16-06-2022

Analysis of Volatile Compounds: a potent multi-faced tool for EVOO quality evaluation

Cecchi, L.,[a] Migliorini, M.,[b] Cane, A.,[b] Giambanelli, E.,[b] Zanoni, B.,[c] Mulinacci, N.,[a]

[a] Dipartimento di NEUROFARBA, Università degli Studi di Firenze

[b] CARAPELLI Firenze S.p.A.

[c] Dipartimento di Gestione dei Sistemi Agrari, Alimentari e Forestali, Università degli Studi di Firenze

INTRODUCTION

the collaboration has been going on for more than 5 years

The collaboration stems to satisfy the needs linked to the main goals of a company leader in the olive oil field.

LEGISLATIVE REQUIREMENTS ➔
- Supporting panel test in virgin olive oil classification according to Reg. CE 2568/91

AUTHENTICATION ➔
- Authentication of the geographic and botanic origin of VOO

QUALITY EVALUATION ➔
- Raw materials selection
- Evolution of volatile compounds over time
- Blends & Products standardisation
- Detection of poor-quality virgin olive oils by only chemical analysis
- Characterization of monocultivar samples

TOOLS ➔
1. Reliable chemical/analytical methods
2. Large data-sets (more than 1000 samples)
3. Suitable statistical tools

THE ANALYTICAL METHOD

Talanta 165 (2017) 641–652

Contents lists available at ScienceDirect

Talanta

journal homepage: www.elsevier.com/locate/talanta

ELSEVIER

Multiple internal standard normalization for improving HS-SPME-GC-MS quantitation in virgin olive oil volatile organic compounds (VOO-VOCs) profile

CrossMark

Martina Fortini[a], Marzia Migliorini[a], Chiara Cherubini[a], Lorenzo Cecchi[b,c,*], Luca Calamai[d]

[a] *PromoFirenze, Azienda Speciale della CCIAA di Firenze, Divisione Laboratorio Chimico, Via Orcagna 70, 50121 Firenze, Italy*
[b] *Dipartimento di NEUROFARBA, Università degli Studi di Firenze, Via Ugo Schiff 6, 50019 Sesto F.no, Firenze, Italy*
[c] *Multidisciplinary Centre of Research on Food Sciences (M.C.R.F.S.- Ce.R.A), Università degli Studi di Firenze, Italy*
[d] *DISPAA, Università degli Studi di Firenze, Piazzale Cascine 28, 50144 Firenze, Italy*

THE STORAGE OF EVOOs UNDER non-ACCELERATED OXIDATIVE CONDITIONS

JOURNAL OF AGRICULTURAL AND FOOD CHEMISTRY

Article

Cite This: *J. Agric. Food Chem.* 2019, 67, 13150−13163

pubs.acs.org/JAFC

New Volatile Molecular Markers of Rancidity in Virgin Olive Oils under Nonaccelerated Oxidative Storage Conditions

Lorenzo Cecchi,[†,‡] Marzia Migliorini,[§] Elisa Giambanelli,[§] Adolfo Rossetti,[§] Anna Cane,[§] and Nadia Mulinacci*[,†,‡]

[†]Dipartimento di NEUROFARBA, Università degli Studi di Firenze, Via Ugo Schiff 6, Sesto F.no, 50019 Firenze, Italia

[‡]Multidisciplinary Centre of Research on Food Sciences (M.C.R.F.S.-Ce.R.A.), Sesto F.no, 50019 Firenze, Italia

[§]Carapelli Firenze S.p.A., Via Leonardo da Vinci 31, Tavarnelle Val di Pesa, 50028 Firenze, Italy

Supporting Information

By studying the evolution of the volatile profile in EVOOs stored under non-accelerated oxidative conditions, it was possible:

- To define volatile molecular markers of rancidity
- To get useful information about EVOO storage over time

AN OVERVIEW OF THE WORKS CARRIED OUT WITHIN THE COLLABORATION

VOLATILE MOLECULAR MARKERS OF RANCIDITY

Correlation between the evolution of 73 VOCs and the rancid defect allowed us to propose the following markers of rancidity for oil stored in different conditions

Sum of the content of:

- Ideal conditions → **Pentanal, hexanal, nonanal, *E*-2-hetpenal, propanoic acid, hexanoic acid**
- Market → **Pentanal, heptanal, nonanal, decanal, *E*-2-heptenal, *E*-2-decenal, *E*,*E*-2,4-heptadienal, *E*,*E*-2,4-decadienal, octane**
- Household → **Pentanal, nonanal, decanal, *E*-2-hetpenal, *E*-2-decenal, *E*,*E*-2,4-heptadienal, nonanol, propanoic acid, octane, 6-methyil-5-hepten-2-one, 1-octen-3-ol**

Simplified marker → Sum of the content of only three molecules
Pentanal, nonanal, *E*-2-heptenal

AN OVERVIEW OF THE WORKS CARRIED OUT WITHIN THE COLLABORATION

THE STORAGE OF OLIVE POMACE IN DIFFERENT AIR/TEMPERATURE CONDITIONS

JOURNAL OF AGRICULTURAL AND FOOD CHEMISTRY

pubs.acs.org/JAFC Article

Volatile Profile of Two-Phase Olive Pomace (Alperujo) by HS-SPME-GC−MS as a Key to Defining Volatile Markers of Sensory Defects Caused by Biological Phenomena in Virgin Olive Oil

Lorenzo Cecchi, Marzia Migliorini, Elisa Giambanelli, Anna Cane, Nadia Mulinacci,* and Bruno Zanoni

Cite This: *J. Agric. Food Chem.* 2021, 69, 5155−5166 Read Online

Research Article SCI

Received: 11 July 2021 Revised: 4 October 2021 Accepted article published: 22 October 2021 Published online in Wiley Online Library: 4 November 2021

(wileyonlinelibrary.com) DOI 10.1002/jsfa.11593

Exploitation of virgin olive oil by-products (*Olea europaea* L.): phenolic and volatile compounds transformations phenomena in fresh two-phase olive pomace ('alperujo') under different storage conditions

Lorenzo Cecchi,[a] Marzia Migliorini,[b] Elisa Giambanelli,[b] Valentina Canuti,[c] Maria Bellumori,[a] Nadia Mulinacci[a*] and Bruno Zanoni[c]

By studying the evolution of the volatile profile in olive pomace stored under different temperature and oxygen exposure, it was possible:

- To define volatile molecular markers of defects of biological origin
- To get useful information for re-using the olive pomace as an ingredient for human diet

