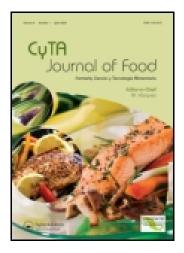
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# Effect of the addition of plant extracts on the microbiota of minimally processed strawberry jam and its physicochemical and sensorial properties

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# Effect of the addition of plant extracts on the microbiota of minimally processed strawberry jam and its physicochemical and sensorial properties

# Efecto de la adición de extractos vegetales en la microbiota y en las propiedades fisicoquíicas y sensoriales de mermelada de fresa mínimamente procesada

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Plant extract addition (pomegranate, rosemary, lemon and balsamic lemon) have been applied to strawberry jam obtained by osmotic dehydration in order to avoid its fast decline. Microbiological, physicochemical and sensorial parameters have been evaluated to compare product quality. Different doses of plant extract were tested to select those with the greatest antibacterial potential. Before determining its antimicrobial potential, the behaviour of the microbiota present in the strawberry jam was analysed using predictive microbiology. Moreover, the samples were analysed for moisture content, soluble solids, pH, water activity, colour and consistency. Strawberry jams were sensorially analysed. Pomegranate extract showed the highest level of antimicrobial activity. Jams with pomegranate showed significantly lower values in colour parameters than the control jam. However, there were no significant colour differences between jams with different doses of pomegranate extract. Jams without extracts were more consistent. In the sensory analysis, tasters found the studied samples to be very similar.

Keywords: strawberry jam; osmotic dehydration; plant extract; antimicrobial; microbiology; sensory evaluation

La adición de extractos vegetales (granada, romero, limón, limón balsámico) ha sido utilizada para mermeladas de fresa obtenidas por deshidratación osmótica con el fin de evitar su rápido deterioro. En este trabajo, se han evaluado los parámetros microbiológicos, físico-químicos y sensoriales del producto para su comparación. Para ello, se testaron diferentes dosis de extractos vegetales en las muestras para seleccionar la de mayor potencial antibacteriano. Anteriormente, se analizó el comportamiento de la microbiota presente en la mermelada (mesófilos aerobios, bacterias ácido lácticas, coliformes, mohos y levaduras). Además, las muestras se analizaron en cuanto a su contenido en humedad, sólidos solubles, pH, actividad del agua, color y consistencia. La memelada de fresa con y sin extractos fue analizada sensorialmente mediante una prueba de comparación pareada con un panel de 26 catadores. El extracto de granada mostró el mayor nivel de actividad antimicrobiana frente a la microbiota de la mermelada de fresa, estableciéndose una concentración mínima inhibitoria de 0,001 g / ml o 0,0005 g / ml si lo definimos como el nivel mínimo de concentración de extracto natural que produce una reducción del 90% en el crecimiento de colonias microbianas o una inhibición completa del crecimiento visible, respectivamente. Las mermeladas con extracto de granada mostraron valores significativamente más bajos en los parámetros de color que la muestra control. Sin embargo, no hubo diferencias significativas de análisis sensorial, las muestras estudiadas fueron muy similares para los catadores.

Palabras clave: mermelada de fresa; deshidratación osmótica; extracto de planta; antimicrobiano; microbiología; evaluación sensorial

#### Introduction

Strawberries have a low calorie content and the major components are water and carbohydrate glucose (417 g/kg), fructose (374 g/kg) and sucrose (209 g/kg) (Hulme, 1970). Strawberries' contribution to detary fibre and content of potassium, phosphorus, calcium, magnesium, ascorbic acid, malic acid and citric acid should be highlighted (Folquer, 1971; Pérez Afonso, 1979; Souci, Fachmann, & Kraut, 2006). Apart from nutrients, strawberry also contains other compounds with health benefits such as anthocyanins and carotenoids. These compounds are mainly responsible for the characteristic colour of strawberries (Holcroft & Kader, 1999; Torreggiani et al., 1999). Strawberries are one of the most consumed fruits and one of the favourites for

Jam is a traditional fruit product that includes heat treatment with high temperatures and long processing times. This kind of process can lead to important losses in beneficial properties (Igual, García-Martínez, Camacho & Martínez-Navarrete, 2010a). Osmotic dehydration uses mild temperatures (30–40°C) and so is a technique that could be used to obtain jam without being so aggressive with the components of the fruit (García-Martínez et al., 2002; Igual, Contreras, & Martínez-Navarrete, 2010b; Shi et al., 1996). Osmotic dehydration is a concentration technique, in which the fruit

consumers, especially young consumers (Abadía, 2004). However, like almost all fruits, strawberries are seasonal and hence there is interest in developing new products to extend their shelf life.

