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Natural Fiano Wines Fermented in Stainless Steel Tanks, Oak Barrels, and Earthenware Amphora

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Abstract: The growing sensitivity toward sustainability is being demonstrated by an increase in sales of natural wines. Natural wines are obtained using exclusively native vines, indigenous yeasts, absence of additives, irregular temperature control during fermentation, and smaller quantities of sulfites even compared to organic wines. In this work, natural wines were obtained from Fiano grape, a historical cultivar of Irpinia (Campania, Italy). The main objective of this study was to compare the chemical and sensory characteristics of natural wines produced using different vessels (10 HL): Test A: stainless steel; Test B: earthenware amphora; Test C: mulberry wood barrel; and Test D: cherry wood barrel without the use of starter yeasts and chemical additives, including sulfites. Our results show a greater concentration of higher alcohols and esters in wines obtained in amphora and wooden barrels. The results of this work reveal that the type of container influences the composition of wine to an important extent. In addition, the Fiano wines obtained have a distinctive sensory profile also due to the ancestral winemaking process used, which did not involve the use of starter yeasts or technological and chemical adjuvants.

Keywords: natural wine; earthenware amphora; wooden barrel; volatile compounds; organoleptic evaluation

Citation: Di Renzo, M.; Letizia, F.; Di Martino, C.; Karaulli, J.; Kongoli, R.; Testa, B.; Avino, P.; Guerriero, E.; Albanese, G.; Monaco, M.; et al. Natural Fiano Wines Fermented in Stainless Steel Tanks, Oak Barrels, and Earthenware Amphora. *Processes* **2023**, *11*, 1273. https://doi.org/10.3390/pr11041273

Academic Editor: Chi-Fai Chau

Received: 29 March 2023
Revised: 17 April 2023
Accepted: 18 April 2023
Published: 19 April 2023

1. Introduction

Increasing attention to issues pertaining to personal health and environmental sustainability has contributed to a growing consumer demand for "natural" agrofood products [1], including on the wine market [2].

The demand for organic wines has increased, especially among the most aware and responsible consumers [3]. This trend suggests that producing and marketing wines with sustainability characteristics are promising strategies for quality differentiation, particularly for locally produced organic wines [4,5].

Therefore, some wine producers have adopted a growing number of initiatives aimed at increasing the sustainability of their practices and the quality of their products [6]. One interesting subset of organic wines is natural wines [7]. Despite the growing market interest in natural wine, scientific information on its composite characteristics and organoleptic properties remains scarce, and the topic requires a more in-depth analysis.

Even in the absence of regulations in this regard [8], natural wine refers to a product obtained in organic winemaking without added pesticides, chemicals, and other additives

It requires the use of organic farming, indigenous yeast, no additives, minimal intervention during fermentation, and low final sulfite concentrations [9].

Most the world's white wine ferments in stainless steel tanks. Stainless steel is completely neutral in the winemaking process and does not confer any characteristics into the wine. In the last decade, the use of different kinds of alternative vessels has become widespread in winemaking, such as wooden barrels [10,11] and earthenware jars [12–15].

Oak barrels, unlike stainless steel, can lead to variations in wine aroma and flavor due to the transfer of oxygen and phenolic and aromatic compounds from wood to wine [16,17].

The use of "kvevri", a large clay amphora for fermentation is a signature feature most popular in Georgia [18]. Earthenware that makes up these amphora gives porosity but, unlike wood, does not give any flavor to the wine. However, there are few studies on the influence that the type of container used for alcoholic fermentation has on the chemical and sensory characteristics of natural wines. Scientific contributions on natural wines are rather scarce, and little research has been conducted on the production of natural wines made from containers of different compositions. In addition, no standard production line has ever been proposed to obtain these wines.

Therefore, an innovative line of organic wine production is proposed in this study to compare the impact of this winemaking process on the chemical and sensory characteristics of natural wines produced with the Fiano cultivar, using three different containers for alcoholic fermentation: stainless steel, earthenware, and wooden vessels. Fiano grape is a typical cultivar widely cultivated in Campania, Puglia, and Sicily in southern Italy [19].

