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Identification of aromatic compounds and their sensory characteristics in cassava flakes and "garri" (*Manihot esculenta* Crantz)

Identificación de los compuestos aromáticos y sus características sensoriales en los copos de mandioca y el "garri" (*Manihot esculenta*)

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Cassava flakes and "garri" are some of the popular cassava products consumed in Africa, Southeast Asia, and Brazil. These products are cherished for their creamy colour and characteristic aroma. The aroma compounds responsible for the unique aroma notes of these products were evaluated by means of solid-phase micro-extraction (SPME). Results confirmed that the divinylbenze-carboxen-polydimethylsiloxane (DVB-CAR-PDMS) fibre was the most appropriate for the isolation of aroma compounds in the cassava products. In addition, the best response was attained when the extraction temperature was 60°C, equilibrium time 20 min, extraction time 10.94 min, and water addition 35%. Analysis of the odorants led to the identification of 21 compounds with an array of odour notes. However, results of the aroma extract dilution analysis (AEDA), and odour activity values (OAVs) showed that guaiacol, 3-methylbutanal, methylpropanal, and butyric acids contributed intensely to the characteristic aroma of both cassava flakes and garri.

Keywords: cassava flakes; garri; aroma compounds; AEDA

Los copos de mandioca y el "garri" son algunos de los productos más populares de la mandioca que se consumen en África, el sureste asiático y Brasil. Estos productos son muy preciados por su color crema y su aroma característico. Se evaluaron los compuestos aromáticos responsables de su aroma único mediante la micro-extracción en su fase sólida. Los resultados confirmaron que la fibra divinilbenzeno-carboxen-polidimetilsiloxano (DVB-CAR-PDMS) fue la más apropiada para el aislamiento de los compuestos aromáticos en los productos de mandioca. Además, la mejor respuesta se alcanzó cuando la temperatura de extracción fue de 60°C, el tiempo de equilibrio 20 min, el tiempo de extracción 10,94 min y un 35% de agua añadida. El análisis de los olores llevó a la identificación de 21 compuestos con una serie de notas de los olores. Sin embargo, los resultados del análisis de disolución del extracto del aroma (AEDA) y los valores de actividad olfativa (OAVs) mostraron que el guayacol, el 3-metilbutanal, el metilpropanal y los ácidos butíricos contribuyeron intensamente al aroma característico de los copos de mandioca y del garri.

Palabras clave: copos de mandioca; garri; compuestos aromáticos; AEDA

Introduction

Cassava (Manihot esculenta Crantz), also called manioc, and tapioca-root, is a woody shrub of the Euphorbiaceae (spurge) family native to South America. It is extensively cultivated as an annual crop in tropical and subtropical regions for its edible starchy tuberous root. Cassava is the sixth most important food source in the tropics after rice, wheat, potato, barley, and maize (Lebot, 2009). The crop has become widespread and all classes of people consume it. It is often considered an inferior food because of its low content of protein, essential minerals, and vitamins (Oboh, Akindahunsi, & Oshodi, 2002). In recent times, food researchers have discovered new ways to use cassava. Some of the new products made from cassava are chips (Nambisan & Sundaresan, 1985; Vitrac, Dufour, Trystram, & Raoult-Wack, 2001), fermented beer (Colehour et al., 2014), bread (Pasqualone et al., 2010), cake (Sanful & Darko, 2010), tapioca (Adebowale, Sanni, Awonorin, Daniel, & Kuye, 2007; Sanni, Atere, & Kuye, 1997), biofuel (Hu, Tan, & Pu, 2006; Nguyen, Gheewala, & Garivait, 2007; Papong & Malakul, 2010), and medicine (La Frano, Woodhouse, Burnett, & Burri, 2013). Cassava, when dried to a powdery (or pearly) extract, is called tapioca; its fermented, flaky version is named "garri" or "gari". Garri, a grated, fermented, and roasted cassava product, is

the most popular cassava product consumed in West Africa. In Brazil, detoxified manioc is made into flakes or ground and cooked to a dry, often hard, or crunchy meal that is used as a condiment. Cassava flakes can be eaten alone, or toasted in butter as a side dish. Apart from colour, aroma is one of the most appreciated characteristics that attract consumers to buying "garri" and flakes.

