

## Volatile constituents of cooked bullfrog (*Rana catesbeiana*) legs

Ian C.C. Nóbrega<sup>a,\*</sup>, Celene S. Ataíde<sup>a</sup>, Onofre M. Moura<sup>a</sup>, Alda V. Livera<sup>b</sup>,  
Paulo H. Menezes<sup>c</sup>

<sup>a</sup> Departamento de Tecnologia Rural, Universidade Federal da Paraíba, Campus III, Bananeiras 58220-000 PB, Brazil

<sup>b</sup> Departamento de Nutrição, Universidade Federal de Pernambuco, Recife, PE, Brazil

<sup>c</sup> Departamento de Química Fundamental, Universidade Federal de Pernambuco, Recife, PE, Brazil

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### Abstract

Volatile extracts obtained from pressure-cooked bullfrog legs by simultaneous steam distillation and solvent extraction were analyzed by gas chromatography–mass spectrometry. Although the raw bullfrog legs used in this study contained 0.6% fat only, the extracts were dominated (qualitatively and quantitatively) by lipid oxidation volatiles such as alkanals, alkenals, alkadienals, alkanols and alkenols. Few Maillard volatiles were found, amongst them 2-acetylthiazole, the only sulfur-containing compound found in the extract. This profile of volatiles may be explained by the high proportion of phospholipids and low concentration of sulfur amino acid cystine in raw bullfrog meat, as previously reported by other researchers. Odour activity values (OAV) of volatiles were also estimated and major OAV compounds were: (*E,E*)-2,4-decadienal, (*E,Z*)-2,4-decadienal, (*E,Z*)-2,6-nonadienal, 1-octanol, and (*E*)-2-nonenal. Some of these potent unsaturated aliphatic aldehydes have also been associated with chicken flavour. This may contribute to flavour resemblances between bullfrog and chicken meat.

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### 1. Introduction

*Rana catesbeiana*, known as bullfrog, is native to North America and was introduced to Brazil in 1935 (Vizotto, 1984). Bullfrogs are commercially reared in frog farms in Brazil, where there are approximately 600 of them (Lima, Cruz, & Moura, 1999). Their legs (including the thigh portion) are the major edible part and are considered by many to be a delicacy. The cooked meat is soft in texture, white in colour and its flavour is described as lightly sweet and bearing a close resemblance to the white meat of a young chicken (Herbst, 1995). However, the sensory qualities of bullfrog meat are rarely studied and the volatile compounds that may contribute to its aroma have not been investigated to date. Taking into account Brazil's potential

for producing and exporting bullfrog meat, such studies are of real economic interest.

Nevertheless, a few studies on the chemical composition of bullfrog meat, particularly fat and amino acid compositions which may be related to its flavour, are found in the literature. Bullfrog meat contains less than 1% fat and it is mainly composed (ca. 90%) of phospholipids (Corrêa, 1988; Lemos & Antunes, 1993; Lindau & Noll, 1988). The most abundant fatty acids reported in bullfrog meat are oleic (C18:1), linoleic (C18:3), arachidonic (C20:4) and palmitic (C16:0) (Coutinho, 2001; Lindau & Noll, 1988). Bullfrog meat has very low content of the amino acid cystine, at non-detectable levels in two studies (Azevedo & Oliveira, 1988; Corrêa, 1988). Phospholipids and cysteine (reduced form of cystine) are considered important flavour precursors as they actively participate in reactions (e.g. Maillard reaction and lipid oxidation) leading to (or controlling) sensorially relevant meat volatiles (Farmer & Mottram, 1990; Mottram & Nobrega, 2002).

\* Corresponding author. Tel.: +55 83 99678669.

E-mail address: [ian.nobrega@uol.com.br](mailto:ian.nobrega@uol.com.br) (I.C.C. Nóbrega).

In this paper we report on the identification and quantification of volatiles generated from cooked bullfrog legs and discuss their potential contribution to aroma.