2. Materials and Methods

The winemaking trials were carried out in "Cantina Giardino" winery (Ariano Irpino, Campania, Italy) during the 2020 vintage. This company specializes in the production of natural wines obtained from historic vineyards of Irpinia, a geographical area of Campania region. Fiano grapes were manually harvested and transported into the cellar using small cases (25 kg). After destemming and crushing, the must (pH 3.5 ± 0.01, sugar content 210.2 ± 0.1 g/L, titratable acidity 8.2 ± 0.2 g/L, and L-malic acid 1.42 ± 0.14 g/L) was stored in a chestnut wood vat for the maceration process at room temperature (about 25 °C). Chemical parameters were determined according to the official methods established by the European Commission (EEC) [20]. The viable yeast cells were 4.77 log CFU/mL. The viability of the yeasts was monitored by plating as reported in Section 2.1. Sulfites were not added, and no inoculation of commercial yeast was carried out. During this phase, two daily pump-overs were carried out, and after 4 days, the must was pressed. The grape juice obtained was divided in four aliquots for fermentation tests using the following different vessels (10 HL): Test A: stainless steel; Test B: earthenware amphora; Test C: mulberry wood barrel; and Test D: cherry wood barrel. Each fermentation test was conducted in triplicate. The experimental winemaking flow chart is depicted in Figure 1.

2.1. Yeast Viability

The viability of the yeasts was monitored by plating. Briefly, 1 mL of wine sample was placed in sterile saline solution and mixed, and serial decimal dilutions were obtained. Finally, an aliquot of 100 µL of these dilutions was plated on WL nutrient agar medium (Thermo Fisher Scientific, Waltham, MA, USA) containing 0.1 g/L chloramphenicol. Plates were incubated for 72 h at 28 °C in aerobic conditions.

2.2. Wine Chemical Analyses

The pH, alcohol, total acidity, volatile acidity, and reducing sugars (g/L) were determined according to the official methods established by the European Commission (EC) [20], while L-malic acid, L-lactic acid, and catechins were determined using enzymatic kit (Boehringer Mannheim GmbH, Ingelheim, Germany) according to the manufacturer's instructions. All the determinations were performed in triplicate. Profiles of volatile compounds in wine were analyzed by GC-MS/QqQ according to a method proposed by

Guerriero et al. [21]. Before proceeding to the GC-MS/QqQ analyses, 2 ng of perdeuterated benzene in methanol was added as internal standard (I.S.) in all the samples. The instrumental analyses were performed by a triple quadrupole gas chromatograph/mass spectrometer (Trace 1310 GC/TSQ 8000 Evo, Thermo Fisher Scientific), and the chromatographic separation was performed by a DB-624 column (60 m × 0.25 mm, 1.40 µm I.D., Agilent Technologies, Milano, Italy). The injected volume was always 1 µL in PTV injector operating in splitless mode (SL time 0.6 min); the injection temperature was 200 °C for 0.05 min and at 14.5 °C s^{-1} up to 300 °C for 0.6 min. Gas carrier was He, and the temperature program started at 2.0 mL min^{-1}, and after 20 min at 1.1 mL min^{-1} rate, it went up to 3.5 mL min^{-1} for 25 min. The optimized chromatographic run was 50 °C (hold time 1 min), ramp 7 °C min^{-1}, 100 °C (0 min), ramp 10 °C min^{-1}, and 240 °C (30 min). Each analysis was performed with MS acquisition in scan mode (29–350 *m/z*) with 0.2 s scan time and emission current of 50 µA and an electron energy of 70 eV in EI+ mode. The source temperature was set up at 260 °C and the transfer line temperature at 240 °C. Data acquisition, processing, and handling were performed using XCalibur software (Thermo Fisher Scientific; Waltham, MA, USA). All chemicals used were of analytical grade (≥99%) and were purchased from Sigma-Aldrich, (Merck KGaA, Darmstadt, Germany).

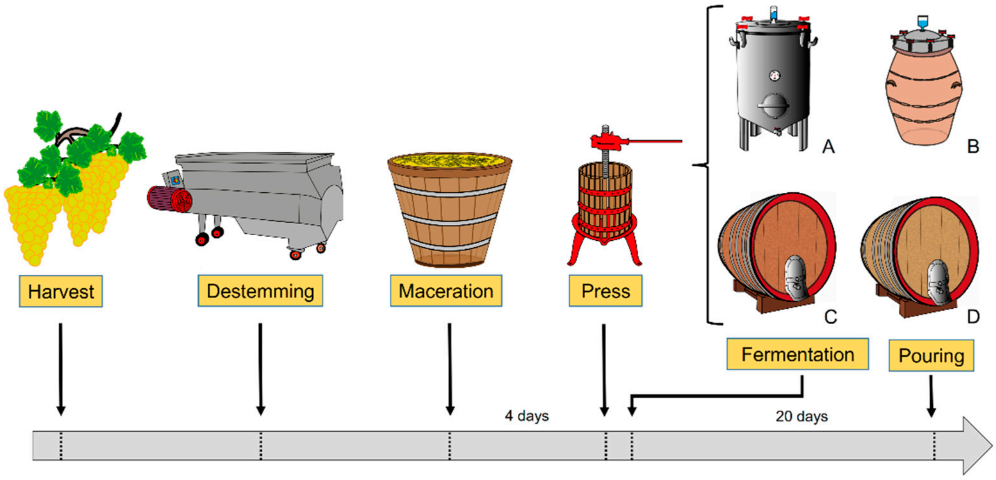


Figure 1. Flow chart of the Fiano winemaking using different fermentation vessels: (**A**) stainless steel; (**B**) earthenware amphora; (**C**) mulberry wood barrel); and (**D**) cherry wood barrel.

