Development and Characterization of Liquors Prepared with Underutilized Citrus By-products, the Peel

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Abstract Three orange liquors were prepared by macerating of the peel of three orange cultivars ("Clemenules", "Valencia Late", and "Sanguinely"), which were by-products of the citrus industry. The following parameters were analyzed in these spirits: the volatile composition by gas chromatography, total polyphenol content, antioxidant activity ($ABTS^+$, $DPPH^•$, and FRAP methods), sensory evaluation by trained and consumers' panel. These products had a great aromatic potential, characterized by proper volatile profiles, mainly monoterpenes and sesquiterpenes, being the "Clemenules" and "Valencia Late" liquors those with the most complex aroma. On the other hand, the liquor obtained from "Sanguinely" oranges had the highest polyphenol content (557 mg GAE/mL) and the highest antioxidant activity by $ABTS^+$ and FRAP (4.77 and 4.90 mmol Trolox/mL, respectively). The "Clemenules" and "Valencia Late" spirits were those preferred by the consumers, and had intensities of the most relevant attributes close to the optimal values, and were the closest ones to the consumers' ideal liquor. Thus, it has been proven that good liquors can be prepared using citrus peels, which are discarded after manufacturing citrus juices, increasing the options of the juice companies and the potential prices of these fruits and the farmers' revenues.

Keywords: *alcoholic beverages, antioxidant activity, spirits, sensory analysis, orange*

1. Introduction

Citrus is one of the world's major fruit crops, with global availability and popularity, widely contributing to consumers' diet, and its annual worldwide production has witnessed strong and fast growth in the last decades [1]. Citrus are the top fruit crops in terms of world trade. For example, in Spain, 26% of the total volume of juices sold are orange juices, and another 5% comes from mandarin orange juices, which is steadily expanding [2]. This enthusiasm for them –citrus in general- is notably due to their high organoleptic quality and high content in nutritional compounds of interest, especially vitamin C and polyphenols.

About 80% of the citrus production is processed into juice; citrus by-products are the processing wastes generated after citrus juice extraction and constitute about 50% of fresh fruit weight [3]. This solid residue is basically comprised of peel (flavedo and albedo), pulp (juice sac residue), rag (membranes and cores), and seeds. In this way, the disposal of the fresh peels is becoming a major problem to many factories [4]. Usually, citrus juice industries dry the residue and either it is sold as raw material for pectin extraction or pelletized for animal feeding, although none of these processes is very profitable. This residual material is a poor animal feed supplement because of its extremely low protein content and high amount of sugar. The application of agroindustrial by-products in new consumer products will help in solving pollution problems related to their disposal [5-7]. In this sense, the production of liquor by using citrus by-products is an interesting alternative to provide increased household income, because the preparation requires simple technology, and the final product has very long shelf-life.

The citrus peel is a tissue rich in secondary plant metabolites and characterized by pleasant flavors; it is commonly used for preparation of marmalades, candied peels, and essential oil [8, 9]. The peels of fruits are major sources of natural antioxidants. In particular, by referring to citrus fruits, previous studies have shown the presence of higher contents of phenolic compounds, such as phenolic acids and flavonoids, in the peels in comparison to the juice [10].

In the context of the European Union, spirits are governed by Regulation (EC) No 110/2008 [11], liquors citrus is a liquor obtained from the alcoholic extraction of the essential oils from the citrus peel. Besides ethanol and water, citrus liquor contains several volatile and non-volatile minor compounds, which are essential for its sensory characteristics. The former are terpenic compounds, which form the essential oils, and the latter include several classes of non-volatile compounds with potential health-related properties, such as flavonoids, coumarins, and psoralens [12, 13].

Citrus fruit, sweet oranges and mandarin oranges specifically, are not frequently used in the liquor industry.

