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A Sustainable Approach Based on the Use of Unripe Grape Frozen Musts to Modulate Wine Characteristics as a Proof of Concept

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Abstract: Aiming to develop a sustainable methodology for must acidity correction in winemaking, particularly needed in warm regions, the present study intends to fulfill the circular economy values. Antão Vaz white wines were produced using two different strategies for must acidity correction: (i) the addition of a mixture of organic acids (Mix*) commonly used in winemaking; and (ii) the addition of previously produced unripe grape must (UM*) from the same grape variety. In addition, a testimonial (T*) sample was produced with no acidity correction. For all wines produced, oenological parameters were determined, and both amino acid (AA) content and volatile composition were evaluated. A higher AA content was found in the Antão Vaz T* wine, followed by UM* wines. The volatile profile was also affected, and LDA demonstrates a clear separation of wines with different acidity corrections. Results obtained indicate that unripe grape musts—a vital waste product containing several compounds with important biological activity—can be used to increase musts acidity without a negative impact on wine characteristics. Furthermore, this work also shows that the use of unripe must may be a valuable tool for reducing the alcoholic content of wines.

Keywords: acidity correction; unripe grape musts; circular economy; Alentejo wine grapes; Antão Vaz grapes; white wines; amino acids; volatile compounds

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1. Introduction

The chemical composition of grapes is influenced by various factors such as degree of maturity, variety, terroir, and year. Organic acids, having essential effects on characteristic fruit flavor, play a significant role in grape quality criteria, and consequently in wine characteristics such as stability, color and flavor. Acidity and sugar balance is fundamental to enhance grape flavor, which determines the wine quality. Grape juice with low acidity often results in unstable musts and wines susceptible to organoleptic degradation. On the contrary, excessive berry acidity is undesirable [1]. Although having the same genotype, grapes harvested under a different climate have different organic acid contents [2]. During grape ripening, continuous warm conditions result in a lower acid content at maturity, primarily due to the increasing degradation of malic acid. Must corrections of the acid-base balance (most often) can improve wine quality, by increasing acidity through organic acid addition [3]. More recently, some research groups have evaluated different winemaking techniques to regulate the ethanol content and pH of wines in response to the effect of global warming on the composition of grapes. High interannual climate variability has been

recorded during the ripening period, which strongly affects the composition of the grape. In particular, high temperatures during the ripening period cause an increased accumulation of sugars and degradation of acidity due to malic acid consumption, impacting the synthesis of polyphenols. In addition, thermal stress during the maturation period causes the degradation and inhibition of anthocyanins accumulation (compounds responsible for the color of grapes, and hence red wines) [4,5].

According to the European Union rules and the International Code of Enological Practices [6], only organic acids can be added to musts and wines to increase the total acidity and thus decrease the final pH. Inorganic acids are forbidden. It is not just a matter of balancing the wine flavor but also promoting good biological evolution and good wine preservation. Indeed, wine acidity is mainly due to the organic acids from grapes, such as tartaric, malic, and citric acids. Among those, tartaric acid is the most stable and has a higher pH impact. Tartaric and malic acids account for 90% or more of the total acidity in grapes. Malic acid is metabolized by lactic acid bacteria during malolactic fermentation [7–10]. The use of citric acid is only allowed in certain non-European winemaking countries. It is usually reserved for wine acidity correction, since lactic bacteria can metabolize it, thus increasing volatile acidity. Adding tartaric acid to the musts has been thought of as the traditional way to adjust the acidity. However, this methodology has limitations, including the costs of quality tartaric acid and the amount required to attain the desired pH decrease. Moreover, the added tartaric acid losses via precipitation of excessive potassium hydrogen tartrate, known as tartaric instability, leads to further acidity corrections. Other alternative strategies to adjust wine acidity include: (1) blending with higher acidity wines; (2) adding acids other than tartaric acid; (3) plastering; (4) the use of cation exchange resins; and (5) applying bipolar membrane electrodialysis [11]. Another option for pH modulation is to blend grapes that have been harvested at different ripeness stages, since it is well known that different grape varieties under the same edaphoclimatic conditions have different behavior, reaching a maturity level at different times [11].

Some studies have been published describing the applications of the unripe grapes in food and beverages [12–17], and also different strategies to reduce the alcohol concentration and pH of wine using unripe grapes [18–24]. Unripe grapes can be picked from the period of bunch closure to *véraison*. During the herbaceous growth phase, the berries are small, green, and complex, increasing their acid content. After *véraison*, the berries begin to soften, and sugars accumulate. The structure, composition, and hard consistency of the unripe berries account for the difficulty encountered in pressing this fruit and justify the low juice yield obtained from this raw material. Unripe grapes are however a rich source of flavonoid compounds, prominent tannins from seeds and skins, flavonols, and hydroxycinnamic acids, but a less rich source of anthocyanins than mature grapes [25]. Unripe grapes are a good waste product that can also be exploited and valorized, since they still contain all their endogenous nutrients and biologically active compounds [25].

Thus, the present work aims to develop a sustainable methodology for musts acidity correction in winemaking to reproduce circular economic values using unripe grape musts as a "green" tool to increase must acidity, so imperative in warm climates, as a novelty. Bearing this in mind, the goal was to evaluate the impact of adding unripe grape musts on wine characteristics.

2. Materials and Methods

2.1. Chemical Reagents and Standards

Methanol (HPLC grade), and glacial acetic acid (analytical grade), were purchased from Fisher Scientific. Acetonitrile used was HPLC grade and purchased from VWR International (Radnor, PA, USA). Hydrochloric acid was purchase from Honeywell, Fluka (Morris Plains, NJ, USA). Sodium azide, boric acid, all amino acid standards, L-2-aminoadipic acid (internal standard), and derivatizing agent diethyl ethoxymethylenemalonate (DEEMM) were analytical grade, purchased from Sigma Aldrich (St. Louis, MO, USA). The water

used in all experiments was distilled and purified by a Milli-Q system (Millipore, Bedford, MA, USA).

2.2. Fermentation Protocols

Wines were produced from white grapes of Antão Vaz, harvested in 2019 from the experimental vineyard of Évora University. Unripe grapes were harvest previously in the summer of the same year from the same experimental vineyard. Their musts were then produced by pressing and then frozen at −80 °C, to be used further on. Before use and after defrosted, total acidity (expressed in tartaric acid (TA)) and pH were measured: Antão Vaz must had a total acidity of 21.76 $g \cdot L^{-1}$ (TA), and a pH of 2.36.

At harvest, grape clusters were destemmed, crushed and pressed to obtain juice and after a cold static settling, must was distributed among glass vessels with a total of 4 L each. 100 mg/L of SO_2 was added, from a commercial 6% aqueous solution of sodium bisulfite (SAI, SOLFOX 6 Nº CE: 231-870-1), and a commercial Saccharomyces cerevisiae (mixture 1:1 of LEVULINE FB from Oenofrance and IOC 18–2007 from Lallemand OEnology) was inoculated. The assay was performed in duplicate, in a total of six final wines. Three groups of two vessels were considered. To each group, a different acidity correction was applied to obtain a similar total acidity value (around 6 $g \cdot L^{-1}$ (TA): (i) 440 mL of frozen unripe grape must addition (UM*); (ii) 19 mL of a mixed solution of tartaric, malic, and lactic acid (Mix*); and (iii) testimonial wine (T*) without any acidity correction. Fermentation took place at 16 °C and, at the end of the alcoholic fermentation process (residual sugar < 2 $g \cdot L^{-1}$), wines were transferred to another glass vessel to eliminate lees. Samples were collected for analysis. Chemical composition of musts and final wines were determined using described methods by the International Organization of Vine and Wine [26].