2.3. Wine Sensory Evaluation

Wine sensory analysis was carried out by a panel composed of twelve panelists (trained tasters, winemakers, and laboratory personnel). The panelists were trained in some preliminary sessions, using different samples of Fiano wines in order to develop a common vocabulary for the description of the sensory attributes of this wine in order to familiarize themselves with scales and procedures. Each attribute term was extensively described and explained to avoid any doubt about the relevant meaning. Therefore, the panelists were invited to evaluate the following parameters: visual evaluation (color intensity), olfactory evaluation (apple, hazelnut, hay, toasted hazelnut, vanilla, fresh wood, fresh fruit, unripe fruits, ginger, sweet cherry, or leather,), taste evaluation (sweetness, acidity, bitterness, softness, sapidity), tactile sensation (astringency), and retronasal olfactory (ro) evaluation (hay, apple, fresh wood, vegetable, wood, fruity, or red fruits). For each wine, based on the frequency of citation (>50%), several descriptors were selected to be inserted on the card thereby finally attributing an overall judgment to the wine. Tasting was organized in three different sessions, and in each session, the panelists evaluated the four wines obtained in the different fermentation tanks (A, B, C, and D). The wine samples (30 mL), served at 13 °C in clear tulip-shaped glasses, were coded with three-digit numbers and distributed in a completely randomized order (International Organization for Standardization, ISO 1997). Unsalted crackers and room temperature water were provided to rinse mouth between samples [22]. The wines were evaluated by assigning a score between 1 (absence of the sensation) and 9 (extremely intense). The resulting scores were averaged and compared.

2.4. Statistical Analysis

Experiments were performed in triplicate (n = 3), and all data are expressed as the mean ± standard deviation (SD). Statistical analysis was performed by analysis of variance (ANOVA) followed by Tukey's multiple comparisons. Statistical significance was set to p-values < 0.05. The software SPSS Statistics 21 (IBM Corp, Armonk, NY, USA) was used for the analysis.

3. Results and Discussion

3.1. Fermentation Dynamics and Main Parameters of Wines

Figure 2 shows the sugar consumption and ethanol production during alcoholic fermentation (AF) in the different trials. The numerical data are shown in Tables S1 and S2 (Supplementary Materials). In all tests, the alcoholic fermentation had a regular course albeit with greater rapidity and completeness in the steel (Test A) and stoneware (Test B) containers. For wines obtained in the wooden containers (Tests C and D), at the end of the fermentation, the residual sugars were higher than 3 g/L resulting in a lower concentration of ethanol in Test C (10.60% vol) and Test D (10.30% vol) compared to the wines obtained in Test A (11.80% vol) and Test B (12.20% vol). These differences could be caused by substances released from the wood (e.g., polyphenol, furfural, and derivatives), which may have influenced the composition of the microbial communities present in the wine and some metabolic processes. In all cases, the course of fermentation and the final alcohol values are within the expected limits based on the composition of the initial must. The analytical data of the wines obtained are shown in Table 1. There were no significant differences in pH values, while for all other parameters, there were significant differences.

