

CHARACTERIZATION OF AROMATIC COMPOUNDS AND α AND β ACIDS FROM HUMULUS LUPULUS L.

INTRODUCTION

The *Humulus lupulus* L. is a species in the Cannabaceae family. Hops, as it is commonly known, is a perennial, dioecious and normally diploid ($2n = 20$) herbaceous plant [1]. It is in brewing that hops have their greatest economic value at an international level. It was in Flanders that it started brewing, since from the beginning of its manufacture in Mesopotamia, until then, the beer was a sweet drink, based on the fermentation of malt, with the name of Ale being its conservation one of the problems. The use of hops has solved this problem, since it produces compounds with bactericidal action, in particular against Gram-negative bacteria [2]. Its production spread throughout Europe and, from there, to the rest of the world.



Currently the demand for new aromas has been increasing, driven by the recent expansion of artisanal beer production in Portugal. Since there is spontaneous hops in much of the country, the collection and analysis of the hops' aromas may lead to the development of new and more fragrant aromas. Volatile components extracted from spontaneous hops collected in the Bragança area were analyzed and compared with those of commercial varieties

METODOLOGY

The vegetal material was harvested in zones of the district of Bragança, different from those of old or current production, next to water courses. The female cones were collected and the volatiles were extracted in a Likens-Nickerson system and then analyzed by CG and CG-MS. From different plants (2 nugget and one spontaneous from France, Bragança) the alpha and beta acids were also extracted and analyzed by HPLC for quantification.

RESULTS

The results obtained show a certain similarity in the monoterpene component, with β -myrcene as the major compound (75 and 64%, respectively in the cultivar sample and in the spontaneous sample) and notorious differences in the sesquiterpene component, clearly evident in cases of α -humulene (12% in the cultivar, 0.2% in the non-spontaneous) and trans- β -farnesene (not detected in the cultivar, 9% in the non-spontaneous). Also worthy of note is the greater richness of the sesquiterpene component of the spontaneous clone, in particular in the oxygenated compounds.

Table 1. Composition of the volatile fraction from samples of *Humulus lupulus*

Compostos	RI	Nugget flower	Sample 3	Cascade	Chinook
Isobutyl isobutyrate	909	0.1			
Methyl hexanoate	914		t		
α -Pinene	930	0.2	0.2		
Camphene	938	t			
β -Pinene	963	1.0	0.7	0.8	0.8
β -Myrcene	975	74.8	64.4	67.5	61.3
α -Phellandrene	995	t			
Isoamyl isobutyrate	999	0.2			
β -Phellandrene	1005	0.3	0.2	0.3	
Limonene	1009	0.3	0.2	0.2	0.2
cis- β -Ocimene	1017	0.1			
trans- β -Ocimene	1027	0.3		0.1	
γ -Terpinene	1035	t			
2-Nonanone	1058		0.7		
Nonanal	1073	t	0.2		
Linolol	1074	0.3	0.7	0.5	0.3
Methyl octanoate	1105	0.2	0.2		0.1
Hexyl isobutyrate	1127	t			
α -Terpineol	1159	t			
2-Decanone	1166		0.5		
Methyl nonanoate	1205	0.1	0.1		
Heptyl isobutyrate	1233	t			
2-Undecanone	1275	0.2	2.2	1.3	
Methyl decanoate	1314	0.1	t		
α -Ylangene	1371	0.1			0.1
α -Copaene	1375	0.1	t		0.3
2-Dodecanone	1389	t	0.3		
β -Caryophyllene	1414	3.5	1.5	3.0	6.2
β -Copaene	1426	0.1		0.1	0.5
Aromadendrene	1428		0.4		
α -Humulene	1447	12.1	0.2	4.9	12.5
trans- β -Farnesene	1455		8.5	6.4	
trans-Cadina-1(6),4-diene	1469	0.1			
γ -Muurolene	1469	0.2	0.6	0.4	1.1
β -Selinene	1476		1.7		0.6
Valencene	1484		2.2		
Viridiflorene	1487	t			
γ -Cadinene	1500	0.3	0.2	0.4	1.1
7-epi- α -Selinene	1500		0.2		
δ -Cadinene	1505	0.5		0.3	1.6
α -Cadinene	1525	t			
α -Cadinene	1529	t			
Germacrene B	1533		1.1		0.7
β -Caryophyllene oxide	1561		0.2		0.1
Viridiflorol	1569		0.2		
γ -Eudesmol	1609		0.2		
T-Cadinol	1616		0.2		0.3
β -Eudesmol	1620		0.6		
Intemediol	1626		0.3		
α -Eudesmol	1634		0.7		
% Identification		95.2	89.6	93.9	81.6
Terpenoid compounds					
Monoterpene hydrocarbons		77.0	65.7	75.6	38.8
Oxygen-containing Monoterpenes		0.3	0.7	0.3	0.2
Sesquiterpene hydrocarbons		17.0	16.6	17.7	41.6
Oxygen-containing Sesquiterpenes		t	2.4	0.0	0.9
Others		0.9	4.2	0.3	0.2

RI - Retention index relative to n-alkanes C₇-C₁₅; t - trace amount (<0.05%)

	α acids		Total	β acids		Total
	Co-Humulone	n-Humulone		Co-Isopulone	n-Hulapulone	
Wild clone	3.00%	4.30%	7.30%	4.10%	4.10%	8.00%
Nugget	2.40%	3.0%	5.20%	3.4%	3.2%	3.1%
Nugget	3.10%	11.2%	14.3%	3.8%	2.1%	3.9%
Cascade	1.45%	3.88%	4.53%	2.93%	2.87%	5.63%
Chinook	3.9%	10.44%	14.40%	2.57%	2.02%	4.58%

Table 2

The cultivar showed results very high for α acid in fact, it is the big amount of all the samples analyzed, besides the Chinook, but had the smallest contribution in the case of the β acids. Nonetheless the results of Cascade had a greater proximity to the values obtained with the wild clone, with smallest amount of α acids but also less β acids, and with values that are not very far from those for the wild clone.

BIBLIOGRAPHY

- [1] Heale J. B., Legg T., Brar J., Fabb A., Bainbridge B.(1989) Application of plant tissue culture and molecular biology techniques to "progressive" wilt of hops caused by *Verticillium albo-atrum*. Eur. Brew. Conv. Monogr., XV, Symposium on Plant Biotechnology, Helsinki, 70:83
- [2] Duke, J. A. (1983). *Humulus lupulus* L. Handbook of energy crops.