AN OVERVIEW OF THE WORKS CARRIED OUT WITHIN THE COLLABORATION

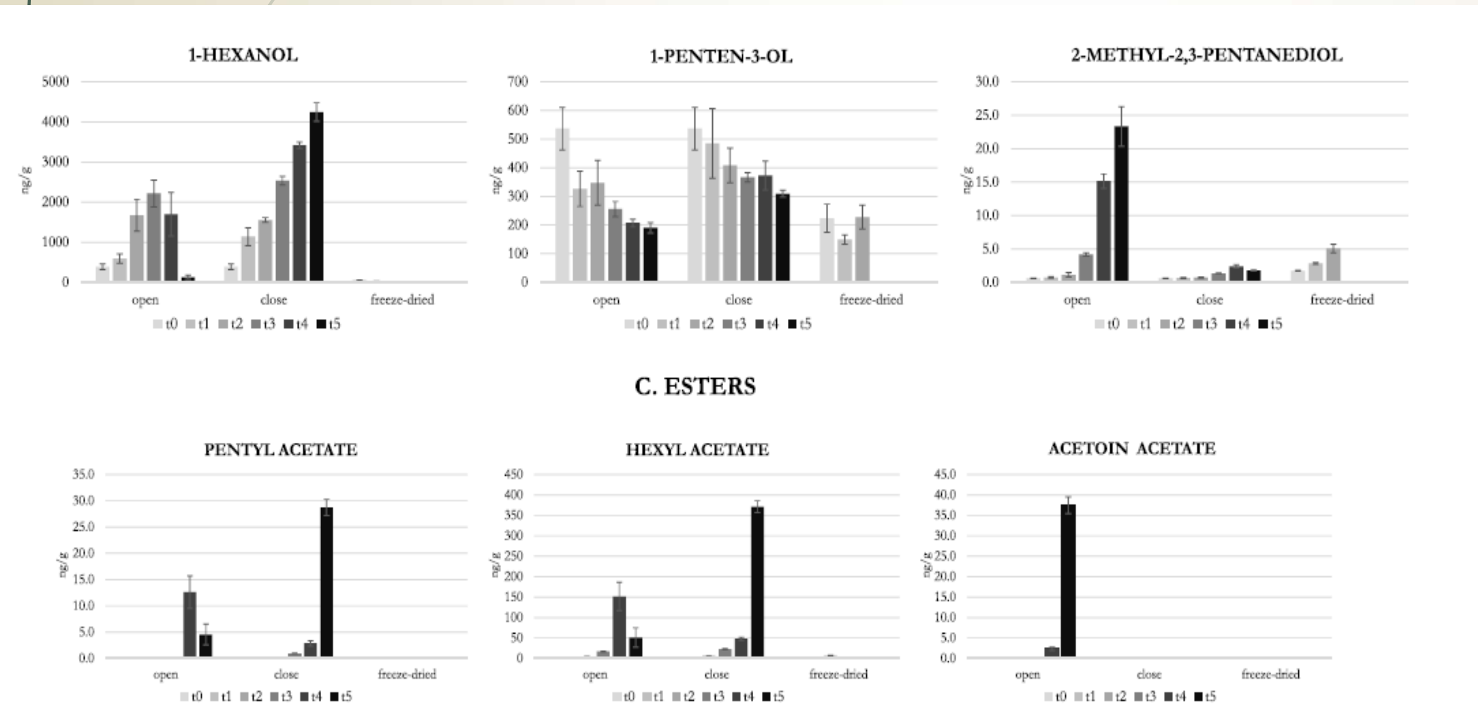
1. **Analysis of olive pomace samples**

a. Freeze-dried

b. In 5 liters containers sealed and stored at 20-23 °C (simulating anaerobic conditions)

c. In 5 liters open containers 20-23 °C (simulating aerobic conditions)

VOCs were analyzed by HS-SPME-GC-MS over 10 days



Some VOCs showed to increase in olive pomace stored in these conditions. We selected these VOCs and investigated on their content in a series of extra virgin olive oil and in a series of virgin olive oils with defectd from microbiological origin

AN OVERVIEW OF THE WORKS CARRIED OUT WITHIN THE COLLABORATION

1. Analysis of olive oil samples

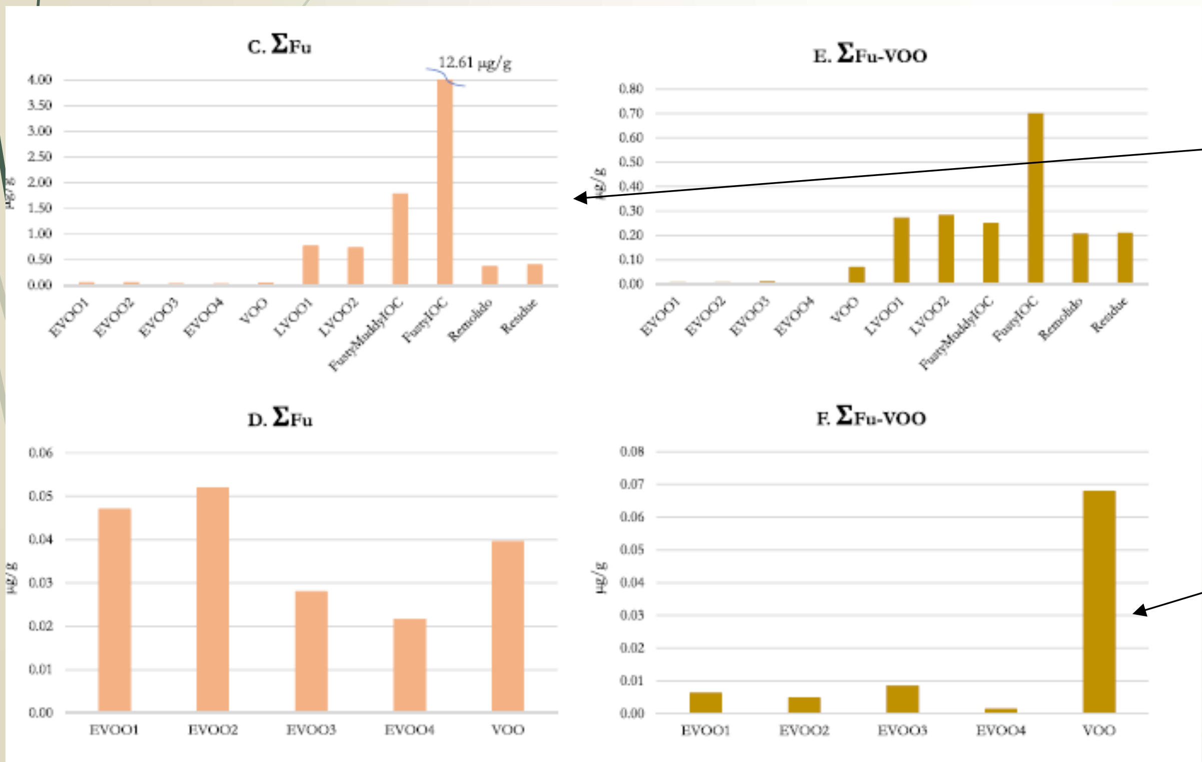
4 Extra virgin olive oils — 1 VOO sample

2 LVOO samples — 1 Remolido sample

1 fusty IOC reference sample — 1 fusty-muddy IOC reference sample

1 sample from the residue precipitated from an unfiltered virgin olive oil

VOLATILE MARKERS OF DEFECTS OF MICROBIOLOGICAL ORIGIN



A group of molecules showed to be able to discriminate among EVOO+VOO and the most defective samples

A second group of molecules present in low concentration also showed to be able to discriminate among EVOO and VOO samples

