Organoleptic and Quality Characteristics of Malagousia Variety Grapes Fermented with Selected Indigenous Yeast Strains

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Commercial Malagousia varietal wines, which are produced in almost all Greek viticultural zones, represent a relatively important part of Greek wine activity. This study presents the results of a profile compilation of volatile aroma compounds of Malagousia musts fermented under identical conditions with selected yeast strains. In total, 62 volatile aroma compounds were identified and separated into their chemical classes (aldehydes, higher alcohols, volatile phenols, terpenes, C13-norisoprenoids, lactones, esters, fatty acids, sulphur compounds, other compounds, and other alcohols). Alcohols and higher alcohols, such as cis-hexen-1-ol and geraniol, terpenes like linalool, esters such as ethyl isovalerate, ketones such us betadamascenone, beta-ionone and zingerone, and fatty acids such as geranic acid and phenylacetaldehyde, were found in all the samples. Among them, linalool and phenylacetaldehyde had the strongest effect on the volatile compound profile of Malagousia wines. The same wine samples were subjected to sensorial analysis by a trained panel of 10 wine tasters, and a statistical analysis of both analyses presents similarities between the two analysis approaches. It is hoped that the results will contribute to a better understanding of the quality potential of the Malagousia variety so as to evaluate possible differences on the basis of the detected aroma concentrations.

INTRODUCTION

There are more than 300 indigenous Greek grape varieties that are cultivated singly or in combination with the well-known international varieties in the nine different wine-growing Greek viticultural zones. Greek wine is trying to find its commercial place in an international environment where the competitiveness and commerce of wines is huge. There is great interest in creating typical products with a strong character and/or in relation to geographical names (Karampatea *et al*., 2021b). In the last decade, the wine produced from indigenous Greek grape varieties has receiving increasing appreciation in the global wine market (Vlachos *et al*., 2017). Malagousia, a white grape variety, has been characterised as the Cinderella of the Greek vineyard (Kourakou, 2016). The variety is mentioned for the first time in the book Oenological (1888) by Othon Roussopoulos and is not related to any protected designation of origin (PDO). Malagousia is authorised to be cultivated in all 11 viticultural departments and, more specifically, is a recommended variety in 43 regional zones of Greece, while is only authorised in the remaining 21 of the total number of 64 regional zones (Karampatea *et al*., 2021a).

The aroma potential of grapes is the consequence of five different systems/pools of specific aroma precursors that release wine varietal aroma during fermentation and/or ageing (Ferreira & Lopez, 2019). It is also well known that grape geographical origin has an influence on wine chemical composition (Francis, 2013; Lambrechts *et al*., 2000). Volatile compounds like terpenes, norisoprenoids, and fermentation-derived by-products (esters, alcohols) are also affected by grape origin (Schreier, 1979; Suomalainen & Lehtonen, 1979; Rapp & Mandery, 1986; Stashenko *et al*., 1992; Mauricio *et al*., 1997). Winemaking techniques also influence wine aroma (Vila *et al*., 2000), and grape aroma potential can be managed by applying adapted vinification protocols (Fraile, 2000). The dominant and major compounds contributing to wine aroma are formed during yeast fermentation (Stashenko *et al*., 1992). These compounds are higher alcohols, fatty acids, acetates, ethyl esters, ketones and aldehydes (Schreier,

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1979). Several studies have demonstrated that fermentation conditions (skin contact time, temperature, yeast, etc.) affect the final aromatic composition (Suomalainen & Lehtonen, 1979; Dubourdieu, 1986; Rapp & Mandery, 1986; Mauricio *et al*., 1997).

In our study we strictly followed the same winemaking protocol for all fermentations. However, the volatile composition of the wines varied with the yeast strain used to do the alcoholic fermentation. Estévez *et al*. (2004) confirmed that different strains from the same yeast family produce the same fermentation metabolites, but in different concentrations. For this reason, those yeasts having the most desired technological properties (producing fruity fermentative aroma, reducing production of higher alcohols or volatile phenols) are being actively sought and selected. This strategy is particularly valuable for obtaining a wine without defects and with the best aromas (Fraile *et al*., 2000; Lambrechts & Pretorius, 2000; Vila *et al*., 2000). The production of wines with different sensory characteristics from the same grape variety may have a commercial advantage by satisfying the different preferences of consumers.

The present paper is an attempt to clarify the aroma characteristics of Malagousia.

MATERIALS AND METHODS

Samples

Yeasts strains of *S. cerevisiae* were isolated from the Malagousia and Assyrtiko Greek white grape varieties (directly from grapes and/or during spontaneous fermentations at controlled temperatures) from five different Greek wine regions with protected geographical indication. A three-year study was undertaken on their isolation and characterisation. After screening their oenological properties, e.g. production of hydrogen sulphide, flocculation properties, fermentation rate expressed as CO_2 g/l losses between first and third day of fermentation, ethanol tolerance, osmotolerance, growth at high temperatures, malic and acetic acid consumption and enzymatic activities, eight strains of *S. cerevisiae* finally were selected as being the most suitable. Cultures of the selected strains were maintained at -20°C with 20% v/v glycerol as a cryoprotectant agent (Monaco *et al*., 2014).