2.3. Analysis of Volatile Compounds

Volatile organic compounds (VOCs) were accessed and identified by HS-SPME sampling experiments [27] using a Divinylbenzene/Carboxen/Polydimethylsiloxane fiber (DVB/CAR/PDMS, 1 cm, 50/30 µm film thickness (df)) supplied from Supelco, (Bellefonte, PA, USA). A GC/MS system consisting of a Bruker GC 456 with a Bruker mass selective detector Scion TQ was used. An automatic sampler injector was used: CTC Analysis auto sampler CombiPAL. The chromatographic conditions were established according to a previous work [28]. Samples were injected in splitless mode, and the chromatographic separation was performed on a ZB-WAX PLUS capillary column (60 m × 0.32 mm i.d., 1.0 µm df) supplied by Phenomenex, Torrance, CA, USA. The linear retention index values (LRI) were calculated through analysis of the commercial alkane standard solution C8-C20, under the same chromatographic conditions. The relative amounts of individual components are expressed as percent peak areas relative to the total peak area (Relative Peak Area—RPA) [28,29]. All analyses were carried out in duplicate.

2.4. Analysis of Amino Acids

The determination of amino acids was carried out following the method described elsewhere [30]. Aminoenone derivatives were accessed by reaction with DEEMM and analyzed by liquid chromatography (HPLC) in a Waters Alliance System 2695 series with a photodiode array detector (2998 PDA Detector) (Waters, Milford, MA, USA). Before injection (10 µL), solutions were filtered through a 0.45 m nylon membranes filters (Whatman) and the detection was performed at 269, 280 and 300 nm. The quantitation was carried out using the internal standard method, and the respective calibration curve of each quantified amino acid was previously described [28] with some modifications on the ranges of the calibration curves. All the analysis were performed in triplicate.

2.5. Statistical Analysis

All computations, and the chemometric analysis, were carried out using SPSS Version 27.0 (IBM, Chicago, IL, USA). First, a one-way analysis of variance (ANOVA) was per-

formed, using for post hoc test comparison of means, Fisher's least significant difference (LSD) test was used at $p < 0.05$ for the oenological parameters, total amino acids, and total volatile compounds. Then, a principal component analysis (PCA) was used to analyze each wine's AA and volatile content to evaluate the systems' discrimination capability towards the different white wines produced. Afterward, a linear discriminant analysis (LDA) was used as a supervised method for the quantitative modeling of the data, which attempts to model differences among samples assigned to specific groups. It was performed based on the significantly different compounds for each wine sample. The method aims to maximize the ratio of the between-group variance and the within-group variance. When this ratio value is at its maximum, the samples within each group present the smallest possible scatter, and the group's separation is maximized [31].

3. Results

3.1. Oenological Analysis

The standard oenological parameters for all wines from both varieties produced with different acidity corrections are summarized in Table 1 for Antão Vaz wines. All parameters are within the legal values (International Organization of Vine and Wine, 2019). Total acidity was always higher than 3.5 $g{\cdot}L^{-1}$ (TA), and volatile acidity was under 1.2 $g{\cdot}L^{-1}$ (expressed in acetic acid). The alcoholic contents for all white wines range from 12.8% to 13.9% (v/v), and pH values range from 3.43 to 3.63.

Table 1. Average values for oenological parameters for all Antão Vaz wines. Frozen unripe grape must addition (UM*); addition of a mixed solution of tartaric, malic, and lactic acid (Mix*); testimonial wine (T*).

Sample Name	Free SO_2 ($mg{\cdot}mL^{-1}$)	Total SO_2 ($mg{\cdot}mL^{-1}$)	Ethanol (% Vol)	Total Acidity ($g{\cdot}L^{-1}$)	Volatile Acidity ($g{\cdot}L^{-1}$)	pH
T*	10.5 ± 0.6	40.0 ^{a} ± 1.1	13.8 ^{a} ± 0.1	4.83 ^{c} ± 0.02	0.29 ± 0.07	3.63 ^{a} ± 0.01
UM*	10.0 ± 0.0	33.5 ^{b} ± 2.1	12.8 ^{b} ± 0.0	6.17 ^{a} ± 0.01	0.24 ± 0.08	3.43 ^{b} ± 0.00
Mix*	12.0 ± 2.8	39.5 ^{a} ± 0.7	13.9 ^{a} ± 0.1	6.00 ^{b} ± 0.03	0.26 ± 0.02	3.43 ^{b} ± 0.01

Total acidity—expressed in tartaric acid; Volatile acidity—expressed in acetic acid. Each value represents the mean ± standard error of the mean. (Different letters in column mean significant differences at $p < 0.05$).

When performing ANOVA on these data, significant differences were obtained for the total content of SO_2 and pH, as UM* wines presented the lowest values in both parameters. Additionally, regarding the total acidity, significant differences were obtained between all wines. An increase for the UM* wine was obtained. Indeed, results indicate that using unripe must increases wine acidity, as expected, but the effect on pH is the same as the one obtained using chemical acidification.

Furthermore, regarding the ethanol content, significant differences were also achieved. In the wines where the acidity correction was performed by adding the unripe grape must (UM*), a decrease in the alcohol content can be observed as the acidity is corrected, obtaining the lowest content compared to Mix* and T* wines. The possibility of lowering the alcoholic range of wines using unripe grape musts is a pertinent achievement due to the expectable raising in ethanol concentration owing to climate change. High ethanol content can also modify the sensory profile of the wine, increasing the perception of bitterness and astringency [32]. Therefore, wines with lower acholic content are continually becoming a trendy market. Reportedly, soft alcohol beverages, such as reduced-alcohol wine, have become increasingly accepted by consumers. Forecasts assume a continuous growth in demand for low-alcohol drinks, reflecting the global trend for healthier lifestyles and awareness about the benefits of drinking wine [33,34].

Different strategies have already been proposed to reduce alcohol concentration, including vineyard management, grape must pre-fermentation practices, microbiological approaches during fermentation, and post-fermentation processing technologies [32,35,36]. Considering the results obtained in this study, a new dual strategy can be explored us-

ing unripe grape musts, enabling modulation of the acidity and alcoholic concentration of wines.

3.2. Amino Acid, VOCs and Aroma Profile of Antão Vaz White Wines

Significant differences were obtained for T* wines compared to the Mix* and UM* wines regarding AA content, with the T* wine showing the higher amounts of AA (1083.82 mg·L^{-1}), followed by the UM* wine (943.15 mg·L^{-1}), and the Mix* wine the lowest content (929.20 mg·L^{-1}), (Table 2). Proline is the primary amino acid responsible for the difference among wines. These values are all in accordance with values reported in previous studies where the amino acid composition of wines from white grapes are quantified [37–41].