Volatiles inherent in the cassava flakes, and "garri" are yet to be characterized. To our knowledge, no data are available on the aroma compounds causing the characteristic aroma of cassava flakes, and "garri". One of the most popular methods for the isolation of flavour compounds from different foodstuff is by solid-phase micro-extraction (SPME). The SPME method has a wide range of applications in the analysis of food components and contaminants. It has been used for the extraction of volatiles from both food raw materials and end-products. Among such applications are determination of volatiles in pasta (Pasqualone, Paradiso, Summo, Caponio, & Gomes, 2014), bread (Poinot et al., 2007; Ying, Lasekan, Naidu, & Lasekan, 2012), rice (Grimm, Bergman, Delgado, & Bryant, 2001), and wheat (Jeleń, Majcher, Zawirska-Wojtasiak, Wiewiórowska, & Wąsowicz, 2003), and the determination of acrolein in French fries (Osório & de Lourdes Cardeal, 2011). Surprisingly, this

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technique has had very little application in cassava products. Osório and de Lourdes Cardeal (2013) applied the technique of SPME-GC/MS to evaluate and quantify acrolein levels in cassava and pork sausage fried in different vegetable oils – palm, canola, soybean, corn, and sunflower oils. The results revealed that the highest concentration of acrolein was produced in samples fried in both canola and sunflower oils. It was then concluded that several parameters such as the fatty acid profile of the oil, viscosity of the oil, and the type of food material contributed to the factors that influenced the concentration of acrolein found in different fried food products. Therefore, this study addressed the application of HS-SPME for sampling volatile constituents in cassava flakes and "garri".

Materials and methods

Chemicals

The following chemical compounds – 3-methylbutanal, furfural, acetyl furan, 1-octen-3-one, benzeneacetaldehyde, pentylfuran, acetyl-1-pyrroline, hexadecanoic acid, ethyl octanoate, butyric acid, guaiacol, methylpropanal, and octadecanoic acid – were purchased from Sigma–Aldrich, Steinheim, Germany. Stock standard solutions of 10^3 or 10^4 µg/mL of each component were prepared by dissolving the pure standard in ethanol (40%) (Lasekan, Khatib, Juhari, Patiram, & Lasekan, 2013). The samples were stored at 4°C. Working standard solutions were prepared daily by mixing an aliquot of each solution and diluting with ultra-pure water (Millipore Co., Bedford, MA, USA) to obtain the desired concentration.

Quantification

Qualitative and semi-quantitative analyses were carried out by comparing the retention times of the chromatographic peaks with those of authentic compounds run under the same conditions and by comparing the retention indices (i.e. Kovats indices) with that obtained from the literature (Cerny & Grosch, 1993; Guth & Grosch, 1994; Jeleń et al., 2003; Pellati, Benvenuti, Yoshizaki, Bertelli, & Rossi, 2005; Poinot et al., 2007; Rychlik, Schieberle, & Grosch, 1998). The comparison of the MS fragmentation

pattern with those of pure compounds and mass spectrum database search was performed using the literature mass spectra (NIST/NBS75K library) (Heller & Milne, 1986).

Cassava flakes and garri production

Low-cyanide (>31 mg/kg of HCN) cassava tubers (10 kg) were obtained from the local market in Malaysia. The cassava tubers were peeled to remove the outer brown skin and the inner thick cream layer and washed thoroughly in water. The washed cassava tubers were grated to form a paste called "massa". The water in the "massa" was squeezed out under hydraulic press. The dried "massa" was re-dissolved in water (2:1). The wet cassava mash was spread onto a Simon Double drum dryer (Nottingham, England) in a thin sheet having a thickness of about 0.2 mm. The resulting dried sheets of flakes with a moisture content of 6% were packaged in aluminium poaches and vacuum sealed (Vasmaster sealer).

For the garri production, the dewatered paste (i.e. Massa) was allowed to ferment at 27°C (ambient temperature) for 24 h. The fermented wet cake was sieved into grits and then roasted in a hot frying tray to form a final dry and granular product (≥1 mm particle size). The production of both cassava flakes and garri was performed in duplicate.

SPME fibre screening

The silica fibres (StableFlex Divinylbenzene/Carboxen/polydimethylsiloxane, DVB/CAR/PDMS 50/30 µm; polydimethylsiloxane-divinylbenzene, PDMS-DVB, 65 µm; StableFlex Carboxen/Polydimethylsiloxane, CAR/PDMS, 65 µm; and StableFlex Carbowax-Divinylbenzene, CW-DVB, 65 µm) purchased from Supelco (Bellefonte, PA, USA) were screened before the optimization study. Each fibre was exposed to the headspace of cassava products for the same time (20 min) and temperature (50°C) and the desorption time was 5 min at 260°C.

