Masking of bitterness in dairy protein hydrolysates: Comparison of an electronic tongue and a trained sensory panel as means of directing the masking strategy

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ARTICLE INFO

Article history:
Received 6 January 2014
Received in revised form
30 January 2015
Accepted 9 March 2015
Available online 17 March 2015

Keywords:
Bitterness masking
Electronic tongue
Sensory analysis
Casein hydrolysates

ABSTRACT

Sodium caseinate hydrolysates (NaCaHs) are known to be a rich source of bioactive peptides but are also known to have a bitter taste which can make them difficult to incorporate into foods. The objective of the study was to identify whether a commercially available electronic tongue (e-tongue) could be used to identify sweeteners and flavours which reduce bitterness perception in model beverages containing NaCaHs. The two NaCaHs analysed in this study were found to be strongly bitter, the sensory panel found sucralose to be the most effective sweetener, reducing bitterness by a minimum of 39% at the lowest concentration assessed, 0.017% w/w, and the flavour vanilla was the most effective flavouring for further reducing bitterness in a sweetened model beverage. The e-tongue was found to be an effective tool for measuring the reduction of bitterness in of NaCaHs due to different sweeteners but not for evaluating the effect of flavourings in the model beverage. The results also showed good agreement between the e-tongue and sensory panel in the measurement of bitterness and sweetness in sweetened NaCaHs suggesting that the e-tongue maybe a useful device for the selection of sweeteners as bitterness masking agents in NaCaHs.

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1. Introduction

Sodium caseinate protein hydrolysates (NaCaH) are known to be a rich source of bioactive peptides (Korhonen, 2009). These peptides can have a positive biological impact on the body's cardiovascular, immune and endocrine systems (Mills, Ross, Hill, Fitzgerald, & Stanton, 2011; Phelan, Aherne, Fitzgerald, & O'Brien, 2009; Phlantor & Fuquay, 2011). However, the process of hydrolysis can negatively affect the taste profile of NaCaHs due to the production of bitter peptides (Favaro-Trindade, Santana, Monterrey-Quintero, Trindade, & Netto, 2010). The bitterness elicited by peptides has been researched by numerous authors who found that bitterness is most commonly associated with; small to medium hydrophobic peptides (Ney, 1979), peptides with high proline content (Ishibashi, Kubo, et al., 1988) and aspects of the peptides specific sequence i.e. presence of hydrophobic amino acids at the N or C termini (Ishibashi, Kougé, Shinoda, Kanehisa, & Okai, 1988).

While much has been communicated on the potential of bioactives found in NaCaHs the difficulty of incorporating them into foods (Neklyudov, Ivankin, & Berdutina, 2000) without negatively impacting on the taste is somewhat under investigated. The various strategies to improve the palatability of bitter ingredients that have been addressed previously include processes such as encapsulation (Mendanha et al., 2009) or enzymatic de-bittering (Minagawa, Kaminogawa, Tsukasaki, & Yamauchi, 1989) which, in the latter case could result in the loss of functionality of bioactive peptides. A common and economical method of suppressing bitterness is to use so called congruent flavourings (Ley, 2008), i.e. mixing the bitter agent with a flavouring that has some inherent level of bitterness associated with it such as chocolate, or grapefruit to reduce the perceived level of bitterness making the food more palatable.

