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# Exploring hotness and pungent odour thresholds among three groups of Thai chilli users

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**Summary** Sensory thresholds of hotness and pungent odour were determined from 120 chilli users. Three groups of Light (L), Moderate (M), and Heavy (H) chilli users were categorised based on their chilli consumption habits and sensitivity to hotness standard solutions. After the screening test, the users participated in 12 Alternative Forced Choice (AFC)-sets, for each of three stimuli (capsaicin and 1-Penten-3-One (1P3O) odour and dried chilli powder). The dried chilli 3-AFC sample's concentration range was calculated to equate the same range of standard capsaicin stimuli for hotness sensation measurements. Concentrations of the three stimuli tested were in a range of (0.08–16.80 dry basis g $L^{-1}$) for standardised dried chilli powder, (0.10–20.16 mg $L^{-1}$) for capsaicin, and (0.01–2.04 μL $L^{-1}$) for 1P3O. Heavy chilli users group threshold, as anticipated, indicated the highest average recognition level of stimuli in terms of pungency odour perceived from dried chilli (5.88 g $L^{-1}$) and 1P3O (1.34 μL $L^{-1}$), as well as for hotness sensation from dried chilli (7.19 g $L^{-1}$) and capsaicin (12.79 mg $L^{-1}$) samples. The magnitudes of heavy user's thresholds were exponentially higher than that of light users. At the recognition thresholds of oral hotness perception, the level of capsaicin presented in dried chilli sample was found to be much lower than the concentration of standard capsaicin sample. It was concluded that pungent odours and other flavours in dried chilli, increase hotness perception as compared with capsaicin stimulus. We also confirmed that 1P3O contributes to chilli pungent odour.

**Keywords** Capsaicin, *Capsicum annuum*, hotness threshold, pungency, spiciness.

## Introduction

Chilli is a small pepper pod belonging to the *Capsicum* genus which delivers an oral hot sensation. Its widespread use is characteristic of tropical climate cuisines, due to the body's physiological adjustment (Mattes & Ludy, 2016; Defrin *et al.*, 2021), or so-called gustatory sweating (Mosley, 2017). Asia-Pacific represents the largest chilli production and consumption market, while the majority of global chilli users are in the food manufacturing industry (Centre for the Promotion of Imports from developing countries (CBI), 2022). In terms of global consumption, Europe has seen the fastest growth of 6.11% in chilli consumption (Mordor Intelligence, 2023). The global growth of chilli usage seen in recent years within the food sector could be attributed to the increases of popularity of veganism (as flavour enhancer), of exotic cuisines including curries and plant-based natural flavours, and of convenient ready-to-cook and ready-to-eat products (The Growth Report, 2020; CBI, Centre for the Promotion of Imports from developing countries (CBI), 2022; Han *et al.*, 2022; Jamaluddin *et al.*, 2022; Yin *et al.*, 2022). Chilli has also attracted more attention due to capsaicin's suggestive benefits on weight control (Hernández-Pérez *et al.*, 2020; Siebert *et al.*, 2022; Rezazadeh *et al.*, 2023).

Cayenne, Long Fed, and Spur peppers are commonly known names of *Capsicum annuum* L. var. *acuminatum* Fingerh. This type of chilli is grown and consumed worldwide. It is mostly and conveniently preserved in dried powder or flake forms for domestic consumption and export market. Food manufacturers use dried chilli for ready-to-cook products such as seasoning mixes, gravies, chilli pastes, and in ready-to-eat snacks and meals (Toontom *et al.*, 2016; Jamaluddin *et al.*, 2022; Seth & Singla, 2022). How much dried chilli can be used in

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the food items is limited by hotness sensation caused by capsaicin compounds, capsaicinoids. Capsaicin when ingested creates levels of irritation in the mouth such as tingling, warm, hot, or even burning or numbing sensations. The perceived hotness intensity is also influenced by other stimuli such as fat, salt, acid, and sugar in the food compositions (Kosmidou *et al.*, 2013; Schneider *et al.*, 2014; Zhang *et al.*, 2018; Ramírez-Rivera *et al.*, 2021) as well as pungent aromas (Toontom *et al.*, 2016; Korkmaz *et al.*, 2020). Other compounds such as sanshools predominantly found in Szechuan pepper also create tingling and numbness via tactile vibration (Hagura *et al.*, 2013) which is deemed to be a popular characteristic among Chinese food lovers (Zhang *et al.*, 2021). The temperature of food matrix also affects the hotness magnitude and duration (Leijon *et al.*, 2019; Han *et al.*, 2022; Wu *et al.*, 2024). These are a few of the factors, in addition to repeated exposure and familiarity with chilli, influencing hotness perception (Toontom *et al.*, 2016; Zhang *et al.*, 2018; Han *et al.*, 2022; Siebert *et al.*, 2022).

Our objective in this research was to measure the hotness sensation caused by capsaicin which is intensely found in dried chilli peppers in *C. annuum* L. Capsaicin acts by activating Transient Receptor Potential Vanilliod Subtype 1 (TRPV1s) receptors in the mouth, triggering piquancy or heat sensation (Nolden & Hayes, 2017), or spicy, tingling, and burning sensations, which are collectively identified from testers as oral hotness (Toontom *et al.*, 2016; Zhang *et al.*, 2020; Lyu *et al.*, 2021). We were also interested to explore the effects of aroma, 1-Penten-3-One (1P3O) which was proposed in a study conducted by Toontom *et al.* (2016) to be responsible for pungent odour in dried chilli (*C. annuum* Linn. Var. *acuminatum* Fingerh). That study analysed the dried chilli samples using Gas Chromatography – Mass Spectrometry (GC–MS) in conjunction with a trained sensory panel ($n = 15$). It was identified that 1P3O gave strong hotness sensation when mixed with capsaicin. The notion that 1P3O acts as a major contributor to pungent odour in fruits and vegetables has also been supported in several studies, as found in bell pepper (Luning *et al.*, 1995; van Ruth *et al.*, 1995), horseradish (McGorrin, 2019), olive oils (Genovese *et al.*, 2018;), rocket leaves (Raffo *et al.*, 2018), tomatoes and oranges (Mall *et al.*, 2018), and kiwifruits (Zhao *et al.*, 2021).

The human perception of stimulus concentration is measured by intensity unit or 'sensory threshold'. Sensory threshold has been defined and measured in several categories such as absolute or detection, recognition, difference, satiation, trade-off, rejection, or rejection tolerance thresholds (Prescott & Stevenson, 1995; Lawless, 2010; Lawless & Heymann, 2010; BS ISO 3972, 2011; Ardoin *et al.*, 2020). Detection threshold is understood as the minimum concentration of a particular stimulus that is detectable by testers, also known as stimulus awareness which may or may not be recognisable, while recognition threshold is specified concentration when the stimulus is correctly identified (Lawless, 2010; BS ISO 3972, 2011; BS ISO 13301, 2018). In this study, we measured recognition thresholds of standard compounds, in addition to measuring hotness perception in dried chilli solutions. Capsaicin was used as standard stimuli for measurement of hotness threshold measurement, and 1P3O for pungency odour. Dried chilli samples were made by calculating capsaicin content's equivalent in order to cover the concentration range of standard capsaicin solutions. We hypothesised that the hotness perception would be higher in dried chilli solutions when compared with standard capsaicin at the same capsaicin concentration.

Dried chilli was also used as stimuli in this study measuring for hotness and pungent odour thresholds, to explore effects of other food compounds in this popularly used condiment. One of our attempts in this study was to explore the hotness and pungency thresholds of mixed stimuli (i.e. not a single standard compound such as capsaicin or 1P3O). Dried chilli has been part of top-up condiments, similar to salt and pepper in western meal pattern. A sachet of dried chilli powder accompanies snacks and ready-to-eat meals in the South-East Asian market; it is commonly provided in instant foods packets as well as in food outlets as complimentary flavour enhancer. It was also anticipated in this study that 1P3O could be used as a case study for using aroma to enhance hotness perception, enabling lowering of the amount of capsaicin or sanshools and their burning sensations. This should appeal to food developers to introduce 'chilli sensation' while optimising chilli content in new markets where consumers are not accustomed to or are sceptical about having chilli in their food.

The literature has reported some inconsistency of thresholds arising from variations in measurement method, stimuli, tester's dietary and habitual amount of chilli consumption (Lawless, 2010; Zhang *et al.*, 2018; Siebert *et al.*, 2022). Frequency and the quantity of chilli normally consumed have been reported to have an impact on hotness perception (Cowart, 1987; Stevenson & Prescott, 1994; Lawless *et al.*, 2000; Reinbach *et al.*, 2007; Ludy & Mattes, 2012; Nolden & Hayes, 2017; Spinelli *et al.*, 2018; Siebert *et al.*, 2022; Su *et al.*, 2022; Zhang *et al.*, 2023). This study was correspondingly designed to recruit testers based on categories of their habitual chilli consumption (drawn from certain age groups to limit variations), and in anticipation that differences in thresholds would be observed among chilli user categories.

It is evident that there is a gap when comparing thresholds of both hotness and pungent odour using a homogeneous and large set of testers. Our study makes a contribution to the literature by focusing not only on standard capsaicin and 1P3O stimuli but also by examining the hotness thresholds in dried chilli samples. We have also explored sensory thresholds across three chilli user categories based on chilli intakes in regular diets: light, moderate, and heavy users.

## Materials and methods

### Recruitment of chilli users

The study took place at the sensory test facilities of Prince of Songkla University (PSU), Department of Food technology, in Thailand, over the period of 5 days. The recruitment criteria were based on the testers' chilli consumption (frequency and approximated declared amount per day) as well as their preference for popular Thai spicy food dishes (PSU Ethical approval No. 228/2551). First, 132 volunteer candidates whose age ranged between 18 and 35 years were invited to participate in a hotness screening test. The selected age range of testers was supported by the literature, which has indicated that young testers tended to have higher sensitivity and were less influenced by physiological factors (Fukunaga *et al*., 2005; Trachootham *et al*., 2018; Siebert *et al*., 2022; Su *et al*., 2022).

The candidates completed a questionnaire adapted from Byrnes & Hayes (2013), Ludy & Mattes (2012), Toontom *et al*. (2016), and Spinelli *et al*. (2018) which included hedonic responses. It comprised of five chilli consumption-related sections, as follows: (i) Frequency of consumption of chilli-containing foods, (ii) Amount of chilli added per dish, (iii) Self-reported hotness level in daily diet, (iv) Liking of chilli flavours in popular Thai spicy foods, and (v) Attitudes (including liking) towards hotness levels of selected food samples with various amount of added capsaicin. They then underwent tasting a series of twelve sets of 3-AFC capsaicin solutions (0.00–10.00 mg $L^{-1}$) to gauge hotness perception by individuals, as well as to give an idea of concentration range in the subsequent threshold tests. Following from this, 120 participants were recruited based on their hotness sensitivity, chilli intake habit and commitment to participate in all of the threshold tests. The recruited subjects received a small amount of incentive to participate on three testing days. Three groups of chilli users were then identified as Light (L), Moderate (M), and Heavy (H) users based on the characteristics shown in Table 1.