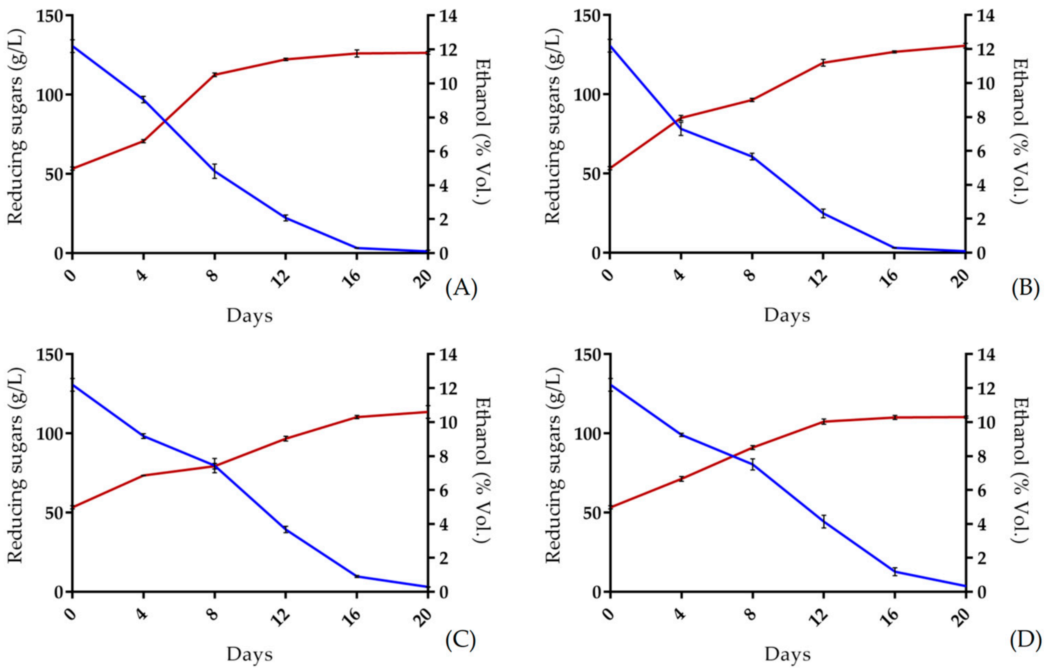


Figure 2. Reducing sugars (g/L) and ethanol (% Vol.) evolution during alcoholic fermentation in the different fermentation vessels: (**A**) stainless steel; (**B**) earthenware amphora; (**C**) mulberry wood barrel; and (**D**) cherry wood barrel. The bars indicate a significant difference (p < 0.05).

Table 1. Chemical parameters of the Fiano wines obtained using the following vessels: Test A (stainless steel tank); Test B (earthenware amphora); Test C (mulberry wood barrel); and Test D (cherry wood barrel).

Parameters	Test A	Test B	Test C	Test D
pH	3.41 ± 0.05 [a]	3.50 ± 0.06 [a]	3.52 ± 0.06 [a]	3.51 ± 0.05 [a]
Reducing sugars (g/L)	1.01 ± 0.08 [c]	0.91 ± 0.07 [c]	3.03 ± 0.11 [b]	3.50 ± 0.10 [a]
Titratable acidity * (g/L)	6.60 ± 0.03 [b]	6.69 ± 0.08 [b]	7.09 ± 0.07 [a]	7.13 ± 0.05 [a]
Alcohol (% *v*/*v*)	11.80 ± 0.08 [a]	12.20 ± 0.14 [a]	10.60 ± 0.37 [b]	10.30 ± 0.06 [b]
Volatile acidity ** (g/L)	0.51 ± 0.02 [b]	0.51 ± 0.04 [b]	0.79 ± 0.09 [a]	0.72 ± 0.09 [a]
Catechins (mg/L)	49.90 ± 0.44 [d]	77.10 ± 0.65 [a]	69.9 ± 0.83 [c]	73.20 ± 0.64 [b]
Malic acid (g/L)	nd	nd	nd	nd
Lactic acid (g/L)	1.23 ± 0.04 [a]	1.27 ± 0.11 [a]	1.17 ± 0.07 [a]	1.24 ± 0.06 [a]

Data are expressed as mean values ± standard deviations (n = 3); different letters (a–d), within a row indicate significant differences ($p < 0.05$); * as tartaric acid; ** as acetic acid.

The quantities of volatile acidity were higher in wines obtained in wooden containers (Test C and Test D); this is probably due to the greater oxygenation allowed by the porosity of wood and clay. The amount of catechins was higher in the Tests B, C, and D (earthenware amphora, mulberry wood barrel, and cherry wood barrel) compared to the levels found in Test A (stainless steel tank); these differences were attributable to the transfer of these compounds by wood and clay to the wine. Malolactic fermentation took place in all tests with final lactic acid amounts between 1.17 g/L (Test C) and 1.27 g/L (Test B).

3.2. Viability of Yeasts after Maceration and during Alcoholic Fermentation

At the end of the maceration process, which lasted 4 days, the viable cell counts were 7.11 log/CFU. Subsequently, in all the containers used for alcoholic fermentation, the vitality of the yeasts increased in the first 4 days and then decreased progressively. After 20 days (end of AF), the viability of the yeasts remained at values between 6.38 log/CFU (Test C) and 6.96 log CFU/mL (Test A). The numerical data of yeast cell counts during the winemaking process are reported in detail in Table S3 (Supplementary Materials). In all tests, the viable yeast counts were within the values normally required to carry out a correct and complete fermentation process of the grape must.