Table 2. Average concentrations of the AA obtained from the analysis of all Antão Vaz wines. Each value represents the mean ± standard error of the mean for the samples analysed, UM*, Mix* and T*. (Different letters in row mean significant differences at $p < 0.05$).

		Antão Vaz Wines (mg·L^{-1})		
Abv.	**Compound**	**UM***	**Mix***	**T***
Asp	Aspartic Acid	6.62 ± 0.38 ^{a}	4.98 ± 0.59 ^{b}	5.96 ± 0.34 ^{a,b}
Glu	Glutamine	32.55 ± 6.15	27.37 ± 2.81	34.34 ± 7.36
Asn	Asparagine	7.01 ± 1.30	5.14 ± 0.69	6.36 ± 1.03
Ser	Serine	7.32 ± 0.73	6.83 ± 0.45	8.57 ± 1.23
His	Histidine	5.51 ± 0.48	5.68 ± 0.89	5.81 ± 0.69
Gln	Glutamic Acid	3.24 ± 0.18	2.92 ± 0.28	3.83 ± 1.83
Gly	Glycine	6.51 ± 0.58	5.83 ± 0.33	7.52 ± 0.92
Thr	Threonine	3.09 ± 0.13	2.91 ± 0.10	3.22 ± 0.18
Arg	Arginine	29.36 ± 2.32	26.36 ± 0.97	28.27 ± 2.25
Ala	Alanine	29.29 ± 2.29	27.02 ± 1.06	29.30 ± 2.44
GABA	Gamma Aminobutyric Acid	<7.88	<7.88	<7.88
Pro	Proline	767.99 ± 129.04	650.12 ± 71.67	1029.78 ± 240.84
Tyr	Tyrosine	6.89 ± 0.35	6.23 ± 0.15	6.91 ± 0.37
Val	Valine	3.30 ± 0.28	2.95 ± 0.13	3.51 ± 0.30
Met	Methionine	4.01 ± 0.13	3.72 ± 0.14	4.00 ± 0.15
Cys	Cysteine	14.48 ± 1.92	12.20 ± 1.00	12.85 ± 2.47
Ile	Isoleucine	3.16 ± 0.23	2.82 ± 0.12	3.27 ± 0.21
Trp	Tryptophan	3.42 ± 0.12 ^{a}	3.19 ± 0.08 ^{c}	3.38 ± 0.19 ^{b}
Leu	Leucine	9.78 ± 1.23 ^{a}	<8.51 ^{b}	9.62 ± 1.31 ^{a}
Phe	Phenylalanine	6.36 ± 0.72	5.19 ± 0.43	6.21 ± 0.81
Orn	Ornithine	10.85 ± 1.84	7.90 ± 0.55	8.93 ± 1.13
Lys	Lysine	11.06 ± 1.51	8.31 ± 0.67	10.81 ± 1.58
	Total	943.15 ± 128.67 ^{b}	929.20 ± 142.14 ^{b}	1083.82 ± 261.61 ^{a}

VOCs, with different polarities and volatilities, are produced in various concentrations. They have a crucial flavour impact and play a central role in defining wine sensorial identity. Each category of flavour compounds varies considerably among different wines with different predominant aromas, which confer specific typicity on each wine [42]. Regarding the ANOVA analysis on the VOCs, no significant differences were obtained among them; nonetheless, results obtained from the dataset for each sample were used to perform a PCA analysis that totalized the amounts of each chemical functional group, as a way to pinpoint the effects of the acidity methodology used on the winemaking process (Figure 1). In addition, loadings for each standardized variable (AAs and VOCs) for the first and second principal components (PC1 and PC2) were obtained. Standardization was performed on a correlation matrix between-groups and matrix plot. A row length normalization was applied to all AAs and VOCs values of each wine where all values were divided by the Euclidean norm of the row. Normalization is needed in order that the magnitude of a particular variable does not dominate the statistical treatment against other variables.

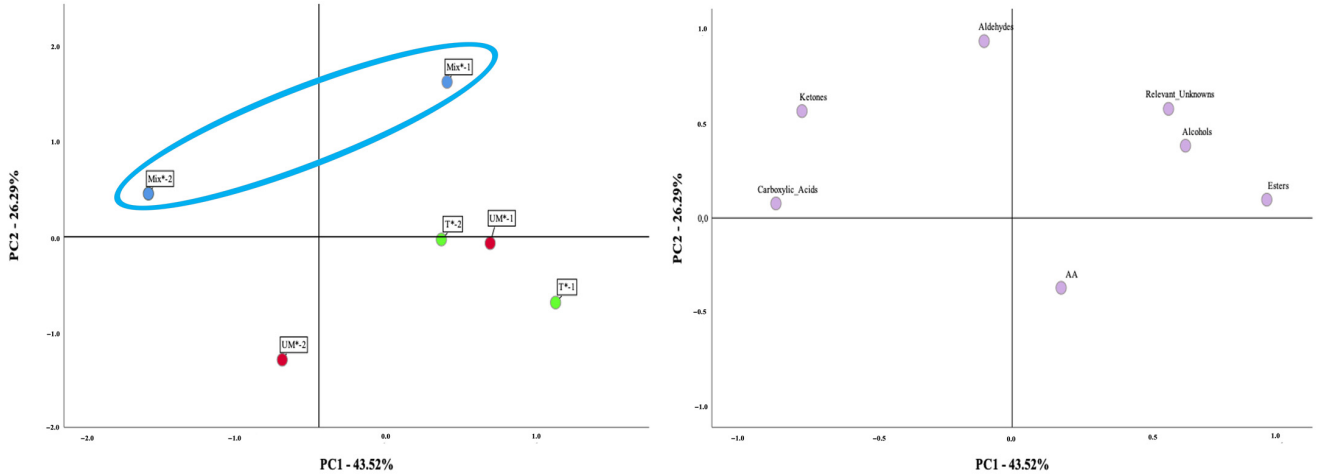

Figure 1. Principal component analysis (PCA) of total content of AA and VOCs data obtained for each Antão Vaz wines separated by chemical functional groups. Blue dots represent Mix* wine samples, green dots represent T* wine samples and red dots represent UM* wine sample.

According to PCA performed for each chemical functional group, Figure 1, PC1 and PC2, accounted for 43.52% and 29.26% of the total system variation, respectively. It is possible to observe that the Mix* white wines group is well separated from the other wine sample groups (UM* and T*), corresponding to the wine samples with higher amounts of aldehydes and ketones.

As for UM* and T* wines, it is not possible to obtain a separation based on the chemical groups, but it is possible to observe that those wine samples are the ones with higher amounts of AAs, esters and relevant unknowns.

Indeed, these results are well corroborated by the results obtained for total concentrations of the AA obtained from the analysis of all Antão Vaz wines, Table 2, and with the total values of VOC obtained from the analysis of all Antão Vaz wines, Table 3, where in both analysis, T* wines show the highest amounts followed by UM* wines.

Table 3. Average area values of VOC obtained from the analysis of all Antão Vaz wines and respective standard deviation of the mean: Frozen unripe grape must addition (UM*); addition of a mixed solution of tartaric, malic, and lactic acid (Mix*); testimonial wine (T*); LRI denotes calculated linear retention indices; LRI (Lit) denotes linear retention indices according to literature; Aroma descriptor denotes the aroma descriptors indicated by the literature. Different letters in the row mean statistically significant differences at $p < 0.05$. N/D—not detected.

