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Comparison on sensory profile, volatile composition and consumer's acceptance for PDO or non-PDO tigernut (Cyperus esculentus L.) milk

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ABSTRACT

Cyperus esculentus tubers are the raw material to prepare tigernut milk (*horchata*), which can be marketed under Protected Designation of Origin (PDO) "C *hufa de Valencia*". The aim of this study was to characterize commercial tigernut milks and compare PDO and non-PDO products. The following aspects were studied: (i) volatile profile, (ii) descriptive sensory profile, (iii) consumer satisfaction degree. The key volatile compounds were limonene, benzaldehyde, linalool and *m*-methoxyanisole. Principal component analysis indicated a mix of PDO and non-PDO samples in the groups formed. The highest consumer satisfactions were observed for 2 PDO samples. Penalty analysis showed that 80% of non-PDO samples needed improvements, while this percentage drastically decreased to 40% for PDO samples. The online study proved that a lot of people drink *horchata*, less people know the PDO *Chufa de Valencia* and even less people consume the protected product consciously. In conclusion, there was not a clear difference among protected and non-protected tigernut milks respect to volatile compounds but there were differences in the degree of consumer preference. So, it is clear that a lack of knowledge regarding the product and its PDO exists and needs attention.

1. Introduction

The tigernut plant (*Cyperus esculentus* L. var. *Sativus* Boeck) produces rhizomes from the base with small spherical tubers (from 8 to 16 mm diameter) (de Vries & Femke, 1991). It can be easily found in wet marshes and edges of streams and ponds (Takhatajah, 1992, pp. 596–610). This plant is commonly known as earth almond, tigernut, *chufa*, yellow nut sedge and zulu nuts (Umerle, Okafor, & Uka, 1997). Nowadays, it is cultivated in Africa and Mediterranean countries such as Burkina Faso, Egypt, Ghana, Ivory Coast, Niger, Nigeria, Mali, Senegal, Spain, and Turkey on a small scale (Coşkuner, Ercan, Karababa, & Nazlıcan, 2002; Djomdi, Ejoh, & Ndjouenkeu, 2007; Matos, Cavalcanti, & Parente, 2008). In Spain, *chufa* cultivation area is around 400 ha, with an average yield of 18,000 to 19,000 kg ha^{-1} and a total production of ~8000 t (MAPA - Ministerio de Agricultura, 2017). The major Spanish tigernut production area is located in Valencia, where it is typified and commercialized as "*Chufa de Valencia*" under a Protected Designation of Origin, PDO (http://www.chufadevalencia.org/). The PDO covers 16 municipalities, being the main ones Valencia and Alboraia (CRDO, 2019).

"*Horchata de chufa*" (tigernut milk), commonly known as "*horchata*", is a traditional Spanish vegetable non-alcoholic beverage, similar in appearance to milk, obtained from *chufa*/tigernut tubers (Real Decreto 1338/1988, 1988). The tigernut milk preparation process includes the following unit operations: washing, selection (eliminating defective tubers), disinfection, rehydration, crushing, pressing, sifting and cooling (Reglamentación, 1988). Due to the high water activity of tigernut tubers, they must be intensively disinfected before starting their processing because of their high microbiological contamination to guarantee final product safety (Roselló-Soto et al., 2018).

More than 50,000,000 L of this beverage are produced annually. This implies that more *chufa* is needed than what is produced in the area covered by the PDO. Consequently, tigernuts are imported from third countries, mainly as dried *chufa* from Burkina Faso, Ghana, Mali, Niger

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Table 1
Description of the tigernut milk (*horchata*) samples under study.

Samples	Name	Ingredients	Fat (g/L)	Protected by PDO
A	*Horchata de chufa*	Water, tigernut (10%), sugar, emulsifiers (citric esters of mono and diglycerides fatty acids and mono and diglycerides of fatty acids), milk proteins and aromas of cinnamon and lemon	28.0	Yes
B	*Horchata de chufa 100% de Valencia*	Water, tigernut, sugar, emulsifier (E−472c), stabilizer (E−407), antioxidant (E−301) and aroma	28.0	Yes
C	*Horchata maestro horchatero*	Water, tigernut, sugar, emulsifier (E−472c), stabilizer (E−407), antioxidant (E−301) and aroma	38.0	Yes
D	*Horchata de chufa de Valencia*	Water, tigernut, sugar, emulsifier (E−472c), milk protein, antioxidant (E−300) and aromas	26.0	Yes
E	*Horchata de chufa*	Water, tigernut (16%), sugar, milk protein, stabilizers (E−472c and E−471), acidity corrector (E−331), lemon and cinnamon aromas	30.0	Yes
F	*Horchata de chufa*	Water, tigernut, sugar, milk protein, stabilizers (E−472c and E−471), acidity corrector (E−331), lemon and cinnamon aromas	22.0	No
G	*Horchata de chufa*	Water, tigernut, sugar, emulsifier (E−472c), milk proteins, stabilizer (E−407), natural aromas of lemon and cinnamon	25.0	No
H	*Horchata premium de chufa*	Water, tigernut (12.8%), sugar, emulsifier (E−472c), milk proteins, acidity corrector (E−331iii), stabilizer (E−407), natural aromas of lemon and cinnamon	30.0	No
I	*Horchata de chufa*	Water, tigernut, sugar, emulsifier (E−472c), milk proteins, antioxidant (E−300) and aromas	20.0	No
J	*Horchata de chufa*	Water, tigernut (9%), sugar, emulsifiers (citric esters of mono and diglycerides of fatty acids, mono and diglycerides of fatty acids), milk proteins and aromas of cinnamon and lemon	22.0	No

Table 2
Lexicon used for descriptive sensory analysis of tigernut milk (*horchata de chufa*).