Headspace SPME

Approximately 2 g of sample was placed in a 20 ml vial closed with a polypropylene cap and PTFE/silicone septum (Supelco,

Table 1. Performance characteristics of the calibration curves obtained with the chosen fibre.

Tabla 1. Características de rendimiento de las curvas de calibración obtenidas con las fibras escogidas.

No	Compounds	Linear range		Limit of detection		Recovery	
		(mg l^{-1})	Reg. Coeff.	LOL, %	(LOD, mg l^{-1})	(%)	Slope±SD
1.	3-Methylbutanal	0.2–3.5	0.97	98.2	0.34	99.6 ± 0.09	0.15 ± 0.00
2.	Furfural	0–25	0.99	99.1	0.12	93.3 ± 0.17	0.71 ± 0.01
3.	Hexadecanoic acid	1–25	0.96	97.5	0.20	94.6 ± 0.10	0.10 ± 0.00
4.	Acetyl furan	0–25	0.98	99.2	0.13	90.4 ± 0.21	1.41 ± 0.02
5.	1-Octen-3-one	5–200	0.99	99.4	1.8	99.7 ± 0.05	3.41 ± 0.05
6.	Benzeneacetaldehyde	0.04–11	0.95	97.8	0.87	93.5 ± 0.15	1.12 ± 0.00
7.	Pentyl furan	0–10	0.97	98.1	0.45	90.8 ± 0.22	0.21 ± 0.00
8.	Acetyl-1-pyrroline	0.5–2.0	0.99	99.1	0.27	90.1 ± 0.41	1.03 ± 0.01
9.	Ethyl octanoate	0–1	0.99	99.0	0.08	92.1 ± 0.15	0.10 ± 0.00
10.	Butyric acid	2–50	0.98	99.2	0.82	91.4 ± 0.20	2.10 ± 0.02
11.	Guaiacol	0–1	0.97	98.3	0.04	95.4 ± 0.50	0.10 ± 0.00
12.	Methyl propanal	0–10	0.94	99.6	0.26	96.2 ± 0.09	0.04 ± 0.00
13.	Octadecanoic acid	0–10	0.98	99.2	0.67	91.0 ± 0.5	1.70 ± 0.00

Note: LOL, Linearity; Reg. Coeff., Regression coefficient.

Nota: LOL, Linealidad; Reg. Coeff., Coeficiente de regresión.

Bellefonte, PA, USA). The SPME extractions of the cassava flakes and garri were performed according to the experimental design outlined in Table 1. After each extraction, the fibre was inserted into the GC injector port using a 0.75 mm i.d. liner (García-Martín, Herrero, Peña, & Barciela, 2010), and the fibre thermally desorbed. A desorption time of 1 min at 250°C was used in split-less mode. Before sampling, each fibre was reconditioned for 5 min in the GC injector port at 250°C.

Gas chromatography (GC) and gas chromatography-mass spectrometry (GC-MS)

The GC-MS analyses were carried out as described by Beaulieu and Grimm (2001). The volatile compounds extracted by HS-SPME procedures from the cassava products were tentatively identified on A QP-5050A GC-MS instrument equipped with a GC-17A ver. 3 GC with a flame ionization detector (FID) (Shimadzu, Kyoto, Japan). The column used was BP X 5 (5% phenylpolysilphenylenesiloxane, 30 m × 0.25 mm i.d., film thickness 0.25 μm) (Scientific Instrument Services, Inc., NJ; USA). Fibre was retained inside the heated injection port for 5 min while the injection port was operated in split-less mode. The initial oven temperature was 50°C, held for 1 min, ramped at 5°C min^{-1}–100°C and at 10°C min^{-1}–250°C. It was held at this temperature for 9 min. A mixture of aliphatic hydrocarbons (C_8–C_{24}) in hexane was loaded onto the SPME fibre and injected under the above temperature programme to calculate the retention indices for all identified compounds and reference standards.

All mass spectra were acquired in electron-impact (EI) mode as described by Lasekan et al. (2013). The data acquisition was carried out with the HP-Chemstation Software (A.03.00) and identified using the NIST/NBS75K database.

