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Antioxidants in Date Fruits and the Extent of the Variability of the Total Phenolic Content: Review and Analysis

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Abstract

The date fruit is economically important agricultural commodity, as well as a staple food in many countries in the Arab world, North Africa, and the Middle East. Recent interest in its nutritional, health, and therapeutic attributes is manifested by the rise in scientific publications. Dates of various cultivars are widely publicized and highly ranked as rich sources of natural antioxidant constituents and antioxidant activity. Such publicity, justified or otherwise, is sometimes accompanied by misconceptions and claims of cultivar- and/or country-wise superiority. This chapter examines these claims using a dataset generated from scientific studies published over the last three decades focusing on the total phenolic (TP) content of three stages of date maturity, with emphasis on the last stage, Tamer. The dataset contains TP values (mg GAE/100 g DM) from 18 countries and 243 cultivars and included 583 entries. It only examines variability of TP values. Statistical analysis indicates a great variability of TP content, both within a particular cultivar and among different cultivars. Claims of cultivar- and country-wise superiority and very high ranking of date antioxidant activity are not substantiated. The chapter also discusses various causes of high variability and calls for a collaboration work to address the issue.

Keywords: antioxidant, antioxidant activity, date palm, dates, dried fruits, phenolic compounds, polyphenolics

1. Introduction

The date palm, the tree and the fruit alike, enjoys a high place in the hearts and minds of the people of the Arab region, in particular, and the Middle East in general and of the three major regional religions (Islam, Christianity, Judaism). A place where the mythological and the cognitive are highly intertwining and intersecting, culturally, religiously, and historically, in a clear indication of the depth of this tree's roots in the soul and civilization of this part of the world. The foundations on which this status is based may be lacking in validity and may be somehow exaggerated but cannot be ignored.

This tree, which enjoys a status of sanctity, due to many religious verses, conversations, and curses, was a staple food for the farmers in their ranches, the divers looking for pearls in the deep sea away from land for several months, and for the mobile

Bedouin in the deserts in the cold winter and in the high heat during their winter/summer traveling trips. With respect to common food consumption, the lives of people, in this area, were centered around few simple things, and their day may begin and end with eating dates supplemented with few additional foods such as milk, meat, and fish. The temporal and spatial presence of dates in the residences was overwhelming in the Arabian Peninsula and, archeologically, very well documented in many locales [1].

At present, global production statistics show increasing interest in dates as an economic commodity with a good financial return [2]. Scientifically, researchers have also increased their academic interest in studying different aspects of the date tree and its fruit using recent approaches and methodologies. The phytochemicals, antioxidant efficacies, and health of common dried fruits, including dates, have recently been reviewed [3]. The phenolic antioxidant properties and benefits in date fruits have recently been reviewed [4]. The biochemistry of the ripening process in dates as the main deriving source of metabolic variation has been recently reported [5].

Nutritionally, date fruits provide quick and high energy (~280–330 kcal/100 g) due to its high content of simple carbohydrates, mainly glucose, fructose, and sucrose [6]. They are also rich sources of fibers and potassium, among other nutrients. In recent years, there have been several reviews of the nutritional attributes of dates [6, 7]. The health and therapeutical attributes of dates have also been recently reviewed by [8–10]. These attributes include anticancer, anti-inflammatory, antimicrobial, antioxidant, antimutagenic, gastroprotective, hepatoprotective, immunostimulant, and nephroprotective activities. Of these, the antioxidant property appears to be of a high interest and, currently, is being explored at different levels using different methodological approaches including the metabolomics studies [11].

Because of this scientific activity, knowledge of the antioxidant properties of dates and the importance of dates as a good source of antioxidants has substantially increased. This knowledge, however, has been accompanied by claims and misconceptions, mostly are unsubstantiated and/or justified, rather, unfortunately confusing and questionable. Such claims include, but not limited to (1) claims of country-wise (or regional-wise) superiority of dates, (2) claims of antioxidant superiority of certain cultivars, and (3) claims of high ranking of dates among other dried fruits and natural products.

The main aim of this chapter is to review the current state of knowledge of the antioxidants in date fruits, with emphasis on dry stage (Tamer), and the issues pertinent to the huge quantitative discrepancy of total phenolic (TP) content in an attempt to find answers to questions that directly address the abovementioned issues/claims. This chapter singles out TP content as the only antioxidant parameter understudy due to space and time limitation; hence the analysis is a preliminary. Further analysis of TP content in relation to other related parameters and factors is highly needed and is to be seen.

2. The date palm

2.1 The tree and cultivars

The date palm (*Phoenix dactylifera* L.) is among the first domesticated perennial plants with some fossil records showing that the tree has existed for about 50 million years [12]. It has been growing in the Arabian Peninsula, the Middle East, North Africa, and South Asia for about 5000 years [12]. Generally, it is characterized by its ability to tolerate relatively high temperatures, salinity, and drought [13]. To produce dates, the date palm tree has a strict requirement to relatively a hot and lengthily summer. The date palm belongs to the family Arecaceae and is a dioecious

flowering plant which can live for 100 years [13, 14]. Usually, artificial pollination of the date palm starts late February to early March [15]. Fruit development and ripening stages are cultivar-dependent (see next section). It is a common practice to classify cultivars into early, mid, and late cultivars where fruit maturation takes place in June, August, and late September, respectively. Worldwide, the number of date cultivars is large (2000–3000) [16], and at the country level, the number may range between 300 and 600 cultivars in the known major country producers [16]. Within each country, the number of cultivars with significant commercial importance is very limited (10–30). Generally, names of cultivars are, country (or regional)-dependent, with some names of cultivars being traded widely. The same cultivar may have different names in different countries.

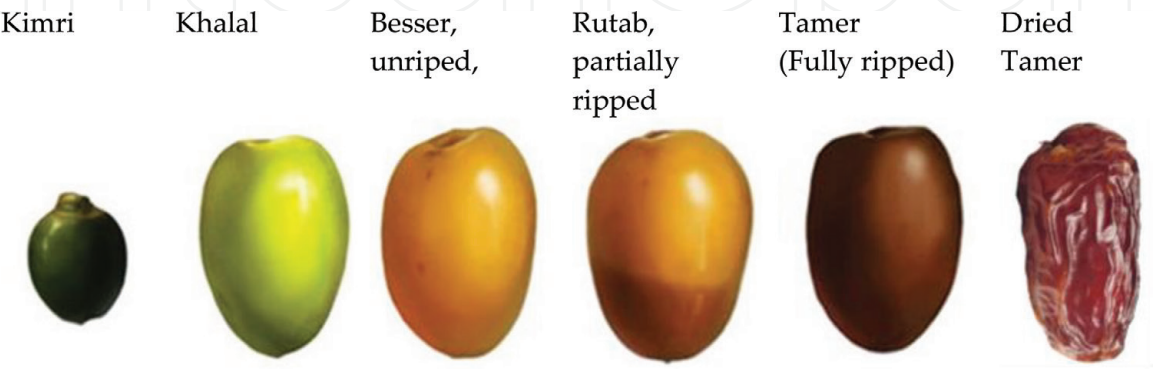


Figure 1.
Stages of maturation of date palm fruits. Re-drawn from Al-Mssallem et al. [17].