3.3. Wine Volatile Compounds and Sensory Profiles

The perceived flavor of wine is the result of complex interactions between all the volatile and nonvolatile compounds present in wine. These compounds are produced or metabolized by yeasts and bacteria during alcoholic and malolactic fermentations or are formed as a result of chemical reactions that occur during wine processing and aging [23–28]. The analytical method proposed allowed the correct identification and quantification of 38 compounds in the volatile fraction of Fiano wines. However, their concentrations in wines vary according to the fermentation tank, as shown in Table 2. Our results show that higher alcohols and esters were quantitatively dominant in the Fiano wines analyzed [21,24,27,28]. These volatile compounds are important in defining the sensory properties of wines [29,30].

Table 2. Volatile organic compounds detected in the Fiano wines obtained using different fermentation vessels: Test (A) stainless steel; Test (B) earthenware amphora; Test (C) mulberry wood barrel; and Test D) cherry wood barrel. Different letters within rows indicate significant differences ($p < 0.05$).

Compound	Fiano White Wine (μg mL^{-1}) mg/L				
	Retention Time (min)	Test A	Test B	Test C	Test D
Ethyl acetate	9.68	1.953 ± 0.011 [d]	5.820 ± 0.055 [a]	4.323 ± 0.04 [b]	4.106 ± 0.122 [c]

Table 2. *Cont.*

Compound	Fiano White Wine (μg mL^{-1}) mg/L				
	Retention Time (min)	Test A	Test B	Test C	Test D
Isobutanol	10.58	3.763 ± 0.005 [d]	4.310 ± 0.034 [c]	5.252 ± 0.055 [b]	8.373 ± 0.132 [a]
Acetic acid	10.74	0 ± 0 [b]	0 ± 0 [b]	0 ± 0 [b]	3.216 ± 0.097 [a]
Ammonium acetate	10.94	2.426 ± 0.005 [d]	3.380 ± 0.030 [c]	8.946 ± 0.110 [b]	11.380 ± 0.568 [a]
Diglycerol	11.17	0 ± 0 [b]	0 ± 0 [b]	0 ± 0 [b]	45.320 ± 1.368 [a]
1-Hydroxypropan-2-one	12.54	0.273 ± 0.020 [d]	0.583 ± 0.025 [c]	1.310 ± 0.040 [b]	2.056 ± 0.138 [a]
Isoamyl alcohol	13.24	20.20 ± 0.952 [d]	39.186 ± 0.721 [a]	29.083 ± 0.886 [b]	25.423 ± 0.979 [c]
Pentanol	13.29	5.413 ± 0.040 [c]	7.900 ± 0.089 [a]	7.876 ± 0.140 [a]	7.203 ± 0.489 [b]
1-Heptene-4-ol	14.17	0.120 ± 0.020 [b]	0.081 ± 0.004 [c]	0.186 ± 0.030 [b]	0.343 ± 0.050 [a]
Dioxirane	14.40	0 ± 0 [c]	0 ± 0 [c]	0.162 ± 0.010 [b]	0.260 ± 0.060 [a]
Propylene glycol	14.54	1.750 ± 0.040 [d]	3.883 ± 0.065 [a]	1.426 ± 0.065 [c]	2.073 ± 0.141 [b]
Ethyl lactate	15.14	1.200 ± 0.020 [d]	1.356 ± 0.020 [c]	1.676 ± 0.061 [a]	1.516 ± 0.095 [b]
2,3-Butanediol	15.32	7.206 ± 0.035 [c]	11.136 ± 0.060 [a]	5.836 ± 0.070 [d]	8.636 ± 0.223 [b]
1,3-Butanediol	15.50	4.383 ± 0.015 [d]	6.960 ± 0.045 [a]	4.726 ± 0.070 [c]	5.640 ± 0.176 [b]
Furan-2-carbaldehyde (or Furfural)	16.07	0.466 ± 0.005 [c]	0.773 ± 0.030 [b]	3.206 ± 0.095 [a]	3.356 ± 0.136 [a]
Hexanol	16.22	0.203 ± 0.023 [b]	0.306 ± 0.026 [a]	0.350 ± 0.055 [a]	0.243 ± 0.060 [a]
2-Furanmethanol	16.53	0.101 ± 0.008 [d]	0.360 ± 0.026 [c]	1.863 ± 0.090 [b]	2.130 ± 0.130 [a]
Lactic acid	16.96	0.053 ± 0.005 [d]	0.130 ± 0.020 [c]	0.313 ± 0.023 [a]	0.210 ± 0.043 [b]
Pyruvic acid	17.1	0.066 ± 0.005 [b]	0.120 ± 0.026 [b]	0.113 ± 0.020 [b]	0.626 ± 0.080 [a]
1-Methoxybutan-2-ol	17.39	0.253 ± 0.025 [a]	0.200 ± 0.020 [b]	0.080 ± 0.006 [c]	0.116 ± 0.015 [c]
1,3-Dioxane-2-methyl-4-methyl	17.58	0 ± 0 [c]	0.150 ± 0.010 [b]	0.216 ± 0.037 [b]	0.413 ± 0.045 [a]
4-Acethylpyrazole	17.67	0.078 ± 0.007 [d]	0.206 ± 0.023 [c]	0.376 ± 0.020 [b]	0.450 ± 0.040 [a]
2,4-Dihydroxy-2,5-dimethyl-3(2H)-Furanone	18.53	0.156 ± 0.005 [c]	0.043 ± 0.011 [c]	0.663 ± 0.020 [b]	1.730 ± 0.090 [a]
Furfural-5-metil	18.84	0.766 ± 0.020 [d]	1.343 ± 0.015 [c]	2.233 ± 0.049 [a]	2.003 ± 0.125 [b]
4-Oxopentanedioic acid	18.91	0.260 ± 0.010 [d]	0.670 ± 0.020 [c]	1.333 ± 0.085 [b]	1.846 ± 0.056 [a]
Dihydroxyacetone (or 1,3-Dihydroxypropan-2-one)	19.04	0 ± 0 [d]	0.236 ± 0.011 [c]	1.036 ± 0.060 [b]	2.363 ± 0.055 [a]
Pyran-2,6(3H)-dione	19.89	0.556 ± 0.030 [b]	0.840 ± 0.134 [a]	0.626 ± 0.035 [b]	0.670 ± 0.050 [a]
2-Acetylfuran (or 2-Furyl methyl ketone)	21.40	0.070 ± 0.005 [d]	0.746 ± 0.015 [c]	0.863 ± 0.025 [b]	1.040 ± 0.065 [a]
Phenethyl alcohol	21.58	15.633 ± 0.164 [a]	12.423 ± 0.411 [c]	14.046 ± 0.311 [b]	9.216 ± 0.134 [d]
Diethyl butanedioate (or Diethyl succinate)	21.73	0.573 ± 0.011 [b]	0.530 ± 0.020 [b]	1.083 ± 0.080 [a]	0.616 ± 0.045 [b]
Glycerin acetate (or 1-acetylglycerol)	21.86	0.320 ± 0.014 [c]	0.710 ± 0.020 [a]	0.696 ± 0.049 [a]	0.550 ± 0.036 [b]
Pyrarone	22.15	1.063 ± 0.050 [c]	4.391 ± 0.010 [b]	6.866 ± 0.050 [a]	7.116 ± 0.251 [a]
Ethyl succinate	22.18	12.723 ± 0.302 [b]	14.606 ± 0.166 [a]	10.680 ± 0.045 [c]	12.410 ± 0.530 [b]
5-Hydroxymaltol	22.43	0.010 ± 0.001 [c]	0.196 ± 0.015 [b]	0.473 ± 0.035 [a]	0.576 ± 0.070 [a]
Succinic acid (or Butanedioic acid)	22.85	7.756 ± 0.035 [a]	5.633 ± 0.055 [b]	0.836 ± 0.025 [d]	3.616 ± 0.080 [c]
2,3-Dihydrobenzofuran	23.18	0.540 ± 0.029 [d]	1.971 ± 0.030 [a]	1.383 ± 0.030 [c]	1.653 ± 0.045 [b]
Hydroxymethylfurfural	23.56	0.090 ± 0.002 [c]	0.501 ± 0.028 [c]	5.396 ± 0.164 [b]	10.250 ± 0.305 [a]
Tyrosol (or 4-(2-Hydroxyethyl) phenol)	27.07	5.906 ± 0.281 [b]	8.833 ± 0.080 [a]	5.510 ± 0.105 [b]	5.270 ± 0.060 [c]

