ABSTRACT

This PhD dissertation thesis has been focus on the development and characterization of antimicrobial packaging films based on the incorporation in the polymer matrix or on the attachment to the film surface of naturally occurring antimicrobial compounds with the purpose of inhibiting the proliferation of microorganisms and extend the microbiological shelf life of packaged food products.

The studied active films are based on the use of ethylene vinyl copolymers (EVOH) containing 29% (EVOH29) or 44% (EVOH44) molar percentage of ethylene as polymeric vehicle for the incorporation of several antimicrobial compounds -oregano essential oil (OEO), citral, ethyl lauroyl arginate (LAE), epsilon-polylysine (EPL), green tea extract (GTE) and lysozyme. These antimicrobial agents have been incorporated in the film-forming solution or immobilized to the film surface by covalent bonding.

Prior to the preparation of the active films, the antimicrobial activity of the selected compounds against selected microorganism was demonstrated, confirming that they could be good candidates to be used as preservatives for active food packaging applications, and an alternative to synthetic additives. The effect of the incorporation of the antimicrobial agents on relevant functional properties of the developed EVOH films was studied. In general, the polymer properties as materials for food packaging were not relevantly affected.

In order to evaluate the potential of EVOH matrices as sustain release systems of active compounds, the release kinetics of the active compounds from the film to different media was evaluated; for that the agent release rate and extend into food simulants was monitored, and it was concluded that the agent concentration, release temperature, type of EVOH, interaction of EVOH with the food simulant, and the solubility of the active compound in the release media were the main controlling factors.

EVOH matrices have also shown good properties to be used for the attachment of active molecules. In this regard, lysozyme was successfully immobilized on the film surface of EVOH.

Several experiments were conducted to determine the antimicrobial properties of the resulting films *in vitro* against different microorganisms responsible for foodborne illness and *in vivo* with real foods –minimally-process salad, infant milk, surimi sticks and chicken stock- to enhance their preservation. All the materials presented a strong *in vitro* antimicrobial activity. Although the results obtained through *in vivo* tests showed activity reductions caused by food matrix effects, all materials presented significant microbial inhibition and, therefore, great potential to be used in the design of active food packaging. They can be applied as an inner coating of the packaging structure, releasing the active agent or acting by direct contact, producing a great protection against contamination with a prolongation of the microbiological food shelf life.