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Effect of pomegranate juice dipping and chitosan coating enriched with *Zataria multiflora* Boiss essential oil on the shelf-life of chicken meat during refrigerated storage

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ABSTRACT

The objective of present study was to investigate the effect of pomegranate juice (PJ) and chitosan (CH) coating enriched with *Zataria multiflora* essential oil (ZEO) on the shelf-life of chicken breast meat during refrigerated storage. Treatments examined were the following: Control, PJ, PJ–CH, PJ–CH–Z 1%, and PJ–CH–Z 2%. The samples were stored at 4 °C for 20 days and analyzed at 5-day intervals. All of treatments significantly decreased total viable counts, *Pseudomonas* spp., lactic acid bacteria, *Enterobacteriaceae*, Psychrotrophic bacteria and yeasts–molds as compared control during the storage period. Peroxide value, Thiobarbituric acid reactive substance values and protein oxidation significantly were lower in all of treatments than control. PJ gave a pleasant effect on sensory attributes and chitosan coating enriched with ZEO significantly improved sensory scores. In conclusion, PJ can be suggested as a replacement to synthetic preservatives as well as synthetic flavorings in chicken breast meat.

Industrial relevance: Chicken meat is susceptible to rapid spoilage due to high level protein and moisture. Then, food industries are recently finding methods to extend its shelf-life. The various chemical preservatives are generally undesired by consumers because of their adverse effects. Therefore, natural additives such as pomegranate juice, not only give appropriate color and flavor to foods but also they can extend the shelf-life of foods. The objective of this study was to introduce a new and palatable product resulting from dipping chicken breast meat in pomegranate fruit juice and its preservation with chitosan coating enriched with *Z. multiflora* Boiss essential oil under refrigerated storage.

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1. Introduction

The fresh meat is very sensitive to spoilage by microbial growth and oxidative reactions. High level protein and moisture cause microbial spoilage of meat while the aerobic condition induces oxidation of lipid and protein. Decreasing microbial growth and delaying lipid and protein oxidation during storage can increase the shelf-life of meat (Vaithiyanathan, Naveena, Muthukumar, Girish, & Kondaiah, 2011). Nowadays, synthetic preservatives are being applied to prevent the microbial growth and as well as retarding the oxidation reactions in meat. The consumers are unsatisfied from various chemical preservatives because of their side effects such as carcinogenicity and teratogenicity. The excessive demand for natural preservatives results in their extended utility (Giatrakou & Savvaidis, 2012).

Acidification using organic acids and natural acidic fruit juices such as pomegranate juice is an extensively used method in food processing to extend the shelf-life (Sengun & Karapinar, 2004; Vijayakumar & Wolf-Hall, 2002). Pomegranate (*Punica granatum* L.) from the Punicaceae family is an important commercial fruit crop that is excessively cultivated in Asia, North Africa, the Mediterranean and the Middle East (Sarkhosh, Zamani, Fatahi, & Ebadi, 2006). Pomegranate juice products are one of the most popular flavorings used to give flavor to several foods such as chicken, fish, salads and appetizers, in Iran and Turkey (Karabiyikli & Kisla, 2012). Pleasant flavor of pomegranate juice results from the combination of various taste, aroma and mouthfeel sensations. The distinguished taste is due to mainly the presence of sugars (glucose and fructose) and organic acids (primarily citric and malic acids). The aroma contains volatile compounds, including alcohols, aldehydes, ketones, and terpenes (Vázquez-Araújo, Koppel, Chambers Iv, Adhikari, & Carbonell-Barrachina, 2011). Recently, the great antioxidant potency from different components of pomegranate fruit such as juice, peel and seeds have been discovered (Gil, Tomás-Barberán, Hess-Pierce, Holcroft, & Kader, 2000). The antioxidant activity of pomegranate juice is higher than other fruit juices and beverages (Seeram et al., 2008). This antioxidant activity is correlated to the high level of phenolic compounds, including anthocyanins (3-glucosides and 3,5-diglucosides of delphinidin, cyanidin, and pelargonidin),

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ellagic acid, punicalin, punicalagin, pedunculagin and various flavanols (Alighourchi, Barzegar, & Abbasi, 2008; Miguel, Fontes, Antunes, Neves, & Martins, 2004). Pomegranate anthocyanins are sensitive compounds to oxidation that will be degraded by enormous destructive reactions during storage and processing (Wrolstad, Durst, & Lee, 2005).