## 2. Materials and methods

### 2.1. Animals and sample preparation

Spawns of *R. catesbeiana* were reared at the frog farm unit of the Universidade Federal da Paraíba (Bananeiras, PB, Brazil) under controlled conditions until froglets (newly metamorphosed tadpoles) were obtained. Then, approximately 1500 animals with average weight of 6 g were placed in a 30 m<sup>2</sup> shallow tank designed for the purpose, according to Lima and Agostinho (1992). The tank had a central pool, shelters and feeders. The experiment was carried out using natural light, with 12 h light/12 h dark, and environmental temperatures ranged from 22.6 to 29.9 °C. Froglets were fed on a diet composed of commercial extruded and granulated fodder (intended for fish). The fodder granules were of the same size (2 × 3 mm), shape and colour. Throughout the experimental period, the fodder granules were moved around by live *Musca domestica* larvae at an initial rate (first 15 days) of 1–1 (wt/wt, larvae-fodder) and then reduced to 1–20. Before slaughter, the animals were kept off their feed for 24 h, receiving only water. Then the animals with weight ranging from 180 to 200 g were randomly chosen in the tank, washed with chlorinated (5 ppm) water and stunned by electrical shock (60 V/60 Hz). Slaughtering was performed according to Moura (1999). Bullfrog legs (with the thigh portion) were separated, then, from the carcasses. Legs from several frogs were mixed and samples of approximately 220 g (equivalent to four pairs of legs), still on the bone, were vacuum packed into plastic laminated bags and stored at –20 °C.

### 2.2. Proximate analysis of raw bullfrog legs

Each sample ( $n = 3$ ), still in the packet, was thawed by immersing it in water at room temperature for approximately 30 min. Then legs were deboned and two replicate analysis of each sample were carried out. Moisture, protein, fat and ash analyses were carried out according to the AOAC (2002).

### 2.3. Analysis of volatiles from cooked bullfrog legs

Each sample ( $n = 3$ ) was thawed as described previously and placed into 500 ml glass bottles fitted with airtight, PTFE-lined screw tops. The sample was cooked at 140 °C in an autoclave for 30 min and afterwards allowed to cool at room temperature. Then the sample was boned and 100 g were used for extraction of volatiles. The cooking method used although not conventional, has the advantage of being reproducible and does not need any frying (i.e. oil, butter, etc.) that would interfere with the identification and

quantification of the sample volatiles. Furthermore, all volatiles generated during cooking could be trapped in the glass bottle.

The volatile compounds were isolated from the cooked meat samples by using simultaneous steam distillation and solvent extraction (SDE) in a Likens–Nickerson apparatus (Likens & Nickerson, 1964). The cooked sample (100 g) was blended with 350 ml of distilled water and the slurry transferred to a 1 l round bottom flask. Extractions were carried out for 2 h using a mixture of redistilled *n*-pentane (27 ml) and diethyl ether (3 ml) at 45 °C. After extraction, an internal standard (130 µg of 1,2-dichlorobenzene in 0.1 ml of diethyl ether) was added to the solvent extract which was concentrated to about 0.5 ml by distilling the solvents off the extract at 45 °C. Additional concentration to 0.2 ml was achieved by placing the extract in a gentle stream of nitrogen. A blank extraction was also carried out in which the meat sample was omitted.

A splitless injection was used to introduce 0.3 µl aliquots of the extract onto a silica capillary column (DB-5, 30 m × 0.25 mm i.d. × 0.25 µm film thickness; J & W Scientific) installed in a Shimadzu CG-17A gas chromatograph coupled to a GCMS-QP5050A mass spectrometer. Helium at 1.0 ml/min (36.3 cm/s) was used as carrier gas. The oven was initially kept at 50 °C for 2 min, and then programmed at 4 °C/min to 280 °C. The injector temperature was 250 °C, and the interface of the GC to the MS was maintained at 280 °C. The MS was operated in the electron impact mode with ionization energy of 70 eV and a scan rate of 2.0 scans/s over the mass range of 29–400 amu. A solution containing C<sub>7</sub>–C<sub>22</sub> *n*-alkanes was also analyzed under the same conditions to allow calculation of linear retention index (LRI) values for each sample component. Volatiles from samples were first identified by comparing their mass spectra with those contained in electronic mass spectral databases (NIST/EPA/NIH and Wiley) or in previously published literature and then comparing LRI values (DB5 column) with published values (DB5 or DB5-like columns). Approximate quantities of the volatiles were estimated by comparing their peak areas with those of the 1,2-dichlorobenzene internal standard, obtained from total ion chromatograms, using a response factor of 1.