Another approach which is common practice is the addition of another strong tastant, such as MSG in savoury or sucrose in sweet foods to “distract” from the bitterness and improve the overall taste profile (Ley, 2008). The utilisation of sugars and sweeteners to suppress bitterness is known as ‘mixture suppression’, as both
tastes are suppressed in their perceived intensity when presented together due to a form of cognitive inhibition (Keast & Breslin, 2002). However, the addition of high levels of sucrose to mask bitterness may be counterproductive to the overall health benefit of a functional food (in particular given the links between excessive intake of sugar and obesity) in these cases the use of low calorie or non-caloric sweeteners maybe useful. The use of artificial sweeteners has increased in the last decade as the food industry tries to match the demand of the consumer for healthier, low sugar foods while maintaining sweetness (Zyglker, Wasik, & Namieśnik, 2009). In the suppression of bitterness in NaCaHs it is important to investigate sweeteners that are suitable for the food industry. These sweeteners should be easily incorporated in foods, widely used to improve food taste profile and well accepted by consumers. Sensory analysis with a trained panel is the best method of quantifying bitterness in a food product; however it can be an inconvenient method of analysis as it is very time consuming due to the time taken to recruit and train a panel to a satisfactory level to generate accurate results (Linforth, Baek, & Taylor, 1999). As a result, there has been increased interest in replacing or reducing the reliance on sensory panels through the use of instrumental analysis. A multi-sensor technology, commonly referred to as the electronic tongue (e-tongue) has been developed to mimic the human taste response and has been utilized in bitterness masking studies in the pharmaceutical sector as an aid to the development of palatable formulations (Janczyk et al., 2010; Lorenz, Reo, Hendl, Worthington, & Petrossian, 2009; Thi, Morel, Ayouni, & Flamant, 2012; Woertz, Tissen, Kleinebudde, & Breitkreutz, 2011). However, the e-tongue has never been used before to measure the efficacy of a bitter masking agent in a complex food system such as NaCaH. Therefore, it was important to verify that the e-tongue is suitable tool to measure the efficacy of bitter masking agents on the taste profile of NaCaH by comparing its analysis with that of a trained human sensory panel.

The aim of this study was to evaluate the suitability of some sweeteners and flavours in reducing the perceived bitterness in a model beverage formulation containing NaCaHs. The degree of bitterness masking was evaluated both by sensory analysis and by an e-tongue and the response of both methods to the various masking agents was compared.

2. Materials and methods

2.1. Chemicals and compositional analysis

Two spray dried sodium caseinate, labelled NaCaH1 and NaCaH2, were produced to food grade specifications by a research partner (Morepark technology Ltd. Cork, Ireland). Composition of NaCaHs was determined by the following methods: protein by means of Kjeldahl analysis (ISO/DIS 9686, 2001), fat by the Rose Gottlieb method (Richardson, 1985), ash was determined using a muffle furnace at 550 °C using the ISO 5545:2008 (IDF 90: 2008) method, the degree of hydrolysis (DH) using the o-phthalaldehyde-hyde (OPA) method (Nielsen, Petersen, & Dambmann, 2001) and pH was also recorded (pH unicum 9450, Cambrige, England). US Pharmacopeia grade caffeine and sucrose were purchased from Sigma Aldrich (Sleeze, Germany). Sucralose and Stevia were donated by J.K Sucralose Inc. (Breda, The Netherlands) and Vitiva (Markovet, Slovenia) respectively. Chocolate, vanilla, grapefruit and grapefruit/orange flavours were donated by Silesia (Silesia Gerhard Hanke GmbH & Co.KG, Neuss, Germany). Solutions of HCl, NaCl and monosodium glutamate (MSG) used in the start-up procedures for the e-tongue; were supplied by the manufacturer (Alpha M.O.S., Toulouse, France).

2.2. Sample preparation

An initial screening of different sweeteners as bitterness masking agents was performed using the e-tongue, on samples prepared as follows: NaCasH1 or NaCasH2 (10 g/100 g) was dissolved in ultrapure water(Milipore, Synergy UV system, Merck) or in ultrapure water containing sucrose (10 g/100 g), sucralose (0.017 g/100 g; Binns, 2003), stevia (0.13 g/100 g; Lekrisompong et al., 2012) or acesulfame K (0.05 g/100 g; Bellisle & Drenowski, 2007); the concentrations of sweeteners were chosen to give equivalent levels of sweetness. Based on the results of this study, which showed acesulfame K and sucralose to be the most effective masking agents, a further set of samples were prepared for analysis by the sensory panel by dispersing the NaCasH1 and NaCasH2 (10 g/100 g) in bottled water or in bottled water to which different concentrations of the two most effective masking agents had been added — i.e. 0.025, 0.05, 0.075, 0.1 g/100 g acesulfame K or 0.017, 0.027, 0.037, 0.047 g/100 g sucralose. Finally, a set of ‘model beverages’ were prepared comprising of 10 g/100 g NaCasH1 or NaCaH2, sweetened with 0.017 g/100 g sucralose and containing 0.06 g/100 g of one of the following flavourings; chocolate, vanilla, grapefruit or a grapefruit/orange blend; this concentration was the dosage suggested by the manufacturer.