### The sample preparations

Based on the capsaicin ranges reported in hotness threshold studies, and the information gathered during the candidate sensitivity screening test, we then pilot-tested 3-AFC sample sets for 1P3O, capsaicin, and dried chilli solutions on small groups of testers recruited into H, M, and L groups ($n$ = 10 each) to finalise the coverage of our stimuli concentration test ranges. An objective was that an individual's thresholds should not fall either outside or near the limits of a range, but well within it.

The serials of increasing concentration dilutions for each stimulus (capsaicin, 1P3O, and dried chilli) were decided and prepared as stated in (a), (b), and (c) below, respectively. The sample serving design followed the 3-AFC method (ASTM E679-04, 2011; BS ISO 13301, 2018) with an increment factor of 1.62 presenting:

(a) Twelve sets of capsaicin (≥95.0%, Sigma, USA) concentrations (0.10, 0.16, 0.26, 0.43, 0.69, 1.12, 1.81, 2.93, 4.74, 7.68, 12.45, and 20.16 mg $L^{-1}$) for hotness threshold measurement.

(b) Twelve sets of 1P3O (97.0%, Aldrich, Germany) samples (0.01, 0.02, 0.03, 0.04, 0.07, 0.11, 0.18, 0.30, 0.48, 0.78, 1.26, and 2.04 μL $L^{-1}$) for pungent odour threshold measurement.

(c) Twelve sets of dried chilli (ground dried chilli *C. annuum* Linn. var. *acuminatum* Fingerh.) solutions (The research chilli peppers (Capsicum annuum Linn. var. acuminatum Fingerh.) were hand-picked, harvested, dried with hot air oven to reach the moisture content of 10%–13%, packed in aluminium laminated bags under vacuum condition, and then stored at –20 °C (Toontom *et al*., 2016). Just prior to each testing session, the chillies were ground to be used for the test. The dried chilli samples were passed through a sieve to get a homogeneous chilli powder (80 meshes), according to Thai Community Product Standard of ground chilli (TCPS 492–2004). The final range of concentrations 0.08–16.08 g $L^{-1}$ dried chilli solutions was decided based on estimated capsaicin content from the information collected from candidate screening and pilot tests. The mixtures of 2% ethanol and chilli powder were stirred under room temperature for 10 min and filtered using filter paper No.4 to prepare the stock solutions before administering them: (0.08, 0.13, 0.22, 0.36, 0.58, 0.93, 1.51, 2.44, 3.95, 6.40, 10.38, and 16.80 g $L^{-1}$). This series of dried chilli solutions also equate to capsaicin concentrations of 0.09, 0.15, 0.26, 0.42, 0.68, 1.09, 1.77, 2.85, 4.62, 7.49, 12.14, and 19.66 mg $L^{-1}$, respectively. These equivalent capsaicin concentrations were calculated based on capsaicin content of the study's dried chilli powder (identified by HPLC

**Table 1** Characteristics of chilli user groups

| | Light (L) chilli users ($n$ = 40) | | Moderate (M) chilli users ($n$ = 40) | | Heavy (H) chilli users ($n$ = 40) | |
|---|---|---|---|---|---|---|
| | Mode | Frequency (%) | Mode | Frequency (%) | Mode | Frequency (%) |
| Frequency of consumption of chilli-containing foods (scores 1–7)[†] | 2 | 88.57 | 4 | 40.00 | 6 | 82.86 |
| Self-classified hotness level in daily spicy food intake (scores 1–3)[‡] | 1 | 68.57 | 2 | 82.86 | 3 | 57.14 |

| | Mean score (L users) | Mean score (M users) | Mean score (H users) |
|---|---|---|---|
| Amount of chilli consumed per meal (estimated capsaicin content (mg g$^{-1}$)) | $0.49 \pm 0.34^{c}$ | $1.03 \pm 0.58^{b}$ | $1.71 \pm 0.42^{a}$ |
| Liking of chilli flavour in popular spicy dishes (scores 1–9)[§] | $3.80 \pm 0.91^{c}$ | $6.53 \pm 0.75^{b}$ | $7.45 \pm 0.94^{a}$ |

Different superscripts ($^{a–c}$) within a row show significant difference ($P \leq 0.05$).
[†]Frequency of chilli consumption (1 = rarely, 2 = less than once a month, 3 = a few times a month, 4 = three to four times a week, 5 = almost every day, 6 = everyday, 7 = many times a day).
[‡]Level of hotness in food in comparison to added dried chilli (1 = a sprinkle or less (less than 2.12 g/dish), 2 = around a teaspoon (up to 4.25 g/dish), 3 = a teaspoon to a tablespoon (4.25–9.71 g/dish), 4 = more than a tablespoon).
[§]Liking of chilli flavour (9-point category hedonic scale 1 = extremely disliked, 9 = extremely liked).

analysis in the dried chilli solutions as 0.98–1.29 mg g$^{-1}$). This analysis is reported in the Appendix S1. This set of dried chilli solutions was used for both hotness and pungent threshold measurements.

## Design of the threshold tests

Individual testers underwent preparation to taste a maximum of twelve sets for each measurement, in four distinct testing sessions (three triads each session) with a break of at least half an hour between them. Sniffing tests were conducted on the first testing day with two stimuli of 1P3O and dried capsaicin solutions for pungent odour threshold measurements. Then oral hotness threshold was tested on two separate days. Capsaicin solutions were tested on the second testing day and dried chilli solutions on the final day with a break between each testing day, that is, three testing days in total spread over 5 days (Fig. 1).

The test design applied sensory ascending 3-AFC method in a Randomised Complete Block Design (RCBD). Each 3-AFC set consisted of three test samples, the triad presented one target sample (i.e. either capsaicin, 1P3O, or dried chilli solutions), and two controls (2% ethanol made with filtered water) for each concentration level and for all hotness and pungent odour test sets. The cleansing materials used included sucrose solution and water for hotness measurements. The testers did not know that each sample set was presented to them in ascending order of concentration, nor were they told what substances (capsaicin, 1P3O, dried chilli, ethanol (control)) would be in the test solutions.

The testers were informed on each testing day that the test samples were from a range of test solutions representing hotness solutions and odourants. On the testing day, testers went through a maximum of four testing sessions, of three triad sets in each session. The participants were instructed to *identify* one sample in each set in which they could detect something different than the other two samples (controls). The testers were also asked to record if they noticed a detectable taste or odour, thus giving a certainty judgement (either guessing or not). If detected, they were then asked to *specify* what the detectable taste or odour of the sample was. The individual Best Estimated Threshold (BET) was calculated using the Geometric Mean (GM) of the concentration at which the highest hotness missed and the next higher concentration (correctly identified) of the hotness sensation or pungent odour. For example, (see Fig. 2 as an example of the results from capsaicin hotness thresholds identified by L users) tester ID2 GM is calculated from capsaicin concentrations of 0.26 and 0.43, which is 0.33.

For hotness threshold tests, samples of 10 mL of capsaicin and dried chilli solutions were presented in porcelain cups. The participants tasted each sample set under a red-lighted booth to mask the sample's colour. They were instructed to, firstly, hold a sample in the mouth for 15 s; secondly, spit the solution out; thirdly, wait for 30 s, and finally carry out the evaluation. After testing each sample, the participants were also required to rinse their mouths with sucrose solution (10% sucrose w/w in water) (Nasrawi & Pangborn, 1990; Lee & Kim, 2013) as the use of sugar contributes to reduction of oral irritations from capsaicin (Schneider *et al.*, 2014; Ramírez-Rivera *et al.*, 2021). Milk was

 

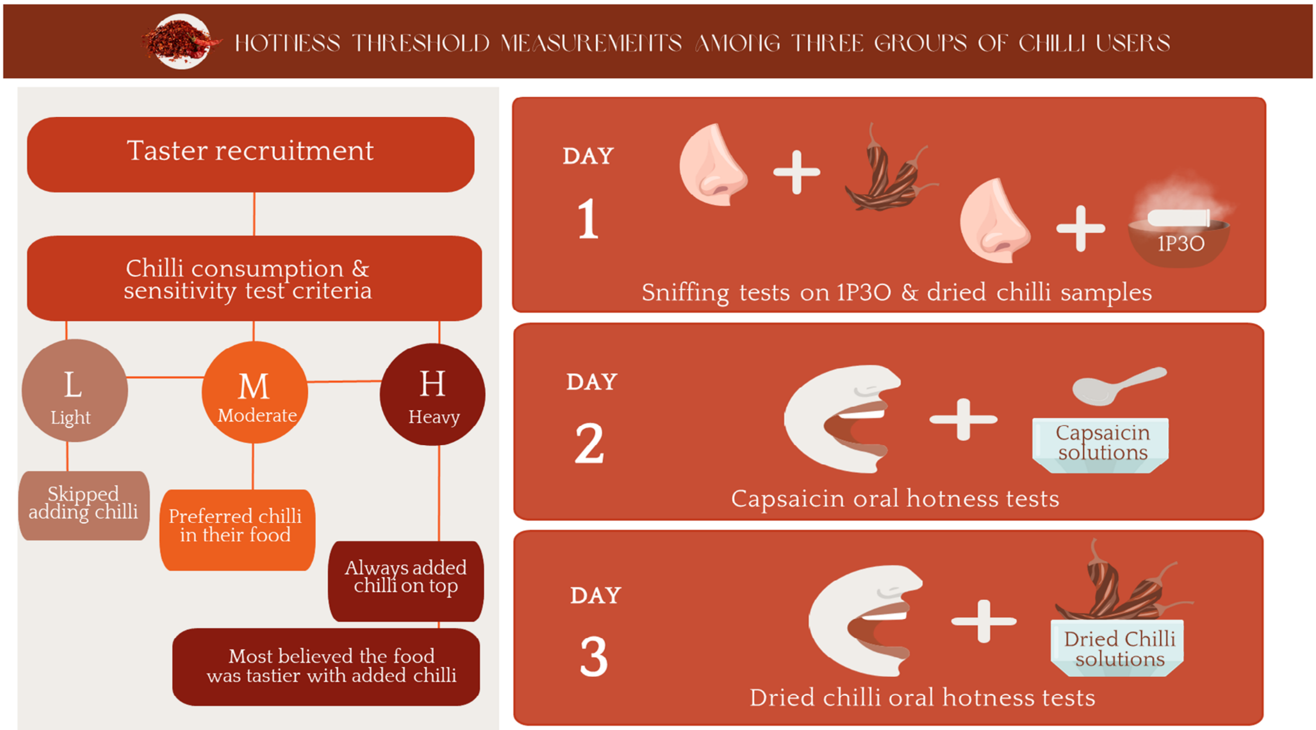


**Figure 1** Hotness and pungency threshold measurement design.

| Group (L) | Set 1 | Set 2 | Set 3 | Set 4 | Set 5 | Set 6 | Set 7 | Set 8 | Set 9 | Set 10 | Set 11 | Set 12 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Capsaicin Conc. (mg/l) | 0.1 | 0.16 | 0.26 | 0.43 | 0.69 | 1.12 | 1.81 | 2.93 | 4.74 | 7.68 | 12.45 | 20.16 |
| ID2 | 1 | 1 | 0 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| | Bitter | Bitter | Bitter | Hot | Hot | Hot | Hot | Hot | Hot | Hot | Hot | Hot |
| ID4 | 0 | 0 | 1 | 0 | 0 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| | Sweet | Sweet | Sweet | Bitter | Hot | Hot | Hot | Hot | Hot | Hot | Hot | Hot |
| ID9 | 1 | 1 | 0 | 0 | 1 | 1 | 0 | 1 | 1 | 1 | 1 | 1 |
| | Sweet | Sweet | Sweet | Sweet | Sweet | Bitter | Bitter | Hot | Hot | Hot | Hot | Hot |
| ID11 | 0 | 0 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| | Sweet | Sweet | Hot | Hot | Hot | Hot | Hot | Hot | Hot | Hot | Hot | Hot |
| ID15 | 0 | 0 | 1 | 0 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| | Sweet | Sweet | Bitter | Bitter | Hot | Hot | Hot | Hot | Hot | Hot | Hot | Hot |
| ID18 | 0 | 0 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| | Sweet | Sweet | Hot | Hot | Hot | Hot | Hot | Hot | Hot | Hot | Hot | Hot |

