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Defining the Typical Aroma of Sherry Vinegar: Sensory and Chemical Approach

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The aroma of the three different classes of Sherry vinegar was evaluated by gas chromatography/mass spectrometry (GC-MS) and gas chromatography/olfactometry (GC-O). GC-O was employed to identify substances responsible for aromatic notes associated with the selected descriptors of the typical aroma of Sherry vinegar and odor activity values (OAV) calculated to measure the single impact effect of different compounds selected by GC-O. Diacetyl, isoamyl acetate, ethyl isobutyrate, isovaleric acid, sotolon, and ethyl acetate reached high OAVs, turning out to be characteristic odor active compounds in Sherry vinegars. A total of 58 compounds were quantified, among them, 7 had not been previously reported in Sherry wine vinegars: ethyl 2-methylbutyrate, ethyl heptanoate, ethyl furoate, and ethyl benzoate, acetophenone, nonanoic acid, and sotolon. Linear discriminant analysis (LDA) reveals that using aroma compounds as variables, we can classify Sherry vinegars with 100% correct scores as different from red wine vinegars.

KEYWORDS: Aroma; Sherry vinegar; sensory analysis; GC-olfactometry; OAV; sotolon

INTRODUCTION

The quality of food products is a multivariate notion in which sensory properties play a crucial role. There is a need for the characterization of the typical sensory properties of traditional products (*1*) not only for the industrialization of food production, but also for laws on food safety and even for the development of innovative products.

Sherry vinegar can be considered a traditional food product used as seasoning and as a condiment. Wine vinegar is a grape-derived product obtained by a double-fermentative process (alcoholic and acetic). From a technological point of view, there are two well defined methods for its production: traditional processes and submerged methods (*2*). The first one is the so-called surface culture fermentation, where the acetic acid bacteria is placed on the air−liquid interface in direct contact with atmospheric air. Thus, oxygen availability to the acetic acid bacteria is not boundless, and a long period of time is required to obtain a high acetic degree. This process usually takes place in wood barrels. As a consequence, chemical modifications related with aging occur at the same time, and a highly appreciated product is obtained. Nowadays, traditional and selected vinegars (Sherry vinegars and traditional balsamic vinegars from Modena, among others) are produced following this method. The sensory complexity of Sherry vinegars is the consequence of chemical composition of the product, and the extraordinary organoleptic properties are acquired thanks to the method of production followed, the so-called "criaderas y solera" system (*3*). This particular method of production consists of a dynamic aging system in contrast with the not so usual static method, in which vinegar is produced and aged in a single butt (*4, 5*).

Therefore, Sherry vinegar regulation also allows the production by submerged culture acetification followed by aging in wood (dynamic or static system). Three qualities for Sherry vinegar are considered according to aging time in oak barrels: "Vinagre de Jerez" (minimum of 6 months), "Reserva" (at least 2 years), and finally, the new category "Gran Reserva" (at least 10 years) (*4*).

The aroma is one of the most important indicators of vinegar quality. For this reason, manufacturers choose the best raw materials as well as the optimum acetification conditions to increase the aromatic quality of wine vinegar and to present new products to the consumers (*2*). Although most of the volatile constituents are already present in wine, the final content is closely related to the genuine characteristics of the vinegar itself (*6*). The flavor of wine vinegars is determined by a series of volatile constituents with three different origins: wine substrate, acetification, and aging. During acetification, volatile compounds from wine may suffer important transformations. The acetic acid bacteria can metabolize high alcohols, in a way similar to that of ethanol, producing an increase in acid concentration. Moreover, ethylic esters are hydrolyzed, and at the same time, acetic esters such as isoamyl and methyl acetates are formed. Acetoin

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Table 1. List of Vinegar Samples

vinegar type	origin	time of aging	samples	acetic degree
red wine vinegar	Winery (Banyuls, France)	0 months	VT1	6
		12 months	VT2	6
	Winery (Priorat, Spain)	0 months	VT3	6
		12 months	VT4	6
balsamic vinegar	Winery (Reggio Emilia, Spain)	0 months	VB1	6
		12 months	VB2	6
Sherry vinegar	Commercial samples (Spain)	6 months ("Vinagre Jerez")	VJ1	7
			VJ2	7
			VJ3	7
		24 months ("Reserva")	VR1	7
			VR2	8
			VR3	8
			VR4	8
			VR5	9
		120 months ("Gran Reserva")	VGR1	7
			VGR2	10
			VGR3	10

also increases during acetification, being higher in traditional vinegars (*3, 7*).

During the aging of vinegar in wood barrels, there are several phenomena taking place as follows: (i) Loss of water through the pores of the casks and, consequently, a concentration of the rest of compounds; (ii) extraction of some compounds from wood (wood-extractables), mainly aromatic aldehydes; (iii) condensation (esterification); and (iv) oxidation (acetoin, diacetyl formation). Ultimately, these processes are responsible for the increase in the aromatic complexity of the vinegar.

Although the aroma composition of Sherry vinegars has been studied by several authors (*6, 8, 9*) as well as changes along aging in wood (*5, 10*), the contribution of individual compounds to the characteristic aroma of Sherry vinegar has not been considered up to now. Hence, the aim of the present work is to describe the aroma profile of the different categories of Sherry vinegar correlating the sensory results with the chemical data by measuring the single impact effect determined by the OAV (odor activity value) of different compounds selected by gas chromatography-olfactometry (GC-O). The use of GC-O allows us to screen and identify substances responsible for aromatic notes associated with the selected descriptors of the typical aroma of Sherry vinegar.

MATERIALS AND METHODS

Samples. We used for this study a total of 17 vinegars. The samples were divided into three categories (red wine, balsamic, and Sherry vinegars) according to the raw material and origin (**Table 1**). Four red and 2 balsamic vinegars were obtained from different wineries. All samples were elaborated by the traditional method (surface culture) in oak wood barrels. Eleven representative Sherry vinegars were purchased in the market (commercial samples), and they belong to the three categories established by Sherry vinegar regulation in accordance with aging time in oak barrels: 3 "Vinagre Jerez", (6 months old), 5 "Reserva" vinegars (2 years old), and 3 "Gran Reserva" vinegars (at least 10 years old).

Reagents and Chemicals. The standards of 58 aroma compounds, given in **Table 3**, were obtained from the commercial sources as follows: 2, 3, 14, 15, 19−21, 23−27, 29−32, 40−42, 45−51, and 53−58 (Sigma-Aldrich, Madrid, Spain); 1, 4, 6−10, 13, 17, 18, 28, 34−39, 44, and 52 (Merck, Darmstadt, Germany); 5, 11, 12, 16, 22, 33, and 43 (Fluka, Madrid, Spain). 3,4-Dimethylphenol (Sigma-Aldrich) and 4-methyl-2-pentanol (Merck) were employed as internal standards (IS). Dichloromethane, hexane, ether, anhydrous sodium sulfate, sodium chloride, and acetic acid were obtained from Merck, and all of them were of analytical quality. Water was obtained from a Milli-Q purification system (Millipore, USA).

