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Phytochemical content, antioxidant potential, and fatty acid composition of dried Tunisian fig (*Ficus carica* L.) cultivars

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Summary

This study reports the main phenolic compounds, as well as phenolic profiles and antioxidant activity in nine sun-dried fig cultivars with different skin color, originating from South-Eastern and Middle-Eastern Tunisia. For all evaluated parameters, a considerable variability with high significant differences was observed among the cultivars studied. Dark fruits exhibited a higher total polyphenol contents (201.77 mg GAE/100g DM in cultivar Saoudi Douiret) compared to green fruits (73.74 mg GAE/100g DM in cultivar Bayoudhi Douiret). Fatty acid methyl esters, identified by GC-MS, distinguished the presence of (C16: 0), (C18: 1), ((C18: 2) 9, 12), ((C18: 3) 9, 12, 15) and (C20: 0). Strong correlations between the amounts of total phenolics, phenolic acids, flavonoids, fatty acids and antioxidant capacity were found. A principal component analysis showed three groups of cultivars regarding their similarity level.

Key Words: Bioactive compounds; fatty acids; *Ficus carica* L.; GC-MS; LC-ESI-MS.

Introduction

Ficus carica L. is considered one of the world's oldest fruit trees, belonging to the Moraceae botanical family (SOLOMON et al., 2006). This species is characterized by various chemical components and biological activities, which allow it to be a typical element of the Mediterranean diet that has been favorable to health for millennia; comparing its phenolic composition with red wine and tea, its level seems to be higher (VALLEJO et al., 2012). All over the world, figs are consumed fresh or dried because of their richness in minerals, carbohydrates, essential amino acids, vitamins and polyphenols (SOLOMON et al., 2006; VEBERIC et al., 2008; VIUDA-MARTOS et al., 2015). Furthermore, they are exploited in traditional medicine and have been studied and proven their potential therapeutic value (KHAIRUDDIN et al., 2017; MONIRUZZAMAN et al., 2017). This plant is cultivated in all Mediterranean regions and countries with similar climatic conditions. About 1,184,884 tons of figs are produced worldwide. Turkey is the leading producer of figs providing approximately 23.5% of the total production in the world; about 51.6% of this crop is marketed in dry form (FAOSTAT, 2015). In Tunisia, total fig production is 26,000 tons/year representing 3% of world production (FAOSTAT, 2015), mainly produced in the South-Eastern regions (34% of the national production) (MARS et al., 2008). Fig cultivars (cvs) and the wild form of the fig can show a wide variety of fruits with respect to color, shape, weight, acidity, sugar and mineral contents, etc, while the region where the figs are mainly cultivated is at the origin of their name (MEZIANI et al., 2015). Thus, a morphological and chemical study must be performed in order to show the variation among cultivars.

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Despite the prominence of this crop in arid, semi-arid and sub-humid regions, research on natural substances and molecules of this species has been a major topic for several years. These molecules, which constitute the active ingredient of medicinal plants, belong predominantly to secondary metabolites such as polyphenols, essential oils and alkaloids. Several research programs on figs are being developed in order to analyze their content in polyphenols (SOLOMAN et al., 2006; CALISKAN and POLAT, 2011; HARZALLAH et al., 2016; KONAK et al., 2017). Since dried figs are a more concentrated form of fruit, and due to its economic benefits which are appreciated by both industrial users and individual consumers, the current work investigated the quantity and quality of antioxidants contained in these fruits to determine the richness in these elements.

Few studies investigated the phytochemical contents in dried figs. The current study evaluates the contents of phytochemical compounds and the antioxidant activity by using ABTS and DPPH tests and characterizing the phenolic and fatty acid profiles of light and dark skin dried fig cultivars cropped in South-East and Mid-East of Tunisia. The aim is to determine which cultivars can be recommended as natural healthy food products basing on their biochemical and nutritional facts, thus allowing their labeling as a superior product.

Materials and methods

Plant material

The nine local fig cultivars were harvested by hand in the summer of 2015 and 2016 in several commercial farms located in southern and central Tunisia during the ripening stage (Tab. 1, Fig. 1). Figs were then traditionally sun-dried for seven days using the direct solar dryer, consisting of a single piece that is both a drying chamber and a solar collector, which produces an uncontaminated final product of good quality on the hygienic and nutritious plan (OUAOUICH et al., 2005). After drying, figs were stored in closed jars in cold rooms.

Tab. 1: Origin and description of studied dry fig cultivars.

Accession name	Label	Geographical origin	Description (Color)
Saoudi douiret	SD	Douiret Tataouine	Black
Bayoudhi douiret	BD	(South-East)	Green – yellow
Wedleni bir amir	WBA	Bir Amir Tataouine	Purple
Hamouri bir amir	HBA	(South-East)	Light purple
Zidi bni khdech	ZBK	Bni Khdech Medenine	Black
Bayoudhi bni khdech	BBK	(South-East)	Light green
Hamouri bni khdech	HBK		Purple
Kahli karkenah	KHK	Karkenah Sfax	Purple
Mahdoui karkenah	MK	(Mid-East)	Light green