## 3. Results and discussion

### 3.1. pH and proximate analysis of raw bullfrog legs

Table 1 shows the results of pH and proximate analyses of raw meat from bullfrog (*R. catesbeiana*) legs. Value for pH (6.2) is similar to that reported by Corrêa (1988) in

Table 1  
pH and proximate analyses<sup>a</sup> (percentage by wet weights) of raw meat from bullfrog legs

pH	Moisture	Ash	Protein	Fat
6.2 ± 0.0	74.1 ± 0.4	1.0 ± 0.0	19.4 ± 0.0	0.6 ± 0.0

<sup>a</sup> Data are expressed as means ± standard deviation.

bullfrog meat. The pH is within the range for most fish (6.2–6.6) and above that for warm-blooded animals (5.3–5.5) (Eskin, 1990). The fat content was low (0.6% wet weight or 2.4% dry weight) and is within the range (0.3–0.8%) reported by other researchers previously (Azevedo & Oliveira, 1988; Corrêa, 1988; Lemos & Antunes, 1993; Lindau & Noll, 1988).

### 3.2. Volatile constituents of cooked bullfrog legs

The relative percentage areas (RPA) and approximate concentrations ( $\mu\text{g}/\text{kg}$ ) of 40 volatile compounds extracted from cooked bullfrog legs are shown in Table 2. These compounds corresponded to 96% of total ion chromatogram area. Ten compounds were identified by mass spectrum matching (MS) and 30 by MS combined with linear retention index (LRI) matching (MS + LRI). Mass spectral data of compounds identified by MS only are also given in Table 2.

Saturated and unsaturated aliphatic aldehydes were quantitatively and qualitatively the most dominant group of volatiles in the SDE extract of cooked bullfrog legs. Twenty two compounds were aliphatic aldehydes (alkanals, alkenals, and alkadienals) and they accounted for 50% of total ion chromatogram (TIC) area. 15-Methylhexadecanal, (*Z*)-13-octadecenal, tetradecanoic acid, and 9-hexadecenoic acid were found particularly in high amounts and they accounted, respectively, for 9.8%, 7.9%, 12.0%, and 18.4% of TIC area (Table 2). However, these long-chain compounds usually have very low volatility and therefore are not expected to make a significant contribution to aroma. Interestingly, the methyl-branched long chain aldehydes 15-methylhexadecanal and 14-methylpentadecanal (Table 2) have been reported previously in beef, pork and chicken volatiles and are believed to contribute to cooked meat aroma giving “tallowy” and “fatty” characters (Werkhoff, Brüning, Emberger, Güntert, & Hopp, 1993), but their odour threshold values have not been reported.

Aliphatic aldehydes, alcohols, hydrocarbons, and ketones, all with straight alkyl chains containing five or more carbons, as well as alkylfurans, are all lipid-derived volatiles. These volatiles are formed by thermally induced oxidation of the fatty acid chains of phospholipids and triglycerides (Forss, 1972; Mottram, 1996). Interestingly, the bullfrog meat used in this study contained 0.6% fat only (Table 1). However, fat from bullfrog meat has a high proportion of phospholipids, approximately 90%, according to Corrêa (1988) and Lemos and Antunes (1993). Phospholipids contain a much higher proportion of unsaturated fatty acids than triglycerides (Farmer & Mottram, 1990; Meynier, Genot, & Gandemer, 1998) and are known to undergo oxidation much more readily than those which are saturated. This may explain the dominance of lipid-derived volatiles and the relatively high number of unsaturated compounds found in the bullfrog extract (Table 2). Additionally, the extraction method (SDE) might have some effect on the profile of volatiles found. The boiling of samples with large amounts of water associated with SDE

could create additional thermal decomposition of meat components, in particular hydrolysis of phospholipids and subsequent oxidation of free fatty acids. However, in this study the SDE was carried out at much lower temperature than the cooking process. Furthermore, it is considered an efficient extraction method in recovering volatile components (Leahy & Reineccius, 1984; Parliment, 1997).