Therefore, due to the economic importance of these citrus fruits in the Mediterranean region of Spain, the aim of the present study was to prepare and characterize citrus liquors based on by-products with the perspective of developing new products with potential benefits for the human health and improving the use of Spanish citrus by-products. The quality of the citrus liquors was assessed by the evaluation of volatile composition, antioxidant capacity, sensory profile, and consumer's satisfaction degree after 3 months of maceration.

2. Materials and Methods

2.1. Citrus Fruits

Three citrus fruits were studied, 2 cultivars of sweet oranges, (i) *Citrus sinensis* (L.), cultivar "Valencia Late", (ii) *Citrus sinensis* (L.), cultivar "Sanguinelli" (red orange), and 1 cultivar of sweet mandarin oranges, (iii) *Citrus reticulata* (L.), cultivar "Clemenules". The citrus fruits used in this study were all grown in the same farm and under identical conditions of soil, irrigation, and illumination in Eastern Spain (Murcia, Spain). The selected trees were about 12 years old and free from diseases. Fruits from these cultivars were selected on the basis of their diameter, pH, total soluble solids content, and maturity index.

2.2. Liquor Manufacturing

Liquor manufacturing were prepared as previously described in recent publications by our research team [14, 15]. Briefly, the citrus peels were macerated during 3 months with 60% abv (alcohol by volume), at room temperature, and, then, adjusted to an alcoholic strength of 30% abv using a sucrose syrup. Liquors were prepared in three different batches, to allow proper replication of the experiment.

2.3. Extraction of Volatile Compounds

The method selected to study the volatile composition of the liquors was headspace solid-phase microextraction (HS-SPME). 10 mL of liquor and 5 mL of ultrapure water were placed in a 50 mL vial with a polypropylene cap and a PTFE/silicone septum. To promote the release of the volatile compounds to the headspace and avoid enzymatic reactions 15% of NaCl was added to the vial. After 10 min of equilibrium at 40 ºC, DVB/CAR/PDMS fiber was exposed to the sample headspace for 50 min at 40 ºC. This fiber was chosen for its high capacity of trapping fruit volatile compounds and 40 ºC was chosen because is a temperature which can be close to the mouth temperature. Similar extraction procedure has been successfully used in maqui liquor [15], pomegranate wine [16], pistachio [17] and jujube fruits [18], among other fruits.

Desorption of the volatile compounds form the fiber coating was carried out in the injection port of the GC-MS, after sampling, for 2 min at 230 ºC.

2.4. Identification and Quantification of Volatile Compounds

A chromatograph Shimadzu GC-17A (Shimadzu Corporation, Kyoto, Japan), coupled with a Shimadzu mass spectrometer detector GC-MS QP-5050A were performed to the isolation and identification of the volatile compounds. The GC-MS system was equipped with a SupraWax-280 column (Teknokroma S. Co. Ltd., Barcelona, Spain). Analyses were carried out using helium as carrier gas at a flow rate of 0.8 mL/min, at a split ratio of 1:10 and the following temperature program: 40 ºC for 3 min; rising at 5 ºC/min to 100 ºC; rising at 4 ºC/min to 220 ºC and held for 1 min; rising at 20 ºC/min to 260 ºC and held for 5 min. Detector and injector were held at 230 ºC and 260 ºC respectively. Mass spectra were obtained by electron ionization (EI) at 70 eV and spectral range of 45–450 m/z was used.

Three analytical methods were using for the identification of the volatile compounds: (i) retention indexes of problem compounds, (ii) GC-MS retention times of standards, and iii) mass spectra of authentic chemicals and those of the database Wiley 229.

The quantification of the volatile compounds was performed on a gas chromatograph, Shimadzu 2010, with a flame ionization detector (FID). The column and chromatographic conditions were those previously reported for the GC-MS analysis. The injector temperature was 250 °C and nitrogen was used as carrier gas (1 mL/min). Results were expressed as percentage (%) of the total area represented by each one of the volatile compounds and volatile composition analysis was run in triplicate.