**Figure 2** An example of how the individual thresholds were identified from capsaicin threshold test in L users.

not chosen to be used as a palate cleanser due to its residual effects and interference to perception of the stimuli with low concentrations (based on our study's pilot test). The cleansing procedure continued by rinsing further five times with water and waiting for at least 3 min before proceeding to the next sample (adapted from Allison *et al.*, 2007; Lawless *et al.*, 2000). In total, there was at least a 30-min break between each sample set.

For pungent odour and dried chilli sample sets, the samples were presented in covered dark glass bottles of 10 mL solution to mask the interfering colour and to control the volatiles in the containers. The testers were instructed to *sniff* the sample for 5 s, and to

| Group (L) | Set 1 | Set 2 | Set 3 | Set 4 | Set 5 | Set 6 | Set 7 | Set 8 | Set 9 | Set 10 | Set 11 | Set 12 | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 1P3O (µl/l) | 0.1 | 0.16 | 0.26 | 0.43 | 0.69 | 1.12 | 1.81 | 2.93 | 4.74 | 7.68 | 12.45 | 20.16 | |
| ID2 | 1 | 1 | 0 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | |
| | Plastic | Plastic | Plastic | Pungent | Pungent | Pungent | Pungent | Pungent | Pungent | Pungent | Pungent | Pungent | smell |
| ID4 | 0 | 0 | 1 | 0 | 0 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | |
| | Plastic | Plastic | Metallic | Metallic | Metallic | Pungent | Pungent | Pungent | Pungent | Pungent | Pungent | Pungent | smell |
| ID9 | 1 | 1 | 0 | 0 | 1 | 1 | 0 | 1 | 1 | 1 | 1 | 1 | |
| | Detectab | Detectab | Metallic | Metallic | Off smell | Off smell | Metallic | Pungent | Pungent | Pungent | Pungent | Pungent | smell |
| ID11 | 0 | 0 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | |
| | Plastic | Plastic | Pungent | Pungent | Pungent | Pungent | Pungent | Pungent | Pungent | Pungent | Pungent | Pungent | smell |
| ID15 | 0 | 0 | 1 | 0 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | |
| | Detectab | Detectab | Detectab | Plastic | Pungent | Pungent | Pungent | Pungent | Pungent | Pungent | Pungent | Pungent | smell |
| ID18 | 0 | 0 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | |
| | Plastic | Plastic | Pungent | Pungent | Pungent | Pungent | Pungent | Pungent | Pungent | Pungent | Pungent | Pungent | smell |

**Figure 3** An example of how the individual thresholds were identified from 1P3O pungent odour test in L users.

rapidly evaluate them. Cleansing procedures between each 1P3O samples started with sniffing a non-scented facial tissue paper before proceeding to test the next sample in two-way air circulated testing rooms (Reilly & York, 2001; Frasnelli *et al.*, 2009). The participants recorded their choice and identified the smell for each set/concentration of the three stimuli. The example of recorded sniffing test results is shown in Fig. 3.

All the sample containers were coded with three-digit random numbers and were presented at a controlled temperature of 25 °C. The order of sample presentation was randomised within each triad, in every session throughout the three testing days.

## Statistical methods

The calculations of group recognition thresholds of hotness and pungent odour were carried out using Average Threshold from individual BET data. Logistic regression on pooled data was also estimated (Group Threshold) to provide a comparison.

ASTM E679-04 (2011) suggests calculation of average group threshold using individual best estimate threshold (BET). The group BETs were correspondingly calculated based on the individual BETs (ASTM E679-04, 2011). The geometric mean threshold is a measure of central tendency calculated by multiplying a series of numbers and taking the *n*th root of the product, where '*n*' is the number of items in the series (Markowitz, 2012). In this study, the Average Threshold was computed as the geometric mean of the highest concentration missed (or incorrect) and the next higher concentration (correctly identified) (Lawless, 2010; Zhang *et al.*, 2023).

Logistic regression was also estimated to measure the probability of correct choices by 3-AFC method (Lawless, 2010; BS ISO 13301, 2018). The concentrations of dried chilli and capsaicin hotness and pungent odour of 1P3O were the predictor variables. A correct identification of an odd sample was the predicted outcome (the dependent variable). To determine the threshold levels using the logistic regression method, the percentage correct (% correct) was converted to the percentage correct response chance (% correct response chance). That was done for every concentration and each sample using Abbott's formula (eqn 1) (Lawless, 2010), and by plotting the percentage correct response chance against the concentration level.

$$\%\text{correct above chance} = 100 \times \frac{(\%\text{correct} - \%\text{correct by chance})}{(100 - \%\text{correct by chance})} \quad (1)$$

Logarithmic trend lines in the form $Y = \mathrm{M}\ \ln(x) + c$ were fitted with data for determining thresholds. Theoretically, the thresholds are usually determined at the probability of a 50% recognition level. Regarding 3-AFC test, with a probability of 33.3% correct by chance, 66.7% of identifications were thus required for a correct answer to represent a true proportion of 50% (Lawless, 2010; Cliff *et al.*, 2011). *The correct identification* was calculated as shown in eqn 2.

$$50 = \frac{100 \times (y - 33.3)}{(100 - 33.3)} \qquad y = 66.7 \quad (2)$$

The proportions of correct response were then plotted and fitted to a logistic regression equation for predicting the individual recognition threshold as displayed in Fig. 7.

Group and Average thresholds were calculated across the three stimuli measured. Threshold values were tested for normal distribution and were log-transformed prior to performing the Analysis of Variance (ANOVA) (Cliff *et al.*, 2011). ANOVA was also employed to evaluate the effects on the likely distinct chilli-user groups. Significant differences between means were estimated by Duncan's new multiple range test (DMRT), with a level of significance of 0.05. Statistical analyses were performed by using the Statistical Package for Social Science (IBM Corp, 2021).

## Results and discussion

### Classification of chilli users

The classification of the participants was based on both quantitative and qualitative information gathered from their chilli consumption (the questionnaire is available upon request). As proposed by Ludy & Mattes (2012) and Byrnes & Hayes (2013), hedonic responses to hotness were included as predictors to classify threshold groups. Chilli consumption per dish was estimated from reported amount of dried and fresh chilli normally used in four popular daily foods (papaya salad, noodle, spicy & sour soups (Tom yum), and spiced salad). Thus, from the analysis of frequency of chilli use, it was possible to identify three categories of chilli users: Light (L), Moderate (M), and Heavy (H) as summarised in Table 1.

Light chilli users consisted of a group of twenty-nine females and eleven males whose ages ranged from 20 to 35 years. They typically added chilli in their foods <1 month$^{-1}$ with an estimated chilli use content of less than 2.12 g/dish in their diet (Ludy & Mattes, 2012; Byrnes & Hayes, 2013). The average chilli liking score for this group was close to the characterisation 'disliked slightly' (score 3.80/9).

Moderate chilli users consisted of twenty-eight females and twelve males whose age ranged from 19 to 35 years who ate spicy foods every other day (3–4 times week$^{-1}$) in amounts of approximately 2.12–4.25 g/dish. Their average chilli liking score was 'liked moderately' (score 6.59/9).

Heavy chilli users consisted of twenty-five females and fifteen males whose age ranged from 20 to 34 years. These users ate chilli-containing foods daily, had a habit of adding chiili into the dishes, and liked chilli 'taste' very much (score 7.45/9). Their estimated chilli consumption ranging 4.25–9.71 g/dish. These chilli users had the least discriminating ability of hotness sensation as established from pre-testing capsaicin stimulus.

In comparison, Othman *et al.* (2011) reported 15.5 g of chilli daily intake from the population of Riyadh city, equating to 7.58 mg of capsaicin/person/day. This study compared its results to other studies, finding reported chilli intake to be higher than reported from samples of Thais (5 g day$^{-1}$) and Indians (2.5 g day$^{-1}$), but lower than Mexicans (20 g day$^{-1}$). Orellana-Escobedo *et al.* (2012) reported average chilli intake among their Mexican testers of 21.92 g day$^{-1}$ (8 kg/year per capita). Mexican chilli users are well known for their regular high chilli intake and would be anticipated to have higher threshold for hotness perception, but it was found to be in close range or lower when compared to American users (Orellana-Escobedo *et al.*, 2012). The authors discussed that this could arise from variations in the preparation of sample as well as possible desensitisation among chronic chilli users, which could be the case for our Heavy users. Self-reported chilli intake frequency is also used to categorise chilli users. Lyu *et al.* (2021) found a significant difference in intake frequency of spicy food per year between their consumers recruited from young Caucasian adults in the Netherlands. The intake of spicy food 'once a week' (fifty-two times a year) was then applied as a cut-off point to separate frequent and infrequent chilli users. The significant difference of hedonic scores between the two chilli users was observed in the study, as well as from our L, M, and H frequent chilli users with significant differences ($P \leq 0.05$) as shown in Table 1. L users agreed that they sometimes avoided having chilli in ready-to-eat/ready-to-cook dishes whilst M users preferred to have chilli, and H users always added chilli and thought the food was tastier with the chilli top-up. Some of our testers categorised as Heavy Users were likely to have a similar amount of chilli intake per day (4.25–9.71 g/dish) as those Saudi Arabian testers reported by Othman *et al.* (2011) whereas the reported 5 g of chilli intake in Thais is consistent with the range of chilli intake reported from our Moderate Users (assuming they ate at least two of the dishes a day). Although the measured amount and frequency of chilli intake were not proportionately accurate predictors of hotness threshold in this study, this categorisation as L, M, and H provides a basis for comparison to chilli users elsewhere in the world.

