

Research Article

Antioxidants Activity and Color Evaluation of Date Fruit of Selected Cultivars Commercially Available in the United States

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Dates (*Phoenix dactylifera* L.) are nutrient-rich fruit consumed throughout the world, either directly or in several food products. Six commercially available date cultivars in the US were analyzed for total phenolics, antioxidant activity using ABTS, DPPH, FRAP, and ORAC assays, and instrumental color. Total phenolics content varied from 33 to 125 mg GAE/100 g dry weight, with the highest in Barni (Saudi Arabia). Antioxidant values as determined by the ABTS in Deglet Nour (Algeria), Deglet Nour (California), Deglet Nour (Tunisia), Shahia (Tunisia), Barni (Saudi Arabia), and Khudri (Saudi Arabia) were 1300, 1047, 796, 452, 776, and 341 $\mu\text{mol TE/g}$ dry weight, respectively. Antioxidative properties as measure by DPPH, FRAP, and ORAC varied from 3.27 to 3.54, 3.29 to 5.22, and 189 to 243 $\mu\text{mol TE/g}$ dry basis, respectively. Fruit and pulp color of Deglet Nour (Algeria) was lighter whereas pulp of Barni (Saudi Arabia) was the darkest. Antioxidant values varied with different techniques used and also followed a different pattern than that of phenolics content.

1. Introduction

The date palm (*Phoenix dactylifera* L.) is grown in over 30 countries. Nutrient-rich dates are relished for their sweet, succulent, and exotic flavor. In recent years, dates have found acceptance among consumers in North America and European countries. Beside fresh consumption, this fruit is also processed into a wide variety of value-added products, such as dry dates, date paste, date syrup, date juice concentrate, date jam, date butter, date bars, date chutney, date relish, and date pickles, whereas date oil and date coffee are some of the by-products produced from date seeds [1, 2].

Phytochemicals are naturally produced, nonnutritive, and bioactive compounds which are synthesized by plants for protection against external stresses and attack by pathogenic microorganisms [3]. Phytochemicals are reported to have various biological effects, such as antimutagenic, anticarcinogenic, antioxidant, antimicrobial, and anti-inflammatory [4].

These compounds can be divided into several classes—phenolics, alkaloids, steroids, terpenes, and saponins. Phenolic compounds are characterized as having potent antioxidants and free radical scavengers, which can act as hydrogen donors, reducing agents, metal chelators, and singlet oxygen quenchers [5]. Phenolic compounds are active antioxidants playing an important role in neutralization of free radicals and decomposition of peroxides [6].

Date fruit has been shown to possess strong antioxidant activity among twenty-eight fruits commonly consumed in China [6]. Studies on various date fruit cultivars demonstrated a linear relationship between antioxidant activity and phenolic content [7]. Dates serve as a good source of natural antioxidants and could potentially be considered as functional food or ingredient [8, 9]. Date fruit lowers the incidence of cancers, especially pancreatic cancer due to antitumor activity or antimutagenic properties, and boosts immune system [10–13]. Consumption of dates may also be

beneficial in glycemic and lipid control in diabetic patients [14–16].

The objective of this study was to evaluate the total phenolics, antioxidant activity, and instrumental color of six commercially available date fruit cultivars in the United States.

2. Materials and Methods

2.1. Materials. Six commercially available date fruits, that is, Deglet Nour cultivated in California (DNC), Deglet Nour imported from Tunisia (DNT), Deglet Nour imported from Algeria (DNA), Khudri imported from Saudi Arabia (KSA), Barni imported from Saudi Arabia (BSA), and Shahia imported from Tunisia (SHT), were purchased from a local source in Lansing, MI, USA. The moisture content of cultivars was variable: DNC (24.1%), DNT (26.4%), DNA (25.3%), KSA (23.78%), BSA (20.4%), and SHT (22.3%). Therefore, all results are reported on dry weight basis to overcome the variation due to moisture content.