Higher alcohols are released during the fermentation process by yeasts; they are produced via two pivotal mechanisms: via an anabolic pathway from glucose or via the Ehrlich pathway from their corresponding amino acids present in grapes particularly under nitrogen-limiting conditions [30,31]. In our work, nine higher alcohols were identified and quantified in Fiano wines: iso-amyl alcohol, phenethyl alcohol, pentanol, isobutanol, 2,3-butanediol, 1,3-butanediol, 1-methoxybutan-2-ol, hexanol, and 2-furanmethanol. Isoamyl alcohol is the major higher alcohol found in Fiano wines in the range of 20.76 (Test A) to 39.24 mg/L (Test B). The only alcohol detected in a concentration above the threshold value (Odor Threshold 10–14 mg/L) is phenethyl alcohol (RT: 21.58), which gives the wine floral, rose, honey, and peach notes [32]. Higher alcohols, due to the concentrations that are found in wines and their high taste thresholds, often do not have direct sensory effects individually in the wine but participate overall in the definition of the fruity aroma. Moreover, the higher alcohols are precursors of the most active esters with a taste that helps to give the wine fruity and floral notes [33]. Our data show higher alcohol production in the wooden and stoneware containers than in the stainless steel tanks and confirmed that the final concentrations of most higher alcohols depend on the oxygenation conditions [34,35]. In steel containers, no aeriform exchanges with the outside or cession of substances that can interfere with the metabolic processes of yeasts take place because it is a completely inert material.

