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Antioxidant packaging with encapsulated green tea for fresh minced meat

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ABSTRACT

A novel approach to incorporate green tea extract into polyethylene by extrusion technology has been studied. Green tea entrapped by inorganic capsules was incorporated into melted material and extruded without compromising its quality. The shelf-life of fresh meat from two types of active packaging was significantly extended for 3 days compared to blank samples. Significant difference was observed after 9th day of study in case of results of CIE L*a*b*, MetMb and organoleptic assay. Migration study of antioxidants from the materials was also performed by UHPLC-MS/TQ using food simulants. Amount of migrated catechins was in the range between 6.3 ± 3.3 and 228.4 ± 15.2 μg/kg (ratio 6:1). Regression coefficients (R) between 0.9925 and 0.9989 were obtained. Minimum LOD (0.001 μg/g) and LOQ (0.004 μg/g) were obtained for epicatechin gallate and catechin gallate while maximum LOD (0.057 μg/g) and LOQ (0.191 μg/g) were obtained for catechin.

1. Introduction

The development of new films for extending the shelf-life of fresh food products is important for the food industry as packaging plays an essential role in limiting the deterioration of food (Prasad & Kochhar, 2014). Recently a lot of attention is given to active packaging (Soares, Silva, Medeiros, Carelli, & Espitia, 2011). Therefore, different active packaging technologies have been developed and applied to decrease meat decay (Contini et al., 2014; Lorenzo, Batlle, & Gómez, 2014; Muppalla, Kanatt, Chawla, & Sharma, 2014; Nerin et al., 2006) and also to limit environmental pollution connected with packaging (McMillin & Belcher, 2012). Although the great interest and the amount of scientific publications dealing with active packaging, there are only a few packaging materials commercially available. This lack of commercial materials can be attributed to either the difficulties in producing the active materials at industrial scale, the low efficiency of most of the developed materials in in vivo tests and the degradation of active agents in extrusion processes.

One of the promising multi-component technologies that can be applied to protect the active ingredients during the packaging production is encapsulation. Encapsulation involves the isolation of a compound from its external environment by entrapping it into an additionally material, which has a protective function. This way the active compounds can be incorporated into melted material and extruded without compromising its quality. A lot of work about encapsulation directly applied into food products, processing and production can be already found in the scientific literature (Gibbs, Kermasha, Alli, & Mulligan, 1999; Nedovic, Kalusevic, Manojlovic, Levic, & Bugarski, 2011; Zuidam & Nedović, 2010). However, the use of active capsules in polymers for extending the shelf-life of food has not been extensively used to date (Augustin & Sanguansri, 2010; Haidong, Fang, Zhihong, & Changle, 2011). The most popular materials used for encapsulation are cyclodextrins that let encapsulate compounds quickly and efficiently (Fuenmayor et al., 2013; Haidong et al., 2011; Silva, Figueiras, Gallardo, Nerin, & Domingues, 2014; Sun et al., 2014). However, inorganic capsules consisting of crystalline microporous aluminosilicates, as those used in this study, can be perfect carriers because of the micropores present on the shell, which facilitate the incorporation and protection of the active agents. Moreover, inorganic capsules are more thermo-resistant and the antioxidant performance could be observed in high humidity environment (Wei et al., 2013; Yu & Xu, 2010).

Natural antioxidants such as alpha-tocopherol (Noronha et al., 2013), eugenol (Woranuch & Yoksan, 2013), thymol and cinnamaldehyde (Ponce Cevallos, Buera, & Elizalde, 2010) have been successfully encapsulated with further possibility of incorporation into plastic films. Catechins are powerful antioxidants present in cocoa, green tea, grapes and green algae (Arts, van De Putte, & Hollman, 2000; Haidong et al., 2011; Ioannone et al., 2015; Rusak, Komes, Likić, Horžić, & Kovač, 2008) among others. They include compounds such as (+)-catechin, (−)-epicatechin, (−)-gallocatechin, (−)-epigallocatechin, (−)-epigallocatechin gallate, (−)-epicatechin gallate, (−)-catechin gallate and

