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Effect of a new remediated substrate on bioactive compounds and antioxidant characteristics of pomegranate (*Punica granatum* L.) cultivar 'Purple Queen'

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ABSTRACT

The maritime ports and terminals periodically carry out dredging activities. This operation is necessary in order to avoid the excessive accumulation of sediments in the areas of maneuvering and berthing of ships. Thus, the objective of this study was to investigate the effect that dredged remediated sediments have on the quality and bioactive components of pomegranate (*Punica granatum* L.) cultivar 'Purple Queen'. Pomegranate trees were grown on three substrate-based treatments: Peat 100% (Pt as a control), dredged remediated sediments 100% (DRS), and a 50% mixture of each (Pt-DRS). Pomegranate fruits and two types of juices (juice of arils and juice of arils plus carpellary membranes) were characterized. The results showed that the use of dredged remediated sediments 100% (DRS) negatively affected yield, numbers of fruit per tree, and fruit weight average of pomegranate fruits. However, neither fruit quality nor composition of pomegranates grown on this substrate and mixture were affected. The tested media showed suitable horticultural properties for pomegranate cultivation, at which the Pt and Pt-DRS had better plant responses than the DRS one.

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Antioxidant activity; dredged remediated sediment; polyphenols; punicalagins; *Punica granatum*

Introduction

The maritime ports and terminals periodically carry out dredging activities. This operation is necessary in order to avoid the excessive accumulation of sediments in the areas of maneuvering and berthing of ships, with the consequent loss of navigability and competitiveness compared to other terminals (Viñes 2011). The potential use of dredge materials for agricultural purposes contributes to reducing the need for off-shore disposal and provides an alternative to disposal the materials in landfills. Because of its variable but unique physical and chemical properties, dredged materials are often viewed by society and regulators as pollutants, even though many have been used for coastal nourishment, land or wetland creation, construction materials, and for soil improvement as amendments (Sigua et al. 2004). Studies on dredged sediments in ports have shown that decontamination techniques such as phytoremediation can be successful for obtaining new substrates for plant growth (Mattei et al. 2016). More than 90% of dredged sediments are not contaminated, which means that in many cases it can be considered as a resource rather than a waste (CEDEX 2012). The recent EU Waste Directive supports the re-use of sediments dredged in

ports which, unlike those dredged from highly contaminated sites, can be re-used without specific remediation actions; if they do not contain excessive levels of organic and inorganic contaminants, as determined by official method. However, current EU legislation is ambiguous about the possibility of using treated dredged sediments as by-products for agriculture. European countries depend on their own national legislation frameworks or on the regulations of local authorities.

To find an ecologically worthwhile use, the LIFE HORTISED project (LIFE14 ENV/IT/000113) tries to demonstrate the suitability of dredged remediated sediments as an alternative substrate to peat for horticultural purposes. Therefore, the main objective of this study was to assess the effect that dredged remediated sediments have on the quality and bioactive components of pomegranate fruit, cultivar '*Purple Queen*'.

Materials and methods

Plant material and experimental design

The experiment was conducted over 2016 and 2017 in an experimental plot belonging to Miguel Hernández University at Orihuela campus (Southeastern Spain, 38°04'N, 0°58'W, 26 masl). The tested plant material consisted of pomegranate trees cv. '*Purple Queen*', an extra-early cultivar which reaches full ripeness between mid-August and late September (Northern hemisphere). After planting, on 2 May 2016, pomegranates were hedged up to 40 cm. and grown in 40 L polyethylene pots. A support system separately collects the water drainage of each experimental block by galvanized steel trays. The growing trees were trained on a trellising system. '*Purple Queen*' pomegranates were grown on 3 different substrates: (i) peat 100% (Pt as a control); (ii) dredged remediated sediments 100% (DRS); and (iii) a 50% mixture of each (Pt-DRS). Each treatment consisted of three replications (5 trees each) in a randomized complete block design (RCBD). A drip irrigation system (delivering 2 L h⁻¹ each dripper) was used for water and mineral nutrition of the plants. All treatments received a complete Hoagland nutrient solution, composed of KNO₃, NH₄NO₃, K₂SO₄, HNO₃, H₃PO₄, and a complex mix of microelements. For irrigation scheduling, matrix voltage in the root zone was measured by installing 3 probes Watermark® in every treatment at 15 cm deep. Irrigation started when the matrix voltage was 10 cb and stopped when the required water volume was reached. To monitor water status, tensiometric readings were daily recorded.

Sediment substrate properties

The sediment tested in this experiment was the same used in a strawberry experiment conducted by our research group and published by Melgarejo et al. (2017). Briefly, the sediments used were obtained from the dredging of Leghorn Port (Italy). They were partially decontaminated (bioremediation) during 3 years; in a previous European project (AGRIPORT Agricultural Reuse of Polluted dredged Sediments, No. ECO/08/239065/S12.532262), using plants (phyto-treatment) and organic amendments at pilot scale. Afterwards, a series of analyses physical, chemical and biochemical were carried out. Also, soil toxicity was assessed according to the ISO standard method (ISO 11348-3, 1998). The results obtained in the analyses complied with the Spanish legislation on growing substrates for agricultural uses (Royal Decree 865/2010, July 2nd).