Gas chromatography-olfactometry (GC-O)

For GC-O analysis, an aliquot (2 μL) of the cassava flakes and garri extracts was injected separately into an Agilent 6890 N instrument equipped with a split–split-less injector, an FID, and an ODP 2 (Olfactory Detector Port) (Gerstel, Mulheim, Germany) (Lasekan et al., 2013). The aliquot (2 μL) was separated on a BP X 5 column (30 m × 0.25 mm i.d., 0.25 μm film thickness) (Scientific Instrument Services, Inc., NJ, USA). The temperature programme and carrier gas conditions used were similar to that of GC-MS. At the end of the capillary column, the effluent was split 1:1 for the FID (250°C).

Three trained assessors, who have demonstrated satisfactory sensitivity, and strong ability to recognize odours in previous sessions, were used for the sniffing experiments. The sniffing analysis was divided into three parts of 20 min and each assessor participated in the exercise. Samples were presented in a completely randomized order blocked by sample replication. The assessors evaluated each of the two replicate samples once for each cassava product. Only the odours detected by all the three assessors were considered valid.

Aroma extracts dilution analysis (AEDA)

GC-O of serial dilutions using the AEDA approach (Schieberle, 1995) was used to determine the flavour dilution (FD) factors of the odour-active compounds. The original cassava product extracts (50 μL) from 50 g of powdered cassava flakes and "garri" were diluted in a stepwise manner by the addition of dichloromethane (1: 1) until no odorant was detectable by sniffing of the highest dilution. GC-O was carried out with 0.5 μL aliquot using capillary BPX5. Three experienced sniffers performed the AEDA experiments. Their response to the individual compounds did not differ by >2 FD factors.

Aroma profile determination

The 20 g samples of cassava flakes and "garri" were presented to the panellists in 100 mL closed containers and preheated to 35°C, 30 min prior to quantitative descriptive analysis (QDA). Samples were orthonasally evaluated by a sensory panel (10). The sensory analysis was developed by 10 trained panellists (six females and four males) recruited from the University Putra Malaysia. In previous training sessions held weekly, the panellists were trained in recognizing orthonasally and retronasally 150 selected odorants at different odorant concentrations according to their odour qualities. Training was at least for 1 year prior to participation in the actual sensory before the panellist participated in the actual sensory experiments. Descriptors used were determined in initial sensory experiments. Each descriptor used was defined based on the odour of a reference compound dissolved in water at a concentration of 100 times above the respective threshold value. Reference odorants used were Methylpropanal (malty), 3-methylbutanal (Malty), butyric acid (rancid butter), 2-acetylpyrroline (popcorn), 1-octen-3-one (mushroom), and guaiacol (smoky). Panellists were asked to rate the intensities of each descriptor on a non-structural scale from 0 to 10, with 0 = not detectable, 5 = weak, and 10 = strong. The results were presented in a web plot.

Experimental design

The optimization of the HS-SPME conditions was performed using a central composite experimental design (CCD) (Lasekan & Abdulkarim, 2012). The analysis was based on a 2^4 factorial design plus 8 axial points and 6 replicates in the centre of the design. The effect of four independent variables, X_1 (extraction temperature), X_2 (equilibrium time), X_3 (extraction time), and X_4 (water addition level), on the total sum of the peak areas of compounds identified in the samples (Y_1) was evaluated using response surface methodology (RSM). Therefore, 30 treatments were assigned based on the second-order CCD with four independent variables. The experimental data of the four factors and three levels are presented in Tables 2 and 3. Statistical analysis was performed using the

Table 2. Factor levels and experimental domain applied to optimize the HS-SPME experimental conditions.

Tabla 2. Niveles de factor y dominio experimental aplicado para optimizar las condiciones experimentales HS-SPME.

Factor	Experimental domain				
	−axial	−1	0	1	+axial
Extraction temperature (°C)	30	40	50	60	70
Equilibrium time (Min)	0.0	10	20	30	40
Extraction time (Min)	0.0	5	10	15	20
Water addition (%)	15	10	35	60	85

Table 3. Experimental conditions and response value (total area) of the central composite design used to optimize the extraction conditions of the cassava products (flakes and "garri") by HS-SPME.

Tabla 3. Condiciones experimentales y valores de respuesta (área total) del diseño compuesto central utilizado para optimizar las condiciones de extracción de los productos de mandioca (copos y "garri") mediante HS-SPME.