2.5. Total Phenolic Content and Antioxidant Activity

Total phenolic content (TPC) was quantified using Folin–Ciocalteu colorimetric method described previously by Gao et al. [19], and used by our research team in recent studies [20]. The results were expressed as gallic acid equivalents (GAE), mg GAE/mL of liquor.

The $ABTS^+$, $DPPH^{\bullet}$, and FRAP assays were prepared as described previously in a recent study by our research team [21]. For all analyses, a standard curve was prepared using different concentrations of Trolox. All determinations were performed in triplicate, using a UV-Vis Uvikon XS spectrophotometer (Bio-Tek Instruments, Saint Quentin Yvelines, France). The results were corrected for dilution and expressed in mmol Trolox/mL.

2.5. Sensory Analysis with Trained Panel

Sensory evaluation with trained panel was used to describe the citrus liquors. A panel of 8 panelists, aged between 25 and 58 years, 50% female, associated with the Research Group "Food Quality and Safety" of the Universidad Miguel Hernández de Elche (Orihuela, Alicante, Spain), with more than 1000 h of testing experience with fruits, vegetables, and beverages, were selected to participate in this study [22-24].

Liquors were assessed using a flavor profile method. Panelists discussed the main attributes of liquors involved in the visual, olfactory and gustative phases, during 3 preliminary orientation sessions, each lasting 45 min. For the evaluation, samples were served in glasses, odor-free, and covered by glass caps. Samples were served at room temperature that was fixed at 20 ºC in a testing room with a combination of natural and fluorescent light.

The panel after orientation sessions, agreed to evaluate the following attributes: (i) *appearance*: color; (ii) *odor* (perception of volatile compounds with the liqueur outside the mouth): citrus, floral, fruity, and alcohol; and (iii) *flavo*r: sweet, bitter, alcohol, citrus, astringent, and aftertaste. For quantifying the intensity of the samples attributes, the panel used a numerical scale where 0 represents none and 10 extremely strong, with 0.5 increments. Three 1.5 h-sessions were held for samples evaluation, and the total number of samples under analysis was 3; all 3 samples were evaluated in each session and thus, each sample was tested in triplicate.

2.5. Sensory Analysis with Consumers' Panel

Consumer acceptance was studied in Spain, in March 2016, with 100 consumers recruited at the Orihuela campus of UMH, (ratio of men:women 58:42), of which 34% were aged 18-24, 21% 25-35, 43% 36-45, and 2% 46-55 years old. All of them drank any kind of liquors at least twice *per* week, had no diet restrictions or allergies and were willing to taste the samples.

Samples were served under the same preparation conditions described above in the section on sensory evaluation with trained panel.

In each questionnaire, consumers were asked using a 9-point hedonic scale (1 = dislike extremely and 9 = like extremely) about their overall liking of the sample and their satisfaction degree for the main liquors attributes: color, citrus (odor and flavor), sweet, bitter, alcohol (flavor), astringent, and aftertaste. The intensity of key attributes (color, citrus odor and flavor, sweet, bitter, alcohol, astringent, and aftertaste) was qualitatively evaluated using JAR (Just-About-Right) questions [25]. Additionally, consumers were asked about what characteristics must have an "ideal" liquor (sweet, natural flavor, herbal flavor, natural color, bitter, citrus flavor, high alcohol and refreshing).

2.8. Statistical Analysis

Results were subjected to one-way analysis of variance (ANOVA) first, and later date were also subjected to Tukey's multiple-range test to compare the means. Differences were considered statistically significant at $p<0.05$. All statistical analyses were performed using StatGraphics Plus 5.0 software (Manugistics, Inc., Rockville, MD).

Table 1. Volatile compounds (% of total area of identified compounds), retention time (min), retention indexes, and sensory descriptors of citrus liquors (*C = "Clemenules"; VL = "Valencia Late"; S = "Sanguinely"*).