Parameter	Developmental (maturation) stages of date fruits				
	Hababouk	Kimri	Khalal	Rutab	Tamar
Moisture (%)		85	50–60	35–45	20–25
Duration (weeks)	5 weeks	9–14	3–5	Varies	Stable
Maturation and growth rate	Very early slow	Fast fruit enlargement	Full size, crunchy	Ripe, soft	Ripe, sun-dried
Color	Green	Green, unripe	Yellow or red	Varies	Golden grown to dark blue
Texture		Hard	Hard	Soft	Soft, semihard, hard
Edibility	Inedible	Inedible	Inedible with exceptions	Edible	Edible
TN (%) ^a			100	58–64	28–36
TP (%) ^a			100	45–57	40–51 ^b
TF (%) ^a			100	40–88	22–70 ^b
FRAP (%) ^a			100	30–72	25–55 ^b
DPPH (%) ^a			100	55–76	39–60 ^b

TN = tannins, TP = total phenolics, TF = total flavonoids, CT = condensed tannins^a All values are expressed as percent since data points of the included parameters vary greatly, with values of Khalal stage taken as 100%. These percentages were calculated from a limited number of published studies.
^bFew published studies reported rather higher values in Tamer than Rutab.

Table 1.
Chemical and physical characteristics of the developmental (maturation) stages of date fruits in relation to antioxidant properties.

2.2 The date palm fruit

The date palm fruit, the date, is a berry or drupe consisting of a single inedible seed (pit) surrounded by a fibrous, parchment-like endocarp, a fleshy mesocarp, and the fruit skin (pericarp) [13, 15]. Usually, the fruit is oblong, though great variation exists in shape, size, color, as well as in quality and texture. Weight of dates ranges between 4 and 36 g; length, 2–7.5 cm; diameter, 1.3–4 cm; and volume, 5–19 cm. The edible part of the date represents 85–92% of the total fruit weight. The development of the date passes through five distinct morphological stages, widely known by their Arabic names: Hababouk (Habanbo), Kimri, Khalal (Besser), Rutab, and Tamer (**Figure 1**, **Table 1**). At full size, Khalal, the color of dates is either yellow or red, with different shading and hues, with the yellow-colored dates representing the majority (> 80%). Date maturation starts 10–15 weeks after pollination and takes place over an extended period lasting for about 6–8 months. This process requires developmental competence and high heat. Dates of some cultivars can be eaten starting from the Khalal stage; others can only be eaten during or after attaining some degree of ripening. Antioxidant properties of dates are directly and indirectly influenced by the physiological aspects and developmental stages (**Table 1**).

3. Antioxidant in dates

3.1 The issue of large variability of total phenolic content

Over the past two decades, several studies on antioxidant properties of date fruits have been published. These studies have contributed to increase our knowledge and understanding of these properties and the importance of dates as a food and its medical, therapeutical, and health virtues. However, the foresight of these studies finds great variations of levels and values of studied parameters of antioxidant of these results, which confuses the researcher/reader, leading to some skepticism and casting doubt.

To illustrate the magnitude of the problem and the importance of attempting to overcome this wide disparity, I will review here what Hammouda et al. [17] recently noted while comparing their findings with results of a previous published. Hammouda et al. [18] estimated the TP of two date cultivars from three geographical origins using an HPLC methodology and reported an average of 154 mg/whole fruit (or 126.3 mg/edible part of the fruit, average weight of a fruit was 10.2 g). When they compared their findings with values reported on the widely cited study of Al-Farsi and Lee [6], which reported that TP ranged between 194 and 240 mg/100 g of fruit, corresponding to ~19–24 mg per fruit, they concluded that (a quote): “We consider that our estimation better reflects the real concentration of total polyphenols in dates as phloroglucinolysis–HPLC is the only quantification method that takes into account the nonextractable PCs which represent the major part of polyphenols in dates and which are not quantified when a colorimetric assay is performed on a methanol extract.”

Many researchers have faced a similar situation and may have reached similar conclusions, as evidenced by many comparative studies. Recently, Mishra et al. [19] reviewed and analyzed the abnormalities associated with reporting the antioxidant activity using DPPH methods. Whether one agrees or disagrees with the above statement made by [18] is not the issue here. The issue is that whether the huge disparity of experimentally obtained and numerically reported values of antioxidants and antioxidant activity on date fruits really reflect the phenomena of natural variability or a manifestation of otherwise.

In the following section, the total phenolic (TP) content reported in the literatures by many groups will be statistically analyzed. Selection of the TP content to illustrate the extent of variability and diversity of the antioxidant in dates is largely based on its commonality and convenience. The majority of the published studies dealing with antioxidants and antioxidant activity of dates (other plant based produces as well) reports TP as the prime parameter. Furthermore, TP content is highly correlated with many assays used to estimate antioxidant activity (ABTS, DPPH, FRAP, etc.).

Estimation of TP is usually performed using the well-established Folin–Ciocalteu (FC) colorimetric method or one of its variants with gallic acid (GA) being widely used as a standard for calibration [19]. Results are usually expressed as mg GAE/g or 100 g. The FC method is based on electron transfer reactions between the phenolic antioxidant(s) and the FC reagent. It is not specific for TP determination and is prone to interfering compounds presented in the sample leading to biased estimation. Reducing sugars which are present in high concentration in dates and ascorbic acid, which is present in substantial level at some developmental stages, are examples of these interfering compounds [20]. Most of the published studies reporting TP in dates did not adequately address this issue.