Physico-chemical determinations

Fifteen fruits per substrate were weighed and sized. Then, pomegranates were washed with water and completely dried. Two types of juice were made: (i) seven fruits were collected and peeled by hand, and the arils extracted while removing all the carpellary membranes; arils were weighed, homogenised and squeezed in two-layered muslin cloth to extract the complete juice; and (ii) other

seven fruits were cut in halves and carefully hand-squeezed using a commercial kitchen juicer, piths carpellary membranes and the arils all together. Freshly squeezed juices were centrifuged at $15,000 \times g$ at 4°C for 20 min (Sigma 3–18 K, Germany), and stored at -80°C until chemical analyses were conducted.

The following variables were determined: fruit weight (g), yield per tree (kg), number of fruits per tree, equatorial diameter (mm), calyx diameter (mm), fruit length without calyx (mm), total fruit length (mm), calyx length (mm), number of carpels (Nc) counted in the equatorial section, rind weight plus weight of carpellary membranes (Rw + Cm) (g), rind thickness (Rt) (mm) (measurements made on two opposite sides of the equatorial zone), arils yield (%), total soluble solids (TSS), titratable acidity (TA) and maturity index (MI). Results were expressed as mean values \pm SE (standard error).

Antioxidant activity ABTS, DPPH and FRAP methods

Antioxidant activity was quantified by spectrophotometry as described by Melgarejo et al. (2017). The free radical scavenging activities were determined using three standard methods: ABTS (2,2-azinobis-3-ethylbenzothiazoline-6-sulfonic acid radical cation), DPPH (2,2-diphenyl-1-picrylhydrazyl radical) and FRAP (ferric reducing antioxidant power). The ABTS, DPPH and FRAP assays were conducted as previously described by Re et al. (1999), Brand-Williams et al. (1995), and Benzie and Strain (1996), respectively. Trolox was used as reference standard, and results were expressed as mM of Trolox equivalent dry weight (dw). Analyses were run in triplicate.

Total phenol

The total phenol content (TPC) of pomegranate juice was estimated using the Folin-Ciocalteu's reagent and following the recommendations of Singleton et al. (1999). Gallic acid (GA) was employed as reference standard, and results were expressed as mg of gallic acid equivalents 100 g^{-1} dw. Analyses were run in triplicate.

Identification and quantification of ellagic acid

Ellagic acid was identified and quantified by high performance liquid chromatography (HPLC) as described by Calín-Sánchez et al. (2013). For the identification of ellagic acid, spectrum and retention times were used and compared with that obtained from authentic standard. The standard curve for ellagic acid was used for quantification. Analysis was run in triplicate and results were expressed as g L^{-1} .

Punicalagin isomers

Punicalagins (α and β) contents were determined as described by Nuncio-Jáuregui et al. (2015). For the identification of punicalagins, absorption spectra and retention times were employed and compared with those obtained from chemical standards. The standard curves for pure punicalagins (Chengdu Biopurify Phytochemicals Ltd., Sichuan, China), with a concentration range between 0.05 and 0.80 g L^{-1} , were used for quantification. Results for individual isomer punicalagins (α and β) were expressed as g L^{-1} . Analysis was run in triplicate.

Organic acids and sugars

Organic acids and sugars profiles were quantified according to Hernández et al. (2016). Briefly, a sample of 2 mL of each type of pomegranate juice was subjected to ultra-sonication extraction for 30 min with 5 mL of Milli-Q water, centrifugation at $15,000 \times g$ for 20 min (Sigma 3–18 K; Sigma,

Osterode am Harz, Germany). Then, one millilitre of the hydrophilic extract centrifuged was filtered through a 0.45 μm Millipore filter, and 10 μL were injected into a Hewlett- Packard HPLC Series 1100 (Wilmington DE, USA) with an autosampler and an UV detector, set at 210 nm and coupled with a refractive index detector (HP 1100, G1362A). A column (Supelcogel TM C-610H column 30 cm \times 7.8 mm) and a pre-column (Supelguard 5 cm \times 4.6 mm; Supelco, Bellefonte, PA) were used for the analyses of both organic acids and sugars. The elution buffer consisted of 0.1% phosphoric acid at a flow rate of 0.5 mL min^{-1} ; organic acid absorbance was measured at 210 nm using a diode-array detector (DAD). The same HPLC conditions (elution buffer, flow rate and column) were used for the analysis of sugars. The detection was conducted using a refractive index detector (RID). Standards of organic acids and sugars were obtained from Sigma (St. Louis, MO). Calibration curves were used for the quantification of organic acids and sugars, showing good linearity ($r^2 \geq 0.999$). Analyses were run by triplicate, and results were expressed as concentrations of g 100 mL $^{-1}$ of fresh weight.

Statistical analyses

A weighted analysis of variance was performed for statistical analysis (ANOVA; statistical software IBM SPSS Statistics v. 24 for Windows). The Shapiro–Wilk test was used to evaluate the normality of the data. Tukey's HSD test was used for mean separation. The significance level was $p \leq 0.05$.