## Individual BET and Group thresholds of pungent odour and hotness

### Individual BETs

The identified concentrations when the substance was detected and when it was correctly recognised were recorded in both hotness and pungent odour tests. Even though we focus on discussing recognition thresholds in this paper, we found interesting patterns of how the hotness and pungent odour were detected (i.e. the odd sample was correctly chosen) but were not recognised (i.e. hotness sensation or pungent odour was not correctly specified) within and among L, M, and H users in the raw data records. For example, results from oral hotness test with dried chilli solutions (0.08–16.8 g $L^{-1}$, equivalent to capsaicin 0.09–19.66 mg $L^{-1}$) indicate a clear perception pattern in L users. Once the sample was correctly detected and recognised, they would be able to correctly recognise the hot sample in other triads with higher concentrations (Fig. 4), whereas H users showed inconsistency (Fig. 5). This could be most likely caused by lower sensitivity to capsaicin in heavy, frequent chilli users. This is also reported in recent studies such as Lyu *et al*. (2021) and Su *et al*. (2022). The 'noise' in other compounds present in dried chilli solutions seems to have greater impact on H group's hotness perception and test performance than on the L group. This is evidently seen in various tastes being incorrectly identified in the H user' results (Fig. 5). Han *et al*. (2022) suggested that capsaicin increased taste sensitivity by lowering recognition threshold at low concentration of capsaicin around 1 μM (0.31 mg $L^{-1}$). While this may be the case of L users at Triads 3 and 4 of the dried chilli solutions (0.22–0.36 g $L^{-1}$ equivalent to capsaicin 0.26–0.42 mg $L^{-1}$) when bitter taste was identified on the correctly detected samples, it is not the case from H group results. In terms of the range of substance concentrations, the maximum of capsaicin concentration used in this study is higher than 64 μM, which gives coverage of most capsaicin concentrations reported in the threshold tests. Results from H group show the same inconsistency pattern when hotness threshold was measured in capsaicin solutions (Fig. 6). Sweet taste was incorrectly identified in low capsaicin concentration, which could be associated with cleansing material (10% sucrose solution), and more bitter taste was incorrectly identified in later triads with higher capsaicin concentrations. No sour taste was identified from capsaicin samples but was identified in dried chilli solutions which could be an influence from several volatile acids and furans found in dried cayenne pepper such as acetic acids and 2-Acetylfuran (Toontom *et al*., 2016; Korkmaz *et al*., 2020).

## Group thresholds

*Average thresholds based on individual BETs*

Based on the results from Individual BETs, the average scores of Geometric means on each measurement of L, M, and H groups are displayed in Table 2. It can be seen clearly that the Average thresholds of oral hotness and pungent odour are significantly different among the three chilli users ($P < 0.05$). The results will be next discussed with Group thresholds calculated from Logistic regression analyses.

*Hotness thresholds from capsaicin and dried chilli stimuli*

The logistic regression predicts the recognition thresholds for capsaicin hotness sensation to be 12.79 mg $L^{-1}$ for heavy users, 2.36 mg $L^{-1}$ for the moderate, and 0.96 mg $L^{-1}$ for light chilli users, respectively (Table 3 and Fig. 7c). As for the stimuli recognition regarding dried chilli samples, we found the predicted Group thresholds of hotness exponentially decline from 7.19 g $L^{-1}$ for the heavy to 2.23 g $L^{-1}$ for the moderate and 0.58 g $L^{-1}$ for the light users (Fig. 7d). While Average BET thresholds agree with the predicted group threshold as the heavy chilli-user group also had the highest hotness threshold of 7.07 g $L^{-1}$ stimulated by dried chilli samples, and 11.75 mg $L^{-1}$ by capsaicin solutions. The two calculation methods present similar range of threshold values across the three user groups despite the possibility of non-linear hotness perception.

The hotness thresholds of capsaicin calculated from BETs of 0.87, 2.09, and 11.75 mg $L^{-1}$ are equivalent to 2.81, 6.74, and 37.90 μM (0.31 mg $L^{-1}$ capsaicin is equal to 1 μM) from our L, M and H users, respectively. The hotness thresholds of dried chilli, represent 0.68, 2.61, and 8.41 mg $L^{-1}$ capsaicin (average of 1.17 mg capsaicin $g^{-1}$ of the researched dried chilli) which are equivalent to 2.19, 8.42, and 27.13 μM for L, M, and H users, respectively.

It can be concluded that the dried chilli solutions affected oral hotness perception with lower capsaicin content when compared with standard capsaicin solution, in L and H users.

*Pungent thresholds from 1P3O and dried chilli stimuli*

From the logistic regression prediction, the predicted recognition thresholds for pungent odour of 1P3O stimulus indicated it was at a concentration of 1.34 μL $L^{-1}$ for the heavy, 0.23 μL $L^{-1}$ for the moderate, and 0.06 μL $L^{-1}$ for the light users, respectively (Fig. 7a). These values are close to the Average BET thresholds (1.27, 0.20, 0.04 μL $L^{-1}$ from H, M, and L groups, respectively). By sniffing, the heavy chilli users also recognised low intensity of pungent odour in dried chilli at a concentration of 5.88 g $L^{-1}$ (Fig. 7b) which is close to the Average BET threshold of 5.76 g $L^{-1}$.

Heavy chilli users tended to identify pungent odour thresholds of 1P3O at exponentially higher levels of over twenty times than that of Light users. This is another indication that heavy chilli users are not only less sensitive to oral hotness but also to pungent odour. This is likely due to long-term use of chilli (Cowart, 1987; Stevenson & Prescott, 1994; Lawless *et al*., 2000; Reinbach *et al*., 2007; Ludy & Mattes, 2012; Nolden & Hayes, 2017; Siebert *et al*., 2022; Zhang *et al*., 2023).

Notably, when testing 1P3O at low concentrations, the testers identified the samples as having 'metallic', 'off smell', and 'irritably strange' odours (Fig. 3). A previous study reported a group of ketone compounds that contributed to metallic aroma perceived by sensory panellists in green tea; however, 1P3O in that

 

| Triad test set (L users) | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Dried chilli (g/l) | 0.08 | 0.13 | 0.22 | 0.36 | 0.58 | 0.93 | 1.51 | 2.44 | 3.95 | 6.4 | 10.38 | 16.8 |
| Capsaicin equivalent (mg/l) | 0.09 | 0.15 | 0.26 | 0.42 | 0.68 | 1.09 | 1.77 | 2.85 | 4.62 | 7.49 | 12.14 | 19.66 |
| ID2 | 0 Sweet | 0 Sweet | 0 Bitter | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot |
| ID4 | 0 Sweet | 0 Sweet | 0 Sour | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot |
| ID9 | 1 Sweet | 0 Sweet | 0 Sour | 0 Sour | 1 Sour | 1 Bitter | 1 Bitter | 0 Bitter | 1 Hot | 1 Hot | 1 Hot | 1 Hot |
| ID11 | 0 Sweet | 1 Sweet | 1 Sour | 1 Sour | 0 Sour | 0 Bitter | 0 Bitter | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot |
| ID15 | 0 Sweet | 1 Sour | 0 Sweet | 1 Sour | 0 Sour | 0 Bitter | 0 Bitter | 0 Bitter | 0 Bitter | 1 Hot | 1 Hot | 1 Hot |
| ID18 | 0 Sweet | 0 Sour | 0 Sour | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot |
| ID20 | 0 Sweet | 1 Sweet | 1 Bitter | 0 Bitter | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot |
| ID25 | 0 Sweet | 0 Sweet | 0 Bitter | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot |
| ID27 | 0 Sweet | 0 Sweet | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot |
| ID30 | 0 Sweet | 0 Sweet | 0 Bitter | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot |
| ID36 | 0 Sweet | 1 Sweet | 1 Bitter | 1 Bitter | 1 Bitter | 1 Bitter | 0 Sour | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot |
| ID40 | 0 Sweet | 0 Sweet | 0 Bitter | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot |
| ID41 | 0 Sweet | 0 Sweet | 0 Sour | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot |
| ID44 | 0 Sweet | 1 Sweet | 1 Hot | 1 Hot | 1 Bitter | 1 Bitter | 0 Bitter | 0 Bitter | 1 Hot | 1 Hot | 1 Hot | 1 Hot |
| ID47 | 1 Sweet | 1 Sweet | 1 Hot | 0 Bitter | 1 Bitter | 0 Bitter | 1 Hot | 1 Hot | 0 Hot | 1 Hot | 1 Hot | 1 Hot |
| ID48 | 0 Sweet | 0 Sweet | 0 Sour | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot |
| ID51 | 0 Sweet | 0 Sweet | 0 Bitter | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot |
| ID59 | 0 Sweet | 1 Sweet | 1 Bitter | 0 Bitter | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot |
| ID60 | 0 Sweet | 0 Sour | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot |
| ID62 | 0 Sweet | 1 Sweet | 1 Sweet | 0 Sour | 0 Sour | 0 Sour | 0 Bitter | 0 Bitter | 1 Hot | 1 Hot | 1 Hot | 1 Hot |
| ID63 | 0 Sweet | 1 Hot | 0 Sweet | 1 Sour | 0 Sour | 1 Sour | 1 Bitter | 1 Hot | 0 Bitter | 1 Hot | 1 Hot | 1 Hot |
| ID66 | 1 Sweet | 1 Hot | 0 Sweet | 0 Sour | 0 Sour | 1 Hot | 0 Bitter | 0 Bitter | 0 Bitter | 0 Bitter | 0 Bitter | 1 Hot |
| ID67 | 0 Sweet | 1 Hot | 0 Sweet | 0 Sour | 0 Sour | 0 Sour | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot |
| ID70 | 0 Sweet | 0 Sweet | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot |
| ID74 | 1 Sweet | 0 Sweet | 1 Sweet | 0 Sweet | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot |
| ID75 | 1 Sweet | 0 Sweet | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot |
| ID78 | 1 Sweet | 1 Sweet | 0 Sour | 0 Sour | 0 Sour | 1 Hot | 1 Hot | 1 Bitter | 1 Bitter | 0 Hot | 1 Hot | 1 Hot |
| ID80 | 1 Sweet | 1 Sweet | 0 Sour | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot |
| ID83 | 1 Sweet | 0 Sweet | 1 Sour | 0 Bitter | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot |
| ID85 | 1 Sweet | 0 Sweet | 1 Sweet | 0 Sweet | 0 Bitter | 0 Bitter | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot |
| ID88 | 0 Sweet | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot |
| ID90 | 0 Sweet | 1 Hot | 1 Sour | 0 Bitter | 0 Bitter | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot |
| ID91 | 1 Sweet | 0 Hot | 0 Sour | 0 Bitter | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot |
| ID97 | 1 Sweet | 0 Bitter | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot |
| ID98 | 0 Sweet | 0 Sour | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot |
| ID100 | 0 Sweet | 0 Sour | 1 Hot | 0 Sour | 0 Sour | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot |
| ID101 | 0 Sweet | 0 Sour | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot |
| ID107 | 0 Sweet | 1 Sour | 0 Sour | 0 Bitter | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot |
| ID110 | 1 Sweet | 0 Bitter | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot |
| ID116 | 1 Sweet | 0 Bitter | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot | 1 Hot |