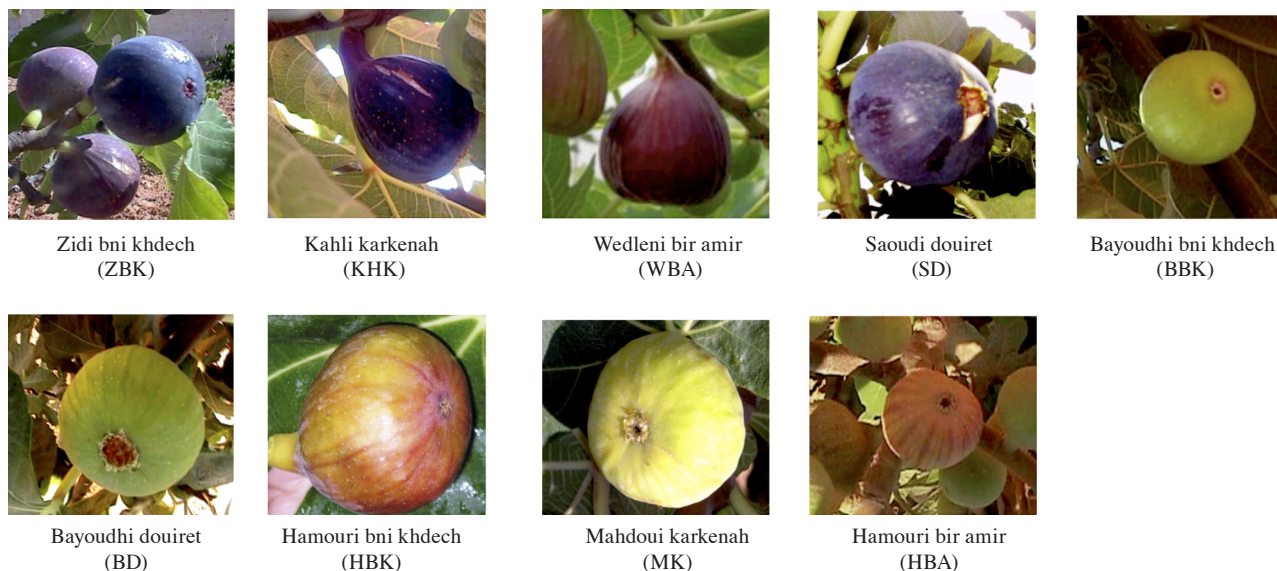


Fig. 1: Nine fig cultivars with different skin colour at ripening stage.

Total polyphenols content determination

The polyphenols were extracted with 80% methanol (1 g of each sample is ground with 6 mL of methanol), then the samples were placed in a box with ice and kept in the dark during the extraction which was performed on an orbital shaker (Stuart, Staffordshire, UK) for one hour at $200 \times g$. After that, fig extracts were centrifuged at 15,000 rpm (4°C) for 10 min and the supernatant was recovered and stored for total phenolic compounds (TPC) and total antioxidant capacity (TAC) quantification. Total phenolic content in fig extracts was evaluated, using the Folin-Ciocalteu method, in 96-well plates, with a spectrophotometer (Tecan Infinite M200 Mannedorf, Switzerland) at the absorbance of 750 nm, estimated by the method of SINGLETON and ROSSI (1965) with some modifications proposed by MARTINEZ-HERNANDEZ et al. (2011). In each sample, the total polyphenol contents were expressed in mg of gallic acid equivalent (GAE) per 100g of dry matter (mg GAE/100g DM) through a calibration curve based on gallic acid at different concentrations (mg/mL).

Total flavonoids content determination

Total flavonoids content in the methanolic extracts of dried figs was monitored by the colorimetric method of aluminum chloride (AlCl_3) (BRIGHENTE et al., 2007) with slight modifications. Then, 1 mL of each fig extract was added to 1 mL of a methanolic solution of AlCl_3 (2%). After 10 min incubation at room temperature and using a spectrophotometer at 430 nm, the absorbance of the mixture was determined. The flavonoid concentrations were deduced from the calibration curve established with quercetin. The results were expressed as mg quercetin equivalents per 100 g of dry matter (mg QE/100g DM).

Antioxidant capacity

DPPH assay

The DPPH assay (2, 2-diphenyl-1-picrylhydrazil) was determined by a spectrophotometric method at 515 nm, adapted from BRAND-WILLIAMS et al. (1995) with slight modifications made by MARTINEZ-HERNANDEZ et al. (2011). The test is based on the capacity of scavenging free radicals. The DPPH radical solution was prepared by dissolving 12 mg of DPPH in 50 mL of methanol and then stored at -5°C in the dark. The absorbance was adjusted to 1.1 ± 0.02 . An aliquot of 21 μL of extract was placed in a 96-well plate and 194 μL of DPPH solution were added.

After incubation for 35 min under dark conditions, the absorbance of the aqueous solution was measured against a blank using a spectrophotometer. The results were expressed as mg TROLOX equivalent antioxidant capacity (TEAC) per 100 g DM through a TROLOX calibration curve at different concentrations (mg/mL). All measurements were performed in triplicate.

ABTS assay

The ABTS assay (2, 2'-Azino-bis (3-ethylbenzothiazoline-6-sulfonic acid)) was determined by a spectrophotometric method at 734 nm, which was adapted from ARNAO et al. (2001) with slight modifications. The assay is based on the ability to reduce the radical cation 2, 2'-Azino-bis (3-ethylbenzothiazoline-6-sulfonic acid) diammonium salt (ABTS) by antioxidant. The mixture solution contained 14 mM ABTS and 4.9 mM $\text{K}_2\text{S}_2\text{O}_8$ solutions diluted in methanol and adjusted to 1.1 ± 0.02 (absorbance), and then incubated for 16 h in the dark. An aliquot of 10 μL of extract was placed in a 96-well plate and added with 190 μL of ABTS solution. After 35 min of incubation, the absorbance of the aqueous solution was measured against a blank using a spectrophotometer. Results were expressed in terms of mg TROLOX equivalent antioxidant capacity (TEAC) per 100 g DM through a TROLOX calibration curve at different concentrations (mg/mL).