The starter cultures were performed by transferring a single colony from YPD agar to YPD liquid medium and incubating it for 24 h at 26°C in an orbital shaker with a stirring rate of 120 rpm. Grape juice obtained from Malagousia grapes was inoculated with 10^6 cells/mL. After manual destemming, the white grapes were pressed at a maximum pressure of 0.5 bar using a vertical water press of 40 L. Each of the juices was mixed with 2 g/hL pectolytic enzyme (Clarizym, Exelcia Burgundia, France) for clarification and maintained at 10°C for 18 h. Just before the inoculation, the grape juice was filtered in order to confirm the dominance of the vaccinated strain. Fermentations in microscale vinifications were carried out in 30 L stainless steel thermoregulated tanks containing 25 L of Malagousia must with the following chemical characteristics: sugars 209 g/L; pH 3.55; titratable acidity 6.1 g/L tartaric acid; and initial yeast assimilable nitrogen 80 mg/L. A concentration of 30 ppm total SO_2 was added to the musts, and nutrient additions were made before inoculation (organic nitrogen 40 g/hL) and after the consumption of 150 g/L sugars (organic and inorganic nitrogen 40 g/hL). Two months after the alcoholic fermentation, the lees was discarded for a second time (the first took place at the end of the alcoholic fermentation) and three bottles were prepared from each tank (Karampatea *et al*., 2021b).

Extraction SPE and GC-MS/MS

Sample preparation and the extraction of free aromatic compounds were performed according to a modification of the method described in López *et al*. (2002) and Vrhovsek *et al*. (2014). The sorbent cartridges were placed in the extraction system and rinsed with 4 mL of dichloromethane, 4 mL of methanol and, finally, with 4 mL of a water-ethanol mixture (12% v/v). A total of 50 mL of wine, containing 25 mL of BHA solution, was put through the solid-phase extraction cartridge at a rate of 2 mL/min. The sorbent was then dried by passing air through it (20.6 Bar, 10 min). The analytes were recovered by elution with 1.3 mL of dichloromethane and 25 mL of the elution solution, and added on top of the eluted sample. The mixture was then hermetically sealed and stored at -25°C until GC-MS analysis. Calibration charts were prepared by GC-MS by analysis of dichloromethane solutions containing known amounts of standards and internal standards.

For the GC-MS/MS analysis, the method used by Paolini *et al*. (2018) was followed, with some modification, using the Agilent Intuvo 9000 fast gas chromatography system, coupled with an Agilent 7010B triple quadrupole mass spectrometer (Agilent Technologies, Santa Clara, CA, USA) equipped with an electronic ionisation source operating at 70 eV. Separation was achieved by injecting 1 μL at running split (1:10) into a DB-WaxUltraInert column (30 m, 0.25 mm and 0.25 m film thickness, Agilent Technology, Santa Clara, CA, USA). The initial gas chromatograph oven temperature was 40°C for 2 min, increased with 10°C/min to reach 55°C, then 20°C/min to 165°C, 40°C/min to 240°C for 1.5 min. Final setting was 50°C/min to 250°C and kept at this temperature for 4 min. Total run time was 16 min (Carlin *et al*., 2022).

Helium was used as the carrier gas, with a flow rate of 1.2 mL/min. Mass spectra were acquired in monitoring and multiple reaction mode. Nitrogen was used as the collision gas, with a flow of 1.5 mL/min, and additional helium with a flow of 4.0 mL/min was used as the quenching gas. The transfer line and source temperature were set at 250°C and 230°C, respectively. Finally, data collection and subsequent analyses were performed using Mass Hunter Workstation software (Carlin *et al*., 2022).

For the analysed wines, the R^2 was in a range from 0.986691 to 0.998692 for all compounds, and indicated good fit and linearity for the calibration curves. The limits of quantification (LOQ) for all compounds were from 0.14 to 25.00, which is suitable for their quantification in all white wines. The linearity limit was from 75 to 2 500 for the major compounds. The chromatographic run of only 16 min allowed high production capacity. The extraction method, together with the fast GC-MS/MS analysis, made it possible to significantly reduce the use of the DCM solvent, with advantages in terms of operator safety as well as time, thereby avoiding further concentration of the extracts and allowing for the quantification of 62 compounds. All the validation parameters are reported in Tables 1 and 2.