Compound	Chemical Group	RT (min)	Retention indexes		ANOVA[†]	Area (%)			Sensory descriptors
			Exp.	Lit.		C	VL	S	
1,1-Diethoxy-ethane	Other	3.34	889	886	NS	0.10	0.13	0.04	Herbal
α-Pinene	Monoterpene	5.78	1016	1014	*	0.18 b	0.19 b	6.64 a	Woody
(+)-3-Carene	Monoterpene	9.26	1142	1151	NS	0.05	0.15	0.52	Lemon
α-Phellandrene	Monoterpene	9.79	1160	1165	NS	0.14	0.13	0.14	Minty, herbaceous
Myrcene	Monoterpene	9.89	1163	1161	*	1.43 b	1.30 b	2.61 a	Grape, fruity, peach
α-Terpinene	Monoterpene	10.25	1176	1183	NS	0.17	0.12	0.75	Berry, citrus, vegetable
Limonene	Monoterpene	10.99	1200	1196	***	53.9 b	44.8 b	78.2 a	Lemon, orange, citrus
γ-Terpinene	Monoterpene	12.31	1244	1243	NS	0.29	0.23	0.05	Citrus, herbaceous
E-*β*-Ocimene	Monoterpene	12.58	1253	1250	NS	0.45	0.07	0.06	Citrus, green
p-Cymene	Monoterpene	13.1	1270	1273	NS	0.03	0.02	0.03	Citrus
Terpinolene	Monoterpene	13.44	1282	1282	NS	0.13	0.11	0.26	Plastic
Octanal	Aldehyde	13.71	1290	1288	*	0.26 b	3.26 a	0.24 b	Honey, fruity, citrus
E-Sabinene hydrate[¥]	Monoterpenoid	16.55	1386	*na*	NS	0.76	0.85	0.23	Woody
Nonanal	Aldehyde	16.82	1395	1398	NS	0.36	0.95	0.12	Citrus, fruity, vegetable
1-Octen-3-ol	Other	18.13	1439	1444	NS	0.60	1.04	0.13	Earthy, vegetable
1-Tetradecene[¥]	Other	18.32	1446	*na*	NS	1.31	0.73	0.14	*nf*
α-Cubebene	Sesquiterpene	18.68	1458	1458	NS	0.12	0.07	0.02	Herbal
α-Copaene	Sesquiterpene	19.71	1492	1493	NS	1.17	0.85	0.26	Honey, woody
Decanal	Aldehyde	20.01	1502	1505	*	7.63 ab	11.5 a	1.22 b	Citrus, waxy, floral
Linalool	Monoterpenoid	21.39	1548	1552	*	2.88 a	1.65 ab	0.56 b	Citrus, floral
Aromadendrene	Sesquiterpene	22.70	1592	1589	NS	0.78	0.59	0.12	Woody
E-Caryophyllene	Sesquiterpene	22.88	1598	1599	NS	0.42	1.00	0.30	Spicy, woody
4-Terpineol	Monoterpenoid	23.00	1602	1602	NS	1.76	1.78	0.30	Lilac
α-Humulene	Sesquiterpene	25.01	1670	1675	NS	1.60	0.66	0.15	Woody
Decyl acetate	Ester	25.39	1683	1692	NS	0.13	0.38	0.08	Orange, pineapple, rose
α-Gurjunene[¥]	Sesquiterpene	25.603	1690	*na*	NS	1.09	0.99	0.26	Woody, balsamic
Dodecanal	Aldehyde	26.28	1714	1709	NS	1.34	2.34	0.21	Herbaceous, floral
Valencene	Sesquiterpene	26.52	1722	1722	*	11.7 ab	17.3 a	5.00 b	Orange
Carvone	Sesquiterpene	26.87	1734	1738	NS	0.76	1.05	0.24	Herbaceous
Farnesene	Sesquiterpene	27.38	1752	1753	NS	1.47	1.07	0.10	Apple, lime, lavender
δ-Cadinene	Sesquiterpene	27.621	1760	1763	*	5.85 a	4.20 a	0.89 b	Herbal
Ethyl laureate	Ester	30.04	1846	1851	NS	0.52	0.24	0.05	Sweet, waxy, floral
Phenylethyl alcohol	Others	31.89	1912	1916	NS	0.31	0.04	0.02	Sweet, floral, fresh
Ethyl myristate	Ester	35.55	2052	2049	NS	0.10	0.04	0.01	Honey, fatty
Ethyl palmitate	Ester	40.66	2256	2255	NS	0.24	0.22	0.06	Waxy