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is immersed in a highly concentrated solution in order to promote water loss in the fruit cells (Lazarides, 2001). The high concentration of solutes on the surface of the product contributes in obtaining a product with good taste and colour, in improving the cellular structure and in preventing the loss of pigments and aromatic compounds and the browning of the products (Heredia, Barrera, & Andres, 2007; Heredia, Peinado, Barrera, & Grau, 2009; Shi et al., 1996). Jams, as well as preserves and jellies, usually preserve well due to their high-sugar content, low-water activity and pH; furthermore, cooking destroys the microorganisms in the raw material. However, osmotic dehydration works at mild temperatures; therefore this technique does not destroy some types of bacteria, mould and yeast and there are osmophilic fungal species that can survive in high-sugar concentration (Pascual & Calderón, 2000). An alternative solution for this problem would be the use of plant extracts with antimicrobial activity. These extracts are phenolic compounds from bark, stems, leaves and flowers, and organic acids in fruits that retard the growth or kill microorganisms and increase the resistance to quality and safety altering (Beuchat, 2001). In this work, pomegranate, lemon, and rosemary extracts have been studied. Pomegranate (Punica granatum L) is a very rich source of anthocyanins, ellagitannins, and other phenolic compounds with proven antioxidant (Madrigal-Carballo, Rodriguez, Krueger, Dreher, & Reed, 2009; Pérez-Vicente, Serrano, Abellan, & Garcia-Viguera, 2004) and antimicrobial activity (Machado et al., 2002; Voravuthikunchai, Lortheeranuwat, & Jeeju, 2004). Pomegranate is effective against various microorganisms: Bacillus cereus, Listeria. monocytogenes, Escherichia. coli, Salmonella enteriditis, Staphylococcus aureus, Aeromonashydrophila and Streptococcus faecalis (Beuchat, 2001; Negi & Jayaprakasha, 2003; Shan, Cai, Brooks & Corke, 2007; Voravuthikunchai et al., 2004; Voravuthikunchai & Kitpipit, 2005), Pseudomonas aeruginosa, Bacillus subtilis (Nascimento, Locatelli, Freitas, & Silva, 2000) and Aspergillus flavus (Krishnamurthy & Shashikala, 2006). Lemon (Citrus limon) has antimicrobial and antifungal activity (Viuda-Martos, Ruiz-Navajas, Fernandez-Lopez, & Perez-Alvarez, 2008) and antioxidant activity (Xu et al., 2008). The antifungal activity is due to the presence of components such as D-limonene, linalool or citral (Alma et al., 2004; Rasooli, Moosavi, Rezaee, & Jaimand, 2002; Rodov, Ben-Yehoshua, Fang, Kim, & Ashkenazi, 1995; Skocibusic, Bezic, & Dunkic, 2006; Tepe et al., 2006; Vekiari et al., 2002; Veldhuizen, Tjeerdsma-Van Bokhoven, Zweijtzer, Burt, & Haagsman, 2006; Veriotti & Sacks, 2001). Finally, rosemary (Rosmarinus officinalis) slows down the growth of some bacteria such as E. coli (Gachkar et al., 2007; Tajkarimi, Ibrahim, & Cliver, 2010), L. monocitogenes (Pandit & Shelef, 1994) and S. aureus (Farbood, Macneil, & Ostovar, 1997). Also, rosemary has antioxidant activities due to rosamarinic acid, carnosic acid, and its phenolic dipteran carnosol, rosmanol and rosmaridiphenol (Inatani, Nakatani, Fuwa, & Seto, 1983; Nakatani & Inatani, 1981; Pérez-Fons, Garzon, & Micol, 2010; Yesil-Celiktas, Sevimli, Bedir & Vardar-Sukan, 2010).