Identification and quantification of phenolic compounds

After pressurized liquid extraction and solid-phase extraction, the polyphenolic patterns of figs were identified and quantified using a liquid chromatography-electrospray ionization-tandem mass spectrometry (LC-ESI-MS). The extract was filtered through a 0.45 μm membrane filter and then injected into the High Performance Liquid Chromatography system (HPLC). Quantification was achieved by injection of standard chemical solutions of known concentrations at the purity $> 98\%$. The mobile phase was composed of a mixture of two solutions: The first contains 0.1% formic acid in H_2O and the second contains 0.1% formic acid in methanol. The column temperature was set at 40°C and the flow rate of the mobile phase was 0.4 mL/min. The auxiliary gas used is the N_2 which was very pure. The results were expressed as mg/100 g DM. All extracts were analyzed in triplicate.

Fatty acids composition

The total lipid content was determined by the Soxhlet method which allows extracting the dry extract as much as possible. The Fatty acid methyl esters (FAMES) were extracted using a hexane solvent and were determined by gas chromatography coupled with GC-MS mass spectrometry (Supelcowax TM10 FUSED SILICA CAPILLARY COLUMN), 30 m × 0.25 mm × 0.25 μm film thickness (Supelco, Milan, Italy), according to MONDELLO et al. (2004) with slight modifications. GC-MS analysis was performed using a GCMS-QP2010 Ultra mass spectrometer (Shimadzu, Kyoto, Japan), with a split/split less injector and a Shimadzu AOC-20i auto-injector and a Shimadzu AOC-20s auto sampler. The data was obtained by the GCSolution / GCMSsolution software. The identification of Fatty acid methyl esters (FAMES) was achieved by comparing their retention time with pure standards analyzed under the same conditions.

Statistical and multivariate analysis

Statistical analyzes were performed using an ANOVA with SPSS version 22.0 software. Duncan test was used to compare significant differences with the level of significance at $P < 0.05$ and Pearson's correlation coefficient (r) involved to determine correlations between studied parameters. Each assay was performed three times from the same extract and the value was expressed as mean ± standard deviation (SD).

The structure of genetic variability among the figs cultivars based on mean values of total polyphenol contents, phenolic compounds such as phenolic acids and flavonoids, fatty acids and antioxidant activity was analyzed using principal component analysis (PCA). These multivariate analyzes were carried out using the Xlstat 9.0 software to evaluate the influence of phenolic compounds in the differentiation of Tunisian dried fig samples.

Results and discussion

Total polyphenol contents (TPC)

The TPC of dried fig cultivars are shown in Fig. 2. The fig cultivars were classified into seven different homogenous subsets according to the Duncan test ($P < 0.05$). Cultivars belonging to different subsets presented significant differences in terms of phytochemical characters. The TPC ranged from 73.74 mg GAE/100g DM in (BD) cv to 201.76 mg GAE/100g DM in (SD) cv. Our findings are inferior

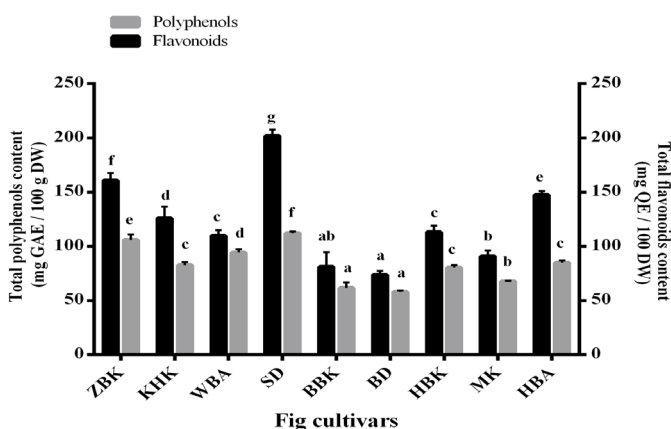


Fig. 2: Contents of total phenolics (mg GAE/100 g DM) and flavonoids (mg QE/100 g DW) in nine cultivars of dry figs. Data are displayed as mean ± standard deviation of three replications for each cultivar. Letters above columns represent the groups given by Duncan test ($\alpha = 0.05$) in arising order. Different letters indicate significant difference at $P < 0.05$.

to those of POURGHAYOUMI et al. (2017), who mentioned that dried Iran fig contained 1120 to 2681.8 mg GAE/100g DM, but are in accordance with the results reported by CAPANOGLU (2014) who found 169.4 mg GAE/100g DM in Turkish dried fig. These differences in TPC can be attributed to differences in crop region and cultivar type, as well as to cultivation practices, geographical origin, experimental conditions, and postharvest storage conditions (SOUFI et al., 2014; HOXHA et al., 2015). Furthermore, the drying processes used (air or sun drying) may be responsible for these differences (ARVANITI et al., 2019). On the other hand, there was no significant difference between cultivars harvested in South-East and Mid-East of Tunisia in terms of TPC, since these two regions have almost the same climatic conditions (arid climate and semi-arid climate, respectively). MK cv (from Mid-East) contained 90.96 mg GAE/100g DM while BBK cv (from South-East) contained 81.21 mg GAE/100g DM). As expected, the polyphenol contents in dark-skinned figs were significantly higher than those in light ones. The highest values were found on dark colored figs (SD-cv, ZBK-cv, HBA-cv and KHK-cv) which shows that the skin color was the major contributor of this difference. These results were confirmed by DEBIB et al. (2014) who found similar findings and reported that dark-colored dried fig varieties contained a higher level of phenolics than the green and yellow ones (HARZALLAH et al., 2016). Comparing the levels found in dried figs with those of fresh figs, several studies have shown that the TPC in dried figs was higher than that of fresh ones (KONAK et al., 2017). However, other studies have shown that drying methods have a negative impact on the phytochemical contents (BACHIR BEY et al., 2016). Nevertheless, these levels remain higher than those of certain fruits (dates for example) (AL-TURKI et al., 2010). It is interesting to note that a large number of biochemical properties are identified in these compounds and are conceivably useful for inhibiting the development of diseases such as cancer, cardiovascular diseases and diabetes (CHANG et al., 2016).

