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A Kinetic Study on Benzoic Acid Pungency and Sensory Attributes of Benzoic Acid

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Abstract

Aqueous solutions of benzoic acid (BA) were evaluated by two methods: (i) sensory profile: a descriptive test of sensory attributes combined with semiquantitative analysis; and (ii) pungency intensity measures as a function of time: a computerized recording using specific software. Kinetic parameters evaluated were maximal intensity (I_{MAX}), total time of pungency (T_{tot}), rates of increase (V_1) and decrease (V_2), half-life ($T_{½}$), area under curve (AUC) and time to maximal intensity (T_{IMAX}). Results were analyzed by ANOVA, LSD test, iterative calculations and adjustment to equations according to mathematical models, regression analysis, principal component analysis (PCA) and clusters analysis. Pungency was the main sensory attribute of BA (3–36 mM) in the tongue and epiglottis. The seven kinetic parameters showed concentration-dependency ($P < 0.001$) and were described by different functions: (i) lineal: $I_{MAX} = 2.24 \pm 0.14C - 3.06 \pm 2.58$, $R^2 = 0.98$; $T_{IMAX} = 0.19 \pm 0.02C + 6.87 \pm 0.47$, $R^2 = 0.92$; $V_1 = 0.68 \pm 0.03C + 0.10 \pm 0.69$, $R^2 = 0.99$; $AUC = 49.10 \pm 3.17C - 230.78 \pm 59.66$, $R^2 = 0.98$; (ii) potency: $T_{½} = 6.62 \pm 0.61C^{0.39\pm0.03}$, $R^2 = 0.97$; $V_2 = 1.07 \pm 0.11C^{0.53\pm0.04}$, $R^2 = 0.98$; $T_{tot} = 8.08 \pm 1.01C^{0.43\pm0.04}$, $R^2 = 0.96$. PCA revealed high correlation between (i) T_{IMAX} and T_{tot}; (ii) $T_{½}$ and V_2; and (iii) I_{MAX} and V_1. Stimuli grouped across three main clusters: (i) 3 and 6 mM; (ii) 9, 12 and 18 mM; and (iii) 24 and 36 mM. Maximal pungency intensity best correlated with both concentration and persistence among kinetic parameters. Prototypical prickling of BA was observed at 12 and 18 mM.

Introduction

Benzoic acid (BA) is a well-known additive used in preserving food and beverages. However, the sensory characteristics of benzoic acid are unknown. In other words, is there any possibility for BA to activate gustatory receptors and/or other, mainly trigeminal, receptors sensitive to chemical substances different from taste receptors (Green and Lawless, 1991)? In particular, there is previous evidence that BA stimulates the common chemical sense through the activation of trigeminal nerve endings (Peleg and Noble, 1995) that coexist with taste cells (Parker, 1912), inducing mild pungency, or prickling.

From an anatomical point of view trigeminal receptors are located on bare free nerve endings associated with taste buds (Bartoshuk, 1993). In view of its chemical structure, BA (an organic acid) might induce either sourness (taste) or irritation (non-taste or pungency) due to common chemical stimulation. Moreover, all its hydroxy derivatives—salicylic (2-hydroxy), *m*-hydroxy (3-hydroxy), gentisic (2,5-dihydroxy), protocatechuic (3,4-dihydroxy) and gallic (3,4,5-trihydroxy) acids—induce sourness, astringency and prickling (pungency) with relative different intensities (Peleg and Noble, 1995). The study by Peleg and Noble (1995) points to BA having a possible pungency.

BA and BA hydroxy derivatives are widespread among spices (hot spices and a majority of seasonings), grains, vegetables and fruits (grapes, figs, plums, raspberries and mulberries in general) (Hermann, 1989; Louekari *et al.*, 1990). Different classes of tannins, i.e. hydroxybenzoic acid-conjugated polyphenols, among other substances, are known astringents in beverages like tea or wine. Yet, it is not clear whether astringency is a trigeminal sensation.

The food industry uses BA as an inhibitor of microbial growth in fruit juice, soft drinks, liquid non-nourishing sweeteners, caviar and packed fish from Germany, and a great majority of preserves (Louekari *et al.*, 1990). However, it has not yet assessed the sensory characteristics of BA.