Attributes	Definition	References and intensities
Flavor		
Sweet	The fundamental taste factor associated with a sucrose solution	Sucrose solution 40 g/L = 2.5; sucrose solution 80 g/L = 5.0; sucrose solution 160 g/L = 9.5
Bitter	The taste stimulated by substances such as quinine or caffeine	Caffeine solution 0.5 g/L = 2.5; caffeine solution 0.8 g/L = 4.0
Sour	The taste stimulated by acids, such as citric and malic	Tartaric acid solution 0.5 g/L = 2.5; tartaric acid solution 0.8 g/L = 4.0; tartaric acid solution 2.0 g/L = 9.5
Astringent	The shrinking or puckering of the tongue surface caused by substances such as tannins or alum	Minute Maid® Orange juice = 2.0
Tigernut ID	Fundamental taste sensation associated to tigernut	25 g of grinded *Cyperus esc.* = 4.0; 100 g of grinded *Cyperus esc.* = 10
Earthy	Green and herbaceous aroma associated with tigernut	250 g of grinded *Cyperus esc.* + 100 mL H_2O = 4.0; 100 g of grinded *Cyperus esc.* + 25 mL H_2O = 10
Vegetal	Fresh, green, slightly sour aromatics associated with green vegetables, newly cut vines, snap peas	Kroger lima beans (canned) = 3.0; Small sprig fresh parsley = 7.0 (aroma); Fresh parsley = 10.0
Tigernut milk ID	Fundamental taste sensation associated to tigernut milk	100 mL tigernut milk + 150 mL water = 4.0; 250 mL tigernut milk = 10
Nut	Spices notes	25 g of grinded *Juglans regia* nut + 100 mL H_2O = 4; 100 g of grinded *Juglans regia nut* + 25 H_2O = 10
Cinnamon	Spices notes	25 g of grinded *Cinnamomum Cassia* bark + 100 mL H_2O = 4; 100 g of grinded *Cinnamomum Cassia* bark + 25 H_2O = 10
Lemon	Characteristic flavor to lemon	Lemonade (Country time) = 4; Lemon juice = 10
Aftertaste	Time in which the specific flavor of the product flavor remains in the mouth after swallowing the sample	10 s = 2.0; 30 s = 8.0
Texture		
Viscosity	The measure of flow as the product moves on the tongue when pressed between the tongue and the palate (2.46 mL of product).	Water = 1.0; Light Cream (Land O'Lakes) = 2.2; Pancake syrup (Vermont Maid) = 6.8
Mouthcoating	The amount of film left on the mouth surfaces	Olive oil (Carbonell) = 10

and Nigeria.

Consumer demand for "*horchata de chufa*" is growing (CRDO, 2019) due to its high nutritional quality and very distinctive organoleptic characteristics (Martín-Esparza & González-Martínez, 2016); with its consumption being concentrated during summer when it is served very cold or even frozen (Codina, Trujillo, & Ferragut, 2016). Its consumption is related to prevention of heart attacks and thrombosis and to blood circulation promotion (Chukwuma, Obioma, & Ononogbu, 2010; Sánchez-Zapata, Fernández-López, & Pérez-Alvarez, 2012), and decreased colon cancer risk (Adejuyitan, Otunola, Akande, Bolarinwa, & Oladokun, 2009). However, it is necessary to bear in mind its high sucrose content (minimum 100 g/L), as established by the Spanish legislation (Real Decreto 1338/1988, 1988).

Considering all the above, the aim of this study was to characterize commercial tigernut milk samples and compare PDO and non-PDO products. To reach this aim, the following parameters were studied: (i) volatile profile (responsible for odor, aroma and flavor notes), (ii) descriptive sensory profile (using a trained panel) and (iii) consumer satisfaction degree (using a consumer panel).

2. Material and methods

2.1. Tiger nut milk samples

For this study, available commercial samples of tigernut milks (n = 10) were acquired at supermarkets in Alicante and Murcia, only UHT

Table 3
Identification and descriptors of volatile compound found in the tigernut milk samples under study.

Compound	C. F.	RT	KI (LIT)	KI (EXP)	*p*-value	ANOVA[†]	*Analytical pooled SD*	Concentration (μg/L *horchata*)										*p*-value	ANOVA	*SD*	Mean (μg/L)		Descriptors
								A	B	C	D	E	F	G	H	I	J				PDO[¥]	Non-PDO	
α-Pinene	T	4.574	932	936	0.0000	***	0.3	1.4 de[¥]	1.7 d	0.0 g	15.7 b	1.1 def	2.7 c	3.0 c	0.7 efg	41.9 a	0.3 fg	0.2213	NS	12.6	4.0	9.7	Woody
Benzaldehyde	A	5.393	980	975	0.0000	***	3.1	152 c	201 b	392 a	39.4 e	0.9 g	10.3 f	0.4 g	7.3 fg	143 c	57.3 d	0.0079	**	108.5	157 a	43.6 b	Almond, fruity, nut
Sabinene	T	5.498	980	981	0.0000	***	0.9	1.6 e	0.7 g	2.8 d	16.9 c	0.4 i	71.6 b	0.5 h	0.4 i	131 a	0.9 f	0.0161	*	38.9	4.5 b	40.8 a	Woody, citrus, green
β-Pinene	T	5.625	990	985	0.0000	***	0.1	6.7 a	2.5 c	1.9 d	0.3 h	0.9 e	0.8 e	0.6 f	0.4 f	0.0 i	5.0 b	0.1682	NS	2.1	2.5	1.4	Woody
Myrcene	T	5.678	998	990	0.0000	***	0.8	10.4 d	0.6 e	8.5 d	117 a	25.0 b	1.5 e	13.4 c	2.0 e	0.6 e	1.5 e	0.0203	*	31.7	32.2 a	3.8 b	Herbaceous, woody, rose
p-Cymene	T	6.646	1034	1029	0.0000	***	0.4	25.7 b	1.0 f	5.2 e	20.9 c	0.9 f	55.1 a	0.4 f	0.8 f	19.2 d	1.0 f	0.4758	NS	17.3	10.7	15.3	Fresh, citrus, woody
Limonene	T	6.759	1036	1033	0.0000	***	8.9	542 c	232 e	149 f	514 d	174 f	871 a	92.6 g	96.1 g	734 b	62.0 h	0.6457	NS	288.3	322	371	Lemon, orange, citrus
γ-Terpinene	T	7.514	1059	1062	0.0000	***	1.5	74.1 c	3.6 hi	7.4 gh	64.6 d	34.9 e	161 a	11.6 f	8.5 fg	107 b	0.5i	0.2811	NS	51.8	36.9	57.7	Woody, citrus, lemon
Linalool	T	8.766	1110	1107	0.0000	***	1.6	46.4 c	55.9 b	26.8 f	59.8 b	48.5 c	58.2 b	33.6 e	39.4 d	209 a	2.2 g	0.2939	NS	53.8	47.8	68.4	Lemon, orange, floral
Nonanal	A	8.856	1107	1109	0.0000	***	1.1	56.8 b	30.7 f	31.9 f	14.5 g	45.0 d	52.7 c	36.3 e	29.8 f	7.2 h	130 a	0.2044	NS	32.5	35.7	51.2	Rose, lemon, citrus
m-Methoxyanisole	E	11.456	1181	1179	0.0000	***	5.0	1.0 c	1.4 c	785 a	63.4 b	0.5 c	1.5 c	0.4 c	1.0 c	0.9 c	1.0 c	0.0494	*	225.7	170 a	1.0 b	Fruity, nutmeg
Terpinen-4-ol	T	11.799	1191	1191	0.0000	***	0.3	4.9 c	0.4 e	3.2 d	16.9 b	3.0 d	2.2 d	0.2 e	0.4 e	52.8 a	0.3 e	0.3493	NS	15.8	5.7	11.1	Woody, citrus
α-Terpineol + methyl salicylate	T + e	12.382	1207	1206	0.0000	***	0.6	2.7 fg	1.6 gh	88.1 a	12.7 b	6.5 d	4.0 ef	5.8 d	5.1 de	10.4 c	0.2 h	0.0548	NS	24.4	22.3	5.1	Lilac
	Total identified volatile compounds (μg/L *horchata)*				0.0000	***	18.2	925 c	533 d	1501 a	956 c	341 e	1292 b	198 fg	191 g	1457 a	262 ef	0.3389	NS	509.9	851	667	