3.2 Sources and preparation of dataset

Data used in this analytical review are of secondary type. They include the total phenolic (TP) content of 243 cultivars from 18 countries, covering the three potentially edible maturity stages, Khalal, Rutab, and Tamer (**Figure 1**). The selection of these datasets was based solely on relatedness and availability at the time of the preparation of this review. Values of TP were either copied and pasted from the published sources or extracted from graphs by using the online site WebPlotDigitizer [21]. A partial list of selected studies with some parameters of antioxidants and antioxidant activity is given in **Table 2**. **Table 3** lists countries of the recruited studies.

Most published values of TF were reported as sampled (i.e., the fresh or dry weight of the edible portion of the date fruits). To make them comparable and meaningful, these weight values were recalculated and presented on a dry matter (DM) basis. When given, moisture content was used to calculate the moisture fraction, hence the DM. In the absence of moisture content, the following general moisture contents were used: Khalal (66%), Rutab (43%), and Tamer (22%). When oven-dried or lyophilized samples were indicated, the moisture content used value was 15%. Samples of date syrup and wasted dates were also included since they usually possess similar TP content.

3.3 Data cleaning

The name of the same date cultivar in different countries may have different spellings. An example of this is the cultivar Barhi which has the following synonymous: Berhi = Burhi = Barhee = Barhy; Deglet Nour = Deglet Noor; Sokary = Sukkari = Sukari; and Sofry = Sufry = Suffry. It was very essential for the analysis to designate a single spelling for the same cultivar.

Outliers were statistically detected and removed from analysis. Removing of statistically detected outliers, at this stage of analysis, was based on convenience, simplifying the analysis, to examine their effects on estimates. Their analysis requires a more rigorous methodology, and perhaps these extreme values may represent a reality.

3.4 Statistical analysis

Excel (Microsoft) and SPSS (IBM, version 23) were used for statistical analysis which included estimates of central tendency and variability.

Country	Author (no. cultivar)	Parameter		Stg.	Mean (SD, range)	References
Algeria	Benmeddour et al., 2013 (10)	TP (mg GAE/100 g DW)		T	493.15 (294.90; 226–954)	[22]
		TF T(mg QE/100 g DW)		T	102.7 (94.45, 15.2–299)	
		CT (mg CE/100 g DW)		T	243.75 (139.62, 82.8–525.1)	
	Mansouri et al., 2005 (7)	TP (mg GAE/100 g FW)		T	4.70 (1.996, 2.5–8.4)	[23]
Egypt	Farag et al., 2014 (21)	TP (mg GAE/ 100 g DW)	(Low)	T	273.57 (40.53, 233–349)	[24]
			(Med)	T	449.57 (115.42, 437–622)	
			(High)	T	1332.83 (271.06, 1100–1898)	
			(Overall)	T	638.48 (473.12, 233–1898)	
Iran	Biglari et al., 2008 (8)	TP (mg GAE/100 g DW)		R	21.61 (45.27, 2.4–141.4)	[25]
		TF (mg CE/100 g DW)		R	12.57 (26.18, 1.6–81.2)	
		FRAP (umol/100 g DW)		R	65.46 (121.79, 11.6–387.3)	
		TEAC (umol TE/100 g DW)		R	97.99 (152.43, 22.8–500.3)	
	Mortazavi et al., 2015 (9)	TP (mg GAE/100 g FW)		K	126.04 (61.58, 57.8–262.8)	[26]
				R	57.41 (20.47, 23.5–94.1)	
				T	76.04 (18.87, 38.2–103.9)	
KSA	Farag et al., 2016 (18)	TP (mg GAE/100 g DW)		T	439.39 (559.37, 93–255)	[27]
	Hamad et al., 2015 (12)	TP (mg GAE/100 g DW)		T	17.52 (3.62, 10.5–22.1)	[11]
		TF (mg CE/100 g DW)		T	2.12 (0.51, 1.2–2.8)	
	Al-Turki et al., 2010 (5)	TP (mg GAE/100 g FW)		T	418.12 (55.18, 315.68–508.01)	[28]
	Hatem et al., 2018 (4)	TP (mg GAE/100 g FW)		K	4.92 (1.00, 3.26–5.94)	[29]
				R	6.95 (2.63, 2.49–9.23)	
				T	6.15 (1.23, 4.25–7.65)	
		DPPH (IC50: mg/ml)		K	4.62 (0.36, 4.1–5.1)	
				R	2.96 (1.53, 2.0–5.4)	
				T	4.0 (0.60, 3.4–5.0)	

Country	Author (no. cultivar)	Parameter	Stg.	Mean (SD, range)	References
Morocco	Taouda et al., 2014 (13)	TP (mg GAE/100 g DW)	T	2.68 (0.86, 1.5–4.5)	[30]
		TF (mg/100 g DW)	T	0.066 (0.094, 0.01–0.38)	
		DPPH (IC50: ug/ml)	T	1743 (6.71, 7.5–33)	
	Bouhlali et al., 2017 (8)	TP (mg GAE/100 g DW)	T	466.26 (66.54, 331.9–537.1)	[31]
		TF (mg RE/100 g DW)	T	124.12 (53.06, 68.9–208.53)	
		CT (mg CE/100 g DW)	T	75.25 (11.86, 57.6–92.1)	
		FRAP (umol TE/100 g DW)	T	640.96 (157.8, 406.6–860.9)	
		DPPH (IC50: mg/ml)	T	3.94 (1.31, 2.1–6.2)	
		ABTS (umol TE/100 g DW)	T	621.54 (124.8, 383.9–846.9)	
Oman	Al-Farsi et al., 2005 (3)	TP (mg GAE/100 g FW)	T	246.67 (80.45, 134–343)	[32]
	Singh et al., 2013 (6)	TP (mg GAE/100 g DM)	T	172.5 (56.84, 81–235)	[33]
Pakistan	Nazeem et al., 2011 (21)	TP (mg GAE/100 g DW)	T	216.22 (45.78, 141.9–297.0)	[34]
	Haider et al., 2013 (10)	TP (mg GAE/100 g DW)	K	459.92 (62.89, 349–571.3)	[35]
			R	211.076 (55.08, 102.8–265.3)	
			T	120.48 (47.46, 50.2–184.1)	
		DPPH (IC50: mg/ml)	K	0.59 (0.13, 0.47–0.86)	
			R	1.06 (0.32, 0.75–0.98)	
			T	1.86 (0.55, 1.4–2.9)	
Tunisia	El-Arem et al., 2012, (4)	TP (mg GAE/100 g FW)	K	482.27 (93.14, 303.17–602.28)	[36] [37]
			R	362.93 (49.57, 278.8–435.4)	
			T	269.96 (60.17, 182.2–375.5)	
	El-Arem et al., 2013 (5)	TF (mg CE/100 g DW)	K	232.0 (53.91, 109.79–307.59)	
			R	144.49 (45.30, 79.6–231.0)	
			T	94.81 (24.77, 52.8–140.5)	
		CT (mg CE/100 g DW)	K	189.82 (68.66, 86.0–276.8)	
			R	121.1 (46.57, 65.3–198.2)	
			T	81.13 (22.24, 40.1–110.5)	