**Figure 4** Individual BET of hotness from dried chilli samples among L users. *Data in bold red text are responses correctly identified as hotness, whereas the ones in bold blue text are the highest concentration missed. **Tester IDs highlighted in colours show inconsistency patterns of identifying hotness samples.

| Triad test set (H users) | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Dried chilli (g/l) | 0.08 | 0.13 | 0.22 | 0.36 | 0.58 | 0.93 | 1.51 | 2.44 | 3.95 | 6.4 | 10.38 | 16.8 |
| Capsaicin equivalent (mg/l) | 0.09 | 0.15 | 0.26 | 0.42 | 0.68 | 1.09 | 1.77 | 2.85 | 4.62 | 7.49 | 12.14 | 19.66 |
| ID6 | 1 | 1 | 0 | 1 | 1 | 0 | 0 | 1 | 1 | 0 | 1 | 1 |
| | Sweet | Sweet | Bitter | Sweet | Sweet | Sweet | Sweet | Sweet | Sweet | Bitter | Hot | Hot |
| ID8 | 1 | 1 | 0 | 1 | 1 | 0 | 0 | 1 | 1 | 0 | 1 | 1 |
| | Sweet | Sweet | Sweet | Sweet | Sour | Sour | Sour | Sour | Sour | Bitter | Hot | Hot |
| ID10 | 0 | 1 | 1 | 0 | 1 | 1 | 1 | 1 | 0 | 1 | 1 | 1 |
| | Sweet | Sweet | Sweet | Sweet | Sour | Sour | Sour | Bitter | Bitter | Hot | Hot | Hot |
| ID16 | 0 | 0 | 0 | 0 | 0 | 0 | 1 | 1 | 0 | 1 | 1 | 1 |
| | Sweet | Sweet | Sweet | Sour | Sour | Sour | Sour | Bitter | Bitter | Hot | Hot | Hot |
| ID17 | 0 | 1 | 1 | 1 | 1 | 0 | 1 | 1 | 1 | 0 | 1 | 1 |
| | Sweet | Sweet | Bitter | Bitter | Sour | Sour | Sour | Bitter | Bitter | Bitter | Hot | Hot |
| ID21 | 0 | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 1 | 1 |
| | Sweet | Sweet | Sweet | Bitter | Bitter | Sour | Sour | Bitter | Bitter | Bitter | Hot | Hot |
| ID22 | 1 | 1 | 1 | 0 | 1 | 1 | 0 | 0 | 1 | 1 | 1 | 0 |
| | Sweet | Sweet | Sweet | Bitter | Bitter | Sour | Sour | Bitter | Bitter | Bitter | Hot | Hot |
| ID24 | 1 | 0 | 0 | 0 | 1 | 1 | 0 | 1 | 1 | 0 | 0 | 1 |
| | Sweet | Sweet | Sweet | Bitter | Bitter | Bitter | Bitter | Bitter | Bitter | Bitter | Hot | Hot |
| ID26 | 1 | 0 | 1 | 0 | 0 | 1 | 1 | 1 | 1 | 1 | 1 | 0 |
| | Sweet | Sweet | Sweet | Sweet | Sweet | Bitter | Bitter | Bitter | Bitter | Bitter | Hot | Hot |
| ID33 | 0 | 0 | 0 | 1 | 0 | 1 | 0 | 1 | 1 | 0 | 1 | 0 |
| | Sweet | Sweet | Sweet | Sour | Sour | Bitter | Bitter | Bitter | Bitter | Hot | Hot | Hot |
| ID35 | 0 | 0 | 1 | 0 | 0 | 0 | 1 | 1 | 0 | 1 | 0 | 1 |
| | Sweet | Sweet | Sweet | Sour | Sour | Bitter | Bitter | Bitter | Bitter | Bitter | Bitter | Hot |
| ID37 | 0 | 0 | 0 | 1 | 1 | 0 | 1 | 0 | 1 | 1 | 1 | 1 |
| | Sweet | Sweet | Sweet | Sweet | Sour | Sour | Sour | Bitter | Hot | Hot | Hot | Hot |
| ID39 | 0 | 1 | 1 | 1 | 1 | 0 | 1 | 1 | 1 | 1 | 1 | 1 |
| | Sweet | Sweet | Sweet | Sweet | Sour | Sour | Hot | Hot | Hot | Hot | Hot | Hot |
| ID42 | 0 | 0 | 0 | 0 | 1 | 0 | 1 | 1 | 0 | 1 | 0 | 1 |
| | Sweet | Sweet | Sweet | Sweet | Sour | Sour | Hot | Hot | Hot | Hot | Bitter | Hot |
| ID43 | 0 | 0 | 0 | 1 | 0 | 1 | 1 | 1 | 0 | 1 | 0 | 1 |
| | Sweet | Sweet | Sweet | Sour | Sour | Sour | Hot | Hot | Hot | Hot | Bitter | Hot |
| ID45 | 0 | 0 | 1 | 0 | 0 | 0 | 1 | 1 | 1 | 1 | 1 | 0 |
| | Sweet | Sweet | Sweet | Sweet | Sour | Sour | Sour | Hot | Hot | Hot | Bitter | Bitter |
| ID46 | 0 | 0 | 0 | 1 | 1 | 1 | 1 | 0 | 1 | 1 | 0 | 1 |
| | Sweet | Sweet | Sweet | Sweet | Sour | Sour | Sour | Sour | Sour | Hot | Hot | Hot |
| ID50 | 0 | 0 | 1 | 0 | 0 | 0 | 1 | 1 | 1 | 1 | 1 | 0 |
| | Sweet | Sweet | Sweet | Sweet | Sour | Sour | Sour | Bitter | Bitter | Bitter | Bitter | Bitter |
| ID52 | 0 | 0 | 0 | 1 | 0 | 0 | 1 | 1 | 0 | 1 | 0 | 1 |
| | Sweet | Sweet | Sweet | Sweet | Sour | Sour | Sour | Bitter | Bitter | Bitter | Hot | Hot |
| ID53 | 0 | 1 | 1 | 1 | 1 | 0 | 1 | 1 | 1 | 1 | 1 | 1 |
| | Sweet | Sweet | Sweet | Sweet | Sour | Sour | Hot | Hot | Hot | Hot | Hot | Hot |
| ID55 | 0 | 0 | 1 | 0 | 0 | 0 | 1 | 0 | 0 | 1 | 0 | 1 |
| | Sweet | Sweet | Sweet | Sweet | Sour | Sour | Sour | Sour | Bitter | Bitter | Bitter | Hot |
| ID57 | 1 | 0 | 0 | 1 | 0 | 0 | 0 | 0 | 1 | 1 | 1 | 0 |
| | Sweet | Sweet | Sweet | Sweet | Sour | Sour | Sour | Sour | Bitter | Bitter | Hot | Hot |
| ID61 | 1 | 0 | 1 | 1 | 0 | 1 | 0 | 0 | 1 | 1 | 0 | 1 |
| | Sweet | Sweet | Sour | Sour | Sour | Sour | Sour | Bitter | Bitter | Bitter | Bitter | Hot |
| ID64 | 0 | 0 | 1 | 0 | 0 | 1 | 1 | 0 | 0 | 0 | 0 | 1 |
| | Sweet | Sweet | Sweet | Sour | Sour | Sour | Sour | Sour | Bitter | Bitter | Bitter | Hot |
| ID68 | 0 | 0 | 0 | 1 | 1 | 0 | 0 | 1 | 0 | 0 | 1 | 0 |
| | Sweet | Sweet | Sweet | Sour | Sour | Sour | Sour | Sour | Bitter | Bitter | Bitter | Hot |
| ID69 | 0 | 1 | 0 | 1 | 1 | 0 | 1 | 0 | 1 | 0 | 1 | 1 |
| | Sweet | Sweet | Sweet | Sour | Sour | Bitter | Hot | Hot | Hot | Hot | Hot | Hot |
| ID71 | 0 | 0 | 1 | 1 | 0 | 1 | 0 | 0 | 1 | 0 | 1 | 1 |
| | Sweet | Sweet | Sweet | Sour | Sour | Bitter | Bitter | Bitter | Bitter | Bitter | Hot | Hot |
| ID73 | 0 | 0 | 0 | 0 | 0 | 0 | 1 | 1 | 0 | 1 | 0 | 1 |
| | Sweet | Sweet | Sweet | Sour | Sour | Bitter | Bitter | Bitter | Bitter | Bitter | Bitter | Hot |
| ID76 | 1 | 0 | 1 | 0 | 0 | 1 | 1 | 0 | 1 | 1 | 1 | 1 |
| | Sweet | Sweet | Sweet | Sour | Sour | Bitter | Bitter | Bitter | Hot | Hot | Hot | Hot |
| ID77 | 1 | 1 | 0 | 0 | 0 | 1 | 0 | 0 | 0 | 1 | 1 | 1 |
| | Sweet | Sweet | Sweet | Sweet | Sour | Bitter | Bitter | Bitter | Bitter | Hot | Hot | Hot |
| ID86 | 1 | 0 | 0 | 0 | 1 | 1 | 0 | 0 | 1 | 1 | 0 | 1 |
| | Sweet | Sweet | Sweet | Sweet | Sour | Bitter | Bitter | Bitter | Bitter | Bitter | Bitter | Hot |
| ID87 | 0 | 0 | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 1 | 1 | 1 |
| | Sweet | Sweet | Sweet | Sweet | Sour | Bitter | Bitter | Bitter | Bitter | Hot | Hot | Hot |
| ID94 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 1 | 0 | 1 | 1 | 1 |
| | Sweet | Sweet | Sweet | Sweet | Sour | Bitter | Bitter | Bitter | Bitter | Hot | Hot | Hot |
| ID95 | 1 | 1 | 1 | 0 | 0 | 1 | 0 | 0 | 1 | 1 | 1 | 1 |
| | Sweet | Sweet | Sweet | Sweet | Sour | Bitter | Bitter | Bitter | Hot | Hot | Hot | Hot |
| ID99 | 0 | 1 | 0 | 0 | 0 | 1 | 0 | 0 | 1 | 0 | 1 | 0 |
| | Sweet | Sweet | Sweet | Sweet | Sour | Sour | Bitter | Bitter | Bitter | Bitter | Hot | Hot |
| ID106 | 0 | 0 | 0 | 1 | 1 | 1 | 0 | 0 | 1 | 1 | 0 | 1 |
| | Sweet | Sweet | Sweet | Sweet | Sour | Sour | Bitter | Bitter | Bitter | Bitter | Bitter | Hot |
| ID109 | 0 | 1 | 0 | 0 | 0 | 1 | 0 | 0 | 1 | 0 | 1 | 1 |
| | Sweet | Sweet | Sweet | Sweet | Sour | Sour | Bitter | Bitter | Hot | Hot | Hot | Hot |
| ID111 | 1 | 0 | 0 | 1 | 1 | 1 | 0 | 0 | 1 | 0 | 1 | 1 |
| | Sweet | Sweet | Sweet | Sweet | Sour | Sour | Bitter | Bitter | Bitter | Bitter | Hot | Hot |
| ID115 | 0 | 0 | 0 | 0 | 1 | 1 | 0 | 1 | 0 | 1 | 0 | 1 |
| | Sweet | Sweet | Sweet | Sweet | Sour | Sour | Bitter | Bitter | Bitter | Bitter | Bitter | Hot |
| ID118 | 1 | 1 | 0 | 0 | 1 | 1 | 0 | 1 | 1 | 0 | 1 | 1 |
| | Sweet | Sweet | Sour | Sour | Sour | Sour | Sour | Bitter | Bitter | Bitter | Hot | Hot |
| ID121 | 1 | 0 | 0 | 1 | 1 | 0 | 1 | 0 | 0 | 1 | 1 | 1 |
| | Sweet | Sweet | Sweet | Sour | Sour | Sour | Sour | Bitter | Bitter | Hot | Hot | Hot |
| ID122 | 0 | 1 | 1 | 0 | 0 | 0 | 1 | 1 | 0 | 0 | 1 | 1 |
| | Sweet | Sweet | Sweet | Sweet | Sweet | Sour | Sour | Bitter | Bitter | Bitter | Hot | Hot |