No	Compound	LRI	LRI (Lit) References [43–59]	Aroma Descriptor	UM*	Mix*	T*
	Esters						
1	Ethyl acetate		(885–898)	Fruity, sweet, pineapple, red fruits [1]	$2.43 \times 10^{10} \pm 7.78 \times 10^{8\,b}$	$2.32 \times 10^{10} \pm 4.60 \times 10^{8\,b}$	$2.84 \times 10^{10} \pm 1.24 \times 10^{9\,a}$
2	Ethyl isobutyrate		(955–984)	Fruity, strawberry, sweet, bubble gum, alcoholic [1]	$4.03 \times 10^{7} \pm 2.30 \times 10^{6}$	$5.02 \times 10^{7} \pm 1.17 \times 10^{7}$	$2.99 \times 10^{7} \pm 2.23 \times 10^{7}$
3	Isobutyl acetate	1027	(1005–1007)	Solvent, alcoholic, ripe fruit [1,2]	$4.48 \times 10^{8} \pm 1.41 \times 10^{7}$	$4.50 \times 10^{8} \pm 1.17 \times 10^{7}$	$5.31 \times 10^{8} \pm 1.05 \times 10^{8}$
4	Ethyl butyrate	1049	(1022–1057)	Fruity, strawberry, sweet, bubble gum, banana [1]	$3.66 \times 10^{9} \pm 1.20 \times 10^{8}$	$3.80 \times 10^{9} \pm 8.13 \times 10^{7}$	$4.30 \times 10^{9} \pm 3.36 \times 10^{8}$
5	Ethyl 2-methylbutanoate	1063	(1041–1069)	strawberry, fruity [2]	N/D	N/D	$5.83 \times 10^{5} \pm 8.24 \times 10^{5}$

Table 3. *Cont.*

No	Compound	LRI	LRI (Lit) References [43–59]	Aroma Descriptor	UM*	Mix*	T*
6	Isoamyl acetate	1127	(1118–1147)	Banana, sweet, fruity, fresh, green [1]	$1.01 \times 10^{11} \pm 2.79 \times 10^{9\,a}$	$9.34 \times 10^{10} \pm 1.38 \times 10^{9\,b}$	$1.03 \times 10^{11} \pm 2.12 \times 10^{9\,a}$
7	Ethyl (Z)-but-2-enoate	1175	(1122–1152)		$1.26 \times 10^{7} \pm 2.35 \times 10^{6}$	$1.64 \times 10^{7} \pm 6.75 \times 10^{6}$	$1.71 \times 10^{7} \pm 3.54 \times 10^{5}$
8	Hexyl acetate	1231	1264	Pleasant fruity, pear [1]	$6.91 \times 10^{10} \pm 4.99 \times 10^{9}$	$6.75 \times 10^{10} \pm 1.84 \times 10^{9}$	$7.16 \times 10^{10} \pm 1.17 \times 10^{9}$
9	Ethyl hexanoate	1269	(1224–1270)	Green apple, fruity, strawberry, anise [3]	$1.73 \times 10^{10} \pm 6.72 \times 10^{8\,a}$	$1.40 \times 10^{10} \pm 3.89 \times 10^{8\,b}$	$1.60 \times 10^{10} \pm 2.83 \times 10^{8\,a}$
10	Ethyl 3-hexenoate	1293	1301		$1.07 \times 10^{8} \pm 1.21 \times 10^{7}$	$1.48 \times 10^{8} \pm 3.22 \times 10^{7}$	$1.05 \times 10^{8} \pm 2.57 \times 10^{7}$
11	(Z)-3-Hexenyl acetate	1309	1308	Fruity, green tea [4]	$1.35 \times 10^{9} \pm 4.95 \times 10^{7\,a}$	$9.71 \times 10^{8} \pm 6.72 \times 10^{7\,b}$	$1.08 \times 10^{9} \pm 2.58 \times 10^{7\,b}$
12	Ethyl heptanoate	1324	1331	Pineapple, fruity [5]	$1.12 \times 10^{8} \pm 9.44 \times 10^{6}$	$1.22 \times 10^{8} \pm 5.30 \times 10^{6}$	$1.27 \times 10^{8} \pm 4.84 \times 10^{7}$
13	Ethyl lactate	1331	1341	Lactic, raspberry [3]	$1.55 \times 10^{7} \pm 1.06 \times 10^{6\,b}$	$4.69 \times 10^{7} \pm 1.37 \times 10^{7\,a}$	$1.15 \times 10^{7} \pm 1.10 \times 10^{6\,b}$
14	Heptyl acetate	1361	(1374–1385)	Pear [6]	$4.22 \times 10^{8} \pm 3.18 \times 10^{6\,b}$	$3.65 \times 10^{8} \pm 8.03 \times 10^{7\,b}$	$7.64 \times 10^{8} \pm 7.11 \times 10^{7\,a}$
15	Methyl octanoate	1378	1378	Fruity, floral, creamy [1]	$2.03 \times 10^{8} \pm 2.26 \times 10^{7}$	$2.23 \times 10^{8} \pm 7.78 \times 10^{6}$	$2.40 \times 10^{8} \pm 1.45 \times 10^{7}$
16	Ethyl octanoate	1418	(1422–1446)	Soapy, fatty, anise, fruity, pineapple, pear, flora, sweet [1]	$1.93 \times 10^{11} \pm 1.7 \times 10^{10}$	$1.81 \times 10^{11} \pm 1.98 \times 10^{10}$	$2.03 \times 10^{11} \pm 1.34 \times 10^{10}$
17	Isopentyl hexanoate	1439	1444	Sweet fruity [7]	$4.28 \times 10^{8} \pm 8.10 \times 10^{7}$	$4.32 \times 10^{8} \pm 7.64 \times 10^{7}$	$4.74 \times 10^{8} \pm 3.96 \times 10^{7}$
18	Ethyl nonanoate	1510	1528	Rose, fruity [1]	$2.90 \times 10^{8} \pm 6.29 \times 10^{7}$	$2.81 \times 10^{8} \pm 3.89 \times 10^{7}$	$4.18 \times 10^{8} \pm 9.05 \times 10^{7}$
19	Butyl octanoate	1520	(1601–1621)	Orange floral, jasmine, pear [1]	$1.98 \times 10^{7} \pm 3.89 \times 10^{6}$	$2.11 \times 10^{7} \pm 3.18 \times 10^{6}$	$1.9 \times 10^{7} \pm 1.48 \times 10^{6}$
20	Propyl octanoate	1524	(1508–1530)		$5.5 \times 10^{7} \pm 1.79 \times 10^{7}$	$6.12 \times 10^{7} \pm 1.21 \times 10^{7}$	$6.71 \times 10^{7} \pm 3.15 \times 10^{7}$
21	Ethyl (E)-oct-2-enoate	1532		Fruity, pineapple, green with a fatty waxy nuance [1]	$1.04 \times 10^{8} \pm 1.20 \times 10^{7}$	$1.39 \times 10^{8} \pm 4.29 \times 10^{7}$	$1.07 \times 10^{8} \pm 3.11 \times 10^{7}$
22	Isoamyl lactate	1539	1583	Fruity creamy nutty [8]	N/D	N/D	$3.18 \times 10^{7} \pm 4.49 \times 10^{7}$
23	Methyl decanoate	1565	(1570–1636)	Fruity, soap, waxy [1]	$6.44 \times 10^{7} \pm 2.37 \times 10^{6}$	$6.53 \times 10^{7} \pm 1.56 \times 10^{7}$	$7.48 \times 10^{7} \pm 2.20 \times 10^{6}$
24	Ethyl decanoate	1603	(1595–1665)	Fruity, grape, fatty, pleasant, floral, sweet [1]	$1.08 \times 10^{11} \pm 8.2 \times 10^{9}$	$1.02 \times 10^{11} \pm 1.59 \times 10^{10}$	$1.03 \times 10^{11} \pm 1.48 \times 10^{9}$
25	Isoamyl octanoate	1622	1642	Sweet, cheese [5]	$2.54 \times 10^{9} \pm 2.86 \times 10^{8}$	$2.39 \times 10^{9} \pm 4.63 \times 10^{8}$	$2.10 \times 10^{9} \pm 2.33 \times 10^{8}$
26	Diethyl succinate	1633	1684	Light fruity [3]	$1.15 \times 10^{8} \pm 8.17 \times 10^{6}$	$1.08 \times 10^{8} \pm 1.69 \times 10^{7}$	$1.87 \times 10^{8} \pm 6.19 \times 10^{7}$
27	Ethyl dec-9-enoate (isomer)	1639	1694	Fruity, fatty [1]	$7.41 \times 10^{7} \pm 1.34 \times 10^{6\,a}$	$5.38 \times 10^{7} \pm 7.88 \times 10^{6\,b}$	$6.35 \times 10^{7} \pm 2.33 \times 10^{6\,a,b}$