Sensory perception is not a static but a dynamic process. Recording the intensity of a sensation as a function of time is an extension of classic scaling methods, producing temporal information with the added advantage of not resorting to numbers to equalize sensations (Lee and Pangborn, 1986). A great majority of studies refer to the burning sensation induced by irritating substances such as pepper, ginger, alcohol or cinnamon (Lawless, 1984; Green, 1988; Hutchinson *et al.*, 1990; Nasrawi and Pangborn, 1990; Cliff and Heymann, 1992). However, there is no kinetic data available on benzoic acid. In fact this type of pungency, i.e. a tickling, crawling, prickling sensation, has not been studied. Some studies have described astringency in organic acids (acetic, lactic, citric, fumaric and malic) and hydrochloric acid (Corrigan Thomas and Lawless, 1995), and prickling in soda (Harper and Mc Daniel, 1993) and in oversaturated solutions of carbon dioxide in water (Cometto-Muñiz *et al.*, 1987; Green, 1992). Pungents are usually used to increase food acceptability since they may enhance some tastes while concealing other, less pleasant tastes. It is to be noted that some sulfur compounds in garlic and onion, menthol and menthone in different classes of mint, gingerol in ginger, and other pungents are generally endowed with medicinal properties. Examples of these are the thermic and arteriolar effects (Otero-Losada, 1997), antiseptic properties (Yamada and Azuma, 1977) and anti-ulcer action reported for ginger (Yoshikawa *et al.*, 1994). Hence their sensory attributes are relevant not only to food technology but also for their putative use as therapeutic agents.

This work studies two different aspects of BA, namely sensory qualities and the dynamic aspects of an outstanding attribute, using psychophysical methodology. The following are described and analyzed: (i) quali-semiquantitative sensory profile, pungency concentration range and topography of stimulation; and (ii) kinetic characteristics of BA oral perception.

Materials and methods

Subjects

Eleven panelists (seven male and four female, age 25–45 years) were paid to participate in the experiments. Most of them reported previous experience with sensory evaluation. They were asked not to smoke or eat (water drink allowed) 1–2 h before experiments.

Stimuli

Aqueous solutions of BA (reagent grade, Anedra S.A., Argentina), of 3, 6, 9, 12, 18, 24 and 36 mM, were prepared in deionized water 24 h before the experiments and kept at 5°C. Solutions were poured into 30 ml disposable cups (sample volume 10 ml). Three-digit random numbers identified solutions.

Preliminary session procedures

The sip–spit technique was used to evaluate the solutions at room temperature. Subjects identified the main attribute perceived in standard solutions of sucrose (0.4 M), NaCl (0.2 M), citric acid (0.02 M), quinine (3×10^{-5} M), carbonated soda (dilution of an oversaturated solution of CO_2 in distilled and deionized water to 40% v/v) and capsaicine (0.5–10 ppm, by dilutions in distilled water of a stock solution of vanillyl nonamide 98–100% in 80% ethanol). The differences between taste and non-taste (pungent) sensations were then explained and subjects were asked to differentiate between them. Several daily drinks and foods and their predominant usual attributes were mentioned and named according to sensory impact. The subjects had no difficulty in discriminating between taste and pungent sensations, and at the end of a series of training sessions were able to distinguish among sweet, salty, bitter, sour and pungent (either tactile, thermic or painful, but non-taste sensation).

Experimental session procedures

First experiment: sensory profile and topography of pungency

The aim of these experiments was to develop a permanent quali-semiquantitative record of the sensory components of BA. This list was combined with semiquantitative analysis and the panel was trained in the use of intensity scales.

A list of possible sensations and attributes for BA detection was elaborated (ASTM, 1968) and analyzed in previous training sessions in order to clarify terminology and to identify each attribute. The list was available to the judges during the experimental sessions.

The intensity of each attribute was evaluated using a graphic scale (an unstructured horizontal line, 15 cm long) on which the panelist placed a vertical mark across the line at that point which best reflected the perceived intensity of the corresponding attribute.

The performance of the judges and the effectiveness of the descriptive terms developed by the group were checked by statistical analysis of about four replicated sets of training data.

The instructions given to judges were: (i) to choose one or more words from the list (as many as needed and/or others of their own) which best described the sensation/s induced after tasting each sample and to place the corresponding mark for evaluation on the graphic scale; (ii) to pay attention at the place on the oral cavity (lips; tongue: tip, sides, middle, back; soft or hard palate; cheeks mucosae; epiglottis; upper throat, etc.) where sensations were perceived. Three digit numbers at random in double blind tests identified BA solutions (3–36 mM). Hence, information obtained from each sample was: sensation/s (quality), percentage (quantity) and location. Interaction was not allowed among judges so that judgements were independent.

To counterbalance possible sequential effects, i.e. any influence from a previous sensation, samples were randomly given to the subjects in quadruplicate. Subjects also rinsed out their mouths with a mouthful of distilled water between each solution sampling (inter-trial time: 2–3 min).

Results obtained from individual questionnaires were openly discussed with each subject.

Data analysis. Percentages referring to the relative intensity of each attribute were obtained from the relative and proportional records obtained by means of the graphical scales totaling 100% of the sensory impact. For each concentration, data gathered from subjects were averaged, both the standard deviation (SD), and standard errors (SE) were calculated, and results were expressed as the mean ± SE of 11 subjects.

