that of segments in nano packaging, respectively. The segments in both the packaging had 30 days of shelf life under refrigerated storage (4⁰C) temperature, possibly due to over lapping of segments in the test packages which become watery and lost the shelf life within 30 days in refrigerated storage (Table 2).

Total aerobic count on nutrient agar from Nagpur mandarin segments juice after 30 day of storage was found to be 2.318 log CFU/ml (208 CFU/ml) in Nano packaging and 2.556 log CFU/ml (360 CFU/ml) in LDPE packaging.

Total aerobic count on Potato Dextrose agar from Nagpur mandarin segments juice samples after 30 day of storage was found to be 2.274 log CFU/ml (188 CFU/ml) in Nano packaging and 2.565 log CFU/ml (368 CFU/ml) in LDPE packaging, indicating that this total aerobic plate count is safe limit because the shelf life of segment is defined as 'the time required to reach a microbial population of 6 log CFU/ml' (Raccach and Mellatdoust, 2007) (Table 3).

Table 1. Effect of ZnO Nano particle containing film on PLW and colour quality of Nagpur mandarin segments under refrigerated storage (4°C).

Parameters		Initial		30 days in refrigerated storage (4°C)		
		ZnO Nanopackaging (50 nm)	LDPE (50 nm)	ZnO Nanopackaging (50 nm)	LDPE (50 nm)	CD at 0.05%
Segment colour	*L	54.29	54.29	53.17	46.68	0.05
	*a	6.75	6.75	9.52	10.67	0.05
	*b	15.73	15.73	26.83	21.78	0.05
	*a/b	0.43	0.43	0.35	0.49	0.05
Weight of Packets (g)		229.52	228.37	228.08 (0.62%)**	228.23 (0.06%)*	NS

*L: lightness/darkness, *a: Redness, *b: yellowness, *a/b: orange ** Physiological loss in weight (%)

Table 2. Effect of ZnO Nano particle containing film on biochemical composition of Nagpur mandarin segments under refrigerated storage (4°C).

Parameters	Initial		30 days in refrigerated storage (4°C)		
	ZnO Nanopackaging (50 nm)	LDPE packaging (50 nm)	ZnO Nanopackaging (50 nm)	LDPE packaging (50 nm)	CD at 0.05%
TSS (%)	9.60	9.60	8.30	8.10	NS
Acidity (%)	0.59	0.59	0.41	0.48	0.05
Vit C (mg/100ml)	32.18	32.18	17.60	17.00	NS
Browning (mg/l)	0.40	0.40	0.60	0.64	NS
Limonin (ppm)	6.00	6.01	10.70	10.78	NS
Carotenoid (mg/100ml)	0.18	0.18	0.10	0.10	NS
Reducing sugar (mg/ml)	1.40	1.40	1.32	1.10	0.16
Non-reducing Sugar (mg/ml)	3.64	3.64	3.10	2.30	0.16
Total Sugar (mg/ml)	5.23	5.23	4.88	4.41	0.24

Table 3. Effect of ZnO Nano particle containing film on total aerobic count of Nagpur mandarin segments juice under storage at 4°C.

Storage period (4°C)	Total aerobic count log (CFU/ml) on Nutrient agar			Total aerobic count log (CFU/ml) on potato dextrose agar		
	ZnO packaging	Nano packaging	LDPE packaging	ZnO packaging	Nano packaging	LDPE packaging
Initial	0.00		0.00	0.00		0.00
30 days	2.318		2.556	2.274		2.565

CONCLUSION

This study showed that application of film containing ZnO nano particles is a new approach for preserving and extending the shelf life of fresh Nagpur mandarin segments under storage condition at 4 °C temperature. The quality of the packaging film including good dispersion of nanomaterials in the polymer matrix free from agglomeration was shown to be very effective on the antimicrobial effects of these packaging materials. Application of packages containing nano-ZnO prolonged the shelf life of Nagpur Mandarin segments up to 30 days without any negative effects on sensorial attributes and physiological parameters. Study is in progress for detailed study.

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REFERENCES

- AOAC. (1990). Official Methods of Analysis. Association of Analytical Chemists, 15th Ed. Washington DC.
- Aryou Emamifar, Mahdi Kadivar, Mohammad Shahedi, Sabihe Soleimani-Zad (2011). Effect of nanocomposite packaging containing Ag and ZnO on inactivation of *Lactobacillus plantarum* in orange juice. *Food Control*, 22: 408-413
- Bull, M. K., Zerdin, K., Goicoechea, D., Paramanandhan, P., Stockman, R., and Sellahewa, J.(2004). The effect of high pressure processing on the microbial, physical and chemical properties of Valencia and Navel orange juice. *Innovative Food Science and Emerging Technologies*, 5, 135–149.
- Chaudhry, Q., Scotte, M., Blackburn, J., Ross, B., Boxall, A., and Castle, L. (2008). Applications and implications of nanotechnologies for the food sector. *Food Additives and Contaminants*, 25(3), 241–258.
- Jin, T., Sun, D., Su, J. Y., Zhang, H., & Sue, H. J. (2009). Antimicrobial efficacy of zinc oxide quantum dots against *Listeria monocytogenes*, *Salmonella enteritidis*, and *Escherichia coli* O157:H7. *Journal of Food Science*, 74(1), 46–52.

- Jones, N., Ray, B., Ranjit, K. T., and Manna, A. C. (2008). Antibacterial activity of ZnO nanoparticle suspensions on a broad spectrum of microorganisms. *FEMS Microbiology Letters*, 279, 71–76.
- Joseph, T., and Morrison, M. (2006). Nanotechnology in agriculture and food. A Nanoforum report www.nanoforum.org, 14 pp..
- Meléndez-Martínez, A. J., Vicario, I. M., and Heredia, F. J. (2007). Review: Analysis of carotenoids in orange juice. *Journal of Food Composition and Analysis*, 20, 638–649.
- MILLER, G.L., 1972, Use of dinitro-salicylic acid reagent for determination of sugar. *Annals of Chemistry*, 31: 426-428.
- Panse, V.G. and Sukhatme, P.V. 1967. *Statistical Methods for Agricultural Workers*. 2nd edn. ICAR. New Delhi.
- Raccach, M. M., and Mellatdoust, M. (2007). The effect of temperature on microbial growth in orange juice. *Journal of Food Processing and Preservation*, 31, 129–142.
- Souza, M. C. C., Benassi, M. T., Meneghel, R. F. A., and Silva, R. S. S. F. (2004). Stability of unpasteurized and refrigerated orange juice. *Brazilian Archives of Biology and Technology*, 47(3), 391–397.
- Ting, S.V. Russell and Rouseff, L. (1986). *Citrus Fruits and their products. Analysis and Technology*, Florida Market Deccar, Inc. ACC-6.
- Vigneshwaran, N., Bharimalla, A.K., Prasad, V., Kathe, A.A. and Balasubramanya, R.H. (2008). Functional behavior of polyethylene- ZnO nanocomposites, *J. Nanosci. Nanotech* vol. 8(7): 1-6.
- Wilson, K.W and C.A. Cruthchfield (1968). Spectrophotometric determination of limonin in orange juice. *J. Agr. Food Chem* 16: 118-124.
- Yan, D., Yin, G., Huang, Z., Yang, M., Liao, X. and Kang, Y. (2009). Characterization and bacterial response of zinc oxide particles prepared by a bio mineralization process. *The Journal of Physical Chemistry*, 113: 6047–6053.

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