The aim of this work was to evaluate the effect of addition of plant extracts (pomegranate, rosemary, lemon, and balsamic lemon) in minimally processed strawberry jam, prepared by osmotic dehydration, on the microbiological, physicochemical and sensorial characteristics.

#### Materials and methods

#### Raw materials

The experiments were made with strawberries (var. Camarosa) from the Huelva region of Spain, with similar characteristics in terms of  $a_w$ , °Brix, and pH (mean values and standard deviation: 0.991 (0.003), 6.5 (0.1) and 3.40 (0.02), respectively). These were purchased in a local supermarket. Fruit pieces were cut into four parts. An osmotic solution (OS) was prepared by mixing common food grade sugar (650 g/kg) with distilled water until it was completely dissolved, thus forming a 65 °Brix syrup. Citrus peel pectin (60% degree of sterification, Fluka Biochemika, Switzerland) was used as a gelling agent.

The following natural extracts were added to the strawberry jam to study the subsequent antimicrobial activity: lemon extract (Nutracitrus functional ingredients, Spain), pomegranate extract (Nutracitrus functional ingredients, Spain), balsamic lemon extract (Nutrafur, Spain) and rosemary extract (Nutrafur, Spain).

#### Processing

The strawberry pieces were submerged in the OS (the OS:fruit ratio was 5:1) and submitted to 50 mbar pressure for 10 min. The atmospheric pressure was then restored for an additional 10 min to promote impregnation with the OS. Samples with the OS were then heated to 30°C in a P-Selecta Precisterm bath (Selecta, Barcelona, Spain) with continuous stirring at 200 rpm (Heidolph Instruments, RZR 2020, Schwabach, Germany) for 3 h, and reaching  $\approx 20$  °Brix. The osmodehydrated samples were then ground together with part of the OS to obtain jam with 500 g fresh fruit/kg jam, with or without natural extracts and pectin (10 g/kg jam) as a gelling agent. The obtained jams were placed in glass jars and stored at room temperature for 24 h until analysis.

#### Analysis

#### Antimicrobial activity

The jam was stored for 20 days at room temperature. At the end of the period, the jams contained moulds and the structure was divided in two phases (liquid and gel phase). Inoculums were prepared from the altered jam. Thirty-five grams of jam were macerated in a peptone water phosphate-buffered (Sharlau) with a lab blender (Masticator, IUL Instruments, Barcelona, Spain). An aliquot of 5 mL of leaf homogenate was transferred to 250 mL of Brain Heart Infusion broth (BHI broth) and incubated overnight at 37°C. The media was concentrated by centrifugation at 11,000 rpm at 4°C for 5 min (Centrifuge 5804R, Eppendorf, Hamburg, Germany) and frozen at  $-80^{\circ}$ C after the addition of 20% of glycerol. The final concentration of microorganisms in the inoculums was approximately  $1.14 \times 10^{10}$  cfu/mL of mesophilic aerobic bacteria and  $1.24 \times 10^{5}$  cfu/mL of moulds and yeast.

Before determining the antimicrobial potential of the plant extracts, the behaviour of the native microflora was studied. The growth curve (at room temperature and BHI media) was shaped by Gompertz and Baranyi models (Baranyi & Roberts, 1994). The sensitivity of microorganisms to the plant extracts was determined by two techniques. The first technique was the agar diffusion method; the natural extract was applied to plate count agar (PCA) to evaluate the antimicrobial activity of the inoculum  $(10^2-10^4-10^6 \text{ cfu/mL})$ when mixed with the agar in the plate. When the agar was solidified, five wells were made. In each well, 100  $\mu$ L of various concentrations of natural extracts were introduced. The plates with PCA were incubated at 30°C for 24 h. After the incubation of the plates, the zones of inhibition were measured. Results of this test are qualitative, so microorganisms are generally termed susceptible, intermediate, or resistant, depending on the diameter of the inhibitory zone (Davidson & Parish, 1989).