Second experiment: measurement of pungency intensity as a function of time (time–intensity records)

A computerized data collecting automatic system (record took place each 0.28 s) was used. The subject moved the computer mouse from left to right according to a rate pattern proportional to the intensity increase of the perceived sensation. The subject's movement was converted into a sliding motion over a monitor screen along a 20 cm (500 pixel) unstructured scale. Experimental sessions consisted of three sessions. These were performed with the subjects after previous training on how to use the mouse and couple mouse motion to cursor motion (motor–visual coordination).

Samples identified by key numbers were evaluated randomly, except for the first one, the content of which had an intermediate concentration (12 mM) and was given as a reference. Its maximum intensity matched a point at mid-scale. After rinsing out their mouths with distilled water, the subjects sipped their samples without swallowing them: as soon as the subjects perceived sample pungency they started moving the mouse to the right until peak intensity was reached and then leftwards according to intensity decrease. An audible signal after 5 s indicated the subjects to spit the sample out (Zamora *et al.*, 1998).

Data analysis. Kinetic parameters evaluated were: maximum intensity (I_{MAX}, %); pungency total time (T_{tot}, s); rates of increase (V_1, %/s) and decrease (V_2, %/s) (rates were obtained considering the straight line that best adjusted a majority of points either preceding or subsequent to I_{MAX} respectively); half life: time at which intensity had decayed 50% of I_{MAX} ($T_{½}$, s); area under curve [AUC, % × s, calculated according to the trapezoid rule ($S(I_i + I_j)/2Dx$, where x = time and I = intensity); and time for attaining I_{MAX} (T_{IMAX}, s).

The above-mentioned parameters were submitted to mathematical iterations in order to find the best fit equation as a model which described the behavior of the parameter across BA concentrations. Results were also submitted to an analysis of variance (ANOVA), followed by the least significant difference test (LSD) in order to determine which BA concentrations behaved different kinetically. Prism and SPSS software packages were used for analyses. A randomized blocks experimental design was used according to a mixed effects model wherein three factors were used: subjects (variable factor), replications and concentrations (fixed factors).

Principal components analysis (PCA) and clusters analysis. Kinetic parameters obtained from each average curve per subject and concentration were submitted to a PCA. Only factors with eigenvalues greater than one were considered to be significant. To identify similar observation groups, a hierarchical group analysis was used according to the average method based on the degree of similarity among records (cluster analysis).

Results

Sensory profile

BA was found to be mildly pungent and slightly sour (Figure 1). The latter attribute was perceived inconsistently only by some subjects in some sessions, and was not always replicated. Some subjects (Figure 1) perceived a smooth sweetness only at low concentrations of BA. A unique, characteristic sensation was induced in the subjects' mouths, quite similar, as described in their own words, to the prickling caused by the prickly pear (the adjective given to the name of that fruit is precisely derived from the sensation it

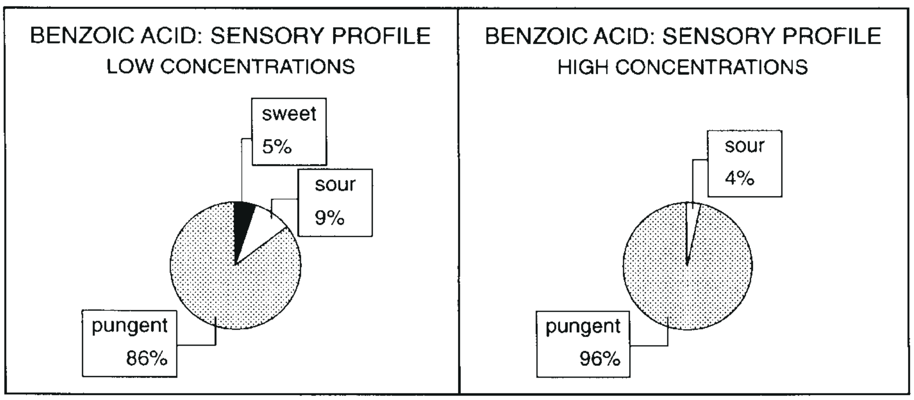

Figure 1 Sensory profile of BA. A list of possible attributes for BA detection was elaborated. The intensity of each attribute was evaluated using a graphic scale and respective percentages for different attributes were obtained from the relative records which totaled 100% of the sensory impact. For more details see Materials and methods.