Sensory evaluation

The evaluation of the wines produced from the experimental microscale vinifications of 25 L was done by a trained panel of 10 judges. The wines were tasted blind, in random order, and each connoisseur had to rate them using a specific given questionnaire. More specifically, 30 mL of wine was served in a 21.5 cL tulip-shaped glass (ISO) at a temperature of 12°C. The tasters scored the intensity and quality of the aroma and taste, trying to distinguish them as fruity and/or flowery aroma, or fruity, floral, sour, stiff wine body, aftertaste and, finally, the overall quality. The rating scale ranged from 1, corresponding to perception threshold, up to 5, corresponding to the maximum estimated intensity.

RESULTS AND DISCUSSION

Once the analysis of several aromatic compounds was done, we examined which of them had a bigger concentration than the olfactory threshold, as cited in the bibliography. Compounds like geraniol, linalool, *cis*-3hexen-1-ol, 1-hexanol, phenylacetaldehyde, betadamascenone, zingerone, beta-ionone and geranic acid have at all eight samples concentration bigger than their threshold as mentioned in the literature. In addition, several ethyl esters were found in remarkable concentrations in the majority of the tasted wines.

In Table 1, the average values of the main volatile compounds chemical classes are presented, showing a significant difference among some wines, while individual volatile compounds are presented in Table 2. The most abundant classes were esters and other alcohol groups.

The primary aroma compounds, linalool and geraniol, usually have a maximum concentration immediately after fermentation and show a sharp decrease afterwards (Francis, 2013). They are considered to be the most important of the monoterpene alcohols, as they are present in greater concentrations and have lower flavour thresholds than other major wine monoterpenes (Etiévant, 1991).

Linalool and geraniol are two compounds with greater concentrations than their perception thresholds and their odour descriptor is citrus. More specifically, samples 9 and 10 had the higher concentrations. In addition, 1-hexanol always, with the exception of one sample, had a greater concentration than the perception threshold, with an odour descriptor of rose. Meanwhile, previous scientific work has described Malagousia wines as having aromas of citrus blossoms, rose and lemon (Nanou *et al.*, 2020).

Taking into account the sum of the weighted concentration values of all the aromatic compounds determined for each sample, we observed a clear superiority of sample 9, followed by samples 10, 5, 7 and 6. We reached the same conclusion taking into account the results of the wine tasting.

The aromatic profile of Malagousia wines ranges from herbal, minty and citrusy to peachy and tropical, as well as floral (Lazarakis, 2018; Karakasis, 2020). In our analysis of the results, we found a bigger coefficient for the samples with a bigger concentration of those aromatic compounds.

TABLE 1
Average volatile compound concentrations (μg L-1) of Malagousia wines fermented with different yeast strains

	Samples															
	3 (ΜΧΒ36)				**4 (ΑΧΒ23)**				**5 (ΑΚΣ23)**				**6 (ΜΑΣ18)**			
Parameters	SUM	**MIN**	**MAX**	Mean ± SD	SUM	**MIN**	**MAX**	Mean ± SD	SUM	**MIN**	**MAX**	Mean ± SD	SUM	**MIN**	**MAX**	Mean ± SD
P Aldehydes	149.976				70.904				24.637				91.427			
P Higher alcohols	260.986	0.183	222.895	52.197 ±96.429	279.118	0.261	234.04	55.824 ±100.828	112.721	0.127	97.345	22.544 ±42.133	254.429	0.248	205.012	50.886 ±87.276
P Volatile phenols	2.377	0.049	2.328	1.189 ±1.611	2.327	0.056	2.271	1.163 ±1.566	1.809	0.061	1.748	0.905 ±1.192	5.406	0.086	5.32	2.703 ±3.702
P Terpenes	129.696	0.025	58.576	13.006 ±19.954	238.262	0.005	100.681	27.862 ±35.754	98.662	0.327	63.805	13.646 ±19.792	140.514	0.014	54.282	16.954 ±20.515
P C13-norisoprenoides	0.247	0.247	0.247		0.296	0.296	0.296		0.275	0.275	0.275		0.28	0.28	0.28	
P Lactones	12.521	0.465	10.09	3.130 ±3.947	16.028	0.681	11.989	4.007 ±4.626	20.991	0.34	18.528	5.248 ±7.385	14.926	0.695	9.411	3.732 ±3.586
P Ketones	3.625	0.043	2.42	0.906 ±49.847	3.696	0.027	1.403	0,924 ±23.306	2.938	0.083	2.27	0.735 ±8.123	3.995	0.068	2.82	0.999 ±30.323
P Esters	394.576	0.025	104.6	26.305 ±35.056	914.686	0.005	480.372	0.924 ±23.306	1165.2	0.171	864.337	89.631 ±218.521	1107.335	0.014	343.815	79.095 ±99.303

TABLE 1 (CONTINUED)