The second method was broth dilution. In this assay, studied compounds were serially diluted and distributed in a nutrient broth (Davidson & Parish, 1989) (BHI). In the mediums, natural extracts were applied, inoculated with inoculums  $(10^2 \text{ cfu/mL})$  of altered jam in tubes and incubated at 37°C for 24–48 h. Then, 1 mL from each tube was put on a PCA plate and these were incubated in the same way as in the first method.

The minimum inhibitory concentration (MIC) is defined as the minimum level of natural extract concentration that produces a 90% reduction in the growth (population) of microbial colonies (Ponce, Fritz, Valle, & Roura, 2003) or a complete inhibition of visible growth (Jia et al., 2010). The minimum bactericidal concentration (MBC) is defined as the minimum level of natural extract concentration that produces at least a 99.9% reduction in the growth of microbial or fungal colonies, respectively (Skandamis, Koutsoumanis, Fasseas, & Nychas, 2001).

#### Physicochemical and sensorial properties

All the samples were analysed for moisture content  $(x_w)$  (AOAC method 934.06, 2000), the soluble solids of the liquid phase of the sample at 20°C (°Brix) (refractometer, Zeiss, ATAGO model NAR-3T refractometer, Tokyo, Japan) and water activity ( $a_w$ ) (dew point hygrometer FA-st Lab, GBX, Lyon, France). The pH was measured using a CRISON pH-meter (Crison Instruments S.A., Barcelona, Spain).

Colour was measured using CIE-L\*a\*b\* coordinates (L\*, a\*, b\*  $C_{ab}$ \* and  $h_{ab}$ \*) obtained with a 10° observer and a D65 illuminant. A CM 3600D espectrophotometer with a cuvette that contained the jam, (Konica, Minolta, Tokyo, Japan) was used. The colour coordinates were then used to calculate colour differences,  $\Delta E$ , (Equation (1)) with respect to the control strawberry jam sample.

$$\Delta E = \sqrt{\Delta L^{*2} + \Delta a^{*2} + \Delta b^{*2}} \tag{1}$$

The parameter used to characterise the consistency of the samples was the distance the samples advanced in the consistometer related to the weight of the sample (mm/g). The flow distance of a controlled sample weight for a constant time was measured using a Bostwick consistometer (Aname, Madrid, Spain), and the distance the sample flowed in 30 s was measured (Bourne, 1982).

All the experiments were replicated three times in each batch.

A panel of 26 assessors carried out the sensory analysis of strawberry jam without extract, and strawberry jam with the various concentrations of selected extracts. This analysis consisted of a paired comparison test (UNE-EN ISO 5495, 2009). The evaluated attributes were colour, smell, taste, texture, product coverage in mouth and preference. During the test session, the panellists worked in individual booths. Samples were served at room temperature in transparent plastic glasses coded with three digit random numbers. Each panellist tasted approximately the same amount of each sample, toast was used to spread the jam and evaluate the texture, and mineral water was provided to the assessors to rinse their mouths.

#### Statistical analysis

Analysis of variance (ANOVA), with a confidence level of 95% (p < 0.05), was applied using Statgraphics Plus 5.1 Software (Statistical Graphics Corporation, USA) to evaluate the differences among the treatments. Friedman analysis for the pairwise ranking test (Meilgaard, Civille, & Carr, 1999) was undertaken on the data from each taster to know in which attributes the samples showed significant differences. The significance of these differences was also determined by applying Tukey's HSD (Honestly Significance Difference) as a multiple comparison procedure (Meilgaard et al., 1999). Correspondence analysis (CA) was applied to the sensorial results using the SPSS program version 16.0. Correlations between sensory and instrumental data were determined using the Pearson correlation coefficient (r).

#### **Results and discussion**

## Determination of the plant extracts' antimicrobial potential

The aim of this study was to determine the potential antimicrobial activity of the four plant extracts (pomegranate, lemon, rosemary and balsamic lemon). Different doses of these compounds were tested to select those with the greatest antibacterial potential. Before the agar diffusion and broth dilution, the behaviour of the microbiota present in the strawberry jam was analysed using predictive microbiology. The results showed a growth curve and associated parameters characteristic of the microbiota present in strawberry jam obtained by osmotic dehydration.