Esters contribute to the sensory features of wines, being responsible for their floral and fruity notes. Their concentrations are dependent on various factors, including yeast species, temperature, and aeration degree during alcoholic fermentation (AF) [23,36].

Our results show the presence in the analyzed Fiano wines of ethyl succinate, ethyl lactate, glycerin acetate, and ethyl acetate. These compounds at low levels give a fruity character to the wine and positively contribute to the aroma of the wines [37–39].

In the tests carried out in wood (Tests C and D), the wines had a greater presence of some components whose origin is attributable to the heating to which the wood is subjected during processing: furfural (RT: 16.07), 2-furanmethanol (RT: 16.53), 5-metil furfurale (RT: 18.84), pyranone (RT: 22.15), and hydroxymethylfurfural (RT:23.56). In the panelists' assessment, the presence of these compounds, at the concentrations detected, did not compromise the positive evaluation of the wines. Overall, our results show a greater concentration of higher alcohols and esters in wines obtained in amphora and wooden barrels, confirming what has been reported in other research [35,40].

The porosity of earthenware and wood favors the micro-oxygenation of the fermenting wines, and, at same time, they allow some yeasts to remain immobilized in the pores, thus producing an increase in the production of alcohols and esters [34,41]. Graphical representations of the sensory profiles of Fiano wine samples are reported in Figure 3 (values indicate the means for each attribute). The descriptors examined for each wine and the relative numerical data of the judgments assigned are shown in Table S4 (Supplementary Materials). The opinions were substantially different among the various wines. All wines did not show off-odors and off-flavors.

Generally, considering Test A (stainless steel) as a reference, the wines obtained in amphora and in wooden barrels were more astringent.

In Test B (earthenware amphora), there was a more intense color and good sapidity and freshness; in the olfactory examination, the panelists found notes of toasted hazelnut and vanilla, slight vegetal nuances, and a persistent sensation of fresh wood. In the Fiano wines obtained in Test C (mulberry wood barrel), persistent notes of fresh and unripe fruit were found. Finally, in the wines obtained in cherry wood (Test D), the panelists found intense and persistent notes of red fruits, apple, and ginger a high softness and flavor that helped to give this wine the best overall rating.

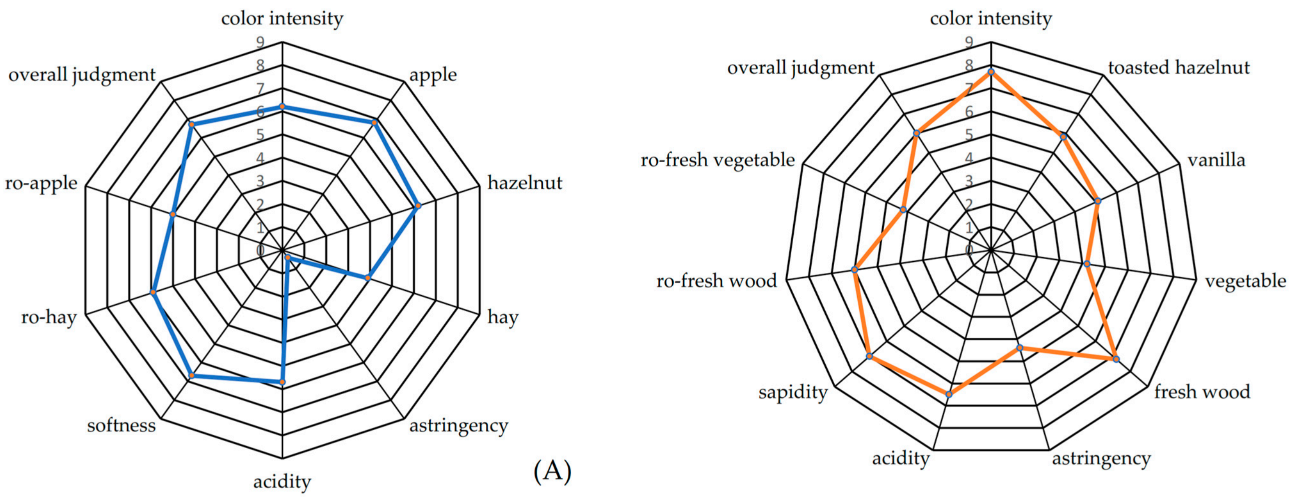
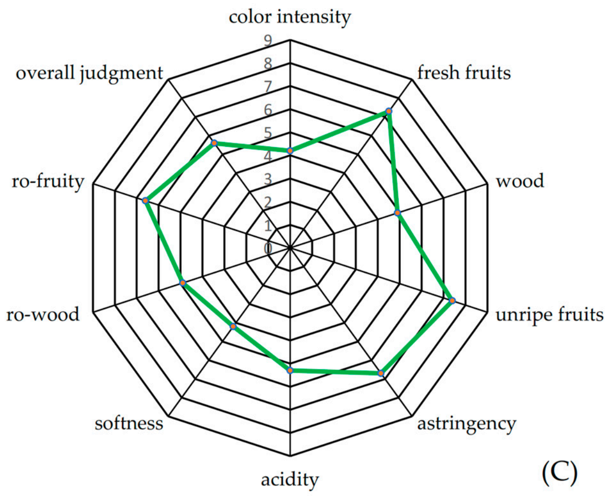