*[†]NS = not significant at p> 0.05; *, and ***, significant at p< 0.05 and 0.001 respectively. [‡]Values (mean of 3 replications) followed by the same letter, within the same row, were not significantly different (p>0.05), Tukey's least significant difference test. [¥] Tentatively identified; Exp.: Experimental; Lit.: Literature; na: not available; nf: not found*

3. Results and Discussion

3.1. Volatile Compounds

Thirty-five compounds were isolated and identified (3 of them were only tentatively identified) in the volatile profile of citrus liquors (Table 1). The relatively high number of volatile compounds found in the headspace of this product is indicative of its complex odor and aroma intensity, which makes this liquor very attractive for consumers. Limonene was the main compound in citrus liquors obtained from "Valencia Late" orange and mandarin orange (~45 and ~54% of total area of identified compounds, respectively) followed by valencene (~17 and ~12%), and decanal (~12 and ~8%). In case of liquors obtained from "Sanguinely" oranges, the main compound was limonene with a ~78% of total area of identified compounds, followed by α-pinene (~7%), and valencene (~5%). All of the mainly compounds obtained in this liquors are directly related to sensory descriptors associated to "citrus" odors and flavors, except α-pinene (woody descriptor) that was found in higher proportion in case of "Sanguinely" liquor, but it is also typical of the aroma profile of citrus products (Table 1).

In addition to these main compounds, there were significantly differences ($p<0.05$) in other four volatile compounds: myrcene, octanal, linalool, and δ-cadinene. These volatile compounds are related to fruity and floral sensory descriptors, and, in general, their contents were higher in the "Clemenules" and "Valencia Late" liquors as compared to the "Sanguinely" one (e.g., the valencene contents were ~12, ~17, and ~5%, respectively), with the exception of myrcene. If volatile compounds are grouped according to their chemical family, the monoterpenes were the main family found in the liquors under analysis, followed by sesquiterpenes in all three products (Figure 1). The liquor that obtained the highest concentration of monoterpenes was that prepared using "Sanguinely" oranges (~90%), followed by those of "Clemenules" and "Valencia Late" (~57 and ~47% respectively).

Monoterpenes are responsible for fruity and floral flavors [26]. In case of sesquiterpenes, the "Sanguinely" liquor had the lowest concentration (~7%) compared to the "Clemenules" and "Valencia Late" spirits (~25 and ~28%).

These compounds are characterized by low polarity values and, therefore, are present at significant concentrations in pulp and peel, and can be a good indicator about the amount of essential oil [27]. A new flavor index has been proposed to assess the quality of citrus juices, but that can be also applied to other citrus products, such as liquors [27]. This index took values of 81%, 75%, and 38%, for "Valencia Late", "Clemenules", and "Sanguinely" liquors, respectively, indicating that the liquor of "Sanguinely" seems of lower aromatic quality, but this hypothesis must be backed up by further data on descriptive sensory analysis and especially consumer studies.

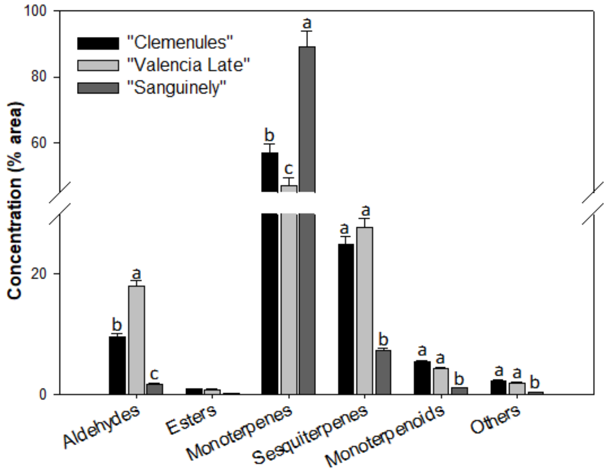

Figure 1. Total concentration (% of total area of identified compounds) of each chemical family of volatile compounds in citrus liquors.