The growth curve showed the typical sigmoid curve of microbial growth, consisting of a lag phase, exponential growth, and finally, a stationary phase. To verify this behaviour, the growth curve was modelled on the Gompertz and Baranyi models, as these are most commonly used for microbial growth. Characteristic parameters of microbial growth were obtained from these models (Table 1) and the highest N (logcfu/mL) population density was highlighted; reaching 8 logcfu/mL.

The study of the antimicrobial activity of the extracts was tested using: the diffusion agar method and by considering charge of microbiota jam of  $10^4$  cfu/mL and  $10^6$  cfu/mL. The first one is the recommended concentration for such assays (Barry, 1986);  $10^6$  cfu/mL is used to determine whether activity was maintained under extreme conditions of pollution. In Figure 1A and B, the antibacterial activity of the extracts tested against a charge of jam microbiota of  $10^4$  and  $10^6$  cfu/mL are shown, respectively.

As can be seen in Figure 1, the most effective natural extract was pomegranate, independently of the dose, followed by lemon extract. These extracts were selected for more detailed study.

Figure 2 shows the results obtained for the pomegranate extract using the diffusion agar method. These indicate that a lower concentration of inoculum increases the effectiveness of

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Table 1. Characteristic parameters of growth for the jam microbiota.

Tabla 1. Parámetros característicos de crecimiento para la microbiota de la mermelada.

Parameters	Moo	del
	Gompertz	Baranyi
$\mu$ [(log ufc/mL)h <sup>-1</sup> ]	0.721	0.572
$\lambda$ [h]	0.000	_
$N_0$ (log ufc/mL)	0.640	1.205
N (log ufc/mL)	8.036	8.009
s.e.	0.348	0.389
$R^2$	0.980	0.976

Note:  $\mu$ : the potential maximum rate;  $\lambda$ : lag phase; s.e.: standard error of fitting;  $N_0$ : initial concentration of cells; N: final concentration of cells;  $R^2$ : Adjusted R-square statistics of the fitting.

Nota:  $\mu$ : velocidad máxima de crecimiento;  $\lambda$ : tiempo de latencia; s.e.: error estándar estimado del ajuste;  $N_0$ : concentración inicial de células; N: concentración final de células;  $R^2$ : coeficiente de correlación.

the extract, regardless of the dose used. In the case of a high levels of inoculum (10<sup>6</sup> cfu/mL), antimicrobial ability was significantly reduced for doses equal to or less than 0.1 g/mL; and disappeared in doses equal to or less than 0.034 g/mL. For this type of inoculum, the minimum concentration with antimicrobial activity was 0.04 g/mL. When the inoculum level was lower, a jam microbiota dose of 0.023 g/mL showed antimicrobial activity. Antimicrobial activity was also observed at the dose of 0.02 g/mL for inoculum of  $10^2$  cfu/mL. The broth dilution technique was used to quantify more exactly the antimicrobial activity of the pomegranate extract (Davison & Parish, 1989). In this case, the extract concentrations tested were 0.012, 0.008, 0.006, 0.004, 0.003, 0.002, 0.001 and 0.0005 g/mL. The result shown by the jam at 24 h was  $6.06 \times 10^8$  cfu/mL and with MIC, defined as the minimum inhibitory concentration of extract that causes complete inhibition of visible growth (Jia et al., 2010), would be 0.001 g/mL. If MIC is considered as the concentration that produces a 90% reduction (Ponce et al., 2003), it would

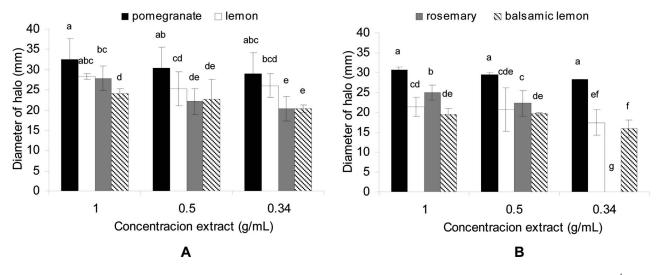


Figure 1. Antimicrobial activity of natural extracts (pomegranate, lemon, rosemary and balsamic lemon) tested in inoculums of: (a)  $10^4$  cfu/mL and (b)  $10^6$  cfu/mL.

