Scientia Horticulturae 128 (2011) 473–478

Contents lists available at ScienceDirect

Scientia Horticulturae

journal homepage: www.elsevier.com/locate/scihorti

Phytochemical and antioxidant properties of selected fig (*Ficus carica* L.) accessions from the eastern Mediterranean region of Turkey

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ARTICLE INFO

Article history:
Received 8 January 2011
Accepted 25 February 2011

Keywords:
Ficus carica
Fig
Phytochemical characters
Antioxidant capacity
Sugars
Fruit skin color

ABSTRACT

Fig has been a typical fruit component of the health-promoting Mediterranean diet for a very long time. Phytochemical characters and antioxidant capacity of green-, yellow-, brown-, purple-, and black-fruited fig (*Ficus carica* L.) accessions were investigated. In this study, total phenolics (TP), total anthocyanins (TA), fructose (FRUC), glucose (GLUC), sucrose (SUC), and variables (such as L*, a*, C*, and hue°) describing fruit skin colors were examined. Also, the antioxidant capacity (TAC) of fig fruits was determined by the ferric reducing antioxidant power (FRAP) assay. Antioxidant capacity was significantly correlated with the polyphenol and anthocyanin ($r = 0.74$ and 0.63, respectively) contents of fruits. Black fig accessions had the highest TAC (range of 7.9–16.1, mean 12.4 Fe^{2+} mmol/kg FW), TA (range of 32.3–356.0, mean 128.4 μg cy-3-rutinoside/g FW), and TP content (range of 69.1–220.0, mean 118.9 mg GAE/100 g FW). These black-fruited accessions had 2-fold greater TAC, 15-fold greater TA, and 2.5-fold greater TP than green and yellow fig accessions. However, the FRUC, GLUC, and SUC content of brown and purple fig accessions were higher than those of other color groups. The predominant sugars present were fructose (~56%) and glucose (~43%), as determined by HPLC.

1. Introduction

Ficus carica, a deciduous tree belonging to the Moraceae family, is one of the earliest cultivated fruit trees and an important crop world wide for both dry and fresh consumption. Seventy percent of the world's fig production of is grown in the countries of the Mediterranean coast. In these countries, figs are an important constituent of the Mediterranean diet, which is considered to be one of the healthiest and is associated with longevity (Trichopoulou et al., 2006).

Antioxidant compounds, such as phenolics, organic acids, vitamin E, and carotenoids scavenge free radicals, thus inhibiting the oxidative mechanisms that may lead to degenerative illnesses (du Toit et al., 2001; Silva et al., 2004). Phenolic compounds are common plant secondary metabolites which have not only physiological functions in plants but also positive effects for human health, because they can act as antioxidants. Phenolic compounds may serve this purpose by reducing or donating hydrogen to other compounds, scavenging free radicals, and quenching singlet oxygen (Merken and Beecher, 2000; Fattouch et al., 2007; Costa et al., 2009). Phenolic compounds are important components of the color, flavor, and aroma of fresh fruits, vegetables, and their products. Phenolic compounds may also, in addition to antioxidative roles, have antimutagenic or anticarcinogenic, anti-inflammatory, or antimicrobial activities (Eberhardt et al., 2000; Kim et al., 2000).

A major benefit of the Mediterranean diet is its high level of natural antioxidants, derived from vegetables and fruits, including figs, which contribute antioxidant vitamins (Solomon et al., 2006) and some of the highest polyphenols levels of commonly available fruits (Vinson, 1999) to the diet. Solomon et al. (2006) showed that the higher the polyphenol content, particularly anthocyanins, in fig fruit, the higher their antioxidant activity. Antioxidants from figs can protect plasma lipoproteins from oxidation and significantly elevate plasma antioxidant capacity for 4 h after consumption (Vinson et al., 2005). Figs are rich in minerals and also sugars (Vinson, 1999), predominantly fructose and glucose (Melgarejo et al., 2003; Genna et al., 2008).

Several phenolic and flavonoid compounds (Teixeira et al., 2006; Vaya and Mahmood, 2006; Del Caro and Piga, 2008; Veberic et al., 2008), in addition to polysaccharides (Yang et al., 2009) anthocyanins, phytosterols, and fatty acids have been characterized in fig fruits and branches of fig trees (Jeong and Lachance, 2001). Sugars and mineral salts have been characterized in fruits (Yahata and Nogata, 1999; Aljane et al., 2007). Antioxidant activity and anthocyanin content of leaf, pulp and peels (Solomon et al., 2006), and the metabolic profile of figs (Konyalıoğlu et al., 2005; Oliveira et al.,

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doi:10.1016/j.scienta.2011.02.023

2009) have been analyzed. However, phytochemical characters such as total phenols, total anthocyanins, total antioxidant capacity, and specific sugars have not been compared among numerous fig accessions.

This study was designed to characterize some of the phytochemical qualities of 76 selected fig accessions from the eastern Mediterranean region of Turkey. We determined the total anthocyanins, total phenolics, total antioxidant capacity, sugar composition, and color profile of these fig accessions. These descriptive data could be useful for the development of health-promoting fig cultivars.

2. Materials and methods

2.1. Plant material and fruit extraction

This study was conducted using 76 local Mediterranean fig accessions. Fruit samples from these accessions were collected during 2008 and 2009 in Hatay province, which is located in the eastern Mediterranean region of Turkey. The accessions 'Bardak'and 'Dolap' are White San Pedro-type figs, and the other accessions are Smyrna-type, or Common figs. Fruits were harvested at their fully mature stage in three replicates of 500 g in each.