3.2. Total Polyphenols and Antioxidant Activity

Three antioxidants methods ($ABTS^+$, $DPPH^•$, and FRAP) were used to properly evaluate the antioxidant activity of the citrus liquors under study. This variety of methods is due to the fact that none of them is able to fully characterize the total antioxidant capacity of a food, including alcoholic beverages. As it can be seen in Table 2, there were no statistically significant differences in the $DPPH^•$ values. In case of $ABTS^+$ and FRAP, the antioxidant activity took the highest values in the "Sanguinely" liquor (4.77 and 4.90 mm Trolox/mL for $ABTS^+$ and FRAP, respectively) (Table 2).

On the other hand, the total polyphenol content (TPC) was determinate using the Folin–Ciocalteu colorimetric method. The "Sanguinely" liquor was the one with the highest TPC value (557 mg GAE/mL), while the "Clemenules" liquor had the lowest TPC value (446 mg GAE/mL).

Table 2. Antioxidant activity (mmol Trolox/mL) and total polyphenols content (mg GAE/mL) of citrus liquors

Orange cultivar	Antioxidant activity (mmol Trolox/mL)			Total polyphenol content (mg GAE/mL)
	$ABTS^+$	$DPPH^•$	FRAP	
ANOVA test[†]				
	*	NS	**	*
Tukey's multiple range test[‡]				
"Clemenules"	3.88 b	2.75	4.43 b	446 b
"Valencia Late"	3.96 b	2.51	4.17 b	513 ab
"Sanguinely"	4.77 a	2.38	4.90 a	557 a

*[†]NS = not significant at $p> 0.05$; *, and **, significant at $p< 0.05$, and 0.01, respectively. [‡]Values (mean of 3 replications) followed by the same letter, within the same column, were not significantly different ($p>0.05$), Tukey's least significant difference test.*

In this particular case, the $DPPH^•$ method seemed less sensitive than the $ABTS^+$ and FRAP assays to the changes observed in the antioxidant activity of citrus liquors; for instance, values in the $DPPH^•$ method varied in a narrow range, 2.38–2.75 mmol Trolox/mL (range of 0.37 units), while the range in the ABTS$^+$ and FRAP methods were of 0.89 and 0.73 units, respectively (Table 2).

Several studies have also shown that total phenols determined by Folin–Ciocalteu method can be positively correlated to the antioxidant activity determined by different methods (*e.g.* ABTS$^+$ and DPPH$^\bullet$ assays) [28]. Proteggente, Saija [29] analyzed the phenolic composition and the total activity antioxidant of fresh Sicilian orange juice from pigmented and non-pigmented varieties of orange (*Citrus sinensis* L., Osbeck). They found that concentrations of anthocyanins and hydroxycinnamic acids were highly correlated with the total activity antioxidant values, while ascorbic acid seemed to play a minor role. Their observations were consistent with a previous report in which the antioxidant action of similar varieties was ascribed to the phenolic content [30]. Besides, Ruby-Figueroa, Cassano [31] assessing the antioxidant capacity of orange liquor, obtained high values which were positively correlated with the levels of phenolic compounds in the samples analyzed.

In the current study, positive correlations were also observed among TPC and the antioxidant activity measured by the ABTS$^+$ (R^2=0.7166), indicating that it is reasonable to think that most of the antioxidant activity of citrus liquors came from their TPC. The high values of TPC in the citrus liquors (>445 mg GAE/mL) in comparison with other types of fruit liquors (apple, persimmon, fig, cherry, passionfruit, pineapple, mango or banana spirits (all of them with TPC contents lower than 445 mg GAE/mL) justified the promising potential of these new alcoholic beverages [32].