Figura 1. Actividad antimicrobiana de los extractos naturals (granada, limón, romero y limón balsámico) testados en inóculos de: a) 10<sup>4</sup>cfu/mL and b) 10<sup>6</sup>cfu/mL.

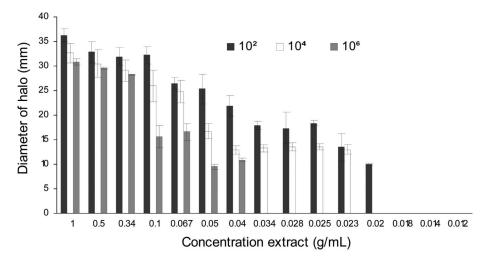


Figure 2. Antimicrobial activity of pomegranate extract tested in inoculums of 10<sup>2</sup> cfu/mL, 10<sup>4</sup> cfu/mL and 10<sup>6</sup> cfu/mL.

Figura 2. Actividad antimicrobiana del extracto de granada testado en inóculos de 10<sup>2</sup>cfu/mL, 10<sup>4</sup>cfu/mL and 10<sup>6</sup>cfu/mL.

correspond to 0.0005 g/mL. If MBC were defined as the concentration causing 99.9% inhibition (Skandamis et al., 2001), then the result would also be a concentration of 0.0005 g/mL.

The minimum dose of antimicrobial activity in the case of lemon extract was 0.14 g/mL (Figure 3). Pomegranate behaves more actively against bacteria in the jam, and so this extract was selected as an antibacterial against the bacterial flora of the jam. The chosen dose for the pomegranate extract was 0.0005 g/mL according to the CMI (Ponce et al., 2003). Using the definition of CMI by Jia et al. (2010), the dose would be 0.001 g/mL.

## Effect of antimicrobial addition on physicochemical and sensory properties of jams

Three batches of strawberry jam were prepared with the selected extracts and dosages in the antimicrobial study: firstly, the control without pomegranate extract (OD); secondly, a mix of jam ingredients with 0.0005 g/mL of pomegra-

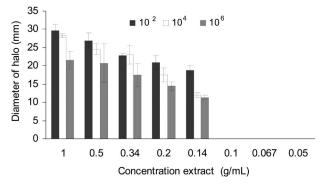


Figure 3. Antimicrobial activity of lemon extract tested in inoculums with  $10^2\ cfu/mL,\ 10^4\ cfu/mL$  and  $10^6\ cfu/mL.$ 

nate (OD + 0.0005 gPG/mL); and thirdly, a (OD + 0.001 gPG/mL) 0.001 g/mL of pomegranate extract was added.

Table 2 shows the values of °Brix,  $a_w$ ,  $x_w$  and pH for the three batches of strawberry jam. There were no significant differences among them. The mean values were  $\approx 44, 0.943,$ 0.53 and 3.45, respectively. Table 2 also shows the values of flow distance corrected for the sample weight. The OD + 0.001 gPG/mL sample was the least consistent (highest distance advanced). However, the sample without extract was significantly (p < 0.05) the most consistent. This may be because pomegranate is composed mostly of phenolic compounds (punicalagin, ellagic acid) (Gil, Tomás-Barberán, Hess-Pierce, Holcroft, & Kader, 2000; Ibrahium, 2010). These compounds can bind pectins in aqueous mediums (Pierzynowka-Korniak, Zadernowski, & Nesterowicz, 1995) as described for other compounds such as proteins, whose interaction with the polar rings of the polyphenol structure occurs through hydrophobic associations (Prigent et al., 2003; Rubino, Arntfield, Nadon, & Bernatsky, 1996). These actions delay the hydration of pectin and reduce the ability to form gel.