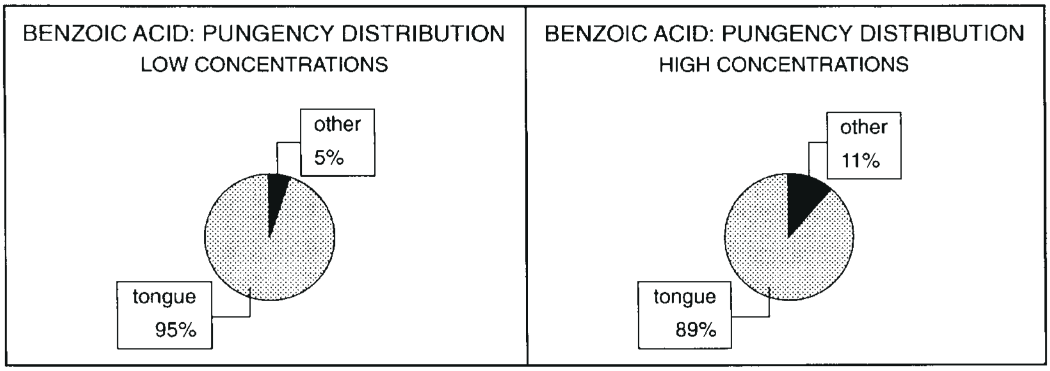

Figure 2 Topography of BA pungency. The places in the oral cavity (lips; tongue: tip, sides, middle, back; soft or hard palate; cheeks mucosae; epiglottis; upper throat; etc.) where BA (mainly pungent) was perceived are shown. For more details see Materials and methods.

induces in the oral cavity). Also some subjects described the sensation as being like that of soda.

The sensory profile could be described as follows: (i) 86% of pungency, 9% of sourness and 5% of sweetness at the lower concentrations (Figure 1, left); (ii) 96% of pungency and 4% of sourness at middle and the higher concentrations (Figure 1, right).

Topography of pungency

BA action was located as follows: (i) at lower concentrations, at the tip of the tongue (95%) and inner cheek mucosa (5%) (Figure 2, left); and (ii) at higher concentrations, at the tip of the tongue, 89%; inner cheek mucosa, 7%; lower lip 3%; gums 1% (Figure 2, right). Some subjects also perceived pungency at a high level of the throat (epiglottis). Consequently, it can be said that pungency was basically located on the tongue (mainly tip, less at sides). At high concentrations, however, pungency was spread all through the mouth.

Intensity/time records

Figure 3 illustrates a typical BA pungency curve obtained at an intermediate BA concentration (18 mM). Pungency started after a short lag period of ~7 s (4–9 s according to concentration). It increased up to a maximum at a rate dependent on concentration and thereafter decreased very slowly (high persistence). The shape of the curve varied according to concentration: an increase in BA concentration speeded up the increase of pungency and delayed its decay. In other words, at high concentrations (36 mM), after a swift increase and a slow decrease of pungency, persistence was reported for ~100 s.

The seven kinetic parameters varied in a concentration-dependent way ($P < 0.001$) and followed different functions (Figures 4–6).

The following equations describe the behavior of parameters as a function of BA concentration: (i) lineal functions: $I_{MAX} = 2.24 \pm 0.14C - 3.06 \pm 2.58$; $R^2 = 0.98$; $T_{IMAX} = 0.19 \pm 0.02C + 6.87 \pm 0.47$; $R^2 = 0.92$; $V_1 = 0.68 \pm 0.03C + 0.10$

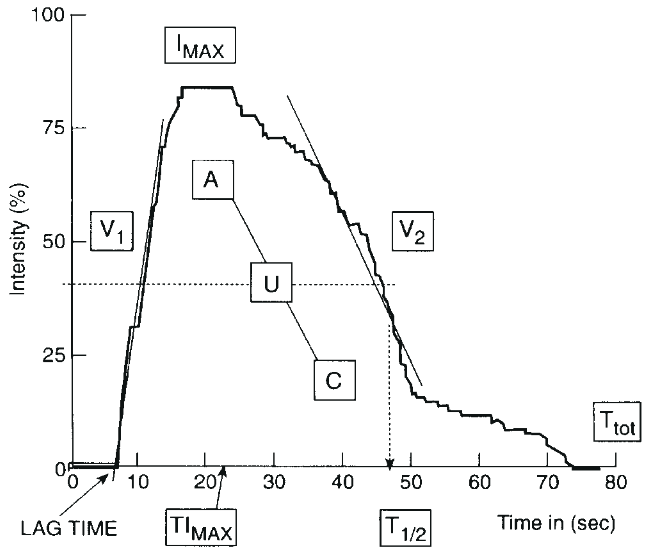

Figure 3 A typical pungency curve obtained at an intermediate concentration (18 mM) of BA. The following kinetic parameters are shown: maximum intensity (I_{MAX}, %); pungency total time (T_{tot}, s); rates of increase (V_1, %/s) and decrease (V_2, %/s); half life: time at which intensity equals 50% of I_{MAX} ($T_{½}$, s); area under curve (AUC, % × s); and time for attaining I_{MAX} (T_{IMAX}, s).

