

Volatile compounds of rehydrated French beans, bell peppers and leeks. Part II. Gas chromatography/sniffing port analysis and sensory evaluation

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The flavours of rehydrated diced French beans, red bell peppers and leeks were characterised by gas chromatography/sniffing port analysis (GC/SP) of volatile compounds released in a mouth model system, and by descriptive sensory analysis. Volatile compounds were identified by combined gas chromatography/mass spectrometry. In French beans, bell peppers and leeks, respectively, 10, 16 and 22 compounds possessed detectable odours. A common odour profile was shown in the three vegetables. It comprised each of the odour active compounds present in French beans: i.e. 2-methylpropanal (chocolate), 2/3-methylbutanal (chocolate), 2,3-butanedione (caramel, fatty), hexanal (grassy, bell pepper), 2-methyl-2-butenal (chemical), octanal (sweet, sickly/musty, grassy, rancid), 1-octen-3-one (mushroom), dimethyl trisulphide (rotten, metal), 1-octen-3-ol (fatty, sickly/musty, mushroom) and one unknown compound (chemical, rotten, rancid). The three vegetables differed markedly in GC/SP patterns and in scores for sensory attributes. Use of nose-clips diminished the scores for attributes in sensory analysis. In principal component analysis, correlation of rehydrated vegetables with sensory attributes and volatile compounds showed considerable contribution of volatile compounds to the flavour of rehydrated vegetables.

INTRODUCTION

Foods have highly complex chemical compositions, containing both volatile and non-volatile substances. Their sensory responses may be significantly affected by their relative concentrations and the temperature of the product as consumed (Heath, 1981). Human taste-buds are capable of differentiating only four stimuli. As the nose is capable of discerning hundreds of different odours, it is not surprising that a major part of flavour research has dealt with analysis of volatile compounds (Hoff *et al.*, 1978).

An instrumental approach to characterise flavour can be regarded as a two-phase arrangement. The first phase involves representative isolation of volatile compounds, because gas chromatographic (GC) patterns are largely influenced by isolation procedures. Release rates of volatile compounds from a product depend upon partition coefficients of the compound, molecular interactions and ambient temperatures (Legger & Roozen, 1994). The second phase involves selection of volatile compounds relevant to the flavour. This

implicates correct determination of the relevant flavour compounds from the whole range of volatiles present in a particular food product (Dirinck & De Winne, 1994). An interesting approach is sniffing the gas chromatographic effluent (GC/sniffing port analysis; GC/SP), in order to associate odour activity with the eluting compounds. Flame ionisation, flame photometric, electron capture, mass spectrometric, fourier transform, infrared and thermal conductivity detectors are not as sensitive as the human nose for many odorants (Acree & Barnard, 1994). The interest in determining the individual contribution of volatile compounds present in foods, has led to a new generation of GC/SP techniques, which can be classified into four categories:

- (i) Dilution analysis methods for producing titre or potency values based on stepwise dilution to threshold, e.g. CHARM (Acree *et al.*, 1984) and aroma extraction dilution analysis (Ullrich & Grosch, 1987).
- (ii) Response interval methods for recording time duration of perceived odours, and the number of assessors with odour perception, it also estimates a titre or potency (Linssen *et al.*, 1993).

- (iii) Time-intensity methods for producing subjective estimates of perceived intensity recorded simultaneously with the elution of the chromatographic peak, e.g. OSME (Sanchez *et al.*, 1992).
- (iv) Posterior intensity methods for producing subjective estimates of perceived intensity, which are recorded after a peak has eluted (Casimir & Whitfield, 1978).

Correlation between sensory and instrumental flavour data has been attempted by several multivariate statistical techniques, like factor and principal component analyses (Noble, 1978). Such techniques have been used to reduce large numbers of sensory and instrumental variables to a smaller number of factors, or to study underlying patterns in the variables (Palmer, 1974; Guedes de Pinho *et al.*, 1994; Luning *et al.*, 1994a).

Recently, the authors reported a dynamic headspace model system for isolation of volatile compounds from vegetables under mouth conditions. Using this model system, release of volatiles from rehydrated French beans, red bell peppers and leeks did not differ significantly from their release in the mouth of 12 assessors (Van Ruth *et al.*, 1995). The present study deals with the relationship between GC/SP detected volatile compounds released in the reported mouth model system from rehydrated French beans, red bell peppers and leeks, and the sensory properties of these vegetables. This relationship was examined by multivariate statistical techniques.

