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PAPER

VOLATILE FINGERPRINT AND PHYSICO-MECHANICAL PROPERTIES OF 'MUSCAT BLANC' GRAPES GROWN IN MOUNTAIN AREA: A FIRST EVIDENCE OF THE INFLUENCE OF WATER REGIMES

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ABSTRACT

The volatile composition of aromatic grape varieties at harvest is a very important criterion in the choice of vinification technique to yield the optimal quality of the final product. The berry mechanical characteristics are important for assessing resistance to fungal attacks and for the estimation of shattering. In this study the effect of irrigation on the volatile fingerprint and the mechanical properties of the *Muscat blanc* (*Vitis vinifera* L.) grapes grown in mountain north-west region of Italy was investigated. Three water regimes were compared: standard irrigation, moderate irrigation and drought.

In the meteorological conditions of the considered season, a significant increase in the amounts of the most representative free volatile components of the *Muscat blanc* variety (linalool and geraniol markers), was observed in standard irrigation treatment. Significantly higher amounts of four C13-norisoprenoid bound compounds were observed in the drought treatment with respect to the standard treatment. Furthermore, no influence of irrigation treatment on berry skin hardness and thickness parameters was noted.

Therefore, in the considered alpine environment, on aromatic *Muscat blanc* variety, the optimum irrigation treatment is an important choose to improve the quality of the grapes.

- Keywords: *Muscat blanc*, volatile components, glycosides, skin hardness, water stress -

INTRODUCTION

Vitis vinifera L. *Muscat blanc* grape variety (synonym of white *Frontignan* and *Muscat* à petit grains blanc), a widespread aromatic grape variety, has been grown since the Middle Ages in the Aosta Valley, a mountain region located in the North-West of Italy. Its cultivation area is mostly concentrated in the South-facing, well exposed mountain slopes around the village of Chambave, at an altitude ranging from 500 to 700 m above sea level. Here it is used for producing the renowned dry wine sold under the Appellation of Origin Vallée d'Aoste - *Chambave Muscat*. The climatic conditions of the area are favourable for wine grape growing: low levels of average annual rainfall (less than 600 mm, even less than 400 mm during particularly dry years, with two seasonal peaks in spring and autumn), low relative humidity, low cloudiness and very high levels of solar radiation. The estimated annual mean temperature is 10°-11°C (MERCALLI, 2003).

Plant water status can greatly affect, either positively or negatively, vegetative growth, berry size, production yield, grape phenolic content and profile, grape aromatic potential, must composition, and wine sensory characteristics depending on its intensity level and timing (HEPNER *et al.*, 1985; MATTHEWS *et al.*, 1990; OJEDA *et al.*, 2001; DELOIRE *et al.*, 2004; CHAPMAN *et al.*, 2005; KOUNDOURAS *et al.*, 2006; QIAN *et al.*, 2009; VAN LEEUWEN *et al.*, 2009; GAMBACORTA *et al.*, 2011). Therefore, in environments where low rainfall is a limiting factor, irrigation practices may be a powerful tool for improving grape quality by modifying berry characteristics and composition; this can often be achieved by inducing moderate levels of water stress in specific stages of berry development and ripening. On the other hand, an excess of irrigation may easily result in poor grape quality (GLADSTONE, 1992). Thus, the assessment of the correct amount of irrigation and its scheduling is essential in order to reach a desired production target.

Volatile aroma compounds of the *Muscat blanc* have been less studied (USSEGLIO-TOMMASET and DI STEFANO, 1980; BUREAU *et al.*, 2000) than other aromatic *Muscat* varieties (RIBEREAU-GAYON *et al.*, 1975; GUNATA *et al.*, 1985; WIRTH *et al.*, 2001) and often many studies treating the aroma profile of the *Muscat* variety were related to aroma wines produced (SANCHEZ PALOMO *et al.*, 2007; del CARO *et al.*, 2012). The terpene content in the grape may be influenced by temperature, light and water availability during ripening (RIBEREAU-GAYON *et al.*, 2000b), thus a knowledge of free and bound precursors of this aromatic variety may help in the selection of best vineyard practices. The effect of the bioclimatic and agronomical conditions, in particular the water regimes, on the aromatic composition of white aromatic berries has not yet been deeply investigated, although the aromatic profile of the 'Muscat' grapes should be considered at least as important as the usual technological ripeness parameters (i.e. sugars content and acidic composition).

Texture Analysis is a analytical technique of survey used for measurement of the physical properties of wine grapes in order to assess their quality (ROLLE *et al.*, 2012). In fact, instrumental texture parameters can be used as phenols extractable markers and ripeness predictors (MAURY *et al.*, 2011; RÍO SEGADE *et al.*, 2011a). Knowing the characteristics of some mechanical parameters such as the ease of detachment of the pedicel, the hardness and thickness of the skin, is important because it is directly related to the phenomena of shattering, resistance to splitting and plant diseases (LANG and DURING, 1990; GABLER *et al.*, 2003), in particular during on-vine withering process (ROLLE *et al.*, 2010). At our knowledge, the effect of irrigation on the physico-mechanical characteristics of the berries has not yet been reported.

During the last ten years the irrigation, normally carried out by drip irrigation systems, has become increasingly popular among the grape growers of the Aosta Valley area. While it is considered a special measure to be adopted only when exceptionally dry conditions occur, it is now often become standard viticultural practice in Aosta Valley.

On the base of all these consideration, the aim of this work was to study the free and bound varietal volatiles and mechanical properties of berries of the *Vitis vinifera* L. grape aromatic variety *Muscat blanc* comparing the effect of three different water regimes on these physico-chemical parameters.

MATERIALS AND METHODS

Plant materials

The experimental site was located on a steep slope with a south aspect, at an altitude of 600 m above sea level, on a loamy sand soil. The vines (*Vitis vinifera* L., *cv Muscat blanc*, clone R6, grafted on 110 Richter rootstock) were 12 years old. The trellis system was Guyot. The vine and row spacing was 0.70 and 1.80 m respectively. The vineyard was equipped with a drip irrigation system using pressure-compensated emitters. The distance between emitters was 60 cm; their flow rate was 2.3 L h^{-1}.

Grapes were collected from the 2008 vintage season. A Randomized Complete Block Design with three replicates was adopted. Plots had three rows each of about 15 m long; only the central row was used for experimental purposes; the other two received the same water treatment and were used as border rows. On the cen-

tral row eight plants were selected and used for sampling berries.