± 0.69; $R^2 = 0.99$ (Figure 4); $AUC = 49.10 \pm 3.17C - 230.78 \pm 59.66$; $R^2 = 0.98$; (ii) power functions: $T_{½} = 6.62 \pm 0.61C^{0.39\pm0.03}$, $R^2 = 0.97$; $V_2 = 1.07 \pm 0.11C^{0.53\pm0.04}$, $R^2 = 0.96$; $T_{tot} = 8.08 \pm 1.01C^{0.43\pm0.04}$; $R^2 = 0.96$ (Figure 5). In the particular case of AUC, best fit was obtained with the equation of a sigmoid (Boltzmann sigmoidal, $AUC = AUC_{min} + (AUC_{max} - AUC_{min})/(1 + \exp((EC_{50} - C)/\text{slope}))$ (Figure 6).

To perform a complete study of kinetic responses, a PCA was carried out using the average values from the group of subjects, which had been calculated for each parameter and each BA concentration. An unidimensional model (Figure 7) reflected the kinetic results: the first component (PC_1) explained 97.52% of the total variation of results; a second

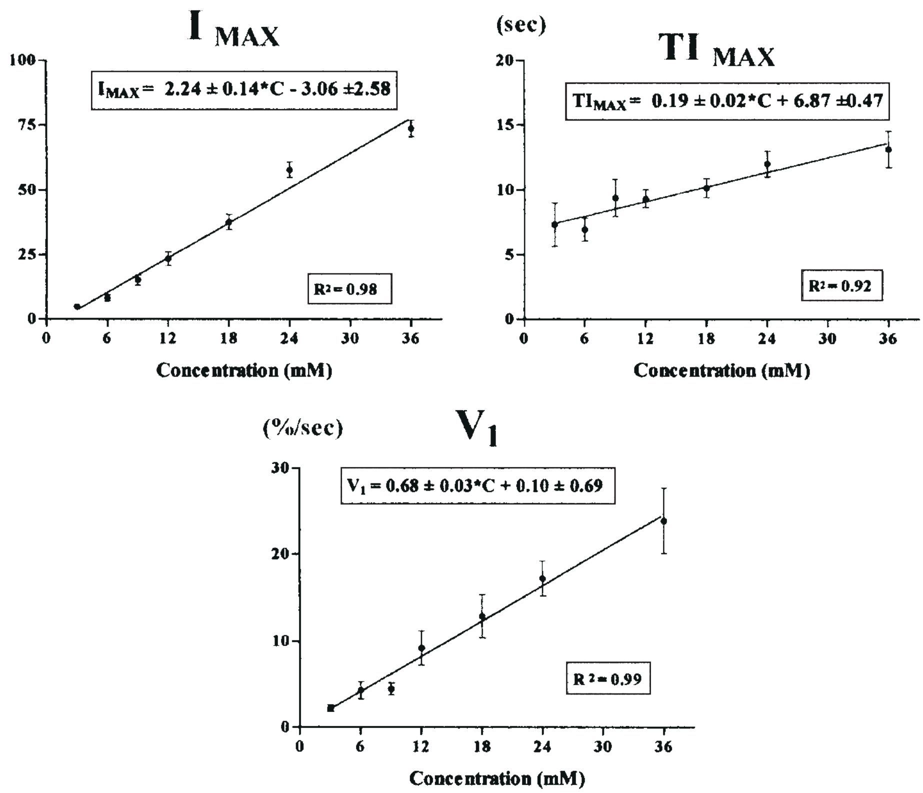

Figure 4 Time–intensity records: kinetic parameters as a function of BA concentration. I_{MAX}, T_{IMAX} and V_1 showed a lineal response to BA concentration increase.