Chitosan, a high molecular weight cationic polysaccharide, produced by the deacetylation of chitin, is widely used in phenolic preservation of some fruits, such as pomegranate, litchi (De Reuck, Sivakumar, & Korsten, 2009) and raspberries (Zhang & Quantick, 1998) because of its excellent film forming and anti-fungal, bio-safe and biochemical properties (Lin & Chao, 2001). Recently, these properties of chitosan have been improved by incorporation of essential oils into chitosan films or coatings. Incorporation of essential oils into chitosan films or coatings can not only increase the film's antimicrobial and antioxidant properties but also decrease water vapor permeability and retard lipid oxidation of the product on which the film is used (Giatrakou, Ntzimani, & Savvaidis, 2010; Ojagh, Rezaei, Razavi, & Hosseini, 2010).

Zataria multiflora Boiss (Persian name: Avishane shirazi) belongs to the family of Lamiaceae, and is a medicinal plant that grows extensively in tropical regions of Iran, Pakistan and Afghanistan. The essential oil of this plant (ZEO) consists great quantities of phenolic oxygenated monoterpens that cause antioxidant, antibacterial and antifungal activities (Aliakbarlu, Sadaghiani, & Mohammadi, 2013; Saei-Dehkordi, Tajik, Moradi, & Khalighi-Sigaroodi, 2010). The use of essential oils for food preservation is often restricted because of their application costs and other defects, such as their vigorous aroma and potential toxicity. An interesting alternative to reduce the amounts of essential oils while maintaining their effectiveness could be to incorporate these compounds into the formulation of edible coatings (Petrou, Tsiraki, Giatrakou, & Savvaidis, 2012). Therefore, the objective of this work was to introduce the new product resulting from dipping chicken breast meat in pomegranate fruit juice and its preservation with chitosan coating enriched with *Z. multiflora* Boiss essential oil under refrigerated storage (4 °C).

2. Materials and methods

2.1. Chicken samples

Skinless and boneless fresh chicken breast meat (ca. 400 g) was purchased from a local poultry processing plant 1 h after slaughter and immediately transferred to the laboratory of Department of Food Hygiene and Quality Control, Urmia University, in insulated polystyrene boxes on ice flakes. The chicken meat samples were subsequently stored at 4 °C until use.

2.2. Preparation of pomegranate fruit juice (PJ)

Fresh pomegranate fruits (*P. granatum* L. cv. Rabbab-e Neyriz) were obtained from a local super market. The fruits were washed and cut into four pieces. The seeds/arils were manually separated and ground in a mixer for 30 s and then passed through muslin cloth. After filtering by a Millipore filter with a 0.22 μm nylon membrane under vacuum at 25 °C, the freshly prepared juice was used for treatments.

2.3. Extraction and analysis of ZEO

The plant, *Z. multiflora* Boiss, was purchased from the local groceries in Shiraz and identified at the Institute of Medicinal Plants, Karaj, Iran. The dried aerial parts were subjected to hydrodistillation for 3 h using a Clevenger-type apparatus. The extracted oil was dried over anhydrous sodium sulfate, followed by filtering and stored in airtight glass vials covered with aluminum foil at 4 °C. Gas Chromatography–Mass Spectroscopy (GC–MS) analysis of ZEO was performed according to conditions reported in the previous work of authors (Bazargani-Gilani, Tajik, & Aliakbarlu, 2014).