C. F., Chemical Family; T, Terpene; A, Aldehyde; E, Ether; e, ester; RT, Retention Time; KI, Kovats Index; LIT, Literature; EXP, Experimental; [†]NS: not significant at $p > 0.05$; *, **, and ***, significant at $p < 0.05$, 0.01 and 0.001, respectively; Pooled SD, Pooled standard deviation; [‡]Values (mean of 3 replications) followed by the same letter, within the same row and factor, were not significantly different ($p > 0.05$). Tukey's least significant difference test. [¥]PDO, Protected Designation of Origin.

(ultra-high-temperature) samples were selected. Among these samples, 5 of them belonged to the Protected Designation of Origin (PDO) "*Chufa de Valencia*". Table 1 shows the codes and the main characteristics of samples under study. The composition of the tigernut milks was similar (fat range between 20 and 30.00 g/L). However, only 4 samples (2 PDO, 2 no PDO), out of a total of 10, showed the percentage of tigernut used. The quantities of ingredients were similar except the amount of chufa. So, it can be stated that the 10 samples under study formed a uniform set of products.

2.2. Volatile composition

The method selected to study the volatile profile of tigernut milk samples was headspace solid phase micro-extraction (HS-SPME). ~15 mL of tigernut milk together with 1.5 g of NaCl and 10 μL of internal standard (benzyl acetate) were placed in a 50 mL vial with a polypropylene cap and a PTFE/silicone septum for the extraction of the volatile fraction. The samples were equilibrated during 15 min at 40 °C on the vials, and a DVB/CAR/PDMS fiber (50/30 μm) was exposed to the sample headspace at 40 °C for 50 min. The extraction conditions (HS-SPME) were optimized to obtain a volatile profile positively correlated with sensory odor characteristics (Alonso, Vázquez-Araújo, García--Martínez, Ruiz, & Carbonell-Barrachina, 2009).

The separation and identification of the volatile compounds were performed using the GC-MS conditions previously described (Issa-Issa et al., 2020). The equipment used consisted of a gas chromatogram Shimadzu GC-17A (Shimadzu Corporation, Kyoto, Japan) coupled with a Shimadzu mass spectrometer detector (MS) QP-5050A and with a DBWax column (30 m length × 0.25 mm internal diameter × 0.5 μm thickness, J&W Scientific, Folsom, California, USA). Finally, a gas chromatograph, Shimadzu GC-2010, with a flame ionization detector (FID) was used for the quantification of the volatile composition of samples, based on the use of 10 μL of internal standard, benzyl acetate. The column and chromatographic conditions were the same as those reported previously by (Issa-Issa et al., 2020). The extraction experiments and volatile studies were run in triplicate.

2.3. Descriptive sensory analysis

A trained panel consisting of 10 highly trained panelists from the research group Food Quality and Safety Group (Universidad Miguel Hernández de Elche, UMH Orihuela, Alicante, Spain) conducted the descriptive sensory analysis in two separated sessions. The panel was selected and trained following the ISO standard 8586-1. The panel is specialized in descriptive sensory evaluation of fruit products (Cano-Lamadrid et al., 2020). Each panelist had more than 600 h of experience with different types of food products. For the present study, the panel worked during 2 orientation sessions (90 min for each one) discussing the main organoleptic characteristics of commercial tiger nut milks. The lexicon used for describing the flavor (n = 12) and texture (n = 2) attributes was based on the previously one developed by other authors (Clemente-Villalba et al., 2020) (Table 2). Lexicons were adapted for studied samples during the orientation sessions. References were chosen and prepared according to previous publications using similar attributes (Lipan et al., 2019), and then, provided to panellists. The methodology used for the descriptive sensory analysis was that previously described (Rutledge & Hudson, 1990) with some modifications. The scale ranged from 0 (no intensity) to 10 (extremely high intensity) with 0.5 increments. Each panelist tested ~25 mL of each sample, coded with 3-digit randomized numbers. Water and unsalted crackers were provided to panellists.

2.4. Affective sensory analysis (consumer panel)

A sample group of 200 consumers was recruited at UMH (Spain), and consisted of 112 men and 88 women aged between 18 and 60 years. The

Table 4
Descriptive sensory analysis of the tigernut milk samples under study, and between PDO and Non-PDO groups.