Total flavonoid contents (TFC)

As shown in Fig. 2, TFC of dried fig extracts ranged from 57.96 mg QE/100g DM to 112.28 mg QE/100g DM. Statistically significant differences in TFC were found between dark and light cultivars ($P < 0.05$) (Fig. 2); SD cv (South-East) appears to have a significantly higher TF content, while a low BD cv (South-East) content was observed.

There is no data available on TFC in black and light dried figs; only one study reported the total number of carotenoids in Algerian figs (OUCHEMOUKH et al., 2012; ARVANITI et al., 2019).

These results are consistent with those obtained by BEY and LOUAILECHE (2015) who indicated that flavonoids content in dark cultivars (126.55 mg/100 g) was higher than that of light ones (87.24 mg/100 g) which confirm that dark-colored figs had more TFC compared to light-colored ones. Regarding the factors that affected this phytochemical composition, our study demonstrated that color and fig cv were responsible for TFC variation, but other studies have shown that other factors were accountable such as drying methods used and the level of maturity of this fruit (PEREIRA et al., 2017; SEDAGHAT and RAHEMI, 2018).

As antioxidants, flavonoids exert therapeutic effects such as antihepatotoxic, antitumor and prevent diabetes and infections (CHANG et al., 2016). In addition, a set of flavonoids provides protection against cardiovascular mortality and heart disease (WANG et al., 2018).

Antioxidant capacity (AC)

The Antioxidant capacity (AC) of dried fig cultivars was assessed by two assays: the DPPH and the ABTS tests. Results shown in Fig. 3 proved that all fig extracts were characterized by antioxidant

activities with significant differences ($P < 0.05$). The antioxidant activity (AAO) against DPPH ranged from 131.55 mg TEAC/100 g DM to 418.51 mg TEAC/100 g DM, respectively for BD cv and SD cv. Similarly, SD cv exhibited the highest level of TEAC using the ABTS test up to 207.43 mg TEAC/100 g DM, while the BD cv presented the lowest level up to 124.53 mg TEAC/100 g DM.

Besides, a significant positive and high correlation was assigned between these two assay methods used to evaluate the antioxidant capacity on the one hand ($r = 0.953$), and between the AC and the amount of phytochemical compounds on the other hand at the $P < 0.05$ (Tab. 2), which infers that the value of antioxidant capacity is often due to the existence of polyphenols and flavonoids. Similarly, KSOUDA et al. (2018) estimated a high and positive correlation between the values of antioxidant capacities obtained by these two methods for the majority of the plant methanolic extracts of 25 Tunisian plant species. Furthermore, OUCHEMOKH et al. (2012) and AMMAR et al. (2015) showed strong correlations between the amount of phytochemical compounds and the antioxidant capacity of figs. According to BREWER (2011), the antioxidant capacity is generally due to the presence of phenolic acids such as Caffeic, Gallic and Chlorogenic acids, as well as to flavonoids namely Rutin, Catechin, Kaempferol and Quercetin.

Moreover, the ANOVA study revealed that the difference between the green-yellow BD cv and the light-green BBK cv was not significant, neither between HBK and WBA purple cultivars nor between HBA and KHK purple cultivars. Significant differences were found only between cultivars owning different skin color (Fig. 3). Thus, figs originated from dark-colored cultivars exhibited a higher antioxidant capacity compared to those from light-colored cultivars with a range of 3 to 4 fold differences between cultivars. These findings were confirmed by KONAK et al. (2017) and PEREIRA et al. (2017). Therefore,

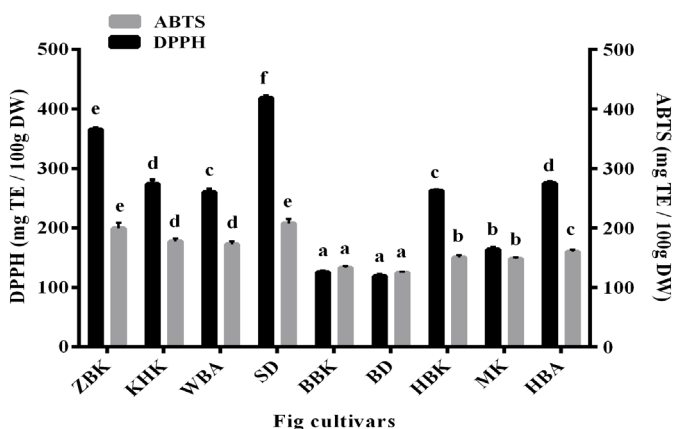


Fig. 3: The total antioxidant capacity of nine dry fig cultivars using DPPH and ABTS radicals. Data are displayed with mean \pm standard deviation of three replications. Letters above columns represent the groups given by Duncan test ($\alpha = 0.05$). Different letters indicate significant difference at $P < 0.05$.

Tab. 2: Correlations between antioxidant capacities by ABTS and DPPH assays, TPC and TFC of fig cultivars studied. Bold Pearson correlation coefficients are significant at 5% of confidence level.