MATERIALS AND METHODS

Plant material

Commercially dried French beans and leeks from The Netherlands and red bell peppers from Hungary were supplied in pieces (8 mm × 8 mm) by Top Foods BV (Elburg, The Netherlands). The vegetables were packed in glass jars and stored at 4°C in the absence of light until sampling. Diced dried vegetables (1.2 g) were rehydrated by adding 10 ml distilled water, followed by heating in a waterbath at 100°C for 10 min and cooling down in a waterbath at 25°C for 4 min.

Instrumental analysis

The rehydrated vegetables were transferred into a dynamic headspace mouth model system with mastication device. Artificial saliva (4 ml) was added and the headspace was flushed with nitrogen gas (250 ml/min) for 12 min in order to trap volatiles in Tenax TA as described previously (Van Ruth *et al.*, 1995). A plunger made about four up and down screwing movements per min to simulate mastication. In GC/SP, desorption of volatile compounds from Tenax was performed by a thermal desorption (210°C, 5 min)/cold trap (-120°C/240°C) device (Carlo Erba TDAS 5000, Interscience BV, Breda, The Netherlands). Gas chromatography was

carried out on a Carlo Erba MEGA 5300 (Interscience BV, Breda, The Netherlands) equipped with a Supelcowax 10 capillary column, 0.25 mm i.d. and 60 m long and a flame ionisation detector (FID) at 275°C. At the end of the capillary column the effluent was split 1:2:2 for FID, sniffing port 1 and sniffing port 2, respectively. Oven temperature programme and sniffing port detection were performed as reported previously (Van Ruth & Roozen, 1994). In preliminary GC/SP experiments, 10 assessors (aged 20–40) generated flavour descriptors for rehydrated French beans, bell peppers and leeks, which were clustered after group sessions of the panel. Besides 'other/I do not know', one of these descriptors (Table 1) had to be used for each compound detected by the assessors at the sniffing port. Tenax tubes without adsorbed volatile compounds were used as dummy samples for determining signal-to-noise level of the group of assessors.

The volatile compounds trapped in Tenax TA were identified by combined GC (Varian 3400, Varian, Walnut Creek, CA, USA) and mass spectrometry (MS; Finnigan MAT 95, Finnigan MAT, Bremen, Germany) equipped with a thermal desorption/cold trap device (TCT injector 16200, Chrompack BV, Middelburg, The Netherlands). Capillary column and oven temperature program were the same as those used in GC/SP analyses. Mass spectra were obtained with 70 eV electron impact ionisation, while the mass spectrometer was continuously scanning from *m/z* 24 to 400 at a scan speed of 0.7 s/decade (cycle time 1.05 s).

Table 1. Comparison of the descriptors of the gas chromatography/sniffing port panel (GC/SP) and the attributes of the analytical sensory panel for three rehydrated vegetables

Descriptors (GC/SP panel)	Attributes (sensory panel)
Bell pepper	Bell pepper
Burned	Bitter
Caramel	Burned
Chemical	Chemical
Chocolate	
Citrus	Citrus
Cooked vegetables	Cooked vegetables
Cucumber	
Detergent	
Fatty	
French bean	French bean
Fruity	
Garlic	
Grassy	Grassy
Herbal	
Leek	Leek
Metal	
Mushroom	Mushroom
Onion	Onion
Rancid	
Rotten	Rotten
	Sharp
Sickly/musty	
Sour	Sour
Spicy	Spicy
Sweet	Sweet

Sensory evaluation

A panel of 21 judges (aged 20–55) was selected and trained for quantitative descriptive analysis (QDA). A computer interactive interviewing system for composing questionnaires was used to gather survey information (Ci2 system; Sawtooth Software Inc., Ketchum, ID, USA). Samples of each of the diced dried vegetables (1.8 g) were rehydrated by adding 15 ml water and heating in a waterbath at 100°C for 10 min. They were cooled down and stored in a waterbath at 50°C until they were served to the panel and assessed for flavour evaluation. Flavour attributes were generated during training sessions, which resulted in a list of 16 attributes (Table 1). Sensory attributes of each of the rehydrated diced vegetables were evaluated with and without use of nose-clips (Jaeger Nederland BV, Breda, The Netherlands) by scoring perceived intensities on a 70 mm visual analogue scale on a portable computer screen.