Table 2. Similarity of the Odors of the Four Extracts to Vinegar Reference (VR1): Scaling and Rank[a]

extracts	similarity values (SV) and SD	rank first	rank second	rank third
dichloromethane	7.80a ± 0.91	5	0	0
hexane	5.3b ± 1.82	0	4	1
ether	3.4bc ± 2.36	0	1	4

[a] Five panelists, discontinuous scale (0−10); SV with the same letter were not significantly different at a level of 5%.

Sensory Analysis. *Sensory Panel.* The expert sensory panel that carried out the different experiments described in this work was composed of seven tasters (five females and two males), all of them belonging to a laboratory and with a lot of experience in wine vinegar sensory analysis (*11*).

Training was performed according to international protocols (ISO 4120:1983 and ISO 6658: 1985) (*12, 13*).

Descriptors Selection. Following methodology for descriptive analysis in wine vinegars (*11, 14, 15*), 10 attributes were chosen by consensus to describe wine vinegar samples as follows: ethyl acetate, pungent sensation, wine character, woody odor, red fruit, sweet aroma, bitter almond, vanilla, raisin, alcohol/liquor, and general impression. This last descriptor can be considered as a hedonic attribute since the sensory panel cannot be trained in it. The selected attributes were compiled in a tasting-card, and panelists were asked to rank each descriptor on a 10-cm unstructured scale (from not noticeable to very strong).

Threshold Determination. There are several published methodologies to calculate thresholds for flavor volatiles (*16, 17*), and we decided to use the method approved by the American Society for Testing and Materials (ASTM) (*18, 19*).

First, an ascending order test was carried out to delimit the proper concentration range to study and familiarize panelists with the odor of the compounds. Five 3-fold dilutions (3*x*, *x*, *x*/3, *x*/9, and *x*/27) were prepared by dispersing the substance whose threshold was to be determined in the medium of interest (acetic acid 7% w/v). Panelists were asked to indicate in which solution they perceived any odor. We fixed the *x* value (concentration of aroma compound) as a concentration 5-fold higher than the correspondent threshold values referenced in literature for wine (*20–22*) due to the marked interference of acetic acid.

Second, according to Plotto et al. (*23*), the three-alternative forced choice (3-AFC) test was used for threshold determination (*19*) (ASTM Designation: E-679, 2004). Four 3-ACFs a day were performed. Thus, three samples were given to panelists: two controls (7% acetic acid solution) and one test dilution (standard in 7% acetic acid solution). The test dilutions differed from the preceding one by a factor of 2 (2*x*, *x*, *x*/2, *x*/4...), and successive dilutions were tested until the lowest was consistently missed. The amount of aroma compound 2*x* corresponds to the minimum concentration of the substance that was perceived by at least 80% of the panel in the ascending order test. In this last case, we employed a factor of 2 since the threshold value was close to the concentration tested.

Then, the best-estimate criterion (*19, 23*) was used to calculate individual thresholds as follows: the threshold for each individual (best-estimate threshold) was an interpolated value determined as the geometric mean between the last concentration missed and the first concentration detected. Finally, the panel threshold was calculated as the geometric mean of the best-estimate thresholds of every individual panelist for each compound.

Selection of a Representative Extract for GC-O. A representative 2 year-old Sherry vinegar (VR1) was extracted with different organic solvents: hexane, ether, and dichloromethane. For each solvent, 50 mL of vinegar was extracted twice with 5 mL. Similarity tests were performed between the aroma of the obtained extracts and the vinegar

Table 3. Range of Volatile Compound Concentrations in Different Groups of Vinegar Samples

