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ACTIVE PLASTIC TRAY INCLUDING ENCAPSULATED ESSENTIAL OILS IMPROVES THE QUALITY OF FRESH CHILLED BLUEBERRIES

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Abstract: *Blueberries production (mainly located in the Huelva area) in Spain has increased in the last decade due to the high health-promoting properties of these blueberries. However, the low optimum temperature for postharvest storage of blueberries implies high energy costs during conservation and transportation. However, plant essential oils (EOs) used as postharvest treatments may preserve the product quality, which may allow to increase in the setting temperature of cold rooms. EO active packaging is an excellent alternative to inefficient EO washing treatments (low EO solubility in water, high sample sensitivity to mechanical damages, removal of the antimicrobial natural waxy layer of blueberries, etc.). Hence, we studied the quality (firmness, microbial and sensorial) of blueberries during refrigerated storage (4 ºC) for up to 7 days using either EO active packaging (with an encapsulated carvacrol:oregano EO:cinnamon EO mix, 70:10:20) or commercial plastic containers. Blueberries packaged with active containers showed 20% higher firmness than control samples on day 7. Yeast loads of samples within the active packaging were 1.1 log units lower than control samples after 7 days. Overall, active packaging better preserved the quality of blueberries during cold storage, which was correlated with the sensory scores of samples.*

Keywords: *shelf life, refrigerated storage, food quality, food safety, β-cyclodextrin inclusion complex.*

1. INTRODUCTION

Blueberry (*Vaccinium corymbosum*) is a fruit belonging to the *Ericaseae* family, being only a few of its species marketable. Blueberries have gained a high worldwide recognition for their high antioxidant activity due to their high concentrations of a large variety of phenolic compounds. In particular, anthocyanins have neuroprotective, antioxidant, anti-inflammatory, and anti-proliferative qualities [1]. Hence, the blueberry production has increased by 72% between 1996 and 2016 [2].

Mechanical damage, which occurs during pre- and postharvest fruit handling, is considered a type of stress that triggers physiological and morphological changes in the fruit product [3]. The softening and bruising of blueberries are some of the most frequent quality disorders that lead to the non-marketability of the product [4]. Hence, a firm blueberry texture is expected by consumers because it is linked to fruit freshness. For that reason, fruit firmness is also an important objective for blueberry breeding because it is beneficial to everyone affected [5].

Essential oils (EOs) are naturally occurring plant extracts with high antimicrobial and antioxidant properties, which are highly accepted among consumers as natural additives. These technological properties are linked to the main EOs constituents: i.e. terpenes, terpenoids, low molecular weight aromatic/aliphatic chemicals, etc. [6]. Previous studies have examined the high *in vitro* antimicrobial activity of EOs and their primary constituents against a variety of spoilage and pathogenic microorganisms [7]. Namely, the primary ingredients of oregano EO are thymol and carvacrol, which are among the EOs components with the highest antimicrobial activities.

Antimicrobial active packaging is an efficient technology that allows increasing the shelf life of the product through a controlled release of antimicrobial compounds, such as encapsulated EOs [8]. Hence, the encapsulation of antimicrobial compounds with cyclodextrins (CDs) is a method that protects EOs from evaporation and degradation reactions (e.g. oxidation, heat degradation, etc.), while allowing its controlled release at the needed antimicrobial doses around the food product [9].
This study aimed to analyze the quality changes of blackberries packaged in recycled polyethylene terephthalate (rPET) packaging including encapsulated EOs during refrigerated storage at 4 ºC.

2. MATERIALS AND METHODS

Blueberries (*Vaccinium corymbosum*, “Biloxi” variety) were purchased in January 2022 in a close (<200 m) local supermarket. Blueberries were produced in Chile. Samples were kept at 7 ºC in a cold room of the food pilot plant of the Universidad Politécnica de Cartagena. Within the next day, the handling and packaging of samples were made in a laminar flow hood (class 100) located inside a clean room (Class 10,000) kept at 12 ºC.
Active packaging consisted of rPET containers (400 mL of capacity) including a polyamide pad on the bottom to avoid mechanical damage to the fruit. This active packaging was fabricated by Ondupet S.A. (Badajoz, Spain) including a coating of an encapsulated mix of 70:10:20 (volume (*v*):*v*:*v*)) carvacrol, oregano EO, and cinnamon EO sprayed on the rPET surface at 1 mg/m^2. The encapsulated (inclusion complex of EO:βCD) EO mix powder was fabricated by Bio-iPack (Fuente Álamo, Murcia, Spain). Carvacrol, oregano EO and cinnamon EO were obtained from Esencias Lozano (Caravaca de la Cruz, Spain). As a control packaging treatment, common commercial plastic non-active containers were used.
A quantity of 150 g of blueberries was introduced in each container and covered with a lid of the same material. Five replicates (packages) were prepared for each packaging treatment and storage time. Subsequently, packaged samples were stored up to 7 days at 7 ºC and 90% relative humidity in the cold room.