2.3. Sensory analysis

2.3.1. Training of the sensory panel and sample presentation

A trained sensory panel of 2 males, 7 female of an age range of 23–45 with over 825 combined hours experience in the analysis of dairy protein hydrolysates was employed in this study. Each panel member had a threshold level for caffeine of less than 1.39 mmol/l and had been trained to use the 15 point spectrum intensity scale, where 5 corresponds to weak and 15 very strong (Meilgaard, Civille, & Carr, 2007). Panelists were required not to smoke, eat, drink caffeinated beverages or wear strong perfumes prior to analysis sessions. The evaluations took place in individual booths and at each evaluation, panelists were supplied with bitter and sweet reference samples to aid scaling (see Table 1). All samples were presented monadically to panelists at room temperature in 20 ml sample cups which were labelled with random three digit codes and panelists were provided with water and plain crackers for palate cleansing between samples (Rétiveau, Chambers, & Esteve, 2005).

For this investigation the formulations were assessed by the trained sensory panel in a complete block design using William Latin square plot to decide presentation order. The test employed for assessing taste intensity was the multi-sample comparison test. To ensure that they were not overloaded, no more than 5 samples were presented to panelists at any one time. Thus, to evaluate the effect of different concentrations of sucralose on NaCasH1, panelists were presented with solutions of NaCasH1 and solutions of NaCasH1 containing the 4 levels of added sucralose and asked to evaluate the samples for both sweetness and bitterness using the 15-point spectrum intensity scale. The procedure was repeated to

Table 1

<table>
<thead>
<tr>
<th>Taste</th>
<th>Compound</th>
<th>Concentration (mmol/l)</th>
<th>Intensity</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bitter</td>
<td>Caffeine</td>
<td>4.1</td>
<td>5</td>
</tr>
<tr>
<td></td>
<td>7.7</td>
<td>10</td>
<td></td>
</tr>
<tr>
<td></td>
<td>10.3</td>
<td>15</td>
<td></td>
</tr>
<tr>
<td>Sweet</td>
<td>Sucrose</td>
<td>146</td>
<td>5</td>
</tr>
<tr>
<td></td>
<td>292</td>
<td>10</td>
<td></td>
</tr>
<tr>
<td></td>
<td>438</td>
<td>15</td>
<td></td>
</tr>
</tbody>
</table>
evaluate the effect of acesulfameK on NaCaH1 and the same procedure applied to evaluate the effect of both sweeteners on NaCaH2.

4. All formulations were analysed in triplicate.

2.4. Electronic tongue

The e-tongue employed in this study was the α-ASTREE II Liquid Taste Analyser (Alpha M.O.S., Toulouse, France). The e-tongue is comprised of a 48 position auto-sampler (25 ml glass sample vials), a 7 sensor array mounted around an Ag/AgCl reference electrode which is connected to an electronic unit which process the voltage response of the sensors.

On the recommendation of the supplier the sensor set used in this study was one developed and marketed for general food applications and exhibited sensitivity to all five basic tastes (Alpha M.O.S. sensor model codes ZZ, JE, BB, CA, GA, HA, JB). This set was used in preference to a set developed solely for the assessment of bitterness in pharmaceutics, also available from the suppliers, for three reasons: 1) the food set had already been successfully used by the authors for evaluation of bitterness in hydrolysates (Newman et al. 2014a, b), 2) it was expected to exhibit better robustness in the face of complex food samples and 3) the sweetness as well as the bitterness of the samples was being assessed in this present study. Prior to analysis the e-tongues sensors were prepared using the start-up procedure as recommended by the manufacturer’s guidelines (Anon, 2010). This involved conditioning and calibration with 0.01 mol/l HCL and a diagnost test of three samples, HCL, NaCl and monosodium glutamate (MSG), all at a concentration of 0.01 mol/l, to ensure that sensors were responding in the correct mV range. Analyses comprised of immersion of the sensor array in the sample for a period of 120 s 10 times during which the mV output was monitored. The first 2 readings were discarded and the average mV reading for the last 20 s of the remaining 8 immersions were used for statistical analysis (Anon, 2010). The outputs of 6 sensors were used for all analyses.