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(−)-gallocatechin gallate. They are efficient free radical scavengers due to the presence of phenolic hydroxyl groups (Colon & Nerin, 2012). As has been previously demonstrated (Carrizo, Gullo, Bosetti, & Nerin, 2014; Carrizo, Taborda, Nerín, & Bosetti, 2016; Nerin, 2010) when scavenging free radicals the release of antioxidants is not required, as they can be active and efficient antioxidants without direct contact with the food (Carrizo et al., 2016; Nerin, 2010). Two mechanisms explain the antioxidant capacity (CAOX) of catechins: hydrogen-atom transfer and single electron transfer (Quideau, Deffieux, Douat-Casassus, & Pouysegu, 2011). In both systems direct contact with the food is not required and thus the use of catechins and other compounds with similar behaviour is a non-migrating performance. Although the chemical activity and antioxidant capacity of individual catechins are not equal, antioxidant synergy of catechins mix was scientifically proved (Sanna et al., 2015). This way, the use of a natural mixture such as green tea, is better option than applying any single catechin.

The stability of green tea during processing and storage can be significantly reduced as they can easily undergo oxidation and thermal decomposition (Ananingsih, Sharma, & Zhou, 2013; Li, Taylor, & Mauer, 2011). Therefore, protection in a capsule would be highly desirable to fully utilize their potential benefits. Several systems based on encapsulation of green tea have been proposed in the literature (Gadkari & Balaraman, 2015) and the properties of catechins as free radical scavengers have been already applied in active packaging to extend the shelf-life of storage products (Carrizo et al., 2014; Carrizo et al., 2016; Colon & Nerin, 2014; Kaewprachu, Osako, Benjakul, & Rawdkuen, 2015; López de Dicastillo, Castro-López, López-Vilariño, & González-Rodríguez, 2013; Wrona, Bentayeb, & Nerín, 2015).

Without a doubt, the quality of fresh minced meat is crucial for consumer's acceptance and one of the most visible decay is the fade of red colour of the meat. That turns brown by oxidation. Deterioration of minced meat is caused by protein oxidation characterised by the red colour loss and lipid oxidation characterised by off-flavours and off-odours formation (Kaewprachu et al., 2015; Nerin et al., 2006). Therefore, the application of active packaging based on natural antioxidants working as free radical scavengers can be a good strategy for improving the quality and freshness of minced meat.

The aim of this work was to develop a new active packaging made by extrusion with encapsulated green tea as free radical scavenger, for extending the shelf-life of fresh meat without compromising its quality. Moreover, the proposed packaging was based on natural active agent rich in catechins that was protected by encapsulation, incorporated into melted polyethylene and extruded as a film, which was further used for packaging fresh minced meat. A pilot plant was used to prepare the new active film. Thanks to the encapsulation step the stabilization of green tea extract and preservation of its antioxidant property were ensured. In vivo test with minced fresh meat demonstrated the feasibility of the new material.

2. Materials and methods

2.1. Reagents

Trolox (6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid; 98%, CAS 53188-07-1), fluorescein (3,6-dihydroxypiro[isobenzofuran-1[3H],9[9H]-xanthen]-3-one; Standard Fluka, CAS 518-47-8), AAPH (2,2′-azobis(2-methyl-propionamidine)dihydrochloride; 97%, CAS 2997-92-4), DPPH (α,α–diphenylpicrylhydrazyl radical, CAS 1898-664), caffeine (CAS 58-08-2) (CAF), (+)-catechin (> 99.0% (HPLC), CAS 154-23-4) (C), (−)-epicatechin (> 95% (HPLC), CAS 490-46-0) (EC), (−)-epicatechin gallate (> 98% (HPLC), CAS 1257-08-5) (ECG), (−)-catechin gallate (> 98% (HPLC), CAS 130405-40-2) (CG), (−)-epigallocatechin (> 95% (HPLC), CAS 970-74-1) (EGC), (−)-gallocatechin (> 98% (HPLC), CAS 3371-27-5) (GC), (−)-gallocatechin gallate (> 98% (HPLC), CAS 4233-96-9) (GCG) and (−)-epigallocatechin gallate (> 95% (HPLC), CAS 989-51-5) (ECG) were from Sigma-Aldrich (Madrid, Spain). Disodium hydrogen phosphate dihydrate (99.5%, CAS 10028-24-7), and sodium dihydrogen phosphate monohydrate (99%, CAS 7558-80-7) were purchased from Merck (Madrid, Spain). Methanol (LC-MS, CAS 67-56-1), ethanol absolute (GC–MS, CAS 64-27-5) and acetone (HPLC grade, CAS 67-64-1) were from Sharlau (Barcelona, Spain). The ultrapure water was obtained from a Millipore Milli-Q_{PLUS} 185 system (Madrid, Spain).

