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# Shelf life extension of ground chicken meat using an oxygen absorber and a citrus extract

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#### ABSTRACT

In the present study the combined effect of an  $O_2$  absorber and a citrus extract (0.1 and 0.2 ml/100 g), on shelf life extension of fresh ground chicken stored at 4 °C was investigated. Microbiological changes [Total viable count (TVC), Pseudomonads, Lactic acid bacteria (LAB), Enterobacteriaceae, and *Clostridium* spp.)], physicochemical changes (pH, total volatile nitrogen (TVB-N), and colour) and sensory changes (colour, odour and taste) were monitored as a function of treatment and storage time (14 days). Aerobically packaged ground chicken meat stored at 4 °C was taken as the control sample. On day 6 of storage TVC was reduced by 0.5, 1.0 and 1.5 log cfu/g by the citrus extract (0.2 ml/100 g), the  $O_2$  absorber and the combination of the  $O_2$  absorber plus citrus extract (0.2 ml/100 g), respectively (p < 0.05). Initial pH (6.38) showed an increase (6.73) or decrease (5.83) depending on specific treatment. TVB-N ranged between 42.5 and 57.5 mg/kg at the time of sensory rejection. Colour parameters remained unaffected in samples containing the  $O_2$  absorber and/or citrus extract. A 4–5 days product shelf life extension using the combination of  $O_2$  absorber and citrus extract (0.1 ml/100 g) as compared to control samples was achieved.

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#### 1. Introduction

Poultry meat is a very popular food commodity around the world due to its low cost of production, low fat content, high nutritional value and distinct flavour (Barbut, 2002; Patsias, Badeka, Savvaidis, & Kontominas, 2008). Processed chicken products' consumption has also dramatically increased over the last decades (Bianchi, Ferioli, Petracci, Caboni, & Cavani, 2009). As a result, extending the shelf life of perishable chicken products is a major concern for the poultry industry (Wang, Chang, & Chen, 2004). Shelf life of fresh chicken and chicken products depends mainly on the initial bacterial load (Fernández-López, Zhi, Aleson-Carbonell, Pérez-Alvarez, & Kuri, 2005). Chopping and/or grinding of poultry products, resulting in an increased surface to weight ratio, may further enhance product quality deterioration due to additional microbial contamination. Furthermore, processing and storage conditions may greatly affect product shelf life (Mead, 1990). Due to the constantly growing consumer awareness regarding the relation between food and health (Viuda-Martos, Ruiz-Navajas, Fernández-López, & Pérez-Álvarez, 2008), the need to preserve foodstuffs using natural preservatives and environmentally friendly technologies is obvious (Coma, 2008).

The concept of combining several preservation technologies has been developed by Leistner into the 'hurdle effect' (Leistner, 1995). No single technology is responsible for making the product stable, but rather stability results from the synergistic or additive action among the combined technologies. Hurdle technology produces minimal sensory changes, which make food products more acceptable than those processed by severe thermal conventional methods (Aguilera & Chirife, 1994). According to Alakomi, Skytta, Helander & Ahvenaine (2002) there are three types of hurdles: physical hurdles such as modified atmosphere/active packaging, chemical hurdles such as organic acids and phenols and microbially derived hurdles such as bacteriocins.

Ageless<sup>®</sup> is the most common O<sub>2</sub> absorber system based on iron (Fe <sup>2+</sup>) oxidation (Nakamura & Hoshino, 1983). It is widely employed in the food industry in Japan as an alternative to nitrogen and vacuum packaging techniques for food preservation. It is promoted as a non-toxic, residue-free method of preserving food-stuffs against mould and aerobic bacterial growth (Grattan & Gilberg, 1994).