2.5. Statistical analysis

Sensory data were pre-processed using a one way ANOVAs to ensure panelist consistency in scoring between sessions with replicates as the factor and two way ANOVAs with panelists and sample treatments as factors. As replicates and panelists were not significant as factors (p > 0.05) the panel data were taken to have been validated. Fisher’s least significant difference test was used to identify significant difference between samples (p < 0.05). The ANOVAs were processed using SAS Version 9.3; SAS Institute Inc, Cary, USA. The effectiveness of the sweeteners and flavours to reduce the perceived intensity of bitterness was calculated as a percentage using the following formula (Keast, 2008):

\[
\text{% bitterness reduction} = \left( 1 - \frac{B - BT}{B} \right) \times 100
\]

where B is the mean bitterness score of NaCaH and BT is the mean bitterness score of NaCaH with added sweetener or flavouring.

The data generated by the e-tongue were autoscaled and processed by multivariate statistical analysis using the statistical software of the e-tongue, AlphaSoft version 12.4 (Alpha M.O.S., Toulouse, France). Data from individual sensors were removed prior to analysis if the relative standard deviation of the sensor was greater than 6% (Zheng & Keeney, 2006). To assess discrimination between samples and masking efficacy of a range of compounds principal component analyses (PCA) were conducted using the covariance matrix. An indicator of the quality of discrimination between samples on the PCA is the discrimination index (DI) which can range from a negative value to 100 (Lorenz et al., 2009). To measure the effectiveness of a masking agent, the Euclidian distances between clusters of the data points for the different samples on the PCA graphs was also recorded. A greater distance between treated and untreated sample groups indicates that the samples are more different in taste profile and that the bitterness has therefore been masked (Campbell et al., 2012; Ito et al., 2013). Partial least square regression (PLS) was conducted to correlate bitterness intensity values from the human sensory panel with those from the e-tongue with the sensory and e-tongue data as x- and y-axis variables, respectively.

3. Results and discussion

3.1. NaCaH composition

The two NaCaHs were produced from the same substrate, sodium caseinate and as a result have similar compositions; both NaCaH samples are high protein (>85 g/100 g), low in fat (<2.5 g/100 g), have moderate ash levels and are neutral in pH (Table 2). However, the samples were produced under different conditions and to differing levels of DH, NaCaH2 is substantially more hydrolysed than NaCaH1.

3.2. Bitterness reduction in NaCaHs by sweeteners

3.2.1. Measuring masking efficacy of different sweeteners on the bitterness of NaCaHs using the e-tongue

The two NaCaHs were analysed separately with and without the addition of four different sweeteners by the e-tongue and two PCAs were constructed using the resulting data which are shown in Fig. 1. The PCA maps were constructed using the first 3 principal components, PC1 contributed 61.68% of the variance in Fig. 1A (sum of the 3 principle components equalling 98.9%) and 88.21% in Fig. 1B (sum of the 3 principle components equalling 99%). In both analyses the e-tongue could clearly discriminate between the sweetened and the unsweetened NaCaHs. However, in both PCAs, there is overlap between the NaCaHs sweetened with the different agents, suggesting that the e-tongue had some difficulty discriminating between the NaCaHs sweetened with different sweeteners, resulting in a negative DI of −21 and −70 values indicative of poor discrimination, Fig. 1 A and B respectively.

Despite the poor discrimination shown on the PCA’s an attempt was made to evaluate the efficacy of each masking agent by comparing the Euclidean distances between the clusters of data points of the unsweetened NaCaHs and the NaCaHs with each of the different added sweeteners which are shown in Table 3. The greater the distances between samples, the greater the masking effect as measured by the e-tongue (Thi et al., 2012). In both NaCaHs, stevia and sucrose were the least successful in masking the taste as measured by the e-tongue, for example NaCaH 2 sweetened with sucrose has a distance of 121.38 and stevia a distance of 101.02 from the unsweetened NaCaH2 relative to the sucralose and acesulfame K with distances of 139.23 and 128.39 respectively. The results from the e-tongue are consistent with those of Lekrisompong, Gerard, Lopetcharat, and Drake (2012) who had

