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Volatile Compounds from Six Varieties of *Ficus carica* from Tunisia

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Abstract: Aroma is one of the essential parameters for the evaluation of fruit quality and consumer acceptance, with volatile components being determinant for this characteristic. During this work, the volatile profile of fresh fruits (pulp and peel) and leaves of Tunisian *Ficus carica* L. white (''Bither Abiadh'', ''Bidi'') and dark (''Bither Kholi'', ''Himri'', ''Kholi'' and ''Tchich Asal'') varieties were characterised by GC and GC-MS. The major components detected among the volatiles of leaves were cedrol (38.9%), manoyl oxide (24.8%), α-terpineol acetate (21.7%), abietatriene (11.8%), γ-muurolene (7.4%), α-pinene (6.1%), pentadecanal (5.2%) and nonadecanal (2.3%). The major components detected in the volatiles of the fruits were cedrol (43.8%), α-terpinyl acetate (22.5%), manoyl oxide (12.9%), α-pinene (9.3%), abietadiene (8.1%),*trans*-calamenene (3.9%) and *n*-heneicosane (3.5%).The results suggest that the varieties could be distinguished on the basis of their volatile fractions composition.

Keywords: *Ficus carica*; leaves; fruits; aromatic compounds; volatiles.

1. Introduction

Thousands of plants are well known in traditional medicine system for their curative and therapeutic potentials worldwide; one of them, *Ficus carica* is a deciduous tree belonging to the Moraceae family. It is one of the earliest cultivated fruit trees and an important crop worldwide for both dry and fresh consumption [1–3]. Its fruits, roots and leaves are used in the folk medicine for the treatment of different ailments, such as gastrointestinal (colic, ulcers, indigestion, loss of appetite and diarrhoea), respiratory (sore throats, coughs and bronchial problems), inflammatory, furuncles, cancer and cardiovascular disorders [4, 5]. The formation of volatile compounds in fruits is a dynamic process, and generally the typical flavour of most of them is not present at harvest, but develops during the ripening process. Volatile compounds present in fresh and processed fruits significantly affect their flavour and aroma quality, which is formed by a complex group of chemical substances [6]. The variability in aroma compounds has been reported to be dependent on climatological conditions, cultivar, maturity and technological factors, such as harvest, post-harvest treatments, processing and storage conditions [7–10].To sampling the volatile compounds, various methods have been developed, including hydrodistillation, supercritical fluid extraction and headspace solid phase micro-extraction (HS-SPME). The objective of this study was to extract the volatiles from the leaves and fruits of *F. carica*, using a simultaneous hydrodistillation and solvent-extraction in a Clevenger-type apparatus, and to identify the fragrant constituents by GC-MS.

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2. Materials and Methods

2.1. Plan material

Ficus carica white (‘‘Bither Abiadh” and ‘‘Bidi”) and dark (‘‘Bither Kholi”, ‘‘Himri”, ‘‘Kholi”, and ‘‘Tchich Asal”) varieties were collected at the same stage of ripeness. The fruits and leaves were gathered in Mahdia region (Central East of Tunisia) in August 2014. All materials were immediately transported to the laboratory of Biochemistry of Faculty of Medicine of Monastir University.The edible parts of the fruits were manually prepared, without using steel knives. Fruits and leaves were homogenised, weighted and separated by aliquots. All the fresh samples were then processed.

2.2. Extraction of volatiles and GC–MS analysis

Each sample (100 g) of fresh fruits and leaves were separately subjected to hydrodistillation for 4 h using a Clevenger-type apparatus (Clevenger, 1928). The volatiles obtained after trapping in hexane were dried over anhydrous sodium sulphate; the solvent was evaporated and concentrated under a gentle stream of N_2 and stored at −20 °C until further analysis. GC analyses were accomplished with an HP-5890 series II instrument equipped with a DB-5 capillary column (30 m × 0.25 mm, 0.25 µm film thickness), working with the following temperature program: 60 °C for 10 min, ramp of 5 °C/min to 220 °C; injector and detector temperatures, 250 °C; carrier gas, nitrogen (1 mL/min); detector FID; split ratio 1:30; injection, 0.5 µL. The identification of the components was performed by comparison of their retention times with those of pure authentic samples and by means of their linear retention indices (LRI) relative to the series of *n*-hydrocarbons. Gas chromatography–electron impact mass spectrometry (GC–EIMS) analyses were performed with a Varian CP 3800 gas chromatograph (Varian, Inc. Palo Alto, CA) equipped with a DB-5 capillary column (Agilent Technologies Hewlett-Packard, Waldbronn, Germany; 30 m × 0.25 mm, coating thickness 0.25 µm) and a Varian Saturn 2000 ion trap mass detector. Analytical conditions were as follows: injector and transfer line temperature at 250 and 240 °C, respectively; oven temperature was programmed from 60 to 240 °C at 3 °C/min; carrier gas, helium at 1 mL/min; split ratio 1:30.