Table 3. *Cont.*

No	Compound	LRI	LRI (Lit) References [43–59]	Aroma Descriptor	UM*	Mix*	T*
28	Decyl acetate	1641	(1691–1692)		$1.32 \times 10^7 \pm 2.16 \times 10^6$	$4.30 \times 10^7 \pm 1.71 \times 10^7$	$2.27 \times 10^7 \pm 7.60 \times 10^6$
29	Isobutyl decanoate	1708	1746		$3.87 \times 10^8 \pm 2.47 \times 10^7$	$3.8 \times 10^8 \pm 2.2 \times 10^7$	$3.47 \times 10^8 \pm 5.3 \times 10^7$
30	Ethyl trans-dec-2-enoate (isomer)	1720			$6.11 \times 10^7 \pm 1.53 \times 10^7$	$6.47 \times 10^7 \pm 3.67 \times 10^7$	$6.98 \times 10^7 \pm 1.2 \times 10^7$
31	Phenethyl acetate	1776	1803	Rose, jasmine, sweet, honey, floral, rosy with a slight green nectar fruity body and mouth feel [1]	$9.67 \times 10^9 \pm 9.55 \times 10^8$	$1.08 \times 10^{10} \pm 5.66 \times 10^8$	$1.18 \times 10^{10} \pm 6.36 \times 10^8$
32	Ethyl laurate	1787	1822	Sweet, floral, fruity, cream [9]	$3.90 \times 10^{10} \pm 5.94 \times 10^9$	$4.39 \times 10^{10} \pm 6.3 \times 10^9$	$3.42 \times 10^{10} \pm 1.0 \times 10^{10}$
33	Isoamyl decanoate	1806	(1840–1871)		$1.00 \times 10^9 \pm 3.61 \times 10^7$	$1.39 \times 10^9 \pm 1.20 \times 10^8$	$1.09 \times 10^9 \pm 4.91 \times 10^8$
34	Ethyl myristate	1964	(2015–2094)		$3.56 \times 10^8 \pm 3.82 \times 10^7$	$4.10 \times 10^8 \pm 4.10 \times 10^7$	$3.94 \times 10^8 \pm 3.01 \times 10^7$
35	Isoamyl laurate	1992	(2048–2110)		$1.20 \times 10^8 \pm 1.42 \times 10^7$	$1.49 \times 10^8 \pm 1.17 \times 10^7$	$1.20 \times 10^8 \pm 4.90 \times 10^7$
36	Ethyl-tetradec-9-enoate (isomer)				$3.71 \times 10^8 \pm 1.38 \times 10^7$	$4.47 \times 10^8 \pm 2.44 \times 10^7$	$2.89 \times 10^8 \pm 1.52 \times 10^8$
37	Ethyl hexadecanoate		2229	Fatty, rancid, fruity, sweet [3]	$9.46 \times 10^8 \pm 3.09 \times 10^8$	$8.39 \times 10^8 \pm 1.81 \times 10^8$	$1.04 \times 10^9 \pm 3.21 \times 10^8$
38	Ethyl hexadec-9-enoate (isomer)				$1.37 \times 10^9 \pm 1.66 \times 10^8$	$1.51 \times 10^9 \pm 1.41 \times 10^7$	$1.18 \times 10^9 \pm 3.35 \times 10^8$
	Alcohols						
39	Ethanol		926		$1.01 \times 10^{11} \pm 4.45 \times 10^9$	$1.01 \times 10^{11} \pm 1.63 \times 10^{10}$	$1.10 \times 10^{11} \pm 4.95 \times 10^9$
40	Isobutyl alcohol	1102	(1085–1125)	Fusel, alcohol, malty, fruity, sweet [1]	$1.23 \times 10^9 \pm 9.90 \times 10^7$	$1.51 \times 10^9 \pm 2.16 \times 10^8$	$1.56 \times 10^9 \pm 1.20 \times 10^8$
41	Butyl alcohol	1150	(1138–1146)	Medicinal, alcohol, spicy, refreshing, sweet [1]	$2.41 \times 10^7 \pm 5.66 \times 10^5$	$2.09 \times 10^7 \pm 9.23 \times 10^6$	$3.11 \times 10^7 \pm 2.05 \times 10^7$
42	Pentan-1-ol	1203	1244		$4.11 \times 10^{10} \pm 2.58 \times 10^9$	$4.64 \times 10^{10} \pm 5.30 \times 10^8$	$4.67 \times 10^{10} \pm 2.23 \times 10^9$
43	Isohexanol	1300	(1301–1316)	Almond, toasted [10]	$9.13 \times 10^7 \pm 1.65 \times 10^7$	$6.25 \times 10^7 \pm 3.04 \times 10^7$	$8.50 \times 10^7 \pm 7.07 \times 10^6$
44	3-Methyl-1-pentanol	1313	(1313–1325)	Herbaceous, cocoa, soil, mushroom [10,11]	$2.43 \times 10^7 \pm 2.44 \times 10^6$	$4.17 \times 10^7 \pm 1.86 \times 10^7$	$3.11 \times 10^7 \pm 1.26 \times 10^7$
45	Hexanol	1337	(1351–1392)	Green, grass, flora, cooked, burnt [1]	$1.27 \times 10^9 \pm 1.94 \times 10^8$	$1.38 \times 10^9 \pm 1.70 \times 10^8$	$1.33 \times 10^9 \pm 1.34 \times 10^8$
46	(Z)-hex-3-en-1-ol	1367	(1378–1407)	Fruity, plant, refreshing, citrus [2,12]	$1.66 \times 10^8 \pm 2.05 \times 10^7$	$1.61 \times 10^8 \pm 2.12 \times 10^6$	$1.43 \times 10^8 \pm 2.83 \times 10^6$
47	1-Heptanol	1430		Rusty, fishy, sweaty, earthy [1]	$5.31 \times 10^8 \pm 8.17 \times 10^7$	$6.32 \times 10^8 \pm 2.83 \times 10^6$	$7.92 \times 10^8 \pm 1.44 \times 10^8$
48	2-Nonen-1-ol	1572			$4.01 \times 10^7 \pm 0.00 \times 10^0$	$5.49 \times 10^7 \pm 1.56 \times 10^7$	$6.69 \times 10^7 \pm 1.12 \times 10^7$

