FILMS OF POLYETHYLENE-AgNPs: STATE-OF-THE-ART, ECOTOXICOLOGICAL ASPECTS AND ANTIMICROBIAL EFFICACY

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Abstract - In recent years, antimicrobial packaging has attracted much attention from the food industry because of the increase in consumer demand for minimally processed, preservative-free-products. Different grades of polyethylene, LLDPE and LDPE play a major role in the film blowing industry and are widely used for packing applications. In our study, blown films of LLDPE / LDPE, and their blends were produced using a twin screw extruder. The combination of PEs as matrix, and silver nanoparticles with oleic acid solution, improved the AgNPs dispersion in the films. The films were evaluated by Scanning Electron Microscopy (SEM), Energy Dispersive Spectroscopy (EDX), and Ecotoxicity. Further, the antibacterial properties of the films were investigated against *Escherichia coli* (Gram-negative) and *Staphylococcus aureus* (Gram-positive) bacteria.
Keywords: *polyethylene, silver nanoparticles, oleic acid, blown films*

Introduction

Polyethylene (PE) has been one of the most widely used polyolefin polymers mainly because of its beneficial properties in low cost and weight, low-temperature toughness, good optical properties, low moisture absorption, strong heat sealing and ease of processing and recycling [1]. PE represents the most popular plastic globally, with annual production projected to total nearly 5 million tonnes in 2017 due to its easy and cheap manufacturing processes, and also represents a versatile biomaterial with significant clinical impact. PE is categorized by its density and branching and as such, several types of PE exist ranging from ultra-high-molecular-weight to medium- and low density PE, each with varying thermal, mechanical, chemical, electrical and optical properties relevant for biological mimicry [2]. Antimicrobial materials play an important role in the academic and technological fields due to applications in food packaging and medicinal devices such as catheters, cannulae, and others. Improvements are achieved at low nanoparticle concentrations (1–10 wt.%), in strong contrast with conventionally filled polymers, which generally require high loads in the range of 25–40 wt.%. In this context the nanocomposites retain the low density of the matrix and are more easily processed [3]. The use of silver in commercial products has proliferated in recent years owing to its antibacterial properties. Food containers impregnated with micro-sized silver promise long food life, but there is some concern because silver can leach out of the plastic and into the stored food. It is important to investigate the release because silver demonstrates neurotoxicity properties in humans, as well as the potential for ecotoxicity this metal[4]. The efficacy and safety of commercial silver-treated food storage containers, for example, are not well characterized regarding the potential life cycle implications of their design and application. There are studies to determine the antimicrobial efficacy and quantify silver leaching from a commercial container product containing micronized silver particles over simulated washing and end-of-life landfill disposal [5]. Particularly AgNPs have received in the last decades a growing interest and have found commercial applications. However, many problems persist in their safe use, as very little is still known about the metabolism, clearance and toxicity of nanoparticles [6].The aim of this research is to verify the biocidal activity of the silver nanoparticles and also to elaborate an ecotoxicological study of these nano-silver-particles with the environment.

Experimental

Materials and Methods

Materials

LDPE with a melt flow index – MFI (190/2.16) of 0.27 dg min^{-1} and density: 0.922 g mL^{-1}, LLDPE with a melt flow index – MFI (190/2.16) of 0.80 dg min^{-1} and density: 0.920 g mL^{-1}, Braskem (Brazil) was provided in the form of pellets. The silver nitrate ($AgNO_3$) and oleic acid (AO) were supplied by Labsynth, which acted as a surfactant for the $AgNO_3$. Antioxidant (BASF Irganox B225ED) was added in small quantities, 1.0 wt%, to prevent the polyethylene from oxidizing and thermally cross-linking at elevated temperatures.

Preparation of the Nanocomposites

The LDPE/LLDPE (90/10) pellet were mixed with Irganox B215ED in a rotary mixer and maintained under this condition for 24 hours. Then the mixture was processed with the addition of silver nitrate solution ($AgNO_3$ = 5000ppm) in a twin-screw extruder Haake co-rotating, model Rheomex PTW 16/25, with the following processing conditions: the temperature profile (feed to die) was 145-170 °C, with a speed of 100 rpm. After processed, the nanocomposites were granulated in a granulator Primotécnica W-702-3. The PE@AgNPs film was produced in planar sheet extruder and the material was placed directly into the hopper of the extruder with a temperature profile (feed to die) of 150-175 ºC, screw speed of 50 rpm and torque of 33-45 Nm. The films were produced with a thickness of ~ 0.05 mm.

Methods

Scanning electron microscopy and Energy dispersive spectroscopy

Specimens were examined with a Hitachi TM 3000, coupled with a Bruker Quantax 70 for the collection of EDS data. SEM coupled with backscattered electron detector (BSE) and energy dispersive X-ray spectroscopy (EDS). Sample sections for the EDS analysis were taken at 15 keV, and the acquisition period was 120 s.