Table 2

<table>
<thead>
<tr>
<th>Composition, degree of hydrolysis (DH) and pH of NaCaHs.</th>
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<tbody>
<tr>
<td>Protein %</td>
</tr>
<tr>
<td>NaCaH1</td>
</tr>
<tr>
<td>NaCaH2</td>
</tr>
</tbody>
</table>
previously used a trained sensory panel to evaluate masking of bitterness in whey protein hydrolysates by the sweeteners sucralose, sucrose and stevia. In that study the sweetener sucralose was also found to be a more effective bitter masking agent than either sucrose or stevia. The fact that the sweetened samples all plot in the same area of the PCA suggest that the suppression of bitterness by the different sweeteners occurs through a similar mechanism of mixture suppression.

The results suggest that the most effective sweeteners for masking the bitterness of the two NaCaHs are sucralose and acesulfame K as determined by the e-tongue. Sucralose and acesulfame K were then selected for further analysis with the e-tongue and sensory panel to identify which of those sweeteners, and at what concentration, was the most effective for reducing the bitter taste of NaCaHs.

### 3.2.2. Scaling bitterness and sweetness intensities of NaCaHs containing either sucralose or acesulfame K using e-tongue and sensory panel

The bitterness and sweetness intensity scores of the NaCaHs and NaCaHs containing the various levels of the sweeteners sucralose and acesulfame K are shown in Table 4. The panel found the unsweetened samples to be intensely bitter both having score of ≈ 11 and to have low levels of sweetness (<1). The addition of even the lowest concentration of either sweetener resulted in a significant increase in sweetness and decrease in bitterness of both NaCaH samples (p < 0.05). Sucralose had the biggest effect on the sweetness of both NaCaHs, increasing the intensities to >10.5. Acesulfame K was less effective at increasing the sweetness intensity in NaCaHs than sucralose and was also a less effective sweetener in NaCaH2 than in NaCaH1 as there was a significant difference between the sweetness scores at the highest concentration of addition (p < 0.001). The results from the sensory panel also indicate that sucralose was the most effective sweetener at reducing bitterness in NaCaHs as the bitterness reduction at the lowest concentration of sucralose (0.017 g/100 g) is greater than that observed at any acesulfame K concentrations in both NaCaHs.

The response of the e-tongue to the NaCaHs and the NaCaHs containing the increasing concentration of acesulfame K and sucralose was correlated with the panels bitterness intensity scores using PLS regression to compare how well the e-tongue could assess bitterness intensity in such complex samples. The correlation between predicted bitterness scores and actual bitterness scores of the sensory panel in the sweetened NaCaHs were generally strong with correlation coefficients (R^2) of 0.78, 0.94, 0.96 and 0.97 for NaCaH1 containing acesulfame K, NaCaH1 containing sucralose, NaCaH2 containing sucralose and NaCaH2 containing acesulfame K, respectively. The weakest correlation observed was NaCaH1 containing acesulfame K (R^2 = 0.78), while this correlation is not as strong as the others observed in the study it was still very close to the R^2 value of 0.8, recommended by Rachid, Simons, Rawas-Qalaji, and Simons (2010) as being indicative of a good correlation between sensory and e-tongue data.

The predicted bitterness intensity scores from the e-tongue, generated by the PLS regression, were used to calculate the percentage reduction in bitterness (BRT), calculated using Eq. (1), due to the sweeteners at their varying level of concentrations of addition to the NaCaHs (Table 4). The BRT are lower for all sweetened NaCaH samples in comparison to the percentage reductions in the scores of sensory panels (BRP). However, the results from the e-tongue are consistent with those of the sensory panel in that there was an increase bitterness reduction with the increasing concentrations of sweetener and that sucralose was more effective than acesulfame K. The results show that the e-tongue and sensory panel correlate reasonably well in the assessment of bitterness intensity in NaCaHs and in assessing the bitterness reduction efficacy of sweeteners. It suggests that the e-tongue could be used in the future as a method of rapid bench testing of bitterness masking agents in NaCaHs.