Table 3. *Cont.*

No	Compound	LRI	LRI (Lit) References [43–59]	Aroma Descriptor	UM*	Mix*	T*
49	Nonan-1-ol	1615	(1619–1624)		$3.04 \times 10^7 \pm 1.17 \times 10^7$	$4.67 \times 10^7 \pm 1.09 \times 10^7$	$5.07 \times 10^7 \pm 2.47 \times 10^5$
50	Methionol	1677	(1738–1745)	Plastic, rubber [13]	$9.35 \times 10^7 \pm 1.16 \times 10^7$	$1.07 \times 10^8 \pm 1.82 \times 10^7$	$9.00 \times 10^7 \pm 2.55 \times 10^7$
51	Phenylethyl alcohol	1858	(1905–1940)	Flowery, pollen, perfume, rose, sweet, honey [1]	$1.38 \times 10^{10} \pm 1.94 \times 10^9$	$1.77 \times 10^{10} \pm 8.49 \times 10^8$	$1.63 \times 10^{10} \pm 1.4 \times 10^9$
52	Nerolidol	1959	(2008–2057)	Floral, fruity, orange, light flavor [3]	$3.22 \times 10^7 \pm 2.40 \times 10^7$	$2.89 \times 10^7 \pm 1.12 \times 10^7$	$3.32 \times 10^7 \pm 4.14 \times 10^6$
	Ketones						
53	2-Nonanone	1384	1397	Fruity, floral, fatty [1]	$9.66 \times 10^6 \pm 4.93 \times 10^6$	$1.70 \times 10^7 \pm 3.29 \times 10^6$	$1.34 \times 10^7 \pm 3.35 \times 10^6$
54	3-Decanone	1488	1491		$1.04 \times 10^8 \pm 1.11 \times 10^7$	$1.61 \times 10^8 \pm 2.97 \times 10^7$	$1.33 \times 10^8 \pm 3.82 \times 10^7$
55	γ-Butirolactone	1627	(1640–1673)	Toasty, wood, caramel, sour, dried floral [10,14]	$4.26 \times 10^7 \pm 3.68 \times 10^6$	$4.29 \times 10^7 \pm 1.66 \times 10^6$	$4.83 \times 10^7 \pm 1.08 \times 10^7$
	Aldehydes						
56	Nonanal	1389	(1402–1415)	Waxy, aldehydic, rose, fresh, orris, orange peel, fatty, peely [1]	$4.02 \times 10^8 \pm 3.78 \times 10^8$	$1.27 \times 10^9 \pm 1.32 \times 10^8$	$6.40 \times 10^8 \pm 7.05 \times 10^8$
57	Furfural	1465	(1458–1485)	Woody, almond, sweet, fruity, flowery, sweet wood, nut, bready, caramel, burnt [15–17]	$4.96 \times 10^8 \pm 7.32 \times 10^7$	$8.21 \times 10^8 \pm 1.77 \times 10^8$	$7.24 \times 10^8 \pm 4.24 \times 10^7$
58	2-Methyl hexadecanal	1695	1654		$4.97 \times 10^7 \pm 1.38 \times 10^7$	$4.84 \times 10^7 \pm 2.97 \times 10^6$	$4.83 \times 10^7 \pm 3.89 \times 10^6$
	Carboyilic acids						
59	2-Hydroxyoctanoic acid	1749			$1.93 \times 10^7 \pm 5.73 \times 10^6$	$2.85 \times 10^7 \pm 3.04 \times 10^6$	$1.97 \times 10^7 \pm 4.88 \times 10^6$
60	Cis-5-Dodecenoic acid	1837			$1.75 \times 10^9 \pm 5.23 \times 10^8$	$1.88 \times 10^9 \pm 3.08 \times 10^8$	$1.22 \times 10^9 \pm 4.63 \times 10^8$
61	Octanoic acid		(2083–2098)	Fatty, unpleasant, cheese, fatty acid, harsh, rancid [1,10]	$1.97 \times 10^{10} \pm 3.71 \times 10^9$	$2.00 \times 10^{10} \pm 3.18 \times 10^9$	$1.50 \times 10^{10} \pm 4.14 \times 10^9$
	Relevant unknowns						
62	Unknown 1				$4.19 \times 10^7 \pm 5.37 \times 10^6$	$5.54 \times 10^7 \pm 1.44 \times 10^7$	$4.58 \times 10^7 \pm 1.03 \times 10^7$
63	Unknown 2	1140			N/D	N/D	$2.53 \times 10^7 \pm 3.57 \times 10^7$
64	Unknown 3	1176			$4.52 \times 10^7 \pm 6.75 \times 10^6$	$4.02 \times 10^7 \pm 2.79 \times 10^6$	$4.69 \times 10^7 \pm 2.97 \times 10^6$
65	Unknown 4	1190			$9.29 \times 10^7 \pm 3.06 \times 10^7$	$7.84 \times 10^7 \pm 4.89 \times 10^7$	$4.86 \times 10^7 \pm 1.97 \times 10^7$
66	Unknown 5	1227			$1.80 \times 10^8 \pm 1.38 \times 10^7$	$1.59 \times 10^8 \pm 1.87 \times 10^7$	$1.76 \times 10^8 \pm 1.10 \times 10^7$
67	Unknown 6	1236			$3.40 \times 10^7 \pm 2.45 \times 10^7$	$2.00 \times 10^7 \pm 2.83 \times 10^7$	$3.11 \times 10^7 \pm 1.15 \times 10^7$

