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Biopolymer-based films as carriers of antimicrobial agents

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Abstract

Films were prepared by incorporating different levels of antibacterial agents such as oregano oil, sodium lactate (NaL) and ε-polylysine (ε-PL) into sorbitol-plasticized whey protein isolate (WPI) films. The moisture uptake behaviour and the water vapour permeability (WVP) of the films were only affected by NaL, as the water sorption and permeability increased with addition of NaL into the protein matrix. An increase of the glass transition temperature of the sorbitol regions, as assessed by Dynamic Mechanical Thermal Analysis (DMTA), was caused by the addition of ε-PL, while incorporation of the oregano oil caused plasticization of the film that was depicted by a decrease in the transition temperature of the polymer-rich regions. On the other hand, incorporation of NaL into the films did not significantly alter their thermo-mechanical properties. However, the addition of NaL or ε-PL in the film forming solution resulted in a decline of maximum tensile strength (σ_{max}). Wrapping of beef cuts with the antimicrobial films resulted in a significant reduction of the bacterial population levels. The maximum specific growth rate (μ_{max}) of total flora (Total Viable Count, TVC) and pseudomonads was significantly reduced ($P<0.05$), with the use of antimicrobial films containing relatively high levels of oregano oil (1.5 % w/w in the film forming solution) or ε-PL (0.75 % w/w in the film forming solution), while the growth of Lactic Acid Bacteria (LAB) was completely inhibited.

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Keywords: Edible films; Beef; Antimicrobial; Thermal properties; Mechanical properties; Whey proteins; Active packaging

1. Introduction

Active packaging remains an on-going development in food preservation technologies aiming at controlling of food spoilage and the production of high quality products with improved sensorial attributes. Apart from acting as selective barriers for moisture exchange, gas and solute migration, edible films may operate as carriers of many functional ingredients such as antioxidants, antimicrobial agents,

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flavours and spices [1]. When applied, antimicrobial packaging materials can effectively control the microbial contamination of solid or semi-solid food products by inhibiting the growth of spoilage or pathogenic microorganisms on the surface of the food item. Incorporation of antimicrobial compounds into the films results in decreased diffusion rates from the packaging material into the product, compared to simple spraying of the antimicrobial agents on top of the food system, thus assisting the maintenance of relatively high concentrations of the active ingredient where it is required [2].

Essential oils (EOs) extracted from plants or spices are rich sources of biologically active compounds such as terpenoids and phenolic acids. It has been long recognised that a lot of the EOs have antimicrobial properties and oregano oil that contains large amounts of carvacrol is considered to be one of the most active plant extracts against pathogens [3]. Lactate salts such as sodium lactate (NaL) are widely used as flavor enhancers in meat and poultry products, contributing to increased cooking yields and water holding capacity of processed meats [4]. Additionally, various lactate salts have demonstrated antimicrobial activity in laboratory media or food products [5]. E-polylysine (ε-PL) is a cationic homopolymer of 25 to 35 L-lysine units interlinked by a peptide bond between the carboxyl and the ε-amino groups of the L-lysine residues. This compound is heat stable even under acidic conditions and exhibits a wide antimicrobial activity against Gram(+) and Gram(-) bacteria, yeasts and moulds [6].

In contrast to the large amount of information on the use of various antimicrobial films for controlling meat pathogens, little is known about their effect on spoilage flora of these products. In the present study, fresh beef cuts were wrapped into whey protein isolate (WPI) films containing different antimicrobial agents (oregano oil, NaL and ε-PL). The effectiveness of the films against the beef's spoilage flora during storage at 5°C was investigated. Additionally, the impact of the antimicrobial agents on the mechanical and physical properties of the films was examined since the overall performance of the films depends strongly on their physicochemical properties.

2. Materials & Methods

Bipro, a whey protein isolate (Davisco Foods International), was dissolved to obtain film-forming solutions of 5% (w/w) concentration. Protein solutions were placed in a water bath at 90ºC for 30 min under stirring. Sorbitol (St. Louis, MO, USA) was added as a plasticizer in the constant concentration of 37.5% (sorbitol/(WPI+sorbitol)). Appropriate amounts of the antimicrobials were added in the film forming solution resulting in a final concentration of 1.0%, 1.5% and 2.0% (w/w) for NaL (50% solution, Merck KGaA, Germany), 0.25%, 0.5% and 0.75% (w/w) for ε-PL (Chisso Int, Japan) and 0.5%, 1.0% and 1.5% (w/w) for oregano oil (*Origanum vulgare* ssp. *Hirtum*, Ecopharm, Greece); the latter were homogenized at room temperature for 2 min at 13000 rpm and 2 min at 19000 rpm using an Ultra-Turrax homogenizer (T-25 basic, IKA, Werbe). Portions of 12.5 g solution were cast on Petri dishes (φ 8.5 cm) and allowed to dry in an oven at 35°C for ~24 h. Moisture sorption isotherms were conducted for all the samples according to [7] and WVP measurements were performed at 25°C using the ASTM (E96-63T) procedure modified for the vapor pressure at film underside [8]. The thermo-mechanical properties of the films were determined by DMTA performed with a Mark III analyzer (Polymer Labs. Loughborough, UK) operated in the single cantilever bending mode (heating rate 2 $^{\circ}$C min^{-1} and a strain level equal to a maximum displacement of 16 μm). The T_g of the samples was determined as the peak in tan δ at 3 Hz. The mechanical properties of the films were evaluated by large deformation mechanical testing using a TA-XT2i instrument (Stable Micro systems, Godalming, Surrey, UK) [9]. In order to determine the antimicrobial properties of the films beef cuts were wrapped with antimicrobial films and stored at 5°C. Throughout storage samples were analyzed and the populations of Total Viable Counts, pseudomonads and Lactic Acid Bacteria were measured. Microbial growth data were modeled as a function of time with the model of Baranyi using the in-house software Dmfit [10].