AN OVERVIEW OF THE WORKS CARRIED OUT WITHIN THE COLLABORATION

1. Analysis of olive oil samples

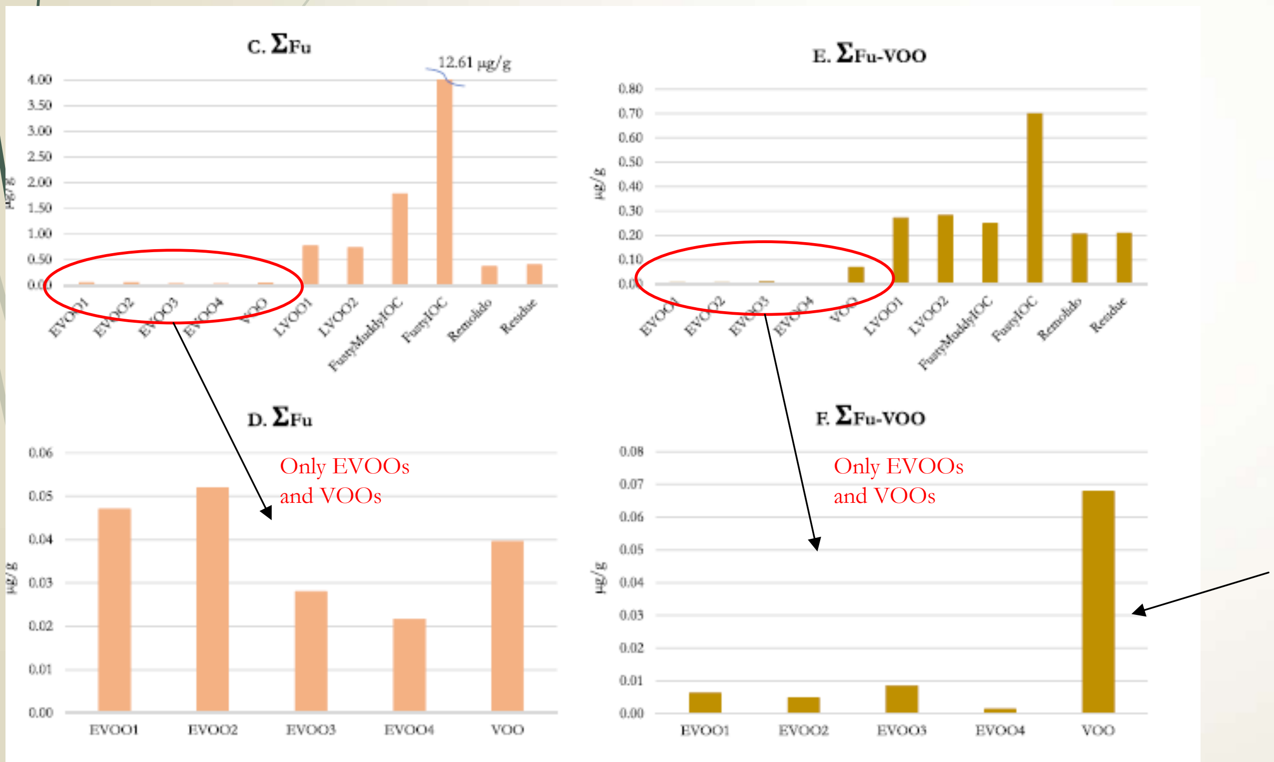
4 Extra virgin olive oils — 1 VOO sample

2 LVOO samples — 1 Remolido sample

1 fusty IOC reference sample — 1 fusty-muddy IOC reference sample

1 sample from the residue precipitated from an unfiltered virgin olive oil

VOLATILE MARKERS OF DEFECTS OF MICROBIOLOGICAL ORIGIN



AN OVERVIEW OF THE WORKS CARRIED OUT WITHIN THE COLLABORATION

VOLATILE MARKERS OF DEFECTS OF MICROBIOLOGICAL ORIGIN

Table 3. List of the VOCs Proposed as Markers for Defects from Biological Origin[a]

marker	VOCs
Σ_{Fu}	1-propanol, 1-butanol, 2-pentanol, 2-heptanol, ethyl propanoate, ethyl 2-methyl propanoate, ethyl pentanoate, methyl hexanoate, ethyl hexanoate, ethyl heptanoate, 2-octanone, propanoic acid, 2-methyl propanoic acid, butanoic acid, pentanoic acid, hexanoic acid
$\Sigma_{Fu\text{-}VOO}$	1-propanol, 1-heptanol, 2-heptanol, 1-octanol, 3-methylbutyl + 2-methylbutyl acetate, pentyl acetate, ethyl propanoate, ethyl 2-methyl propanoate, ethyl butanoate, methyl 3-methyl butanoate, ethyl 3-methyl butanoate, methyl hexanoate, ethyl hexanoate, ethyl benzoate, 2-octanone, acetoin

[a](i) Σ_{Fu} for oils with intense fusty/muddy sediment defect, (ii) $\Sigma_{Fu\text{-}VOO}$ for oils with low-intensity fusty/muddy sediment defect (i.e., the oils were difficult to classify by the panel test).

Only EVOOs and VOOs

AN OVERVIEW OF THE WORKS CARRIED OUT WITHIN THE COLLABORATION

QUALITY CONTROL: SUPPORTING VIRGIN OLIVE OIL CLASSIFICATION

JOURNAL OF AGRICULTURAL AND FOOD CHEMISTRY

Article

Cite This: *J. Agric. Food Chem.* 2019, 67, 9112–9120

pubs.acs.org/JAFC

Headspace Solid-Phase Microextraction–Gas Chromatography–Mass Spectrometry Quantification of the Volatile Profile of More than 1200 Virgin Olive Oils for Supporting the Panel Test in Their Classification: Comparison of Different Chemometric Approaches

Lorenzo Cecchi,[†] Marzia Migliorini,[‡] Elisa Giambanelli,[‡] Adolfo Rossetti,[‡] Anna Cane,[‡] Fabrizio Melani,[†] and Nadia Mulinacci*[,†]

[†]Dipartimento di NEUROFARBA, Università degli Studi di Firenze, Via Ugo Schiff 6, 50019 Sesto Fiorentino, Florence, Italy

[‡]Carapelli Firenze S.p.A., Via Leonardo da Vinci 31, 50028 Tavarnelle Val di Pesa, Florence, Italy

Supporting Information

By studying the volatile profile of more than 1200 virgin olive oils, it was possible to develop 4 approaches for supporting the panel test in virgin olive oil classification as extra virgin, virgin or lampante

AN OVERVIEW OF THE WORKS CARRIED OUT WITHIN THE COLLABORATION

QUALITY CONTROL: ORIGIN AND CULTIVAR AUTHENTICATION

Food Control 112 (2020) 107156

Contents lists available at ScienceDirect

Food Control

journal homepage: www.elsevier.com/locate/foodcont

ELSEVIER

Authentication of the geographical origin of virgin olive oils from the main worldwide producing countries: A new combination of HS-SPME-GC-MS analysis of volatile compounds and chemometrics applied to 1217 samples

Lorenzo Cecchi[a], Marzia Migliorini[b], Elisa Giambanelli[b], Adolfo Rossetti[b], Anna Cane[b], Nadia Mulinacci[a,*], Fabrizio Melani[a]

[a] Dipartimento di NEUROFARBA, Università degli Studi di Firenze, Via Ugo Schiff 6, 50019, Sesto F.no (Firenze), Italy
[b] Carapelli Firenze S.p.A., Via Leonardo da Vinci 31, 50028, Tavarnelle Val di Pesa (Firenze), Italy