Attribute	*p*-value	ANOVA[†]	*Analytical pooled SD*	Tigernut milk										*p*-value	ANOVA	*SD*	Mean intensity	
				A	B	C	D	E	F	G	H	I	J				PDO[¥]	Non-PDO
Sweet	0.0103	*	1.7	4.5 b[‡]	5.0 ab	4.7 ab	5.5 ab	6.0 ab	6.7 ab	4.7 b	7.6 a	6.1 ab	4.6 b	0.0673	NS	0.9	5.2	6.0
Bitter	0.3490	NS	1.0	0.7	1.2	1.2	2.1	2.0	0.7	1.7	1.0	1.5	0.6	0.0052	**	0.5	2.6 a	1.5 b
Sour	0.0027	**	0,4	0.5 ab	0.2 b	0.0 b	0.2 ab	0.5 ab	1.0 a	0.1 b	0.1 b	0.0 b	0.3 ab	0.8026	NS	0.3	0.3	0.3
Astringent	0.3416	NS	0.3	0.2	0.0	0.0	0.3	0.2	0.0	0.2	0.1	0.1	0.0	0.3756	NS	0.1	0.2	0.1
Tigernut ID	0.0000	***	1.0	2.5 cd	4.3 ab	2.8 bcd	2.2 d	5.1 a	4.1 abc	1.6 d	5.3 a	5.2 a	2.0 d	0.5194	NS	1.4	3.4	3.7
Earthy	0.0000	***	1.1	2.5 bcd	3.9 abcd	2.5 bcd	2.2 cd	5.0 a	3.5 abcd	3.0 abcd	4.1 abc	4.2 ab	2.1 d	0.6370	NS	1.0	3.3	3.4
Vegetal	0.0000	***	1.1	0.0 c	2.2 b	1.6 bc	0.0 c	0.0 c	0.0 c	5.7 a	0.0 c	0.0 c	0.0 c	0.4436	NS	1.8	0.8	1.1
Tigernut milk ID	0.0000	***	0,9	2.8 de	4.2 bcd	1.7 ef	2.1 ef	5.7 ab	4.1 cd	1.2 f	5.0 abc	6.0 a	2.0 ef	0.4658	NS	1.7	3.4	3.7
Nut	0.0511	NS	0,9	0.7	0.6	0.1	1.7	0.2	0.7	0.1	0.2	1.0	0.5	0.4970	NS	0.5	0.7	0.6
Cinnamon	0.0000	***	0.4	3.0 a	0.1 b	0.0 b	0.2 b	0.0 b	2.4 a	0.0 b	0.0 b	0.3 b	0.2 b	0.7946	NS	1.1	0.7	0.6
Lemon	0.0000	***	0.3	0.1 c	0.0 c	0.0 c	0.2 c	0.0 c	2.5 a	0.0 c	0.0 c	1.0 b	0.2 c	0.0004	***	0.7	0.1 b	0.8 a
Viscosity	0.0001	***	0.9	2.2 c	2.9 abc	4.0 a	2.1 c	2.7 abc	2.6 abc	2.4 bc	3.7 ab	2.7 abc	1.5 c	0.4490	NS	0.7	2.8	2.6
Mouth coating	0.0111	*	1.1	2.5 ab	2.7 ab	2.5 ab	2.2 ab	4.0 a	3.1 ab	2.3 ab	3.7 ab	3.0 ab	2.0 b	0.7609	NS	0.6	2.8	2.9
Aftertaste	0.6744	NS	1.3	3.2	3.6	3.3	3.5	4.0	3.7	3.8	4.1	4.0	2.8	0.5453	NS	0.4	3.5	3.7

[†]NS: not significant at $p > 0.05$; *, **, and ***, significant at $p < 0.05$, 0.01 and 0.001, respectively; [‡]Values (mean of 10 replications/trained panelists) followed by the same letter, within the same row and factor, were not significantly different ($p > 0.05$). Tukey's least significant difference test. [¥]PDO, Protected Designation of Origin. Pooled SD, Pooled standard deviation.

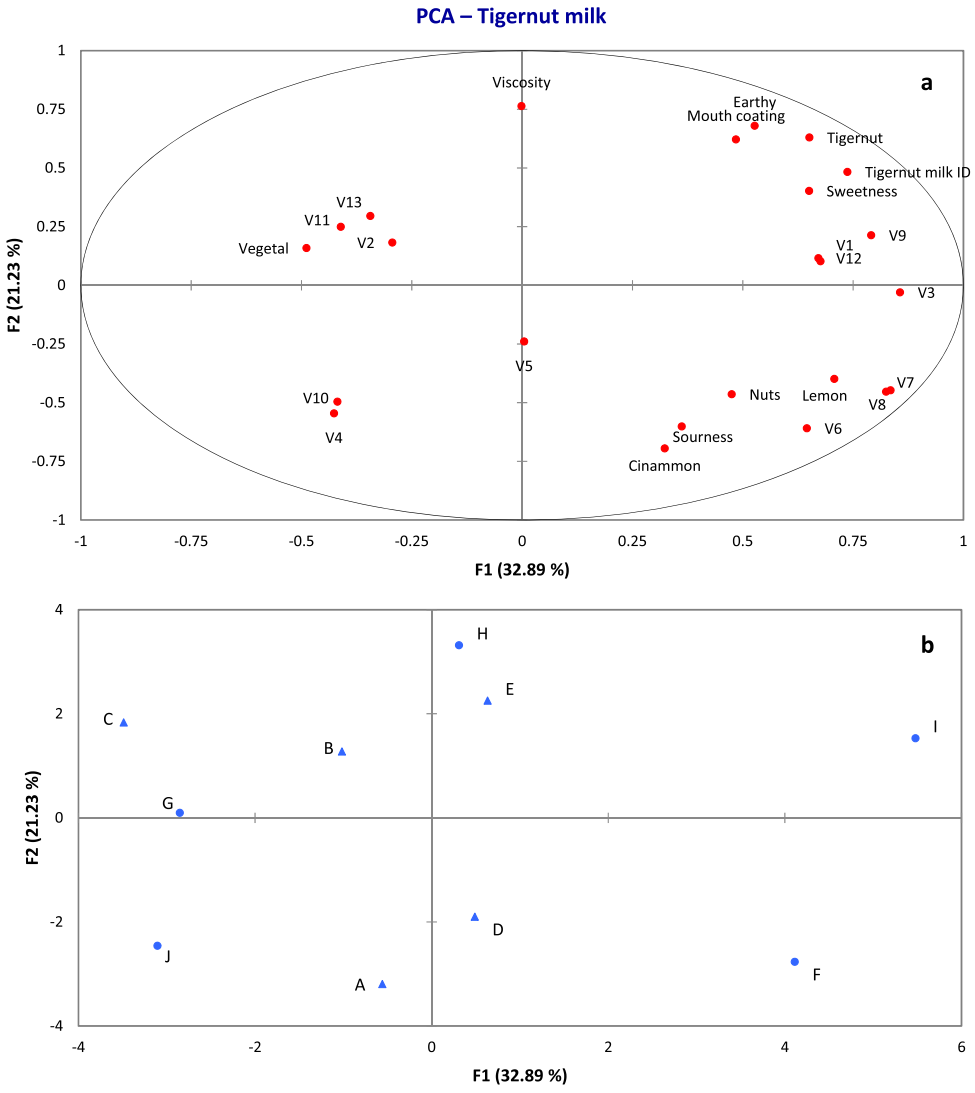

Fig. 1. Principal Component Analysis of tigetnut milk (PCA). a) volatile compounds and descriptive sensory attributes; b) samples map, Δ PDO and • non-PDO.

main requirement for their recruitment was that they consumed regularly this type of beverage. The consumer study was conducted at the UMH facilities in Orihuela, during 2 sessions of 1.5 h. In each session, consumers tested only 5 tigernut milk samples. The samples were presented according to a balanced incomplete block design (split-plot): all samples are analyzed by the same number of panelists but not all panelists study all samples. Each consumer was served ~25 mL of each sample coded with 3-digit numbers, together with the questionnaire. Water and unsalted crackers were provided to consumers between samples for palate cleansing.

