

Epicuticular Wax in Developing Olives (*Olea europaea*) Is Highly Dependent upon Cultivar and Fruit Ripeness

Stefania Vichi^{a,*}, Nuria Cortés-Francisco^b, Josep Caixach^c, Gonçal Barrios^d, Jordi Mateu^d, Antonia Ninot^e, Agustí Romero^e

^a Food Science and Nutrition Department, INSA - XaRTA (Catalonian Reference Network on Food Technology), University of Barcelona, Food and Nutrition Torribera Campus, Av. Prat de la Riba, 171. 08921, Santa Coloma de Gramenet, Spain.

^b Laboratory of the Public Health Agency of Barcelona, Av. de les Drassanes, 13. 08001, Barcelona, Spain.

^c Mass Spectrometry Laboratory/Organic Pollutants, IDAEA-CSIC. Jordi Girona, 18-26. 08034, Barcelona, Spain.

^d Section of Agriculture and Plant Health of Tarragona. Ministry of Agriculture, Livestock, Fisheries and Food, Catalan Government. Av. dels Països Catalans, 5-7. 43007 Tarragona, Spain.

^e Olive Production, Oil Processing and Nut Trees, IRTA, Mas de Bover Crta. de Reus, El Morell. 43120, Constantí (Tarragona), Spain.

Table S1. Attribution, exact mass, mass accuracy and relative abundance of the main signals observed in positive UHR mass spectra of olive EW extracted by different solvents.

Theoretical mass ^a	Delta (ppm)	RDB equiv. ^b	Formula [M+NH ₄] ⁺	Tentative identification ^c	Extraction solvent ^d					Theoretical mass	Delta (ppm)	RDB equiv.	Formula [M+NH ₄] ⁺	Tentative identifcation	Extraction solvent						
					1	2	3	4	5						1	2	3	4	5		
ESI+																					
glycerides																					
346.2952	0.2	0.5	C ₁₉ H ₄₀ O ₄ N	MAG					*	850.7858	0.8	2.5	C ₅₃ H ₁₀₄ O ₆ N	TAG	*	*	*	*	*		
372.3108	0.1	1.5	C ₂₁ H ₄₂ O ₄ N	MAG					*	872.7702	1.3	5.5	C ₅₅ H ₁₀₂ O ₆ N	TAG			*	*	*		
374.3265	0.1	0.5	C ₂₁ H ₄₄ O ₄ N	MAG					*	876.8015	0.8	3.5	C ₅₅ H ₁₀₄ O ₆ N	TAG	*	*	*	*	*		
402.3578	0.2	0.5	C ₂₃ H ₄₈ O ₄ N	MAG				*	*	900.8015	1.2	5.5	C ₅₇ H ₁₀₆ O ₆ N	TAG	*	*	*	*	**		
430.3891	0.1	0.5	C ₂₅ H ₅₂ O ₄ N	MAG				*	*	902.8171	0.9	4.5	C ₅₇ H ₁₀₈ O ₆ N	TAG	*	*	*	***	***		
586.5405	0.4	0.5	C ₃₅ H ₇₂ O ₅ N	DAG				*		916.8328	0.8	4.5	C ₅₈ H ₁₁₀ O ₆ N	TAG				*	*		
612.5562	1.0	1.5	C ₃₇ H ₇₄ O ₅ N	DAG				*		928.8328	0.0	5.5	C ₅₉ H ₁₁₀ O ₆ N	TAG					*		
614.5718	0.2	0.5	C ₃₇ H ₇₆ O ₅ N	DAG				*		930.8484	0.7	4.5	C ₅₉ H ₁₁₂ O ₆ N	TAG				*	*		
638.5718	0.5	2.5	C ₃₉ H ₇₆ O ₅ N	DAG				*	*	942.8484	0.9	5.5	C ₆₀ H ₁₁₂ O ₆ N	TAG				*	*		
668.6188	0.7	1.5	C ₄₁ H ₈₂ O ₅ N	DAG				*	*	Wax esters											
764.6763	0.6	3.5	C ₄₇ H ₉₀ O ₆ N	TAG				*		468.4775	0.4	0.5	C ₃₀ H ₆₂ O ₂ N	AE				*			
766.6919	1.4	2.5	C ₄₇ H ₉₂ O ₆ N	TAG				*		476.4462	1.0	3.5	C ₃₁ H ₅₈ O ₂ N	BE			*	*			
768.7076	1.4	1.5	C ₄₇ H ₉₄ O ₆ N	TAG				*		490.4619	0.3	3.5	C ₃₂ H ₆₀ O ₂ N	BE			*	*			
780.7076	0.8	2.5	C ₄₈ H ₉₄ O ₆ N	TAG				*	*	504.4775	0.6	3.5	C ₃₃ H ₆₂ O ₂ N	BE	*	*	***	**	*		
782.7232	1.0	1.5	C ₄₈ H ₉₆ O ₆ N	TAG				*	*	518.4932	0.5	3.5	C ₃₄ H ₆₄ O ₂ N	BE			*	*	*		
792.7076	0.8	3.5	C ₄₉ H ₉₄ O ₆ N	TAG				*	*	532.5088	0.5	3.5	C ₃₅ H ₆₆ O ₂ N	BE	*	*	*	**	*		
794.7232	1.0	2.5	C ₄₉ H ₉₆ O ₆ N	TAG				*	*	546.5245	0.3	3.5	C ₃₆ H ₆₈ O ₂ N	BE				*	*		
796.7389	1.5	1.5	C ₄₉ H ₉₈ O ₆ N	TAG				*	*	550.5558	0.5	1.5	C ₃₆ H ₇₂ O ₂ N	AE				*			
806.7232	0.8	3.5	C ₅₀ H ₉₆ O ₆ N	TAG				*	*	560.5401	0.2	3.5	C ₃₇ H ₇₀ O ₂ N	BE				*	*		
808.7389	1.4	2.5	C ₅₀ H ₉₈ O ₆ N	TAG				*	*	triterpenic acids											
810.7545	1.8	1.5	C ₅₀ H ₁₀₀ O ₆ N	TAG				*	*	474.3942	0.7	5.5	C ₃₀ H ₅₂ O ₃ N	ursolic/oleanolic acid maslinic acid	*	*	***	***			
820.7389	0.8	3.5	C ₅₁ H ₉₈ O ₆ N	TAG				*	*	490.3891	0.6	5.5	C ₃₀ H ₅₂ O ₄ N			****	****	****	***		
822.7545	1.5	2.5	C ₅₁ H ₁₀₀ O ₆ N	TAG				*	*	fatty acids											
824.7702	1.7	1.5	C ₅₁ H ₁₀₂ O ₆ N	TAG				*	*	304.2482	0.7	0.5	C ₁₆ H ₃₄ O ₄ N	diOHC16:0 acid	*	*	*	**	**		
834.7545	0.9	3.5	C ₅₂ H ₁₀₀ O ₆ N	TAG				*	*	332.2795	0.8	0.5	C ₁₈ H ₃₈ O ₄ N	diOHC18:0 acid			*	**	**		
836.7702	1.4	2.5	C ₅₂ H ₁₀₂ O ₆ N	TAG				*	*	360.3108	0.4	0.5	C ₂₀ H ₄₂ O ₄ N	diOHC20:0 acid				*	*		
838.7858	1.4	1.5	C ₅₂ H ₁₀₄ O ₆ N	TAG					*												
846.7545	1.0	4.5	C ₅₃ H ₁₀₀ O ₆ N	TAG				*	*												
848.7702	0.9	3.5	C ₅₃ H ₁₀₂ O ₆ N	TAG				*	*												