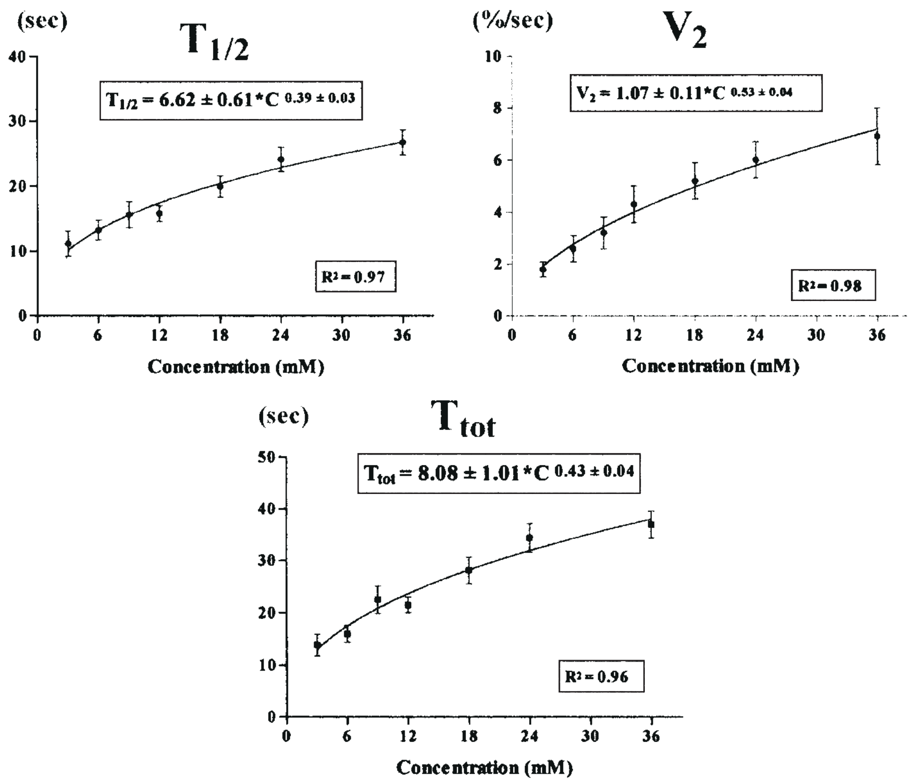

Figure 5 Time–intensity records: kinetic parameters as a function of BA. $T_{½}$, V_2, and T_{tot} described power functions in response to BA concentration increase.

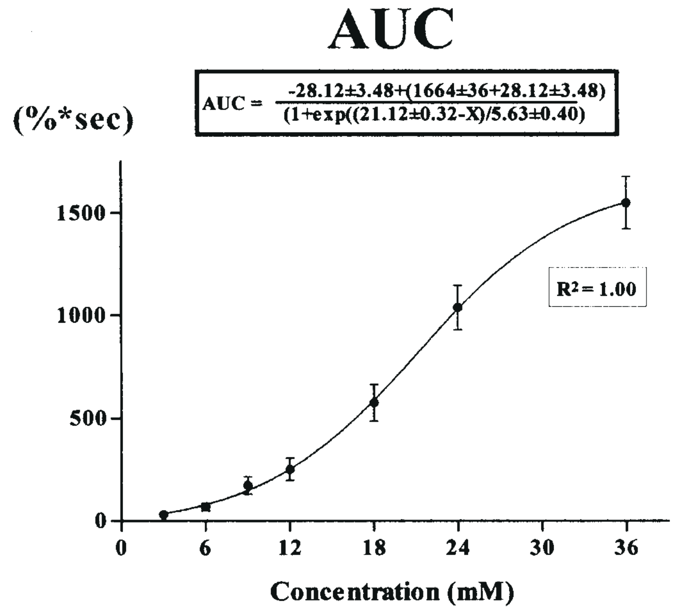

Figure 6 Time–intensity records: kinetic parameters as a function of BA. AUC described a sigmoid in response to BA concentration increase (Boltzmann's sigmoid).

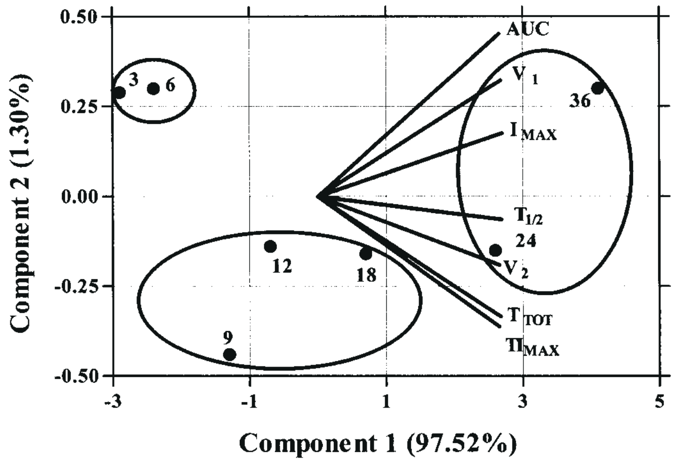

Figure 7 Principal component analysis and cluster analysis of kinetic results.

component (PC_2: 1.30%) was considered as negligible. The seven parameters showed positive values, the vectors of which were projected with a similar magnitude on PC_1, thus evidencing a similar weight in the total variation of results. A high correlation was observed among: (i) T_{IMAX} and T_{tot}; (ii) $T_{½}$ and V_2; and (iii) I_{MAX} and V_1, showing proportionality among parameters.

Stimuli were grouped according to their concentrations along PC_1: 3, 6, 9 and 12 mM to the left, 18, 24 and 36 mM to the right (Figure 7) of the origin of coordinates. Cluster analysis revealed that as a result of such a distribution, stimuli were grouped into three clusters according to BA concentration: (i) low concentrations (3 and 6 mM); (ii) intermediate concentrations (9, 12 and 18 mM); and (iii) high concentrations (24 and 36 mM).