Results

Effect of the substrates on yield and main morphological parameters of 'Purple Queen' pomegranate

The results obtained in the study showed that the use of different substrates definitely affected yield, numbers of fruit per tree, and fruit weight average of pomegranate fruits. The data shown in Table 1 revealed that treatment DRS (dredged remediated sediments 100%) significantly decreased the average fruit weight (227.4 g), number of fruit per tree (18) and yield (4.3 kg), while no significant differences were established between the control substrate (peat) and the Pt-DRS mixture.

Regarding to equatorial diameter, calyx length, number of carpels, rind weight plus weight of carpillary membranes, rind thickness and arils yield, there were no significant differences among the substrates tested (Table 2). However, with regard to total fruit length and calix diameter, the lowest values were obtained for peat-DRS and DRS substrates, with no statistical differences between them; the highest value of calyx diameter was obtained for peat substrate.

Effect of the substrates on chemical properties of 'Purple Queen' pomegranate

Table 3 showed the chemical properties of the pomegranate juices analyzed. Regarding pH, TA and MI for both types of juice, no significant differences were found among the substrates assayed. About pH, the values ranged from 4.6 (Pt) to 5.4 (Pt-DRS) in aril juice, and between 4.3 (DRS) to 4.8 (Pt-DRS) in aril juice plus carpillary membranes. Concerning to titratable acidity (TA) (Table 3), both types of juices analyzed showed TA values higher than 3.1 (g citric acid 100 mL $^{-1}$). On the other hand, statistically significant

Table 1. Yield per tree, fruits per tree and fruit weight average on different substrates.

| | Yield tree $^{-1}$ (Kg) | N $^{\circ}$ fruits tree $^{-1}$ | Fruit weight average (g) |
|--------|-------------------------|----------------------------------|--------------------------|
| Pt | 6.63 \pm 0.35 a | 22 \pm 1.33 a | 261.8 \pm 11.32 a |
| Pt-DRS | 6.29 \pm 0.28 a | 25 \pm 1.29 a | 248.8 \pm 11.64 a |
| DRS | 4.30 \pm 0.37 b | 18 \pm 1.86 b | 227.4 \pm 10.29 b |

Different letters next to a value within columns indicate significant differences among treatments according to Tukey's HSD test ($p < 0.05$). Pt: Peat 100% (control); DRS: Dredged remediated sediments 100%; (Pt-DRS) mixture 50% each.

Table 2. Mean values of the principal morphological parameters of pomegranate fruits on different substrates.

| | Pt | Pt-DRS | DRS |
|---------------------------------|---------------------------|----------------|----------------|
| Equ. Diameter (mm) | 81.88 ± 1.18a | 79.93 ± 1.48a | 78.30 ± 1.34a |
| Calix diameter (mm) | 25.16 ± 0.89a | 20.43 ± 0.89b | 16.48 ± 0.56b |
| Fruit length without calyx (mm) | 73.91 ± 1.16a | 70.05 ± 1.34ab | 67.77 ± 1.05b |
| Total fruit length (mm) | 92.47 ± 1.50a | 86.07 ± 1.83ab | 82.24 ± 1.16b |
| Calyx length (mm) | 18.56 ± 1.33a | 16.02 ± 0.80a | 14.47 ± 0.98a |
| Nc | 5.73 ± 0.18a | 5.93 ± 0.18a | 6.20 ± 0.14a |
| Rw + Cm (g) | 123.80 ± 5.90a | 116.34 ± 5.76a | 115.73 ± 8.84a |
| Skin thickness (mm) | 4.81 ± 0.17a | 4.34 ± 0.23a | 4.61 ± 0.36a |
| Arils yield (%) | 54.22 ± 0.80a | 56.21 ± 1.44a | 52.17 ± 1.43a |
| MC (%) | 83.89 ± 0.60 ^a | 83.60 ± 0.70a | 83.44 ± 0.62a |

Different letters next to a value in within rows indicate significant differences among treatments according to Tukey's HSD test ($p < 0.05$). Nc: number of carpels; Rw + Cm: rind weight plus weight of carpellary membranes. Mc: moisture content; Pt: Peat 100% (control); DRS: Dredged remediated sediments 100%; (Pt-DRS) mixture 50% each.

Table 3. Main chemical properties of two types of pomegranate juice: pH, total soluble solids (TSS, °Brix), titratable acidity (TA, g citric acid 100 mL⁻¹) and maturity index (MI = TSS/TA) ratio.

| Substrate | Pt | Pt-DRS | DRS |
|---|---------------|----------------|---------------|
| Aril juice | | | |
| pH | 4.67 ± 0.25a | 5.41 ± 0.31a | 4.98 ± 0.09a |
| TSS | 14.60 ± 0.15a | 15.83 ± 0.64ab | 16.40 ± 0.25b |
| TA | 3.18 ± 0.13a | 3.20 ± 0.11a | 3.13 ± 0.14a |
| MI | 45.99 ± 1.60a | 49.43 ± 0.80a | 52.67 ± 2.44a |
| Aril juice plus carpellary membranes | | | |
| pH | 4.75 ± 0.19a | 4.85 ± 0.17a | 4.35 ± 0.15a |
| TSS | 14.70 ± 0.1a | 15.00 ± 0.05ab | 16.23 ± 0.49b |
| TA | 3.94 ± 0.16a | 3.53 ± 0.21a | 4.19 ± 0.18a |
| MI | 38.23 ± 1.49a | 42.01 ± 2.68a | 38.91 ± 2.00a |

Different letters next to a value in within rows indicate significant differences among treatments according to Tukey's HSD test ($p < 0.05$). Pt: Peat 100% (control); DRS: Dredged remediated sediments 100%; (Pt-DRS) mixture 50% each.

differences were established among substrate treatments with respect to the total soluble solids content; DRS treatment yielded the highest value (>16° Brix) while peat (Pt) showed the lowest (<15° Brix).