Figure 3. *Cont.*



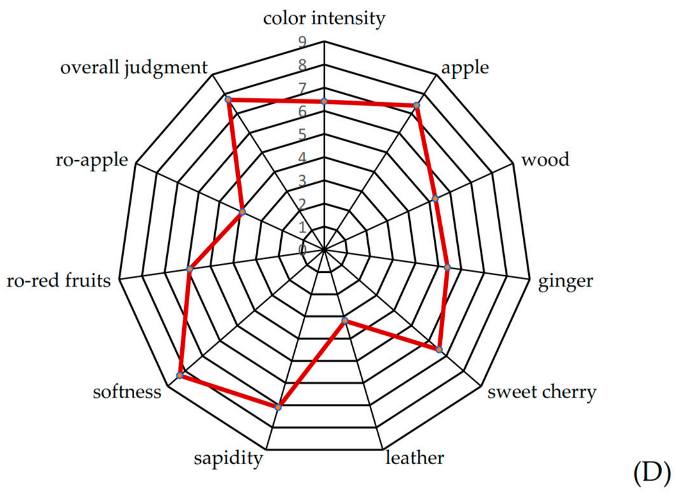


Figure 3. Spider plot of sensory profiles of the Fiano wines obtained using different fermentation vessels: (**A**) stainless steel; (**B**) earthenware amphora; (**C**) mulberry wood barrel; and (**D**) cherry wood barrel.

4. Conclusions

Natural wine is made from grapes produced under organic or biodynamic management without using additives in the cellar.

The results of this work reveal that the type of vessel used for the fermentation of Fiano wine influences the composition of the wine to an important extent. It is very likely that this may be because the wood of the barrels and the earthenware of the amphora are porous materials and interact with the must by releasing some substances during fermentation, unlike what happens with stainless steel tanks, which is a completely stable and inert material. The proposed innovative production line involving 4-day maceration seems to favor the predominance of fermentative yeasts.

Further studies will be conducted on the knowledge of yeast communities using dependent and independent cultivation methods (e.g., Next-Generation Sequencing—NGS) to understand their correlation with the chemical and sensory characteristics of organic wines obtained using containers of different composition. In our study, Fiano wines have a unique sensorial profile, due to the ancestral technique of vinification that originates distinctive wines, full of character and identity. The technologies used can be adopted to obtain good quality wines to be offered to consumers who are increasingly sensitive to chemical-free organic products. Therefore, this work is a preliminary step to further scientific studies on the quality of natural wines and on their possible certification.

In the bigger picture, the promotion and growth of natural wines would be positive in terms of environmental sustainability.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/pr11041273/s1, Table S1: Reducing sugar (g/L) evolution during alcoholic fermentation in the different vessels. Table S2: Ethanol production (% Vol.) during alcoholic fermentation in the different vessels; Table S3: Viable yeast cell counts during alcoholic fermentation in the different vessels; Table S4: Sensory perceptions detected in Fiano wines obtained using different vessels.

Author Contributions: Conceptualization, M.I. and M.D.R.; methodology, M.D.R., B.T. and M.I.; software, F.L. and G.A.; validation, G.A. and F.L.; formal analysis, B.T., E.G., P.A. and M.M.; investigation, C.D.M.; data curation, M.D.R., M.I. and F.L.; writing—original draft preparation, M.D.R. and M.I.; writing—review and editing, M.I.; visualization, M.M., J.K. and R.K.; supervision, M.I. All authors have read and agreed to the published version of the manuscript.

Funding: This research received no external funding.

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.

Data Availability Statement: Not applicable.

Acknowledgments: The authors thank Antonio Di Gruttola for his collaboration in conducting the experimental phases at the "Cantina Giardino" winery Via Petrara 21/B, Ariano Irpino, Campania, Italy.

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

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