Variables	DPPH (TEAC)	ABTS (TEAC)	TPC	TFC
DPPH (TEAC)	1			
ABTS (TEAC)	0.953***	1		
TPC	0.961***	0.906***	1	
TFC	0.973***	0.963***	0.916***	1

a strong correlation between skin color and TPC and AC should be expected. These results make it possible to deduce that the two dark-colored figs, SD and ZBK, harvested in South-Eastern Tunisia, are recommended for their phytochemical contents and their antioxidant potential in order to improve their health-promoting properties.

Regarding the impact of drying on the antioxidant capacity, CHANG et al. (2016) reported that sun-drying process had a positive effect on antioxidant activity, which increases the antioxidant activity of dried figs compared to that of fresh figs, although BACHIR BEY et al. (2016) found that it reduced the antioxidant capacity of figs.

Despite the great interest in the AC of dry figs, it is difficult to know which of the phenolic compounds has the most important antioxidant activity. That's why we use to isolate and identify the antioxidant components in order to evaluate the mechanisms of activity.

LC-ESI-MS analysis

Individual antioxidants found in our extracts are shown in Tab. 3 and seventeen isolated antioxidants were detected. These compounds included different classes of phenolic acids and flavonoids (flavan-3-ol, flavanones, flavonols, and flavones). Among the phytochemicals, in dried figs, each extracted fraction revealed high contents of Rutin (38.05 mg/100 g DM) for SD cv, followed by Protocatechuic acid (10.6 mg/100 g DM) for WBA cv, Cirsiliol (2.72 mg/100 g DM) for SD cv and 4-O-Caffeoylquinic acid (2.42 mg/100 g DM) for KHK cv. Moreover, other phenolic compounds were detected, such as Quercetin (1.49 mg/100 g DM) for SD cv and Kaempferol (1.13 mg/100 g DM) for ZBK cv. A number of phenolic acids were also detected in dried figs, such as Gallic acid, p-Coumaric, trans Cinnamic and trans-Ferulic, but at very low concentrations (< 1 g/100 g DM). In addition, Salviolinic acid was detected only in WBA and SD cultivars with respective amounts of 0.89 and 0.939 mg/100 g DM at a retention time of 20.90 min. The Apigegen was presented in low amounts except in ZBK cv with 4.85 mg/100 g DM. According to ARVANITI et al. (2019), Gallic acid, and Chlorogenic acid were the most predominant phenolic acids in dried figs, while Rutin, Quercetin-3-O-rutinoside, and Epicatechin exhibited the highest concentration levels among flavonoids. Comparing these compounds to those of fresh figs, the same target compounds were found, but with high levels (ARVANITI et al., 2019). Many factors were responsible for these variations; The ripening phase and drying process used seem to significantly affect the content of phytochemical compounds (PEREIRA et al., 2017; ARVANITI et al., 2019), as well as the nature of the solvent used for extraction, the fruit color, the harvest season, the weather conditions, and the fruit variety (SEDAGHAT and RAHEMI, 2018).

Moreover, the dark-colored varieties (SD and ZBK cultivars) had higher phenolic compounds compared to those from light-colored ones (BD and BBK cultivars). Furthermore, it should be emphasized that dark-colored dried figs have the highest content of phytochemicals and exhibited the highest antioxidant capacity due to these compounds.

According to POURGHAYOUMI et al. (2017) and based on our current results, dried figs own one of the highest concentrations of polyphenols among the commonly consumed fruits. In addition, the dark-colored dried varieties, especially the SD and ZBK cultivars, harvested in South-Eastern Tunisia, represent a rich source of two phytochemical classes such as phenolic acids and flavonoids and can offer valuable value for food and pharmaceutical industries, and should be recommended as a natural and healthy food product.

Fatty acids determination (FAMES)

FAMES composition is shown in Tab. 4. The total lipid content (TLC) analyzed for the cultivars studied, expressed in gram per 100 g of dry matter (g/100 g DM), was in the order of 1.5 g/100 g. This

Tab. 3: Phenolic acid and flavonoid profiles in the nine Tunisian dry fig cultivars.