No	compound	mean concentration (μg/L) VJ ($n = 3$)	VR ($n = 5$)	GR ($n = 3$)	VT ($n = 4$)	VB ($n = 2$)	previously reported in Sherry wine vinegar
				Aldehydes			
1	acetaldehyde[c,a]	7.9 − 23.3	14.2 − 98.4	18.7 − 51.2	5.2 − 70.3	24.8 − 56.3	*5, 8*
2	hexanal[d]	n.d. − 9.41	n.d. − 10.7	17.0 − 35.6	12.5 − 46.7	10.7 − 60.6	*9*
3	2-furfuraldehyde	329 − 1358	336 − 1701	1189 − 7841	0.0 − 598	1056 − 3703	*9, 10, 59*
4	benzaldehyde	0.0 − 99.4	58.2 − 160	148 − 1561	0.0 − 89.0	0.0 − 115	*9, 46, 59, 60*
5	5-methyl-2-furfuraldehyde[d]	59 − 248	n.d. − 133	133 − 458	n.d.	729 − 2282	*9, 10, 59*
6	vanillin	n.d. − 1271	n.d. − 8875	2572 − 3926	n.q. − 3587	1494 − 4368	*4, 10*
	total aldehydes[c]	10.8 − 23.8	24.9 − 99.4	22.8 − 65.0	5.8 − 73.2	41.0 −64.0	
				Acetal			
7	acetaldehyde diethylacetal[c,d]	2.0 − 9.6	n.d. − 61.7	3.5 − 115	47.8 − 193	194 − 223	*46*
				Acetic Esters			
8	methyl acetate[c]	12.0 − 19.1	8.7 − 23.9	19.2 − 44.5	10.1 − 40.1	10.2 − 16.6	*5, 46*
9	ethyl acetate[c,a,d]	289 − 712	140 − 2210	351 − 452	132 − 3955	1929 − 3751	*7, 46, 59*
10	propyl acetate[d]	193 − 727	61 − 2354	886 − 3665	385 − 2207	3923 − 4605	*8, 9, 46*
11	isobutyl acetate	662 − 1458	290 − 2513	1241 − 4719	967 − 2284	2394 − 3083	*9, 46*
12	butyl acetate	n.d.	n.d. − 119	21.1 − 350	0.0 − 83.8	77 − 108	*9, 46*
13	isoamyl acetate[c]	1.02 − 3.72	0.36 − 5.59	2.03 − 11.6	2.47 − 7.26	3.97 − 5.30	*7, 9, 46, 60*
14	hexyl acetate	n.d.	n.d.	n.d.	n.d. −82	n.d.	*9, 46*
15	benzyl acetate	n.d.	n.d.	n.d. −190	n.d. − 142	n.d.	*9, 46, 60*
16	2-phenylethyl acetate	309 − 984	527 − 1491	1343 − 2090	765 − 2051	1035 − 2241	*9, 46, 60*
	total acetic esters[c]	304 − 737	159 − 2229	392 − 524	147 − 3986	1951 − 3783	
				Ketones			
17	diacetyl[c]	13.1 − 23.9	14.9 − 32.5	42.5 − 197	n.d.	18.5 − 55.7	*5, 6*
18	acetoin[c,d]	276 − 597	270 − 979	358 − 601	194 − 740	930 − 1020	*9, 46, 60*
19	acetophenone	n.d. − n.q.	n.d. − n.q.	n.q. − 62.1	n.d. − n.q.	n.q.	
	total ketones[c]	289 − 621	294 − 1007	401 − 798	194 − 496	949 − 1076	
				Ethyl Esters			
20	ethyl propanoate[d]	214 − 665	n.q. − 1493	700 − 6396	118 − 1142	3969 − 4547	*46*
21	ethyl isobutyrate[d]	269 − 361	n.q. − 671	330 − 1379	176 −1033	1202 − 1653	*9*
22	ethyl butyrate	50.7 − 209	n.q. − 338	98.6 − 1061	n.q. − 143	387 − 770	*9, 46*
23	ethyl 2-methylbutyrate	n.q. − 71.1	n.q. − 156	49.9 − 401	n.d.	n.d.	
24	ethyl isovalerate	371 − 788	n.q. − 1015	466 − 3317	n.d. − 491	969 − 1492	*9, 46*
25	ethyl valerate	n.d.	n.d. − 13.3	n.d. − 42.5	n.d. − 11.4	18.1 − 25.7	*9, 60*
26	ethyl hexanoate	n.d.	n.d. − 63	n.d. − 248	n.d. −121	98.5 − 143	*9, 46, 60*
27	ethyl heptanoate	n.d	n.d	n.d. − 7.46	n.d.	n.d.	
28	ethyl lactate[c,d]	1.24 − 9.19	0.0 − 9.23	2.04 − 30.4	1.23 − 10.8	41.4 − 48.9	*5, 9*
29	ethyl octanoate	n.d.	n.d.	n.q. −91.4	n.d.	n.d.	*9, 46*
30	ethyl furoate	25.1 − 88.0	36.6 − 255	251 − 422	34.1 − 122	220 − 313	
31	ethyl benzoate	n.d.	n.d. − 7.6	n.d. − 41.6	n.d.	n.d.	
32	ethyl phenylacetate	n.d.	n.q. −136	86.6 − 200	n.d.	328 − 512	*9*
33	diethyl succinate[c,d]	0.08 − 1.7	0.09 − 0.53	0.1 − 0.53	2.15 − 21.8	4.9 − 8.2	*5, 7, 9, 46, 60*
	total ethyl esters[c]	2.86 − 12.7	0.35 − 12.9	4.21 − 44.8	5.24 − 33.6	53.2 − 66.0	
				Alcohols			
34	methanol[c,a,d]	15.9 − 30.4	30.8 − 53.1	19.7 − 68.5	69.9 − 193	22.1 − 78.6	*5, 8*
35	ethanol[c,a,d]	425 − 1135	1002 − 3022	944 − 3412	3284 − 9479	4616 − 12387	*5, 8*
36	1-propanol[c,a,d]	n.d. − 0.97	n.d. − 14.4	0.30 − 19.2	1.19 − 34.7	19.6 − 62.6	*5, 8*
37	isobutanol[c,d]	3.16 − 5.56	2.27 − 5.85	3.45 − 8.53	7.98 − 12.9	9.97 − 10.8	*9*
38	2-methyl-1-butanol[c]	7.64 − 9.88	2.24 − 13.5	6.13 − 12.5	6.91 − 14.4	8.54 − 9.70	*5, 7–9, 46, 60*
39	3-methyl-1-butanol[c,d]	4.77 − 18.1	1.49 − 26.6	7.58 − 48.2	29.3 − 78.3	31.7 − 35.7	*5, 7, 8, 46, 60*
40	1-hexanol	n.d. −88	n.d.	n.d. − 88	n.q. − 449	n.q.	*9, 46*
41	*cis*-3-hexen-1-ol	14.2 − 52.5	15.9 − 51.8	27.0 − 43.7	31.7 − 55.0	18.3 − 20.4	*9, 46*
42	benzyl alcohol	137 − 624	133 − 737	378 − 1236	184 − 4407	529 − 563	*9, 46, 60*
43	furfuryl alcohol	289 − 413	134 − 1142	255 − 1124	0.0 − 1004	635 − 1147	*10, 46*
44	2-phenylethanol[c,d]	5.93 − 11.5	4.99 − 11.2	12.7 − 18.9	23.4 − 30.3	20.7 − 22.9	*5, 7–9, 46, 60*
	total alcohols[c]	475 − 1212	1063 − 3134	997 − 3560	3435 − 9840	4730 − 12609	
				Terpene			
45	α-terpineol	n.d. − n.q.	n.d. − 69.6	n.d. − 121	n.q.	n.d.	*9*
				Acids			
46	isovaleric acid[c,d]	38.4 − 57.2	39.6 − 55.0	58.7 − 121	1.16 − 11.2	24.5 − 33.2	*9, 60*
47	hexanoic acid	784 − 1325	683 − 2185	1860 − 2269	437 − 3322	1424 − 2296	*9, 46, 60*
48	heptanoic acid	n.d.	n.d. − 150	114 − 302	n.d. − 153	n.d. − 237	(*46*)
49	octanoic acid	144 − 531	182 − 704	350 − 774	160 − 732	299 − 546	*9, 46, 60*
50	nonanoic acid	n.d.	n.d.	n.d.	n.d. − 86.3	n.d. −52.8	
51	decanoic acid	24.4 − 113	21.6 − 92.0	68.1 − 106	27.0 − 136	66.3 − 80.8	*9, 46, 60*
	total acids[c]	39.9 − 58.2	42.3 − 58.1	61.1 − 62.6	1.79 − 20.8	26.2 − 36.4	
				Lactones			
52	γ-butyrolactone	682 − 1655	924 − 6583	2693 − 5385	1154 − 2238	1674 − 3337	*7, 46*
53	*trans*-β-methyl-γ-octalactone [d]	64 − 77	65 − 88	75 − 117	102 − 313	85 − 237	*10*

Table 3. Continued

No	compound	mean concentration (μg/L) VJ ($n = 3$)	VR ($n = 5$)	GR ($n = 3$)	VT ($n = 4$)	VB ($n = 2$)	previously reported in Sherry wine vinegar
54	*cis*-β-methyl-γ-octalactone[d]	n.q. − 136	n.q. − 152	125 − 155	363 − 1534	204 − 1179	*10*
55	sotolon[b,d]	n.d.	n.d. − 748	663 − 939	n.d.	n.d.	
	total lactones	894 − 1829	1167 − 7478	3628 − 6536	1619 − 3823	1963 − 4753	
				Phenols			
56	guaiacol[d]	n.d. − 9.8	n.q. − 16.1	11.3 − 21.3	8.4 − 30.6	149 − 301	*10*
57	eugenol	n.d.	n.d.	n.d.	n.q. − 118	n.q. − 81.0	*9, 10*
58	4-Ethylphenol[d]	290 − 1652	427 − 1516	901 − 2382	94.3 − 427	405 − 509	*9, 60*
	total phenols	290 − 1652	427 − 1530	912 − 2397	103 − 441	554 − 891	
	total amounts[c]	1155 − 2576	2305 − 6105	2211 − 5127	6059 − 14405	7754 − 17640	
				Relative Area (Abundance)			
	2-acetylfuran	0.006 − 0.017	0.008 − 0.016	0.019 − 0.039	0.002 −0.006	0.087 − 0.114	
	2,3-butanediol diacetate	0.085 − 0.163	0.115 − 0.315	0.189 − 0.529	0.027 − 0.140	0.199 −0.209	
	TDN	0.044 − 0.105	0.011 − 0.049	0.016 − 0.030	0.008 − 0.415	0.008 − 0.014	

[a] GC-FID n.d.: below detection limit. [b] LLE-GC-MS n.q.: below quantification limit. [c] Concentration in mg/L. [d] Significant differences ($p > 0.05$) among Sherry, red, and Balsamic vinegars.