Percentage reduction of colony forming units

Microbiological assays were performed following the JIS Z 2801 [7] standard, the bacteria used were *E.coli* ATCC 8739 and *S. Aureus* ATCC6538. The cell suspension for the inoculum was 900 x 10^6 mL^{-1} CFU for each tested step. The following procedure was performed separately for each microorganism: samples of the films of PE@AgNPs were placed in a sterile Petri dish and inoculated on the surface of 50 mL of suspension of each organism in an area of 40 x 40 mm^2. All of them were incubated for 24h at 37ºC.

Toxicity tests with sea urchin *Echinometra Lucunter*

The toxicity tests for evaluation of chronic effects on the embryological development of *Echinometra Lucunter* sea urchin were carried out according to the procedures described by USEPA [7], with adaptations to the species by NBR 15350 [8], regarding the exposure time and temperature (ABNT).

Results and Discussion

A magnification of SEM and EDX for PE@AgNPs is shown in Fig.1.

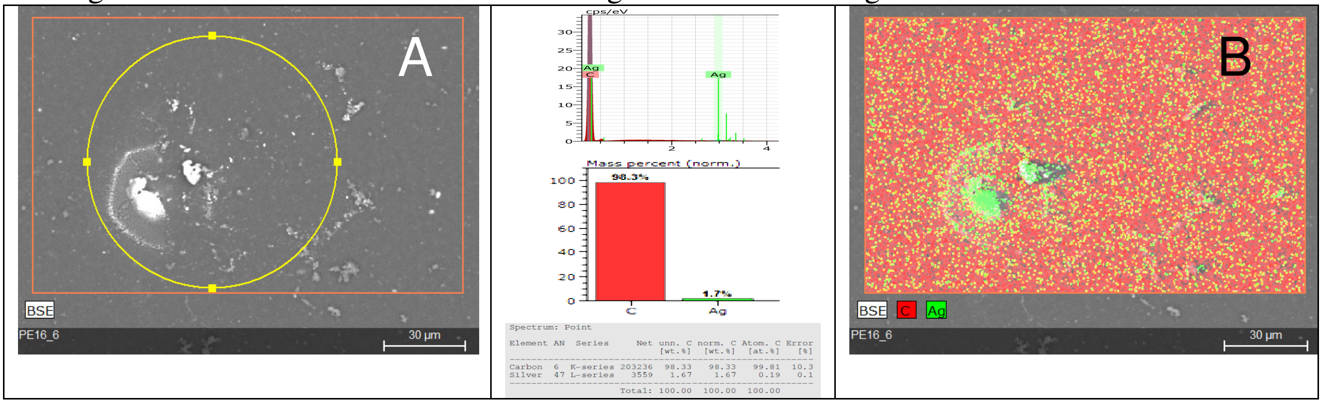

Figure 1. Magnification of SEM image (A) and EDX spectrum of a silver additive conglomerate on the surface of PE@AgNPs composite. The surface of the nanocomposite film showing the distribution of carbon (red) and silver (green) atoms (B).

Representative PE@AgNPs composites have been investigated by SEM spectroscopy. The EDX analyses confirmed, by a semi-qualitative-analysis, the chemical composition of composites and also determined the level of additive microscopic dispersion in the samples analyzed. High resolution SEM images and EDX spectra show that the silver-containing aggregates are as a matter of fact randomly distributed and well isolated conglomerates on the surface. The evidence of the elemental analysis suggests that the nanofillers were well dispersed into the bulk of PE matrix and have dimensions with approximately 50nm (AgNPs) and in some points have agglomerates with dimension of 10µm.

The results show that good dispersion and antimicrobial properties were obtained with AO as a surfactant against *S. aureus* and *E.coli*. The addition of coated $AgNO_3$ to the PE matrix represented an interesting solution for increasing protection against *S. aureus* and *E.coli*. The percentage reduction for the CFU assay showed positive 100% biocidal results for *S. aureus* and *E.coli* in film PE@AgNPs.

The sea-water toxicity tests with embryos of a sea urchin species for PE@AgNPs nano-silver polyethylene film showed no toxic effects, ie, embryos developed normally and had similar control results (water uncontaminated sea).

Conclusions

The addition of coated $AgNO_3$ to a PE matrix during the extrusion process represents an interesting solution for increasing the protection against *S. aureus* and *E. coli.* Silver in PE film properly hindered the bacterial activity in the bulk, furthermore, with the use of AO surfactant, indicates a better dispersion of AgNPs in the PE film and biocidal properties. Another important factor was the absence of contamination of the nano-silver to the environment and therefore non-toxic.

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