Consumers were asked about their global satisfaction degree using a 9-point hedonic scale (1 = dislike extremely, 5 = neither like nor dislike, and 9 = like extremely), together with questions regarding attributes intensity using a Just About Right (JAR) scale. At the end of the session, consumers responded an online-questionnaire after sensory test (Table 1S, Supplementary material) regarding their knowledge on tigernut milks and the PDO "*Chufa de Valencia*". The questionnaire consisted of questions approaching the following topics: (i) basic knowledge; (ii) what makes tigernut milk different from other vegetal drinks; (iii) their own consumption of the product; (iv) specific knowledge about "*horchata de chufa*"; and, (v) knowledge about the PDO "*Chufa de Valencia*".

2.5. Statistical analysis

Data were subjected to two-ways-ANOVA (Factor 1: commercial tiger nut milks; and, Factor 2: tigernut protected quality label, PDO), after checking the normality and homogeneity of the variance and to Tukey's multiple-range test to compare means. This analysis was performed for descriptive sensory analysis, hedonic questions from consumers study and volatile compounds. Differences were considered statistically significant at $p < 0.05$. All statistical analyses were performed using StatGraphics Plus 5.0 software (Manugistics Inc., Rockville. USA), and XLSTAT (Microsoft Corporation, Redmond, Washington, USA).

3. Results and discussion

3.1. Volatile compounds

A total of 14 volatile compounds were isolated, identified and quantified. They were grouped into 4 chemical families: terpenes (n = 10), aldehydes (n = 2), ether(n = 1), and esters (n = 1) (Table 3). The quantification of volatile compounds showed significant differences among samples. The main volatile compounds found in all samples were

Table 5
Consumer satisfaction degree of tigernut milk samples under study, and between PDO and Non-PDO groups.

Attribute	*p*-value	ANOVA†	*Analytical pooled SD*	Tigernut milk										*p*-value	ANOVA	*SD*	Mean	
				A	B	C	D	E	F	G	H	I	J				PDO¥	Non- PDO
Color	0.0004	***	1.7	5.3 bc‡	5.9 abc	5.4 abc	6.1 ab	6.5 a	4.8 c	5.8 abc	5.9 abc	6.0 abc	5.8 abc	0.3413	NS	0.5	5.9	5.7
Appearance	0.0000	***	1.5	5.6 abc	6.1 ab	5.3 bc	6.3 ab	6.6 a	5.0 c	6.3 ab	6.0 ab	6.2 ab	6.1 ab	0.6497	NS	0.5	6.0	6.0
Tigernut milk ID	0.0000	***	2.0	4.3 d	5.5 abcd	4.3 cd	6.2 a	5.8 ab	5.5 abcd	5.2 abcd	4.6bcd	5.3 abcd	5.7 abc	0.9109	NS	0.6	5.3	5.3
Vegetal	0.0006	***	1.7	4.4 bc	5.3 ab	4.1 c	5.4 ab	5.6 a	5.2 abc	5.0 abc	4.7 abc	4.9 abc	4.9 abc	0.7501	NS	0.5	5.0	5.0
Earthy	0.0596	NS	1.5	4.9	5.3	4.7	5.4	5.5	5.5	5.1	4.8	4.9	5.4	0.9760	NS	0.3	5.2	5.2
Sweet	0.0002	***	1.9	5.1 ab	5.6 ab	4.7 b	6.0 a	5.7 ab	5.4 ab	5.3 ab	4.5 b	4.8 ab	6.0 a	0.2353	NS	0.5	5.5	5.2
Viscosity	0.0439	*	1.5	5.1 b	5.3 ab	5.3 ab	5.6 a	6.0 a	5.1 b	5.4 ab	5.4 ab	5.1 b	5.7 a	0.2647	NS	0.3	5.5	5.4
Overall liking	0.0000	***	2.0	4.3 c	5.6 ab	4.2 c	6.1 a	6.2 a	5.4 abc	5.1 abc	4.6 bc	5.1 abc	5.7 ab	0.6327	NS	0.7	5.3	5.2

†NS: not significant at $p > 0.05$; *, **, and ***, significant at $p < 0.05$, 0.01 and 0.001, respectively; ‡Values (mean of 200 replications/consumers) followed by the same letter, within the same row and factor, were not significantly different ($p > 0.05$). Tukey's least significant difference test. ¥PDO, Protected Designation of Origin. Pooled SD, Pooled standard deviation.

limonene, benzaldehyde, *m*-methoxyanisole, linalool and nonanal (Table 3), some of them are added aromas. In general, limonene was the most abundant compound (mean for all samples of 325 μg L^{-1}), and content ranging between 62 and 871 μg L^{-1} in samples J and F, respectively. It is important to highlight that both J and F belong to the non-PDO group, implying that the factor "PDO" does not play a key role in volatile composition, or at least, in the limonene content. A similar behavior was found for linalool, which content range from 2.2 to 209 μg L^{-1} in the non-PDO samples J and I, respectively. Identical situation was found for the nonanal content, with the lowest and highest values being found in the non-PDO samples I and J, respectively. Benzaldehyde ranged between 0.4 and 392 μg L^{-1} in samples G and C, respectively. The content of this aldehyde was 3 times higher in PDO samples than in non-PDO ones; showing significance difference between PDO and non-PDO groups. A similar trend was also observed for *m*-methoxyanisole, with samples C (785 μg L^{-1}) and D (63.4 μg L^{-1}), both samples under the PDO label, having a significantly higher content than the other samples (<2.0 μg L^{-1}). A previous study comparing raw tigernut and toasted tigernut identified 143 volatiles compounds by GC-MS, using a previous extraction by high-vacuum distillation (Cantalejo, 1997). In this study, limonene was also one of the predominant compounds and benzaldehyde, linalool and nonanal were key compounds. These volatile compounds have been widely reported in other products such as almonds, *Prunus dulcis* (Nawade et al., 2019), while *m*-methoxyanisole was not detected previously in tigernut products but it was found in yellow mushrooms (*Agaricale*).

As a conclusion, non-PDO tigernut milks presented the highest content of 8 of a total of 14 volatile compounds. However, PDO samples showed a higher total content of volatile compounds (4256 μg L^{-1}) compared to 3400 μg L^{-1} of the non-PDO samples.