**Figure 5** Individual BET of hotness from dried chilli samples among H users. *Data in bold red text are responses correctly identified as hotness, whereas the ones in bold blue text are the highest concentration missed. **Tester IDs highlighted in colours show inconsistency patterns of identifying hotness samples.

 

| Triad test set (H users) | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Capsaicin conc (mg/l) | 0.1 | 0.16 | 0.26 | 0.43 | 0.69 | 1.12 | 1.81 | 2.93 | 4.74 | 7.68 | 12.45 | 20.16 |
| ID6 | 0 Sweet | 0 Sweet | 1 Sweet | 0 Sweet | 0 Sweet | 1 Sweet | 0 Sweet | 1 Bitter | 1 Bitter | 1 Bitter | 1 Bitter | 0 Bitter |
| ID8 | 0 Sweet | 1 Sweet | 0 Sweet | 0 Sweet | 1 Sweet | 0 Sweet | 0 Sweet | 0 Bitter | **0 Bitter** | **1 Hot** | **1 Hot** | **1 Hot** |
| ID10 | 0 Sweet | 1 Sweet | 1 Sweet | 0 Sweet | 0 Bitter | 1 Bitter | 0 Bitter | 1 Bitter | **0 Bitter** | **1 Hot** | **1 Hot** | **1 Hot** |
| ID16 | 0 Sweet | 0 Sweet | 0 Sweet | 1 Sweet | 1 Bitter | 0 Bitter | 0 Bitter | 0 Bitter | 1 Bitter | **1 Bitter** | **1 Hot** | 0 Hot |
| ID17 | 0 Sweet | 1 Sweet | 0 Sweet | 1 Sweet | 0 Bitter | 1 Bitter | 0 Bitter | 1 Bitter | 0 Bitter | 0 Bitter | 0 Bitter | 0 Bitter |
| ID21 | 0 Sweet | 0 Sweet | 0 Bitter | 0 Bitter | 1 Bitter | 0 Bitter | 0 Bitter | 0 Bitter | 1 Bitter | 0 Bitter | 1 Bitter | 0 Bitter |
| ID22 | 0 Sweet | 1 Sweet | 0 Bitter | 0 Bitter | 0 Bitter | 0 Bitter | 0 Bitter | 1 Bitter | 1 Bitter | 0 Bitter | **0 Hot** | **1 Hot** |
| ID24 | 1 Sweet | 0 Sweet | 0 Bitter | 0 Bitter | 1 Bitter | 1 Bitter | 1 Bitter | 0 Bitter | **0 Bitter** | **1 Hot** | **1 Hot** | **1 Hot** |
| ID26 | 0 Sweet | 1 Sweet | 0 Bitter | 0 Bitter | 1 Bitter | 1 Bitter | 0 Bitter | 1 Bitter | **1 Bitter** | **1 Hot** | 0 Hot | **1 Hot** |
| ID33 | 0 Sweet | 0 Sweet | 1 Sweet | 0 Sweet | 1 Bitter | 0 Bitter | 0 Bitter | 0 Bitter | 0 Bitter | 0 Bitter | **0 Bitter** | **1 Hot** |
| ID35 | 1 Sweet | 1 Sweet | 0 Sweet | 1 Bitter | 0 Bitter | 1 Bitter | 0 Bitter | 1 Bitter | 1 Bitter | 1 Bitter | **0 Bitter** | **1 Hot** |
| ID37 | 0 Sweet | 0 Sweet | 0 Sweet | 1 Sweet | 0 Sweet | 1 Bitter | 1 Bitter | 1 Bitter | 1 Bitter | 0 Bitter | **0 Bitter** | **1 Hot** |
| ID39 | 0 Sweet | 1 Sweet | 1 Sweet | 0 Sweet | 0 Sweet | 1 Bitter | 0 Bitter | 0 Bitter | 1 Bitter | 1 Bitter | 0 Bitter | 0 Bitter |
| ID42 | 0 Sweet | 1 Bitter | 0 Sweet | 0 Bitter | 1 Sweet | 1 Bitter | 1 Bitter | 1 Bitter | 1 Bitter | 1 Bitter | 1 Bitter | 0 Bitter |
| ID43 | 0 Sweet | 0 Bitter | 1 Bitter | 0 Bitter | 0 Sweet | 1 Sweet | 0 Bitter | 0 Bitter | 1 Bitter | 1 Bitter | **0 Bitter** | **1 Hot** |
| ID45 | 0 Sweet | 0 Bitter | 0 Bitter | 1 Bitter | 0 Sweet | 0 Sweet | 1 Bitter | 1 Bitter | 1 Bitter | **1 Bitter** | **1 Hot** | 0 Hot |
| ID46 | 0 Sweet | 0 Sweet | 0 Bitter | 1 Bitter | 0 Sweet | 1 Sweet | 1 Bitter | 1 Bitter | 1 Bitter | 0 Bitter | **0 Bitter** | **1** Hot |
| ID50 | 0 Sweet | 0 Sweet | 0 Sweet | 0 Bitter | 1 Bitter | 0 Sweet | 1 Sweet | 0 Sweet | 0 Bitter | 0 Bitter | 1 Bitter | 0 Bitter |
| ID52 | 0 Sweet | 1 Sweet | 0 Sweet | 0 Bitter | 0 Bitter | 0 Sweet | 0 Sweet | 0 Sweet | 1 Bitter | 0 Bitter | **0 Bitter** | **1 Hot** |
| ID53 | 0 Sweet | 0 Sweet | 1 Sweet | 0 Bitter | 1 Bitter | 1 Sweet | 1 Sweet | 1 Sweet | **0 Bitter** | **1 Hot** | **1 Hot** | **1 Hot** |
| ID55 | 1 Sweet | 0 Sweet | 0 Sweet | 0 Bitter | 1 Bitter | 0 Sweet | 1 Sweet | 0 Sweet | **0 Bitter** | **1 Hot** | **1 Hot** | **1 Hot** |
| ID57 | 0 Sweet | 0 Sweet | 1 Sweet | 0 Bitter | 1 Bitter | 1 Sweet | 1 Sweet | 1 Sweet | 1 Bitter | 1 Bitter | **0 Bitter** | **1 Hot** |
| ID61 | 0 Sweet | 1 Sweet | 0 Sweet | 0 Sweet | 1 Bitter | 0 Bitter | 0 Sweet | 1 Sweet | 1 Bitter | 0 Bitter | **0 Bitter** | **1 Hot** |
| ID64 | 1 Sweet | 0 Sweet | 0 Sweet | 1 Sweet | 0 Bitter | 0 Bitter | 1 Sweet | 1 Sweet | 0 Bitter | 1 Bitter | 1 Bitter | 0 Bitter |
| ID68 | 0 Sweet | 0 Sweet | 1 Sweet | 1 Bitter | 0 Bitter | 1 Sweet | 1 Sweet | 0 Sweet | **0 Bitter** | **1 Hot** | **1 Hot** | **1 Hot** |
| ID69 | 0 Sweet | 0 Sweet | 1 Sweet | 0 Bitter | 1 Bitter | 0 Sweet | 1 Sweet | 1 Sweet | **0 Bitter** | **1 Hot** | **1 Hot** | **1 Hot** |
| ID71 | 0 Sweet | 0 Sweet | 0 Sweet | 1 Sweet | 0 Bitter | 1 Bitter | 0 Sweet | 1 Sweet | 1 Bitter | 0 Bitter | 0 Bitter | 0 Bitter |
| ID73 | 0 Sweet | 1 Sweet | 1 Sweet | 0 Sweet | 0 Bitter | 0 Bitter | 0 Sweet | 1 Sweet | 1 Bitter | 0 Bitter | 0 Hot | 0 Hot |
| ID76 | 0 Sweet | 1 Sweet | 0 Sweet | 1 Bitter | 0 Bitter | 1 Sweet | 1 Sweet | 0 Sweet | **0 Bitter** | **1 Hot** | **1 Hot** | **1 Hot** |
| ID77 | 0 Sweet | 0 Sweet | 0 Sweet | 0 Bitter | 1 Bitter | 1 Sweet | 1 Sweet | 1 Sweet | **0 Bitter** | **1 Hot** | **1 Hot** | **1 Hot** |
| ID86 | 0 Sweet | 0 Sweet | 0 Bitter | 1 Bitter | 1 Bitter | 1 Sweet | 0 Sweet | 1 Sweet | 1 Bitter | **0 Bitter** | **1 Hot** | **1 Hot** |
| ID87 | 0 Sweet | 0 Sweet | 1 Sweet | 0 Sweet | 0 Bitter | 1 Bitter | 1 Sweet | 1 Sweet | 1 Bitter | **0 Bitter** | **1 Hot** | **1 Hot** |
| ID94 | 0 Sweet | 0 Sweet | 0 Bitter | 1 Bitter | 0 Bitter | 0 Bitter | 0 Sweet | 0 Sweet | 1 Bitter | **0 Bitter** | **1 Hot** | **1 Hot** |
| ID95 | 1 Sweet | 0 Sweet | 1 Sweet | 0 Bitter | 1 Bitter | 1 Sweet | 1 Sweet | 1 Sweet | **0 Bitter** | **1 Hot** | **1 Hot** | **1 Hot** |
| ID99 | 0 Sweet | 0 Sweet | 0 Bitter | 1 Bitter | 0 Bitter | 0 Bitter | 1 Sweet | 0 Sweet | 1 Bitter | **1 Bitter** | **1 Hot** | 0 Hot |
| ID106 | 1 Sweet | 0 Sweet | 1 Sweet | 0 Bitter | 0 Bitter | 0 Sweet | 1 Sweet | 1 Sweet | **0 Bitter** | **1 Hot** | **1 Hot** | **1 Hot** |
| ID109 | 1 Sweet | 0 Sweet | 1 Sweet | 1 Bitter | 0 Bitter | 1 Sweet | 1 Sweet | 0 Sweet | **0 Bitter** | **1 Hot** | **1 Hot** | **1 Hot** |
| ID111 | 0 Sweet | 0 Sweet | 0 Bitter | 0 Bitter | 1 Bitter | 0 Bitter | 1 Sweet | 0 Sweet | 1 Bitter | 0 Bitter | 1 Bitter | 0 Bitter |
| ID115 | 0 Sweet | 0 Sweet | 0 Sweet | 1 Bitter | 0 Bitter | 0 Sweet | 1 Sweet | 0 Sweet | **0 Bitter** | **1 Hot** | **1 Hot** | **1 Hot** |
| ID118 | 0 Sweet | 0 Sweet | 0 Sweet | 0 Bitter | 1 Bitter | 0 Sweet | 1 Sweet | 0 Sweet | **0 Bitter** | **1 Hot** | **1 Hot** | **1 Hot** |
| ID121 | 0 Sweet | 0 Sweet | 0 Bitter | 1 Bitter | 0 Bitter | 0 Bitter | 0 Sweet | 1 Sweet | 1 Bitter | 0 Bitter | **0 Bitter** | **1 Hot** |
| ID122 | 1 Sweet | 0 Sweet | 0 Bitter | 0 Bitter | 1 Bitter | 1 Bitter | 1 Sweet | 1 Sweet | 1 Bitter | **0 Bitter** | **1 Hot** | **1 Hot** |