Country	Author (no. cultivar)	Parameter	Stg.	Mean (SD, range)	References
Tunisia	El-Arem et al., 2017 (3)	ABTS (mmol TE/100 g FW)	K	1.36 (0.03, 1.3–1.4)	[38]
			R	1.26 (0.07, 1.2–1.4)	
			T	1.13 (0.10, 1.0–1.3)	
		DPPH (AE = 1/EC50)	K	3.54 (0.63, 2.7–4.1)	
			R	2.54 (0.36, 2.1–2.7)	
			T	1.76 (0.42, 1.4–2.4)	
USA	Al-Turki et al., 2010 (10)	TP (mg GAE/100 g FW)	T	318.19 (61.75, 22.7–491.3)	[28]
Information are alphabetically arranged based on country.					

Table 2.
Mean, SD, and range of selected parameters of antioxidant constituency (TP, TF, CT) and antioxidant activity (ABTS, DPPH, FRAP) extracted from selected published studies demonstrating the large reported variability.

Country	References	No.	Country	References	No.
Algeria	[22, 23, 39, 40]	4	Morocco	[30, 31, 65–68]	5
Bahrain	[41, 42]	2	Oman	[32, 33, 69, 70]	4
Egypt	[24, 43–45]	4	Palestine	[74]	1
Iran	[25, 26, 46–49]	6	Pakistan	[34, 35, 71–73]	5
Iraq	[50]	1	Spain	[75–77]	3
Israel	[51]	1	Sudan	[78]	1
KSA	[11, 27–29, 52–62]	14	Tunisia	[36–38, 78–86]	13
Malaysia	[63]	1	USA	[28, 87]	2
Mauritania	[64]	1	Yemen	[89]	1

Table 3.
Countries and number of recruited studies used to collect and analyze data points of TP content in date fruits.

4. Result

4.1 Descriptives

The total TP entries was 583, from 74 studies collected from 18 countries, consisting of 102 (17.9%), 118 (20.7), and 350 (61%) entries for Khalal, Rutab, and Tamer stages, respectively. More than 50% of the entries came from five countries (n, %): Pakistan (126, 21.61), KSA (125, 21.44), Tunisia (59, 10.12), Iran (47, 8.06), and Algeria (35, 6). The apparent number of included cultivars was 250, with Khalas (5.5%), Khadhrawi (4.3%), Barhi (3.1%), Hallawi (2.7%), Deglet Nour (2.4%), and Medjool (2%) being the most represented. Descriptive statistics, including estimates of centrality and dispersion, are presented in **Table 4** for data with and without outliers. The proportion of detected outliers was 1.96, 8.5, and 6.3% for the three maturation stages, respectively. As expected, the mean and median of TP content were higher in Khalal stage than the final maturation stage, Tamer. Removing outliers greatly improved the statistics of dispersion (SD, SEM, range, variance) as well as the kurtosis, skewness, and CL.

	Khalal		Rutab		Tamer	
	WO	NO	WO	NO	WO	NO
Count	104	102	119	108	360	339
Mean	994.92	935.98	368.67	228.62	446.97	240.93
SEM	111.83	105.85	38.74	18.05	59.00	10.18
Median	791.37	777.28	247.98	177.71	232.05	217.65
SD	1140.46	1068.99	422.65	182.33	1119.40	187.45
Variance	1300651.78	1142745.95	178630.03	33244.77	1253049.41	35137.96
Kurtosis	34.75	46.91	5.59	−0.71	47.66	−0.12
Skewness	4.95	5.85	2.21	0.55	6.55	0.71
Range	9785.41	9785.41	2228.61	663.80	10303.70	858.83
Minimum	9.59	9.59	4.33	4.33	0.14	0.14
Maximum	9795.00	9795.00	2232.94	668.13	10303.85	858.97
CL (95.0%)	221.79	209.97	76.72	35.81	116.02	20.03
CV	114.63	114.21	114.64	79.75	250.44	77.80
Q1	343.18	336.73	102.54	87.72	93.89	84.90
Q2	791.37	777.28	247.98	233.64	233.33	217.65
Q3	1298.02	1281.76	470.23	440.04	388.64	351.12

WO = with outliers included, NO = outliers not included.

Table 4.
Estimates of centrality and variability of the values of TP content (expressed as mg GAE/100 g DM) recruited in this work and obtained from studies listed on Table 2.

Khalal stage exhibited higher variation than Rutab and Tamer. The distribution of the TP values was not normal and rightly skewed for the three stages. **Figure 2** depicts the frequency and cumulative frequency density (CFD) of the TP values of the Tamer stage. Similar patterns are also seen for the Khalal and Rutab stages (not shown). More than 50% of the values of TP content were below 260 mg GAE/100 g DM.

Figure 3A and B depicts the spread of numerical values of the TP content in dates at Tamer stages against the country of origin of dates and cultivar, respectively. These figures, as well as data given in **Table 2**, provide clear evidence against claims and misconceptions of the antioxidant superiority of a particular date cultivar due to its country of origin or cultivar. Low and high values of TP content can be found for a specific date cultivar in a single country.