No.	Extraction temp (°C)	Equilibrium time (min)	Extraction time (min)	Water addition (%)	Total area[a]
1	50.00	20.00	10.00	35.00	1.96517E+008
2	60.00	30.00	15.00	10.00	2.84101E+008
3	40.00	30.00	15.00	60.00	4.87865E+008
4	40.00	30.00	5.00	10.00	2.19232E+008
5	40.00	10.00	5.00	60.00	4.87865E+008
6	60.00	30.00	5.00	10.00	2.09232E+008
7	40.00	10.00	15.00	10.00	1.28908E+008
8	60.00	10.00	15.00	10.00	2.13103E+008
9	40.00	30.00	15.00	10.00	2.43462E+008
10	40.00	30.00	5.00	60.00	5.88991E+007
11	50.00	20.00	10.00	35.00	2.56805E+008
12	60.00	10.00	5.00	10.00	6.17491E+008
13	40.00	10.00	15.00	60.00	1.48908E+008
14	50.00	20.00	10.00	35.00	2.32175E+008
15	60.00	10.00	15.00	60.00	2.54207E+008
16	60.00	10.00	5.00	60.00	1.93477E+008
17	60.00	30.00	5.00	60.00	2.43918E+008
18	60.00	30.00	15.00	60.00	2.68341E+008
19	40.00	10.00	5.00	10.00	2.36805E+008
20	50.00	20.00	10.00	35.00	2.39821E+008
21	50.00	20.00	10.00	35.00	2.96482E+008
22	50.00	0.00	10.00	35.00	1.86508E+008
23	30.00	20.00	10.00	35.00	6.74155E+008
24	50.00	20.00	20.00	35.00	2.46713E+008
25	50.00	20.00	0.00	35.00	8.41861E+006
26	50.00	40.00	10.00	35.00	2.57813E+008
27	50.00	20.00	10.00	35.00	2.22144E+008
28	70.00	20.00	10.00	35.00	2.64782E+008
29	50.00	20.00	10.00	−15.00	2.82819E+008
30	50.00	20.00	10.00	85.00	2.24628E+008

Note: [a]Total area is expressed in arbitrary units.

Nota: [a]El área total está expresado en unidades arbitrarias.

Minitab v. 13.2 statistical packages (Minitab Inc., Pennsylvania, USA).

Results and discussion

Choice and SPME fibre screening

The SPME process is based on the partitioning of analytes between the phase coating, sample, and the headspace above it. Microextraction is completed when the analyte concentration reaches distribution equilibrium between these three phases. Therefore, selecting the best coating is an essential part of the optimization process and this invariably depends on the chemical nature of the target analytes. In the present study, four fibres – polydimethylsiloxane-divinylbenzene (PDMS-DVB, 65 μm), Stableflex divinylbenzene-carboxen-polydimethylsiloxane (DVB-CAR-PDMS, 50/30 μm), Stableflex carbowax-divinylbenzene (CW-DVB, 65 μm), and carboxen-polydimethylsiloxane (CAR-PDMS, 65 μm) – were evaluated for the analysis of cassava flakes and "garri" aroma. The peak areas of all aroma compounds isolated by each fibre were summed up and referred to as the "total peak area". The results of the fibre screening are shown in Figure 1. The total peak area value obtained with the DVB-CAR-PDMS fibre was divided by a factor of two, since the fibre is twice as long as the other fibres (Bicchi, Drigo, & Rubiolo, 2000). The results of the fibre screening as shown in Figures 1 and 2 revealed that the DVB-CAR-PDMS fibre produced the best profile since it extracted more odorant that the rest of the fibres

and captures the highest number of volatiles. Therefore, we decided to select this coating for the evaluation of the characteristic volatile constituents of cassava flakes and garri.

Optimization of the HS-SPME

Analysis of headspace volatile compounds by HS-SPME has been shown to be greatly influenced by the vapour pressure of flavour compounds in the vial (Pellati et al., 2005). In the present
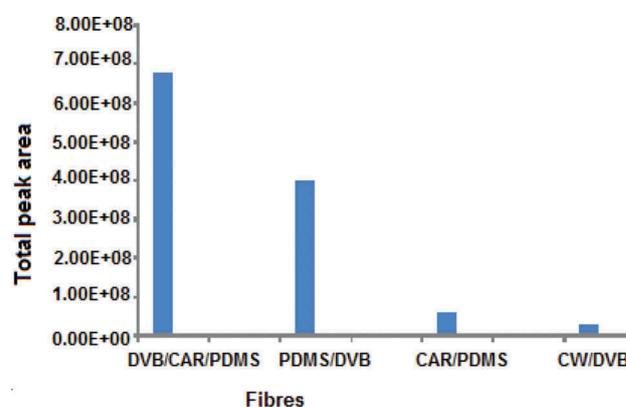
Figure 1. Plot of extraction efficiency of different SPME fibres.