Table 3. *Cont.*

No	Compound	LRI	LRI (Lit) References [43–59]	Aroma Descriptor	UM*	Mix*	T*
68	Unknown 7	1244			$1.08 \times 10^7 \pm 9.49 \times 10^6$	$5.32 \times 10^6 \pm 7.52 \times 10^6$	$1.62 \times 10^7 \pm 7.07 \times 10^6$
69	Unknown 8	1260			$7.07 \times 10^7 \pm 9.26 \times 10^6$	$8.16 \times 10^7 \pm 1.30 \times 10^7$	$8.22 \times 10^7 \pm 4.36 \times 10^6$
70	Unknown 9	1273			$1.37 \times 10^7 \pm 7.35 \times 10^5$	$1.69 \times 10^7 \pm 1.61 \times 10^7$	$1.94 \times 10^7 \pm 6.93 \times 10^6$
71	Unknown 10	1287			$8.01 \times 10^7 \pm 5.23 \times 10^{6\,a,b}$	$9.11 \times 10^7 \pm 7.00 \times 10^{6\,a}$	$6.75 \times 10^7 \pm 2.40 \times 10^{6\,b}$
72	Unknown 11	1341			$1.03 \times 10^8 \pm 1.13 \times 10^{6\,b}$	$1.24 \times 10^8 \pm 2.47 \times 10^{6\,a}$	$1.05 \times 10^8 \pm 2.83 \times 10^{5\,b}$
73	Unknown 12	1470			$1.28 \times 10^8 \pm 4.24 \times 10^{6\,b}$	$1.54 \times 10^8 \pm 1.06 \times 10^{6\,a}$	$1.21 \times 10^8 \pm 6.93 \times 10^{6\,b}$
74	Unknown 13	1495			$1.04 \times 10^8 \pm 2.54 \times 10^7$	$9.35 \times 10^7 \pm 2.12 \times 10^7$	$1.03 \times 10^8 \pm 7.28 \times 10^6$
75	Unknown 14	1523			$2.23 \times 10^7 \pm 8.96 \times 10^6$	$2.51 \times 10^7 \pm 2.09 \times 10^6$	$2.50 \times 10^7 \pm 1.51 \times 10^7$
76	Unknown 15	1548			$2.54 \times 10^7 \pm 3.89 \times 10^6$	$2.97 \times 10^7 \pm 1.02 \times 10^7$	$3.15 \times 10^7 \pm 7.21 \times 10^6$
77	Unknown 16	1561			$1.52 \times 10^7 \pm 1.10 \times 10^{6\,a}$	$1.90 \times 10^7 \pm 1.10 \times 10^{6\,a}$	$9.53 \times 10^6 \pm 2.29 \times 10^{6\,b}$
78	Unknown 17	1597			$6.11 \times 10^6 \pm 1.77 \times 10^5$	$6.65 \times 10^6 \pm 3.90 \times 10^6$	$1.00 \times 10^7 \pm 8.41 \times 10^5$
79	Unknown 18	1611			$2.65 \times 10^7 \pm 1.34 \times 10^7$	$1.67 \times 10^7 \pm 9.10 \times 10^6$	$1.59 \times 10^7 \pm 2.28 \times 10^6$
80	Unknown 19	1651			$1.30 \times 10^{11} \pm 1.59 \times 10^{10}$	$1.31 \times 10^{11} \pm 1.41 \times 10^{10}$	$1.25 \times 10^{11} \pm 2.12 \times 10^9$
81	Unknown 20	1661			$2.15 \times 10^8 \pm 3.64 \times 10^7$	$2.10 \times 10^8 \pm 1.52 \times 10^7$	$2.21 \times 10^8 \pm 2.40 \times 10^7$
82	Unknown 21	1680			$3.23 \times 10^7 \pm 1.03 \times 10^6$	$2.98 \times 10^7 \pm 7.85 \times 10^6$	$3.27 \times 10^7 \pm 3.75 \times 10^6$
83	Unknown 22	1728			$6.35 \times 10^6 \pm 4.60 \times 10^5$	$6.94 \times 10^7 \pm 8.71 \times 10^7$	$6.85 \times 10^6 \pm 1.18 \times 10^6$
84	Unknown 23	1729			$4.56 \times 10^7 \pm 1.32 \times 10^{7\,a,b}$	$7.11 \times 10^7 \pm 8.13 \times 10^{6\,a}$	$2.70 \times 10^7 \pm 2.09 \times 10^{6\,b}$
85	Unknown 24	1753			$1.89 \times 10^7 \pm 4.49 \times 10^6$	$2.49 \times 10^7 \pm 6.36 \times 10^5$	$1.91 \times 10^7 \pm 2.44 \times 10^6$
86	Unknown 25	1757			$1.16 \times 10^8 \pm 2.12 \times 10^7$	$1.13 \times 10^8 \pm 3.64 \times 10^7$	$1.05 \times 10^8 \pm 3.25 \times 10^7$
87	Unknown 26	1826			$2.05 \times 10^9 \pm 5.69 \times 10^8$	$1.99 \times 10^9 \pm 2.93 \times 10^8$	$1.40 \times 10^9 \pm 4.45 \times 10^8$
88	Unknown 27	1893			$5.60 \times 10^7 \pm 2.72 \times 10^7$	$7.36 \times 10^7 \pm 7.18 \times 10^6$	$4.82 \times 10^8 \pm 5.78 \times 10^8$
		Total			$1.76 \times 10^{12} \pm 7.35 \times 10^{10}$	$1.74 \times 10^{12} \pm 8.50 \times 10^{10}$	$1.80 \times 10^{12} \pm 5.10 \times 10^{10}$

Since esters were the most abundant compound family in the studied wines, the information obtained from the PCA, and for each chemical functional group, allow us to observe that, after higher alcohols, esters are known to be wine's second most crucial component of volatile aromas in these samples. Esters are also known to strongly contribute to the floral and fruity characteristics of the final product [42]. Ethyl esters were the largest group of VOCs in all Antão Vaz wines. They have a strong influence on wine aroma. They are usually found in high concentrations and contribute to wine aroma because they

are the primary source of fruity aromas [9,10,27–30]. Most esters in alcoholic beverages are secondary metabolites produced by yeast during alcoholic fermentation. Ethyl esters content depends on different factors, such as sugar content, fermentation temperature, aeration, and yeast strain [60].

In the present study, isoamyl acetate (6) and ethyl octanoate (16) were the prominent esters found, followed by ethyl decanoate (24).

Data were also treated using LDA analysis, a supervised statistical analysis used when the groups are known in advance. First, the method measures the distance from each point to each group's centroid. Then, it classifies the point to the closest group considering the variance and covariance between the variables [9,10,28,29,61,62].

The significantly different compounds responsible for the discrimination of the wine's samples—determined by one-way analysis of variance (ANOVA)—are AA, such as Asp, Leu and Trp, and also various groups of VOCs such as seven esters: Ethyl acetate (1), isoamyl acetate (6), ethyl hexanoate (9), (*Z*)-3-hexenyl acetate (11), ethyl lactate (13), heptyl acetate (14), and ethyl dec-9-enoate (isomer) (27); and four relevant unknown compounds, such as, Unknown 10 (71), Unknown 11 (72), Unknown 12 (73) and Unknown 16 (77).

The linear discriminant analysis (LDA), obtained from the combined data of AAs and volatile compounds (Figure 2), showed a clear separation between all Antão Vaz wines. A clear separation between Mix* and the other two group wine samples were obtained along F1; separation between unripe wines (UM*) and T* wines was possible along F2.