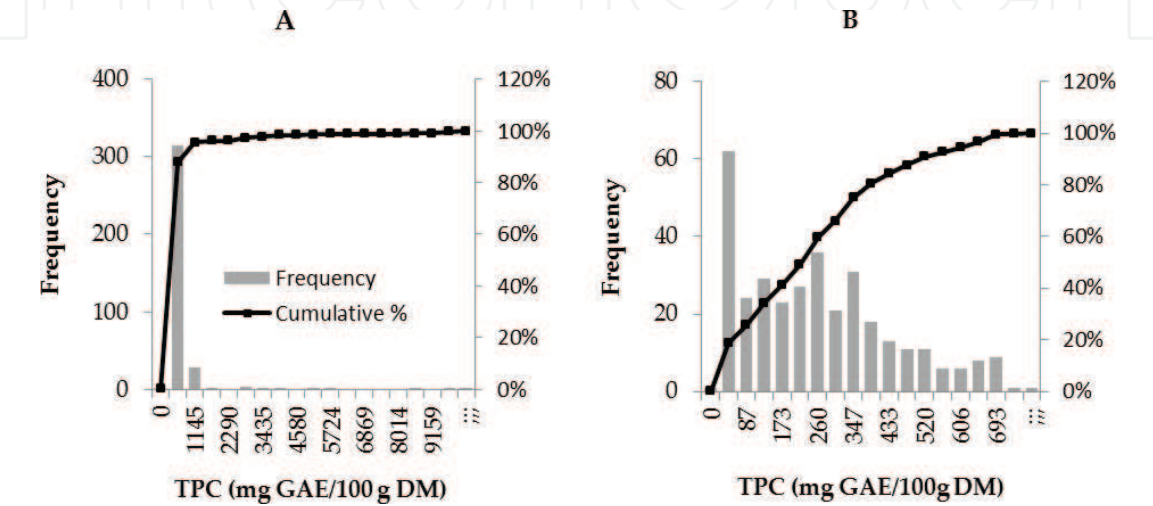


Figure 2.
Histogram and cumulative frequency density (CFD) of the TP content of the Tamer stage. (A) With outliers included, n = 360 and (B) outliers removed, n = 339.

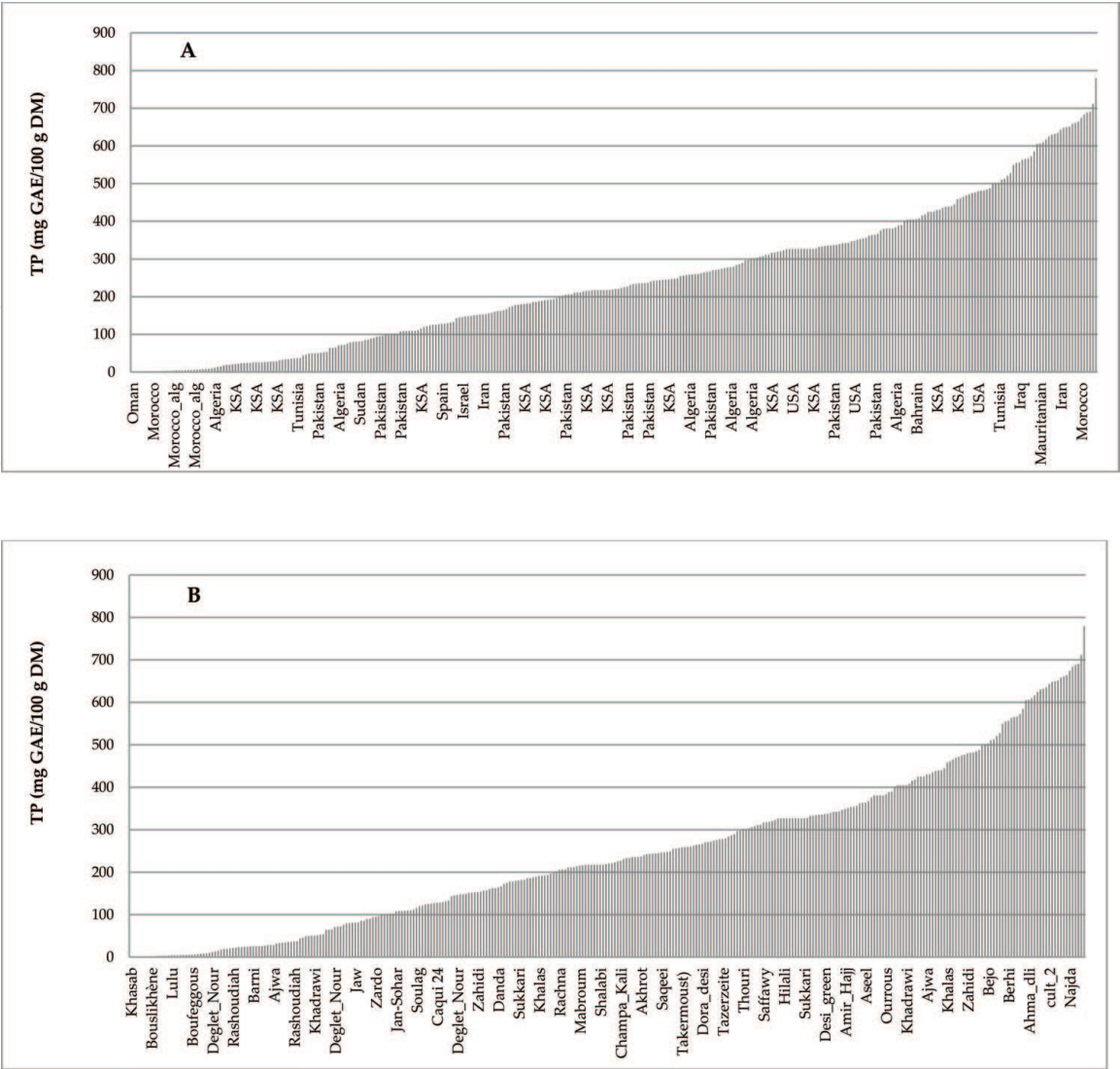


Figure 3. Distribution of the values of TP content in Tamer stage arranged ascendingly according to the country of origin (A) and cultivar (B). Outliers were removed.

	Ajwa	Barhi	Khadhrawi	Khalas	Mejdool	Zahidi	Deglet Nour	Average
Count	9	4	10	20	7	7	13	10
Mean	178.48	254.50	274.79	158.10	265.45	251.96	159.52	220
SEM	70.05	89.86	61.51	34.75	61.84	64.58	34.47	60
Median	49.94	253.26	266.58	112.48	289.46	153.12	108.55	176
SD	210.15	179.71	194.50	155.39	163.62	170.87	124.28	171
Variance	44162.2	32295.9	37829.8	24145.3	26772.8	29195.4	15445.5	29,978
Range	577.66	319.39	477.41	458.12	468.4	407.66	363.74	439
Minimum	6.80	96.05	50.28	0.20	3.33	72.11	3.33	33
Maximum	584.46	415.44	527.69	458.32	471.72	479.77	367.07	472
CL (95.0%)	161.53	285.96	139.14	72.72	151.32	158.02	75.10	149
CV (%)	117.74	70.61	70.78	98.28	61.64	67.82	77.91	81
Country	2	5	7	4	5	5	5	5

Table 5. Estimates of centrality and variability of reported TP values (expressed as mg GAE/100 g DM) of selected date cultivars from different countries.