3.3. Sensory Analysis with Trained Panel

This study is the first one reporting data on the descriptive analysis of Spanish citrus liquors. Table 3 shows the sensory profiles of the three liquors under study and the list and explanation of the main sensory attributes used in the description of the products. Sensory data proved that the citrus liquors studied had appropriate sensory characteristics (Table 3) based on recognizable intensities of key citrus notes in the liquors, such as color, citric odor and taste, astringency, and aftertaste, which should lead to good consumer acceptance.

The trained panel reported that the “Sanguinely” liquor had the highest color intensity (6.2, 4.4, and 2.0 respectively) because this fruit is pigmented. However, citrus and floral odor and flavor notes took higher values in the “Clemenules” and “Valencia Late” liquors as compared to the “Sanguinely” one. The intensity of the citrus notes is linked to the contents of odor-active compounds, such as sesquiterpenes, and their content are depended on citrus cultivar, agricultural practices, and maturity index, among other factors [23, 33]. A similar trend to that previously described for the citrus odor and flavor was also found in the astringency of the liquors, with the highest intensities being found in the “Clemenules” and “Valencia Late” liquors (Table 3).

Table 3. Visual odor, and flavor attributes, definitions and results of the descriptive analysis of citrus liquors. (*C = “Clemenules”; VL = “Valencia Late”; S = “Sanguinely”*)

Attribute	Definition	References and intensity	ANOVA[†]	C	VL	S
Visual						
Color	Visual evaluation of the color intensity of the sample	Pantone 137C = 1 Pantone 1595C = 8	**	4.4 b[‡]	2.0 c	6.2 a
Odor						
Citrus	Odor characteristic of citrus such as orange, lemon, etc.	Limonene (200 mg/L) = 6.0	*	7.0 a	7.4 a	5.0 b
Floral	Heavy aromatic odor of a combination of flowers	Geraniol (1000 mg/L) = 5.0	*	5.2 a	5.4 a	3.8 b
Fruity	A odor blend which is sweet and reminiscent of a variety of fruits	*E*-2-Hexenal (10 g/L) = 5.0	NS	4.4	4.0	3.8
Alcohol	Perception of ethanol with the liquor outside the mouth	Ethanol (30 % v/v) = 6.0	NS	5.4	5.0	5.2
Flavor						
Sweet	The taste stimulated by sugars, such as sucrose	Sucrose solution 100 g/L = 5.0	NS	5.3	4.8	5.0
Bitter	The taste stimulated by substances such as quinine or caffeine	Caffeine solution 0.5 g/L = 3.0	NS	4.0	3.8	4.2
Alcohol	Perception of ethanol with the liquor in the mouth	Ethanol (30 % v/v) = 8.0	NS	6.2	6.5	6.4
Citrus	Flavor characteristic of citrus such as orange, lemon, etc.	Limonene (200 mg/L) = 6.0	**	6.8 a	7.0 a	6.0 b
Astringent	The complex of puckering, drying, shrinking sensations in the oral cavity	Alum solution 0.75 g/L = 4.0	*	3.5 a	3.8 a	2.0 b
Aftertaste	Time in which the characteristic flavor or the liquor remains in the mouth after swallowing the sample	5 s = 1 12 s = 6	*	5.4 a	5.2 a	4.0 b

*[†]NS = not significant at p> 0.05; *, and **, significant at p<0.05, and 0.01, respectively. [‡]Values (mean of 3 replications) followed by the same letter, within the same row, were not significantly different (p> 0.05), Tukey's least significant difference test.*

In summary, it can be state that the "Sanguinely" liquor had the lowest intensities of citrus (odor and flavor), floral (odor), astringent, and aftertaste attributes. These results showed that "Clemenules" and "Valencia Late" liquors, although had the lowest content of monoterpenes, had the highest contents of the key chemical families (sesquiterpenes, aldehydes, and monoterpenoids), leading to a more complex and structured odor and flavor than that of the "Sanguinely" liquor, although its color was more intense.