Effect of the substrates on antioxidant activity and total phenols of 'Purple Queen' pomegranate

The antioxidant activity and total phenol content (TPC) of pomegranate juices are shown in Table 4. To characterize the different mechanisms naturally involved in the antioxidant activity, three antioxidant assay methods (ABTS, DPPH, and FRAP) were performed. The antioxidant activity of pomegranate juices was not affected by the type of growing substrate. Aril juice showed lower antioxidant contents compared with the juice of arils plus carpellary membranes. Aril juices antioxidant activity ranged as follows: ABTS methodology yielded values from 1.27 (DRS) to 1.40 (Pt-DRS); the DPPH one showed values ranging from 0.79 (Pt) to 0.88 (Pt-DRS), and FRAP methodology yielded values from 39.45 (Pt) to 40.92 (DRS). However, regard to aril juice plus carpellary membranes antioxidant activity ranged from 2.20 (Pt-DRS) to 2.43 (Pt); from 1.32 (Pt-DRS) to 1.46 for DRS, and from 41.46 (DRS) to 41.47 (Pt) for ABTS, DPPH and FRAP methodologies, respectively.

Likewise the results for antioxidant activity, the type of substrate did not affect the TPC either. Pomegranate juices of only arils showed lower TPC contents than aril juices plus carpellary membranes (88.97–101.31 mg GAE 100 g⁻¹dw among vs. 259.78–316.62 mg GAE 100 g⁻¹dw, respectively). Neither aril juice not aril juice plus carpellary membranes showed significant differences among their treatments.

Table 4. Antioxidant properties of two type pomegranate juices using different methodologies: ABTS, DPPH and FRAP assays (mM Trolox dw) and total phenols content (mg GAE 100 g⁻¹ dw).

| | ABTS | DPPH | FRAP | TOTAL PHENOLS |
|---|--------------|--------------|---------------|-----------------|
| Aril juice | | | | |
| Pt | 1.29 ± 0.08a | 0.79 ± 0.03a | 39.45 ± 1.93a | 88.97 ± 8.23a |
| Pt-DRS | 1.40 ± 0.04a | 0.88 ± 0.01a | 40.47 ± 1.05a | 95.72 ± 2.31a |
| DRS | 1.27 ± 0.05a | 0.82 ± 0.03a | 40.92 ± 2.07a | 101.31 ± 8.96a |
| Aril juice plus carpellary membranes | | | | |
| Pt | 2.43 ± 0.05a | 1.39 ± 0.03a | 41.47 ± 0.78a | 316.62 ± 26.36a |
| Pt-DRS | 2.20 ± 0.03a | 1.32 ± 0.04a | 41.54 ± 0.79a | 259.78 ± 20.38a |
| DRS | 2.44 ± 0.14a | 1.46 ± 0.06a | 41.46 ± 0.74a | 295.21 ± 19.35a |

Different letters next to a value in within columns indicate significant differences among treatments according to Tukey's HSD test ($p < 0.05$). Pt: Peat 100% (control); DRS: Dredged remediated sediments 100%; (Pt-DRS) mixture 50% each.

Effect of the substrates on ellagic acid (EA) and punicalagin isomers of 'Purple Queen' pomegranate

The content of EA and total punicalagins are given in Figures 1 and 2, respectively. Regarding the EA content, the highest value was obtained for pomegranate juice of arils with the substrate Pt-DRS, followed by substrate Pt and DRS. On the other hand, the EA in pomegranate juice of arils plus carpellary membranes were not affected by the type of substrate used (Figure 1). With regard to total punicalagins contents, the results presented in Figure 2, showed that growing substrates did not affect punicalagins contents of pomegranate aril juice plus carpellary membranes. Conversely, arils juice punicalagins content was definitely affected by the type of growing substrate, showing the Pt one the highest value at all (Figure 2).