Food Control 139 (2022) 109092

Contents lists available at ScienceDirect

Food Control

journal homepage: www.elsevier.com/locate/foodcont

ELSEVIER

Is the volatile compounds profile a suitable tool for authentication of virgin olive oils (*Olea europaea* L.) according to cultivars? A study by using HS-SPME-GC-MS and chemometrics

Lorenzo Cecchi[a], Marzia Migliorini[b], Elisa Giambanelli[b], Anna Cane[b], Bruno Zanoni[c], Valentina Canuti[c], Nadia Mulinacci[a,*], Fabrizio Melani[a]

[a] Department of NEUROFARBA, University of Florence, Via Ugo Schiff 6, 50019, Sesto F.no, Florence, Italy
[b] Carapelli Firenze S.p.A., Via Leonardo da Vinci 31, Tavarnelle Val di Pesa, 50028, Firenze, Italy
[c] Department of Agricultural, Food and Forestry Systems Management (DAGRI), University of Florence, Piazzale Delle Cascine 16, 50144, Florence, Italy

By studying the volatile profile of large groups of samples, it was possible:

- to develop chemometric approaches for the authentication of the geographic origin of virgin olive oils from the main worldwide producing countries
- to develop chemometric approaches for the authentication of the botanic origin of monovarietal virgin olive oils of some of the main cultivars
- to define clusters of cultivars based on patterns of similarity in the volatile profile.

The works carried out up to now within the collaboration was focused on the qualità control of virgin olive oil samples, with particular focus on the commercial classification and on varietal and geographic authentication

Let's take a look at this study

The last study on the varietal authentication also was also the starting point to study the sensory profile of monovarietal high-quality virgin olive oil.

Variety Authentication - Introduction

Why it is important?

EVOO is the highest-value commercial category

The sensory characteristics of EVOO samples can be very different depending on cultivar

An increasing number of consumers is more and more willing to pay a premium price for

- EVOOs with specific healthy properties
- EVOOs labeled for specific attributes that indicate a clear identity (organic EVOO, monocultivar EVOO or EVOO with indications of specific geographic origin)

Increased interest of producers towards monocultivar EVOOs in the last years

Increased risk of mislabeling

Setting up chemometric methods able to authenticate monocultivar EVOOs is strongly required

Variety Authentication - Introduction

Clusters of cultivars?

Monocultivar oils from some cultivar present sensory characteristics very similar to each other

The volatile profile of VOOs may allow clustering cultivars in specific groups

Variety Authentication - Introduction

What possible markers?

legal analytical parameters: free acidity, peroxide n°, UV indices

Not suitable

Other possible markers

- Tocopherols
- Sterols
- Phenolic compounds
- Volatile compounds
- Hydrocarbons
- Pigments
- Fatty acids
- DNA analysis

Among them, **phenolic compounds, volatile compounds** and some hydrocarbons such as **sesquiterpene hydrocarbons,** have been proposed as those likely most related to the varietal sensory typicity

Lukic et al., 2019; Campestre et al., 2017; Torres-Cobos et al., 2021

Aim of the study

Volatile profile analysis of monovarietal extra virgin olive oils and stuitable statistical tools were combined to

- discriminate monocultivar samples according to some of the main widespread cultivars
- clustering cultivars based on patterns of similarity

The samples

320 monovarietal samples classified as Extra virgin olive oils after chemical and sensory analysis, from different cultivars and geographic origins were selected

Cultivar	Geographic origin								Harvest year				
	Croatia	Italy	Greece	Morocco	Portugal	Spain	Tunisia	Unkown	2016-17	2017-18	2018-19	2019-20	Total
Picual					**5**	**69**			**16**	**3**	**53**	**2**	**74**
Arbequina				**1**	**19**	**22**		**4**	**3**	**11**	**30**	**2**	**46**
Coratina		**42**								**25**	**7**	**10**	**42**
Hojiblanca						**18**		**2**		**6**	**12**	**2**	**20**
Nocellara		**19**						**1**		**4**	**7**	**9**	**20**
Frantoio	**1**	**13**								**3**	**7**	**4**	**14**
Moraiolo		**12**								**2**	**7**	**3**	**12**
Koroneiki			**10**							**8**	**2**		**10**
Leccino		**10**								**1**	**7**	**2**	**10**
Peranzana		7								4		3	7
Arbosana					5						5		5
Cobrancosa					2			2		2	2		4
Leccio del Corno		4								1	3		4
Ogliarola		4										4	4
Ravece		4									2	2	4
Tonda iblea		4										4	4
Maurino		3								2	1		3
Morisca						3					3		3
Pendolino		3									3		3
Ascolana		2										2	2
Athinolia			2								2		2
Biancolilla		2								1	1		2

The chemical analysis

The volatile fraction of all samples was analyzed with the validated HS-SPME-GC-MS method

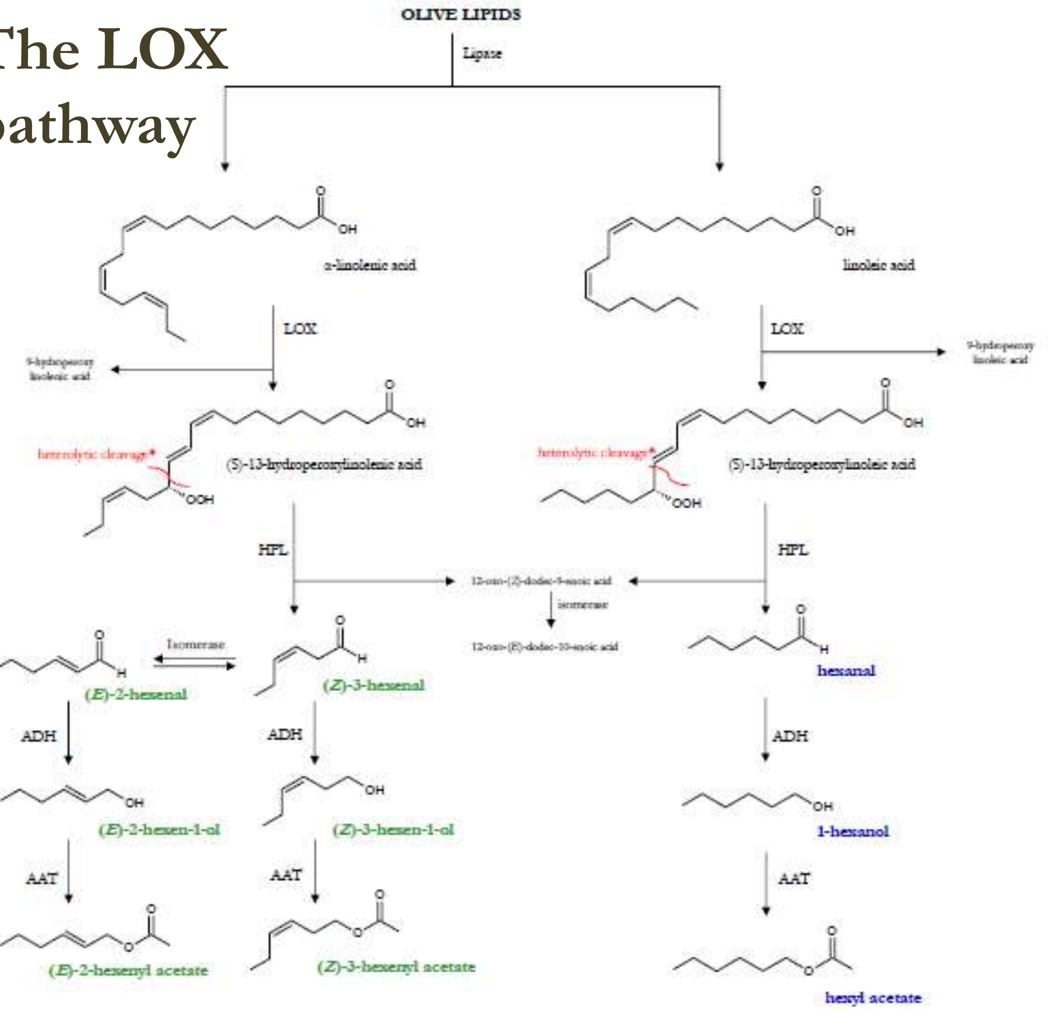
heptane
octane
methyl acetate
ethyl acetate
methanol
2-butanone
methyl propanoate
2-methyl butanal
isovaleraldehyde
ethanol
ethyl propanoate
3-pentanone
valeraldehyde
2-butanol
ethyl vinyl ketone
propanol
ethyl butanoate
buthyl acetate