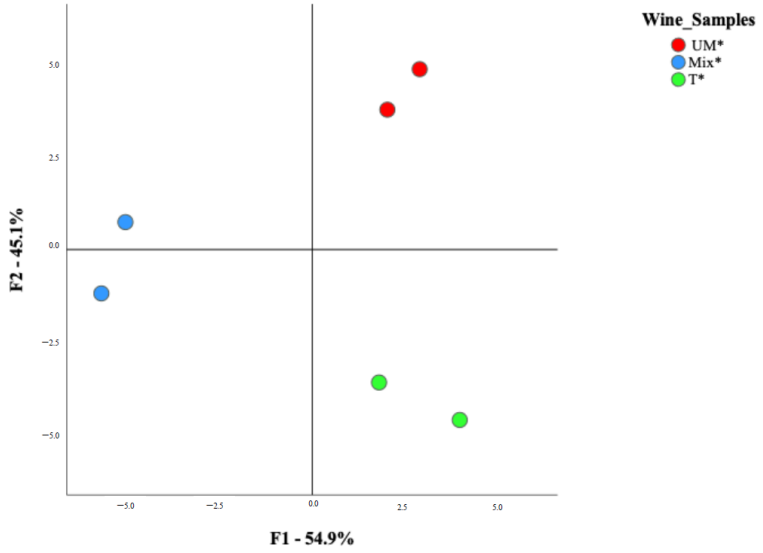

Figure 2. Score plot of the two first discriminant functions obtained after linear discriminant analysis (LDA) analysis of the different Antão Vaz wines.

The results of the flavour profile of Antão Vaz wines are shown in Figure 3. As can be observed, the spider graph represents the six major chemical group compounds present in these wines. Esters represent the major abundant group of VOCs, followed by alcohols; in contrast, ketones are the least abundant group.

The results clearly showed a different volatile profile for the Mix* wines compared to the T* and UM* wines concerning the aldehydes and ketones content. Moreover, concerning the carboxylic acid content, Mix* and UM* wines present higher contents compared to T* wines. Nonetheless, the three wines have similar amounts of the other three chemical groups. These results support the analytical results from the ANOVA test, Table 3, where no significant differences were found between total content of VOCs of Antão Vaz wines, and the PCA obtained for the separation by chemical function groups, Figure 1.

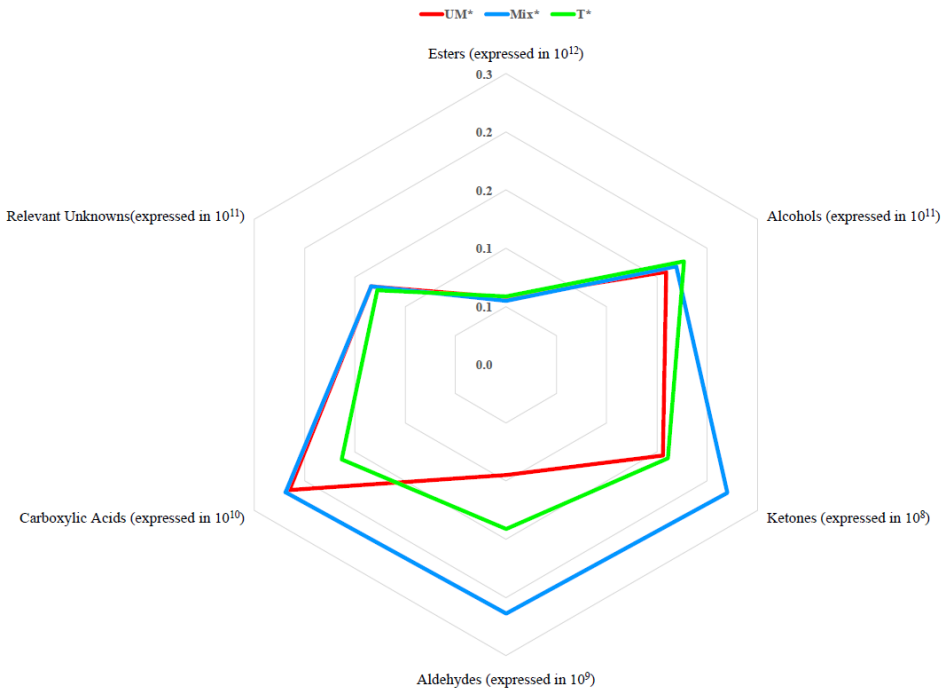

Figure 3. Spider graph of the volatile compounds for all Antão Vaz white wines (UM*, Mix*, and T*), distributed according to their flavour profile.

4. Discussion

As mentioned previously, the chemical composition of grapes is influenced by various factors such as variety, terroir, and year [1]. High temperatures during grape ripening result in low acidity and high sugar content musts, which lead to high pH, high ethanol, faded color, and no freshness in wines [62,63]. Another critical factor in the vineyard environment is grape maturity. The ripening process is highly complex, with concentrations of precursors and metabolites increasing or decreasing significantly over time. However, our understanding of how changes during ripening influence the aroma profile of wine remains limited [64,65].

Amino acids (AAs), precursors of VOCs, are another factor influencing wine aroma. The VOCs derived from yeast sugar and amino acid metabolism are higher alcohols, esters, carbonyl compounds, volatile fatty acids, and sulfur compounds that contribute widely to the wine aroma. However, grape maturity and variety have been reported as the most determinant variables in the content of amino acids that accumulate in the grape berries' tissues [28,42,66].

In our work, using unripe grape musts to correct the final acidity of the wines also allowed us to study the influence of vineyard potential and grape maturity on the flavour compound profile of the studied wines.

Higher concentrations of other volatile phenols distinguish wines from grapes with a lower degree of maturity, which is the case for the Antão Vaz wines [64]. Yet, according to the literature, these results are expected since, in wines produced from grapes with low maturity, a low proportion of linear esters relative to their acid homologs can be found. However, this proportion tends to increase as the grapes ripen [64].

In this case, LDA analysis clustered the wine samples in separate quadrants, showing that the methodology applied to correct the final acidity of the wines played an essential and fundamental role in the final wine. Additionally, looking at ANOVA results, the fact that no significant differences were found in the different studied wines allows us to conclude that unripe grape musts can be used to correct the final acidity of the wines without damaging their flavour profile. Hence, higher amounts of ethyl esters of other fatty acids, such as isoamyl acetate (6), ethyl octanoate (16), and ethyl decanoate (24), were also reported in other highly flavoured white wines [67]. These compounds are responsible for

fermented beverages' highly desired fruity, candy, and perfume-like aromas. Moreover, their lower threshold values compared to other aroma compounds can strongly impact the sensory quality of the wine [41].

5. Conclusions

The use of unripe grape musts to correct the acidity of the Antão Vaz wines is interlinked with the vineyard potential and grape maturity of the grapes used for the wines produced. These three factors together have a significant influence on the final aroma compounds profile of the wines. From the results obtained in this work, one can conclude that grape maturity is a critical factor in the final wine characteristics. Additionally, it influences the results of the acidity correction methodology applied. The results showed wines made from grapes with a lower degree of maturity reported a low proportion of linear esters relative to their precursor organic acids. This is characteristic of wines produced from grapes with low maturity, even though this proportion tends to increase as the grapes ripen. Despite results found for amino acids and volatiles of wines, and the influence of unripe must addition on these wine characteristics, this technique proved to be effective in increasing the total acidity and decreasing pH, and more importantly, it appears that may be further explored as a tool to reduce alcoholic content of wines. Therefore, using unripe grape musts as a "green" tool to increase musts acidity, as an alternative sustainable methodology for musts acidity correction in the winemaking process, is an up-and-coming alternative from the traditional methods. It can enhance the final wine characteristics and lower alcoholic content while contributing to the circular economic values.

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Conflicts of Interest: The authors declare no conflict of interest.

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