Water treatments

Three water regimes were compared: standard irrigation (S), moderate irrigation (M), drought (D). Treatment D did not receive any irrigation, while S and M irrigation strategies were based primarily on pre-dawn water potentials (Ψ_{PD}). Treatment S was kept at Ψ_{PD} levels above -0.2 MPa (absence of water stress) until two weeks before harvest. Treatment M was kept above -0.2 MPa until veraison; after veraison a moderate water stress (-0.2 to -0.4 MPa) was allowed. No irrigation treatment was carried out during the last two weeks before harvest. Before veraison, even in the absence of water stress, treatments S and M received 50 and 25% of the estimated crop evapotranspiration (ET_c) respectively, at approximately 10 day intervals. ET_c was estimated from meteorological data logged by a weather station (Vantage Pro, Davis Instruments Corp., Hayward, California 94545 USA) situated in the experimental site, by multiplying the reference evapotranspiration (ET_0, directly calculated by the station) by a crop coefficient K_c (varying from 0.6 to 0.7 from the start of treatments to veraison).

Plant water status measurements

Plant water status was monitored by measuring the pre-dawn water potential Ψ_{PD} with a SKYE pressure chamber, model SKPM 1400 (SKYE Instruments, Llandrindod Wells, Powys LD1 6DF UK). Ten measurements were made from fruit set to harvest. For each plot, the Ψ_{PD} was estimated by measuring six fully expanded leaves taken from the primary shoots. Measurements took about two hours and terminated before dawn.

Cluster thinning

The crop load of the eight selected plants per plot was balanced by cluster thinning at veraison, aiming at a yield of 1.6-1.8 kg/vine, which is generally considered optimal for the usual local requirements, given the plant density of the experimental vineyard.

Technological parameters analysis

The total soluble solids concentration (°Brix, as SSC) was measured using an Atago 0-32 °Brix temperature compensating refractometer (Atago Co., Tokyo, Japan), pH was determined by potentiometry using a Crison electrode (Carpi, Italy) on the grape must. Titratable acidity (TA), expressed as gL^{-1} tartaric acid, was estimated using the official OIV method. The must contents of citric, tartaric and malic acid were analyzed using an HPLC system (P100-AS3000, Thermo Electron Corporation, Waltham, MA, USA) equipped with a UV detector (UV3000) set to 210 nm. The analyses were performed isocratically at 0.8 $mLmin^{-1}$ flow and 65°C column temperature, with a 300x7.8 mm i.d. Aminex HPX-87H cation exchange column and a Cation H^+ Microguard cartridge (Bio-Rad Laboratories, Hercules, CA, USA), using 0.0013 mol/L H_2SO_4 as mobile phase (GIORDANO *et al.*, 2009). Data treatment was carried out using the ChromQuest™ chromatography data system (ThermoQuest, Inc, San Jose, CA, USA).

Volatile analysis

Sample preparation

Analysis of free and glycosidically bound components was carried out according to the previous method proposed by DI STEFANO (1991), with some modifications. For each plot, two subsamples of 100 berries were taken and analysed separately. They were de-seeded and the pulp (about 200 g) was separated from the skin with the addition of $Na_2S_2O_5$ (80 mg). Skins were placed in 20 mL of methanol for 1 h to inactivate glycosidase enzymes. The pulps and skins were then separately crushed under a nitrogen atmosphere with a laboratory blender (Waring Laboratory, Torrington, USA). The skin suspension and pulp homogenate were then combined. The mixture was centrifuged twice (7,000 g; 10 min; 4°C), washing the solid pellet with tartaric acid buffer (pH=3.2) and the liquid extract was then clarified with pectolytic enzyme (100 mg) without secondary glycosidase activity (Rapidase X-Press, DSM, The Netherlands) at room temperature for 2 hours. Aliquots of 500 mL were collected from this juice. Afterwards, each replicate of the grape juice (100 mL) (n=2), added of 2-octanol as internal standard (200 µL of 41 mg L^{-1} solution in 10% ethanol), was loaded onto a 1g tC18 reversed-phase SPE cartridge (Sep-Pak, Waters, Ireland), previously activated with 10 mL of methanol and 20 mL of water, with a flow rate of ca. 3 mL min^{-1}. The cartridge was then rinsed with 30 mL of pure water to eliminate sugars, acids and other low molecular weight polar compounds. The free fraction was then eluted with 30 mL of dichloromethane. The eluate was dried over Na_2SO_4 and concentrated to 200 µL under a stream of nitrogen. The glycoconjugates were finally eluted from the cartridge with 50 mL of methanol and concentrated to dryness using a vacuum rotavapor (Buchi R-210, Switzerland) at 35°C. The dried glycosidic extract was dissolved in 3 mL of citrate-phosphate buffer (0.2 M, pH=5). The enzymatic hydrolysis was carried out using 50 mg of an AR-2000 commercial preparation with glycosidase side activities (DSM Oenology, The Netherlands) and incubating at 40°C for 24 h. After

addition of 200 µL of 2-octanol (41 mg L^{-1} solution in 10% ethanol), glycosidic precursors were then extracted following the SPE method previously described. The dichloromethane extract obtained was dried over anhydrous Na_2SO_4, concentrated to 200 µL under nitrogen and kept at -20°C until analysed. All analyses were performed in duplicate.

GC/MS analysis

Analysis was performed with a Shimadzu GC-2010 gas chromatograph equipped with a Shimadzu QP-2010 Plus quadrupole mass spectrometer. The gas chromatograph was equipped with a DB-WAX capillary column (30 m x 0.25 mm i.d., 0.25 µm film thickness, J&W Scientific Inc., Folsom, CA, USA). The temperature program started at 35°C for 5 min; 2°C min^{-1} to 190°C; 3°C min^{-1} to 230°C for 5 min. Carrier gas (He) was at 1mL min^{-1}. Injections of 1 µL were performed in split mode 1:20. The injection port temperature was 230°C, the ion source temperature was 240°C and the interface temperature was 230°C (solvent delay of 4.5 min). The detection was carried out by electron impact mass spectrometry (70 eV) in total ion current (TIC) mode in a mass range *m/z* 30-350. Identification of compounds was carried out by comparing their mass spectra and retention indices (RI) (a mixture of a homologous series of C5-C28 was used) with those of standard compounds or by comparing their mass spectra and Kovats with those reported in literature or by comparing mass spectrum with mass spectral databases NIST12, NIST62 (National Institute of Standards and Technology, Gaithersburg, MD, USA), Adams and on line (http://webbook.nist.gov/chemistry/). Semiquantitative data ($\mu g L^{-1}$) were obtained by measuring the relative peak area of each identified compound in relation to that of the added internal standard.