Identification of the constituents was based on comparison of the retention times with those of the authentic samples, comparing their LRI relative to the series of *n*-hydrocarbons, and on computer matching against commercial [NIST 2014(U.S. National Institute of Standards and Technology) and ADAMS (Adams, 2007)] and homemade library mass spectra built from pure substances and components of known samples and MS literature data [11–16].

3. Results and Discussion

The results of GC-MS analysis of the volatile compounds present in the six Tunisian varieties of *Ficus carica* leaves and fruits (peels and pulps) obtained by hydrodistillation are given in Table 1. A total of 92 volatile compounds belonging to eight chemical classes were identified; namely, 19 non-terpene derivatives, five monoterpene hydrocarbons, 21 oxygenated monoterpenes, 17 sesquiterpene hydrocarbons, 21 oxygenated sesquiterpenes, four diterpene hydrocarbons, two oxygenated diterpenes and 3 apocarotenes. Identification accounted for 78.0 to 95.2% of total volatiles. To the best of our knowledge, this is the first report involving the above-mentioned varieties and comparing the volatile composition of *F. carica* leaves and fruits.

Comparing the fruits and leaves samples of Himri, Bither Abiadh, Kholi, Tchich Asal, Bidhi, and Bither Kholi varieties, it can be observed that their volatiles composition was quite similar, even if some qualitative and quantitative differences can be noticed. Also, the number of total volatiles in the leaves was higher than in fruits.

Many more volatiles for both leaves and fruits of *F. carica* have been reported in the present study than in previous ones, with 21 constituents for the leaves and 17 for the fruits. The leaves and fruits were characterized by some different constituents, but others were shared (Table 1). The major components detected among the leaves volatiles were cedrol (38.9%), manoyl oxide (24.8%), α-terpinyl acetate (21.7%), abietatriene (11.8%), γ-muurolene (7.4%), α-pinene (6.1%) and pentadecanal (5.2%). In the case of the fruits, they were cedrol (43.8%), α-terpinyl acetate (22.5%), manoyl oxide (12.9%), α-pinene (9.3%) and abietadiene (8.1%). In general, all the varieties showed a similar profile, although some differences were apparent. In particular, this was true for the fruits samples. They presented almost all the classes of compounds, with the only exceptions of oxygenated diterpenes, diterpenes hydrocarbons and apocarotenes. On the contrary, such classes were most represented in the leaves samples.

Oxygenated sesquiterpenes were the main class of compounds in *F. carica* fruits, except for the 'Bither Kholi' and 'Bither Abiadh' varieties; however, in these samples an oxygenated sesquiterpene, cedrol, was the main compound.

A similar behaviour was observed for the leaves samples. In contrast, Oliveira et al. [17] reported some sesquiterpene hydrocarbons, i.e. germacrene D, β-caryophylene and **τ**-elemene as the major constituents in leaves of *F.carica*. Sesquiterpenes, such as β- and α-caryophyllene are known to be released in response to attacks by *Spodopteraexigua* larvae in cotton plantlets. This seems to support the hypothesis that these compounds may be important in the defence against insects [18].

In fact, the aroma of fruits and leaves is the result of a complex mixture of esters, alcohols, aldehydes, terpenoids compounds and others, at low concentrations that reach the olfactory epithelium, strongly contributing to the taste of foods [19]. Many of the identified compounds certainly contribute to different extents to the pleasant aroma and taste of fig fruits.

Hydrodistillation in a Clevenger-type apparatus combined with solvent-extraction as been revealed to be a useful method to identify and determine the volatiles from the leaves and fruits of *F. carica*. A total of 21 constituents have been identified in the leaves and 17 in the fruits samples. 54 volatiles are shared by leaves and fruits. Further study may evaluate the application of other techniques, such headspace SPME analysis, for a better characterization of the volatile constituent and other studies may be addressed for the search of some new bioactive components in various parts of *F. carica*.