phenol
4-ethylguaiacol
4-ethyl-phenol

hexanal
isobutanol
2-pentanol
E-2-pentenal
1-penten-3-ol
2-heptanone
heptanal
limonene
2-methyl-1-butanol + 3-methyl-1-butanol
Z-3-hexenal
E-2-hexenal
pentanol
hexyl acetate
2-octanone
octanal
1-octen-3-one
E-2-pentenol
Z-2-pentenol
2-heptanol
Z-3-hexenyl-acetate
E-2-hexenyl-acetate
E-2-heptenal
6-methyl-5-hepten-2-one
hexanol
E-3-hexenol

Z-3-hexenol
2-nonanone
nonanal
E-2-hexenol
Z-2-hexenol
2,4-hexadienal
2-octanol
E-2-octenal
1-octen-3-ol
heptanol
acetic acid
decanal
2,4-heptadienal
benzaldehyde
E-2-nonenal
octanol
propanoic acid
butanoic acid
E-2-decenal
nonanol
2,4-nonadienal
pentanoic acid
2,4-decadienal
hexanoic acid
guaiacol
phenyl ethanol

The LOX pathway

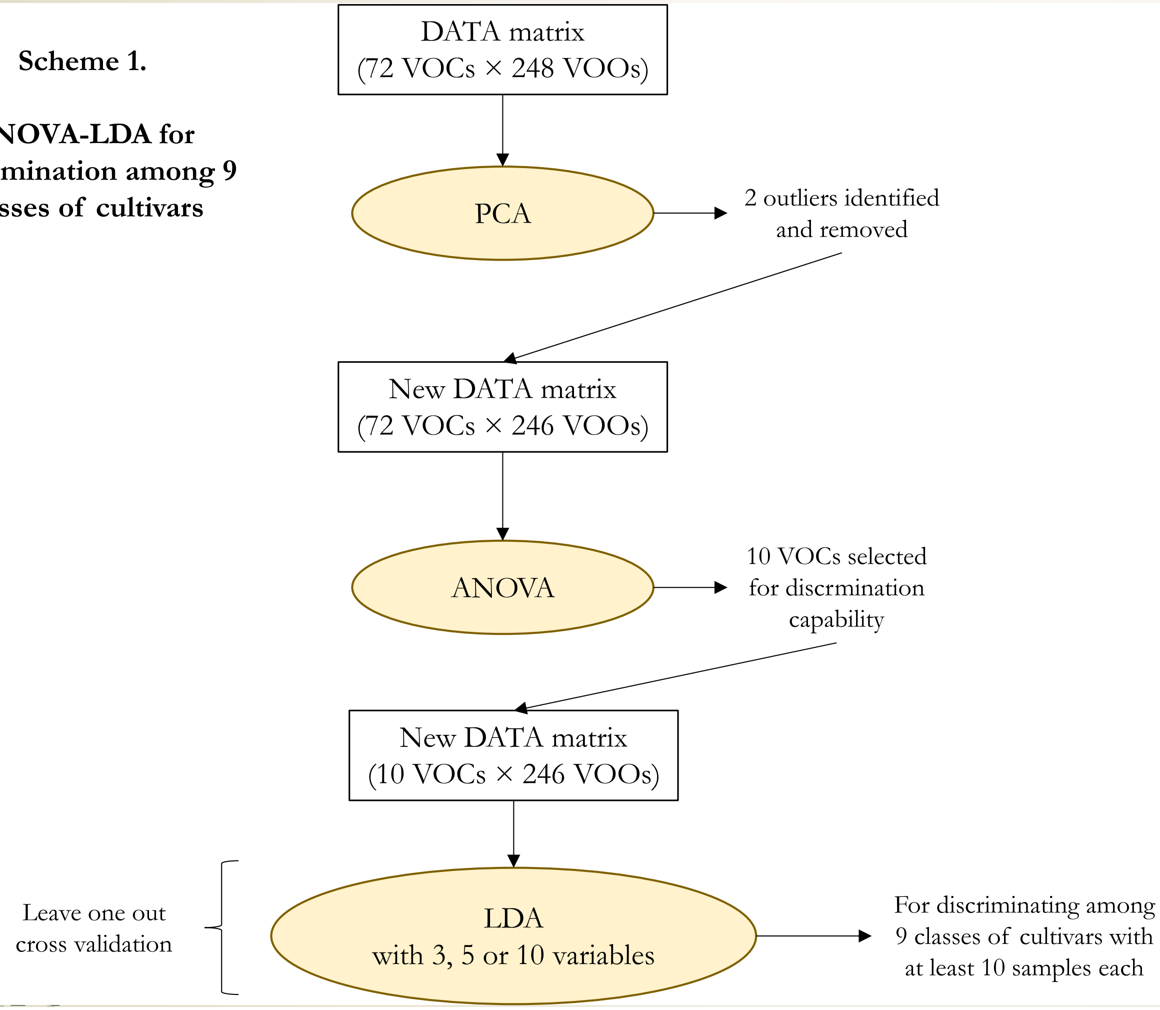


Cultivar authentication: The statistical approaches

Several statistical approaches were defined in search of the best one

Scheme 1.