3.2. Descriptive sensory analysis

Significant differences between the PDO and non-PDO groups were found only in bitter and lemon attributes (Table 4); with non-PDO samples being sweeter and having higher lemon intensity than the samples under the PDO "*horchata de Valencia*". However, 11 attributes showed significant differences among all studied samples. There were not statistically significant differences for bitterness, astringency and aftertaste. All samples can be considered as sweet, although the intensity of this attribute ranged between ~4.5 (samples A, C, G and J) and 7.6 (sample H). PDO samples were sweeter than the non-PDO ones (Table 4). The earthy note could be one of the most distinctive flavor attributes of this special beverage. Its intensity ranged from 2.1 (non-PDO sample J) up to 5.0 (PDO sample E). The vegetal flavor note was not found in 7 out of 10 the samples studied (3 PDO, 4 non-POD), and only reached a significant high level (5.7) in the non-PDO sample G. Surprisingly, there were samples with very low intensity of the most important sensory attribute of this type of vegetal beverage, tigernut milk ID, with only 3 samples (E, H and I) having intensities above 5.0. The low values of some of the PDO samples (A, C and D) were especially worrying. Even though all the samples contained cinnamon in their composition, only two samples (A and F) had measurable levels of this flavor. A similar situation was found for the lemon attribute, which intensity could be properly quantified in two samples (F and I).

3.3. Principal components analysis (PCA)

Principal component analyses (PCAs) was done to get a better understanding of the relationships among the 10 tigernut milk samples, using (i) volatile compounds and (ii) descriptive sensory attributes. In Fig. 1, the F1 axis explained 32.89% of the total data variance; while the axis F2 explained 21.23% of the total variance. Here are the most relevant associations:

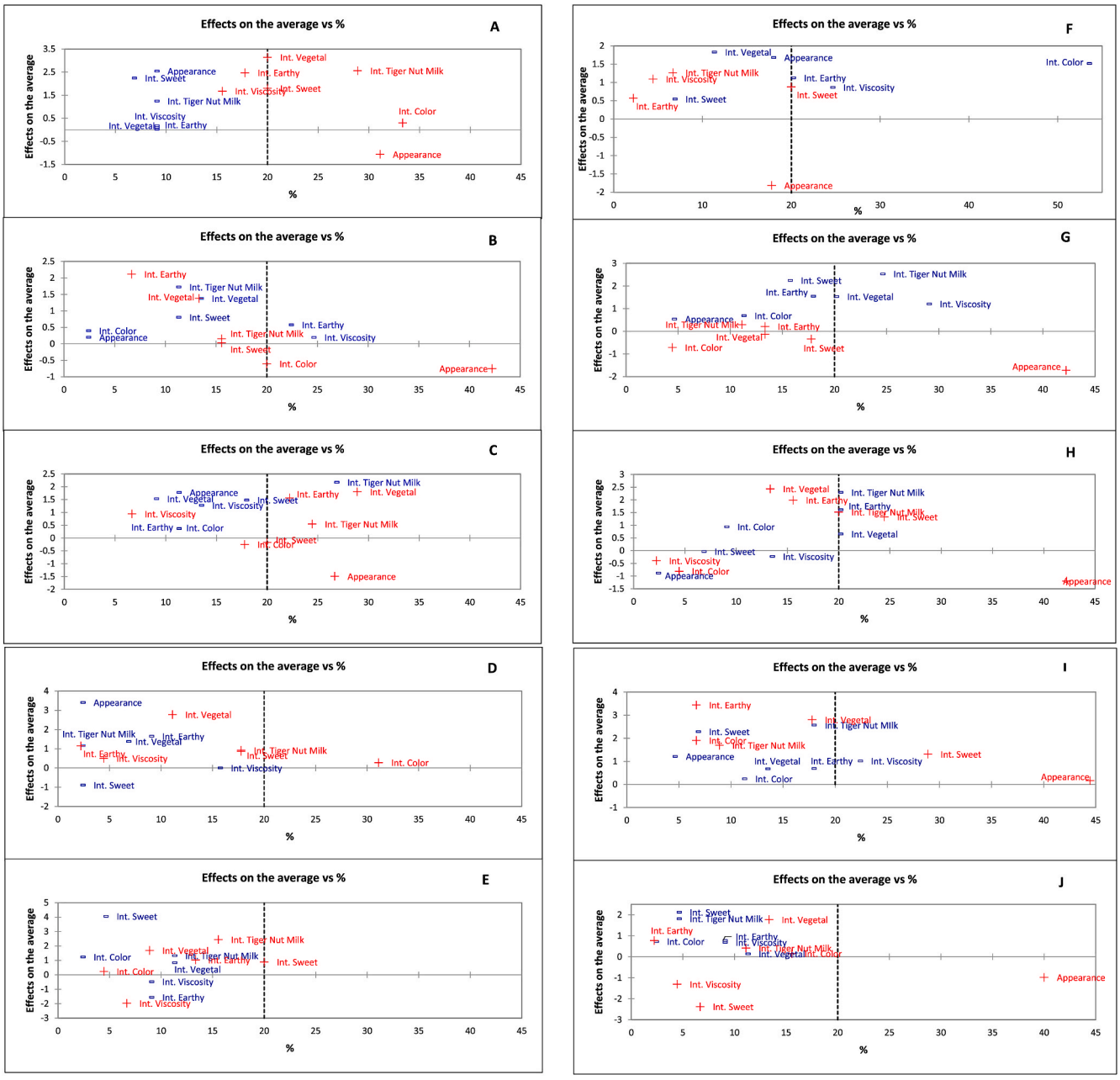

Fig. 2. Penalty analysis of attribute intensity assessed by consumers of the POD and Non-PDO tigernut milk samples (sample code indicated on the top right of each figure; "too low intensity" is indicated. by the symbol "−", and "too high intensity" is indicated by the symbol "+"). Right column PDO samples, left column Non-PDO samples.

- Sample I was linked to V1 (α-pinene), V3 (sabinene), V9 (linalool) and V12 (terpinene-4-ol) and with descriptors such as sweet, tigernut tubers ID, tigernut milk ID, mouth coating and earthy.
- Sample F was linked to V6 (*p*-cymene), V7 (limonene) and V8 (γ-terpinene) and with descriptors sour and lemon.
- Samples A and J were linked to V4 (β-pinene), V10 (nonanal), although no sensory descriptor was linked to these two samples.
- Finally, samples B, C and G were linked to V2 (benzaldehyde), V11 (*m*-methoxyanisole) and V13 (α-terpineol and methyl salicylate) and with the descriptor vegetal.