Effect of the substrate on organic acids and sugar contents of 'Purple Queen' pomegranate

Table 5 showed the organic acid and sugar contents obtained from the two pomegranate juices analysed. Three organic acids (oxalic, citric and quinic acids) and two sugars (glucose and fructose)

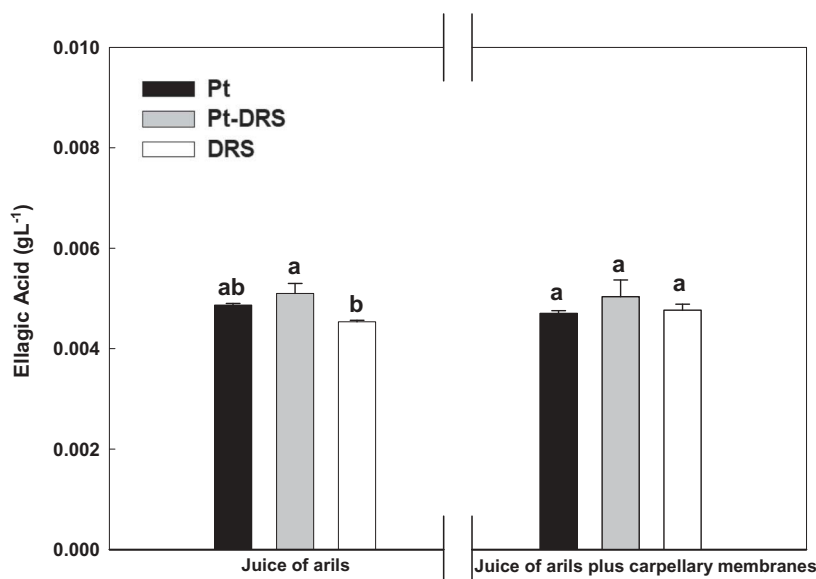


Figure 1. Ellagic acid in two types of pomegranate juices for each substrate, Pt (black bars), Pt-DRS (gray bars) and DRS (white bars). Different letters on top of bars indicate significant differences according to Tukey's HSD test ($p < 0.05$).

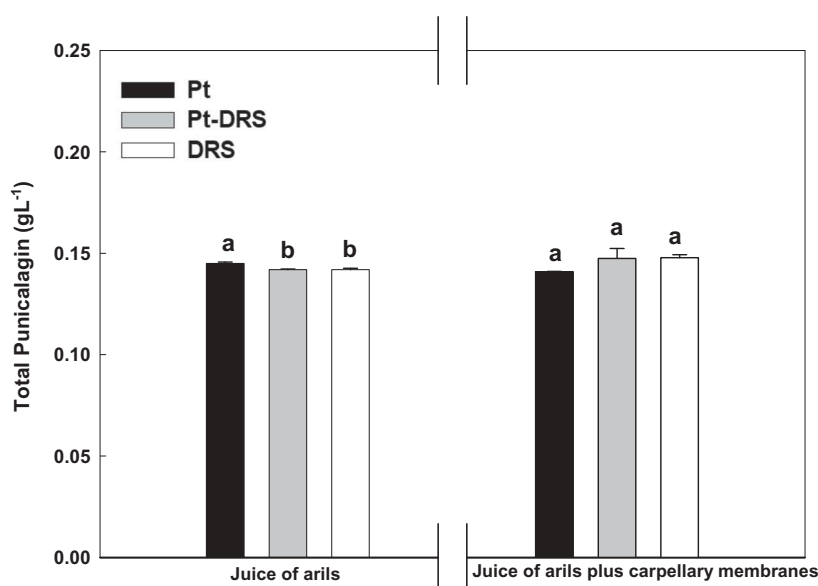


Figure 2. Total punicalagins ($\alpha + \beta$) in two types of pomegranate juices for each substrate, Pt (black bars), Pt-DRS (gray bars) and DRS (white bars). Different letters on top of bars indicate significant differences according to Tukey's HSD test ($p < 0.05$).

Table 5. Organic acids and sugar content of two type of pomegranate juices ($\text{g } 100 \text{ mL}^{-1}$).

| | Acids | | | Sugars | |
|---|--------------------|-------------------|-------------------|---------------------|---------------------|
| | Oxalic | Citric | Quinic | Glucose | Fructose |
| Aril juice | | | | | |
| Pt | 0.08 ± 0.00 ab | 0.09 ± 0.00 a | 0.67 ± 0.11 a | 10.51 ± 0.17 a | 12.79 ± 0.35 a |
| Pt-DRS | 0.08 ± 0.01 a | 0.09 ± 0.00 a | 0.74 ± 0.05 a | 10.58 ± 0.45 ab | 12.73 ± 0.36 ab |
| DRS | 0.09 ± 0.00 b | 0.09 ± 0.01 a | 0.42 ± 0.03 b | 10.92 ± 0.21 b | 13.73 ± 0.23 b |
| Aril juice plus carpellary membranes | | | | | |
| Pt | 0.08 ± 0.00 ab | 0.09 ± 0.00 a | 0.75 ± 0.05 a | 10.73 ± 0.31 a | 12.76 ± 0.33 ab |
| Pt-DRS | 0.09 ± 0.01 a | 0.10 ± 0.01 a | 0.64 ± 0.05 b | 10.43 ± 0.45 a | 12.72 ± 0.36 a |
| DRS | 0.08 ± 0.00 b | 0.09 ± 0.01 a | 0.52 ± 0.09 b | 10.69 ± 0.32 b | 13.24 ± 0.58 b |

Different letters next to a value within columns indicate significant differences among treatments according to Tukey's HSD test ($p < 0.05$). Pt: Peat 100% (control); DRS: Dredged remediated sediments 100%; (Pt-DRS) mixture 50% each.

were identified and quantified in both types of pomegranate juices. Quinic acid was the main organic acid present in both types of juices and for all substrates assayed, being the Pt the highest yielder in aril juice plus carpellary membranes (0.75 mg g^{-1}). The DRS substrate showed the lowest values of quinic acid in both pomegranate juices. Citric acid content was not influenced by the nature of the growing substrate. With reference to sugar content, the results reported in Table 5 showed the fructose as the main sugar, followed by glucose in both types of pomegranate juices. The DRS substrate yielded the highest values of fructose in both pomegranate juices (13.24 and $13.73 \text{ g } 100 \text{ mL}^{-1}$). Glucose content followed the same tendency for pomegranate aril juice.