For the phytochemical analyses, harvested fruit samples were frozen and stored at −20 °C until analyzed. Triplicate 500 g lots of fig fruits from each cultivar were homogenized in a blender at room temperature. All triplicates were screened for their total anthocyanins, total phenolic contents, and antioxidant capacity following a single extraction procedure (Beccaro et al., 2006). For this procedure, 10 g aliquots of each homogenate were transferred to polypropylene tubes and extracted with 25 mL of extraction buffer containing methanol, deionized water, and hydrochloric acid (357:17:1.4, v/v/v) for 1 h at room temperature.

2.2. Total phenolics (TP)

Total phenolic (TP) contents of each sample were measured according to Slinkard and Singleton (1977). To determine TP, 0.5 g of each extract was combined with Folin-Ciocalteu's phenol reagent and water 1:12 (v/v) and incubated for eight minutes at room temperature, followed by the addition of 10 mL of 15% (w/v) sodium carbonate. After 2 h, the absorbance of each was measured at 750 nm using a spectrophotometer (Shimadzu UV-1208, Japan). Values of TP were estimated by comparing the absorbance of each sample with a standard response curve generated using gallic acid. Results are expressed as mg gallic acid equivalents (GAE) on a fresh weight (FW) basis (mg GAE/100 g FW).

2.3. Total antioxidant capacity (TAC)

To determine total antioxidant capacity (TAC), FRAP, the ferric reducing antioxidant power method, was conducted according to Pellegrini et al. (2003). To conduct the assay, a 9 mL aliquot of FRAP reagent (a mixture of 25 mL acetate buffer, 2.5 mL TPTZ [2,4,6-tris(2-pyridyl)-1,3,5-triazine], and 2 mL ferric chloride (10:1:1.25, v/v/v)) was combined with 9 mL of methanolic fruit extract prepared by the protocol above. The samples were incubated at 37 °C for 30 min, and absorbance at 593 nm was determined on a spectrophotometer (Shimadzu UV-1208, Japan). To determine the antioxidant capacity of samples, absorbance values were compared with those obtained from standard curves of $FeSO_4 \times 7H_2O$ (10–100 μM). Antioxidant capacity values were expressed as Fe^{2+} equivalents mmol/kg fruit weight (FW).

2.4. Total anthocyanins (TA)

Total anthocyanin (TA) content was quantified according to the pH differential method (Cheng and Bren, 1991). Absorbance was measured at 520 and 700 nm in buffers at pH 1.0 and pH 4.5 where $A = (A_{520}-A_{700})_{pH\ 1.0} - (A_{520}-A_{700})_{pH\ 4.5}$ Results were expressed as μg cyanidin-3-rutinoside (molar extinction coefficient of 28.800 and molecular weight of 595.2) (Solomon et al., 2006) equivalents per g fresh weight of fruit.

2.5. Sugar composition

Fig fruit homogenates (10 g) were diluted with purified water (40 mL) to prepare solution for detection of individual sugars. The homogenate was centrifuged at 10,000 rpm for 10 min. Supernatants were filtered through Whatman No. 42 filter paper. Aliquots of two milliliters of filtered homogenate per tube were then combined with 6 ml of acetonitrile. These solutions were then filtered through 0.45 mm membrane filters (Millipore, USA) prior to high-performance liquid chromatography (HPLC) analysis. Mobile-phase solvents were degassed before use. All the samples and standards were injected three times each, and the mean values for all chromatography runs were used. The HPLC analyses were conducted using a Shimadzu HPLC system, with an LC-10AT pump and RID-10 A detector (Shimadzu, Japan).

Analysis of sugars was performed according to the method described by Camara et al. (1996) with some modifications. The separation was carried out on an EC 250/4 Nucleosil C18 carbohydrate column (250 mm–4.0 mm i.d.) (Macherey–Nagel, USA). The elution solvent used contained 75% acetonitrile and 25% deionized water. The column was operated at 30 °C with a flow rate of 1.8 mL min^{-1}. Sample injection volume was 20 μL.

2.6. Fruit skin color

Fruit skin color was measured using a colorimeter (Chroma Meter CR-300, Minolta Co., Osaka, Japan), standardized with calibration plate set CR-A47 against a white background. Color parameters were expressed as tristimulus colorimetric measurements, that is, L*, a*, C*, and hue°. Negative L values indicate darkness, and positive L* values indicate lightness. Negative a* values indicate green color, and positive a* values indicate red color. The chroma (C*) value, calculated as $C = (a^2 + b^2)^{1/2}$, indicates color intensity. Hue°, a parameter that has been shown to be effective in predicting visual color appearance, was calculated using the formula hue° = $\tan^{-1}$ (b/a), where 0° or 360° = red-purple, 90° = yellow, 180° = green, and 270° = blue (Zerbini and Polesollo, 1984). Skin color was measured at three random positions per fruit.

2.7. Statistical analysis

Data were analyzed using SAS software and procedures (SAS, 2005). Means and standard deviations were calculated using PROC TABULATE. Analysis of variance tables were constructed using Tukey's Honestly Significant Difference (HSD) method at $p = 0.01$. Correlation coefficients and their levels of significance were calculated using PROC CORR.

3. Results

All results were expressed on a fresh weight basis. Selected fig accessions in this study had high levels of TP, TA and TAC. The amount of TP ranged from 28.6 to 211.9 mg GAE/100 g FW, with an average of 51.8 mg GAE/100 g FW; the amount of TA ranged from 0.0 to 298.9 μg cy-3-rutinoside/g FW, with an average of 18.2 μg cy-3-rutinoside/g FW; and the amount of TAC ranged from 3.9 to