	Samples															
	3 (MXB36)				**4 (AXB23)**				**5 (AKΣ23)**				**6 (MAΣ18)**			
Parameters	SUM	**MIN**	**MAX**	Mean ± SD	SUM	**MIN**	**MAX**	Mean ± SD	SUM	**MIN**	**MAX**	Mean ± SD	SUM	**MIN**	**MAX**	Mean ± SD
P Fatty acids	60.058	21.548	38.51	30.029 ±11.994	92.407	25.654	66.753	60.979 ±120.182	180.861	58.569	122.292	90.431 ±45.059	57.567	23.057	34.51	23.783 ±8.098
P Other alcohols	125.842	9.558	1121.625	314272 ±541.251	2515.692	14.961	2231.749	46.203 ±29.061	32.611	0.298	23.342	8.222 ±10.880	1568.533	10.131	1431.596	392.203 ±695.307
P Sulfur compounds	2.634				14.897				1.999				2.339			
P Oxanes	0.226	0.058	0.168	0.113 ±0.077	0.469	0.0127	0.342	0.234 ±0.152	0.096	0.022	0.074	0.048 ±0.037	0.33	0.087	0.243	0.165 ±0.110

	7 (AΔΣ21)				**8 (MKΣ19)**				**9 (MΔΣ25)**				**10 (MΔΣ30)**			
Parameters	SUM	**MIN**	**MAX**	Mean ± SD	SUM	**MIN**	**MAX**	Mean ± SD	SUM	**MIN**	**MAX**	Mean ± SD	SUM	**MIN**	**MAX**	Mean ± SD
P Aldehydes	40.270				44.744				55.635				54.211			
P Higher alcohols	219.149	0.301	189.126	43.830 ±81.741	100.22	0.125	68.978	20.044 ±29.523	145.837	0.496	76.226	29.167 ±35.570	137.78	0.332	87.714	27.556 ±37.652
P Volatile phenols	3.746	0.208	3.538	1.873 ±2.355	0.647	0.047	0.6	0.323 ±0.391	1.883	0.068	1.815	0.941 ±1.235	2.713	0.38	2.334	1.357 ±1.381
P Terpenes	150.146	0.163	63.528	17.780 ±23.098	247.419	0.011	152.07	31.409 ±46.947	454.422	0.013	287.661	57.990 ±88.611	359.359	0.209	209.791	45.479 ±65.858
P C13-norisoprenoides	0.306	0.306	0.306		0.435	0.435	0.435		0.681	0.681	0.681		0.457	0.457	0.457	
P Lactones	21.44	0.573	14.543	6.360 ±5,682	23.694	0.525	19.434	5.924 ±7.652	42.813	0.051	27.9	8.563 ±10.611	19.909	0.658	12.987	4.977 ±5.085
P Ketones	5.719	0.019	4.416	1.430 ±13.262	5.007	0.031	4.259	1.252 ±14.771	7.392	0.034	3.641	1.848 ±18.298	6.361	0.013	4.915	1.590 ±17.876
P Esters	1442.035	0.162	585.274	103.003 ±172.330	767.34	0.011	447.166	51.156 ±110.7681	900.297	0.013	268.247	60.020 ±87.495	1185.465	0.2	414.35	84.676 ±120.541
P Fatty acids	68.371	30.069	38.302	34.186 ±5.822	61.623	30.004	31.619	30.811 ±1.142	90.634	30.784	59.851	45.317 ±20.553	103.271	45.749	57.522	51.635 ±8.324
P Other alcohols	2743	16.099	2146.42	685.826 ±1010.43	2143.173	12.5	1469.854	535.902 ±694.873	2591.265	18.439	2313.756	647.987 ±1116	2299.618	13.004	1678.125	575.019 ±788.235
P Sulfur compounds	1.331				1.291				1.097				1.25			
P Oxanes	0.3	0.072	0.227	0.150 ±0.109	0.194	0.045	0.149	0.097 ±0.073	0.377	0.093	0.283	0.188 ±0.134	0.659	0.171	0.488	0.329 ±0.225

TABLE 2
Individual volatile compound concentrations (μg L−1) of Malagousia wines fermented with different yeast strains