Statistical evaluation

Sensory data were subjected to Student's *t*-tests to determine significant differences between the three vegetables. The SPSS/PC+ program (SPSS Inc., Chicago, IL, USA) was used for principal component analysis (PCA) of the sensory data as well as for correlation of instrumental and sensory data. A significance level of $P < 0.05$ was used throughout the study.

RESULTS AND DISCUSSION

Volatile compounds of rehydrated diced French beans, bell peppers and leeks were isolated in the mouth model system and analysed by GC/SP. Figure 1 represents the sniffing chromatograms of each of the three vegetables. The volatile compounds were identified by GC/MS and their retention times, and characterised by their FID peak areas and the odours described by the assessors of the sniffing panel (Table 2). GC sniffing of dummy samples showed that detection of an odour at the sniffing port by three or less out of 10 assessors can be considered as 'noise'. GC/SP revealed that 10, 16 and 22 volatile compounds isolated from French beans, bell peppers and leeks, respectively, possessed detectable odours. Although differences in GC/SP patterns across the vegetables were obvious, a common GC/SP profile was shown in the three vegetables. It incorporates all odour active compounds isolated from French beans: 2-methylpropanal (chocolate), 2/3-methylbutanal (chocolate), 2,3-butanedione (caramel, fatty), hexanal (grassy, bell pepper), 2-methyl-2-butenal (chemical), unknown (chemical, rotten, rancid), octanal (sweet, sickly/musty, grassy, rancid), 1-octen-3-one (mushroom), dimethyl trisulphide (rotten, metal) and 1-octen-3-ol (fatty, sickly/musty, mushroom). The presence of 2-methylpropanal and 2/3-methylbutanal is typical for dried vegetables. These volatile compounds can be formed by Strecker degradation of valine and (iso)leucine during