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Table 1. Volatile constituents of leaves and fruits of *Ficus carica*.

		Himri		**Bither Abiadh**		**Kholi**		**Tchich Asal**		**Bidi**		**Bither Kholi**	
Constituents	**l.r.i.**[a]	***Fruits***	***Leaves***	***Fruits***	***Leaves***	***Fruits***	**Leaves**	***Fruits***	***Leaves***	***Fruits***	***Leaves***	***Fruits***	***Leaves***
Non-terpene derivatives													
(E)-3-hexen-1-ol	853	-	-	-	-	-	0.4	-	-	-	-	-	-
benzaldehyde	962	-	-	0.5	-	-	-	-	-	-	-	0.6	-
2-pentyl furan	993	0.3	-	1.1	-	-	-	-	-	-	-	0.9	-
(E)-2-octenal	1063	-	-	0.5	-	-	-	-	-	-	-	0.3	-
nonanal	1102	-	-	0.6	-	-	-	-	0.2	0.2	-	0.3	-
methyl salicylate	1192	0.3	-	-	-	-	-	-	-	-	-	-	-
(E.Z)-2.4-decadienal	1293	-	-	-	-	-	-	-	-	-	-	0.3	-
4-vinylguaiacol	1313	-	-	-	-	-	-	-	-	-	0.3	-	-
(E.E)-2.4-decadienal	1316	0.9	-	1.8	-	0.3	-	-	-	0.3	-	1.2	-
(Z)-3-hexenyl benzoate	1572	-	-	-	-	-	0.4	-	-	-	-	-	-
pentadecanal	1716	-	-	-	5.2	-	-	-	-	-	0.4	-	0.4
ambroxide	1750	-	-	0.4	-	0.4	-	-	-	0.2	-	-	0.2
methyl hexadecanoate	1931	0.6	-	0.8	-	0.6	-	-	-	0.4	-	0.7	-
hexadecanoic acid	1971	1.8	-	0.8	-	0.7	-	-	-	-	-	1.1	-
n-eicosane	2000	0.4	-	-	-	-	-	-	-	-	-	-	-
isopropyl hexadecanoate	2025	-	-	0.5	-	-	-	-	-	-	-	-	-
methyl linoleate	2099	0.5	0.3	-	-	-	-	-	-	-	-	-	-
n-heneicosane	2100	3.5	1.0	-	-	-	-	-	-	-	-	-	-
nonadecanal	2110	-	2.3	-	-	-	-	-	-	-	-	-	-
% identified		**8.3**	**3.6**	**7.0**	**5.2**	**2.0**	**0.8**	**0.0**	**0.2**	**1.1**	**0.7**	**5.4**	**0.6**
Monoterpene hydrocarbons													

α-pinene	941	2.0	0.4	2.8	-	0.9	1.3	1.0	6.1	4.5	3.2	9.3	0.3
camphene	955	-	-	-	-	-	-	-	0.2	0.2	-	-	-
β-pinene	982	-	-	-	-	-	-	-	0.3	0.3	-	0.5	-
δ-3-carene	1013	-	-	-	-	-	-	0.3	1.7	0.4	-	-	-
limonene	1032	-	-	-	-	-	-	-	0.2	-	-	-	-
% identified		**2**	**0.4**	**2.8**	**0.0**	**0.9**	**1.3**	**1.3**	**8.5**	**5.4**	**3.2**	**9.8**	**0.3**
Oxygenated monoterpenes													
linalool	1101	-	0.4	-	-	0.4	-	1.7	0.8	-	-	0.5	-
cis-*p*-menth-2-en-1-ol	1123	-	-	-	-	-	-	-	0.2	-	0.3	-	-
trans-*p*-menth-2-en-1-ol	1142	-	-	-	-	-	-	-	0.2	-	0.3	-	-
trans-verbenol	1144	0.4	-	-	-	0.3	0.4	-	0.2	0.2	0.3	0.4	-
karahanaenone	1155	-	-	-	-	-	-	0.5	0.7	-	-	-	-
umbellulone	1173	-	-	-	-	-	-	-	0.3	-	-	-	-
4-terpineol	1179	0.9	1.4	0.7	-	1.2	0.4	1.8	2.8	1.0	2.8	1.5	0.3
m-cymen-8-ol	1184	-	-	-	-	-	-	-	-	0.2	-	-	-
p-cymen-8-ol	1185	0.7	0.3	0.8	-	0.8	0.7	0.4	0.7	1.2	0.6	0.4	0.4
α-terpineol	1191	1.0	0.7	0.8	-	1.2	0.5	1.5	1.7	0.9	1.4	1.1	0.6
cis-piperitol	1194	-	-	-	-	-	-	-	-	-	0.3	-	-
verbenone	1206	0.3	-	-	-	0.4	-	-	-	0.2	0.3	-	-
(Z)-ocimenone	1233	-	-	-	-	-	0.4	-	-	-	-	-	-
(E)-ocimenone	1241	-	-	-	-	-	0.3	-	-	-	-	-	-
methyl carvacrol	1244	-	-	-	-	-	-	-	0.2	-	-	0.5	-
linalyl acetate	1259	-	-	-	-	-	-	0.5	0.2	-	-	-	-
bornyl acetate	1287	1.4	0.5	1.4	-	1.5	0.4	0.7	0.9	0.6	1.2	1.6	0.6
4-terpinyl acetate	1299	2.2	4.0	-	0.6	2.4	-	4.9	2.2	0.2	0.4	0.2	-
trans-piperitol acetate	1342	-	-	-	-	2.4	1.6	3.4	2.4	2.7	1.8	-	0.8