Compounds	ODT (μg/L)	Odour Descriptor	Sample03	Sample04	Sample05	Sample06	Sample07	Sample08	Sample09	Sample10	SD	Average
Terpenes												
1,4-cineole			NF	NF	NF	NF	NF	NF	NF	NF		
1,8-cineole	3.2 Francis L. (2012)	Eucalyptus Francis L. (2012)	0.025	0.005	NF	0.014	NF	0.011	0.013	NF	0.009	0.014
Linalool oxide A (linalool oxide)	3000 Ferreira V. et al., (2002)	Flower,clove,curry Ferreira V. et al., (2002)	2.056	3.503	3.011	1.897	1.665	4.622	5.170	3470	1.279	3.174
Linalool oxide B (trans-linalool oxide)	6000 Ferreira V. et al., (2002)	Flower Ferreira V. et al., (2002)	1.425	4.733	2.354	1.482	1.207	8.369	9.676	3.188	3.294	4.054
Linalool	25 Ferreira V. et al., (2019)	Citric, flowery, fresh Gómez-Míguez M. J., et al., (2007) ;Noguerol-Pato R.,(2013)	58.576	100.681	63.805	54.282	63.528	152.070	287.661	209.791	86.104	123.799
Terpinen-4-ol	44		0.373	0.369	0.327	0.260	0.305	0.474	0.948	0.440	0.218	0.437
Safranal		Saffron Delgrado J.A. et al., (2020)	0.165	0.226	0.145	0.176	0.163	0.220	0.336	0.209	0.061	0.205
alpha-Terpineol	250 Ferreira V. et al., (2019)	Floral, Citrus, Sweet Falcao L et al., (2012)	12.819	24.524	19.791	13.477	17.056	44.557	69.765	39.716	19.846	30.213
beta-Citronellol	100 Toci A. et al., (2012)	Rose, spicy, flowery Peinado R.A. et al., (2016) Falcao L. et al., (2012)	37.150	74.388	5.221	45.854	49.616	26.751	55.542	74.808	23.457	46.166
Nerol	300 Niu Y. et al., (2020)	Violet, Floral, Rose Guth H., (1997)	17.106	29.834	4.010	23.072	16.607	10.345	25.311	27.738	8.933	19.253
Monoterpenoid												
Geraniol	30 Framcis L. (2012)	Citric, Geranium Noguerol-Pato R.,et al., (2013)	33.417	37.790	12.668	34.193	22.797	26.878	58.371	41.415	13.588	33.441
trans-terpin (Trans-1,8-terpin)			0.360	0.308	1.178	0.246	0.428	1.385	1.626	0.389	0.559	0.740

TABLE 2 (CONTINUED)

Compounds	ODT (µg/L)	Odour Descriptor	Sample03	Sample04	Sample05	Sample06	Sample07	Sample08	Sample09	Sample10	SD	Average
C13-norisoprenoids												
TDN (1,1,6-Trimethyl-1,2-dihydronaphthalene)	2 Carlin S. et al., (2019)	Gasoline, kyrosine Carlin S. et al., (2019); Escudero A. et al., (2004)	0.247	0.296	0.275	0.280	0.306	0.435	0.681	0.457	0.147	0.372
C6 alcohols												
1-Hexanol	8000 Gómez-Míguez M. J., et al., (2007)	Flowery, Rose Cullere L., et al., (2004)	1121.625	2231.749	23.342	1431.596	2146.420	1469.854	2313.756	1678.125	751.649	1552.058
trans-3-Hexen-1-ol	1000 Peinado R.A., et al., (2004)	Grass, resinous, cream Peinado R.A., et al., (2004)	9,.58	14.961	0.298	10.131	16.099	12.500	18.439	13.004	5.536	11.874
cis-3-Hexen-1-ol	400 Peinado R.A., et al., (2004)	Grass, Green Peinado R.A., et al., (2004)	125.659	268.981	8.972	126.805	580.481	660.818	259.070	608.489	252.019	329.909
Higher alcohols												
Guaiacol	9.5 Boidron J.N., et al., (1988)	Medicinal Flque E. et al., (2001)	0.183	0.439	0.127	0.278	0.301	0.125	0.945	0.332	0.267	0.341
Benzyl alcohol	200000 Lopez R., et el., (1999);	Floral, Sweet Flque E. et al., (2001)	222.895	234.040	97.345	205.012	189.126	68.978	76.226	87.714	71.263	147.667
4-ethyl guaiacol	33 Boidron J.N., et al., (1988)	Toasted bread, smoky, clove Peinado R.A., et al., (2004)	0.220	0.261	0.225	0.248	0.390	0.398	0.496	0.719	0.173	0.369
4-vinylguaiacol	1100 Boidron J.N., et al., (1988)	Leather, clove, curry Cullere L., et al., (2004), Peinado R.A., et al., (2004)	4.271	6.588	2.357	14.698	6.535	3.842	9.799	7.601	3.906	6.961
Volatile phenols												
4-ethyl-phenol	Niu Y., et al., (2020), Peinado R.A., et al., (2004)	phenol, stable Peinado R.A., et al., (2004)	0.049	0.056	0.061	0.086	0.208	0.047	0.068	0.380	0.118	0.119
eugenol	6 Gómez-Míguez M. J., et al., (2007)	Cinnamon, clove Peinado R.A., et al., (2004)	2.328	2.271	1.748	5.320	3.538	0.600	1.815	2.334	1.403	2.494
Ethyl esters												
Ethyl butyrate	125 Sáenz-Navajas M.P., (2015)	Apple Falcão L.,et el., (2010)	28.319	57.350	0.679	141.368	97.957	52.610	97.063	125.128	48.472	75.059
Ethyl 2-methylbutyrate	18 Gómez-Míguez M. J., et al., (2007)	Apple Falcão L.,et el., (2010)	0.362	1.540	0.926	1.296	2.169	2.831	4.007	4.102	1.390	2.154