Phenolic contents	m/z	ZBK		KHK		WBA		SD		BBK	
		CC (mg/100g)	Rt (min)	CC (mg/100g)	Rt (min)	CC (mg/100g)	Rt (min)	CC (mg/100g)	Rt (min)	CC (mg/100g)	Rt (min)
Phenolic acids											
Gallic acid	169	0.47 ± 0.02d	4.14	0.12 ± 0.07b	4.14	0.54 ± 0.14e	4.14	0.09 ± 0.01ab	4.12	0.082 ± 0.00a	4.13
Protocatechuic acid	153	4.47 ± 0.23e	7.33	1.85 ± 0.05bc	7.31	10.59 ± 0.13f	7.32	1.96 ± 0.10c	7.32	1.91 ± 0.02c	7.32
4-O-Caffeoylquinic acid	353	0.92 ± 0.03c	12.70	2.42 ± 0.13d	12.71	0.55 ± 0.02b	12.70	0.88 ± 0.05c	12.71	0.27 ± 0.02a	12.75
p-Coumaric acid	163	0.36 ± 0.01b	22.20	0.39 ± 0.01bc	22.18	0.46 ± 0.01cd	22.19	0.89 ± 0.04f	22.20	0.68 ± 0.06e	22.22
trans Cinnamic	147	0.49 ± 0.08b	33.56	0.61 ± 0.03bcd	33.56	0.58 ± 0.02bc	33.60	0.85 ± 0.16e	33.61	0.41 ± 0.03ab	33.55
trans Ferulic acid	193	0.73 ± 0.05de	24.45	0.53 ± 0.05c	24.46	0.46 ± 0.00c	24.46	0.79 ± 0.03e	24.43	0.25 ± 0.01b	24.50
Salviolinic acid	515	N.D	-	N.D	-	0.89 ± 0.00b	29.63	0.94 ± 0.00c	29.63	N.D	-
Flavonoids											
Rutin	609	34.68 ± 0.74g	25.23	22.34 ± 0.62f	25.35	10.28 ± 0.10cd	25.36	38.05 ± 1.75h	25.36	9.29 ± 0.24c	25.37
Luteolin-7-o-glucoside	447	0.43 ± 0.03f	26.07	0.18 ± 0.01d	26.06	0.07 ± 0.01bc	26.06	0.08 ± 0.01c	26.06	0.02 ± 0.00a	26.04
Naringin	579	0.02 ± 0.01b	27.60	0.05 ± 0.00d	27.58	0.03 ± 0.00c	27.59	0.03 ± 0.00c	17.59	0.06 ± 0.01e	27.59
Apigenin-7-o-glucoside	431	0.91 ± 0.06 d	28.53	0.07 ± 0.00ab	28.52	0.02 ± 0.00 a	28.52	N.D	-	N.D	-
Quercetin	301	0.45 ± 0.01b	33.56	1.05 ± 0.03d	33.55	0.24 ± 0.02a	33.56	1.49 ± 0.09e	33.57	0.68 ± 0.05c	33.57
Kaempferol	285	1.13 ± 0.04g	33.56	0.24 ± 0.01e	33.56	0.11 ± 0.01c	33.57	0.10 ± 0.01c	33.60	0.08 ± 0.03bc	33.58
Naringenin	271	0.43 ± 0.02d	35.62	0.24 ± 0.03c	35.61	0.17 ± 0.01b	35.62	0.06 ± 0.01a	35.62	0.20 ± 0.02bc	35.63
Apigenin	269	4.85 ± 0.21d	36.22	0.15 ± 0.01ab	36.21	0.23 ± 0.03ab	36.22	0.05 ± 0.01ab	36.29	0.04 ± 0.00ab	36.26
Luteolin	285	0.07 ± 0.01c	36.67	0.08 ± 0.02c	36.66	N.D	-	0.15 ± 0.01d	36.66	N.D	-
Cirsiliol	329	1.79 ± 0.05bc	37.29	1.23 ± 0.06a	37.30	1.46 ± 0.17abc	37.29	2.72 ± 0.07d	37.30	1.89 ± 0.40c	37.30
Phenolic contents	m/z	BD		HBK		MK		HBA			
		CC (mg/100g)	Rt (min)	CC (mg/100g)	Rt (min)	CC (mg/100g)	Rt (min)	CC (mg/100g)	Rt (min)		
Phenolic acids											
Gallic acid	169	0.07 ± 0.01a	4.10	0.08 ± 0.01a	4.16	0.12 ± 0.01b	4.12	0.38 ± 0.01c	4.15		
Protocatechuic acid	153	0.95 ± 0.02a	7.32	2.60 ± 0.13d	7.32	1.50 ± 0.06 b	7.31	4.28 ± 0.15e	7.34		
4-O-Caffeoylquinic acid	353	0.10 ± 0.01a	12.73	2.33 ± 0.10d	12.72	0.54 ± 0.00 b	12.71	0.25 ± 0.02a	12.72		
p-Coumaric acid	163	0.49 ± 0.01d	22.20	0.36 ± 0.01b	22.23	0.32 ± 0.01 b	22.23	0.23 ± 0.01a	22.22		
trans Cinnamic	147	0.28 ± 0.01a	33.57	0.71 ± 0.01cde	33.51	0.42 ± 0.03 ab	33.58	0.79 ± 0.06de	33.57		
trans Ferulic acid	193	0.42 ± 0.07c	24.45	N.D	-	0.68 ± 0.01d	24.45	N.D	-		
Salviolinic acid	515	N.D	-	N.D	-	N.D	-	N.D	-		
Flavonoids											
Rutin	609	2.17 ± 0.16a	25.36	17.99 ± 0.14e	25.35	12.19 ± 0.23d	25.35	6.87 ± 0.33b	25.37		
Luteolin-7-o-glucoside	447	0.04 ± 0.00ab	26.03	0.28 ± 0.01e	26.06	0.04 ± 0.00a	26.02	0.24 ± 0.01e	26.08		
Naringin	579	N.D	-	N.D	-	0.02 ± 0.01b	27.59	0.01 ± 0.01a	27.62		
Apigenin-7-o-glucoside	431	N.D	-	0.08 ± 0.01bc	28.52	N.D	-	0.14 ± 0.01c	28.55		
Quercetin	301	0.25 ± 0.00a	33.55	0.45 ± 0.01b	33.54	0.95 ± 0.03 d	33.58	0.28 ± 0.01a	33.57		
Kaempferol	285	0.02 ± 0.00a	33.58	0.17 ± 0.00d	33.56	0.041 ± 0.00ab	33.58	0.43 ± 0.01f	33.60		
Naringenin	271	0.01 ± 0.01a	35.63	0.17 ± 0.03b	35.61	0.17 ± 0.01b	35.63	0.22 ± 0.02 bc	35.65		
Apigenin	269	N.D	-	0.25 ± 0.04b	36.22	N.D	-	0.53 ± 0.04c	36.25		
Luteolin	285	N.D	-	0.04 ± 0.01b	36.68	0.15 ± 0.01d	36.67	N.D	-		
Cirsiliol	329	0.95 ± 0.04a	37.31	1.49 ± 0.18abc	37.30	1.27 ± 0.09ab	37.31	1.38 ± 0.10abc	37.33		