Figura 1. Gráfico de eficiencia de extracción de diferentes fibras SPME.
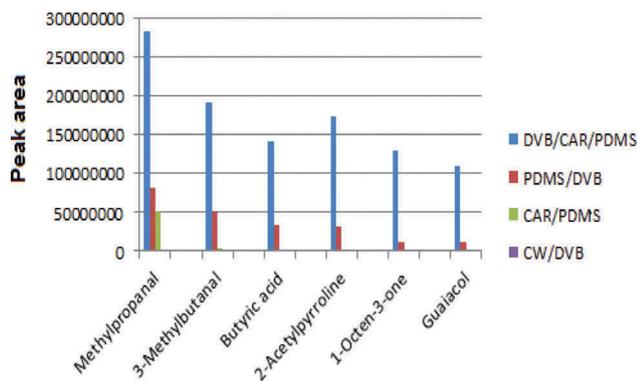
Figure 2.   Effect of the SPME fibre coating on the extraction of key odorants in cassava flakes and "garri".

Figura 2. Efecto del recubrimiento de fibra SPME en la extracción de los olores clave en los copos de mandioca y de garri.

study, factors that have strong effect on the vapour pressure and equilibrium of the aroma compounds in the headspace of the flakes and "garri" are extraction temperature (°C), equilibrium time (min), extraction time (min), and water addition (%). These four factors were therefore chosen and optimized. Thirty experiments of the design were performed at random and the responses are shown in Table 3. Analysis of variance (ANOVA) was used to analyse the obtained data. The procedure was used to evaluate the statistical significance of each factor and interactions between the different factors. To aid visualization, the 3D response surface plots for (A) total area versus extraction time and equilibrium time, (B) total area against water addition and extraction temperature, and (C) total area against equilibrium time and extraction temperature are presented in Figure 3.

Results showed that the main effect of water addition and the interaction effect of extraction temperature and water addition were the most significant parameters ($P < 0.05$), with strong positive influence on the total area. It is a known fact that an increase in sampling temperature increases headspace concentration of the aroma compounds (Zhang, Yang, & Pawliszyn, 1994). In this study, high temperature increased the response (total area). Similarly, the addition of water to cassava flakes and garri samples prior to the micro-extraction process produced an increase in peak area of the

extracted compounds (Figure 3B). The best SPME efficiency was observed when 10% water was added to the sample. However, addition of a higher amount of water (>40%) reduced the extraction efficiency significantly. From the results, the best response within the range studied was achieved when the extraction temperature was 60°C, the equilibrium time 20 min, extraction time 10.94 min, and water addition 35%. Therefore, these values were used for the extraction of volatile compounds from the cassava flakes and garri.

Calibration and linearity

The range of linearity studied for each compound is shown in Table 1. Excellent linearity and good recovery were achieved in all cases followed by a good correlation coefficient ($r^2 > 0.94$). In addition, the limit of detection (LOD) achieved was low enough to detect the volatile compounds in the cassava products. The linearity, which is the "on-line linearity" (LOL), was determined the in following equation in which RSD is the relative standard deviation of the slope (expressed as a percentage):