TABLE 2 (CONTINUED)

Compounds	ODT (μg/L)	Odour Descriptor	Sample03	Sample04	Sample05	Sample06	Sample07	Sample08	Sample09	Sample10	SD	Average
Ethyl isovalerate	3 Gómez-Míguez M. J., et al., (2007)	Fruity, lemon, anise Lopez R., et al., (1999);	1.449	1.609	8.336	1790	3.578	4.296	7.201	4.444	2.579	4.088
Ethyl capronate	0.08 Wu Q., et al., (2016)	green apple, aniseed, banana Gamero A., et al., (2020) Noguerol-Pato R.,et al., (2013)	0.025	0.005	NF	0.014	NF	0.011	0.013	NF	0.009	307.599
Ethyl heptanoate	0.08 Wu Q., et al., (2016)	green apple, aniseed Gamero A., et al., (2020) Noguerol-Pato R.,et al., (2013)	1.283	1.932	0.171	3.477	7.547	2.209	2.732	9.540	3.240	3.611
Ethyl caprylate	0.014 Lopez R., et al., (1999);	fruity, sweet, floral, banana, pear Lopez R., et al., (1999);	69.435	84.588	1.952	215.681	456.334	125.841	268.247	313.642	150.285	191.965
Ethyl leucate	400 Falcão L.,et el., (2010)	fresh blackberry	39.559	8.769	38.487	66.075	76.123	29.521	32.124	60.595	22.157	43.907
Ethyl caprate (decanoate)	200 Gómez-Míguez M. J., et al., (2007)	apple, floral, soap Jakatic Korenika A.M. J.,(2018)·	5.917	7.197	1.509	23.684	49.344	8.103	20.551	41.027	17.574	19.666
Diethyl succinate	200 Zhao P. et al., (2017)	overripe, aged Escudero A.; et al., (2004)	104.600	136.505	1.525	343.815	585.274	447.166	238.734	414.350	197.751	283.996
Ethyl phenylacetate	250 Cullere L., et al., (2004),	Fruity, floral, rose, honey Guth H., (1997)	1.941	2.400	3.433	1.488	2.407	2.154	2.805	3.523	0.705	2.519
Ethyl cinnamate	1.1 Gómez-Míguez M. J., et al., (2007)		0.099	0.094	NF	NF	0.162	0.130	0.085	0.200	0.071	0.128
Acetate esters												
Isobutyl acetate	1.600 Peinado R.A. et al., (2004)	Sweet, fruity, apple, banana	96.306	480.372	864.337	138.085	26.033	42.353	46.027	80.665	298.523	221.772
Hexyl acetate	1.500 Etievant X.P. et al., (1991), Sidhu D. et al., (2015)	Floral, green, fruity Escudero A.; et al., (2004)· Lopez R., et al., (1999);	4.965	19.975	1.823	19.184	11.206	4.827	15.409	8.070	6.903	10.682
Methyl salicylate	50 Cullere L., et al., (2004)	Aniseed Flque E. et al., (2001)	3.893	2.588	1.198	8.290	3.289	1.191	2.217	2.615	2.271	3.160
Phenylethyl acetate	250 Maga J.A., (1973);· Guth H., (1997) Sidhu D., et al., (2015);		36.424	109.763	240.826	143.088	120.613	44.097	163.083	117.567	65.264	121.932

TABLE 2 (CONTINUED)