3.4. Consumer study

The most relevant conclusion was that the consumer satisfaction degree was not influenced by the factor "PDO". PDO sample E got the highest scores for 7 attributes under study; with samples D, J and B showing also good consumer acceptance. On the other hand, the samples with the lowest satisfaction degree, including overall liking, were A, C and H (see Table 5).

The highest overall liking was that of the PDO samples E and D, followed by that of the samples J and B, with the lowest values being found for the samples C, A and H. The color and appearance showed similar behaviors, with sample E getting the best scores. Regarding tigernut milk ID, again the best satisfaction degrees were obtained for samples D, E, and J, while the lowest ones were those of the samples A, C and H (see Table 5).

Results of the online questionnaire on consumer habits and tigernut consumption show that 44% of respondents think that tigernuts are tubers, 28% fruits, 24% do not know, and the other 4% was distributed among roots, stems or leaves. It is important to highlight that 77.5% of

respondents do not eat tigernut tubers, only 8.5% of respondents consume dried tubers and 8.2% fresh tigernuts. Although 100% of respondents knew *horchata* (this was the participation criteria), only 75.4% of them drink it. In addition, 89% of respondents believe that flavor is what makes *horchata de Valencia* different from tigernut milk without PDO, while 41.1% think that authenticity and tradition are the key factors. The consumption of this product is seasonal, with 81.4% of respondents not drinking it all year round, and 100% consuming it in summer. During this season, the consumption frequency reaches 2-3 times a week for 32.1% of respondents. The two most important consumption reasons are: flavor (90.5% of respondents) and tradition (45.2%). Tigernut *horchata* is mainly drunk as a fully liquid product (82.8% of respondents) or frozen (64.7%); almost half of respondents (42.5%) drink it with *fartons* (a typical sweet to dip into the beverage). *Horchata de Valencia* is known and consumed by 61.1% of respondents. Almost 30% of respondents knew about the product but do not consume it. Although ~58% of respondents know that "*horchata de Valencia*" is a Protected Designation of Origin (PDO), only 20% of them knew what the term PDO meant. On the other hand, 40.3% of respondents believe that PDO protection is for the tigernut tubers while 21.2% think if for the drink, 38.6% believe that both, the beverage and the tigernut tubers, are covered by the PDO. Finally, no significant differences were found among age, gender, educational level or income level groups regarding knowledge or consumption.

As a conclusion, it can be stated that there is a clear lack of knowledge among consumers regarding this product and the PDO "*Chufa de Valencia*" even for people living close to the PDO location. Producers should develop a strong information campaign to increase knowledge on their product, its strengths and advantages and promote its consumption.

3.5. Penalty analysis

Penalty analysis is a method that helps interpreting the data of the JAR analysis, describing how close to the optimal is the intensity of key sensory attributes for consumers (Lipan et al., 2019; Pagès, Berthelo, Brossier, & Gourret, 2014). The attributes that would need to be improved, by excess or defect, will be those producing a drop of, at least, 1 unit of satisfaction grade for at least 20% of consumers, reflecting a negative effect on the taste of the samples.

In PDO samples (Fig. 2), it can be observed that no attributes were found in the critical corner in samples B, D and E. On the other side, consumers indicated an excess of intensity of several attributes in sample A (vegetal, sweetness and tigernut milk ID). There were 3 attributes needing improvement in the C sample (excess of vegetal and earthy notes, but defect of tigernut milk ID).

Non-PDO samples (Fig. 2) showed attributes in the critical corner, meaning that all one of them needs to be improved, with the exception of sample J. For example, sample F needed improvements due to low intensity in color and earthy flavor; sample G was low in the intensity of tigernut milk ID, vegetal flavor and viscosity.

Summarizing results from this section, it can be stated that 4 out of 5 of the non-PDO samples need improvements while only 2 out of 5 of PDO samples need improvements. However, it cannot be said that PDO products ensure proper consumer acceptance as 2 out of 5 of the protected products were not fully satisfying regular consumers of this specific drink.

4. Conclusions

This study is the first that carries out a comparison between commercial tigernut milks. It shows a high variability in the volatile profiles of the tigernut milks available in the Spanish market. Also, it has identified liking drivers among consumers. The predominant volatile compounds were limonene, benzaldehyde and *m*-methoxyanisole (which was not previously reported in this product). It was observed that samples protected under the PDO *Chufa de Valencia* did not show a full uniformity regarding volatile and sensory profiles, or even consumer preference. In the descriptive analysis, significant differences were observed in 10 of 14 attributes. However, only bitter and lemon presented significant differences between the DOP and non-PDO groups. The key sensory attributes in this type of vegetal beverage (tigernut tubers ID, tigernut milk ID, earthy and vegetal flavor notes) reached intensities above 5.0 in most of the studied samples. No statistically significant differences were found among PDO and non-PDO samples. The online survey demonstrated the lack of knowledge of consumers about this beverage and its legal protection. Producers organizations (e.g. Regulating Council of PDO) will benefit from harmonizing quality parameters and getting a deeper knowledge on key sensory attributes as well as consumer preferences. In the future, an interesting research line would be to study the odor-active compounds of this product (e.g. non-pasteurized, pasteurized, etc.) and link them with consumer preferences.

CRediT authorship contribution statement

Jesús Clemente-Villalba: Methodology, Software, Writing - original draft, Preparation, Writing - review & editing. **Marina Cano-Lamadrid:** Conceptualization, Methodology, Software, Formal analysis, Writing - original draft, Preparation, Writing - review & editing. **Hanán Issa-Issa:** Methodology, Writing - original draft, Preparation. **Pablo Hurtado:** Methodology. **Francisca Hernández:** Methodology. **Ángel A. Carbonell-Barrachina:** Conceptualization, Formal analysis, Writing - review & editing. **David López-Lluch:** Conceptualization, Formal analysis, Writing - original draft, Preparation, Writing - review & editing, Project administration, All authors have read and agreed to the published version of the manuscript.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.lwt.2020.110606.