Discussion

As far as we know, there are no published studies regarding the potential effect of growing fruit trees on these alternative types of substrates. The substrate DRS induced the lowest fruit weight, production per tree and number of fruits per tree, compared with the other 2 substrates assayed. The Pt-DRS (50% mixture) one had no effect on the evaluated parameters (Table 1). These

differences may be due to the composition of the substrate. The sediment with a bulk density 3–4 times greater than peat caused aeration problems because of its poor porosity, and pauper root systems induce therefore smaller plants and fruits (Zou et al. 2001). The average fruit weights of 'Purple Queen' pomegranates cultivated on these substrates are slightly lower than those presented by the Spanish cultivars (251.0 g to 421.1 g) cultivated under commercial farm conditions (Martínez et al. 2006). These differences could be due to: (i) different pomegranate cultivars, and (ii) different soil and climates. Several studies indicate that the pomegranate tree under stress responds by decreasing the production, number of fruits per tree and average fruit weight. Galindo et al. (2014) reported that deficit irrigation in pomegranate tree led to a significant decrease in total fruit yield, a decrease in the number of total fruits per tree and mean fruit weights.

On the other hand, titratable acidity (TA) and maturity index (MI) were not affected by the type of substrate used (Table 3). The values of TA and MI of cv. 'Purple Queen' grown on these substrates were similar to those reported by Calín-Sánchez et al. (2013) and Hernández et al. (1999) and (2014) grown in commercial farms. Palencia et al. (2016) studied strawberry cultivation on different substrates: cocopeat, perlite, sand, agrotexile, coir fibre and rock wool. These authors also concluded that the nature of substrates did not affect TA and MI. Nevertheless, total soluble solids content (TSS) was affected by the type of substrate used, being the highest values obtained for Peat-DRS (>15°Brix) and DRS (>16°Brix) substrates. These results completely agreed with those reported by Melgarejo et al. (2017), who reported that the same growing substrates also induced higher soluble solids contents in strawberries. This may be due, as mentioned before, to the apparent density of the sediment, which caused a stress on the plant, and increasing then the accumulation of dry matter in the fruit. Regarding this parameter, there is no tacit agreement among researchers, and many reports frequently show contradictory results. For example, Tzortzakis and Economakis (2008) reported that the growing substrate affects the TSS in tomato, while Recamales et al. (2007), Marinou et al. (2013) and Palencia et al. (2016) did not observe any differences in TSS contents depending on the type of substrate used for strawberry. The values of TSS obtained for cv. 'Purple Queen' were similar to those reported by Calín-Sánchez et al. (2011) and Hernández et al. (1999), (2014) for commercial pomegranate cultivation. Thus, the TSS in fruits can be influenced by several factors such as genotype and substrate nature.

The edible part of the fruit is the arils, which can be freshly consumed or as processed products as mainly juice. Pomegranate juice is composed of antioxidative phenolics like punicalagins, hydrolyzable tannins, anthocyanins and ellagic acids (Gil et al. 2000). Numerous studies suggest that these phenolic compounds can be used for the prevention and treatment of diseases like cancer and chronic inflammation (Lansky and Newman 2007). The total phenols and antioxidant activity of pomegranate fruit, evaluated by three different analytical methods as ABTS, DPPH, and FRAP were not affected by the type of substrates (Table 4). Melgarejo et al. (2017) also concluded that the type of substrate did not affect strawberry total phenols and antioxidant activity. Total phenols contents from 'Purple Queen' cv. aril juices agreed with those reported by Galindo et al. (2014), who reported values of total phenols between 176 and 260 mg of GA 100 mL⁻¹. Likewise, the total phenols contents of arils juice plus carpellary membranes of cv. 'Purple Queen' agreed with those reported by Nuncio-Jáuregui et al. (2015) who reported values ranging from 777 to 1660 g GAE kg⁻¹. Li et al. (2006) reported that rind and carpellary membranes are a richer source of antioxidants than the edible arils. The pomegranate cv. *Purple Queen* showed antioxidant activity values that agreed with those previously published by other researchers dealing with other cv. as 'Mollar de Elche' and 'Wonderful'. For instance, Carbonell-Barrachina et al. (2012) reported values from 3.3–3.9 TEAC, whereas Gil et al. (2000) reported values of 18–20 TEAC for commercial juices from 'Wonderful' pomegranates. This wide range in the antioxidant activity can be attributed to the fact that changing climatic conditions and cultural factors may affect the nutritional composition of foods, including bioactive compounds (Scalzo et al. 2005).