(*24*). A drop of extract was placed on a perfume sampling paper, and the aroma was compared with the original vinegar as a pair. Five members of our sensory panel were asked to rate the similarity on a discontinuous scale from 0 (no similarity) to 10 (equal) of each extract with the VR1 vinegar.

Gas-Chromatography (GC) Analysis. We used three different methods to determine the volatile compounds of interest in Sherry vinegar samples. A total of 52 compounds were determined by headspace sorptive extraction (HSSE) gas chomatography-mass spectrometry (HSSE-GC-MS). This method was not adequate for the determination of some major compounds such as ethyl acetate, ethanol, methanol, acetaldehyde, and propanol because of their high concentrations, among others. Hence, these 5 compounds were quantified by GC-flame ionization detector (GC-FID). For the special case of sotolon (polar compound), the HSSE-GC-MS method was not suitable because of the apolar nature of the sorbent in the stir bar, polydimethylsiloxane (PDMS). For this reason, sotolon was determined by liquid−liquid extraction GC-MS (LLE-GC-MS).

GC-FID Analysis. Ethyl acetate, acetaldehyde, methanol, ethanol, and propanol were quantified by GC-FID using the method proposed by Morales et al. (*7*). A 1 mL sample was filtered through Millex-GV_{13} filters of 0.22 μm, and 1 μL of 4-methyl-2-pentanol at 102.14 mg/L was added as internal standard (IS). Filtered samples were analyzed using a Hewlett-Packard 6890 gas chromatograph equipped with a flame ionization detector (FID). One microliter was injected in the split mode (1:60) into a CP-Wax 57 CB column, 50 m × 0.25 mm DI × 0.2 μm film thickness (Varian, Middelburg, Netherlands). The carrier gas was H_2 at 1 mL/min. The program temperature was 35 °C for 5 min, ramped at 4 °C/min to 150 °C held for 17.5 min. The injector was set to 220 °C and the detector to 250 °C. Data acquisition software was HPChemstation data processing system (Agilent Technologies).

Liquid−Liquid Extraction GC-MS (LLE-GC-MS). 4,5-Dimethyl-3-hydroxy-2(5*H*)-furanone (Sotolon) was quantified by LLE-GC-MS using the method proposed and validated by Silva Ferreira et al. (*24, 25*). To 50 mL of the samples, 5 g of anhydrous sodium sulfate was added and extracted twice with 5 mL of dichloromethane. The two organic phases obtained were blended and dried over anhydrous sodium sulfate. Then, 2.5 mL of the organic extract was concentrated 5 times under a nitrogen stream, and 5 μL of 3,4-dimethylphenol in dichloromethane at 0.55 mg/L was added as internal standard (IS). Four microliters of extracts were analyzed by GC-MS, using the conditions described elsewhere with minimum changes (*24*). The column employed was a CPWax- 57CB, with 50 m × 0.25 mm and 0.20 μm film thickness (Varian, Middelburg, Netherlands). The injector port was heated to 220 °C in splitless mode for 1 min, with a total flow rate of 53.5 mL. The carrier gas was He at a flow rate of 1 mL/min. The oven temperature was 40° (for 1 min), which was then increased at 2 °C/min to 220 °C and held for 30 min. The quadrupole, source, and transfer line temperatures were maintained at 150, 230, and 280 °C, respectively. The analysis was performed in SIM mode, and the ions selected for each compound studied were *m/z* 83 (sotolon) and *m/z* 107 (IS).

Headspace Sorptive Extraction GC-MS Analysis (HSSE-GC-MS). The HSSE sampling conditions were as follows (*26*): 5 mL of sample (wine vinegar) and 10 μL of 4-methyl-2-pentanol (IS) at 1045 mg/L was placed into a 20-mL headspace vial with 1.67 g of NaCl. A 10 mm long stir bar coated with 0.5 mm polydimethylsiloxane (PDMS) layer (Twister, Gerstel, Müllheim an der Ruhr, Germany) was placed in an open glass insert and placed into the vial to achieve the extraction in the headspace. Then, the vial was tightly capped and heated for 60 min at 62 °C in a thermostatic bath. The stir bar was removed with tweezers, rinsed with Milli-Q water, and dried with lintfree tissue paper. Finally, for the thermal desorption (TD), the stir bar was placed into a glass tube of 60 mm length, 6 mm o.d., and 4 mm i.d., which was placed in the autosampler tray of the thermo desorption unit for GC-MS analysis.

Gas chromatography analysis was carried out with a 6890 Agilent GC system coupled to a quadrupole mass spectrometer Agilent 5975inert and equipped with a Gerstel, Thermo Desorption System (TDS2) and a cryo-focusing CIS-4 PTV injector (Gerstel). The thermal desorption was performed in splitless mode and with a flow rate of 90 mL/min. The desorption temperature program was the following: 35 °C for 1 min, ramped at 60 °C/min to 250 °C, and held for 5 min. The CIS-4 PTV injector, with a Tenax TA inlet liner, was held at −35 °C with liquid nitrogen for total desorption time and then raised at 10 °C/s to 290 °C, and held for 4 min. Solvent vent mode was employed for the transfer of sample to the analytical column. A CPWax-57CB column, 50 m × 0.25 mm and 0.20 μm film thickness (Varian, Middelburg, Netherlands), was used, and the carrier gas was He at a flow rate of 1 mL/min. Oven temperature program was 35 °C for 5 min, then raised to 220 at 2.5 °C/min (held 5 min). The quadrupole, source, and transfer line temperatures were maintained at 150, 230, and 280, respectively. Electron ionization mass spectra in the full-scan mode were recorded at 70 eV electron energy in the range 35 to 350 amu.

All data were recorded using a MS ChemStation. The identity of 64 peaks (52 of them quantified) was assigned using the NIST 98 library and confirmed by retention index of standards when they were available. Quantification was performed employing the relative area to internal standard of the target ion of each compound. We built the respective calibration curves for each compound, plotting concentration versus relative areas. The samples were analyzed by triplicate, and blank runs of empty glass tubes were done before and after each analysis.

