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Development of new active packaging films coated with natural phenolic compounds to improve the oxidative stability of beef



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ABSTRACT

The aim is to develop active packaging films containing natural antioxidants and to evaluate their capacity to enhance the oxidative stability of beef during refrigeration. The antioxidant activity of a natural extract obtained from a brewery residual waste was evaluated and compared with that of a commercial rosemary extract and two synthetic antioxidants (BHT and propyl gallate). Different concentrations of each antioxidant were also added directly to beef samples, resulting in a reduction in lipid oxidation of up to 70–80% relative to the control. Active antioxidant films coated with PVPP-WS extract reduced lipid oxidation by up to 80%, relative to the control, during cold storage. The use of active packaging films containing natural extracts could improve the oxidative stability of meat products and should therefore be of great interest in the food industry.

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1. Introduction

Lipid peroxidation is a major cause of deterioration of meat quality during processing, distribution and refrigeration, thereby reducing shelf stability and acceptability. Lipid oxidation can produce changes in meat quality parameters, such as organoleptic properties and nutritional value, which leads to the generation and accumulation of compounds that may pose risks to human health (Gray, Gomaa, & Buckley, 1996).

The oxidative stability of meat can be extended by using antioxidants and proper packaging materials (Zhou, Xu, & Liu, 2010).

Synthetic antioxidants have long been used in the food industry to prevent or minimize lipid oxidation in food products. Because of growing concerns about the potential health hazards associated with synthetic antioxidants commonly used in the food industry (e.g. BHT, BHA and PG) and increased consumer demand for natural products, there is a growing interest in the use of naturally occurring antioxidants for use in food processing (Shahidi & Zhong, 2010).

Residual waste substances obtained from agro-industrial byproducts offer a practical and economic source of potent antioxidants that could replace synthetic preservatives (Balasundram, Sundram, & Samman, 2006).

* Corresponding author. *E-mail address:* jmcruz@uvigo.es (J.M. Cruz). Polyvinylpolypyrrolidone washing solution (PVPP-WS) extract is a natural extract obtained from a brewery waste stream. During storage of beer, colloidal haze can develop as a result of the formation of complexes between polypeptides and polyphenols (Siebert, 1999). The negative impact of polyphenols on haze stability is minimized by using polyvinylpolypyrrolidone resin (PVPP) to stabilize beer and extend its shelf life. PVPP stabilization removes a substantial part of the polyphenols in beer (both haze and non-haze active polyphenols), which can be recovered from the PVPP by an alkaline treatment (Mitchell, Hong, May, Wright, & Bamforth, 2005).

The high antioxidant activity of the extract has been demonstrated in different in vitro experiments (Barbosa-Pereira, Angulo, Paseiro-Losada, & Cruz, 2013). The antioxidant activity of PVPP-WS extract is related to the high concentrations of phenolic compounds, such as flavonols (catechin, gallocatechin and epigallocatechin) and hydroxycinnamic and hydroxybenzoic acids (gallic acid, caffeic acid, p-coumaric acid and ferulic acid), which act as free radical acceptors and chain breakers and are therefore responsible for the high free radical scavenging activity of the extract (Barbosa-Pereira, Angulo, et al., 2013; Barbosa-Pereira, Pocheville, Angulo, Paseiro-Losada, & Cruz, 2013).

Natural extracts from many herbs and spices have been studied and used to extend the shelf life of foods (Balasundram et al., 2006; Fernández-Lopez, Zhi, Aleson-Carbonell, Pérez-Alvarez, & Kuri, 2005; Sánchez-Escalante, Djenane, Torrescano, Beltrán, & Roncales, 2003). Rosemary extract has been shown to possess strong antioxidant activity because of the high contents of phenolic diterpenes (e.g. carnosic acid,



carnosol and rosmanol) and phenolic acids (e.g. rosmarinic acid), which act as oxidative chain breakers via electron donation (Zheng & Wang, 2001).

Rosemary extracts have been widely used in the food industry, and many authors have reported their effectiveness in reducing lipid oxidation in meat products. Nevertheless, most studies reported involve the direct addition of rosemary to the packaged food, and relatively few deal with the inclusion of rosemary in the packaging material (McBride, Hogan, & Kerry, 2007; Nerín et al., 2006; Sánchez-Escalante et al., 2003).

Active packaging is currently one of the most dynamic technologies used to preserve the quality of food via the release of active agents from the packaging film. Release of the active agents can be controlled over an extended period of time to maintain or extend the quality and shelf-life of products, without the need for direct addition of any substances to the foodstuff (Lee, 2010; Zhou et al., 2010).

Some studies have evaluated how antioxidants (such as BHT, BHA, alpha-tocopherol and natural extracts) incorporated in packaging film migrate out of the film and retard lipid oxidation in the stored foodstuff (Barbosa-Pereira, Cruz, et al., 2013; Moore et al., 2000). The incorporation of rosemary extract in active packaging film has been described by Nerín et al. (2006), with promising results in relation to extending the shelf life of beef.