The most abundant polyphenolic compound in pomegranates is punicalagin. Along with the ellagic acid, they are potent antioxidants and potential anticancer properties and also have anti-atherosclerotic

biological properties (Lu et al. 2008). The contents of ellagic acid are significantly lower than those of punicalagins in this cultivar (Figures 1 and 2). The punicalagins and ellagic acid concentrations obtained were lower than those presented by Nuncio-Jáuregui et al. (2015).

Fructose and glucose were the main sugars found in both pomegranate juices under study (Table 5). Besides, both types of pomegranates juices showed the highest values of fructose for the DRS substrate. The sugars concentrations obtained for cv. 'Purple Queen' were similar to those previously reported by others researchers; Carbonell-Barrachina et al. (2012) reported values of fructose ranging from 2.52 to 17.6 g 100 mL⁻¹ and from 2.42 to 13.8 g 100 mL⁻¹ for glucose. However, our sugar contents are higher than those obtained by Melgarejo et al. (2000), who reported fructose values ranging from 5.9 to 7.04 g 100 g⁻¹, and values of 6.14 g 100 g⁻¹ for glucose. These differences can be well attributed to different extraction methods, analyses, fruit cultivars and growing media. Likewise, Repo-Carrasco-Valencia and Arana (2017) indicated that the environmental stress affects fruit sugar concentrations. With respect to organic acids profile, quinic acid was the main acid present in both types of juices, and for all substrates assayed followed by citric acid. Nuncio-Jáuregui et al. (2015) also reported that fruit thinning in pomegranates led to major concentrations in citric and quinic acids. However, the current study showed organic acids contents lower than those reported by Nuncio-Jáuregui et al. (2015).

Conclusions

The current study is the first for pomegranate cultivation in Spain dealing with the use of dredged remediated sediments as alternative growing substrates for horticultural purpose. It can be clearly concluded that the different growing substrates tested did not affect at all the final fruit composition and quality of 'Purple Queen' pomegranates. The substrates showed suitable properties for their use in pomegranate production, although, Pt and Pt-DRS (50%) showed the best plant response. This sediment can be a good alternative growing media to cope with the progressive shortage of the traditional substrates. Consequently, the current study reveals the appropriateness of these dredged remediated sediments to cultivate pomegranates on them. Fruit nurseries can benefit from their suitability as an alternative growing media for producing pomegranate trees. Furthermore, this research could be very useful for the use of alternative substrates in the field of horticulture, crops, fruits and ornamental propagation.

Disclosure statement

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References

- Benzie IFF, Strain J. 1996. The ferric reducing ability of plasma (FRAP) as a measure of antioxidant power. The FRAP assay. *Anal Biochem.* 239(1):70–76. doi:10.1006/abio.1996.0292.
- Brand-Williams W, Cuvelier ME, Berset C. 1995. Use of free radical method to evaluate antioxidant activity. *Lebensm Wiss Technol.* 28(1):25–30. doi:10.1016/S0023-6438(95)80008-5.
- Calín-Sánchez A, Figiel A, Hernández F, Melgarejo P, Lech K, Carbonell-Barrachina AA. 2013. Chemical composition, antioxidant capacity, and sensory quality of pomegranate (*Punica granatum* L.) arils and rind as affected by drying method. *Food Bioprocess Tech.* 6:1644–1654. doi:10.1007/s11947-012-0790-0.
- Carbonell-Barrachina AA, Calín-Sánchez A, Bagatar B, Hernández F, Legua P, Martínez-Font R, Melgarejo P. 2012. Potential of Spanish sour-sweet pomegranates (cultivar C25) for the juice industry. *Food Sci Technol Int.* 18:129–138. doi:10.1177/1082013211414783.