Citrus essential oils are the most widely used essential oils in the world (Bousdia, Vian, Ferhat, Meklati, & Chemat, 2009). They are extracted from the peel of citrus fruit (Dharmawan, Kasapis,

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Sriramula, Lear, & Curran, 2009). The phenolics that occur in citrus oil include the flavonoids (flavanones, flavones, and flavonols), anthocyanins and coumarins among others (Berhow, Tisserat, Kanes, & Vandercook, 1996, p. 158). Flavonoids have shown antimicrobial activity in several studies (Tim Cushnie & Lamb, 2005). Citrus essential oils have been found to inhibit Gram-positive and Gram-negative bacteria as well as yeasts, moulds and foodpoisoning bacteria (Deans & Ritchie, 1987) both in the form of liquid and vapour.

Organic acids are natural constituents of plant and animal tissues; they exhibit antibacterial activity while many of them have been classified as: generally recognized as safe [GRAS] (Friedly et al., 2009). The antimicrobial activity of organic acids is attributed to pH reduction, depression of internal pH of microbial cells by ionization of acid molecules, and disruption of substrate transport by altering cell membrane permeability (Jay, Loessner, & Golden, 2005, chap. 2, 4).

Based on the above, the objective of the present work was to study the combined effect of the  $O_2$  absorber "Ageless<sup>®</sup>" and "Citrox<sup>®</sup>" (a commercial citrus extract containing: 18 g citric acid/100 ml, 18 g malic acid/100 ml, and 5 g ascorbic acid/100 ml) on shelf life extension of fresh ground chicken meat stored at 4 °C.

#### 2. Materials and methods

#### 2.1. Raw materials and storage conditions

Fresh chicken breast meat was provided by a local poultry processing plant (Pindos SA. Joannina, Greece) within 1 h after slaughter in insulated polystyrene boxes on ice. It was then ground in a laboratory meat grinder (Kenwood model MG510, Havant, UK) previously sterilized using hot water. O2 absorber sachets [oxygen absorption capacity: 50 ml O<sub>2</sub>] and the Ageless<sup>®</sup> eye oxygen indicators were supplied by Mitsubishi Gas Chemical Company, Tokyo, Japan. Selection of the most suitable type of Ageless® sachet to produce a low  $O_2$  atmosphere depends on the water activity of the packaged commodity. Ageless® FX type oxygen absorbers recommended for food products, like ground chicken meat, with a water activity value higher than 0.85 were selected for this study. Both LDPE and LDPE/EVOH/LDPE films were used to package ground chicken meat. The oxygen permeability of LDPE and LDPE/EVOH/ LDPE films was measured using the Oxtran 2/20 oxygen permeability tester (MOCON, Inc., MN, USA) at 25 °C. These data were used to calculate the concentration of oxygen which may permeate the films during storage (14 days) in order to choose an absorber with an ability to absorb the initial oxygen within the package and the oxygen expected to permeate in the package during storage. Based on this, an oxygen absorber with an absorption capacity of 50 ml of O<sub>2</sub> was chosen. Ageless-eye<sup>®</sup>, a low O<sub>2</sub> concentration indicator tablet, was used as a simple qualitative test to check for the absence of oxygen inside the sealed packages. The eye oxygen indicator turns from purple to pink in colour when O<sub>2</sub> concentration falls to 0.1 ml/ 100 g. Citrox® extract was supplied by Polypan Group, Athens, Greece. Pouch dimensions were 29 × 10 cm, pouch capacity was 200 ml and pouch headspace volume was 50 ml. Six lots of samples were prepared at room temperature: The first lot comprised the air packaged (control) samples (150 g/pouch). Citrox® extract was pipetted to the second and third lot so as to obtain a final concentration equal to 0.1 and 0.2 ml/100 g, respectively. Samples were massaged by hand so as to obtain a homogeneous distribution of the citrus extract in chicken meat. In lot four, the O2 absorber and the Ageless® eye oxygen indicator were added to the inner surface of the package wall. Lots five and six contained both citrox® extract (0.1 and 0.2 ml/100 g, respectively) and the O<sub>2</sub> absorber and Ageless<sup>®</sup> eye oxygen indicator. All samples were aerobically packaged. Lots 1, 2 and 3 were packaged in LDPE pouches, 75 μm in thickness, having an oxygen permeability of 5500 ml<sup>3</sup>/(m<sup>2</sup> day atm) at 75% relative humidity (RH), 25 °C. Lots 4, 5 and 6 were packaged in low density polyethylene/ethylene vinyl alcohol/low density polyethylene (LDPE/EVOH/LDPE) high barrier pouches, 75 μm in thickness, having an oxygen permeability of 2 ml<sup>3</sup>/(m<sup>2</sup> d atm) under same RH and temperature conditions as above. LDPE was chosen (treatments Lots 1, 2 and 3) due to its very high oxygen permeability to maintain an aerobic environment during the entire storage period. On the other hand the oxygen absorber cannot function properly unless it is used in combination with a high barrier material such as LDPE/EVOH/ LDPE. This material was chosen to create an oxygen free atmosphere in combination with the oxygen absorber in treatments 4,5 and 6. Pouches were heat-sealed using a BOSS model N48 thermal sealer (BOSS, Bad Homburg, Germany) and stored in a commercial glass door display cabinet at 4 °C. Sampling was carried out at 2-day intervals for 14 days.