Apparently, 2-acetylthiazole and 2-phenylacetaldehyde were the only Maillard volatiles found in the bullfrog extracts (Table 2). Apart from 2-acetylthiazole, no other sulfur-containing volatile was found. Maillard volatiles typically include heterocyclic nitrogen and sulfur compounds, such as pyrazines, thiophenes, and thiazoles, as well as furanones and furfurals. However, certain nonheterocyclic compounds are also Maillard-derived and these include Strecker aldehydes (e.g. phenylacetaldehyde) from the thermal decomposition of aminoacids in the presence of  $\alpha$ -dicarbonyl compounds. The low incidence of sulfur volatiles in the extract could be related to the very limited amounts of cystine in raw bullfrog meat, as reported by Azevedo and Oliveira (1988) and Corrêa (1988). Another possible reason could be the relatively high concentration of phospholipids in bullfrog meat (Corrêa, 1988; Lemos & Antunes, 1993) and their quenching effect on Maillard volatiles, as observed by several researchers in meat and meat-like systems (Farmer, Mottram, & Whitfield, 1989; Farmer & Mottram, 1990; Mottram & Edwards, 1983).

### 3.3. Potentially important aroma compounds and their origin

Reported threshold values (in water), reported odour descriptions, and estimated odour activity values (OAV, ratio of concentration of compound to threshold value) of 23 compounds found in cooked bullfrog legs are shown in Table 3.

On the basis of the estimated OAVs, the seven most potent compounds (OAV > 100) in cooked bullfrog legs from strong to weak were: (*E,E*)-2,4-decadienal, (*E,Z*)-2,4-decadienal, (*E,Z*)-2,6-nonadienal, 1-octanol, (*E*)-2-nonenal, 1-octen-3-ol, and (*E*)-2-decenal. (*E,E*)-2,4-Decadienal is by far the most important compound identified in the extract and its odour has been reported as “deep fried” (Table 3). Possible precursors of these high OAV compounds found in cooked bullfrog legs are  $n - 6$  and  $n - 9$  fatty acids (Grosch, 1987). The presence of these compounds correlates well with Lindau and Noll (1988) and Coutinho (2001) data, who reported oleic acid ( $n - 9$ ), linoleic ( $n - 6$ ), and arachidonic acids ( $n - 6$ ) as the most abundant fatty acids in bullfrog meat.

1-Octen-3-ol and (*E*)-2-nonenal may be formed from the oxidation of linoleic acid (C18:2), via 10-HPOD (10-hydroperoxy-8,12-octadecadienoic acid), or from the oxidation of arachidonic acid (C20:4), via 12-HPETE (12-hydroperoxy-5,8,10,14-eicosatetraenoic acid). (*E*)-2-Decenal and 1-octanol may be formed from the oxidation of oleic acid (C18:1), via 9-HPOE (9-hydroperoxy-10-octadecenoic acid) and 10-HPOE (10-hydroperoxy-8-octadecenoic acid),

Table 2

Relative percentage areas (RPA) and approximate concentrations ( $\mu\text{g}/\text{kg}$  of meat) of compounds found in SDE (simultaneous distillation-extraction) extracts of cooked bullfrog legs