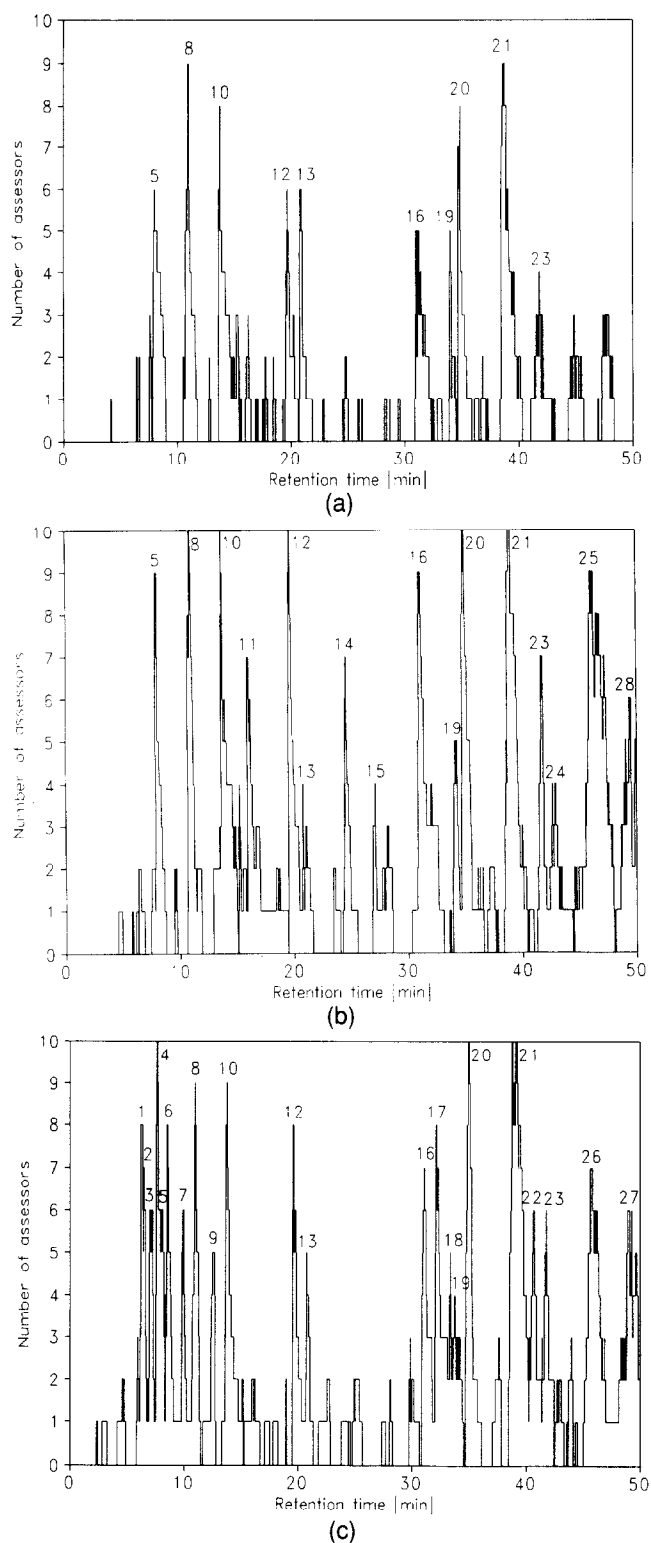


Fig. 1. Sniffing chromatograms of volatile compounds of rehydrated diced (a) French beans, (b) bell peppers, and (c) leeks. Numbers in the chromatograms refer to compounds in Table 2.

the drying process. The other components of the common odour profile can be formed mostly in lipid oxidation reactions (Tressl *et al.*, 1981; Whitfield, 1992). Most compounds of the profile possess low thresholds in water (< 5 mg/kg) (Leffingwell & Leffingwell, 1991).

Many of the compounds with detectable odours were identified in Part 1 of the present study (Van Ruth *et*

Table 2. Odour active volatile compounds of rehydrated diced French beans, bell peppers and leeks; their retention times and odour descriptions

No.	Retention time (min)	Compound	Odour description		
			French beans	Bell peppers	Leeks
1	6.3	Methanethiol	—	—	Rotten
2	6.7	Carbon disulphide	—	—	Sweet, chemical
3	7.2	Dimethyl sulphide	—	—	Rotten, cooked vegetables, leek, spicy
4	7.7	Propanal	—	—	Cooked vegetables, grassy, chemical
5	8.0	2-Methylpropanal	Chocolate	Chocolate	Chocolate
6	8.7	1-Propanethiol	—	—	Onion
7	10.0	2-Propene-1-thiol	—	—	Garlic
8	11.1	2/3-Methylbutanal	Chocolate	Chocolate	Chocolate, spicy
9	12.5	3-(Methylthio)-1-propene	—	—	Onion, rotten, herbal, chemical
10	13.9	2, 3-Butanedione	Caramel, fatty	Caramel	Caramel
11	16.1	1-Penten-3-one	—	Chemical, metal	—
12	19.8	Hexanal	Grassy, bell pepper	Grassy, bell pepper	Grassy
13	20.9	2-Methyl-2-butenal	Chemical	Chemical, metal, leek	Rancid, cooked vegetables, onion
14	24.9	Unknown	—	Citrus, fruity, rotten	—
15	27.2	2-Heptanone/heptanal	—	Bell pepper, cooked vegetables, cucumber	—
16	31.5	Unknown	Chemical, rotten, rancid	Rotten, rancid, sickly/musty, chemical, citrus, leek	Rancid, rotten, chemical, sickly/musty, onion
17	32.6	(Z)-1-Methyl-propenyldisulphide	—	—	Chemical, onion, rotten, herbal, burned
18	33.4	(E)-1-Methyl-propenyldisulphide	—	—	Rotten, garlic, leek
19	34.0	Octanal	Sweet, sickly/musty, grassy, rancid	Citrus, cucumber, leek	Fatty, citrus, sweet
20	35.2	1-Octen-3-one ^a	Mushroom	Mushroom	Mushroom
21	39.3	Dimethyl trisulphide	Rotten, metal	Rotten, metal	Rotten, onion, leek, metal
22	40.6	Propenyl propyl disulphide	—	—	Herbal, garlic, leek, fatty
23	41.7	1-Octen-3-ol	Fatty, sickly/musty, mushroom	Sickly/musty, fatty, rancid, bell pepper	Fatty, herbal, leek
24	42.7	2, 4-Heptadienal	—	Fatty, sickly/musty	—
25	46.1	2-Isobutyl-3-methoxypyrazine	—	Bell pepper	—
26	46.6	Unknown	—	—	Onion
27	49.1	Dipropyl trisulphide	—	—	Onion, garlic
28	49.6	2, 4-Nonadienal	—	Cucumber, cooked vegetables, grassy	—

^a1-Octen-3-one eluted simultaneously with 3-hepten-2-one in bell peppers.