Mechanical properties

A Universal Testing Machine (UTM) TAxT2i Texture Analyzer (Stable Micro Systems - SMS, Surrey, UK) equipped with a HDP/90 platform (perforated or not) and a 5 kg load cell was used. All the acquisitions were made at 400 Hz; data were evaluated using the Texture Expert Exceed software package (vers. 2.54 in Windows 2000). The operating conditions applied, probe and mechanical parameters measured, are summarized in Table 1, according to methods proposed by LETAIEF *et al.* (2008). Skin hardness was evaluated using the puncture test. The berries (n=30, MAURY *et al.*, 2009) were placed on the metal plate of the UTM with the pedicel in a horizontal plane in order to be consistently punctured in the lateral face. For the measurement of berry skin thickness, a piece of skin of almost 0.25 cm^2 was removed from the lateral side of all the 30 berries of each sample with a razor blade. Especial care was taken when removing the pulp from the skin and when positioning the skin sample on the UTM platform to prevent folds in the skin. After calibration of the instrumental probe position, the skin thickness was calculated as the distance between the point corresponding to the probe contact with the berry skin (trigger) and the platform base during a compression test. It was convenient to insert an instrumental trigger threshold equal to 0.05 N that enabled the plane surface of the probe to adhere completely to the skin sample before the acquisition started. It allowed a reduction or elimination of the 'tail' effect due to the postponement of the point. Peduncle detachment resistance was determined by a traction test (ROLLE *et al.*, 2009). In this test the peduncle was anchored to the pliers of the A/PS probe. During the traction, the peduncle passes through the perforated platform of the UTM (diameter of the hole 5mm), while the berry is blocked, permitting the determination of the force of peduncle detachment.

Statistical analysis

Statistical analyses were performed using the statistical software package SPSS (version 17.0; SPSS Inc., Chicago, IL, USA).

Table 1 - Operating conditions used in the texture analysis of grapes.

Test	Probe - Platform	Test speed	Force	Mechanical properties
Berry skin hardness	SMS P/2N Needle; HDP/90 platform	1 mm s^{-1}	compression 3 mm	F_{sk} = Berry skin break Force (N) W_{sk} = Berry skin break Energy (mJ) E_{sk} = Skin Young's modulus (N/mm)
Berry skin thickness	SMS P/2 Ø 2mm; HDP/90 platform	0,2 mm s^{-1}	-	Sp_{sk} = Berry skin thickness (µm)
Peduncle detachment resistance	SMS A/PS modified with rigid arm; HDP/90 perforated (Ø 5mm) platform	1 mm s^{-1}	traction 10 mm	F_{ped} = Peduncle detachment Force (N) W_{ped} = Peduncle detachment Energy (mJ)

RESULTS AND DISCUSSION

Meteorological conditions

The season was characterised by an unusually high amount of rainfall during spring and early summer followed by low precipitation levels until harvest. Total precipitation between flowering to veraison was 160 mm; only 32 mm were registered from veraison to harvest. Before veraison the pre-dawn leaf water potentials ψ_{PD} of all three treatments remained at relatively high levels (within the 0 to -0.2 MPa range), showing an absence of water stress even in the non-irrigated plots. After veraison, the ψ_{PD} of moderately irrigated and non-irrigated vines began to slowly drop, showing an effective differentiation of the relative water status. Only the D treatment reached pre-dawn ψ_{PD} below -0.6 MPa, which can be considered severe water stress threshold, but only in the last days before complete ripening, when these levels are not unusual. No statistically significant differences in ψ_{PD} were observed among treatments before 54 DAF (days after flowering). From 54 DAF until harvest treatment S showed significantly higher ψ_{PD} values than treatment D. Treatment M showed intermediate ψ_{PD}, values, often comparable or closer to D for most of the veraison-harvest period.

The S treatment received a total of 110 mm of additional water by drip irrigation distributed in ten applications from day 8 AF to day 90 AF; M treatment received 34 mm of additional water in five applications from day 8 to 68 AF. Differences in yield were not statistically significant; yields/vine ratios ranged from 1.67 kg (treatment D) to 1.82 (treatment M). Harvest date was 108 DAF.

Technological parameters of ripeness

Sugar content, pH, acid composition, yield and berry weight of grapes from the three different water treatments exhibited no significant statistical differences. At harvest, on average, the grape musts were characterized by soluble solid contents of 23.4 ± 0.6 as °Brix, titratable acidity expressed as tartaric acid of 7.53 ± 0.52 gL^{-1} and by a pH of 3.36 ± 0.05. The contents of the main organic acids were 0.26±0.04, 4.58 ±0.16, 3.90 ±0.51 for citric, tartaric and malic acid, respectively.

Free volatile compounds

A total of 33 free volatile compounds were detected (mean values of concentrations ± standard deviations) in *Muscat blanc* grape juices. They were classified into three chemical categories: four C6 compounds (alcohols and aldehydes), 27 monoterpenoids and two aromatic alcohols (Table 2). All C6 compounds and 18 of 27 monoterpenoids were always observed in all treatments.