3.4. Sensory Analysis with Consumers' Panel

After the analysis of the liquors by a trained sensory panel, a consumer study was also used. The consumer study evaluated consumers' satisfaction degree on the product as a whole and on specific key sensory attributes. Besides, the intensity of the main attributes was qualitatively evaluated using JAR questions. It is important to mention that the floral odor attribute was removed, because it was considered to be too complex to be evaluated by consumers (untrained panelists).

The results obtained are shown in Table 4, and significant differences in five of the studied attributes were obtained: color, citrus odor, citrus flavor, aftertaste, and overall satisfaction degree. The highest values of the satisfaction degree were found in the "Clemenules" liquor for color and citrus odor, and in "Valencia Late" for citrus flavor, aftertaste, and especially global. These affective results agreed quite well with descriptive sensory data and also with the instrumental data (volatile profiles), in which "Sanguinely" liquor had the lowest contents of key volatile compounds and the lowest intensities of key sensory attributes.

The JAR questions indicated that the intensities of most of the attributes were close to the optimal values in the "Clemenules" and "Valencia Late" products, although the color of the "Valencia Late" liquor was considered below the optimal, and needs to be improved.

In addition, 53% of consumers stated that an "ideal liquor" must have citric flavor, 47% said that it had to be refreshing, and 45% said that it had to be sweet. These attributes had proper intensities, as can be seen in the descriptive profiles of the products (Table 3), especially in the "Clemenules" and "Valencia Late" spirits.

4. Conclusions

In this study, three citrus liquors were obtained by macerating the peel of three orange varieties ("Clemenules", "Valencia Late", and "Sanguinely") obtained as by-products of the Spanish citrus industry. These products had a great aromatic potential, characterized by having high contents of volatile compounds, mainly monoterpenes and sesquiterpenes, being the "Clemenules" and "Valencia Late" liquors those with more complex aromatic profile (volatile compounds, and sensory odor and flavor). On the other hand, the liquor obtained using the peel of "Sanguinely" pigmented oranges had the highest content of antioxidant activity and total polyphenol content. In a consumers' study, "Clemenules" and "Valencia Late" spirits were those obtaining the higher satisfaction degree values and also appropriate (close to the consumers' optimum) intensities of most of the sensory attributes, being these liquors the closest ones to what consumers defined as an "ideal liquor". The experimental results obtained in this research suggested that these new citrus alcoholic beverages have a promising future and are another alternative to the citrus peel by-products. After checking that "Clemenules" and "Valencia Late" showed good properties and consumers' acceptance it is the time to compare their behavior with those of commercial available products, such as *limoncello*.

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Table 4. Satisfaction degree of Spanish consumers on the main sensory attributes of citrus liquors, using a 9-point hedonic scale.

Orange cultivar	Color	Citrus (o)	Citrus (f)	Sweet	Bitter	Alcohol	Astringent	Aftertaste	Global
					ANOVA Test†				
	***	***	**	NS	NS	NS	NS	***	*
					Tukey's Multiple Range Test‡				
"Clemenules"	6.0 a	6.1 a	4.9 ab	5.0	4.8	4.7	5.2	5.7 a	5.4 ab
"Valencia Late"	4.2 b	5.8 a	5.5 a	5.0	5.0	5.0	5.4	6.0 a	5.7 a
"Sanguinely"	5.7 a	4.7 b	4.1 b	4.4	4.6	4.3	4.9	4.7 b	4.7 b

*†NS = not significant at $p > 0.05$; *, ** and ***, significant at $p < 0.05$, 0.01, and 0.001, respectively. ‡Values (mean of 3 replications) followed by the same letter, within the same column, were not significantly different ($p > 0.05$), Tukey's least significant difference test.*

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