$$LOL \ (\%) = 100 - RSD \tag{1}$$

Identification of the key odorants in cassava flakes and garri

Sensory evaluation of the cassava flakes and garri samples revealed characteristic aroma patterns. While the cassava flakes were characterized by the malty, popcorn nuances, the garri was dominated by the smoky, rancid buttery and mushroom notes (Figure 4). The reasons for this observation are not far-fetched since the cassava products were produced differently. To elucidate the molecular principles for the observed aroma impressions, the odorants from cassava flakes and garri were sequentially analysed by HS-SPME, GC-MS, and GC-O. This protocol led to the detection and identification of 21 odorants (Table 4). The odorants identified included heterocyclic compounds, aldehydes, esters, alcohols, aromatic hydrocarbons, sesquiterpenes, and an acid. Interestingly, none of the compounds was representative of the characteristic aroma of garri or flakes. The aroma of the flakes and garri appeared to be an integration of a complex mixture of aroma compounds. To distinguish the volatile constituents responsible for the overall odour-impression obtained
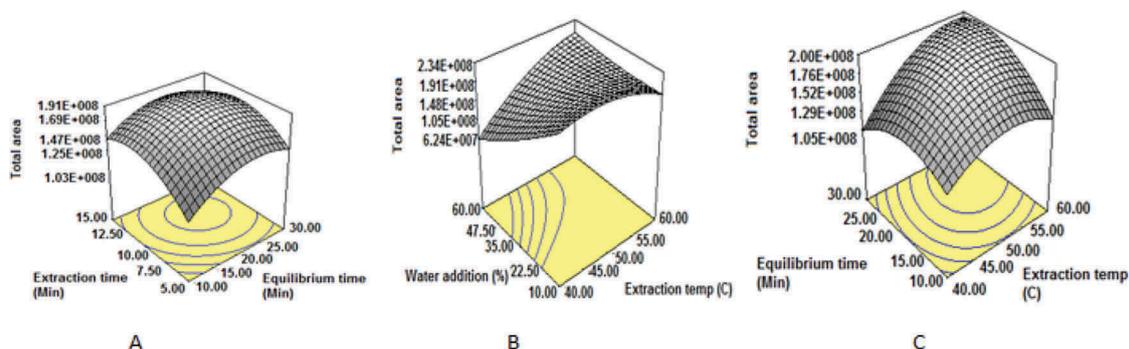
Figure 3.   Response surface plot for: (A) total area versus extraction time and equilibrium time; (B) total area versus water addition and extraction temperature; and (C) total area versus equilibrium time and extraction temperature.

Figura 3. Gráfico de superficie de respuesta para: (A) área total frente a tiempo de extracción y tiempo de equilibrio; (B) área total frente a agua añadida y temperatura de extracción; (C) área total frente a tiempo de equilibrio y temperatura de extracción.
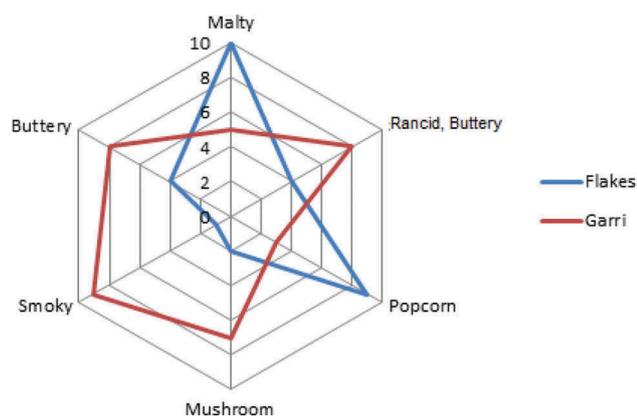
Figure 4.   Orthonasal sensory evaluation of the main aroma attributes of cassava flakes and garri.

Figura 4. Evaluación sensorial ortonasal de los principales atributos aromáticos de los copos de mandioca y el garri.

from the flakes and garri, concentrated distillates from both samples were analysed by the AEDA (Table 4). Results of the AEDA showed that 2-acetyl-1-pyrroline (FD = 512) was the most significant odorant in the cassava products. This was closely followed by 3-methylbutanal (FD = 256) and 1-octen-3-one (FD = 128). Other compounds with appreciable FD factors were guaiacol (FD = 32), butyric acid (FD = 16), and methylpropanal (FD = 16).

The six odorants with the highest FD ≥ 16 factors were selected for further investigation to ascertain their respective contribution to the aroma profile of the products. The odour

activity values (OAVs) of the odorants were calculated based on their orthonasal odour thresholds in starch (Table 5). The OAVs showed that guaiacol, 3-methylbutanal, methylpropanal, and butyric acid contributed intensely to the cassava flakes and garri aroma. Interestingly, odorants such as 2-acetylpyrroline and 1-octen-3-one, which exhibited high concentrations in the samples, gave relatively low OAVs. For instance, 2-acetylpyrroline produced <1 OAVs in both flakes and garri. It is therefore assumed that this odorant is not contributing to the overall aroma of these products. It is worth noting that series of interaction phenomena, such as additive, synergistic or suppressive effects, are well-documented, so that the present OAVs do not allow a direct prediction of the odorant's contribution to the cassava products' aroma sensations. However, they can be regarded as the key to further investigations and sensory experiments in this field of research.

Conclusion

This study has shown that the DVB-CAR-PDMS is the best fibre for the extraction of odorants from cassava flakes and garri. In addition, analysis of the odorants led to the identification of 21 odorants. These odorants exhibited an array of odour notes such as malty, rancid/buttery, popcorn, balsamic, mushroom, ammonia-like, chocolate, cocoa-like, and fatty. However, the results of the AEDA and OAVs revealed that guaiacol, 3-metylbutanal, methylpropanal, and butyric acid contributed intensely to the characteristic aroma of cassava flakes and garri. Moreover, odorants such as 2-acetylpyrroline

Table 4.   Concentrations (µg kg^{-1}) of odorants in cassava flakes and "garri" by means of HS-SPME.