Among monoterpenoids, free linalool was the most abundant compound found in all water regimes. Other components, that contribute with more than 5% to the terpenoids content, were geraniol, 3,7-dimethyl-1,5-octadien-3,7-diol (diendiol I) and 3,7-dimethyl-1,7-octadien-3,6-diol (diendiol II). Linalool, that provides characteristic sweet and flowery notes, together with geraniol, α-terpineol and ho-trienol are considered the marker compounds for the terpene-like character of Muscat wines (RIBEREAU-GAYON *et al.*, 1975; MARAIS, 1983). The content of linalool in *Muscat blanc* grape was found at similar average levels by other authors (BUREAU *et al.*, 2000; SANCHEZ-PALOMO *et al.*, 2006; LAMBRI *et al.*, 2012). Another terpene alcohol, geraniol, was found at high content in all treatments, above all in S treatment. This component is known as a floral, rose-like character in berries (LUND and BOHLMANN, 2006). Nerol was not found in any treatments in the free form, but at high concentration as bound-glycosidic precursor. Citronellol was present at low amount only in the S treatment, at amount similar to that of Muscat "a petit grains" grapes (SÁNCHEZ PALOMO *et al.*, 2007). Among polyoxygenated terpenes, diendiol I is predominant in grape juices, followed by diendiol II. Even if diendiol I is not considered to contribute directly to *Muscat* aroma due to its low sensory relevance, it is a precursor of monoterpenol odorants such as hotrienol and nerol oxide in wines (WILLIAMS *et al.*, 1980). Among C6 compounds, the major compounds of the *Muscat blanc* grape were the C6 alcohols in particular 1-hexanol and (E)-2-hexen-1-ol, known as responsible for the green and herbaceous aromas of grape and wines (GOMEZ *et al.*, 1995). A predominance of C6 alcohols and aldehydes has also been reported in musts of Semillon grapes, that presented a herbaceous note, when influenced by an excess of water (URETA and YAVAR, 1982).

Generally, the standard irrigation treatment seemed to increase the level of free and bound volatile compounds compared to the other treatments. In total, the content of five free varietal monoterpenoids (linalool, citronellyl formate, geraniol, (*Z*)-8-hydroxy-linalool and geranic acid), 2 C6-compounds (1-hexanol and (*E*)-2-hexen-1-ol) and three C13-norisoprenoids were significantly higher in the S treatment compared to the other water regimes. In fact linalool increased about 1.8 times compared to the D treatment ($P<0.05$ with ANOVA). Linalool represented about 28% of free monoterpenoids in the S treatment, about 29% in the M treatment and a low content in the drought regime (about 23%). The geraniol level in the S treatment was almost 2 times higher than in other treatments, accounting for 19% of all monoterpenoids ($P<0.05$ with ANOVA). Thus, the significant major contribution of linalool and geraniol, both the most distinctive Muscat-like varietal compounds, observed in the standard

Table 2 - Free volatile compounds (μgL^{-1})* in mountain *Muscat* grapes influenced by different irrigation treatments.

Compounds*	LRI$_{calc}$§ (LRI$_{lit}$@)	ID⊥	Concentration (μgL^{-1}) S	M	D	P ξ
C6 compounds						
(*E*)-2-Hexenal	1201 (1192)	I	4.0±0.6	6.3±8.5	2.8±1.6	NS
1-Hexanol	1352 (1356)	I	38.3±5.9^{a}	21±10.2^{b}	18.2±8.7^{b}	0.002
(*Z*)-3-Hexen-1-ol	1380 (1386)	I	6.8±1.9	5.2±2.0	4.0±1.9	NS
(*E*)-2-Hexen-1-ol	1404 (1409)	I	38.0±7.0^{a}	22.7±8.9^{b}	20.3±9.4^{b}	0.005
Total			87.1±15.4^{a}	55.2±29.6^{b}	45.3±21.6^{b}	0.006
Monoterpenoids						
β-Myrcene	1154 (1145)	I	2.2±2.1	2.8±3	0.7±0.8	NS
α-Phellandrene	1163 (1166)	I	ND	0.3±0.8	ND	NS
Limonene	1181 (1218)	I	3.3±2.4	3.5±4.5	2±0.6	NS
(*Z*)- β-Ocimene	1226 (1225)	II	1.2±1	1±1.7	ND	NS
γ-Terpinene	1229 (1238)	I	ND	0.2±0.4	ND	NS
(*E*)- β-Ocimene	1241 (1250)	II	2.3±1.9	2.5±3.3	ND	0.040
Terpinolene	1266 (1287)	I	1.5±1.6	1.8±2	ND	NS
(E)-Furan linalool oxide	1432 (1437)	I	4.8±2.2	4.2±1.9	2.7±0.8	NS
(Z)-Furan linalool oxide	1460 (1476)	I	6.5±1.9	6.5±2.3	5.3±2	NS
Linalool	1546 (1562)	I	211.2±60^{a}	167.7±41.7^{ab}	118.5±41.2^{b}	0.016
ho-Trienol	1605 (1623)	II	3.8±1.9	6.8±11.4	2.2±1.5	NS
α-Terpineol	1686 (1688)	I	4.2±1.2	3.5±0.5	3.2±0.8	NS
Ethoxy-diol I	1702	III	4.5±1	3±1.3	3±2.1	NS
(E)-Pyran linalool oxide	1728 (1710)	II	44.8±7.6	45±8.5	41.8±10.3	NS
(Z)-Pyran linalool oxide	1757 (1754)	II	4.8±1.2	6.3±2.3	5±1.3	NS
Citronellyl formate	1764	III	11.3±3.3^{a}	5±3.5^{b}	5.5±4^{b}	0.014
β-Citronellol	1765 (1775)	II	1.8±4.5	ND	ND	NS
Geraniol	1844 (1858)	I	142.7±33.2^{a}	75.5±23^{b}	82.5±53.3^{b}	0.016
Diendiol I	1948 (1949)	I	95±22.4	94.8±18.3	97.5±30.5	NS
Endiol	1980 (1986)	II	1.2±1.8	ND	ND	NS
Diendiol II	2126 (2134)	I	68.8±19.5	64.8±14.8	52.5±10.3	NS
Hydroxy citronellol	2205		12.3±2.8	ND	8.7±3	0.001
8-Hydroxy-6,7-dihydrolinalool	2208 (2220)	II	3.8±5.9	5.2±5.7	7.2±6.6	NS
(E)-8-Hydroxylinalool	2269 (2270)	II	18.2±5.5	16.5±5.3	14.5±3.9	NS
(Z)-8-Hydroxylinalool+ hydroxy-geraniol	2308 (2310)	II	35.5±6^{a}	17.7±14.3^{b}	35.8±9.9^{a}	0.014
Geranic acid	2348 (2329)	I	58±14.2^{a}	43.5±10.3^{ab}	34.5±16.5^{b}	0.032
Total			743.7±205.1^{a}	578.1±180.8^{ab}	522.6±199.4^{b}	0.020
Alcohols						
Benzyl alcohol	1862 (1882)	I	4.7±4.3	1.2±2.9	1.8±2.9	NS
2-Phenylethanol	1895 (1929)	I	3.8±2.8	1.7±2	ND	0.014
Total			8.5±7.1	2.9±4.9	1.8±2.9	0.053