ANOVA-LDA for discrimination among 9 classes of cultivars



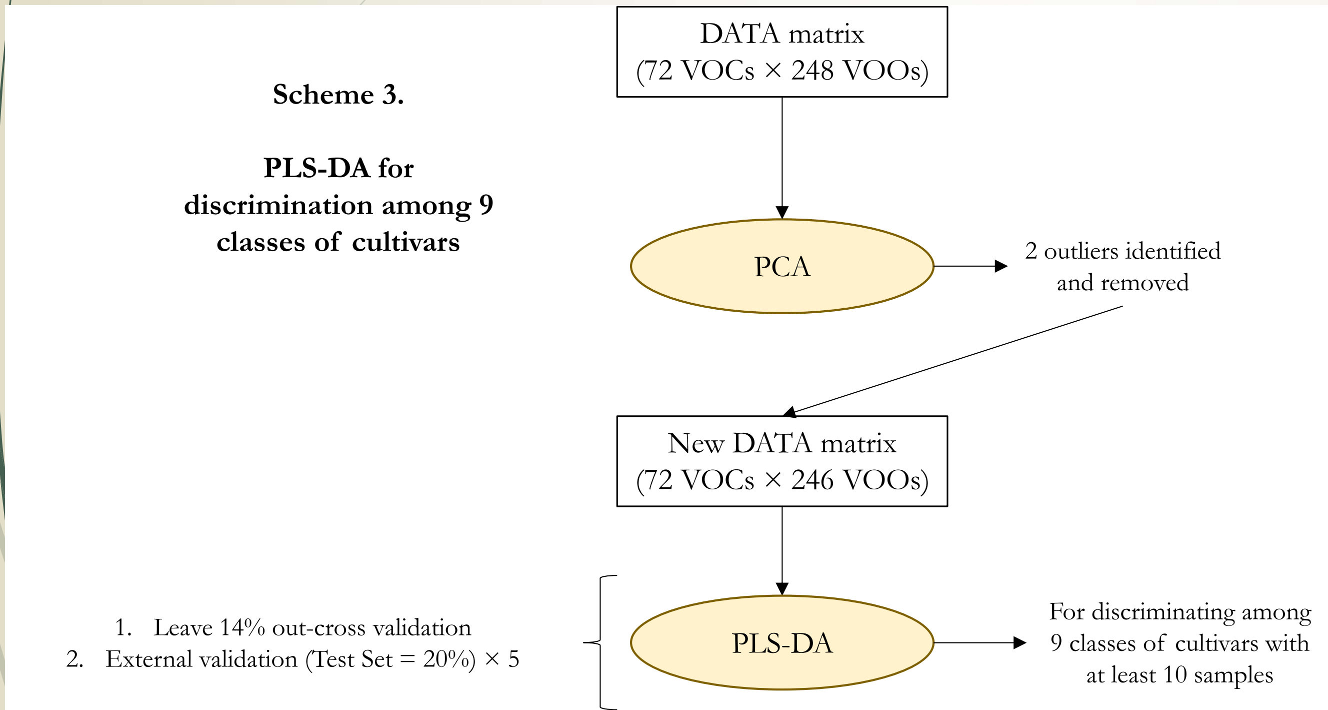
1. ANOVA-LDA for 9 classes of cvs
2. PLS-DA for 9 classes of cvs

The cultivars with at least 10 samples

- Picual
- Arbequina
- Coratina
- Hojublanca
- Nocellara
- Frantoio
- Moraiolo
- Koroneiki
- Leccino

Cultivar authentication: The statistical approaches

Several statistical approaches were defined in search of the best one



Scheme 3.

PLS-DA for discrimination among 9 classes of cultivars

1. ANOVA-LDA for 9 classes of cvs
2. PLS-DA for 9 classes of cvs

The cultivars with at least 10 samples

- Picual
- Arbequina
- Coratina
- Hojublanca
- Nocellara
- Frantoio
- Moraiolo
- Koroneiki
- Leccino

Results

1. Cultivar characterization

VOC (µg/kg)	Arbequina	Nocellara	Hojiblanca	Picual	Koroneiki	Coratina	Frantoio	Leccino	Moraiolo
Hexyl acetate	477.8	107.9	392.0	247.9	992.5	39.3	90.2	37.8	83.6
(*E*)-2-hexenal	26452.7	24033.3	5389.6	4303.2	12355.0	40395.4	39945.8	38309.1	33097.5
Hexanal	1125.5	2284.7	673.1	239.8	691.4	1118.4	685.9	634.2	754.0
(*Z*)-3-hexenyl acetate	2649.4	589.0	4209.8	3244.7	4016.5	249.4	319.6	243.0	1001.7
(*E*)-2-hexen-1-ol	1353.5	369.9	192.8	233.6	955.2	1775.1	414.2	477.4	261.9
1-Penten-3-one	196.0	924.8	354.4	183.4	258.6	468.8	681.6	475.2	543.9
1-Penten-3-ol	374.6	548.7	408.9	223.9	488.2	626.6	550.7	394.6	463.2
6-methyl-5-hepten-2-one †	0.8	2.5	2.6	3.2	24.4	1.1	2.2	1.1	0.6
(*Z*)-3-hexenal	714.7	2279.8	818.8	186.0	253.7	701.6	584.5	479.4	954.6
Phenyl ethanol †	485.3	101.5	159.3	160.7	719.6	364.1	141.8	145.5	120.2
Other LOX VOCs									
(*E*)-2-pentenal	48.0	115.9	76.7	26.6	64.5	68.3	79.7	84.7	70.0
(*Z*)-2-penten-1-ol	17.3	312.5	20.8	15.5	18.3	128.7	59.9	64.7	77.1
(*E*)-2-penten-1-ol	55.5	141.7	103.5	31.0	70.8	90.0	142.6	75.4	125.9
1-Hexanol	1305.8	742.8	755.9	453.6	604.6	1412.0	408.3	419.7	410.1
(*Z*)-3-hexen-1-ol	2046.7	2219.7	2674.8	1557.9	1484.4	894.1	554.4	573.8	1134.1
(*E*)-2-hexenyl acetate	35.7	2.7	10.1	8.5	13.0	7.7	11.7	3.3	7.3
Total LOX VOCs	36803.8	34673.3	16081.2	10955.5	22266.8	47846.7	44529.1	42272.4	38984.9

8 out of the 10 selected VOCs were originated from the LOX pathway

The total content of LOX VOCs was very different considering different groups of cvs

(*E*)-2-hexenal was the most abundant VOC in all cultivars

(*E*)-2-hexenal represented
- more than 84% of LOX-VOCs in several Italian cvs, except Nocellara
- Less than 40% of LOX-VOCs in Picual and Hojiblanca
- Approx 70% of the LOX-VOCs in Arbequina and Nocellara
- Approx 50% of LOX-VOCs in Koroneiki

The cultivars with the higher (*E*)-2-hexenal content were also those with the lowest (*Z*)-3-hexenyl acetate content and vice versa

Cultivar authentication: Results

The nine cvs
Arbequina, Coratina, Frantoio, Hojiblanca, Koroneiki, Leccino, Moraiolo, Nocellara, Picual.