Gas Chromatography−Olfactometry. To identify substances responsible for the aromatic notes associated with the selected descriptors of the typical aroma of Sherry vinegar, GC olfactometric analysis was applied to three representative samples corresponding to the three different qualities of Sherry vinegars: VJ2, VR1, and GR2. Extraction was performed according to the methodology previously described for

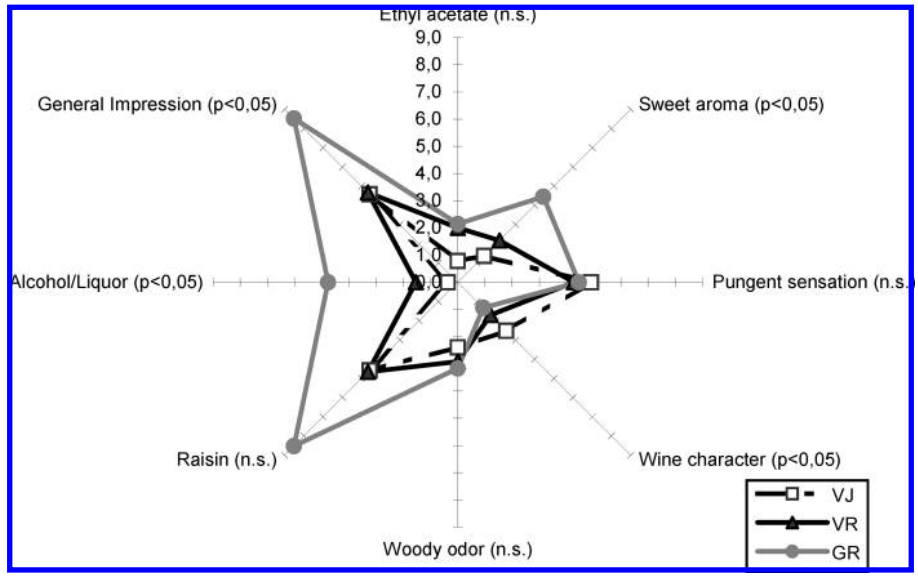

Figure 1. Aroma profile of three representative Sherry vinegars, one from each category ("Vinagre Jerez", "Reserva", and "Gran Reserva").

LLE-GC-MS. Then, 2 mL of this organic phase was concentrated 5 times under a nitrogen stream. Several dichloromethane extracts from different vinegars were submitted to GC-O. Chromatographic conditions were the following: Hewlett-Packard HP 5890 gas chromatograph; BP-21 column (50 m × 0.25 μm), fused silica (SGE, France); hydrogen (5.0, Air−liquid, France); flow, 1.2 mL/min; injector temperature, 220 °C; oven temperature, 40 °C for 1 min programmed at the rate of 2 °C/min to 220 °C, maintained during 30 min. Extract aliquots of 1 μL were injected into the GC in splitless mode (0.5 min); split flow, 30 mL/min. The makeup gas employed on the olfactometric device (SGE, France) was air (80% N_2; 20% O_2) (Air−liquid, France). Two streams were used: one was bubbled in water, nose moistener, the other was applied at the exit of the GC column to lower the temperature of the effluent.

Compound Identification. Identification of odorants was performed by comparison of MS spectra, chromatographic retention indices (RIs), and odor description with experimental and literature data. RIs were calculated in GC-FID-O and HS-SBSE-GC-MS from the retention times of *n*-alkanes by linear interpolation, according to the literature (*25*).

Statistical Analysis. All statistical analysis were performed by means of Statistica software, version 7.0 (Statsoft, Tulsa, USA).

RESULTS AND DISCUSSION

Sensory Descriptive Analysis of Sherry Vinegars. The 11 Sherry vinegar samples considered in this study were described by the expert sensory panel (*15*). The selected descriptors were ethyl acetate, sweet aroma, pungent sensation, wine character, woody odor, raisin, alcohol/liquor, and general impression. The attributes bitter almond, red fruit, and vanilla were not considered as they reached very low scores in all of the samples. Representative spider charts are shown in **Figure 1**. Pungent sensation and general impression reached the highest scores, while ethyl acetate and wine character accounted for the lowest marks. The scores obtained in the three categories for ethyl acetate, woody odor, and pungent sensation were similar, which suggests that these are generic characteristics of Sherry vinegars. Raisin reached similar marks in "Vinagre Jerez", and "Reserva" vinegars; however, "Gran Reserva" samples accounted higher marks to this attribute. On the contrary, the scores given to the descriptors general impression, alcohol/liquor, sweet aroma, and wine character were significantly different among the three different qualities (according to the $p \leq 0.05$ value obtained in the ANOVA). General impression, alcohol/liquor, and sweet aroma reached the highest values for "Gran Reserva" (the oldest vinegars). Hence, these three sensory descriptors are related with the time spent by vinegars in wood, that is to say, with the aging of vinegars. Conversely, wine character reached the highest scores for "Vinagre Jerez" (the youngest vinegars), being inversely correlated with aging.

Selection of a Representative Extract for GC-O. A total of five panelists were asked to score the similarity between the odor of three extracts (dichloromethane, hexane, and ether extracts) and the odor of the VR1 vinegar itself. Results indicate that among the four extracts tested, the dichloromethane extract is the most representative since it reached the highest similarity values and was ranked by all the panelists in the first place (**Table 2**). In addition, the dichloromethane extract showed significant differences ($p < 0.05$) with the other extracts. Hence, this solvent was chosen to perform the GC-Olfactometry analysis.

Quantification and Identification of Aroma Compounds. **Table 3** shows ranges of aroma compounds determined in 11 Sherry vinegars, 4 red, and 2 balsamic vinegars, and as can be seen, a total of 58 compounds were quantified. To our knowledge, among them, 7 had not been previously reported in wine vinegars. These new compounds are principally esters (ethyl 2-methylbutyrate, ethyl heptanoate, ethyl furoate, and ethyl benzoate), ketones (acetophenone), acids (nonanoic acid), and lactones (sotolon). Some of them have been previously identified and quantified in Sherry wines (ethyl heptanoate, ethyl furoate, ethyl benzoate, and sotolon) (*27, 28*) and red wines (ethyl 2-methylbutyrate) (*29, 30*). Sotolon was identified as a key aroma compound in flor wines (*20, 31–33*). This compound is a very powerful odorant, which contributes to the characteristic sensory impression of several foods, and it is also present in other types of wines such as the Botrytised wines, Jura wines, ("vins jaunes" and "vins doux naturelles"), Port, and Tokay (*34*). The odor of sotolon is described as nutty at low concentrations and curry at higher levels. Therefore, the presence of this molecule is closely related with aging in Port wines (*24, 35*), and according to Moreno et al. (*28*), sotolon along with acetaldehyde diethylacetal can be used as markers of the changes in "fino" Sherry wine during its biological aging. The formation mechanism of sotolon is not totally clarified in wines (*33, 36–41*). Nevertheless, some studies have demonstrated that oxygen has an important role on the rate of formation of this key odorant (*22, 24, 25, 35, 42*). In addition, it seems that sotolon is originated in biologically aged wines by chemical reaction

between α-ketobutyric acid and the acetaldehyde produced by flor yeasts (*20*), as proposed by Pham et al. (*33*).