Diendiol I: 3,7-Dimethyl-1,5-octadien-3,7-diol; Endiol: 3,7-dimethyl-1-octen-3,7-diol or 2,6-dimethyl-7-octen-2,6-diol; Diendiol II : 3,7-dimethyl-1,7-octadien-3,6-diol ; *(E)*-8-Hydroxylinalool: (*E*)-2,6-dimethyl-2,7-octadien-1,6-diol; Hydroxy citronellol: 3,7-dimethyl-octan-1,7-diol; 8-Hydroxy-dihydrolinalool: 2,6-dimethyl-7-octen-1,6-diol); *(E)*-8-Hydroxylinalool: (*E*)-2,6-dimethyl-2,7-octadien-1,6-diol; hydroxyl-geraniol: 3,7-dimethyl-2-octen-1,7-diol.

*Values are mean ± standard deviation (*n*=6 samples per treatment).

§LRI: Linear retention index on column DB-Wax calculated (@LRI literature).

⊥ID: Compounds identified by comparing I: their mass spectra and retention indices (RI) with those of standard compounds; II: their mass spectra and Kovat's index with those reported in literature; and III: mass spectrum with mass spectral database.

ξ P values are referred to one-way ANOVA: Values of means followed by the same letter are not significantly different (P≤0.05). NS= not significant.

ND, not detected.

irrigation treatment could help to differentiate this treatment from the other water regimes.

Glycosidically-bound compounds

A total of 46 bound volatile compounds were detected and classified into four chemical categories: three C6 compounds (alcohols), 33 monoterpenoids, eight C13-norisoprenoid and two aromatic alcohols (Table 3).

Among the monoterpenoids, high levels of geranic acid (accounting for about 17% of all terpenoids), followed by nerol (~16%), geraniol (~12%), linalool (~11%), (*E*)-8-hydroxy linalool (~11%) and hydroxy-geraniol (~7%), were found in all treatments.

It was observed, in all bound fraction, the presence of a cyclic ether, *cis*-rose oxide, at very low content. This compound is already known as a potent varietal odorant in Scheurebe and

Table 3 - Glycosidically-bound compounds (μgL^{-1}) * in the juice of mountain *Chambave Muscat* grapes influenced by different irrigation treatments.

Compound*	LRI_{calc} § (LRI_{lit} @)	ID⊥	Concentration (μgL^{-1})			P ξ
			S	M	D	
C6 compounds						
1-Hexanol	1352 (1356)	I	18.7±6.8	21.5±3	27.0±11.2	NS
(Z)-3-Hexen-1-ol	1380 (1386)	I	2±2.4	3±2	3.8±1.3	NS
(*E*)-2-Hexen-1-ol	1404 (1409)	I	1.5±1.6	3.3±0.5	3.5±1.8	NS
Total			22.2±10.8	27.8±5.5	34.3±14.3	NS
Monoterpenoids						
β-Myrcene	1154 (1145)	I	18±16.4	8.7±3.8	6.2±1.9	NS
α-Phellandrene	1163 (1166)	I	2.3±4.4	1.5±1	0.3±0.8	NS
Limonene	1181 (1218)	I	15.5±12.1	10.5±3.7	7.3±2.1	NS
β-Phellandrene	1189 (1287)	II	2.3±2.1	0.5±0.8	ND	0.011
(*Z*)-β-Ocimene	1226 (1225)	II	9.7±9.4	4.2±1.5	2.8±1.3	NS
γ-Terpinene	1229 (1238)	I	3.2±3.1	2±0	1±1.1	NS
(*E*)-β-Ocimene	1241 (1241)	II	17.2±15.4	8.3±3.1	5.7±2.2	NS
Terpinolene	1266 (1287)	I	12.5±13.7	4.5±1.9	3.2±0.8	NS
cis-Rose oxide	1339 (1355)	I	3.2±1.2	2.3±1.6	2.3±0.8	NS
(E)-Furan linalool oxide	1432 (1437)	I	68.2±19.7	78.2±31.7	74.3±28.7	NS
(Z)-Furan linalool oxide	1460 (1476)	I	50.8±28.4	41.2±14.4	46.7±21.1	NS
Linalool	1546 (1562)	I	539.8±100.7	640.3±223.2	468.2±161	NS
ho-Trienol	1605 (1623)	II	8.5±5.6	6±3	5.3±1.2	NS
α-Terpineol	1686 (1688)	I	37.5±14.3	35.3±14.5	32.8±14.5	NS
(E)-Pyran linalool oxide	1728 (1710)	II	108.5±26.7	127.8±52.3	132.2±37.9	NS
(Z)-Pyran linalool oxide	1757 (1754)	II	7.7±3.6	9.3±5.4	10.8±5.8	NS
β-Citronellol	1765 (1775)	II	39.8±7.9	38±12.8	39.2±5.4	NS
Nerol	1797 (1811)	I	788.2±187.2	699.2±139.2	690.5±190.9	NS
Geraniol	1844 (1858)	I	588±125.9	607.3±90.5	576.8±166.9	NS
exo-2-Hydroxy-1,8-Cineole	1850 (1860)	II	ND	ND	1.8±2.6	NS
Diendiol I	1948 (1949)	I	235.5±277.9	157±51.7	172.5±43.4	NS
Endiol	1980 (1986)	II	20.7±16.5	16±6.1	17.7±5.5	NS
Diendiol II	2126 (2134)	I	79±57.8	67±24.2	69±13.2	NS
Hydroxy citronellol	2206 (2143)	II	103±98.7	82.3±18.8	100.3±23.9	NS
8-Hydroxy-6,7-dihydrolinalool	2208 (2220)	II	127.2±89.3	96±23.5	115.5±20.7	NS
Hydroxy-nerol	2266	III	36±25.7^{ab}	30.2±5.8^{b}	45±8.1^{a}	0.030
(E)-8-Hydroxylinalool	2269 (2270)	II	565.5±471.8	421.8±122.3	446.5±55.9	NS
(Z)-8-Hydroxylinalool	2277 (2310)	II	16.8±17.7	15.2±3.3	18±1.7	NS
Hydroxy-geraniol	2308 (2310)	II	288.8±182.9	272.7±59	348.2±21.7	NS
Geranic acid	2348 (2329)	I	857.5±192.7	966.7±144.5	969.3±193.3	NS
p-Menth-1-ene-7,8-diol	2501 (2517)	II	295±583.1	48±19.6	50.2±7.2	NS
(Z)-8-Hydroxy-nerol	2582	III	39.3±23.5	30.8±6.2	37.3±8	NS
(E)-8-Hydroxy-nerol	2590 (2613)	II	82.2±62.8	63±17.4	79.8±14.7	NS
Total			5067.4±2698.2	4591.8±1107	4576.7±1064.3	NS
C13-norisoprenoids						
3-Hydroxy-β-damascone	2520 (2537)	I	ND	0.2±0.4	0.7±0.5	0.030
3-Oxo-α-ionol	2606 (2635)	I	54.7±17.4	70.5±22.6	66.3±19	NS
3,4-Dehydro-7,8-dihydro-β-ionone	2623	III	71.5±53.4	53.7±17.3	46.7±10.1	NS
3,9-Dihydroxy-megastigman-5-ene	2629	III	158±95.9	116.8±33.2	119.2±18.5	NS
6,7-Dehydro-7,8-dihydro-3-oxo-α-ionol	2660	III	1.7±4.1^{b}	12.3±4.8^{a}	22.8±14.7^{a}	0.006
3-Hydroxy-7,8-dihydro-β-ionol	2676 (2675)	II	ND	11.3±9.4	18.2±4.1	0.007
Homovanillic alcohol	2889 (2892)	II	ND	8.5±19.4	1±0	0.005
Dihydroconiferyl alcohol	-	III	38±37.7	24.5±18.7	24±10.3	NS
Total			323.9±208.5	297.8±125.8	298.9±77.2	NS
Alcohols						
Benzyl alcohol	1862 (1882)	I	23±6.4	27.3±4	25.7±11.6	NS
2-Phenylethanol	1895 (1929)	I	29.5±4.3	29.7±3.2	28±6.2	NS
Total			52.5±10.7	57±7.2	53.7±17.8	NS