### 3.2.3. Selection of a sweetener for a model beverage

Acesulfame K is 200 hundred times as sweet as sucrose and has been reported to have a fast sweetness onset and when used in high concentrations can elicit an unpleasant bitter after-taste (O’Brien, 2001). One possible reason why it was less successful at reducing the bitterness in NaCaHs than sucralose is that upon analysis, the

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**Table 3**

<table>
<thead>
<tr>
<th>Sweetener (w/w)</th>
<th>NaCaH 1</th>
<th>NaCaH 2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Acesulfame K 0.05%</td>
<td>183.9</td>
<td>128.39</td>
</tr>
<tr>
<td>Sucralose 0.017%</td>
<td>170.52</td>
<td>139.23</td>
</tr>
<tr>
<td>Sucrose 10%</td>
<td>109.65</td>
<td>121.38</td>
</tr>
<tr>
<td>Stevia 0.13%</td>
<td>124.37</td>
<td>101.02</td>
</tr>
</tbody>
</table>

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Fig. 1. Three dimensional principal component analysis of the e-tongues sensor response to A) NaCaH1 (■) and B) NaCaH2 (■) and NaCaHs sweetened with Stevia 0.13% (▲), Sucralose 0.017% (▲), Sucrose 10% (▲) and Acesulfame K 0.05% (▲).
panellists tasted an initial sweet taste which was then succeeded by a bitter after-taste.

Sucralose was the most effective sweetener to improve the palatability of NaCaHs and has been previously reported to be an effective bitterness masking agent in whey protein hydrolysates (Leksrisompong et al., 2012). It is 600 times sweeter than sucrose and as a result is effective at extremely low concentrations with no associated off tastes and as a result it is already widely used to improve the taste profile of a wide range of foods from baked beans to vanilla milk (O'Donnell and Kearsley, 2012). The wide spread applications of sucralose is due not only to its palatability but also to its stability over time and at extreme temperatures making it an ideal sweetener in foods that undergo thermal heat treatment for long shelf life products (Blins, 2003). As sucralose was the most effective sweetener, it was selected for further analysis in combination with various flavourings as a ‘model beverage’ and used at the concentration of 0.017 g/100 g.

3.3. The effect on bitterness intensity of NaCaHs of the addition of flavouring agents in a ‘model beverage’

3.3.1. Assessment of ‘model beverages’ by the trained sensory panel

The sensory panel was required to score the bitterness intensity in a series of model beverages which comprised of NaCaH 1 or NaCaH2, sweetened with 0.017 g/100 g of sucralose and containing 0.06 g/100 g of one of the following added flavourings; chocolate, vanilla, grapefruit or grapefruit/orange. Fig. 2 shows the bitterness scores of the NaCaHs, the NaCaHs with (0.017 g/100 g) sucralose and each model beverage as assessed by the sensory panel. As discussed in Section 3.2, the main effect observed was that the addition of sucralose caused the bitterness of the NaCaH1 to be reduced from 10.96 to 7.11 and NaCaH2 from 11.23 to 6.65, representing a bitterness reduction of <39%. The only flavouring to result in a significant (p < 0.05) further decrease in the bitterness score of both NaCaHs was vanilla which reduces the bitterness intensity in NaCaH1 to 5.5 and NaCaH2 to 4.77, representing a further bitterness reduction from the sweetened NaCaH of bitterness reduction of ~18% (Fig. 2). The flavourings grapefruit and the grapefruit/orange combination had no additional masking effect in the model beverages containing either NaCaHs.

As with the sweeteners, the flavourings appear to have had a different level of efficacy for the reduction of bitterness in each of the two NaCaHs. The model beverage containing NaCaH1 and chocolate flavouring was actually slightly more bitter than the NaCaH1 containing sucralose alone (p = 0.0193). In the model beverage containing NaCaH2 however, the flavouring chocolate had the opposite effect that it significantly reduced the bitterness (p = 0.0038).

3.3.2. Assessment of ‘model beverages’ by the electronic tongue

The model beverages formulated with either NaCaH1 or NaCaH2 were analysed by the e-tongue and the results were expressed by PCAs (Fig. 3). The PCA’s demonstrate that the e-tongue could not distinguish between the model beverages containing the different flavourings; the poor discrimination resulted in negative DI values of −135 and −199 for the analyses.