2.2. Sample Preparation. Samples were deseeded and crushed in Waring blender (20 g of date in 40 mL of distilled water); then, 2.5 g of the sample were taken in centrifuge tubes to which 20 mL of 80% methanol was added. The samples were stirred for 1 h at 200 rpm in a water bath shaker, followed by centrifugation at 10,000 \times g for 10 min. Supernatants were collected and the pellet was reextracted twice with 10 mL 80% methanol by vortexing and centrifugation at 10,000 \times g for 5 min. Finally, all the supernatants were pooled and volume was made to 50 mL with 80% methanol.

2.3. Total Phenolics. Total phenolic contents were determined using the procedures described by Zieslin and Ben-Zaken [17]. Briefly, 0.5 mL of methanolic extract was mixed with 0.5 mL Folin-Ciocalteu reagent by manual shaking for 15–20 sec. After 3 min, 1 mL saturated sodium carbonate and 1 mL of distilled water were added. The reaction mixture was incubated in the dark at room temperature for 2 h and its absorbance was measured at 725 nm against deionized water using spectrophotometer. Results were expressed in mg gallic acid equiv. (GAE)/100 g dry weight (dw).

2.4. Antioxidant Capacity by DPPH. Analysis was carried out following the methods of Brand-Williams et al. [18]. The stock solution (24 mg DPPH/100 mL methanol) was diluted with methanol to obtain an absorbance of 1.1 at 515 nm using spectrophotometer (Milton Roy, Warminster, PA, USA). 0.6 mL of the sample extracts, blank, or Trolox solution as standard was allowed to react with 3 mL of the DPPH working solution for 20 min under dark conditions. Then, the absorbance was taken at 515 nm. Radical scavenging capacity was calculated from the absorbance of sample, blank, or Trolox solution as standard. The standard curve was prepared using Trolox as standard versus radical scavenging activity and the results were expressed in terms of micromole Trolox Equivalence (μ mol TE)/g (dw).

2.5. Antioxidant Capacity by ABTS. ABTS radical cation (ABTS^{•+}) was produced by reacting ABTS stock solution (7 mM) with potassium persulfate (2.45 mM) in the dark at room temperature for 12–16 h. The ABTS^{•+} Solution was diluted with methanol (80%) to an absorbance of 1.1 at 734 nm. Diluted ABTS^{•+} solution (3 mL) was added to 30 μ L of sample or methanol for blank or Trolox solution as standard and the absorbance reading was taken at 30°C at 1 min interval up to 6 min. The oxidation index at 6 min and then percent antioxidant activity were calculated. Antioxidant activity was plotted against the concentration of Trolox to get standard curve. The results of samples were computed as μ mol TE/g dw.

2.5.1. Ferric Reducing Antioxidant Power (FRAP). The ferric reducing ability of dates extract was measured calorimetrically according to the method developed by Benzie and Strain [19]. The stock solutions included 300 mM acetate buffer (3.1 g C₂H₃NaO₂·3H₂O and 16 mL C₂H₄O₂) pH 3.6, 10 mM TPTZ solution in 40 mM HCl, and 20 mM FeCl₃·6H₂O solution. The fresh experimental solution was prepared by mixing 2 mL acetate buffer, 2.5 mL TPTZ solution, and 2.5 mL FeCl₃ solution. A 3 mL FRAP reagent was taken in test tubes and 300 μ L of standard, blank, or sample was added. After 5 min, the absorbance was recorded at 593 nm. Standard curve was constructed using Trolox and the results were expressed in μ mol TE/100 g dw.

2.6. Oxygen Radical Absorbance Capacity (ORAC). The ORAC assay for extracted dates samples was conducted on FLx800 Fluorescence Microplate Reader and Gen5 Data Analysis Software (BioTek Instruments Inc., Winooski, VT, USA). Exterior wells were filled with 300 μ L water; then, 150 μ L diluted fluorescein solution was added to all the experimental wells. For blank wells, 25 μ L 75 mM sodium phosphate buffer (pH 7.4) was added. A 25 μ L of Trolox dilutions (6.25–100 μ M) was added to Trolox wells. A 25 μ L of the sample dilutions was added to sample wells. After incubation at 37°C for 30 min, 25 μ L AAPH was added to all the wells. The fluorescence was recorded for 3 h and results computed using software were expressed as μ mol TE/100 g dw color measurement.