	Ajwa (KSA)	Rashoudiah (KSA)	Sukkari (KSA)	Khalas (KSA)	Khalas (Oman)	Faradh (Oman)	Khasab (Oman)	Khalas (Oman)	Khalas (KSA)	Ajwa (KSA)
Count	7	3	11	13	5	5	4	5	13	7
Mean	195.48	141.60	280.46	148.11	109.03	155.57	71.68	109.03	148.11	195.48
SEM	89.26	60.87	54.46	43.82	61.34	90.67	59.66	61.34	43.82	89.26
Median	28.35	185.90	327.06	26.12	33.94	35.63	18.92	33.94	26.12	28.35
SD	236.17	105.42	180.63	157.99	137.17	202.74	119.31	137.17	157.99	236.17
Variance	55776.74	11114.00	32625.96	24961.62	18814.52	41102.45	14235.21	18814.52	24961.62	55776.74
Range	577.66	196.39	557.09	450.47	295.96	439.52	248.57	295.96	450.47	577.66
Minimum	6.8	21.26	8.44	7.85	0.20	0.23	0.14	0.20	7.85	6.8
Maximum	584.46	217.65	565.53	458.32	296.15	439.74	248.72	296.15	458.32	584.46
CL (95.0%)	218.423	261.89	121.35	95.47	170.31	251.73	189.85	170.31	95.47	218.423
CV (%)	120.81	74.45	64.40	106.67	125.80	130.32	166.46	125.80	106.67	120.81

Table 6.
Statistics of centrality and variability of reported TP values (expressed as mg GAE/100 g DM) of selected date cultivars taken from different studies from two countries (KSA and Oman).

4.2 Variability of TP values of selected date cultivars from different countries

Table 5 presents estimates of variability and central tendency of TP content of selected date cultivars reported from different countries. Normally, in nutritional epidemiology, the variance represents the true variability of nutrient content. The variability of continuous type of results produced experimentally by some assays is evaluated by the CV rather than SD, since the CV is a standardization of the SD ($CV = SD/\text{mean} \times 100$). Using CV allows for direct comparison of estimates of variability regardless of the magnitude of the level of analyte under investigation. In many biological fields, a twofold difference in measurements of the same sample can be acceptable as the upper limit of variability. Furthermore, a CV of 40% can be tolerated in nutrient estimation for food labeling and nutrient intake calculation [90]. Since there is no reference value or a benchmark for the variability of TP content in dates to compare with, the above recommendation may be used to facilitate comparison. The variance and CV, as well as other estimates of dispersion, are very large. The largest variance was found for Ajwa, whereas Deglet Nour exhibited the lowest variance. The CV was even more pronounced as an evidence of the vast variability, with some cultivar possessing CV values of more than 100%. Estimates presented on **Table 5** demonstrate the extent of variability of the TP content values regardless of the country.

Table 6 presents similar statistics based on data obtained from studies originated from a single country for a particular date cultivar. This table illustrates the extent of variability of the TP content within a country. For example, TP values of Khalas cultivar from two countries (Saudi Arabia and Oman) showed large variation within cultivar and between the two countries, while the TP values of selected date cultivars taken from different studies carried out within that country are similar. Again, all estimates of variability are indicative of the large disparity of the published TP values. Notably, Ajwa cultivar of Saudi Arabia, which is grown almost exclusively in the holy city, Al-Madina Al-Munawara, possessed the largest CV (%) among the listed four cultivars.

5. Discussion

5.1 Variability of TP value and its implication

In the fields of public health, nutrition, and nutritional epidemiology, reliable and accurate estimates of concentration of a nutrient in a food commodity is important for estimating the daily consumption (intake) for an individual within a population, as well as for setting the average, upper, and lower limits of that nutrient for official recommendations and guidelines. The TP content is neither (yet) considered as a nutrient nor as a single chemical compound that can be reduced to the level of an officially declared nutrient such as ascorbic acid and treated similarly. TP is rather an experimentally measured value representing a chemical measure for an inherently great numbers of diverse groups of secondary metabolites or phytochemicals, simple and polyphenols, with many biological functions vital for the survival of their producers (plants) and for their consumer. Although TP is not a single entity, but, theoretically (and hypothetically) speaking, it is similar, in a way, to the groups of foods (proteins, carbohydrates, and lipids) which are characterized by high diversity of its nature, structure, and consistency. For this, one may be allowed to deal with TP content in a similar way, taking into consideration that TP content, at present, is not among the macro- and micronutrients.

Admittedly, large variation widely exists in biological measurements. In nutritional sciences, nutrient variability is a common place. A nutrient may vary in its numerical values for many reasons, and the magnitude of variation can be very large [92]. In the analysis of the already published values of the TP content date fruits for a large number of cultivars from different countries, regions, and continents, it can be concluded that the magnetite of variation in all edible stages, and in the Tamer stage in particular, is very high, in the order of hundreds, when extreme values and outliers are removed, and perhaps in thousands when these values are included.

This situation represents an unfavorable challenge for researchers, nutritionists, end users, and policy makers alike. To illustrate, a researcher may ask of the typical value of TP content of the date fruit in general or a typical value for a specific cultivar. In fact, in the literature, it is common to declare nutritional values of dates based on one or two cultivars with the assumption that these are true representative of the vast majority of cultivars, i.e., [91].

Does such variability is due to natural variation, or should we take into consideration the uncertainty, or a combination of both? This remains unclear and needs to be answered. While variability is defined as the occurrence of multiple values for a quantity at different locations and refers to the inherent heterogeneity or diversity of data in an assessment, uncertainty refers either to the lack of knowledge of the value of some quantity (qualitative uncertainty) or the usage of non-precise measurement methods of (quantitative uncertainty) may come from the use [93]. The source of uncertainty can be of many types including random errors, sampling, and measurement errors. Variability can be characterized but cannot be reduced, whereas uncertainty can be reduced, which, if appropriately applied, can lead to increased confidence in the estimates [93].

The variability of the values of the TP content for date fruits is evident by the various estimates of dispersion (see **Tables 4–6**). Causes of such dispersion are not known nor can be investigated unless the experimental conditions of the actual analysis can be traced back. In such situation, with little or no knowledge about of the data quality and the associated errors, one may speculate that data of the values of TP of dates do not merely reflect a natural variation, but element(s) of uncertainty cannot be excluded.