Another concept in active packaging is the addition of bioactive substances by a coating process. The development of coatings with antimicrobial capacity has been studied in relation to preservation of meat products (Bonilla, Atarés, Vargas, & Chiralt, 2012; Kerry, O'Grady, & Hogan, 2006; Lee, 2010; Zhou et al., 2010). Antioxidant additives can also be incorporated by coating them onto food packaging materials to control spoilage by oxidation and to preserve food quality (Vermeiren, Dvelieghere, van Beest, de Kruijf, & Debevere, 1999; Lee, An, Lee, Park, & Lee, 2003).

The aim of this study was to evaluate and compare the antioxidant effect of two natural extracts (PVPP-WS and rosemary extracts) and two synthetic antioxidants (BHT and PG) on the oxidative stability of beef during cold storage. The natural extracts were used to coat active films with antioxidant properties. Finally, the effectiveness of the new antioxidant active packaging films in delaying lipid oxidation of beef during cold storage was evaluated in samples in which the packaging was with and without direct contact with the food sample.

2. Materials and methods

2.1. Chemicals

Butylated hydroxytoluene (BHT) (99.0%, CAS No. [128-37-0]), sodium azide (99.0%, CAS No. [26628-22-8]), 2-thiobarbituric acid (TBA) (≥98%, CAS No. [504-17-6]) and trichloroacetic acid (TCA) (puriss. p.a. 99.5%, CAS No. [76-03-9]) were purchased from Sigma-Aldrich (Steinheim, Germany). Propyl gallate (PG) (98%, CAS No. [121-79-9]), 1,1,3,3tetraethoxypropane (TEP) (purum \geq 95% (GC), CAS No. [122-31-6]) and 2, 2-diphenyl-1-picrylhydrazyl (DPPH) (TECHN ≥85%, CAS No. [1898-

Table 1a

Table Ta	
Total and individual phenolic compounds (mg g	⁻¹) in the PVPP-WS extract.

66-4]) were supplied by Fluka Chemie AG (Buchs, Switzerland). Methanol $(GC \ge 99.9\%, CAS No. [67-56-1])$, orthophosphoric acid (85% GR for analysis, CAS No. [7664-38-2]) and ethanol (absolute for analysis, CAS No. [64-17-5]) were provided by Merck (Darmstadt, Germany).

2.2. Natural extracts

2.2.1. PVPP-WS extract

This extract, which contains natural antioxidants, was obtained from a residual stream generated during the PVPP cleaning process in the brewing industry by a process described by Barbosa-Pereira et al. (2014). In beer production, a clarification step is essential to improve beer stability. As a result of this process, a PVPP sludge is obtained. The PVPP sludge loaded with polyphenolic compounds was washed with a NaOH solution (2% w/w) at room temperature. After the NaOH-PVPP was filtered, a clean PVPP resin and a PVPP washing solution (PVPP-WS) containing phenolic compounds were obtained.

The PVPP washing solution (PVPP-WS) was acidified to pH 1.5 with HCl (37%), and polyphenolic compounds were extracted with ethyl acetate by stirring for 30 min at room temperature. Organic and aqueous phases were separated by decantation and the organic phase was collected and evaporated to dryness at 40 °C. Residual water was removed from the extract by lyophilization to yield the dry crude extract (Barbosa-Pereira, Angulo, et al., 2013). The content of phenolic compounds is shown in Table 1a (Barbosa-Pereira, Pocheville, et al., 2013).

2.2.2. Rosemary extract

Commercial rosemary extract (MS-198-08 GIN 601331 Rosemary Extract P) was supplied in powder form by INGRENAT (Ingredientes Naturales SL., Murcia, Spain). The composition of the rosemary extract is shown in Table 1b.

2.3. Instruments

The concentration of 2-thiobarbituric acid reactive substances (TBARs) and the absorbance in the DPPH method were determined in a dual-beam spectrophotometer (Uvikon XL, Bio-Tek Instruments, Milan, Italy).

Secondary oxidation compounds were extracted from beef by use of a T 25 ULTRA-TURRAX digital homogenizer and an MS2 Mini Vortex Shaker (both from IKA).

2.4. Antioxidant activity of natural extracts – DPPH method

The antioxidant activity of the natural extracts was determined by the DPPH (2,2-diphenyl-1-picrylhydrazyl) radical scavenging method described by von Gadow, Joubert, and Hansmann (1997), with slight modifications.

Standard solutions of the different antioxidants and of two synthetic compounds with antioxidant properties usually used in the food industry, PG and BHT, were prepared in methanol. An aliquot of antioxidant (50 µL) was added to 2 mL of DPPH radical methanolic

Phenolic compound (mg g^{-1})					
Benzoic acid derivates	38.1	Cinnamic acids	59.2	Flavonols	48.9
Gallic acid	20.1	Caffeic acid	14.1	Isoquercetin	28.3
Protocatechuic acid	15.5	p-Coumaric acid	11.4	Quercetin	14.5
4-Hydroxybenzoic acid	2.54	Ferulic acid	33.7	Kaempferol	6.06
Flavan-3-ols	214	Flavanones	14.6	Acetophenone derivates	14.6
Gallocatechin	132	Naringenin		Acetosyringone	
Epigallocatechin	30.6	-			
Catechin	29.9	Flavones	8.10	Stilbenoids	5.35
Epicatechin	21.4	Apigenin		Resveratrol	

Total phenolic compounds present in the PVPP-WS extract, 403 mg g^{-1} .

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