Discussion

Sensory profile

According to the first experiment few subjects found sourness and sweetness in any sessions at 3 and 6 mM of BA. These results were, however, inconsistent and not always replicated. At most concentrations (9, 12, 18, 24 and 36 mM) the main attribute of BA was pungency (prickling) and BA was practically tasteless. In relation with chemical perception, compounds structurally related to BA such as benzoate or benzaldehyde show different attributes. Benzaldehyde features an almond smell and taste (Cometto-Muñiz, 1981), while sodium benzoate has been reported as sweet, salty and bitter (Fox, 1954). Yet this perception may vary not only among individuals and concentrations (Peryam, 1960) but also for the same individual in different circumstances (Hoover, 1956). Saltiness and lack of sourness for benzoate (the derived anion from BA, i.e. BA salt, chemically speaking) may be contrasted with BA's slight sourness. We have observed that benzoate does not induce pungency (unpublished data) and that the carboxylic group of BA is needed to induce it.

No trigeminal reflexes could be observed, such as cutaneous vasodilation with reddening of the face and conjunctiva, tearing, perspiration from the head and neck or increased salivation, which are usually induced by intense pungent substances (Moncrieff, 1951; Lee, 1954; Lawless, 1984). Hence we are dealing with a mild, non-aggressive pungent stimulus.

Lower psychophysical (Stevens) coefficients are generally found for taste stimuli than those observed for pungent stimuli. Thus the ratio between the proportion of perceived intensity increase (psychological magnitude) and the increase in stimulus concentration (physical magnitude) is higher for pungents. Interestingly, in relation to the chemical structure and related psychophysical properties of substances, the taste coefficient(Stevens' β coefficient) for benzaldehyde is unusually high (0.92), practically twice that observed for other stimuli such as vanillin (0.52) or piperonal (0.62) (Cometto-Muñiz, 1981). Thus the β value for benzaldehyde (taste) is quite near to that of BA (pungency), which is >1, thus suggesting a probable structure–activity relationship.

Topography of stimulus

At low concentrations pungency was specifically induced at the tip of the tongue and, to a lesser extent and with lower intensity, in the cheek mucosa. The most sensitive areas to BA were the tip and anterior sides of the tongue. With higher concentrations, pungency was also observed on the lower lip, palate and throat (epiglottis). As regards sensitive areas, the present findings are in agreement with reports for other pungents such as ethanol (Green, 1988), capsaicin (Lawless, 1984; Düner-Engstrom *et al.*, 1986; Lawless and Stevens, 1988) and piperine (Lawless and Stevens, 1988). Anatomical studies have found that trigeminal receptors are of a free nature—they are not organized in particular structures but are located on free nerve endings associated to taste buds at the deeper layers of oral mucosa (Dixon, 1962; Farbman and Hellekant, 1978; Whitehead *et al.*, 1985). A high density of trigeminal receptors is found at the tip and anterior sides of the tongue. The anterior and posterior palate (nasopalatine nerve and posterior palatine nerve respectively), cheek mucosa (buccal nerve) and gingival mucosa are endowed with a low density of trigeminal receptors. The epiglottis receives vagal innervation (Silver and Finger, 1991). Consequently, areas where BA pungency was perceived are relevant to areas innerved by trigeminal branches and the vagus nerve, which carry information related with non-taste sensations induced by chemicals (i.e. pungency) upon activation of the common chemical sense.

Kinetic study of pungency

Pungency was detected no earlier than 4–9 s after presentation of BA, depending on the concentration. After dissolution into membrane phospholipids, hydrophobic stimuli cross through the successive layers of the epithelium until the deepest levels are reached, wherein the trigeminal receptors are located (Silverman and Kruger, 1989; Silver and Finger, 1991). The time interval needed for a stimulus to penetrate across the epithelium before contacting the trigeminal receptors is the limiting step (the largest event) in the chain of events that occur before signal detection (electrophysiological data) (Dixon, 1962; Farbman and Hellekant, 1978; Whitehead *et al.*, 1985). We suggest that the lag time to the onset of pungency involves these events. Thus, the expected reaction time (RT) to pungency (s) should be longer than the RT to taste stimuli (ms) due to the different anatomical location of pungency (deepest mucosa layers) and taste receptors (exposed papillae) (Buratti *et al.*, 1996; Otero-Losada *et al.*, 1997). In a sole study carried out on this topic, it could be observed that in fact the reaction time to pungency averaged 5.9 s (ethanol, 35–85%) in small areas of the tip of the tongue (0.38 cm^2) (Green, 1988). Thus the present results of BA lag, as compared with ethanol lag, are in keeping with what would be expected for a pungent substance. Lag time and spatial variation of pungency observed with regard to BA are both in agreement with the anatomical location of trigeminal terminals, respectively: (i) deep in mucosa and thus far away from a direct contact with saliva; and (ii) ending within epithelial tissue or neighboring tissues only in the anterior part of the tongue and coexisting with the location of taste buds (Dixon, 1962; Farbman and Hellekant, 1978; Whitehead *et al.*, 1985).