Hydroxy-nerol: 3,7-dimethyl-2-octen-1,7-diol, (*m/z*: 69, 121, 136, 43, 59).

*Values are mean ± standard deviation (*n*=6 six samples per treatment).

§LRI: Linear retention index on column DB-Wax calculated (@LRI literature).

⊥ID: Compounds identified by comparing I: their mass spectra and retention indices (RI) with those of standard compounds; II: their mass spectra and Kovat's index with those reported in literature; and III: mass spectrum with mass spectral database.

ξ P values are referred to one-way ANOVA: Values of means followed by the same letter are not significantly different (P≤0.05). NS= not significant.

ND, not detected.

Gewürztraminer wines, and was even found recently, in the black Muscat Hamburg grape variety (FENOLL *et al.*, 2009). Its stereoselective biosynthesis in grape berry mesocarp has recently been demonstrated (LUAN *et al.*, 2005). Moreover, grapes from drought treatment were characterized by the presence of a low level of 2-*exo*-hydroxy-1,8-cineole too, a component of grape *cv* Sauvignon (BITTEUR *et al.*, 1990).

The different water regimes did not modify the levels of monoterpenoids except for the hydroxynerol that was found at a higher level ($P< 0.05$) in the D treatment, and for β-phellandrene that was not present in D water regimes. In contrast, compared to the S treatment, the drought treatment seemed to increase the levels of four C13-norisoprenoids.

Both positive and negative effects of water stress on the content of free and glycoconjugated aromatic components have been reported in previous investigations. KOUNDOURAS *et al.* (2006) compared three non-irrigated sites widely differing in their water status and found higher levels of bound volatile compounds in wines from stressed locations. However, in this case, the observed differences may also be due to the fact that three different environments were compared. On the other hand, on non-aromatic grape *cv* of Sauvignon blanc, non volatile S-cysteine conjugate precursors were assessed during optimising irrigation, and, according to our results, severe water deficit stress seemed to limit aroma potential (PEYROT DES GACHONS *et al.*, 2005).

Mechanical properties

Table 4 shows the mechanical properties of berry skins. Higher average values of F_{sk} were found in parcels S, although no statistical differences were observed. In this treatment, the berries skins also show a higher springiness (lower value of E_{sk}). No differences were found in skin thickness (Sp_{sk}). The skin of the grape plays a critical role, regulating gas exchange between the berry and the surrounding environment, serving as a protective barrier against fungal disease and protecting the grape from UV light and physical injuries (LANG and DURING, 1990; GABLER *et al.*, 2003). Therefore, skin hardness evaluated by break skin force, is a positive parameter for the grape quality. This characteristic can also be used to characterize the *Muscat blanc* cultivars because this properties is a varietal marker, even if the parameters are strongly conditioned to the climatic course of the year (ROLLE *et al.*, 2011) and growing locations (LE MOIGNE *et al.*, 2008; RÍO SEGADE *et al.*, 2011b; RÍO SEGADE *et al.*, 2011c). In this study on *Muscat blanc* variety, no influence of irrigation treatments on F_{sk} and Sp_{sk} parameters were found. Therefore, this practice of vineyard management would not seem to induce significant changes on the resistance at the splitting and plant diseases.

Instead, in spite of the high dispersion of the data, significant differences in peduncle detachment resistance were present among all three different levels of irrigation. In particular, F_{ped} showed the lowest average values in the treatments S (-0.377N) and M (-0.478) (Table 5). However, these values are higher in comparison with other varieties (ROLLE *et al.*, 2010) and, in fact, no problems of shattering were observed.

The high resistance to shattering is an important property for the Muscat blanc grapes, because of the severe environmental conditions of growing present in mountain areas. In young berries the shatter is caused by the hydrolysis of pectins of the middle lamella of the cell walls

Table 4 - Mechanical properties of berry skin.