α-terpinyl acetate	1352	14.4	16.5	13.0	13.2	19.2	8.3	22.5	21.7	12.1	11.0	11.7	8.0
geranyl acetate	1383	-	-	-	-	-	-	0.3	-	-	-	-	-
% identified		**21.3**	**23.8**	**16.7**	**13.8**	**29.8**	**13.0**	**38.2**	**35.2**	**19.3**	**20.7**	**17.9**	**10.7**
Sesquiterpene hydrocarbons													
longifolene	1404	0.7	-	0.5	-	0.4	-	-	-	0.6	0.5	0.7	0.3
(Z)-caryophhylene (syn. isocaryophyllene)	1406	-	-	-	-	-	-	-	-	0.2	-	-	-
α-cedrene	1410	0.9	0.4	1.0	-	1.2	-	0.6	0.7	0.6	1.0	1.0	0.4
β-caryophyllene	1419	-	-	-	-	-	6.5	0.6	1.2	0.9	7.3	-	0.3
β-cedrene	1419	1.1	4.5	0.5	0.6	0.8	-	-	-	-	-	0.8	-
aromadendrene	1440	-	-	-	-	-	0.6	-	-	0.3	0.4	-	-
α-humulene	1456	-	0.6	-	-	-	0.6	0.7	0.6	0.5	0.6	0.8	-
alloaromadendrene	1462	-	-	-	-	-	-	-	-	-	0.3	-	-
cis-muurola-4(14).5-diene	1463	-	-	0.6	-	0.2	0.4	0.3	0.3	0.2	0.3	1.0	-
γ-muurolene	1478	-	6.2	0.5	4.1	-	5.7	0.3	3.1	-	7.4	-	3.1
bicyclogermacrene	1496	-	0.9	-	-	0.3	-	-	-	-	1.8	-	-
α-muurolene	1499	-	-	-	-	0.4	-	0.4	0.5	0.6	0.3	1.1	0.2
α-alaskene	1511	-	0.4	1.2	-	1.2	0.5	0.6	0.8	1.1	0.6	-	0.3
trans-γ-cadinene	1514	-	-	-	-	-	-	-	-	-	-	-	0.3
trans-calamenene	1526	0.7	-	2.0	0.9	0.8	1.0	-	-	2.7	0.9	3.9	-
δ-cadinene	1524	-	0.6	-	-	-	-	1.1	2.1	-	-	-	0.6
α-calacorene	1543	-	-	-	-	0.3	-	-	-	-	-	-	-
% identified		**2.0**	**0.4**	**2.8**	**0.0**	**0.9**	**1.3**	**1.3**	**8.5**	**5.4**	**3.2**	**9.8**	**0.3**
Oxygenated sesquiterpenes													
epi-cubebol	1495	-	-	-	-	-	1.5	0.3	0.4	0.4	-	0.5	0.2
germacrene D-4-ol	1575	-	0.3	-	-	0.2	-	0.3	-	0.3	-	-	0.3
spathulenol	1577	-	-	-	-	-	0.3	-	-	-	0.5	-	-