Color of whole and crushed dates was measured by Hunter Color Meter (Hunter Associates Lab, Reston, VA, USA). Instrument was calibrated using standard black and white tiles. Samples were placed in the standard cup and color values were recorded as *L* (0, black; 100, white), *a* ($-a$, greenness; $+a$, redness), and *b* ($-b$, blueness; $+b$, yellowness). Eight readings of each sample were taken from different sides.

2.7. Statistical Analysis. The data from three replicates were statistically analyzed using analysis of variance (ANOVA) following K. A. Gomez and A. A. Gomez [20]. The treatment means were compared using the least significant difference (LSD) at the 5% level and were used to examine multiple comparisons between means according to Waller and Duncan [21]. All statistical analysis was performed using SAS software package, version 8.0 [22].

TABLE 1: Antioxidant activity of different date fruit cultivars as determined by ABTS, DPPH, FRAP, and ORAC assays.

Cultivars	Antioxidant activity ($\mu\text{mol Trolox equiv./g dry weight}$)			
	ABTS	DPPH	FRAP	ORAC
Deglet Nour (Algeria) (DNA)	1300.50 \pm 73.36 ^a	3.54 \pm 0.85 ^a	5.22 \pm 0.34 ^a	235.05 \pm 9.39 ^a
Deglet Nour (California) (DNC)	1047.34 \pm 61.20 ^b	3.33 \pm 0.86 ^a	3.80 \pm 0.77 ^b	189.11 \pm 14.61 ^c
Deglet Nour (Tunisia) (DNT)	795.74 \pm 59.36 ^c	3.50 \pm 0.88 ^a	4.65 \pm 0.83 ^{ab}	243.05 \pm 19.35 ^a
Shahia (Tunisia) (SHT)	451.80 \pm 32.19 ^d	3.27 \pm 0.81 ^a	3.70 \pm 0.47 ^b	202.49 \pm 17.74 ^c
Barni (Saudi Arabia) (BSA)	775.97 \pm 65.87 ^c	3.31 \pm 0.80 ^a	4.03 \pm 0.22 ^{ab}	224.28 \pm 17.77 ^b
Khudri (Saudi Arabia) (KSA)	341.38 \pm 71.79 ^d	3.35 \pm 0.78 ^a	3.29 \pm 0.41 ^{ab}	206 \pm 15.33 ^b

Data is mean of three replicates \pm standard error. Means with different letters in the same column are significantly different at $p \leq 0.05$.

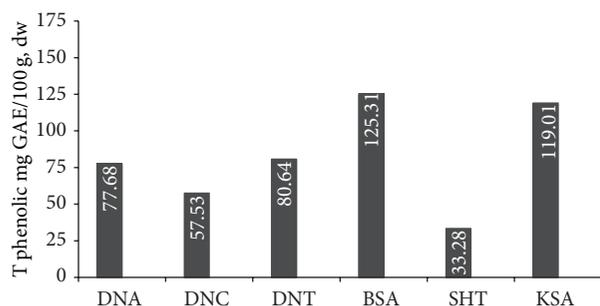


FIGURE 1: Total phenolic content of six date cultivars as gallic acid equivalent (GAE).

3. Results and Discussion

3.1. Total Phenolics. The total phenolics contents of six cultivars varied from 33 to 125 mg GAE/100 g dw (Figure 1). The highest total phenolic content was observed in BSA whereas the lowest content was found in SHT. Wide variation was observed in the total phenolics contents among the date cultivars. ANOVA of total phenolics revealed significant differences ($p \leq 0.05$) among the various date cultivars. Dates of the Deglet Nour cultivar (DNA, DNC, and DNT) from different countries showed significant difference ($p \leq 0.05$), indicating the possible effect of location, weather, and agricultural practices. Al-Turki et al. [23] found that the range of total phenolics content in fresh dates was 225.0 to 507.0 mg GAE/100 g, which was significantly higher than that observed in the present study. Similarly, Al-Farsi et al. [24] reported the total phenolics content of 172.0 to 246.0 mg GAE/100 g fresh weight in selected Omani date varieties. In another study, Al-Farsi et al. [9] reported the phenolics content to be in the range of 217.0 to 343.0 mg GAE/100 g fresh weight. Mohamed et al. [25] reported total polyphenols content of 35.82 and 99.34 mg GAE/100 g in six varieties of dates cultivated in Sudan. Wu et al. [26] reported that dates contain relatively higher amounts of total phenolics as compared to other fresh or dried fruits in a comparative study of total phenolics in different fruits. Present results indicated lower phenolics content as compared to the previous reports which might be due to the genetic makeup, agricultural practices, and analytical procedures.