	S	M	D	*P*ξ
F_{sk} (N)	0.522±0.122[a]	0.487±0.101[a]	0.492±0.112[a]	NS
W_{sk} (mJ)	0.589±0.195[a]	0.478±0.167[b]	0.479±0.170[b]	0.018
E_{sk} (N/mm)	0.202±0.029[b]	0.229±0.521[a]	0.236±0.050[a]	<0.001
Sp_{sk} (μm)	199±47[a]	196±34[a]	200±47[a]	NS

F_{sk} = Berry skin break force; W_{sk} = Berry skin break energy; E_{sk} = Young's modulus of Skin; Sp_{sk} = Berry skin thickness. Average value ± standard deviation (n=30). ξP values are referred to one-way ANOVA; NS= not significant. Values of means followed by the same letter are not significantly different (P≤0.05).

Table 5 - Peduncle detachment resistance.

	S	M	D	*P*ξ
F_{ped} (N)	2.393±0.592[ab]	2.292±0.550[b]	2.770±0.684[a]	0.024
W_{ped} (mJ)	2.402±1.077[a]	2.157±0.897[a]	2.616±1.104[a]	NS

F_{ped} = Peduncle detachment force; W_{ped} = Peduncle detachment energy. Average value ± standard deviation (n=30). Average value ± standard deviation (n=30). ξP values are referred to one-way ANOVA; NS= not significant. Values of means followed by the same letter are not significantly different (P≤0.05).

forming a separate layer at the base of pedicel (RIBEREAU-GAYON *et al.*, 2000a). During grape maturation adverse meteorological conditions and a varietal-specific sensitivity are responsible of this phenomenon (RIBEREAU-GAYON *et al.*, 2000a). Therefore, the force of detachment of the pedicel (F_{ped}) is an effective parameter that should be monitored to assess this characteristic. Although the irrigation effect on this parameter at harvest cannot be considered a problem, these grapes could not be adapted to on-vine drying process where the decrease of F_{ped} is high (ROLLE *et al.*, 2009).

CONCLUSION

The assessment of free volatile compounds and their precursors together with the mechanical properties in white aromatic grapes may be important in selecting the best irrigation strategy in order to obtain the highest grape quality.

In this study the contributions of free linalool and geraniol, the most characteristic volatile varietal compounds of Muscat grape, were more dominant in the standard than in the drought regime, with an increase in standard irrigation of about 78% for free linalool and 73% for free geraniol respect to drought treatment.

Berry mechanical characteristics are important for assessing the resistance to fungal attacks and for the estimation of shattering. In particular, no influence of irrigation treatment on F_{sk} and Sp_{sk} parameters was noticed. Therefore, apparently, this practice of vineyard management does not result in significant changes to the resistance towards splitting and spread of plant diseases. Finally, the values of technological parameters of ripeness as well as berry weight and production yield of grapes from the three different water treatments showed no significant differences.

REFERENCES

Adams R.P. 2001. Identification of essential oil components by gas chromatography/quadrupole mass spectroscopy. Allured Publishing Corporation, Carol Stream.

Bitteur S.M., Baumes R.L., Bayonove C.L., Versini G., Martin C.A. and Dalla Serra A. 1990. 2-exo-Hydroxy-1,8-cineole: a new component from grape var. Sauvignon. J. Agric. Food Chem. 38:1210.

Bureau S.M., Razungles A.J. and Baumes R.L. 2000. The aroma of Muscat of Frontignan grapes: effect of the light environment of vine or bunch on volatiles and glycoconjugates. J. Sci. Food Agric. 80: 2012.

Chapman D.M., Roby G., Ebeler S.E., Guinard J. and Matthews M.A. 2005. Sensory attributes of Cabernet sauvignon wines made from vines with different water status. Aust. J. Grape Wine Res. 11: 339.

Del Caro A., Fanara C., Genovese A., Moio L., Piga A. and Piombino P. 2012. Free and enzymatically hydrolysed volatile compounds of sweet wines from Malvasia and Muscat grapes (*Vitis vinifera* L.) grown in Sardinia. S. Afr. J. Enol. Vitic. 33: 115.

Deloire A., Carbonneau A., Wang Z.P. and Ojeda H. 2004. Vine and water: a short review. J. Int. Sci. Vigne Vin. 38:1.

Di Stefano R. 1991. Proposal for a method of sample preparation for the determination of free and glycoside terpenes of grapes and wines. Bull. OIV. 721-722: 219.

Fenoll J., Manso A., Hellín P., Ruiz L. and Flores P. 2009. Changes in the aromatic composition of the *Vitis vinifera* grape Muscat Hamburg during ripening. Food Chem. 114: 420.

Gabler M.G., Smilanick J.L., Mansour M., Ramming D.W. and Mackey B.E. 2003. Correlation of morphological, anatomical and chemical features of grape berries with resistance to *Botrytis cinerea*. Phytopathology. 93:1263.

Gambacorta G., Antonacci D., La Gatta M., Faccia M., La Gatta B., Pati S., Coletta A. and La Notte E. 2011. Phenolic composition of Aglianico and Nero di Troia grapes and wines as affected by cover cropping and irrigation. Ital. J. Food Sci. 23: 381.

Giordano M., Rolle L., Zeppa G. and Gerbi V. 2009. Chemical and volatile composition of three Italian sweet white passito wines. J. Int. Sci. Vigne Vin. 43:159.

Gladstone J.S. 1992. Viticulture and Environment, Winetitles, Adelaide.

Gomez E., Martinez A. and Laencina L. 1995. Changes in volatile compounds during maturation of some grape varieties. J. Sci. Food Agric. 67: 229.

Gunata Y.Z., Bayonove C., Baumes R. and Cordonnier R. 1985, The aroma of grapes. Localisation and evolution of free and bound fractions of some grape aroma components *cv* Muscat during first development and maturation. J. Sci. Food Agric. 36: 857.

Hepner Y., Bravdo B., Loinger C., Cohen S. and Tabacman H. 1985. Effect of irrigation and crop level on growth, yield and wine quality of Cabernet sauvignon. Am. J. Enol. Vitic. 36:132.

Koundouras S., Marinos V., Gkoulioti A., Kotseridis Y. and van Leeuwen C. 2006. Influence of vineyard location and vine water status on fruit maturation of nonirrigated *cv* Agiorgitiko (*Vitis vinifera* L.). Effects on wine phenolic and aroma components. J. Agric. Food Chem. 54: 5077.

Lambri M., Dordoni R., Silva A. and De Faveri 2012. Comparing the impact of bentonite addition for both must clarification and wine fining on the chemical profile of wine from Chambave Muscat grapes. Int. J. Food Sci. Sci. Tech. 47:1.