1. The ANOVA-LDA model

Results of the LDA models built using the data from 10 VOCs

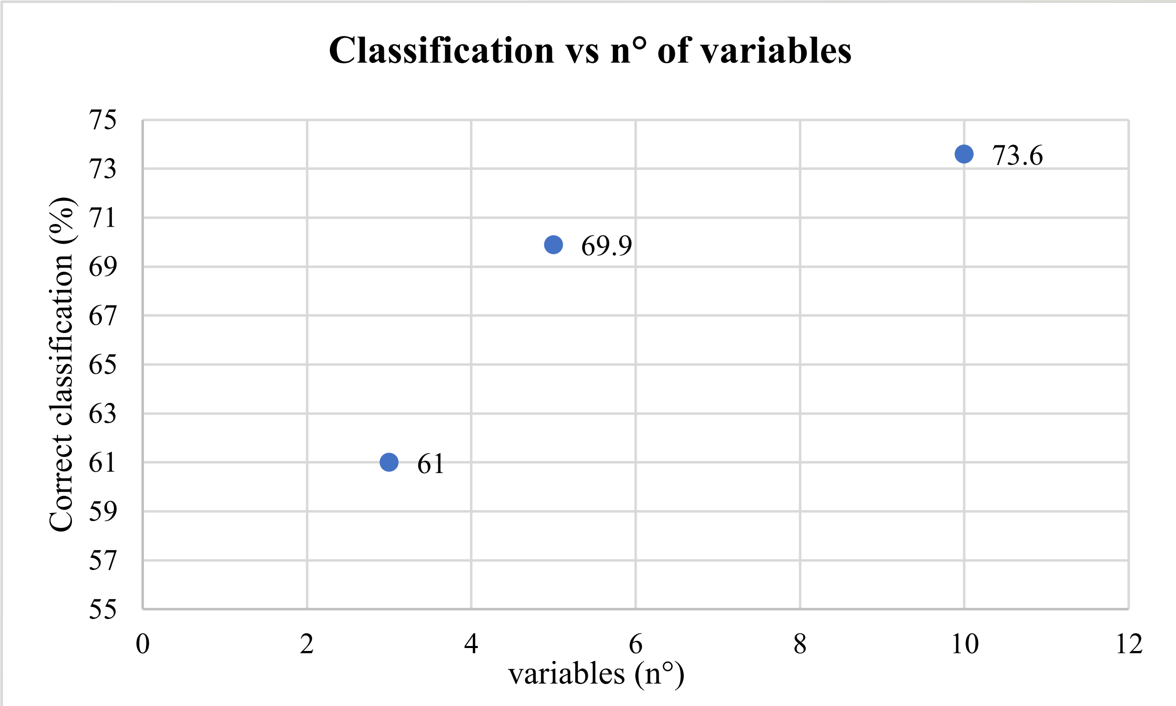
Cultivar	Samples	Arbequina	Coratina	Frantoio	Hojiblanca	Nocellara	Picual	Koroneiki	Leccino	Moraiolo
Arbequina	45	**39 (86.7%)**	1 (2.2%)	-	-	-	3 (6.7%)	-	-	2 (4.4%)
Coratina	41	1 (2.4%)	**23 (56.2%)**	11 (26.8%)	-	1 (2.4%)	-	-	1 (2.4%)	4 (9.8%)
Frantoio	14	-	-	**8 (57.1%)**	-	-	-	-	4 (28.6%)	2 (14.3%)
Hojiblanca	20	-	-	-	**14 (70.0%)**	3 (15.0%)	3 (15.0%)	-	-	-
Nocellara	20	-	1 (5.0%)	-	-	**17 (85.0%)**	-	-	-	2 (10.0%)
Picual	74	-	1 (1.4%)	-	12 (16.2%)	-	**61 (82.3%)**	-	-	-
Koroneiki	10	-	-	-	-	-	-	**10 (100%)**	-	-
Leccino	10	-	-	2 (20.0%)	-	-	-	-	**7 (70.0%)**	1 (10.0%)
Moraiolo	12	-	-	3 (25.0%)	-	1 (8.3%)	-	-	6 (50.0%)	**2 (16.7%)**
Total	**246**	**Total Correct Classification: 181 samples = 73.6%**								

Cultivar authentication: Results

The nine cvs
Arbequina, Coratina, Frantoio, Hojiblanca, Koroneiki, Leccino, Moraiolo, Nocellara, Picual.

1. The ANOVA-LDA model

Results of the LDA models built using the data from 10 VOCs



Cultivar authentication: Results

The nine cvs
Arbequina, Coratina, Frantoio, Hojiblanca, Koroneiki, Leccino, Moraiolo, Nocellara, Picual.

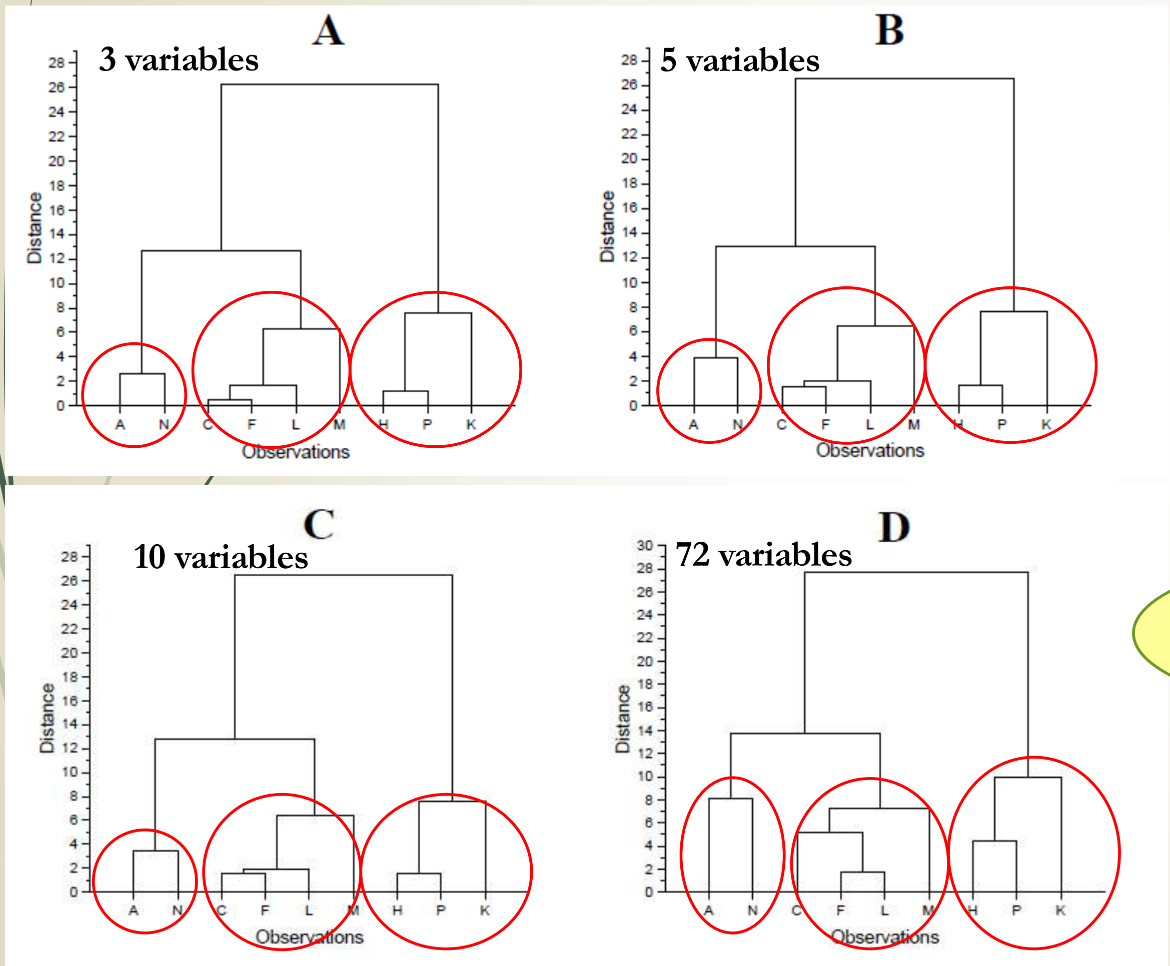
2. Cultivar authentication: the PLS-DA model