Compound [ $m/z$ (relative intensity)] <sup>a</sup>	Amounts <sup>b</sup>		LRI <sup>d</sup>	Identification <sup>e</sup>
	RPA <sup>c</sup>	$\mu\text{g}/\text{kg}$		
Heptanal	3.8 $\pm$ 0.1	145 $\pm$ 33	900	MS <sup>2</sup> + IRL <sup>3,4</sup>
Octanal	2.7 $\pm$ 0.7	105 $\pm$ 49	1001	MS <sup>2</sup> + IRL <sup>5</sup>
Nonanal	5.0 $\pm$ 0.3	190 $\pm$ 50	1102	MS <sup>2</sup> + IRL <sup>3</sup>
Decanal	0.3 $\pm$ 0.0	13 $\pm$ 3	1204	MS <sup>2</sup> + IRL <sup>5</sup>
Dodecanal	0.2 $\pm$ 0.0	9 $\pm$ 1	1406	MS <sup>2</sup> + IRL <sup>5</sup>
Tetradecanal	0.9 $\pm$ 0.2	34 $\pm$ 1	1610	MS <sup>2</sup> + IRL <sup>3,4</sup>
Pentadecanal	0.5 $\pm$ 0.2	18 $\pm$ 3	1711	MS <sup>2</sup> + IRL <sup>8</sup>
14-Methylpentadecanal [57, 43(79), 82(77), 55(72), 56(61), 69(58), 83(56), 41(55), 68(45), 96(41)]	0.6 $\pm$ 0.3	20 $\pm$ 6	1776	MS <sup>9</sup>
15-Methylhexadecanal [57, 43(84), 82(81), 55(68), 41(67), 96(50), 68(48), 83(44), 69(42), 67(41)]	9.8 $\pm$ 3.3	358 $\pm$ 46	1815	MS <sup>9</sup>
Octadecanal [57, 43(87), 82(79), 55(63), 41(61), 96(50), 68(47), 83(42), 69(38), 71(38), 67(36), 29(36)]	0.5 $\pm$ 0.4	18 $\pm$ 11	2021	MS <sup>2</sup>
<i>Total alkanals (10)</i>	24.3	910		
( <i>E</i> )-2-Hexenal	0.2 $\pm$ 0.0	8 $\pm$ 1	844	MS <sup>2</sup> + LRI <sup>5</sup>
( <i>E</i> )-2-Octenal	3.6 $\pm$ 0.3	134 $\pm$ 17	1055	MS <sup>2</sup> + LRI <sup>5</sup>
( <i>E</i> )-2-Nonenal	1.6 $\pm$ 0.4	61 $\pm$ 29	1158	MS <sup>2</sup> + LRI <sup>5</sup>
( <i>E</i> )-2-Decenal	1.1 $\pm$ 0.0	40 $\pm$ 7	1260	MS <sup>2</sup> + LRI <sup>5</sup>
( <i>E</i> )-2-Undecenal	0.9 $\pm$ 0.0	35 $\pm$ 9	1362	MS <sup>2</sup> + LRI <sup>7,8</sup>
( <i>E</i> )-11-Hexadecenal [55, 41(73), 69(59), 67(52), 43(45), 81(42), 82(38), 83(38), 98(34), 29(33), 70(32)]	1.2 $\pm$ 0.1	45 $\pm$ 7	1793	MS <sup>2</sup>
( <i>E</i> )-9-Tetradecenal [55, 41(66), 69(55), 67(46), 83(42), 43(42), 98(41), 81(40), 54(32), 82(31), 56(29)]	0.3 $\pm$ 0.0	13 $\pm$ 2	1893	MS <sup>2</sup>
( <i>Z</i> )-13-Octadecenal [55, 41(72), 69(58), 43(53), 57(48), 83(46), 67(40), 98(37), 82(37), 81(35), 70(34)]	7.9 $\pm$ 1.1	293 $\pm$ 22	1995	MS <sup>2</sup>
<i>Total alkenals (8)</i>	16.8	629		
( <i>E,Z</i> )-2,6-Nonadienal	0.2 $\pm$ 0.1	5 $\pm$ 1	1152	MS <sup>2</sup> + LRI <sup>5</sup>
( <i>E,Z</i> )-2,4-Decadienal	0.8 $\pm$ 0.0	29 $\pm$ 5	1292	MS <sup>2</sup> + LRI <sup>5</sup>
( <i>E,E</i> )-2,4-Decadienal	3.6 $\pm$ 0.6	139 $\pm$ 9	1315	MS <sup>2</sup> + LRI <sup>5</sup>
( <i>Z,E</i> )-9,17-Octadecadienal [67, 81(71), 55(53), 41(49), 95(46), 54(42), 68(39), 82(38), 79(34)]	4.0 $\pm$ 1.6	144 $\pm$ 27	1989	MS <sup>2</sup>
<i>Total alkadienals (4)</i>	8.6	317		
Benzaldehyde	3.3 $\pm$ 0.2	123 $\pm$ 18	956	MS <sup>2</sup> + LRI <sup>6</sup>
2-Phenylacetaldehyde	0.5 $\pm$ 0.4	19 $\pm$ 12	1041	MS <sup>1</sup> + LRI <sup>6,7</sup>
4-Pentylbenzaldehyde [120, 91(97), 119(95), 92(72), 176(49), 65(28), 29(24), 105(21), 77(20), 41(18)]	0.3 $\pm$ 0.1	13 $\pm$ 7	1459	MS <sup>2</sup>
<i>Aromatic aldehydes (3)</i>	4.1	155		
2-Heptanone	0.3 $\pm$ 0.1	11 $\pm$ 6	886	MS <sup>1</sup> + LRI <sup>5</sup>
2-Nonanone	0.2 $\pm$ 0.0	7 $\pm$ 1	1090	MS <sup>2</sup> + LRI <sup>8</sup>
2-Decanone	0.5 $\pm$ 0.0	18 $\pm$ 5	1192	MS <sup>1</sup> + LRI <sup>3</sup>
<i>Total alkanones (3)</i>	1.0	36		
Dodecanoic acid	0.4 $\pm$ 0.1	15 $\pm$ 6	1565	MS <sup>2</sup> + LRI <sup>3</sup>
Tetradecanoic acid	12.0 $\pm$ 2.9	465 $\pm$ 208	1767	MS <sup>2</sup> + LRI <sup>3</sup>
Pentadecanoic acid [43, 73(99), 60(98), 57(74), 41(69), 55(63), 129(48), 69(44), 71(37), 87(34), 85(27)]	0.3 $\pm$ 0.1	12 $\pm$ 7	1826	MS <sup>2</sup>
9-Hexadecenoic acid	18.4 $\pm$ 3.3	708 $\pm$ 271	1949	MS <sup>2</sup> + LRI <sup>10</sup>
<i>Total alkanolic acids (4)</i>	31.1	1200		
1-Octen-3-ol	3.6 $\pm$ 0.5	139 $\pm$ 47	977	MS <sup>2</sup> + LRI <sup>3,5</sup>
1-Octanol	1.6 $\pm$ 0.2	60 $\pm$ 22	1070	MS <sup>2</sup> + LRI <sup>4</sup>
Nonanol	0.2 $\pm$ 0.1	9 $\pm$ 4	1172	MS <sup>2</sup> + LRI <sup>8</sup>
1-Nonen-3-ol	0.3 $\pm$ 0.1	11 $\pm$ 5	1280	MS <sup>2</sup> + LRI <sup>4</sup>
<i>Total alkanols and alkenols (4)</i>	5.7	219		
2-Pentylfuran	2.9 $\pm$ 0.2	108 $\pm$ 32	990	MS <sup>2</sup> + LRI <sup>3,6</sup>
2-Acetylthiazole	0.5 $\pm$ 0.3	18 $\pm$ 7	1016	MS <sup>2</sup> + LRI <sup>5,6</sup>
2-(1,1-dimethylethyl)-phenol [135, 107(94), 150(43), 91(24), 115(20), 77(13), 136(11), 65(11)]	0.2 $\pm$ 0.0	8 $\pm$ 4	1274	MS <sup>2</sup>