2.1. Fruit firmness

A texture analyzer (model TA.XT plus; Stable Micro Systems, Surrey, UK) with a load cell of 5 kg was used to analyze the berry firmness. A penetration test of 5 mm from the berry surface was carried out using a probe of 2 mm diameter. The pre- and post-test velocities were 2 and 5 mm/s, respectively. Ten berries from each replicate (package) with uniform colour and consistency were assayed. Firmness was expressed in g.

2.2. Microbial analysis

Microbial quality was conducted by mixing (stomacher for 30 s) samples in buffered peptone water (1:10 weight:*v* ratio). Subsequently, pertinent dilutions in buffered peptone water were made before pour plating in the correspondent agar media for mesophiles (plate count agar), psychrophiles (plate count agar) and enterobacteria (violet red bile dextrose agar), followed by incubation at 31 ºC/48 h, 7 ºC/7 days and 37 ºC/48 h respectively [10]. Results were expressed in colony-forming units (CFU) per sample surface area (cm^2).

2.3. Sensory analysis

A sensory analysis panel test was composed of researchers (4 women and 4 men between the ages of 21 and 39) from the UPCT, who were regular consumers of blueberries. Prior to sensory evaluation, each sample was randomly assigned a 3-digit code. The following sensory attributes were analyzed: visual appearance, color, texture, aroma, flavor, sweetness and acidity, as well as an overall quality score. A 9-point hedonic scale was used as follows: 1, extremely bad; 5, the limit of consumption; 9, excellent. Additionally, the presence of strange odours, flavors or colors were evaluated (1, absence of alteration; 9, intense alterations).

2.4. Statistical analysis

The SPSS software (v.19 IBM, New York, USA) was used for the statistical analysis, which involved multivariate analysis of variance (ANOVA) (packaging treatment × storage time) with Tukey's test (p=0.05).

3. RESULTS

3.1. Firmness

After 7 days of storage, the firmness of blueberries on active packages was 24.5 % higher than control packaging (Figure 1). In addition, no differences ($p>0.05$) were observed for every treatment during storage time. It indicates the better firmness quality of blueberries, which may be due to the resistance of their epidermis probably due to the inhibitory activity of EOs against cell wall degrading enzymes (i.e. polygalacturonase and pectinmethylesterase) as previously observed by our Group in other fruits [11] [12].

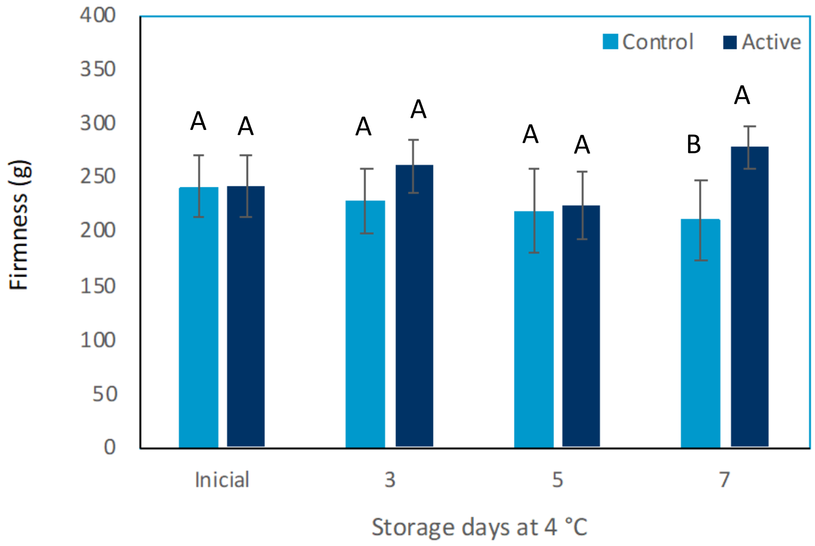

Figure 1. Blueberry firmness during storage at 4 º C for up to 7 days using active and control packaging treatments (mean (n=5) ±SD). Different uppercase letters denote significant differences ($p<0.05$) between packaging treatments for the same sampling time.