2.2. Apparatus

Ultra-Turrax T18 from IKA (Staufen, Germany) was used to homogenize the meat samples. Ultrasonic bath Branson 3510 (Branson Ultrasonic Corporation, USA) and centrifuge CENTROMIX model S-549 were also utilized.

Nikon Coolpix 4300 camera (Tokyo, Japan) and Konika Minolta chroma meter C-400 (Tokyo, Japan) were used for colour evaluation.

2.3. Equipment

UV-1700 of Shimadzu Pharmaspec Ibérica (Madrid, Spain) spectrophotometer was used for antioxidant capacity methods.

Non-volatile compounds were analysed by Acquity™ system with use of an Acquity UPLC BEH C18 column of 17-μm particle size (2.1 mm × 100 mm), both from Waters (Milford, MA, USA). MS detector consisted of a hexapole, a quadrupole, a collision cell and time of flight analyser (Xevo G2) from Waters (Milford, MA, USA).

Volatile compounds were analysed by CTC Analytics CombiPal autosampler coupled to an Agilent 6890 N gas chromatograph with an MS mass spectrometer detector. All instruments were from Agilent Technologies (Palo Alto, CA, USA). Chromatographic separations were carried out on a BP-20 (30 m × 0.25 mm × 0.25 μm) from SGE analytical science (Madrid, Spain). DVB/CAR/PDMS fiber from Supelco (Bellefonte, PA, USA) for SPME adsorption was used.

Antioxidants were analysed by Acquity Ultra Performance LC TQ detector (triple quadrupole; Waters). An ESI probe was used in positive (ESI+) and in negative (ESI−) mode as the ionization source. Chromatography was carried out in the Acquity system using an Acquity UPLC BEH C18 column of 1.7 μm particle size (100 mm × 2.1 mm) from Waters. MassLynx (v. 4.1) software (Waters) was used to acquire and process the chromatographic and MS data.

2.4. Samples

Samples of minced pork meat were bought in a local supermarket. Samples were obtained on PP trays closed with a lid of a high barrier plastic. The trays were provided with absorbent pad. All samples were stored in a refrigerator at 4 °C.

2.5. Active packaging

Active polyethylene (PE) films with incorporated capsules loaded with green tea extract were provided by NUREL. Processes of preparation of inorganic capsules and active materials are protected by the patents EP1564242B1 and EP1923423B1. Two different types of capsules developed by the company were incorporated into the polymer structure: polyethylene with type I capsules (PE I) and polyethylene with type II capsules (PE II). PE I consisted of 20% of masterbatch containing active capsules while PE II consisted of 40% of masterbatch containing active capsules. Both materials were prepared by extrusion. These materials with green tea extract at concentrations of 6.4 mg/g (mg of active agent per g of material) in case of PE I and 12.8 mg/g (mg of active agent per g of material) in case of PE II were tested. The same materials but without green tea were used as blank samples. 22 g of minced pork meat were placed in small plastic Petri dish (d = 5 cm). Then meat was covered by PE active film (6 cm × 6 cm) and packed at vacuum bag (7 cm × 9 cm) under normal atmosphere using a thermo-