ESI-																	
Triterpenic acids									Phenols								
455.3531	0.9	7.5	C ₃₀ H ₄₇ O ₃	ursolic/oleanolic	*	*	*	*	195.0663	0.8	5.5	C ₁₀ H ₁₁ O ₄	hydroxytyrosyl acetate	***	***	***	***
471.3480	1.0	7.5	C ₃₀ H ₄₇ O ₄	maslinic acid	*	*	*	**	241.0718	0.7	5.5	C ₁₁ H ₁₃ O ₆	elenolic acid	*	*	*	*
Fatty acids									319.1187	0.4	8.5	C ₁₇ H ₁₉ O ₆	decarboxymethyl oleuropein	***	***	***	***
367.3582	0.8	1.5	C ₂₄ H ₄₇ O ₂	C ₂₄ :0	*	*	*	*	361.1293	0.9	9.5	C ₁₉ H ₂₁ O ₇	ligstroside aglycone	*	*	*	*
395.3895	0.9	1.5	C ₂₆ H ₅₁ O ₂	C ₂₆ :0	*	*	*	*	377.1242	0.7	9.5	C ₁₉ H ₂₁ O ₈	oleuropein aglycone	****	****	****	****
423.4208	0.5	1.5	C ₂₈ H ₅₅ O ₂	C ₂₈ :0		*	*	*									*

^a: m/z values of NH₄⁺ adduct ions; ^b: Rings and double bond equivalents; ^c: compound class according to the number of carbon, oxygen and hydrogen atoms, according to Vichi et al.²³; ^d: extraction solvent 1: acetone; 2: chlorofom; 3: hexane:dichloromethane 50:50; 4: hexane:dichloromethane 90:10; 5: hexane. *: >0,1%; **>20%; ***>50%; ****>80%

Table 2. Attribution, exact mass, mass accuracy and relative abundance of the main signals observed in negative UHR mass spectra of olive epicuticular waxes extracted by different solvents.