Tabla 4. Concentraciones (µg kg^{-1}) de los olores en los copos de mandioca y el "garri" mediante HS-SPME.

No	Compounds	ID[a]	LRI (BPx5)	FD	Flakes	"Garri"	Odour impression
1	Methylpropanal	A	>600	16	497	141	Malty
2	3-Methyl butanal	A	645	256	918	1012	Malty
3	2-Methyl-2-butanamine	C	714	nd	37	67	nd
4	Unknown	-	715	4	114	317	Ammonia-like
5	Butyric acid	A	801	16	108	919	Rancid, buttery
6	Furfural	A	829	4	175	297	Almond-like
7	Acetyl furan	A	893	2	48	54	Balsamic
8	Methoxy phenyl oxime	B	897	nd	36	61	nd
9	2-Acetyl-1-pyrroline	A	923	512	1129	798	Popcorn
10	1-Octen-3-one	A	971	128	144	211	Mushroom
11	2-Pentyl furan	B	993	4	41	53	Green, beany
12	Benzeneacetaldehyde	A	1044	4	128	98	Cocoa-like
13	Guaiacol	A	1085	32	86	345	Smoky
14	4,5-Dimethyl-2-phenyl-1,3,2-dioxaborolane	C	1398	nd	134	131	nd
15	2,4-Di-tert-butyl phenol	C	1512	nd	67	75	nd
16	6-Epi-shyobunol	B	1518	nd	34	54	nd
17	2-Hydroxy-2(5-methylfuran-2yl)-1-phenylethanone	B	1519	2	214	112	Chocolate-like
18	*tert*-Hexadecanethiol	C	1522	4	54	78	Fatty
19	2-Methyl-1-hexadecanol	A	1534	2	32	43	Medicinal
20	9-Octadecenal	A	1999	4	18	56	Fatty
21	N-Benzyl-2-aminocinnamate. Methyl ester	C	2010	2	157	65	Flowery
22	2,2-Dimethoxy-1.2-diphenyl, ethanone	C	2016	nd	3 2	35	nd

Note: [a]Dilution factor = 2^{n-1}, where n is the number of dilution (factor 2) until no odour was perceived.

The reliability of the identification process is indicated by the following: A, Mass spectrum and Kovats index agreed with standards; B, Mass spectrum and Kovats index agreed with the literature data; C, Mass spectrum agreed with mass spectral.

Nota: [a]Factor de disolución = 2^{n-1}, donde n es el número de disolución (factor 2) hasta que no se percibió ningún olor.

La fiabilidad del proceso de identificación se ha indicado mediante lo siguiente: A, Espectro de masas e Índice de Kovats en correspondencia con los estándares; B, Espectro de masas e Índice de Kovats en correspondencia con los datos de la literatura; C, Espectrometría de masas en correspondencia con el espectro de masas.

Table 5. Relationships between the key odorants (FD ≥ 16) of cassava flakes and "garri" and their odour thresholds (µg kg^{-1}) in starch.

Tabla 5. Relaciones entre los olores clave (FD ≥ 16) de los copos de mandioca y el "garri" y sus umbrales de olor (µg kg^{-1}) en el almidón.

Compounds	Odour threshold in starch	(Reference)	OAVs for Flakes	OAVs for "garri"
Methylpropanal	20	1	24.85	7.05
3-Methylbutanal	32	2	28.69	31.63
Butyric acid	45	2	2.4	20.42
2-Acetylpyrroline	1205	3	<1	<1
1-Octen-3-one	57	4	2.53	3.70
Guaiacol	4.2	5	20.48	82.14

Note: [1]Rychlik et al. (1998).

[2]Guth and Grosch (1994).

[3]Buttery, Ling, Juliano, and Turn-Baugh (1998).

[4]Konopka, Guth, and Grosch (1995).

[5]Cerny and Grosch (1993).

[OAVs]Odour Activity Values.

[OAVs]Valores de actividad olfativa.

and 1-octen-3-one, which exhibited high concentrations in the products, produced relatively low OAVs. Therefore, it is assumed that they are not contributing to the overall aroma of cassava flakes and garri.

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