BA solubility in water is low (g/l): 2.9 at 20ºC; 3.4 at 25ºC (*Chem. Abstr.*, 1993, p. 170); and 6.6 at 20ºC (Louekari *et al.*, 1990). This fact may well determine the slow BA removal by saliva flow. The observed high BA persistence may be related to its low dissolution and remotion by saliva. It has been demonstrated that pungency duration depends on both the lipophilic and hydrophobic properties of chemestesic compounds that are scarcely soluble in the aqueous phase (Green and Lawless, 1991). Recognition and binding of pungent stimuli to specific pungency receptors proceeds by means of adsorption–desorption mechanisms where weak forces (van der Waals, hydrogen bonds) develop at relatively low velocities, compared with taste where strong forces participate in high-speed molecular interactions (Cliff and Heymann, 1994). These differences are likely related to the different reaction times (s versus ms) and kinetics of pungency and taste perceptions respectively. The present kinetic results on BA prickling are the psychophysical evidence of underlying neurochemical processes. Therefore these results may be useful in studying the mechanisms underlying prickling at the molecular level, a matter beyond the scope of this study.

It should be mentioned that, after sampling BA solutions, a majority of subjects informed about a sensation of tongue and/or whole mouth dryness. In relation to this, astringency would embody a particular case of non-gustatory stimulation, a mixed case apparently including both gustatory and tactile sensations (Iiyama *et al.*, 1994).

Up to now, a sole study has reported BA prickling sensation. In that study, soda was used as the reference substance (Peleg and Noble, 1995) for BA induced a characteristic sensation quite similar to the sensation caused by the bubbling of soft drinks, or pop. As far as our study is concerned, BA-induced pungency is a mild pungency in between the effervescence of soda and a tingling, prickling sensation. The precise 'prickling' experienced in oral mucosae upon ingestion of unripe figs has given its name to the 'prickly' pear. The chemical structure of BA, an organic acid (pK_a = 4.2, an approximate proton concentration in an equilibrium condition, namely 0.1 mM) may be responsible for such a sensation, potentially sour to the sense of taste, with slight irritating effects.

Linearity found for I_{MAX} , T_{IMAX}, V_1 and total impact (AUC) with respect to BA concentration may facilitate the estimations made by inexperienced panelists, since lineal reasoning arises intuitively and spontaneously, more easily than when more complex functions describe the increase of a parameter (this is particularly important for perceived intensity) as a function of concentration.

Extrapolation of maximum intensity in the absence of BA (intersection of the graph at zero concentration) yielded negative values for both maximal intensity and total impact, thus reflecting the characteristics of the smooth pungency of BA which is not particularly intense, and scarcely detectable at low concentrations. The ordinate, at the origin of T_{IMAX}, represents an extrapolation, with no strict physical meaning, that is likely to forecast a detection time (i.e. proportional to the reaction time) to be expected with regard to infinitesimal dilutions of stimulus (zero concentration). Its value of 8.7 ± 0.47 is in keeping with the lag time (≥7 s) that was observed for the lowest BA concentration under study (i.e. 3 mM). In this sense, not all subjects perceived pungency when sampling the 3 mM BA solution. On the contrary, detection thresholds were higher in the great majority of cases.

Among the lineal parameters, maximum intensity (I_{MAX}) and total impact (AUC) evidenced the highest slopes: 2.24 ± 0.14 and 49.10 ± 3.17 respectively; in other words, both parameters were the most sensitive to increases in concentration. Slope values of T_{IMAX} and V_1 (0.19 ± 0.02 and 0.68 ± 0.03 respectively) suggest these parameters are less sensitive to the varying concentrations of BA.

T_{IMAX} values for BA ranged from 8 to 13 s, higher than T_{IMAX} values observed for the pungency of CO_2 in water (~5 s; Cometto-Muñiz and Noriega, 1985). Consequently, BA evidences a slower development of prickling (compared with the development of soda prickling). Also BA prickling is smoother, yet more persistent. Persistence is much more closely related to the chemical structure of a stimulus than sensation is *per se*. Thus, CO_2 and BA induce a similar sensation (quality) but differ in sensation persistence (temporal integration of the stimulus). Different solubility in the aqueous phase as shown above may play an important role in such difference.

The findings of the present study suggest that BA could be considered as a prototypical mild pungent stimulus. BA induces prickling and may be perceived as slightly sour by some individuals. Pungency is mainly located at the tip of the tongue, starts after a lag time (s) and persists for an average 1 min. Different kinetic parameters respond differently to increases in BA concentration. Pungency intensity depends on concentration, and is directly proportional to persistence and inversely proportional to the lag time for detection. The prototypical prickling sensation of BA is observed around 12 and 18 mM.

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