**Figure 6** Individual BET of hotness from capsaicin samples among H users. *Data in bold red text are responses correctly identified as hotness, whereas the ones in bold blue text are the highest concentration missed. **Tester IDs highlighted in colours show inconsistency patterns of identifying hotness samples.

**Table 2** Average threshold based on geometric means

| | Geometric means[†] | | |
|---|---|---|---|
| | **Light chilli users ($n$ = 40)** | **Moderate chilli users ($n$ = 40)** | **Heavy chilli users ($n$ = 40)** |
| Pungent odour of dried chilli (g L) | 0.61 ± $0.04^{c}$ | 1.68 ± $0.04^{b}$ | 5.76 ± $0.40^{a}$ |
| Pungent odour of 1P3O (μl $L^{-1}$) | 0.04 ± $0.01^{c}$ | 0.20 ± $0.05^{b}$ | 1.27 ± $0.30^{a}$ |
| Hotness of dried chilli (g $L^{-1}$) | 0.58 ± $0.06^{c}$ | 2.16 ± $0.04^{b}$ | 7.07 ± $0.33^{a}$ |
| Hotness of capsaicin (mg $L^{-1}$) | 0.87 ± $0.53^{c}$ | 2.09 ± $0.43^{b}$ | 11.75 ± $0.28^{a}$ |

Different superscripts ($^{a–c}$) within a row show significant difference ($P \leq 0.05$).

[†]Geometric mean ± $\text{Log}_{10}$ standard deviation.

study was identified as 'train oil-like' (Zhai *et al.*, 2022). Genovese *et al.* (2018) included metallic and pungent odours as 1P3O odour descriptors found in olive oil. The metallic smell could also be caused by aluminium film used in wrapping the sample containers in this study. We feel that there is insufficient evidence to conclude an association between 1P3O and metallic odour perceived from dried chilli sniffing test.

A summary of predicted Group thresholds is presented in Table 3. It is worth highlighting that recognition thresholds of the experimental stimulus Capsaicin were found to be higher when compared with reported thresholds in other studies. For example, the hotness threshold of Thai Light user group is 0.96 ± 0.38 mg $L^{-1}$ capsaicin whereas European consumers were reported at 0.08 mg $L^{-1}$ capsaicin (Schneider *et al.*, 2014) and Japanese consumers at 0.70 mg $L^{-1}$

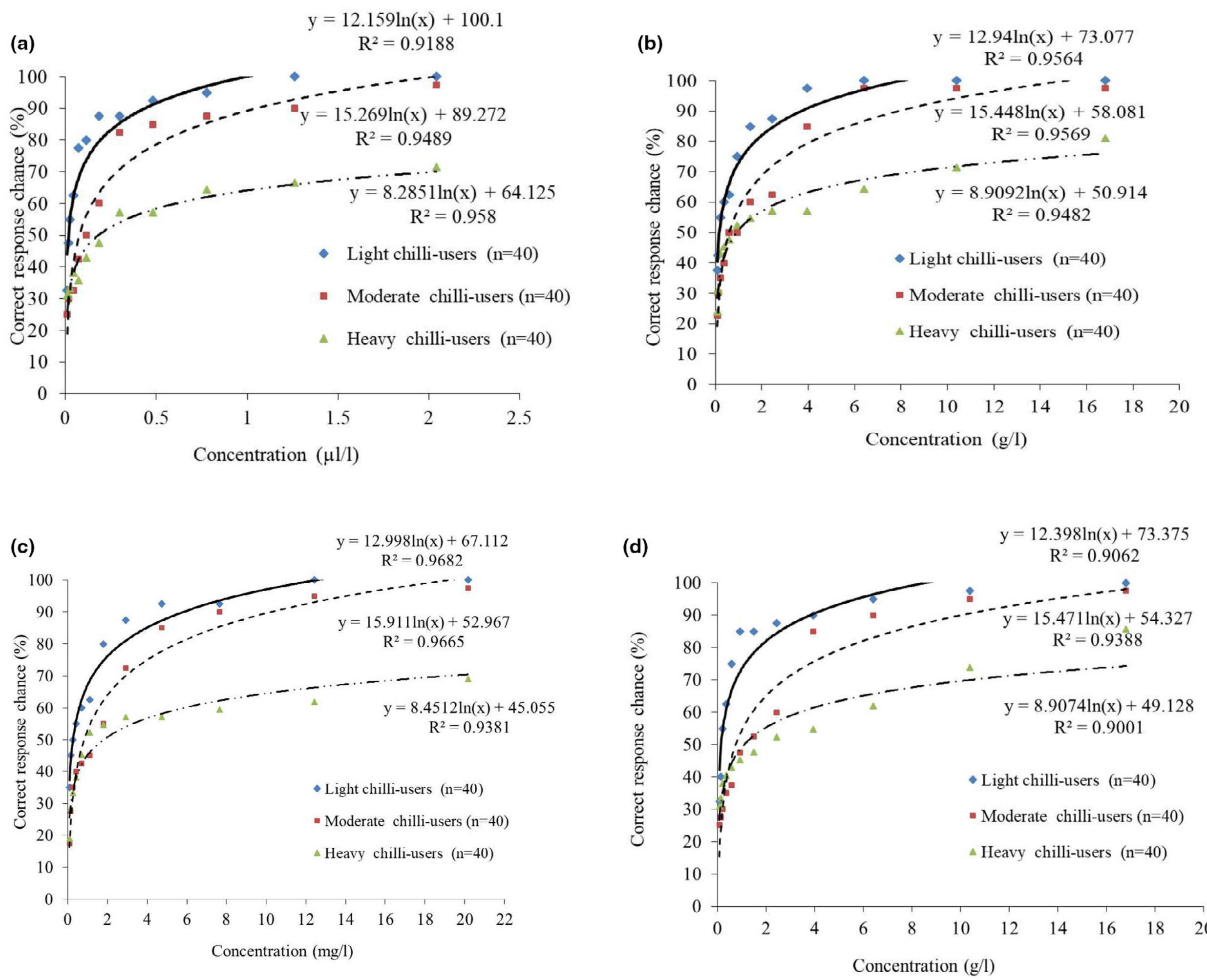


**Figure 7** Logistic regression on (a) 1P3O pungent odour, (b) dried chilli pungent odour, (c) capsaicin hotness, and (d) dried chilli hotness threshold values among three groups of chilli users.

 

capsaicin (Fukunaga *et al.*, 2005). Yet, those in the moderate group shared similar range of capsaicin threshold level (2.36 mg $L^{-1}$) with Turkish consumers (1.53 mg $L^{-1}$ capsaicin) (Mavi *et al.*, 2000). A recent study on threshold measurement among Chinese assessors reported their capsaicin threshold range as between 0.85 and 6.4 mg $L^{-1}$ (Zhang *et al.*, 2023) which is in the range of our recognition thresholds measured from Light and Moderate users. Orellana-Escobedo *et al.* (2012) conducted a large study of 250 Mexican subjects aged 18–60 years using 2-AFC method, and reported an absolute threshold of 0.05 mg $kg^{-1}$ total capsaicinoids. It was discussed that the low threshold from Mexican subjects in this study could be attributed to desensitisation. A comprehensive review of *Capsicum annum*'s potential by Hernández-Pérez *et al.* (2020) supports this notion, as the review suggested that 1 mg of capsaicin $kg^{-1}$ of sample would be classified as non-pungent. Han *et al.* (2022) determined oral hotness threshold at capsaicin concentration of 0.28 mg $L^{-1}$ (0.9 μM) as not detectable or only barely detectable. This study also raises for discussion about subject recruitment, as it involves naïve consumers who participated without prior knowledge of the study and who entered the threshold test with no training session, unlike other threshold tests where staff or students in academia participated. The review from Hernández-Pérez *et al.* (2020) also gives an overview of the chilli daily intake among frequent users and reported that Thais had third-highest chilli consumption after Mexicans and Saudi Arabians.

Based on the geometric means, most of the Average BET thresholds (Table 2) tended to be slightly lower than the predicted Group thresholds (Table 3). Light chilli users presented very close values between Average and Group thresholds regarding pungent odour and hotness of dried chilli. This very close proximity between the predicted and average thresholds in the Light chilli users could point to the fact that they were least exposed to chilli/capsaicin stimuli, and were also the most sensitive users, hence the variations of results within this group are not as high as the Heavy users. Having said that, the two calculated threshold values are not far off in the case of Medium and Heavy users either.