(continued on next page)

Table 2 (continued)

Compound [ <i>m/z</i> (relative intensity)] <sup>a</sup>	Amounts <sup>b</sup>		LRI <sup>d</sup>	Identification <sup>c</sup>
	RPA <sup>c</sup>	µg/kg		
Gamma dodecalactone	0.4 ± 0.1	17 ± 8	1676	MS <sup>2</sup> + LRI <sup>5</sup>
Total miscellaneous (4)	4.0	151		

<sup>a</sup> The most abundant ions of some compounds are cited in order of decreasing relative intensity.

<sup>b</sup> Amounts are expressed as means ± standard deviation.

<sup>c</sup> Relative percentage areas (RPA) were obtained by dividing peak areas of each compound by the area of the total ion chromatogram, excepting residual solvent, and then multiplied by 100.

<sup>d</sup> Linear retention index (LRI) values calculated in relation to the C<sub>7</sub>–C<sub>25</sub> *n*-alkanes for a DB-5 column.

<sup>e</sup> Mass spectrum (MS) or mass spectrum and linear retention index (MS + LRI) of compound agree with: <sup>1</sup>Wiley MS Database, <sup>2</sup>NIST/EPA/NIH MS database, <sup>3</sup>Lee et al. (1991), <sup>4</sup>Ramarathnam et al. (1993), <sup>5</sup>Rychlik et al. (1998), <sup>6</sup>Beal and Mottram (1994), <sup>7</sup>Gómez et al. (1993), <sup>8</sup>Acree and Heinrich (2004), <sup>9</sup>Werkhoff et al. (1993), and <sup>10</sup>Kondjoyan and Berdagué (1996).