Compounds	ODT (μg/L)	Odour Descriptor	Sample03	Sample04	Sample05	Sample06	Sample07	Sample08	Sample09	Sample10	SD	Average
Oxanes												
Rose oxide I	80-160 Escudero A.; et al., (2004)·	Floral, Green Escudero A.; et al., (2004)·	0.168	0.342	0.074	0.243	0.227	0.149	0.283	0.488	0.128	0.247
Rose oxide II	100 Francis L., (2012)	Lychee, rose Francis L., (2012)	0.058	0.127	0.022	0.087	0.072	0.045	0.093	0.171	0.047	0.084
Mercaptan												
Furfurylthiol	0.0004 Tominaga T. Et al., (1998)		NF	12.491	NF	NF	NF	NF	NF	NF	4.416	12.491
Benzylmercaptan			NF	NF	NF	NF	NF	NF	NF	NF	0.000	
Pyrazines												
2-sec-Butyl-3-Methoxypyrazine	0.0015 Shinohara T., (1985);	Bell pepper Smyth H.E., (2005);	NF	NF	NF	NF	NF	NF	NF	NF	0.000	
Aldeyde												
Phenylacetaldehyde	1 Guth H. (1999)	Fresh, flowery Ferreira V., et al., (2019);	149.976	70.904	24.637	91.427	40.270	44.744	55.635	54.211	39.264	66.475
Ketones												
Beta-Damascone			NF	NF	NF	NF	NF	NF	NF	NF	0.000	
Beta-Damascenone	0.05 Francis L., (2012)	Baked Fruit, Rape Fruit Gómez-Míguez M. J., et al., (2007)	2.420	1.395	2.270	2.820	4.416	4.259	3.641	4.915	1.229	3.267
Beta-ionone	0.09 Ferreira V., et al., (2019);	Violet Carlin S. et al., (2019)	0.689	0.872	0.418	0.641	0.599	0.279	0.349	0.551	0.194	0.550
2-aminoacetophenone	0.2 Buttery R.G., et al., (1994);	Sweet, Caramel Niu Y., (2020)	0.043	0.027	0.167	0.068	0.019	0.031	0.034	0.013	0.050	0.050
Zingerone	7	Toasty, Dry fruit Delgado J.A., et al., (2020);	0.473	1.403	0.083	0.466	0.686	0.438	3.367	0.883	1.041	0.975
Lactones												
trans-whiskey lactone	790 San-Juan F., et al., (2011);	Nutty, Coconut Zhao P., et al., (2017)	NF	NF	NF	NF	NF	NF	NF	NF	0.000	
g-octalactone	7 Noguerol-Pato R.,et al., (2013)		0.817	1.608	1.558	1.797	1.401	1.039	7.099	1.311	2.053	2.079
cis-whiskey lactone	67 San-Juan F. et al., (2011)	Nutty, Coconut Zhao P. et al., (2017)	NF	NF	NF	NF	NF	NF	NF	NF	0.000	

TABLE 2 (CONTINUED)

Compounds	ODT (μg/L)	Odour Descriptor	Sample03	Sample04	Sample05	Sample06	Sample07	Sample08	Sample09	Sample10	SD	Average
g-nonalactone	30 Ferreira V. et al., (2019)	Coco Ferreira V. et al., (1999)	10.090	11.989	18.528	9.411	14.543	19.434	27.900	12.987	6.144	15.610
g-decalactone	88 Ferreira V. et al., (2019)	Peach/Sweet Carlin S. et al., (2019)	0.465	0.681	0.565	0.695	0.573	0.525	1.695	0.658	0.397	0.732
d-decalactone	386 Ferreira V. et al., (2019)	Coconut, Peach Chearny M. et al., (2018) ; Ferreira V. et al., (1999)	1.148	1.751	0.340	3.024	4.922	2.696	6.068	4.953	2.035	3.113
Menthalactone			NF	NF	NF	NF	NF	NF	0.051	NF	0.018	0.051
g-dodecalactone			NF	NF	NF	NF	NF	NF	NF	NF	0.000	
Fatty acids												
Valeric acid	33 Toci A., et al., (2012)	Sweet, pungent, herbaceous Toci A., et al., (2012)	21.548	25.654	122.292	23.057	30.069	30.004	30.784	45.749	33.619	41.145
Geranic acid	7	Green, Floral Delgrado J.A. et al., (2012)	38.510	66.753	58.569	34.510	38.302	31.619	59.851	57.522	13.777	48.204
Benzothiazoles												
Benzothiazole	50 Gayon-Riberau P.,(2006)	Rubber Gayon-Riberau P.,(2006)	2.634	2.406	1.999	2.339	1.331	1.291	1.097	1.250	0.617	1.793

🟥 under LOQ 🟦 Over ODT 🟨 Over linerity limit ODT odour detection treshold SD-standard deviation

Samples 7, 9 and 10 had the biggest concentration of most of the examined aromatic compounds.

For the aroma, the conclusion of the overall assessment coincides with the individual assessments. From the average of the four indicators related to the aroma, some samples stand out (sample 5, followed by samples 10 and 9, are those with the highest averages). Among them, however, one (sample 9) had extremely low variability (standard deviation), which means that all its features were consistently high (and it did not have any excellent features, with some being mediocre). The same sample (9) is the one that had the best performance in the parameter, 'overall aroma rating'. This is important, because it demonstrates the objectivity and effectiveness of the grading/evaluation method followed.

The wine made with strain 10 had the highest mean (4.00; SD = 0.33). At the same time, Table 3 shows that the wine made with strain 10 was statistically significantly different from all the other wine strains (sig. = 0.00 < 0.05), which the post hoc test shows is the best strain among all in terms of 'fruity aroma'. This is followed by strains 3 and 7 both scoring 3.4 points.