Theoretical mass	Delta (ppm)	RDB equiv.	Formula [M-H] ⁻	Tentative identifcation ^a	1	2	3	4	5	Theoretical mass	Delta (ppm)	RDB equiv.	Formula [M-H] ⁻	Tentative identification	1	2	3	4	5
<i>Acids</i>										<i>Unidentified</i>									
133.0142	1.6	2.5	C ₄ H ₅ O ₅	MALIC ACID?	*					231.0874	1.1	3.5	C ₁₉ H ₁₅ O ₆		*				
455.3531	0.9	7.5	C ₃ OH ₄₇ O ₃	oleanolic acid	*	*	*	**		289.0870	0.7	14	C ₁₉ H ₁₃ O ₃				*	*	
471.3480	1.0	7.5	C ₃ OH ₄₇ O ₄	maslinic acid	*	*	**	**		293.1758	0.4	5.5	C ₁₇ H ₂₅ O ₄				*	*	
<i>Pol. Fragm.</i>										317.0819	1.8	15	C ₂₀ H ₁₃ O ₄					*	
165.0557	1.2	5.5	C ₉ H ₉ O ₃	fragments polyphenols	*	*	*	*		351.1449	1.1	7.5	C ₁₈ H ₂₃ O ₇		*			*	
181.0718	1.2	0.5	C ₆ H ₁₃ O ₆	fragments polyphenols	*	*				387.1144	1.8	2.5	C ₁₃ H ₂₃ O ₁₃		*				
191.0561	1.1	2.5	C ₇ H ₁₁ O ₆	fragments polyphenols	***	***	***			395.3895	0.6	1.5	C ₂₆ H ₅₁ O ₂		*	*	*		
199.0612	0.8	4.5	C ₉ H ₁₁ O ₅	fragments polyphenols	*	*	*	*		423.4208	0.8	1.5	C ₂₈ H ₅₅ O ₂		*	*	*	*	
213.0557	0.2	9.5	C ₁₃ H ₉ O ₃	fragments polyphenols			*	*		431.2439	1.2	8.5	C ₂₅ H ₃₅ O ₆		*	*	*	**	
225.0616	1.1	1.5	C ₇ H ₁₃ O ₈	fragments polyphenols	*	*	*			447.1238	1.1	21	C ₂₉ H ₁₉ O ₅		*	*	*	*	*
227.0772	1.2	0.5	C ₇ H ₁₅ O ₈	fragments polyphenols	*	*	*			473.1817	2.0	12	C ₂₅ H ₂₉ O ₉		*	*			
271.0823	0.8	5.5	C ₁₂ H ₁₅ O ₇	fragments polyphenols				*		499.4521	1.6	5.5	C ₃₄ H ₅₉ O ₂		*			*	
275.0925	0.8	8.5	C ₁₅ H ₁₅ O ₅	fragments polyphenols	*	*	*	*		501.3585	0.5	7.5	C ₃₁ H ₄₉ O ₅		*	*	*	**	
301.1081	1.4	9.5	C ₁₇ H ₁₇ O ₅	fragments polyphenols	*	*	*	*		518.3554	1.6	16	C ₃₈ H ₄₆ O		*	*	*		
307.0823	0.9	8.5	C ₁₅ H ₁₅ O ₇	fragments polyphenols	*	*	*	*		533.3484	1.1	7.5	C ₃₁ H ₄₉ O ₇		*	*	*		
349.1293	1.2	8.5	C ₁₈ H ₂₁ O ₇	fragments polyphenols	*	*	*	*		543.2083	1.9	8.5	C ₂₅ H ₃₅ O ₁₃		*	*			
<i>polyphenols</i>										556.2314	2.0	13	C ₃₀ H ₃₆ O ₁₀		*	*	*		
195.0663	0.8	5.5	C ₁₀ H ₁₁ O ₄	Oh-tyr acet	***	***	***	***		616.1950	0.5	19	C ₃₄ H ₃₂ O ₁₁		*	*	*		
241.0718	0.7	5.5	C ₁₁ H ₁₃ O ₆	ea aglyc	*	*	*	*		632.1899	1.2	19	C ₃₄ H ₃₂ O ₁₂		*	*	*		
319.1187	0.4	8.5	C ₁₇ H ₁₉ O ₆	dcm ol aglyc	***	***	***	***		635.4165	1.4	7.5	C ₃₆ H ₅₉ O ₉		*	*			
361.1293	0.9	9.5	C ₁₉ H ₂₁ O ₇	lig aglyc	*	*	*	*		637.4321	1.4	6.5	C ₃₆ H ₆₁ O ₉		*	*			
377.1242	0.7	9.5	C ₁₉ H ₂₁ O ₈	ol aglyc	****	****	****	****	*	639.2447	1.4	16	C ₃₄ H ₃₉ O ₁₂		*	*			
539.1770	0.9	11	C ₂₅ H ₃₁ O ₁₃	oleuropein	**	*	*			651.4114	0.6	7.5	C ₃₆ H ₅₉ O ₁₀		**	*			
<i>Sugars?</i>										653.4270	1.4	6.5	C ₃₆ H ₆₁ O ₁₀		*	*			
179.0561	1.1	1.5	C ₆ H ₁₁ O ₆	GLUCOSE?	**					697.2502	1.2	17	C ₃₆ H ₄₁ O ₁₄		*	*			
371.1195	0.8	2.5	C ₁₉ H ₂₁ O ₈	sugar?	*					775.4791	1.4	15	C ₄₇ H ₆₇ O ₉		*	*			
373.1351	1.1	1.5	C ₁₃ H ₂₅ O ₁₂	sugar?	*					791.4740	1.0	15	C ₄₇ H ₆₇ O ₁₀		*	*			