3.2. Antioxidant Activity of Date Fruit. The results of the antioxidant assays showed that the antioxidant activity of date cultivars analyzed by ABTS, DPPH, FRAP, and ORAC varied to a large extent. Antioxidant values in terms of Trolox equivalent were low in DPPH assay than the highest observed by ABTS method. It indicated that components of the date cultivars reacted differently with chemicals involved in different antioxidant analytical protocols.

ABTS assay indicated high antioxidant activity that varied from 341 to 1300 $\mu\text{mol TE/g dw}$ (Table 1). DNA showed about four-time higher antioxidant activity than KSA. Dates cultivars had more than 600 $\mu\text{mol TE/g dw}$ activity except for KSA and SHT. Statistical analysis revealed that DNA had significantly higher antioxidant activity followed by DNC ($p \leq 0.05$). The difference between DNT and BSA cultivars was nonsignificant in antioxidant activity. Similar observations were noted between KSA and SHT cultivars. DPPH assay revealed the antioxidant values in the range of 3.27–3.54 $\mu\text{mol TE/g dw}$. Statistical analysis showed that the antioxidant values by DPPH method did not vary significantly ($p > 0.05$) among the six cultivars. It indicated that DPPH decolorizing reaction was not supported by date constituents and thus showed a little activity.

Antioxidant activity assayed by FRAP showed that values varied from 3.29 to 5.22 $\mu\text{mol TE/g dw}$. DNA cultivar also exhibited the highest activity while KSA showed the minimum activity. The general pattern of antioxidant activity was comparable to ABTS but values were quite low. Statistical analysis showed significant difference ($p \leq 0.05$) in FRAP values among the six cultivars.

The ORAC values of six date cultivars varied from 189 to 243 $\mu\text{mol TE/g}$ with higher activity in DNA and DNT. The lowest in ORAC values were observed in DNC and SHT. Statistical analysis showed that ORAC values varied significantly among the cultivars ($p \leq 0.05$).

The ORAC values were higher than DPPH and FRAP but lower than ABTS values. The variations in antioxidant activities of different date cultivars were expected due to variation in agroclimatic conditions, variety, and country of origin. In addition, temperature, relative humidity, maturity, and processing can have a significant impact on phytochemicals profile of the date fruit [8, 27, 28]. The antioxidant values measured by different techniques did not give similar results. ABTS and FRAP analysis indicated that DNA had the highest

TABLE 2: Hunter color values of whole dates and date paste from different cultivars.

Date cultivars	Whole fruit			Ground fruit (paste)		
	<i>L</i>	<i>a</i>	<i>b</i>	<i>L</i>	<i>a</i>	<i>b</i>
Deglet Nour (Algeria) (DNA)	19.35 ± 2.59 ^{abc}	5.04 ± 0.93 ^a	4.54 ± 0.62 ^a	42.13 ± 1.14 ^b	3.98 ± 0.49 ^c	9.71 ± 0.47 ^b
Deglet Nour (California) (DNC)	21.06 ± 0.85 ^a	4.28 ± 1.35 ^a	4.21 ± 0.45 ^{ab}	47.05 ± 1.37 ^a	3.89 ± 1.43 ^c	11.50 ± 3.51 ^b
Deglet Nour (Tunisia) (DNT)	18.40 ± 0.61 ^{bc}	4.35 ± 0.44 ^a	3.61 ± 0.43 ^b	37.58 ± 3.68 ^c	6.15 ± 1.66 ^{ab}	8.63 ± 1.68 ^b
Shahia (Tunisia) (SHT)	20.19 ± 0.34 ^{ab}	4.51 ± 1.34 ^a	3.68 ± 0.70 ^b	45.46 ± 1.04 ^{ab}	4.44 ± 0.20 ^{bc}	10.85 ± 0.33 ^b
Barni (Saudi Arabia) (BSA)	19.26 ± 1.36 ^c	1.80 ± 0.37 ^b	2.20 ± 0.35 ^c	37.56 ± 4.30 ^c	5.85 ± 2.14 ^{abc}	9.35 ± 2.83 ^b
Khudri (Saudi Arabia) (KSA)	17.81 ± 0.41 ^c	4.81 ± 0.81 ^a	2.58 ± 0.09 ^c	42.84 ± 1.44 ^b	6.86 ± 1.56 ^{bc}	15.23 ± 3.82 ^a