3.2. Microbial analysis

The mesophilic and psychrophilic counts of samples ranged from 4 to 5 log CFU g^{-1} (Figure 2). Between packaging methods, no differences are observed between control and active packaging treatments for mesophilic, psychrophilic and mould loads. There were also no differences in Enterobacteriaceae with ≈2 log CFU g^{-1}. In the case of yeasts, samples within active packaging showed 1.1 lower log units than control packaging.

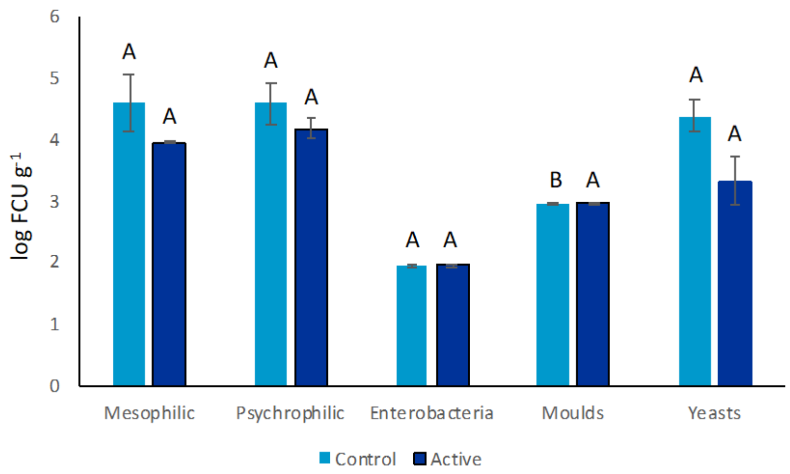

Figure 2. Microbial load for total mesophilic aerobes, psychrophiles, enterobacteria, molds and yeasts in blueberries under different packaging treatments after 7 days of conservation at 4 º C (mean (n=3) ± SD). Different letters denote significant differences ($p<0.05$) between control and active packaging for the same type of red fruit.

The low microbial variations among treatments could be the natural physical barrier present in blueberries: the bloom wax. This whitish surface coating occurs from the formation of a partial or continuous layer of amorphous or crystalline wax on the fruit surface during its ontogeny. The bloom wax could protect the underlying plant tissues from desiccation, pathogen infection and insect attack. The bloom wax is not only present in blueberries, as it can be found in other fruits such as grapes, as well as in other plant organs, such as stems and leaves. Its main function is to defend the fruit from the attack of entomological pests, ultraviolet light and also diseases caused by humidity since it acts by repelling droplets that may adhere to its surface [4].

3.3 Sensory analysis

Sensory scores of samples are shown in Figure 3. The product acidity increases or decreases depending on the maturity of the fruit [13]. Acidity is also an important factor when choosing an attractive product for the consumer. Total acidity is made up of the sum of numerous organic acids, the main ones being malic acid, ascorbic acid and citric acid [13]. Blueberries under the active packaging showed higher acidity scores, which denotes that this packaging treatment better preserved the fruit acidity, which indicates a lower senescence rate. In addition, blueberries under the active packaging were better scored for texture, flavour and overall quality.

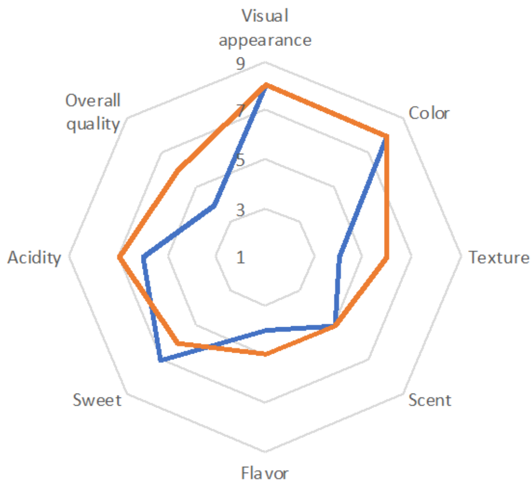

Figure 3. Sensory analysis of blueberries under different packaging treatments (control, blue line; active packaging, orange line) after 7 days of conservation at 4 ºC (mean (n=3)).

4. CONCLUSIONS

The use of active packaging with encapsulated essential oils in blueberries did not negatively affect the sensory properties of blueberries, which were even better preserved. Hence, the higher firmness and texture scores of samples within the active packaging indicate a higher potential resistance to possible damages generated during transportation and conservation. Although no significant microbial differences were observed, probably owed to the bloom wax barrier, blueberries of active packages showed ≈ 1 log unit lower counts than samples within the control packaging. As observed, the controlled release of essential oils from the active packaging preserved the fruit quality, specifically for key quality parameters of this fruit like firmness/texture and acidity.

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