Colour analysis (Table 3) showed a significant decrease in all the parameters for the samples with pomegranate extract, while jams with two different doses of pomegranate extract showed no significant variations between them. The addition of pomegranate extract in any dose caused with less red tonality (a\* decrease) but the samples were darker (L\* decreased) and with less colour purity ( $C_{ab}$ \* decrease). This change is probably due to the anthocyanin content (Zafrilla, Valero, & García-Viguera, 1998). There are slight colour differences between the jams when compared with the control and there were no significant (p < 0.05) differences between the jams formulated with pomegranate extract.

The results of Friedman's T test to know the significant evaluated attributes revealed that colour and product coverage in mouth showed statistically significant differences ( $\alpha = 0.05$ ) in the studied samples. Friedman's T for these attributes were 6.3 and 15.2, respectively, and the theoretical T value ( $\alpha = 0.05$ ) was 5.99. The HSD method was used to

Table 2. Mean values (and standard deviation) of "Brix, pH, water activity ( $a_w$ ) and moisture content ( $x_w$ ) of formulated jams.

Tabla 2. Valores medios (y desviación estandar) de °Brix, pH, actividad del agua  $(a_w)$  y contenido en humedad  $(x_w)$  de las mermeladas formuladas.

Sample	°Brix	$a_{\mathrm{w}}$	$X_{ m W}$	pH	Distance/weight (mm/g)
OD OD + 0.0005gPG/mL OD + 0.001gPG/mL	44.5 (0.2) <sup>a</sup> 44.0 (0.2) <sup>a</sup> 44.1 (0.2) <sup>a</sup>	$\begin{array}{c} 0.944 \; (0.003)^{a} \\ 0.944 \; (0.003)^{a} \\ 0.942 \; (0.003)^{a} \end{array}$	$\begin{array}{c} 0.5290  (0.0001)^a \\ 0.534  (0.006)^a \\ 0.539  (0.001)^a \end{array}$	$\begin{array}{c} 3.47 \ (0.02)^a \\ 3.45 \ (0.02)^a \\ 3.47 \ (0.02)^a \end{array}$	0.96 (0.06) <sup>c</sup> 1.116 (0.012) <sup>b</sup> 1.176 (0.03) <sup>a</sup>

Note: The same letter in superscript within columns indicates homogeneous groups established by ANOVA (p > 0.05).

Nota: La misma letra en superíndice de las columnas indica los grupos homogéneos establecidos por el ANOVA (p > 0.05).

Table 3. Mean values (and standard deviation) of colour parameters of formulated jams.

Tabla 3. Valores medios (y desviación estándar) de los parámetros de color de las mermeladas formuladas.

Sample	L*	a*	b*	C* <sub>ab</sub>	h* <sub>ab</sub>	$\Delta E$
OD OD + 0.0005gPG/mL OD + 0.001gPG/mL	29.9 (0.3) <sup>a</sup> 28.8 (0.3) <sup>b</sup> 28.67 (0.17) <sup>b</sup>	16.4 (0.2) <sup>a</sup> 13.7 (0.7) <sup>b</sup> 13.4 (0.2) <sup>b</sup>	$\begin{array}{c} 6.77 \ (0.15)^{a} \\ 5.3 \ (0.4)^{b} \\ 5.19 \ (0.14)^{b} \end{array}$	17.7 (0.3) <sup>a</sup> 14.7 (0.8) <sup>b</sup> 14.4 (0.3) <sup>b</sup>	$\begin{array}{c} 22.44 \ (0.13)^{a} \\ 21.4 \ (0.3)^{b} \\ 21.1 \ (0.2)^{b} \end{array}$	$3.2 (1.2)^{\rm b}$ $3.61 (0.25)^{\rm b}$

Note: The same letter in superscript within columns indicates homogeneous groups established by ANOVA (p > 0.05).

Nota: La misma letra en superíndice de las columnas indica los grupos homogéneos establecidos por el ANOVA (p > 0.05).

Figura 3. Actividad antimicrobiana del extracto de limón testado en inóculos con  $10^2$  cfu/mL,  $10^4$ cfu/mL and  $10^6$ cfu/mL.

perform multiple comparisons among the samples. Tukey's HSD value was calculated according to assay conditions and the result was 14.62. Differences between the sum of ranks for OD jam when it was compared with the other jams with pomegranate extract only existed for the attribute product coverage in mouth (OD + 0.0005 gPG/mL was 22 and

Table 4. Contribution of dimension to inertia of attributes and jams.