According to Table 4, the wine made with the strain 9 had the highest average (mean = 3.25; SD = 0.35), followed by strain 7 (3.0) and strain 10 (3.0). At the same time, Table 5 shows that the wine made with strain 9 was statistically significantly different from that made with strains 4 (sig. = 0.00 < 0.05; mean difference = 1.45) and 8 (sig. = 0.00 < 0.05; mean difference = 0.85), with strain 9 being better in 'overall aroma evaluation' than strains 4 and 8 (Fig. 1).

Overall in relation to taste, sample 10 stands out positively and sample 3 negatively. The post hoc test found that wine made with strain 3 was the worst, also in terms of aroma quality. It is impressive how negatively sample 3 stands out (it is displayed as a data outlier, as if it were an error of observation), as also shown in Fig. 1. In the evaluation of taste, there was great homogeneity in five of the samples (samples 5, 6, 7, 9 and 10).

CONCLUSIONS

Some aromatic compounds were quantified in all eight white wines and their odour description was based on published data. The volatile profile of each wine is coherent with the organoleptic profile formed from the sensory analysis. Data from each wine gave us the opportunity to differentiate their quality and categorise them. Esters and other alcohols were the most dominant compounds in all of the wines, as they accounted for the largest proportion of the total aroma. The wine with the higher scores in terms of its organoleptic quality had the highest concentration of

TABLE 3
Post hoc for the association of wines on 'fruity aroma'

Dependent variable: Fruity aroma

(I) Sample	(J) Sample	Mean Difference (I-J)	Std. error	Sig.
10	**3**	0.60*	0.18	0.03
	4	1.40*	0.18	0
	5	1.40*	0.18	0
	6	0.80*	0.18	0
	7	0.60*	0.18	0.03
	8	1.20*	0.18	0
	9	0.75*	0.18	0

*The mean difference is significant at the 0.05 level.

TABLE 4
Means of wines regarding the 'overall aroma rating'

Sample	Mean	Std. deviation
3	2.80	0.42
4	1.80	0.67
5	2.60	0.57
6	2.60	0.21
7	3.00	0.41
8	2.40	0.57
9	3.25	0.35
10	3.00	0.41
Total	2.68	0.62

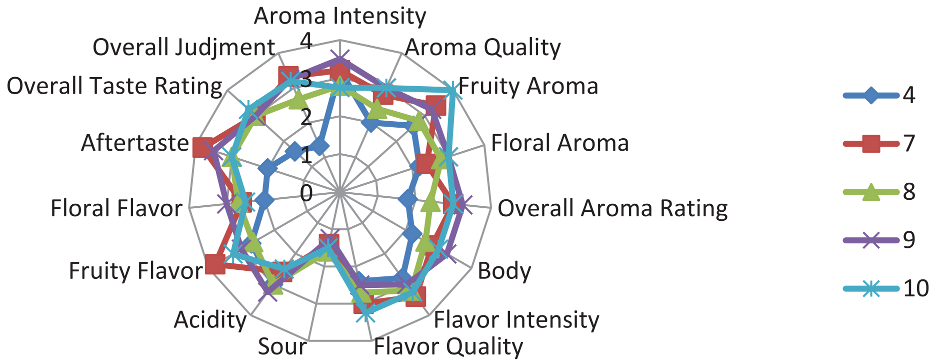

FIGURE 1
Organoleptic evaluation of different vinifications, using selected *S. cerevisiae* strains as starting cultures (information is not shown for all wine samples).

TABLE 5
Post hoc for the association of wines on the ‘overall aroma rating’

Dependent variable: Overall aroma rating

(I) Sample	(J) Sample	Mean Difference (I-J)	Std. error	Sig.
9	**3**	0.45	0.21	1.00
	4	1.45*	0.21	0.00
	5	0.65	0.21	0.08
	6	0.65	0.21	0.08
	7	0.25	0.21	1.00
	8	0.85*	0.21	0.00
	10	0.25	0.21	1.00

* The mean difference is significant at the 0.05 level.

terpenes, and this could explain the floral aroma and flavour descriptors. Significant differences in aroma were found among the white wines studied, helping to differentiate the fermentation results of the eight selected yeast strains. The major differences in aroma among these eight wines could be attributed to the variation in the intensity of fruity and floral notes, principally due to their content of ethyl esters, acetates, monoterpenes and 2-phenyl ethanol.

Such data could lead to a better understanding of Malagousia aromatic characteristics. However, this work also provides a basis for future research in terms of the variations in volatile aroma compounds within Malagousia wines, and for the development of models that better explain these variations. These variations could be due to geographic origin, which is associated with similar climatic conditions or soils, and the effect of using different oenological practices. Furthermore, it is important to better understand which precursors are present in Malagousia grapes and how to extract them or express them in wines.

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