- CEDEX. 2012. Clave 23-409-5-003 Guía para la caracterización y destino de los materiales de dragado de acuerdo a la ley 22/2011. Ministerio de Agricultura, Alimentación y Medio Ambiente. España
- Galindo A, Calín-Sánchez A, Collado-Gonzales J, Ondoño S, Hernández F, Torrecillas A, Carbonell-Barrachina AA. 2014. Phytochemical and quality attributes of pomegranate fruits for juice consumption as affected by ripening stage and deficit irrigation. *Sci Food Agric*. 94:2259–2265. doi:10.1002/jfsa.6551.
- Gil MI, Tomas-Barberán FA, Hess-Pierce B, Holcroft DM, Kader AA. 2000. Antioxidant activity of pomegranate juice and its relationship with phenolic composition and processing. *J Agric Food Chem*. 48:4581–4589.
- Hernández F, Legua P, Martínez R, Melgarejo P, Martínez JJ. 2014. Fruit quality characterization of seven pomegranate accessions (*Punica granatum* L.) grown in Southeast of Spain. *Sci Hortic*. 175:174–180. doi:10.1016/j.scienta.2014.05.035.
- Hernández F, Melgarejo P, Tomás-Barberán FA, Artés F. 1999. Evolution of juice anthocyanins during ripening of new selected pomegranate (*Punica granatum*) clones. *Eur Food Res Technol*. 210:39–42. doi:10.1007/s002170050529.
- Hernández F, Noguera-Artiaga L, Burló F, Wodjdylo A, Carbonell-Barrachina AA, Legua P. 2016. Physico-chemical, nutritional, and volatile composition and sensory profile of Spanish jujube (*Ziziphus jujuba* Mill.) fruits. *J Sci Food Agric*. 96(8):2682–2691. doi:10.1002/jfsa.7386.
- Lansky EP, Newman RA. 2007. *Punica granatum* (Pomegranate) and its potential for prevention and treatment of inflammation and cancer. *J Ethnopharmacol*. 109:177–206. doi:10.1016/j.jep.2006.09.006.
- Li Y, Guo CH, Yang J, Wei J, Xu J, Cheng S. 2006. Evaluation of antioxidant properties of pomegranate peel extract in comparison with pomegranate pulp extract. *Food Chem*. 96:254–260. doi:10.1016/j.foodchem.2005.02.033.
- Lu J, Ding K, Yuan Q. 2008. Determination of punicalagin isomers in pomegranate husk. *Chromatographia*. 68:303–306. doi:10.1365/s10337-008-0699-y.
- Marinou E, Chrysargyris N, Tzortzakis N. 2013. Use of sawdust, coco soil and pumice in hydroponically grown strawberry. *Plant Soil Environ*. 59(10):452–459. doi:10.17221/297/2013-PSE.
- Martínez JJ, Melgarejo P, Hernández F, Salazar DM, Martínez R. 2006. Seed characterisation of five new pomegranate (*Punica granatum* L.) varieties. *Sci Hortic*. 110:241–246. doi:10.1016/j.scienta.2006.07.018.
- Mattei P, D'Acqui LP, Nicese FP, Lazzerini G, Masciandaro G, Macci C, Doni S, Sarteschi F, Giagnoni L, Renella G. 2016. Use of phytoremediated sediments dredged in maritime port as plant nursery growing media. *J Environ Manage*. 186:225–232. doi:10.1016/j.jenvman.2016.05.069.
- Melgarejo P, Legua P, Pérez-Sarmiento F, Martínez-Font R, Martínez-Nicolás JJ, Hernández F. 2017. Effect of a new remediated substrate on fruit quality and bioactive compounds in two strawberry cultivars. *J Food Nutr Res*. 8(5):579–586.
- Melgarejo P, Salazar DM, Artés F. 2000. Organic acids and sugars composition of harvested pomegranate fruits. *Eur Food Res Technol*. 211:185–190. doi:10.1007/s002170050021.
- Nuncio-Jáuregui NP, Munera-Picazo S, Calín-Sánchez A, Wojdylo A, Hernández F, Carbonell-Barrachina AA. 2015. Bioactive compound composition of pomegranate fruits removed during thinning. *J Food Compos Anal*. 37:11–19. doi:10.1016/j.jfca.2014.06.015.
- Palencia P, Bordonaba JG, Martínez F, Terry LA. 2016. Investigating the effect of different soilless substrates on strawberry productivity and fruit composition. *Sci Hortic*. 203:12–19. doi:10.1016/j.scienta.2016.03.005.
- Re R, Pellegrini N, Proteggente A, Pannala A, Yang M, Rice-Evans C. 1999. Antioxidant activity applying an improved ABTS radical cation decolorization assay. *Free Radical Biol Med*. 26(10):1231–1237.
- Recamales AF, López-Medina J, Hernanz D. 2007. Physicochemical characteristics and mineral content of strawberries grown in soil and soilless system. *J Food Qual*. 30(5):837–853. doi:10.1111/j.1745-4557.2007.00154.x.
- Repo-Carrasco-Valencia R, Arana JV. 2017. Carbohydrates of kernels. *Pseudocereals*. New Jersey (EEUU): John Wiley & Sons Ltd.; p. 49–70.
- Scalzo J, Politi A, Pellegrini N, Mezzetti B, Battino M. 2005. Plant genotype affects total antioxidant capacity and phenolic contents in fruit. *Nutrition*. 21:207–213. doi:10.1016/j.nut.2004.03.025.
- Sigua GC, Holtkamp ML, Coleman SW. 2004. Assessing the efficacy of dredged materials from Lake Panasoffkee, Florida: implication to environment and agriculture. Part 1: soil and environmental quality aspect. *Environ Sci Pollut Res Int*. 11(5):321–326.
- Singleton VL, Orthofer R, Lamuela-Reventos RM. 1999. Analysis of total phenols and other oxidation substrates and antioxidants by means of Folin-Ciocalteu reagent. *Methods Enzymol*. 299:152–178.
- Tzortzakis NG, Economakis CD. 2008. Impacts of the substrate medium on tomato yield and fruit quality in soilless cultivation. *Hortic Sci*. 35(2):83–89. doi:10.17221/642-HORTSCI.
- Viñes M. 2011. Estudio de la legislación ambiental en licencias de dragado de puertos en Brasil y España. Estudio de caso. Trabajo fin de carrera. Universidad Politécnica de Valencia. España.
- Zou C, Penfold C, Sands R, Misra RK, Hudson I. 2001. Effects of soil air-filled porosity, matric potential and soil strength on primary root growth of radiata pine seedlings. *Plant Soil*. 236:105–115. doi:10.1023/A:1011994615014.