Note: Mean values ± SE, followed by different letters in the same column indicate significant difference at $P < 0.05$ according to Duncan's multiple range test. Rt: Retention time; CC: Concentration (mg/100 g); N.D: not detected

content varied between 1 g/100 g and 2.25 g/100 g. These levels are slightly higher than those indicated by VIDAUD et al. (1997) and BOSTAN et al. (1998) who estimated that the content was 1 g/100 g. The analysis of fatty acids presented in Tab. 3 led to the following results; Palmitic acid (C16: 0) has an average content of 0.27 g/100 g DM, the maximum content of which is detected in HBA cv and the lowest in HBK cv of a value equal to 0.91 g/100 g DM and 0.15 g/100 g DM respectively for these two cultivars. The average content of Oleic acid (C18: 1) is equal to 0.31 g/100 g DM, the highest of which is detected in HBA cv (0.51 g/100 g DM), Linoleic acid (C18: 2) has an average of 0.35 g/100 g DM. The high content of Linolenic acid (C18: 3) is observed in WBA cv (0.79 g/100 g), Ara-

chidic acid is present in all cultivars studied but in trace form. Its average content is low and equal to 0.08 g/100 g DM. Linolenic and Linoleic acid were the dominant saturated fatty acids in all samples. These levels are higher than those estimated in the table of nutritional composition of CIQUAL FOODS (2013) (Palmitic acid 0.118 g/100 g DM, Linoleic acid 0.39 g/100 g DM, Oleic acid 0.18 g/100 g DM, Arachidic acid absent). Fatty acids play a key role in metabolism as a metabolic fuel, a necessary component of all membranes and as a regulator of genes. In addition, fatty acids are used for many industrial purposes. The level of vitamin E generally depends on the part of the plant. Indeed, this content is higher in vegetable leaves and especially those that are green when ripe (CHUN et al., 2006).

Tab. 4: Fatty acid contents and composition (g/100g DM) of studied dry fig cultivars.

Cultivar	Lipids	SFA		MUFA C18:1	PUFA	
		C16:0	C20:0		((C18:2) 9, 12)	((C18:3) 9, 12, 15)
HBK	1.41 ± 0.04b	0.152 ± 0.01a	0.02 ± 0.00ab	0.32 ± 0.01de	0.41 ± 0.01d	0.43 ± 0.01d
ZBK	1.60 ± 0.02c	0.163 ± 0.00ab	0.04 ± 0.02ab	0.27 ± 0.01cd	0.47 ± 0.03f	0.60 ± 0.02e
BBK	1.73 ± 0.09c	0.190 ± 0.01abc	0.04 ± 0.01ab	0.38 ± 0.01e	0.46 ± 0.01ef	0.56 ± 0.01e
WBA	2.06 ± 0.07d	0.180 ± 0.01abc	0.04 ± 0.00a	0.38 ± 0.02e	0.57 ± 0.01g	0.79 ± 0.03f
SD	1.39 ± 0.02b	0.153 ± 0.00a	0.01 ± 0.00a	0.33 ± 0.00de	0.42 ± 0.00de	0.40 ± 0.01d
MK	1.01 ± 0.03a	0.203 ± 0.00bc	0.01 ± 0.02d	0.24 ± 0.03bc	0.19 ± 0.00ab	0.07 ± 0.03a
HBA	2.25 ± 0.05e	0.913 ± 0.02e	0.28 ± 0.09e	0.51 ± 0.01f	0.23 ± 0.01c	0.23 ± 0.01b
BD	1.00 ± 0.03a	0.256 ± 0.00d	0.21 ± 0.01c	0.17 ± 0.01a	0.21 ± 0.00bc	0.30 ± 0.01c
KHK	1.12 ± 0.04a	0.223 ± 0.01cd	0.13 ± 0.02d	0.19 ± 0.02ab	0.15 ± 0.00a	0.12 ± 0.01a
Total	1.5 ± 0.05	0.27 ± 0.04	0.08 ± 0.13	0.31 ± 0.02	0.35 ± 0.02	0.39 ± 0.04
ANOVA	68.92**	262.15***	404.21***	27.26**	104.59***	124.31***

SFA: Saturated fatty acids; MUFA: Monounsaturated fatty acids; PUFA: Polyunsaturated fatty acids

One-way ANOVA ($\alpha = 0.05$) is expressed in F-values followed by the significance level (***) at $P < 0.05$. Mean values \pm SE, followed by different letters in the same column indicate significant difference at $P < 0.05$ according to Duncan's multiple range test.

Chemometric analysis: Multivariate classification of dried fig cultivars studied

The weight and the impact of the different tested parameters in the dry fig cultivars classification were established using the PCA technique. The PCA relies on the extraction of the most important quantitative variables in a data table representing the observations called 'components', in order to construct the pattern of similarity between the variables and the observations as points on a map (SAPORTA and NIANG, 2009). In the PCA, a multivariate technique was used to distinguish between different cultivars in terms of phytochemical contents (TPC, TFC), and their antioxidant capacity (DPPH and ABTS assays) on the one hand, and the phenolic composition, such as phenolic acids and flavonoids, and fatty acids, on the other hand, to establish the possible grouping of the dried fig cultivars studied. The cluster analysis was performed on the basis of the first two main PCs (PC1 and PC2) (Fig. 4). A total of 55.42% of the whole varia-

bility was explained by the relationship between PC1 vs PC2. PC1 was responsible for 32.61%, and represented the most important component which was positively related to the following variables: Phenolic acids, flavonoids, TPC, TFC, antioxidant capacity using both ABTS and DPPH assays, C18:2, C18:3, and C18:1, that were positively correlated with SD and ZBK cultivars and were therefore primarily responsible for the discrimination of dark dried figs. However, light dried figs were negatively associated with PC1 and positively with PC2 which was responsible for 22.81% of the difference. In addition, both PC1 and PC2 were negatively related with C16:0 and C20:0.