Fig. 3A shows the PCA analysis of NaCaH1, NaCaH1 with sucra-lose and the model beverages containing NaCaH1, PC1 accounts for 90% of the total variance. It can be seen on the PCA that the samples are separated across PC2 and PC3 which accounts for <10% of the total variance, suggesting that the difference between these samples, as measured by the e-tongue is not substantial. The only real discrimination on this PCA was between NaCaH1 and all the other samples containing sucralose, indicating that in this analysis of NaCaH1, only the sucralose appears to strongly influence the response of the e-tongue.

In the analysis of NaCaH2 based beverages, shown in Fig. 3B, the distribution of variance on the PCA is similar to that in Fig. 3A as PC1 accounts for >90%. The separation of samples occurs across PC2 and PC3, which accounts for <8% of the total variance. It appears from this PCA that the e-tongue can discriminate between samples with or without flavouring i.e. NaCaH2 and NaCaH2 sweetened with sucralose are both separated from the flavoured model bev-erages. As was the case for NaCaH1, there was little discrimination between model beverages containing different flavouring agents as analysed by e-tongue.
While the e-tongue did not discriminate well between the NaCahs containing different sweeteners (Fig. 1A and B), the inter-group distances recorded from those PCAs (Table 3) were considerably larger than those recorded between the model beverages containing different flavourings. These differences in the level of discrimination between Figs. 1 and 3 may suggest that the e-tongue is more effective in the measurement of the masking effect from sweeteners rather than between different flavourings.

The poor discrimination between the model beverages containing NaCahs demonstrates the limitations of the e-tongue in assessing possible bitterness masking ingredients. The composition of the NaCahs may have contributed to the poor discrimination between the samples; they contain a moderate amount of salt which may have interfered with the potentiometric sensors of the e-tongue and contain fat, albeit at a low level, (Table 2) which may have coated the sensors causing interference for subsequent analysis of samples. However, it should be noted that the scores from the sensory panel also indicated that there was little difference in the bitterness masking efficacy of flavourings, therefore (if the e-tongue is truly measuring masking efficacy) the results of the e-tongue reflect very well those of the sensory panel.

While the e-tongue has been reported to be useful in measuring the masking effect of different flavours in pharmaceutical preparations (Campbell et al., 2012) in those studies the flavourings were added to mask the effect of a single pure compound known to be responsible for a specific unpleasant taste. In contrast, the NaCahs in the present study contain a large diversity of peptide species of differing composition, size and hydrophobicity and have complex flavour profiles. It may be that such matrices may just be too complex in nature for a device such as the e-tongue to pick up any effects of flavourings. On the other hand, a recent study by Laureati, Buratti, Bassoli, Borgono, and Pagliarini (2010) combined sensory data with those from e-nose and e-tongue devices to successfully discriminate between extracts prepared from different varieties of an oriental herb Perilla frutescens. Perhaps a similar approach of combining the sensory and instrumental data — from both an e-nose and e-tongue — in the analysis might have allowed better discrimination between the different flavoured model beverages in the present study. However the primary aim of the present study was to compare the effectiveness of the e-tongue with that of the panel rather than to combine them in the analysis.

4. Conclusion

Sucralose was found to be the most effective sweetener in the study for reducing the bitterness of bioactive containing NaCahs. The bitterness masking effect was found to be further increased by the addition of vanilla flavouring in a model beverage. The e-tongue was shown to be a useful tool for identifying suitable sweeteners to reduce bitterness in sweetened and unsweetened NaCah and was also shown to correlate well with the sensory panel in rating of bitterness and sweetness intensities in NaCahs. However, the e-tongue proved to be of limited use for further formulation development specifically for evaluating any additional masking of bitterness using flavourings. The e-tongue is an objective analysis tool and so can offer no hedonic value; therefore, if the effectiveness of a 'masking agent' arises from a general improvement of the overall taste profile then the e-tongue is unlikely to be able to detect this — for this assessment by a sensory panel is necessary. The findings of this study indicate that while the e-tongue shows potential as a useful tool for the screening of bitter masking ingredients for functional foods it cannot fully replace sensory analysis for the formulation of palatable ready to use products.

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