#### 2.2. Microbiological analysis

The following groups of microflora were monitored: Total viable counts (TVC), Pseudomonads, Lactic acid bacteria (LAB), Enterobacteriaceae, as well as *Clostridium* spp. Twenty five grams of fresh ground chicken meat were removed aseptically from the package using a spoon, transferred to a stomacher bag (Seward Medical, Worthing, West Sussex, UK), containing 225 ml of sterile quarterstrength Ringer's solution, and homogenized using a stomacher (Lab Blender 400, Seward Medical) for 60 s at room temperature. For microbial enumeration, 0.1 ml samples of serial dilutions (1:10. diluent, quarter-strength Ringer's solution) of ground chicken meat homogenates were spread on the surface of agar plates. TVC were determined using plate count agar (PCA; Merck, Darmstadt, Germany), after incubation for 3 days at 30 °C. Pseudomonads were determined on cetrimide fusidin cephaloridine agar (Oxoid code CM 559, supplemented with SR 103, Basingstoke, UK) after incubation at 25 °C for 2 days (Mead & Adams, 1977). For members of the Enterobacteriaceae family, 1.0 ml sample was inoculated into 10 ml of molten (45 °C) violet red bile glucose agar (Oxoid code CM1082, Basingstoke, UK). After setting, 10 ml overlay of molten medium were added and incubated at 30 °C for 24 h. The large colonies with purple haloes were counted (Mossel, Eelderink, Koopmans, & Rossen, 1979). Lactic acid bacteria were determined on de Man Rogosa Sharpe medium (Oxoid code CM1153, Basingstoke, UK) after incubation at 35 °C for 3 days. All plates were examined visually for typical colony types and morphological characteristics associated with each growth medium. In addition, the selectivity of each medium was checked routinely by Gram staining and microscopic examination of smears prepared from randomly selected colonies from all of the media. Testing for the presence of *Clostridium* spp. in samples containing the O<sub>2</sub> absorber was carried out using meat broth for enrichment at 30 °C for 7 days. Clostridia were enumerated using Reinforced Clostridium Medium (RCM, Merck code 1.05410, Darmstadt, Germany) after incubation at 35 °C for 2 days under anaerobic conditions. The anaerobic conditions were attained by the use of Anaeropack® GENbox Jars combined with Pack-Anaero oxygen absorbers (Mitsubishi Gas Chemical Company, Tokyo, Japan).

#### 2.3. Sensory evaluation

The samples were evaluated, by a 51-member panel (acceptability test). Panelists (28 females, 23 males) were chosen among graduate students and faculty of the Department of Chemistry, University of Ioannina using the following criteria: ages between 18 and 60, non-smokers, who consume chicken products regularly.

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