The cumulative variance of 55.42% shows that the fig cultivars were well discriminated by their biologically active compounds levels, fatty acids and their TAC. In fact, PC1 allowed differentiation between dark and green skinned cultivars (Fig. 4). The dispersion of the cultivars on the biplot defined by the first two components revealed

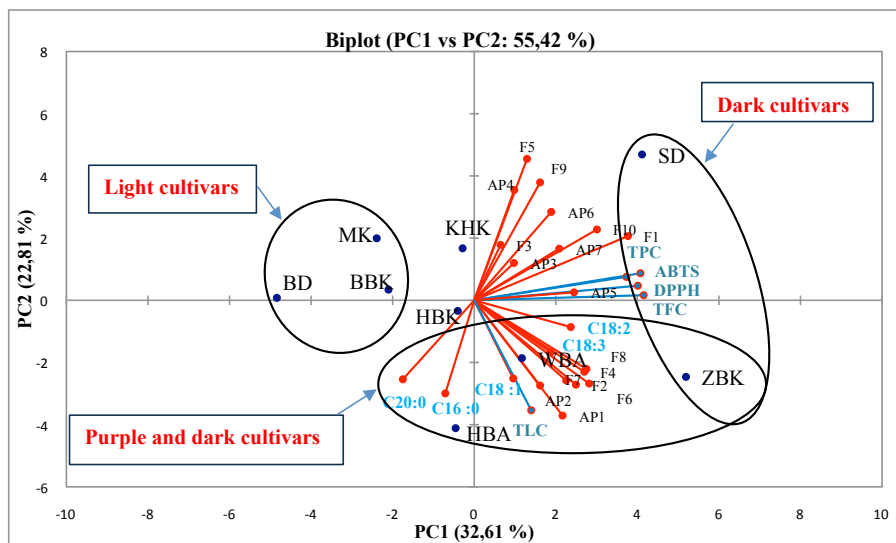


Fig. 4: Principal component analyses of nine dry fig Tunisian cultivars. Flavonoids: Rutin (F1), Luteolin-7-o-glucoside (F2), Naringin (F3), Apigenin-7-o-glucoside (F4), Quercetin (F5), Kaempferol (F6), Naringenin (F7), Apigenin (F8), Luteolin (F9), Cirsiliol (F10), and phenolic acids: Gallic acid (AP1), Protocatechuic acid (AP2), 4-O-Caffeoylquinic acid (AP3), p-Coumaric acid (AP4), trans Cinnamic acid (AP5), trans Ferulic acid (AP6), Salvianolic acid (AP7).

a strong heterogeneity among cultivars. Indeed, the cultivars are significantly individualized. The graphical distribution of the dried fig cultivars presented in Fig. 4, depending on their factor scores, indicates that the dark cultivars were distinguished from light ones; the first group on the right side of the graph includes SD and ZBK cultivars, while the second on the left includes BD, BBK and MK cultivars, and the third one contains HBA, ZBK and WBA cultivars in the center of the graph.

The highest values of TP, TF and AC were achieved on dark colored figs (SD and ZBK cultivars) that showed different flavonoids and phenolic acids in the polyphenol profile which are antioxidant compounds. Our results are consistent with those of BEY and LOUAILECHE (2015) who reported that PCA plot shows a clear separation between the dark and light dried fig cultivars. The highest values of TLC and fatty acids were detected in the HBA, ZBK and WBA cultivars, which contained also fairly high levels of TP, TF and TAC. The third group which included BD, BBK and MK cultivars was poor in all compounds.

In our research, using 24 parameters relative to bioactive compounds, fatty acids and antioxidant activity, PCA applied to the considered fig cultivars proved that three phenotype groups of Tunisian dried figs (black, green, and purple) can be distinguished with significant differences. Obtained results showed that dark fig (ZBK) cv, harvested from the South-East, is a relevant food, which provides interesting antioxidant compounds in sufficient amounts to the human diet; especially samples rich in Rutin (F1), Cirsiliol (F10), Apigenin (F8) and Kaempferol (F6) (the black cultivars), characterized by a large amount of polyphenols, mainly flavonoids, and fatty acids, particularly polyunsaturated ones.

Conclusions

The nine fig cultivars studied herein have a huge potential and constitute an important heritage that deserves to be valued. They are characterized by an attractive range of fruit skin colors, ranging from dark black to green-yellow. Dark cultivars contained the highest levels of flavonoids and phenolics and exhibited high antioxidant capacity, while light skinned cultivars contained the lowest levels. The determined total phenolic content indicates the richness of Southern Tunisian figs, in particular SD and ZBK cultivars. In these, main compounds were Rutin, Protocatechuic acid, Cirsiliol, 4-O-Caffeoylquinic acid, Kaempferol and Quercetin. These dried fruits also contained fatty acids, mainly polyunsaturated. Therefore, dried figs, notably SD and ZBK cultivars, can be recommended as a natural healthy food product to be added to the diet by dieticians and nutritionists, thus allowing their labeling as a superior product.

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