References

Adejuyitan, J., Otunola, E., Akande, E., Bolarinwa, I., & Oladokun, F. M. (2009). Some physicochemical properties of flour obtained from fermentation of tigernut (*Cyperus esculentus*) sourced from a market in Ogbomoso, Nigeria. *African Journal of Food Science, 3*, 51–55. https://doi.org/10.5897/AJFS

Alonso, A., Vázquez-Araújo, L., García-Martínez, S., Ruiz, J. J., & Carbonell-Barrachina, Á. A. (2009). Volatile compounds of traditional and virus-resistant breeding lines of Muchamiel tomatoes. *European Food Research and Technology, 230* (2), 315–323. https://doi.org/10.1007/s00217-009-1173-2

Cano-Lamadrid, M., Tkacz, K., Turkiewicz, I. P., Clemente-Villalba, J., Sánchez-Rodríguez, L., Lipan, L., … Wojdyło, A. (2020). How a Spanish group of millennial generation perceives the commercial novel smoothies? *Foods, 9*(9), 1213. https://doi.org/10.3390/foods9091213

Cantalejo, M. J. (1997). Analysis of volatile components derived from raw and roasted earth-almond (*Cyperus esculentus* L.). *Journal of Agricultural and Food Chemistry, 45* (5), 1853–1860. https://doi.org/10.1021/jf960467m

Chukwuma, E., Obioma, N., & Ononogbu, C. (2010). The phytochemical composition and some biochemical effects of Nigerian tigernut (*Cyperus esculentus* L.) tuber. *Pakistan Journal of Nutrition, 9*, 709–715. https://doi.org/10.3923/pjn.2010.709.715

Clemente-Villalba, J., Ariza, D., García-Garví, J. M., Sánchez-Bravo, P., Noguera-Artiaga, L., Issa-Issa, H., … Carbonell-Barrachina, Á. A. (2020). Characterization and potential use of Diplotaxis erucoides as food ingredient for a sustainable modern cuisine and comparison with commercial mustards and wasabis. *European Food Research and Technology, 246*(7), 1429–1438. https://doi.org/10.1007/s00217-020-03501-3

Codina, I., Trujillo, A. J., & Ferragut, V. (2016). Horchata. In K. K. J. Oliveira (Ed.), *Traditional foods - 2016; general and consumer aspects*. New York: Springer US. https://doi.org/10.1007/978-1-4899-7648-2, 2016.

Coşkuner, Y., Ercan, R., Karababa, E., & Nazlıcan, A. N. (2002). Physical and chemical properties of chufa (*Cyperus esculentus* L) tubers grown in the Çukurova region of Turkey. *Journal of the Science of Food and Agriculture, 82*(6), 625–631. https://doi.org/10.1002/jsfa.1091

CRDO, Consejo Regulador de Denominación de Origen. (2019). Horchata de Chufa de Valencia. Retrieved from http://www.chufadevalencia.org/.

Djomdi, D., Ejoh, A., & Ndjouenkeu, R. (2007). Soaking behavior and milky extraction performance of tiger nut (*Cyperus esculentus*) tubers. *Journal of Food Engineering, 78*, 546–550. https://doi.org/10.1016/j.jfoodeng.2005.10.022

Issa-Issa, H., Guclu, G., Noguera-Artiaga, L., López-Lluch, D., Poveda, R., Kelebek, H., … Carbonell-Barrachina, Á. A. (2020). Aroma-active compounds, sensory profile, and phenolic composition of Fondillón. *Food Chemistry, 316*, 126353. https://doi.org/10.1016/j.foodchem.2020.126353

Lipan, L., Cano-Lamadrid, M., Corell, M., Sendra, E., Hernández, F., Stan, L., … Carbonell-Barrachina, Á. A. (2019). Sensory profile and acceptability of HydroSOStainable almonds. *Foods, 8*(2), 64. https://doi.org/10.3390/foods8020064

MAPA - Ministerio de Agricultura, P. y. A. (2017). Anuario de estadística 2017. https://www.mapa.gob.es/estadistica/pags/anuario/2017/CAPITULOSPDF/CAPITULO13/pdfc13_3.3.3.pdf.

Martín-Esparza, E., & González-Martínez, C. (2016). Horchata de Chufa: A traditional Spanish beverage with exceptional organoleptic, nutritive, and functional attributes. In K. Kristbergsson, & S. Ötles (Eds.), *Functional properties of traditional foods* (pp. 371–375). Boston, MA: Springer US.

Matos, F., Cavalcanti, F., & Parente, J. (2008). Estudo agronômico qualitativo e quantitativo de *Cyperus esculentus* L. (junça) - Uma fonte inexplorada de alimento energético. [Spanish language] Estudio agronómico cualitativo y cuantitativo de *Cyperus esculentus* L. (juncia) - una fuente inexplorada de alimento energético. *Revista de Ciencias Agronomicas, 39*, 124–129.

Nawade, B., Yahyaa, M., Reuveny, H., Shaltiel-Harpaz, L., Eisenbach, O., Faigenboim, A., … Ibdah, M. (2019). Profiling of volatile terpenes from almond (*Prunus dulcis*) young fruits and characterization of seven terpene synthase genes. *Plant Science, 287*, 110187. https://doi.org/10.1016/j.plantsci.2019.110187

Pagès, J., Berthelo, S., Brossier, M., & Gourret, D. (2014). Statistical penalty analysis. *Food Quality and Preference, 32*, 16–23. https://doi.org/10.1016/j.foodqual.2013.07.008

Reglamentación Técnico-Sanitaria para la Elaboración y Venta de Horchata de Chufa. (1988). Royal Decree of Spanish Government, 1338/1988 C.F.R.

Roselló-Soto, E., Poojary, M. M., Barba, F. J., Koubaa, M., Lorenzo, J. M., Mañes, J., et al. (2018). Thermal and non-thermal preservation techniques of tiger nuts' beverage "horchata de chufa". Implications for food safety, nutritional and quality properties. *Food Research International, 105*, 945–951. https://doi.org/10.1016/j.foodres.2017.12.014

Rutledge, K. P., & Hudson, J. M. (1990). Sensory evaluation: Method for establishing and training a descriptive flavor analysis panel. *AGRIS-FAO, 44*(12), 78–84.

Sánchez-Zapata, E., Fernández-López, J., & Pérez-Alvarez, J.Á. (2012). Tiger nut (*Cyperus esculentus*) commercialization: Health aspects, composition, properties, and food applications. *Comprehensive Reviews in Food Science and Food Safety, 11*(4), 366–377. https://doi.org/10.1111/j.1541-4337.2012.00190.x

Takhatajah, A. (1992). *Angiosperms (the flowering plants). New encyclopedia britannica, macropaedia* (15th ed.). Chicago: Encyclopedia Britannica.

Umerle, S. C., Okafor, E. O., & Uka, A. S. (1997). Evaluation of the tubers and oil of *Cyperus esculentus*. *Bioresource Technology, 61*(2), 171–173. https://doi.org/10.1016/S0960-8524(97)00045-X

de Vries, F. T., & Femke, T. (1991). Chufa (*cyperus esculentus, cyperaceae*): A weedy cultivar or a cultivated weed? *Economic Botany, 45*(1), 27–37. https://doi.org/10.1007/BF02860047