Table 3

Some volatiles found in cooked bullfrog legs with their published odour thresholds, odour description, and estimated odour activity value

Compound <sup>a</sup>	Odour threshold <sup>b</sup> (µg/kg)	Odour description <sup>c</sup>	Odour activity value <sup>d</sup>
( <i>E,E</i> )-2,4-Decadienal	0.07	Deep-fried	1986
( <i>E,Z</i> )-2,4-Decadienal	0.04	Deep-fried	725
( <i>E,Z</i> )-2,6-Nonadienal	0.01	Cucumber-like	530
1-Octanol	0.13	Moss, nut, mushroom	462
( <i>E</i> )-2-Nonenal	0.15	Tallowy, cucumber-like	410
1-Octen-3-ol	1.0	Mushroom-like	139
( <i>E</i> )-2-Decenal	0.4	Tallowy, orange-like	101
Heptanal	3.0	Fatty	48
Nonanal	5.0	Tallowy, fruity	38
( <i>E</i> )-2-Octenal	4.0	Fatty, nutty	33
2-Pentylfuran	6.0	Buttery, green bean-like	18
Octanal	8.0	Fatty	13
Decanal	2.0	Orange skin-like, flowery	6
2-Phenylacetaldehyde	4.0	Honey-like, flowery	5
Dodecanal	2.0	Fatty, citrus-like	4
Gamma dodecalactone	7.0	Green, fruity	2.5
2-Acetylthiazole	10.0	Roasty, sulfury	1.8
Benzaldehyde	350	Almond, burnt sugar	0.4
( <i>E</i> )-2-Hexenal	50	Apple-like	0.2
2-Heptanone	140	Soapy, fruity	0.08
2-Nonanone	200	Soapy, fruity	0.04

<sup>a</sup> Compounds were sequenced (decreasing order) according to their estimated odour activity values.

<sup>b</sup> Orthonasal odour thresholds (in water) of compounds obtained from Rychlik et al. (1998), excepting (*E,Z*)-2,4-decadienal (Kerscher & Grosch, 2000), 1-octanol, and benzaldehyde (van Gemert & Nettenbreijer, 1977).

<sup>c</sup> Odour description of compounds obtained from Rychlik et al. (1998), excepting 1-octanol and benzaldehyde (Acree & Heinrich, 2004).

<sup>d</sup> Odour activity values were estimated by dividing the concentrations of compounds (µg/kg, Table 2) by their published odour threshold in water.

respectively. (*E,E*)-2,4-Decadienal may be formed from oxidation of either linoleic acid, via 9-HPOD (9-hydroperoxy-9,11-octadecadienoic acid), or arachidonic acid, via 11-HPETE (11-hydroperoxy-5,8,12,14-eicosatetraenoic acid) (Frankel, 1982; Grosch, 1987). Origins of (*E,Z*)-2,4-decadienal and (*E,Z*)-2,6-nonadienal are less clear, but they are oxidation products of linoleic and linolenic acids, respectively (Grosch, 1987).

It is of interest to highlight the literature survey by Mottram (1991) on the profile of aldehydes that had been reported in the flavour of beef, pork, lamb and chicken. He showed that chicken meat had proportionally higher levels of unsaturated aliphatic aldehydes, in particular alkadienals and alkenals, and this may be important to its characteristic aroma. The three most important aroma compounds identified in the bullfrog meat were alkadienals (Table 3) and this might explain the resemblances between the flavour of bullfrog and chicken meat, as described by

Herbst (1995). Interestingly, in a comparison of the aromas of cooked beef, pork and chicken by Kerscher and Grosch (2000), (*E,Z*)-2,6-nonadienal showed relatively high OAV in chicken, whereas in pork and beef the values for this compound were negligible, implying that this compound may be important to the characteristic aroma of chicken meat. (*E,Z*)-2,6-Nonadienal was the third most potent aroma compound found in the bullfrog meat (Table 3) which may particularly contribute to its resemblance to chicken meat.

#### 4. Conclusions

Despite the very low levels of lipids in raw bullfrog meat, its aroma extracts were quantitatively and qualitatively dominated by saturated and unsaturated aliphatic volatiles derived from lipid oxidation. Few Maillard volatiles were found, amongst them 2-acetylthiazole. These results might correlate to the high proportion of phospholipids and low



concentrations of cystine in raw bullfrog meat, as reported by other authors previously.

Based on estimated odour activity values, the three most potent aroma compounds identified in the extracts were (*E,E*)-2,4-decadienal, (*E,Z*)-2,4-decadienal, and (*E,Z*)-2,6-nonadienal. Alkadienals and other unsaturated aliphatic aldehydes are frequently associated with chicken meat and therefore this may contribute to flavour resemblances between bullfrog and chicken meat.

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