caryophyllene oxide	1582	0.7	1.2	1.7	1.2	0.8	1.9	0.5	0.8	2.2	3.4	2.4	1.3
globulol	1584	-	-	-	0.7	-	-	-	-	-	-	-	-
viridiflorol	1591	-	-	-	-	0.6	0.4	-	-	-	-	-	-
cedrol	1597	43.8	38.9	30.2	30.5	33.3	22.1	36.9	20.7	28.6	29.5	22.1	35.6
humulene epoxide II	1607	0.3	0.4	1.7	0.9	0.6	1.1	0.5	0.3	1.9	0.7	-	0.8
epi-cedrol	1613	0.7	0.6	0.7	0.5	0.6	0.5	-	0.7	-	0.7	0.7	-
1.10-di-*epi*-cubenol	1614	-	-	0.7	-	-	-	-	-	0.7	-	0.9	1.1
humulane-1.6-dien-3-ol	1615	0.7	-	0.5	-	0.7	1.1	1.5	0.7	-	-	1.5	-
1-*epi*-cubenol	1629	-	-	-	-	-	-	1.0	0.8	1.1	-	0.9	-
α-acorenol	1631	0.8	0.8	0.8	0.6	0.7	0.6	0.2	-	-	0.7	-	0.9
β-acorenol	1633	-	-	-	-	-	-	-	-	-	-	-	0.2
T-cadinol	1641	0.4	0.6	2.0	1.2	0.6	1.1	1.0	1.4	4.6	0.8	3.3	0.8
T-muurolol	1642	-	-	0.7	-	0.3	0.4	0.5	0.5	1.2	0.3	0.9	0.5
α-cadinol	1654	0.7	0.9	3.7	5.3	1.1	2.4	2.2	2.6	6.4	1.2	5.8	2.0
cadalene	1675	-	-	-	-	-	-	-	-	-	-	0.3	-
cis-α-santalol	1682	-	-	0.8	4.9	0.6	0.6	0.5	0.9	0.5	0.4		0.9
cis-14-muurol-5-en-4-one	1685	0.6	-	-	-	-	-	-	-	-	-	1.4	-
oplopanoyl acetate	1883	-	-	-	-	-	-	0.2	-	-	-	-	-
% identified		**48.7**	**43.7**	**43.5**	**45.8**	**40.1**	**34.0**	**45.6**	**29.8**	**47.9**	**38.2**	**40.7**	**44.6**
Diterpene hydrocarbons													
pimaradiene	1941	-	-	-	-	-	0.4	-	-	-	-	-	-
sandaracopimara-8(14).15-diene	1962	0.4	0.8	1.9	3.0	1.2	3.8	0.7	0.8	2.1	0.7	0.3	1.7
abetatriene	2055	3.1	4.4	6.5	8.2	2.5	5.5	3.4	2.2	8.1	1.6	2.4	11.8
abetadiene	2081	-	-	-	-	-	0.6	0.6	0.9	0.9	-	-	0.3
% identified		3.5	5.2	8.4	11.2	3.7	10.3	4.7	3.9	11.1	2.3	2.7	13.8
Oxygenated diterpenes													

manoyl oxide	1991	8.0	5.0	10.0	13.6	12.9	24.8	2.5	8.6	3.2	9.3	5.7	19.7
epi-13-manoyl oxide	2009	-	-	-	-	-	-	-	-	-	-	-	0.5
% identified		**8.0**	**5.0**	**10.0**	**13.6**	**12.9**	**24.8**	**2.5**	**8.6**	**3.2**	**9.3**	**5.7**	**20.2**
Apocarotenes													
β-cyclocitral	1222	-	-	-	0.5	-	-	-	-	-	0.4	-	0.4
(E)-β-damascenone	1382	-	-	-	-	-	-	-	0.3	-	-	0.3	-
(E)-β-damascone	1412	-	-	-	-	-	-	-	0.2	-	-	-	0.3
% identified		**0.0**	**0.0**	**0.0**	**0.5**	**0.0**	**0.0**	**0.0**	**0.5**	**0.0**	**0.4**	**0.3**	**0.7**
Total identified		**93.8**	**82.1**	**91.2**	**90.1**	**90.3**	**85.5**	**93.6**	**95.2**	**93.4**	**78**	**92.3**	**91.2**

[a] *:* linear retention indices

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