Table 4. Contribución de la dimension a la inercia de los atributos y mermeladas

Attribute/Jam	D1	D2	Total
Colour	0.969	0.031	1.000
Smell	0.443	0.557	1.000
Texture	0.610	0.390	1.000
Taste	0.043	0.957	1.000
Product coverage in mouth	0.926	0.074	1.000
Preference	0.213	0.787	1.000
OD	1.000	0.000	1.000
OD + 0.0005G	0.741	0.259	1.000
OD + 0.001G	0.730	0.270	1.000

OD + 0.001 gPG/mL was 20). In general, the studied samples were very similar for tasters. A CA was carried out to relate the jams obtained by the different formulations with evaluated attributes and assessor preferences. From this analysis, two dimensions that explained 100% of the variability of the results were obtained. The first dimension (D1) explained 87.7% of the variability, while the second (D2) explained 12.3%. Table 4 shows that both jams as the attributes are well represented along the first two dimensions, since the obtained values for the sum of the relative contributions equalled 1 in all cases. Figure 4 shows the projection along the plane of the jams and attributes. D1 clearly separated the colour attribute on left-hand side and product coverage in mouth and texture on right-hand side with an opposing relationship and this fact is in accordance with instrumental measurements. According to the distribution of attributes and samples along the plane, there were three groups that characterised the jam as follows: firstly, OD was identified with texture and product coverage in mouth; secondly, OD + 0.0005 gPG/mL was identified with taste, smell and preference attributes; and finally, OD + 0.001gPG/mL was identified with colour attributes. Furthermore, it can be observed that taste and smell attributes were closely

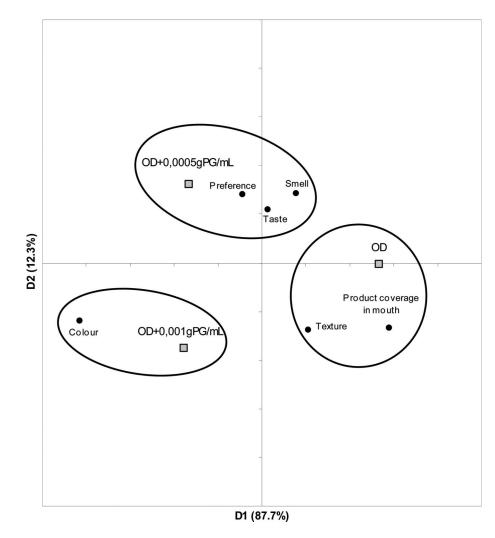


Figure 4. Correspondence analysis. Representation of attributes and samples tested in plain with two dimensions.

Figura 4. Análisis de correspondencia. Representación de los atributos y muestras analizados en un plano con dos dimensiones.

related to the preference of the judges, since they were represented as being very near to each other.

Pearson analysis was used to establish correlations between colour and consistency parameters measured by instrumental and sensory methods. The results showed a significant negative correlation between sensory colour and parameter L\*, a\*, b\*,  $C^*_{ab}$  and  $h^*_{ab}$ . The highest correlation coefficients (r) were between colour attribute and a\*, b\* and  $C^*_{ab}$ , 0.9183, 0.9093 and 0.9167, respectively, meaning that tasters identified the sensory attribute colour with colour purity. However, the relationship between the attributes is negative since the assessors related darkness with intensity of colour. As expected, the texture and product coverage in mouth of the studied jams showed a significant and negative correlation with flow distance (mm/g), r = 0.8712 and r = 0.8934, respectively.

#### Conclusion

In vitro studies indicate that of the plant extracts tested in this study, it was pomegranate extract that showed the greatest antimicrobial activity. According to problems presented by the use of osmotic dehydration for the production of jams in terms of speed deterioration since it is a non-thermal concentration, this study concludes that the addition of pomegranate extract increases its stability during storage at room temperature without affecting their sensory properties.

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