2.4. Preparation of coating solutions and treatment of chicken meat

Chitosan (medium molecular weight, Sigma-Aldrich Chemical Co.) solution was prepared with 1.5% (w/v) chitosan in 1% (v/v) acetic acid. To achieve complete dispersion of chitosan, the solution was stirred at room temperature for overnight. The solution in beakers was placed on a hotplate/magnetic stirrer and glycerol was added to chitosan at 0.75 mL/g concentration as a plasticizer and stirred for 10 min. The resultant chitosan coating solution was filtrated through a Whatman No. 3 filter paper to remove any undissolved particles. Then the ZEO, mixed with Tween 80 (Aldrich Chemical Co., Steinheim, Germany), was added to the chitosan solution (Ojagh et al., 2010; Petrou et al., 2012). The final coating solution consisted of 1.5% chitosan, 1% acetic acid, 0.75% glycerol, 0.2% Tween 80 singly or incorporated with 1% and 2% ZEO. Meat samples were randomly assigned into five groups including the control (sterile distilled water) and four groups treated with the following solution: PJ, PJ–CH, PJ–CH–ZEO 1% and PJ–CH–ZEO 2%. Each sample was immersed for 2 min in dipping solution (sterile distilled water and PJ) and then drained well and followed by a second immersion in the coating solution for 2 min (Ojagh et al., 2010; Vaithiyanathan et al., 2011). Then the meat samples were removed and allowed to drain for 5 h at 10 °C on a pre-sterilized metal net under a biological containment hood in order to form the edible coatings and then the treated samples were aerobically packaged in a low density polyethylene pouches and stored at 4 °C for subsequent quality assessments. Chemical, microbiological and sensorial analyses were performed at 5-day intervals to determine the overall quality of samples for 20 days.

2.5. Microbiological analysis

Chicken meat (10 g) was mixed with 90 mL of 0.1% sterile peptone water (Merck, Darmstadt, Germany) in a sterile stomacher bag and stomached for 1 min. For microbial enumeration, 0.1 mL of serial dilutions of chicken homogenates was spread on the surface of agar plates. Total viable counts (TVC) were determined using Plate Count Agar (PCA, Merck, Darmstadt, Germany), after incubation for 2 days at 30 °C. *Pseudomonas* spp. were enumerated on cetrimide fusidin cephaloridine agar (CFC, Fluka, Germany) and incubated at 20 °C for 2 days. Lactic acid bacteria (LAB) were counted on de Man Rogosa Sharpe agar (MRS, QUELAB, Canada) incubated at 35 °C for 2 days. *Enterobacteriaceae* were enumerated by the pour-overlay method using Violet Red Bile Glucose (VRBG) agar (Merck, Darmstadt, Germany). The plates were incubated at 37 °C for 24 h. Psychrotrophic bacteria were determined on Plate Count Agar and the plates were incubated at 7 °C for 10 days. Finally, yeasts–molds were enumerated on Rose Bengal Chloramphenicol (RBC) selective agar (Merck, Darmstadt, Germany) plates using surface spreading technique and plates were incubated at 25 °C for 3–5 days in the dark. Three replicates of at least three appropriate dilutions depending on the sampling day were enumerated. Microbiological data were transformed into logarithms of the number of colony forming units (cfu/g).

2.6. Physicochemical analysis

2.6.1. Determination of pH value

The pH value was recorded using a pH meter (PH-Meter E520, Metrohm Herisau, Switzerland). Meat sample (5 g) was homogenized with 25 mL of distilled water for 30 s and the homogenate was used for pH determination.

2.6.2. Determination of peroxide value

Peroxide value (PV) was measured as suggested by the International Dairy Federation (IDF) (Shantha & Decker, 1993). The sample (0.30 g) was mixed with 9.8 mL chloroform–methanol in a glass tube and vortexed for 2–4 s. Ammonium thiocyanate solution (10 mM)

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