al., 1995). Some of these compounds contribute to the flavour of the vegetables, despite their very low FID responses. 2-Isobutyl-3-methoxypyrazine is present in the rehydrated bell peppers and has a characteristic bell pepper odour. Several sulphur-containing volatile compounds in leeks possess leek odours. However, no volatile compounds identified in rehydrated French beans possessed a particular French bean flavour. The odour active compounds 2-methylpropanal and 2/3-methylbutanal, present in the French beans, have been reported in dried beans previously (Lovegren *et al.*, 1979). Stevens *et al.* (1967) and Toya *et al.* (1974) reported that 1-octen-3-ol belongs to the most important flavour compounds of canned snap beans. Several of the odour active compounds of bell peppers were reported previously (Wu *et al.*, 1986; Luning *et al.*, 1994b). The bell pepper odour of 2-isobutyl-3-

methoxypyrazine is well known (Buttery *et al.*, 1969; Luning *et al.*, 1994a). Sulphur-containing components are the main odour active compounds of rehydrated leeks. Several of these compounds were reported by Schreyen *et al.* (1976a,b), i.e. 1-propanethiol, methyl propenyl disulphide, propenyl propyl disulphide and dipropyl trisulphide, each of them possessing leek/onion odours.

An analytical sensory panel of 21 judges evaluated the rehydrated diced French beans, red bell peppers and leeks. Although individual judges responded differently to the three vegetables, the panel was consistent across replications. Most of the attributes generated by the sensory panel were similar to the descriptors of the GC/SP panel (Table 1). Apparently, volatile compounds contribute to the flavour of French beans, bell peppers and leeks. The results of the sensory evaluation

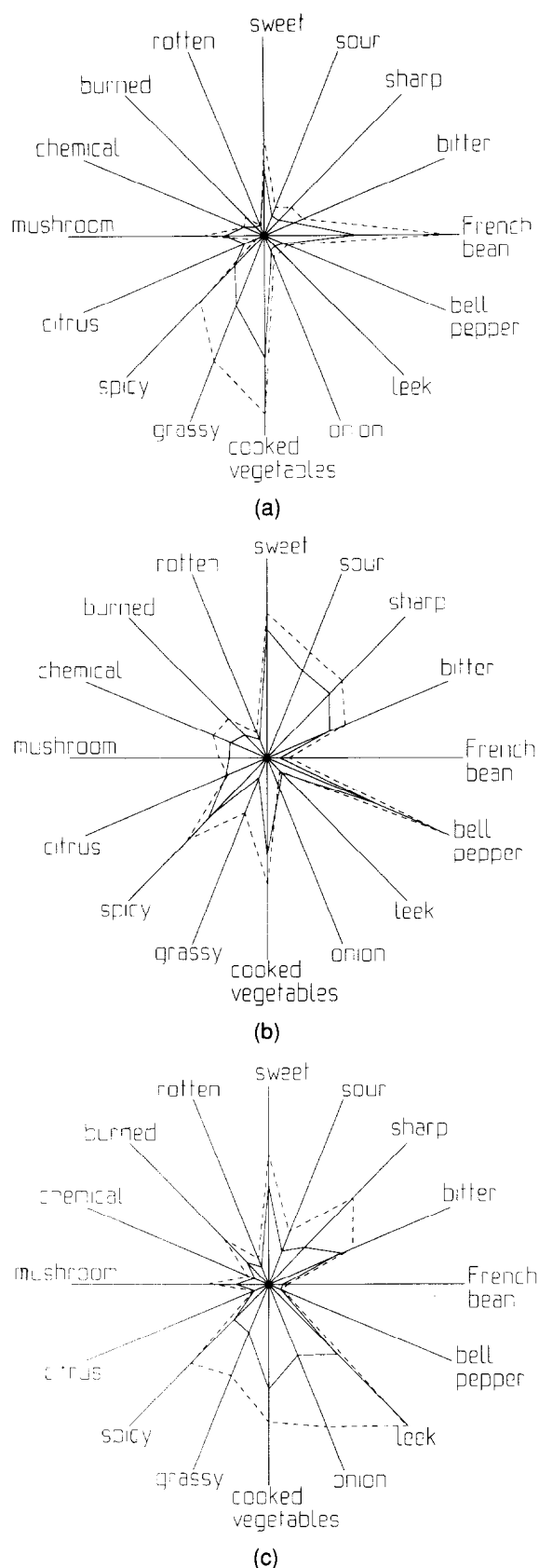


Fig. 2. Spider-web diagrams of scores for sensory attributes of rehydrated diced (a) French beans, (b) bell peppers, and (c) leeks assessed (—) with and (-----) without nose-clips.