Data is mean of eight replicates ± standard error. Means with different letters in the same column are significantly different at $p \leq 0.05$.

antioxidant activity while ORAC analysis showed that DNT and DNA had the highest antioxidant activity.

The antioxidant activity data is not supported by the total phenolics content as reported by some researchers. Study also indicated that the composition of the date cultivars varied significantly. Moreover, dates participate in reaction involving antioxidant estimation differently in different techniques used.

Al-Turki et al. [23] reported that the antioxidant activity measured by ABTS was higher than that measured by DPPH. Their results support the present findings where the antioxidant activity values of DPPH were lower than the ABTS values.

3.3. Date Fruit and Paste Color. Color is the important parameter affecting the consumer acceptability. It is also an important quality parameter in commercial date fruits. The results of color measurement of whole fruit dates and date paste of different cultivars, in terms of Hunter *L*, *a*, and *b* values, are presented in Table 2. Measuring of whole date fruits color is useful for the comparison between different date cultivars and for quality control of processed date products in the international trade. In whole date fruits, Hunter *L* values varied in the range of 18.44 to 21.06 among the cultivars. Higher *L* values indicate "lighter" color; thus, KSA had dark color whereas DNC had light color. Statistical analysis showed a significant difference ($p \leq 0.05$) in *L* values of whole fruit. The Hunter color *L* values for date pastes showed a similar trend but were in the range of 37.57–47.05. The *L* values of the paste increased indicating that color became lighter. Among the cultivars BSA and DNT had dark color whereas DNC had lighter color.

The Hunter color *a* values, of the whole dates ranged from 1.8 to 5.04. Statistical analysis showed a significant ($p \leq 0.05$) difference of "*a*" value in whole date. Hunter values of paste varied from 3.9 to 6.9. None of the dates or pastes having Hunter values were in the negative (–) range, which reflected the absence of any greenish tint. Hunter *b* values varied from 2.20 to 4.53 in whole fruits and from 8.63 to 15.23 in ground paste of date. The Hunter color *b* values of dates were observed to be high for DNA whole fruit while being low for BSA. Similarly, KSA cultivars had the highest *a* value whereas DNT had the lowest. Measuring the color of date fruit paste is useful in comparing different dates. Overall, the difference in color is mainly due to the genetic variability [27].

Hasnaoui et al. [29] evaluated the fruit color for twenty-seven of date palm cultivars collected from Moroccan oases at Tamar stage (fully ripened) and found the *L* values in the range of 12.12 to 38.93, *a* values in the range of 1.35 to 15.29, and *b* in the range of 0.86 to 35.12.

4. Conclusion

Characterization of date fruit for antioxidant properties can expand this fruit's consumption, as consumers are increasingly looking for healthy food. Barni and Khudri cultivars had relatively higher phenolics content than other four cultivars. With respect to assessing antioxidant capacity, the ABTS method gave higher antioxidant values than other three methods used in this study. Deglet Nour from Algeria has higher antioxidant properties than others cultivars. Light colored dates are considered better; Deglet Nour from California showed a better color with higher *L* values. Based on the results of this study, it can be concluded that date cultivars had medium phenolic contents and significant antioxidant activity. However, relationship between phenolic contents and the antioxidant activity of the respective date cultivars could not be established conclusively. Our work adds expanded scientific information on antioxidant analysis of different cultivars. We used different methods to measure the antioxidants capacity of dates.

Conflict of Interests

The authors declare that there is no conflict of interests regarding the publication of this paper.

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