In this work, sotolon was detected in 7 of the 11 Sherry vinegars (all the "Reserva" samples, except for VR2, and the three "Gran Reserva" vinegars) with a concentration ranging 663−939 μg/L. This compound is found at levels from only few dozen μg/L in young wines to about 100 μg/L in 10-year-old wines and up to 200 μg/L after 10 additional oxidative aging years (*20, 24*). In agreement with this, except for VR2, sotolon was detected and quantified in the most aged Sherry vinegar categories ("Reserva" and "Gran Reserva"). As can be seen in **Table 3**, most aged vinegars (GR) account for the highest concentrations. Hence, the formation of this compound is favored by time in an oxidative medium. Sotolon was not quantified in any "Jerez Vinegar" class samples. This is in agreement with Zea et al. (*43*) since "fino" Sherry wines aged for less than 2.5 years showed very low concentrations of sotolon, below its odor threshold in wine (5 μg/L). After this time, concentrations of sotolon were higher, ca. 700 μg/L, and were increasing with aging. However, it is remarkable that sotolon was not quantified in any of the red and balsamic vinegars (**Table 3**).

Being present in concentrations between 132 and 3955 mg/L, ethyl acetate was by far the major volatile compound in all of the samples, followed by considerable amounts of acetoin (194−1020 mg/L). For Sherry wine vinegars, outstanding concentrations for diacetyl (13.7−197 mg/L) and isovaleric acid (38.4−121 mg/L) were also determined. Among these compounds, ethyl acetate has a particular relevance due to its great influence on the final sensory profile of Sherry vinegars. In addition, we observed a correlation between amounts of ethyl acetate and ethanol ($r = 0.8$). Other authors also observed that acetoin and isovaleric acid were two of the major volatile compounds quantified in commercial Sherry vinegars (*8, 9*). Diacetyl (produced by the oxidation of acetoin) has been reported to increase with aging and is proposed as an indicator of the age of Sherry vinegars (*5*). This finding has been confirmed in our samples. This compound was not present in red wine vinegars (**Table 3**).

Acetaldehyde diethylacetal is formed in Sherry wine from the acetaldehyde produced by flor yeast and exhibits a strong odor impact on wines under biological aging, to which it contributes with green fruit and liquorice aroma notes (*20*). This compound was quantified in most of the studied Sherry vinegars, with concentrations ranging between 2.0 and 223 mg/L.

Hexyl acetate and eugenol could not be quantified in the Sherry vinegars since their concentrations were under their limits of detection (LOD). Vanillin, eugenol, guaiacol, and *cis*- and *trans*-β-methyl-γ-octalactone, also named oak lactones, could be effectively extracted from oak wood and oak chips during experimental aging of wine vinegars (*10*). Eugenol has a clove-like aroma, and its concentration increases when barrels are heated at medium or heavy toast levels. This compound is present at very low concentrations in Sherry vinegars (*9*), usually under detection limits (LOD). Vanillin was quantified in 7 of the 11 Sherry vinegars in a concentration ranging between 1271 and 8875 μg/L. This compound is considered an important contributor to the quality of barrel-aged wines, and its content in wood barrels depends on differences in heat penetration, rather than the intensity of toasting. *cis*-β-Methyl-γ-octalactone was quantified in eight Sherry vinegars and in all of the red and balsamic vinegars. The *cis*/*trans* ratios were higher than 5 for red and balsamic samples aged in new barrels and lower than 2 for Sherry vinegars, as opposed to wine aged in American oak barrels, whose ratios are always greater than 5 (*44*). This is probable because of the fact that Sherry vinegars, in general, are produced in very old wood barrels.

Besides sotolon (previously mentioned), ethyl-2-methylbutyrate, ethyl heptanoate, ethyl octanoate, ethyl benzoate, acetophenone, and α-terpineol were only quantified in Sherry vinegars since their concentrations were under the limits of quantification (LOQ) in red and balsamic samples. In addition, ethyl heptanoate, ethyl octanoate, α-terpineol, and acetophenone were only determined in "Gran Reserva" vinegars.

Concentrations found for hexanal, 2-phenylethyl acetate, ethyl furoate, 2-phenylethanol, and sotolon were significantly different ($p \leq 0.05$) among the three different Sherry qualities, reaching the highest concentrations for "Gran Reserva" vinegars.

On the other hand, as was expected, red vinegars accounted for the highest amounts of methanol since red wines have higher concentration (152 mg/L) than rosés (91 mg/L), while white wines have even less (63 mg/L) (*45*). Consequently, the concentration of methyl acetate in red vinegars was also proportionally higher in relation to the time of aging.

Other volatile compounds identified in the samples and confirmed with their corresponding mass spectra of respective standards were acids such as propanoic (RI 1544), isobutyric (RI 1572), butyric (RI 1648), and pentanoic (RI 1751) acids, and *cis*-3-hexen-1-ol acetate (RI 1304), isomers of linalool oxide (RI 1355 and 1377), methyl salicylate (RI 1766), and 5-hydroximethyl-2-furfulraldehyde (RI 2357). Most of them had been previously reported as constituents of Sherry vinegar aroma (*9, 46*). Methyl salicylate and isomers of linalool oxide have been identified in wines, but to our knowledge, it is the first time they are described in wine vinegars. 5-Hydroximethyl-2-furfulraldehyde (5-HMF), which is primarily a Maillard reaction product, increased in the oldest samples according to other authors (*4*). This compound can be extracted from oak wood, although its presence in vinegars has been traditionally attributed to the legal practice of must caramel addition.

Other compounds were tentatively identified with the aid of the NIST library and RIs, since their corresponding standards were not available: isomers of 2,3-butanediol diacetate (RI 1380 and 1488), acetylfuran (RI 1504), and 1,1,6-trimethyl-1,2-dihydronaphthalene (TDN) (RI 1734). TDN if present at concentrations above 20 μg/L causes an unpleasant kerosene or petrol-like note, contributing to the off-flavor of wine. Nevertheless, several authors pointed out a positive influence in the wine aroma complexity when TDN is present at concentrations lower than the threshold limit (*47*). This compound seems to be a genuine compound of long aged cavas (*48*) and Riesling wines (*49*), in which it increases with maturation. Nevertheless, as can be seen in **Table 3**, we obtain relative areas of TDN decreasing with time of aging in Sherry vinegars. This finding is similar to that in Madeira wines in which this compound decreases with oxidative aging (*47*).

Correlations between sensory descriptors and aroma compounds (data not shown) demostrate that sweet aroma is the descriptor better correlated with a major number of volatile compounds, a total of 16, with most of them being esters, such as isoamyl acetate, ethyl furoate, and isobutyl acetate ($r > 0.7$). On the contrary, raisin and wine character are not correlated with any single compounds. It is also remarkable that *trans*-β-methyl-γ-octalactone is correlated with woody odor ($r = 0.74$) as was expected since it is responsible for the oak wood odorant note present in barrel-aged alcoholic beverages (*50, 51*). Besides, the sensory attribute alcohol/liquor is correlated with sotolon ($r = 0.7$).