of each of the vegetables are presented in spider-web diagrams (Figs 2(a)–(c)). Use of nose-clips diminished scores for sensory attributes in general, which is in

agreement with packaging migration studies of Linssen *et al.* (1991). This confirms our suggestion that the contribution of volatile compounds to the flavour of the vegetables is significant. As expected, the scores for taste attributes, like ‘sweet’, ‘sour’, ‘sharp’ and ‘bitter’, as well as for the particular vegetable less relevant attributes (indicated by low intensity scores with use of nose-clips) decreased less than others.

Mean scores for the sensory attributes for each of the vegetables and the statistical significances of mutual differences (Students’s *t*-test, $P < 0.05$) are shown in Table 3. Intensity scores for the bell peppers were similar to those reported previously (Van Ruth & Roozen, 1994). French beans scored high in ‘French bean’, ‘cooked vegetables’, ‘grassy’, ‘sweet’ and ‘spicy’ attributes, the bell peppers in ‘bell pepper’, ‘sweet’, ‘cooked vegetables’, ‘spicy’, ‘sour’, ‘sharp’ and ‘bitter’ and leeks in ‘leek’, ‘onion’, ‘cooked vegetables’, ‘sweet’, ‘sharp’, ‘spicy’ and ‘grassy’. The three vegetables did not differ significantly in scores for the attributes ‘spicy’ and ‘mushroom’, however, each of the other attributes revealed significant mutual differences between the vegetables. Sensory data were subjected to PCA, the first five principal components of which are shown in Table 4. PCA shows relationships between attributes: ‘leek’ and ‘onion’ loaded high on the first component, indicating that these attributes were strongly associated with each other. ‘Sweet’, ‘sour’, ‘citrus’ and ‘bell pepper’ loaded high on the second component. Two associated attributes were determined for each of the other three components.

PCA was performed on combined GC/SP (number of assessors perceiving an odour active compound) and QDA sensory data sets (intensities of sensory attributes perceived without nose-clips) (Fig. 3). The first two principal components explained 75% of the variance in the data sets. Vegetable sample scores in the PCA map

Table 3. Scores for sensory attributes of rehydrated diced French beans, bell peppers and leeks, assessed without nose-clips (mean, $n = 21$)^a

Code	Attribute	French beans	Bell peppers	Leeks
A	Sweet	30.2a	42.6b	39.3b
B	Sour	9.1a	35.8b	16.6a
C	Sharp	11.8a	33.4b	37.4b
D	Bitter	12.6a	26.4b	28.1b
E	French bean	56.6b	7.2a	6.3a
F	Bell pepper	7.1a	59.0b	4.9a
G	Leek	5.6a	5.9a	60.9b
H	Onion	5.5a	8.8a	47.5b
I	Cooked vegetables	54.0b	38.4a	42.3a
J	Grassy	41.1c	18.3a	30.0b
K	Spicy	28.7a	36.0a	34.3a
L	Citrus	4.0a	12.9b	4.5a
M	Mushroom	19.4a	16.2a	17.5a
N	Chemical	7.1a	17.5b	7.5a
O	Burned	7.9a	16.8b	19.3bc
P	Rotten	3.6a	8.1ab	8.2b

^aDifferent following letters within a row indicate significant differences (Student’s *t*-test, $P < 0.05$).

Table 4. Attributes of the first five principal components concerning rehydrated diced French beans, bell peppers and leeks (loadings in parentheses)

First 25% ^a	Second 16%	Third 13%	Fourth 9%	Fifth 7%
Leek (0.9)	Sweet (0.7)	Grassy (0.9)	Burned (0.8)	Mushroom (0.7)
Onion (0.9)	Sour (0.7)	Cooked vegetables (0.8)	Chemical (0.7)	Rotten (0.7)
	Citrus (0.7)			
	Bell pepper (0.7)			

^aPercentage of explained variance.