Results of the PLS-DA models built with 15 latent variables

Cultivar	Samples	Arbequina	Coratina	Frantoio	Hojiblanca	Nocellara	Picual	Koroneiki	Leccino	Moraiolo
Arbequina	45	**88.9%**	-	-	-	2.2%	6.7%	2.2%	-	-
Coratina	41	2.4%	**95.2%**	2.4%	-	-	-	-	-	-
Frantoio	14	-	21.4%	**42.9%**	-	-	-	-	28.6%	7.1%
Hojiblanca	20	-	-	-	**45.0%**	10.0%	45.0%	-	-	-
Nocellara	20	-	-	-	5.0%	**90%**	-	-	5.0%	-
Picual	74	-	1.4%	-	2.7%	-	**95.9%**	-	-	-
Koroneiki	10	-	-	-	-	-	10.0%	**90%**	-	-
Leccino	10	-	10.0%	20.0%	-	-	-	-	**50.0%**	20.0%
Moraiolo	12	-	8.3%	8.3%	-	16.7%	8.3%	-	25.0%	**33.3%**
Total	**246**	**Average Total Correct Classification: 81.7%**								

Cultivar Clustering: The statistical approaches

Agglomerative Hierarchical Clustering



AHC was run on the mean of the variables for each cultivar for clustering cultivars based on their patterns of similarity. It was run four time:

- With 3 variables
- With 5 variables
- With 10 variables
- With all the 72 variables

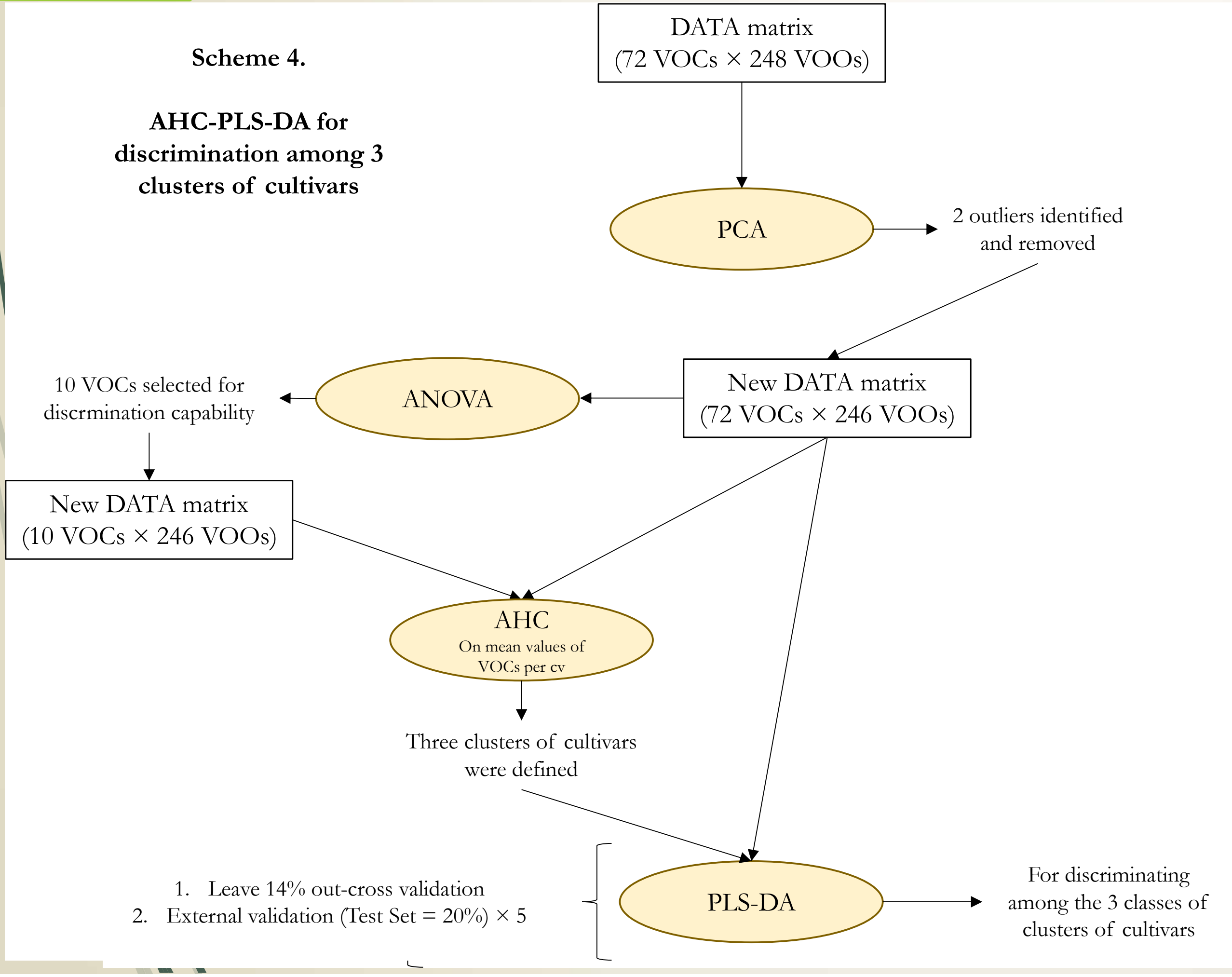
Arbequina, Nocellara

Coratina, Frantoio, Moraiolo, Leccino

Hojiblanca, Picual, Koroneiki

Cultivar Clustering: The statistical approaches

Several statistical approaches were defined in search of the best one



Scheme 4.

AHC-PLS-DA for discrimination among 3 clusters of cultivars

1. ANOVA-AHC-LDA for 3 clusters of cvs
2. AHC-PLS-DA for 3 clusters of cvs

The cultivars with at least 10 samples

- Picual
- Arbequina
- Coratina
- Hojublanca
- Nocellara
- Frantoio
- Moraiolo
- Koroneiki
- Leccino

Cultivar clustering: Results

A	Average Samples	AN	CFLM	HPK
AN	13	**89.2%**	6.2%	4.6%
CFLM	15.4	5.2%	**93.5%**	1.3%
HPK	20.8	2.9%	1.0%	**96.1%**
Total	49.2	**Average Correct Classification: 93.5%**		

Results with the LDA model built using 10 VOCs

Results with the PLS-DA model

B	Average Samples	AN	CFLM	HPK
AN	13	**86.2%**	3.1%	10.7%
CFLM	15.4	5.2%	**94.8%**	0%
HPK	20.8	1.0%	1.0%	**98.0%**
Total	49.2	**Average Correct Classification: 93.9%**		

AND NOW?

NEXT GOAL: to define the sensory profile of monocultivar EVOOs

- We are investigating on EVOOs of specific cultivars through the characterization of both the sensory and volatile profile. A group of volatile hydrocarbons that seem able to complete the characterization of the cultivars is also included in the study
- We are also investigating if EVOO samples of the defined clusters resemble each other or not even from a sensory point of view
- Further cultivars are being included in the study. They will be included in one of the three defined cultivars or new clusters, if any, will be defined

THANK YOU FOR YOUR ATTENTION

Dr. Lorenzo Cecchi
Lo.cecchi@unifi.it