Table 4. Classification and Cross-Validation Results of LDA

	predicted group membership (%)	
	Sherry vinegar	red vinegar
Original Model		
Sherry vinegar	100	0
red vinegar	0	100
Cross-Validation Model		
Sherry vinegar	100	0
red vinegar	0	100

Multivariate Statistical Analysis. To perform multivariate statistical analysis, we made a substantial reduction of variables. First, those compounds accounting for a high number of no detected or no quantified scores in the samples were eliminated. Moreover, redundant variables with high correlation coefficients ($r > 0.7$) were eliminated. Finally, we used 14 variables: acetaldehyde, ethanol, methyl acetate, ethyl isobutyrate, diacetyl, hexanal, 2-methyl-1-butanol, acetoin, γ-butirolactone, *trans*-β-methyl-γ-octalactone, 4-ethylphenol, decanoic acid, and sotolon.

Linear discriminant analysis (LDA) was performed considering two groups of samples: Sherry vinegars and red vinegars. Balsamic vinegars ($n = 2$) were excluded because of a very low number of samples ($n = 2$). LDA is a supervised chemometric method widely used for classification purposes. This method minimizes the variance within categories and maximizes the variance between categories. LDA renders a number of orthogonal linear discriminant functions equal to the number of categories minus one; when two classes are considered, one linear discriminant function is obtained.

When LDA is applied to a set of samples, the samples are usually divided into a training set and a test set, the first one to find discriminant functions and the second one to check the utility of those discriminant functions to correctly classify new samples. In our case, we have used the so-called leave-one-out method (*52*) consisting in dividing the whole set of samples into two groups: a training set holding all the samples except one which is used then as a test set. Thus, LDA was applied as many times as the number of samples.

One discriminant function that includes the variables ethanol, diacetyl, hexanal, *trans*-β-methyl-γ-octalactone, 4-ethylphenol, and sotolon was obtained when the LDA forward stepwise method was applied. We have obtained 100% of correct classifications of samples in the cross-validation analysis by the leave-one-out method, and the results are reported in **Table 4**.

GC-O. GC-O experiments were conducted with dichloromethane extracts obtained from three Sherry vinegars, representative of each quality (VJ2 "Vinagre Jerez", VR1 "Reserva", and GR2 "Gran Reserva"). This olfactometry study as screening procedure was performed by a panel of four individuals, and sniffing of samples were carried out in triplicate to increase the robustness of data. Results of the screening are summarized in **Table 5**. We attempted to correlate the chemical molecules identified by MS with the aroma perceived with the same RI. The descriptors were selected according to their frequency of citations. Hedonic terms (good/bad) and their analogues were not considered and were replaced by the most cited.

A total of 80 odors were obtained in the sniffing of the three samples. Among them, 25 were found in the three vinegars, and only 8 of them were detected by all of the panelists: glue (RI 1063), butter (RI 1084), cherry/strawberry (RI 1118), banana/mulberry/strawberry (RI 1123), strawberry/banana (RI 1414), pungent (RI 1422), cheese (RI 1705), and curry/liquorice (RI 2201). These odor-active regions were identified as ethyl acetate, diacetyl, butyl acetate, isoamyl acetate, ethyl octanoate, acetic acid, isovaleric acid, and sotolon, respectively. In addition, other 9 odors perceived in all of the samples reached a frequency $\geq 50\%$: strawberry (RI 1080, ethyl isobutyrate), river water/lake/vapor (RI 1532, unknown), cheese/feet (RI 1595, isobutyric acid), burned/burned hair (RI 1655, unknown), cheese/vomit (RI 1811, unknown), boiled vegetable (RI 1875, unknown), clove (RI 2054, eugenol), sweet/vanilla (RI 2076, unknown), and flower/fruit/banana (RI 2151, unknown).

By comparison of the three vinegars, we can see that sample GR2 revealed 64 odor-active areas with 18 of them reaching the maximum frequency (100%). VR1 presented 62 odors, 14 of them being perceived by all the assessors, and VJ2 was the sample with a minor number of odor-active regions up to 46, and only 10 of them obtained the maximum frequency. Hence, a greater aromatic complexity is observed when the time of aging in wood increases.

Odor Activity Values. After screening by detection frequency in GC-O, calculation of odor activities values (OAVs) enables a more reliable evaluation of potent odorants for a given product, despite its limitations. OAVs are obtained by dividing the concentration of the compound by its recognition threshold in a suitable matrix (*53*). Hence, OAV is linearly proportional to concentration and threshold (*54*). However, it is known that the slope of the psychometric function of a compound varies markedly between different compounds (*55*). So, the intensity of some volatile compounds will rise rapidly after exceeding their odor threshold (OT), while the intensity increment of other volatile compounds can be very small over many orders of concentration magnitude (*56*).

In relation to the OAV concept, although it is not a psychophysical measure for perceived odor intensity, it is assumed that the odorants showing high OAVs contribute strongly to the overall aroma (*54, 57*). However, because of masking, a compound showing an OAV > 1 can still be insignificant in a mixture and has to be examined further by sensory analysis (*53*).

First of all, we had to calculate our own odor thresholds for the special case of vinegar matrices. For that, we selected those odorants which either reached high detection frequency in GC-O, or high concentrations in Sherry vinegar, or even those with important impact in wines. To estimate the odor contributions of the selected odorants, their OAVs were calculated on the basis of their nasal thresholds in a 7% (w/v) acetic acid solution (**Table 6**). Compounds in the table are ranked according to the maximum odor activity values (OAV max) reached in the three Sherry wine vinegars under study in GC-O experiments (VJ2, VR1, and GR2). Altogether, 20 of the 27 odorants showed in **Table 6** reached concentrations above their odor thresholds in this set of Sherry vinegars. Data in the table confirm the results obtained in the olfactometry study to almost all of the selected odorants and in fact support the usefulness and validity of the GC-O approach in this work. Hence, it can be seen that nearly all of the compounds with high GC-O scores also had high OAV. The single exception to this observation is butyl acetate, with OAV < 1.

The highest odor activity value of 4899 was calculated for diacetyl, followed by isoamyl acetate, which was the second in rank. In addition, an increase in OAV for diacetyl and isoamyl acetate was observed in vinegars with longer aging in wood. Hence, GR2 displayed the highest OAVs. Other compounds such as acetaldehyde diethylacetal, ethyl isovalerate, ethyl

Table 5. Detection Frequency (%) of the Odors of VJ2, VR1, and GR2 Sherry Vinegars Detected and Described by the Sniffing Panel