show that leeks (LEEK) was separated from bell peppers (BELL) and French beans (BEAN) along the first component. BELL and LEEK were separated along the second component. The diagram reveals three sites in which vegetable sample, volatile compounds and sensory attributes are closely related to each other. The following numbers refer to volatile compounds in Table 2 and capitals refer to sensory attributes in Table 3. 'French bean' (E) and 'cooked vegetables' (I) showed high negative loadings on both the first and second component and correlated well with BEAN. BELL correlated well with 2-methoxy-3-isobutylpyrazine (25) and 'bell pepper' (F). The compound 2-methoxy-3-isobutylpyrazine was described by the GC/SP panel as bell pepper. Sensory attributes 'onion' (H) and 'leek' (G), as well as several volatile compounds having leek

or onion odours according to GC/SP, have high loadings on the first component and low loadings on the second. These sensory attributes and volatile compounds, among which are methanethiol (1), 3-(methylthio)-1-propene (9) and propenyl propyl disulphide (22), correlated with LEEK. Correlation between the rehydrated vegetables, sensory attributes and several volatile compounds having corresponding odours according to GC/SP, showed that volatile compounds contribute considerably to the flavour of rehydrated vegetables. This is in agreement with studies of Noble (1978), in which a correlation between sensory and instrumental data of wines was determined and Rothe *et al.* (1994), who reported relationships between volatile compounds and blue cheese flavour. Investigations of Teule and Crouzet (1994) showed a relationship between volatile compounds and formation of cooked aroma in dehydrated apple products. Burdach and Doty (1987) reported that persons who have lost their sense of smell, frequently perceive this loss as one of taste, rather than one of smell. This indicates that stimulation of the olfactory receptors by foods via the retronasal route is a primary determinant of their flavour. In GC/SP the volatile compounds are sampled separately by the assessors in GC elution sequence. Nevertheless the instrumental data of GC/SP of volatiles released in a mouth model system with mastication device correlated quite well with sensory data.

CONCLUSIONS

The presence of a common odour profile was shown in the three rehydrated vegetables by GC/SP. This profile

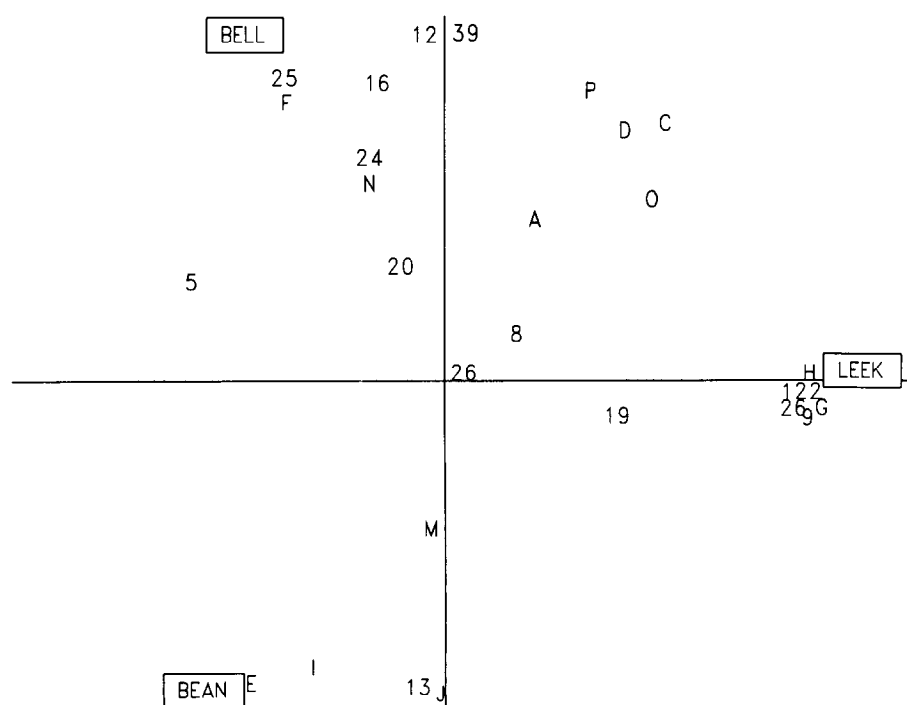


Fig. 3. Scores of vegetable samples (BEAN, French beans; BELL, bell peppers; and LEEK, leeks) and loadings of volatile compounds and sensory attributes on the first (horizontal) and second (vertical) principal component axes. Numbers refer to volatile compounds in Table 2 and capitals to sensory attributes in Table 3.

incorporates each of the odour active compounds of rehydrated French beans. Correlation between rehydrated vegetables, sensory attributes and several volatile compounds isolated in a mouth model system, showed major contribution of volatile compounds to the flavour of rehydrated vegetables.

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