5.2 Sources of variability of antioxidant activity in dates

Variation in antioxidants and antioxidant activity is not limited to variation due to cultivars, maturity stage, and geographical or agronomical conditions. Rather, antioxidant activity varies between dates within the same bunch and even within the same fruit. In the following section, some of causes of the antioxidant will be presented.

5.2.1 Variation of antioxidant properties due to maturity stage

Many studies examined the effect of maturity stage in the antioxidant consistency and activity [25, 35–38, 41, 54, 73, 74]. There is a general agreement that the highest antioxidant activity is found in Khalal stage and the lowest in Tamer stage. Sourial et al. [94] reported data of five cultivars exhibiting a sigmoidal decline of tannins. The remaining tannin content at Tamer stage represented 33–43% of that at Rutab and Khalal stages. The kinetics of degradation of total phenolic content during these three stages was also reported [42]. TP content declines to follow first-order reaction in the Tamer stage which represents between 25 and 40% of the Khalal stage. Generally, red cultivars at Khalal stage possess greater antioxidant activity than yellow cultivars.

5.2.2 Variation of antioxidant properties within a single date fruit

Date fruits harvested from the same bunch at the same time may possess different levels of antioxidants, though may be statistically insignificant (need further studies). Within the same bunch, dates are differentially exposed to sunlight. Sunlight affects biosynthesis of simple and polyphenolic compounds including flavonoids. In many fruits, biosynthesis of polyphenolic compounds is an adoptive process [95]. High light induces the expression of many early and late genes involved in biosynthesis of flavonoids. Dates located inside the bunch are the least to receive sunlight, compared to those at the peripheral. This is also valid with regard to different bunches within the same tree.

Within a single date fruit, the distribution of antioxidants in the tissues is not homogenous. Guo et al. [96] reported that the peel of unspecified date cultivar possessed 2.4 times higher antioxidant activity than the pulp, 16.69 compared to 6.98 mmol/100 g WW (FRAP assay), respectively. A recent study by Djouab et al. [97], using Tamer of the yellow Algerian cultivar Mesh Degla, showed that the level of TP in the whole flesh, peel, brown tissue, and white tissue was 206, 247.3, 185.2, and 66.63 mg GAE/100 g DM, respectively. In this study, the antioxidant activity followed the same trend. Generally, fruit peels possess higher antioxidant than the flesh [96]. Depending on date cultivar, peel may contribute between 50 and 70% of the antioxidant, despite constituting only 3–5% of the total edible weight. Due to their vital biological role as protectants, many potent polyphenolic antioxidants are essentially localized in the peel, particularly during Khalal stage, leading to higher antioxidant activity. Furthermore, the white tissue of the flesh, the most inner part, possesses the least antioxidant/activity as compared to other tissues. Within the brownish tissue, condensed tannins are, usually, stored in the stone cells.

5.2.3 Variation of antioxidant activity due to diverse polyphenolic composition

Antioxidant property in plant-based food is largely due to the natural polyphenolic antioxidants. Redox properties of these natural antioxidants make them function as reducing agents, free radical scavengers, hydrogen donors, chelators, and metal. The phenolic consistency of date fruits, including flavonoids, has been recently studied by many research groups [18, 22–24, 38, 62, 68–70, 74, 98–100].

Phenolic acids found in dates belong mainly to benzoic or cinnamic acid derivatives. However, the distribution of phenolic acids varies considerably among different date cultivars. El-Arem et al. [38] reported a significant difference in the phenolic compounds amounts between maturation stages for the majority of cultivars. These groups identified two newly described phenolics in dates (hydroxyphenylacetic and phenylacetic acids). A contrasting example of the dynamic nature (or fluctuation) of phenolic acids in date fruits is cinnamic acid (CNA) which was also reported by this group [38]. CNA was not detected in the three maturity stages (K, R, T) of cultivars Gondi and Rotb Ahmar; however, it significantly increased during maturation of cultivar Gosbi and detected in comparable amount in R and T stages but not in K in the cultivar Khalt Dhahbi.

Farag et al. [24] recently identified 44 metabolites in 18 Saudi cultivars, of which 20 were flavonoids and 4 were hydroxycinnamates but also noted that several of previously reported predominant phenolic acids were not found in their study. While free phenolic acids are present in Rutab stage in most cultivars, albeit at lower concentration, the semidry cultivar Sukkari had no detectable free and glycosylated phenolic acids at Rutab stage and contained only esterified phenolics. Moreover, the fate of a particular phenolic acid or flavonoid differs between date cultivars (**Figure 4**). Among six different cultivars, Kursinszki et al. [98] reported that rhamnosyl

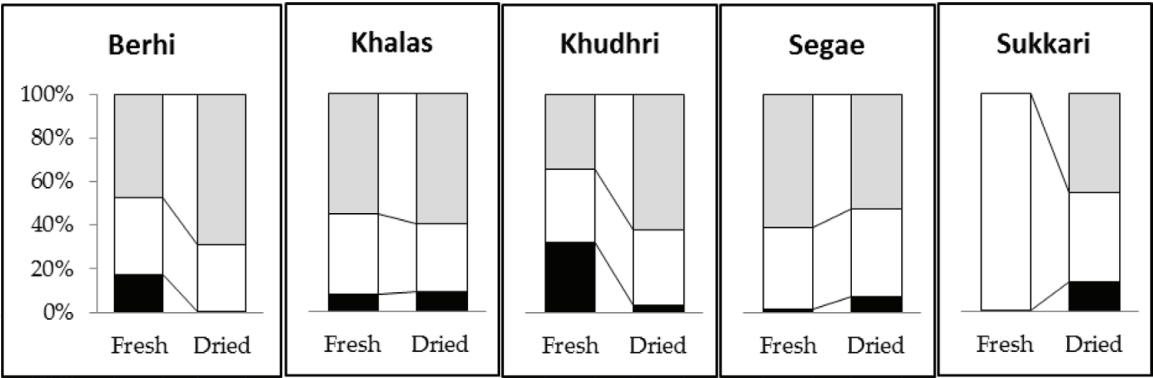


Figure 4. Distribution of free (■), esterified (□), and glycosylated (▒) phenolic acids in five date fruits at two maturation stages Rutab (fresh) and sun-dried Tamer. Data extracted from Khojah [53].

hexosyl methyl luteolin was a major constituent in all of them, albeit at different levels, whereas hexosyl methyl luteolin sulfate was a major constituents in only three cultivars Khenazizi, Khalas, and Lulu. Among these cultivars, Lulu was characterized by being relatively low in flavonoid content. The Al-Medina dates were distinct by the presence of rhamnosyl hexosyl luteolin. A very recent detailed study by Abu-Reidah et al. [101] has identified 52 phenolic compounds in five various parts of the date palm tree including the edible portion (skin and pulp). The distribution (and the quantification) of phenolic compounds in the edible portion of the date fruit is of particular interest on this review. The combined number of peaks identified in pulp and skin was 22, of which 17 were found in pulp and 16 in skin, with 12 peaks being shared (~55%). Interestingly, the edible portion of dates was lacking of ferulic acid derivatives despite its known abundant in both, the skin and pulp. To the contrary, luteolin was only found in the skin, while its derivatives may be found, unequally, in both tissues. The methyl glycoside derivatives, which is consistent with specialization and functionality of the plant part, were also lacking from the edible portion.