RI[b]	RI[c]	odor quality	odorant (tentative identification)	VJ2	VR1	GR2
1063		glue	ethyl acetate	100	100	100
1070		alcohol	ethanol	75	50	50
1072		rancid	unknown	25	50	75
1076	928	chemical, alcohol, grass, plastic	acetaldehyde diethylacetal	25	50	0
1080	962	strawberry	ethyl isobutyrate	50	100	100
1084	969	butter	diacetyl	100	100	100
1089	998	plastic, medicinal, chemical	isobutyl acetate	0	100	50
1097	1014	strawberry	ethyl butyrate	0	50	75
1105	1028	fruit, banana	ethyl 2-methylbutyrate	0	75	50
1110	1044	strawberry	ethyl isovalerate	75	0	25
1118	1046	cherry, strawberry	butyl acetate	100	100	100
1123	1112	banana, mulberry, strawberry	isoamyl acetate	100	100	100
1173	1150	fruit, banana	amyl acetate	0	75	50
1181		banana	unknown	0	50	0
1220	1207	rancid	3-methylbutanol	25	50	0
1239	1208	banana, fruit, mulberry	ethyl hexanoate	0	75	50
1254	1257	mulberry, banana	hexyl acetate	0	0	75
1277		rancid	unknown	0	0	25
1297		boiled potato	unknown	50	50	0
1327	1271	sweet, yogurt, dairy product	acetoin	0	50	25
1360		toasted maize	3-hydroxy-2-pentanone	0	25	50
1414	1432	strawberry, banana	ethyl octanoate	100	100	100
1418		boiled potato	unknown	75	0	75
1422		pungent	acetic acid	100	100	100
1438		fruit, flower, strawberry	linalool oxide (isomer)	50	0	100
1439		feet	unknown	0	75	50
1461		strawberry, sweet, mulberry	unknown	0	25	75
1484		boiled potato	methional	0	75	100
1496		strawberry, sweet	unknown	0	100	50
1510		toasted maize, fried chicken, burned	2,3-butanediol diacetate	50	25	25
1513		boiled potato	unknown	0	0	25
1517		strawberry	unknown	0	0	50
1520		plastic	unknown	25	0	25
1532		river water	unknown	75	100	100
1537		strawberry, alcohol, roses, sweet	unknown	0	50	50
1545		banana, mulberry	ethyl 3-hydroxybutanoate	25	50	0
1553		flower, roses, sweet	unknown	50	25	75
1557	1518	aspirin, mulberry, fruit	benzaldehyde	100	75	0
1563	1536	aspirin, mulberry, cherry	ethyl nonanoate	100	100	0
1586		rancid, cheese, feet	propanoic acid	0	50	75
1595		cheese, feet	isobutyric acid	75	75	100
1655		burned, burned hair	unknown	75	75	50
1659		strawberry	unknown	50	0	25
1661		cheese, vomit	butyric acid	0	100	50
1671	1664	burned, burned hair	furfuryl alcohol	0	25	25
1679	1659	sweet	ethyl benzoate	50	0	0
1685		roses, talcum power, perfume	unknown	0	0	50
1705	1670	cheese	isovaleric acid	100	100	100
1722		strawberry	unknown	0	0	25
1747		rancid, cheese	pentanoic acid	25	50	50
1762		boiled vegetable or potatoes	methionol	50	50	0
1763		strawberry, fruit	unknown	75	0	25
1780	1770	plasticine, wax pencil	ethyl phenylacetate	0	50	25
1786		urine	ethyl salicylate	0	25	0
1789	1800	grass, feet, humidity	2-phenylethyl acetate	0	25	25
1796		roses, floral perfume	unknown	50	0	100
1802		metallic	unknown	0	25	0
1809		boiled vegetable	unknown	0	50	0
1811		cheese, vomit	unknown	75	50	100
1842		sweet, fruit, fruit preserve	unknown	25	25	0
1858		stewed apples, apple juice	β-damascenone	0	50	75
1875		boiled vegetable	unknown	50	50	50
1878		cheese, feet	unknown	25	25	75
1880		fruit, fruit preserve	unknown	0	25	75
1889		sweet, vanilla	2-methyl-3-hydroxy-4-pyrone	75	50	0
1896		metallic	unknown	50	0	25
1932	1927	cheese	heptanoic acid	0	50	50
2017		flower (daisy), chamomile tea	4-ethylguaiacol	0	25	25
2028	2009	urine, chamomile tea, chemical	octanoic acid	0	0	25
2033		flower, honey, roses	unknown	75	0	100
2051		coconut, sweet	γ-decalactone[a]	0	50	100
2054	2087	clove	eugenol	50	50	75
2076		sweet, vanilla	unknown	75	75	100
2098		clove, vanilla, pepper	4-vinylguaiacol	25	50	75
2105		toasted, dried fruit	unknown	25	0	25
2113		liquor, "oloroso sherry wine", sweet	unknown	50	50	0
2137	2098	sweet, vanilla	nonanoic acid	25	25	75
2149	2097	cardboard, metallic, meta	4- ethylphenol[a]	0	25	0
2151		flower, fruit, banana	unknown	75	75	75
2201		curry, liquorice, "oloroso sherry wine", toffee, syrupy sugar	sotolon	100	100	100

[a] Possibly identified compounds. [b] BP-21 column. [c] CPWax- 57CB column.

Table 6. Odour-Activity Values (OAV) and Odor Thresholds

odorant	odor threshold (μg/L)	OAV max	VJ2	VR1	GR2
diacetyl	40	4899	595	807	4899
isoamyl acetate	12	1146	118	365	1146
acetaldehyde diethylacetal	133	865	15	464	865
isovaleric acid	150	807	380	359	807
ethyl isovalerate	4.4	754	84	321	754
ethyl isobutyrate	3.66	377	73.5	149	377
4-ethylphenol	4	326	72	297	326
acetaldehyde[a]	402	155	43	155	130
acetoin	8800	68	68	65	68
2-phenylethyl acetate	88	65	4.7	13	65
ethyl octanoate	1.5	62			62
sotolon	16	59		47	59
vanillin[a]	94	47		47	42
isobutyl acetate	177	27	4.7	10	27
ethyl acetate[a]	91000	14	1.5	10	14
ethyl propanoate	516	12.3	0.5	2.5	12.3
benzaldehyde[a]	158	9.9		0.77	9.9
trans/cis-oaklactones	78	2.7	1.5	1.6	2.7
hexanoic acid	2600	1.8	0.3	0.8	1.8
furfural[a]	6200	1.26	0.22	0.14	1.26
butyl acetate	453	0.8			0.8
octanoic acid	987	0.8	0.15	0.37	0.8
furfuryl alcohol	1415	0.79	0.29	0.28	0.79
propyl acetate	6708	0.5	0.03	0.19	0.5
bencyl alcohol	16900	0.3	0.04	0.04	0.3
ethyl benzoate	210	0.20		0.03	0.20
eugenol	0.17				

[a] Threshold reported in a previous work (*15*).

isobutyrate, and isobutyl acetate also reached high OAVs in the three samples, and their values increased with time.

However, OAV of ethyl acetate was >1 in the three samples but only reached an OAV max of 14 because of its high odor threshold. In spite of this, this compound has a great influence on the final sensory profile (*58*). In addition, ethyl acetate presents a characteristic (glue) aroma, very easy to recognize, and it is one of the selected sensory descriptors for vinegars.

Ethyl propanoate and hexanoic acid showed OAV <1 in VJ2 samples (the youngest vinegar). However, these OAVs increased with aging reaching values major than 1 in the oldest vinegars.

Finally, according to the results of OAV, GC-O, and GC-MS, diacetyl, isoamyl acetate, isovaleric acid, sotolon, and ethyl acetate are characteristic odor active compounds in Sherry vinegars since they showed concentrations far above their odor thresholds and were detected in the three samples analyzed by all panelists. These results confirmed the active role of the five compounds in the tipicity of Sherry vinegars.

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