Lang A. and During H. 1990. Grape berry splitting and some mechanical properties of the skin. Vitis. 29:61.

Le Moigne M., Maury C., Bertrand D. and Jourjon F. 2008. Sensory and instrumental characterisation of Cabernet franc grapes according to ripening stages and growing location. Food Qual. Prefer. 19: 220.

Letaief H., Rolle L. and Gerbi V. 2008. Mechanical behaviour of winegrapes under compression tests. Am. J. Enol. Vitic. 59: 323.

Luan F., Mosandl A., Münch A. and Wüst M. 2005. Metabolism of geraniol in grape berry mesocarp of *Vitis vinifera* L. *cv* Scheurebe: demonstration of stereoselective reduction, E/Z-isomerization, oxidation and glycosilation. Phytochem. 66: 295.

Lund S.T. and Bohlmann J. 2006. The molecular basis for wine grape quality. A volatile subject. Science. 311: 804.

Marais J. 1983. Terpenes in the aroma of grapes and wines: A review. S. Afr. J. Enol. Vitic. 4: 49.

Matthews M.A., Ishii R., Anderson M.M. and Omahony M. 1990. Dependence of wine sensory attributes on vine water status. J. Sci. Food Agric. 51: 321.

Maury C., Madieta E., Le Moigne M., Mehinagic E., Siret R. and Jourjon F. 2009. Development of a mechanical texture test to evaluate the ripening process of Cabernet franc grapes. J. Text. Stud. 40: 511.

Mercalli L. 2003. Atlante climatico della Valle d'Aosta. Società Meteorologica Subalpina, Torino.

Ojeda H., Deloire A. and Carbonneau A. 2001. Influence of water deficits on grape berry growth. Vitis. 40:141.

Peyrot des Gachons C.P., Van Leeuwen C., Tominaga T., Soyer J.P., Gaudillere J.P. and Dubourdieu D. 2005. Influence of water and nitrogen deficit on fruit ripening and

aroma potential of *Vitis vinifera* L *cv* Sauvignon blanc in field conditions. J. Sci. Food Agric. 85: 73.

Qian M.C., Fang Y. and Shellie K. 2009. Volatile composition of Merlot wine from different vine water status. J. Agric. Food Chem. 57:7459.

Ribéreau-Gayon P., Boidron N. and Terrier A. 1975. Aroma of Muscat grape varieties. J. Agric. Food Chem. 23:1042.

Ribéreau-Gayon P., Glories Y., Maujean A. and Dubourdieu D. 2000a. Handbook of Enology, Vol. 1, The microbiology of wine and vinifications, John Wiley and Sons Ltd. Baffins Lane, Chichester.

Ribéreau-Gayon P., Glories Y., Maujean A. and Dubourdieu D. 2000b. Handbook of Enology, Vol. 2, The chemistry of wine stabilization and treatments, John Wiley and Sons Ltd. Baffins Lane, Chichester.

Río Segade S., Giacosa S., Gerbi V. and Rolle L. 2011c. Berry skin thickness as main texture parameter to predict anthocyanin extractability in winegrapes. LWT - Food Sci. Technol., 44: 392.

Río Segade S., Orriols I., Giacosa S. and Rolle L. 2011a. Instrumental texture analysis parameters as winegrapes varietal markers and ripeness predictors. Int. J. Food Prop. 14: 1318.

Río Segade S., Soto Vázquez E., Orriols I., Giacosa S. and Rolle L. 2011b. Possible use of texture characteristics of winegrapes as markers for zoning and their relationship with anthocyanin extractability index. Int. J. Food Sci. Technol. 46: 386.

Rolle L., Gerbi V., Schneider A., Spanna F. and Río Segade S. 2011. Varietal relationship between instrumental skin hardness and climate for grapevines (*Vitis vinifera* L.). J. Agric. Food. Chem. 59: 10624.

Rolle L., Siret R., Río Segade S., Maury C., Gerbi V. and Jourjon F. 2012. Instrumental texture analysis parameters as markers of table-grapes and winegrape quality: a review. Am. J. Enol. Vitic. 63, 11.

Rolle L., Torchio F., Cagnasso E. and Gerbi V. 2010. Evolution of mechanical variables of the winegrapes for icewine production during on-vine drying. Ital. J. Food Sci. 22:143.

Rolle L., Torchio F., Giacosa S. and Gerbi V. 2009. Modification of mechanical characteristic and phenolic composition in berry skins and seeds of Mondeuse winegrapes throughout the on-vine drying process. J. Sci. Food Agric. 89:1973.

Sánchez Palomo E., Díaz-Maroto M.C., González Viñas M.A., Soriano-Pérez A. and Pérez-Coello M.S. 2007. Aroma profile of wines from Albillo and Muscat grape varieties at different stages of ripening. Food Control. 18: 398.

Ureta C.F. and Yavar O.L. 1982. Influence de quelques pratiques culturales sur la qualite des raisins. Conn. Vigne Vin. 16:187.

Usseglio-Tommaset L. and Di Stefano R. 1980. Profilo aromatico del Moscato bianco del Piemonte. Riv. Vitic. Enol. 33: 58.

van Leeuwen C., Tregoat O., Choné X., Bois B., Pernet D. and Gaudiliére J.-P. 2009. Vine water status is a key factor in grape ripening and vintage quality for red Bordeaux wine. How can it be assessed for vineyard management purposes? J. Int. Sci. Vigne Vin. 43:121.

Williams P.J., Strauss C.R. and Wilson B. 1980. Hydroxylated linalool derivatives as precursors of volatile monoterpenes of Muscat grapes. J. Agric. Food Chem. 28: 766.

Wirth J., Guo W., Baumes R. and Günata Z. 2001. Volatile compounds released by enzymatic hydrolysis of glycoconjugates of leaves and grape berries from *Vitis vinifera* Muscat of Alexandria and Shiraz cultivars. J. Agric. Food Chem. 49: 2917.

Zarrouk O., Francisco R., Pinto-Marijuan M., Brossa R., Santos R.R. Pinheiro C., Costa J.M., Lopes C. and Chaves M.M. 2012. Impact of irrigation regime on berry development and flavonoids composition in Aragonez (Syn. Tempranillo) grapevine. Agric. Water Manage. 114: 18.

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