5.2.4 Variation of antioxidants due to pollination, bagging, and thinning

Date palm tree is a dioecious monocotyledonous, and fertilization occurs either naturally or is carried out artificially. Pollens obtained from one cultivar can fertilize another cultivar. However, pollination has significant impacts in the physical and chemical properties of the resultant dates. It affects, among other things, the fruit set, size, time of ripening, seeds, eating quality, as well as the chemical constituency of the date including antioxidants, an effect known in plant science as metaxenia. Maryam [72] reported that pollen patents had the potential to significantly influence total phenolics in dates. Using eight male pollen patents to fertilize two different cultivars, the TP of Hallawi cultivar increased from 190 mg GAE/100 g in the control to 491 mg GAE/100 g and from 212 to 480 mg GAE/100 g in Khadhrawi cultivar. Similar effect was also found with ascorbic acid. Farag et al. [102] found that one of two pollinator types significantly increased the content of anthocyanin and ascorbic acid, but not tannins, over the other.

The practice of fruit thinning, either by reducing the number of fruits per bunch or the number of bunches per tree, leads to significant quality enhancement in dates. Several methods of thinning are available for date palm trees. This practice was found to reduce the tannins content in some date cultivars [103]. Bunch bagging of the same cultivar with perforated blue polyethylene increased ascorbic acid level, decreased the total soluble tannins concentrations and peroxidase activity, and had no significant effect on total phenolic content [104].

5.2.5 Variation of antioxidant priorities due to abiotic stress

In a study of the effect of sewage water irrigation of date palm tree in the antioxidant constituency of three Saudi date cultivars, Abdulaal et al. [105] reported higher levels of TP; TF; increased antioxidant activity using ABTS, DPPH, and the formation of phosphor-molybdenum complex test; as well as higher activities of peroxidase, polyphenol oxidase, and glutathione-S-transferase in dates irrigated with sewage water as compared to irrigation with municipal water. The increased level of these parameters was accompanied with higher accumulation of heavy metals (Cr, Cu, Fe, Mn, Pb, and Zn) in the sewage water-irrigated dates. The three studied cultivars showed differential responses regarding TP and TF. TP level in Agwa and Safawi increased by 28–30% over the control, while in Anbr cultivar it increased by only 8%. Furthermore, the extent of increase in TF in the three cultivars was somewhat similar (Agwa, 41%; Anbr, 50%; and Safawi, 50%). These results are suggestive of different response mechanisms and need further investigation.

Al-Busaidi et al. [106] recently reported that while the levels of Fe, Zn, and Ni were relatively higher in the treated sewage water irrigated than the groundwater irrigated, whereas the levels of Cu, Cd, Pb, and B were significantly higher in date fruits irrigated with groundwater than sewage water irrigated. These contradicting findings may be partially attributed to the level of treatment of sewage water used, i.e., secondary or tertiary treatment. In our own findings (unpublished) with locally grown several date cultivars, no significant difference was found in the accumulation of several heavy metals between groundwater and secondary-treated sewage water-irrigated dates.

5.2.6 Association of antioxidant consistency and antioxidant activity in date fruits

In vitro methods commonly used to estimate antioxidant activity include ABTS, DPPH, FRAP, and ORAC. Like many other plant-based foods, a clear relationship between the antioxidant content and antioxidant activity exists in date fruits, though its extent varies widely. For example, the DPPH method, widely used to estimate the radical scavenging activity of antioxidants was found [54] to be highly correlated to TP content in four Saudi cultivars, namely, Barhee ($R^2 = 0.96$), Khenazy (0.89), Helali (0.85), Lonet-Mesaed (0.64), but was not significantly correlated in Mejdool (0.46). In contrast, Medjool exhibited high correlation ($R^2 = 0.91$) between DPPH and total soluble tannin concentration. The correlation of DPPH and phenols, tannins, and flavonoid content of 12 products made from two Spanish date cultivars was also high, 0.765, 0.747, and 0.822, respectively [76]. On the other hand, plotting the IC₅₀ (amount in $\mu\text{g/ml}$ which gives 50% inhibition of DPPH quenching) of 18 cultivars from Saudi Arabia against their total phenolics showed a weak correlation ($R^2 = 0.0341$) [27]. These findings not only indicate that phenolic content plays as the major antioxidant in date fruits but also as a cause of the apparent variability of the date antioxidant activity.

6. Limitations of this work

Due to many constraints, this chapter addresses only one aspect of the variability of antioxidants in dates, namely, the TP content. The purpose of this chapter is to shed light and to expose the problem in the hope that other opportunities will be available to address the issue more comprehensively. The issue can be treated in depth with the inclusion of published values of other antioxidants as well as antioxidant

activities. Potential and appropriate statistical tools to investigate the issue are within and between subject analysis of variability and multivariate analysis. Uncertainty as a potential source of variability of date antioxidants can also be examined.

7. Conclusion

Variability of levels of phytochemicals (plant-based) is a common phenomenon. However, the magnitude of such variability is influenced by natural and artificial causes. Examination of values of the TP content in dates published over the last two decades reveals wide disparity that needs to be seriously addressed. This large variability creates a challenge that makes it difficult to deal with the validity and reliability of published values and may hinder or reduce its practical usefulness. Overcoming this problem and related issues requires collaboration between many groups from different countries. With many research teams interested in the date palm and its fruit (dates), this is possible and achievable and requires someone who takes the initiative.

Acknowledgements

The author wishes to thank Mrs. Batool Abdul Wahab, Department of Biology, College of Science, University of Bahrain, for her tremendous efforts during the preparation of this chapter, doing literature searching and collecting and making them available.

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