**Table 3** Group threshold from pooled data (logistic regression) among three groups of chilli users

| | Group threshold[†] | | |
|---|---|---|---|
| | Light chilli users (*n* = 40) | Moderate chilli users (*n* = 40) | Heavy chilli users (*n* = 40) |
| Pungent odour of dried chilli (g $L^{-1}$) | 0.61 ± 0.08[c] | 1.75 ± 0.18[b] | 5.88 ± 0.17[a] |
| Pungent odour of 1P3O (μl $L^{-1}$) | 0.06 ± 0.02[c] | 0.23 ± 0.09[b] | 1.34 ± 0.92[a] |
| Hotness of dried chilli (g $L^{-1}$) | 0.58 ± 0.09[c] | 2.23 ± 0.14[b] | 7.19 ± 0.20[a] |
| Hotness of capsaicin (mg $L^{-1}$) | 0.96 ± 0.38[c] | 2.36 ± 1.17[b] | 12.79 ± 1.78[a] |

Different superscripts (a–c) within a row show significant difference ($P \le 0.05$).

[†]Calculated using $P(x) = a \ln(x) + b$ from logistic regression when $P(x) = 0.667$.

## Conclusions

The aims of this study were to test (i) oral hotness threshold levels caused by a standard capsaicin compound compared to hotness perceived from capsaicin with other complex compounds in chilli powder samples, and (ii) nasal pungent odour thresholds caused by a standard compound 1P3O compared with complex compounds in dried chilli samples. The testers were classified into three groups, based on their chilli intake habit, their sensitivity to capsaicin solutions, and commitment to participate through the tests (with small amount of incentive), into High ($n$ = 40), Moderate ($n$ = 40), and Light users ($n$ = 40). The test results from oral hotness of capsaicin samples (0.10–-20.16 mg $L^{-1}$ (ppm) or 0.33–66.01 μM) show thresholds of L users at 0.96 ppm, M users at 2.36 ppm, and H users at 12.79 ppm. In comparison, hotness thresholds of dried chilli samples with capsaicin equivalent were 0.68 ppm for L users, 2.61 ppm for M users, and

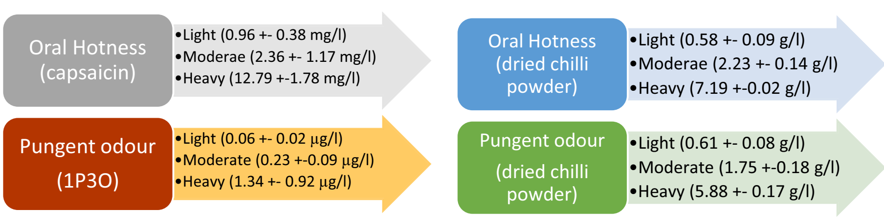


**Figure 8** A summary of hotness and pungent odour thresholds measured from L, M, and H users.

8.41 ppm for H users. The hotness thresholds from the two stimuli clearly indicate the impacts of substances in dried *C. annuum* L. var. *acuminatum* Fingerh other than capsaicin content which lower recognition hotness level (i.e. at lower capsaicin concentration). It is also noted that frequent and long-term spicy food consumption, as seen from H users, had reduced sensitivity to capsaicin in 2-AFC and 3-AFC methods (Orellana-Escobedo *et al.*, 2012; Su *et al.*, 2022).

The volatile 1P3O is a ketone compound with low volatility. It was hypothesised that the compound was a principal pungent odour found in the hot-air dried chilli from *C. annuum* Linn. Var. *acuminatum* Fingerh. used in this study. It has been confirmed with previous literature that 1P3O was perceived as pungent odour by human subjects. Our finding reports pungent recognition thresholds of 1P3O at 0.06, 0.23, and 1.34 μL - $L^{-1}$ in L, M and H users, respectively. Although the information on 1P3O quantification in the dried chilli is yet to be analysed and confirmed, the results suggest the potential for formulating food items for those who are not used to having chilli in their daily diet to have the 'flavour' of chilli through the use of pungent volatiles such as 1P3O.

A summary of the concentration of each stimulus which reaches recognition threshold of each user group (based on logistic regression model) is displayed in Fig. 8. We determined baselines of the dried chilli amount at the recognisable concentration of capsaicin compound that would affect recognisable sensation while not resulting in overbearing hotness through oral and nasal perceptions. This study also reports concentration guidelines for dried chilli usage in top-up spice mix and other ready-to-eat and ready-to-cook products. An example from Andersen *et al.* (2017) showed effective level of oral heat sensation which significantly increased appetite, satisfaction, and satiety of the test subjects. The added chilli in tomato soup was equivalent to capsaicin concentration of 1.13 mg $L^{-1}$. The use of chilli and its aroma for enhancing food flavours or creating new sensation in food and beverages at low concentration of capsaicin is also supported by Han *et al.* (2022), Lyu *et al.* (2021), and Su *et al.* (2022).

Limitations of this study must be acknowledged. First, participants in this study were initially screened and recruited for their chilli intake and sensitivity to capsaicin solutions. They were not tested for taste or smell sensitivity (i.e. diagnostic of anosmia or hypergeusia), which is especially relevant for those who were classified as high chilli users. Some of our H users could potentially have chronic effects from long-term, high amount of chilli intake on oral and/or nasal perception (Lyu *et al.*, 2021; Han *et al.*, 2022; Su *et al.*, 2022) as we observed inconsistent patterns of identifications in the hotness and pungent odour threshold tests discussed in Recruitment of chilli users.

Second, it is acknowledged that Thai chilli users are not representative of chilli users in other parts of the world. However, each of the H, M, and L groups consisted of more than thirty-two assessors (four panels of selected eight) as suggested for estimated number of 3-AFC assessors (BS ISO 13301, 2018). They also underwent preparation based on the BS ISO guidelines prior to commencement of the tests. The set of concentrations exceeded recommendation for the total testing set ($n$ = 192). We anticipate that by capturing three groups of users with varying levels of chilli use (rarely have chilli, sometimes add chilli, and usually have chilli in daily diet), we have provided insight that is useful in other settings as well. The results indicate that the lowest recognition thresholds from the Thai Light and Moderate chilli users were much higher than those of other countries belonging to similar age bracket (18–35 years), particularly compared with European and Japanese consumers (Fukunaga *et al.*, 2005; Schneider *et al.*, 2014; Han *et al.*, 2022) but rather close to Chinese consumers (Su *et al.*, 2022).

Third, the effects of capsaicin interacting with 1P3O on hotness perception in a food matrix were not measured in this study. We had insufficient information on volatiles in the dried chilli (*C. annuum* Linn. Var. *acuminatum* Fingerh) we were analysing. The chemical and sensory analyses of volatile compound 1P3O have rarely been reported, especially in dried cayenne chilli. Aromatic compounds of capsicum chilli and Sichuan peppers were reviewed and compared in Zhang *et al.* (2021) but the study focused on alcohols such as linalool as aroma found in fruits and pericarps, whereas Korkmaz *et al.* (2020) reported derivatives of 1P3O such as (E)-3-Penten-2-one (ketone compound), and 1-Penten-3-ol (alcohol compound) in dried capsicum chilli. 1P3O in this study's dried chilli was detected by Gas Chromatography Mass-Spectrometry (GC–MS) method using Liquid–Liquid Extraction (LLE) technique (details of the analysis methods are reported in Toontom *et al.*, 2016), whereas it was not detected by Solid Phrase Microextraction (SPME) technique (Toontom *et al.*, 2016; Korkmaz *et al.*, 2020). The volatile was also reported by analysis using LLE/APCI-MS in virgin olive oil (Genovese *et al.*, 2018). This suggests that sample extraction and concentration techniques such as LLE followed by GC–MS analysis would enhance detection sensitivity of 1P3O volatile compound. Our research finding confirms the effects of 1P3O on nasal pungency. We are also able to compare the oral hotness thresholds from standard capsaicin stimuli with the dried chilli quantity from HPLC analysis. We acknowledge that oral and nasal perception of hotness from dried chilli sample could be influenced by other capsaicinoids (dihydrocapsaicin, nordihydrocapsaicin, homocapsaicin, and homodihydrocapsaicin) and other pungent volatile compounds found in *C. annuum*

 

L. (cayenne chilli) (Casquete et al., 2021; Zamljen *et al*., 2022). Nevertheless, the results we report contribute additional information relevant to the understanding of cayenne chilli profile. We also acknowledge that if both substances had been tested in a food model, the food compositions such as NaCl, sucrose, amino acids, etc. could enhance or supress the hotness perception (Kosmidou *et al*., 2013; Schneider *et al*., 2014; Andersen *et al*., 2017).

As far as capsaicin and 1P3O are concerned, both substances generate irritating and discomfort effects to human senses. Design elements in threshold test such as serving plan, selected range of concentrations, sample preparation and presentation, measurement tools, as well as the choice of cleansing materials and resting time between 3-AFC triads, should be chosen to reduce discomfort and fatigue of human subjects in the study. Sucrose solutions have been used to reduce the irritating effects from capsaicin in sensory tests, Ramı́rez-Rivera *et al*. (2021) mixed sugar with habanero sample and water to reduce discomfort in their participants from testing chilli solutions. Whilst the use of 2% ethanol solution as control in the 3-AFC tests may not represent the actual scenario of chilli usage, capsaicin in the dried chilli powder can be extracted to the capsaicin concentrations we planned for the threshold tests in this study. This was not unusual as low concentration of ethanol was also used as solvent in recent hotness studies (Lyu *et al*., 2021; Su *et al*., 2022). We also consider that the 'heat profile' of five attributes as proposed by Guzmán & Bosland (2017) applies in relevant future work that study cayenne chilli was characterised in terms of 'rapid development, sharp rising, located in front to middle of the mouth, dissipates quickly, and hot intensity'.

The growing international market for new chilli-flavoured products provides a growth opportunity for food manufacturers. By analysing recognition thresholds of oral hotness and nasal pungency caused by the three stimuli, this study has particularly contributed to understanding how dried chilli powder can be utilised in the food items, and optimised for the new market. Further research examining how the dried chilli powder, capsaicin, and 1P3O interact within food models would be valuable.

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## Author contributions

**Nitchara Toontom:** Investigation; writing – original draft; methodology; project administration; resources; formal analysis; visualization; software; validation. **Mutita Meenune:** Conceptualization; funding acquisition; writing – original draft; project administration; supervision; validation; methodology. **Luis Kluwe Aguiar:** Conceptualization; writing – review and editing; resources; data curation; validation. **Wilatsana Posri:** Data curation; supervision; formal analysis; conceptualization; investigation; funding acquisition; methodology; writing – review and editing; software; resources; visualization; validation; project administration; writing – original draft.

## Ethical approval

The participants were informed of the test samples, procedures, and duration of the test upon the recruitment. The study received ethical approval from Prince of Songkla University (PSU) ethics committee (PSU No. 228/2551).

## Peer review

The peer review history for this article is available at https://www.webofscience.com/api/gateway/wos/peer-review/10.1111/ijfs.17177.

## Data availability statement

The data that support the findings of this study are available on request from the corresponding author.

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## Supporting Information

Additional Supporting Information may be found in the online version of this article:

**Appendix S1.** Supporting information.

**Table S1.** Physical and chemical qualities of Fresh (F), Hot-air Drying (HD) and Sun Drying sample (SD) samples.

**Table S2.** Capsaicin content of fresh and dried chilli samples.

**Table S3.** Volatile flavour compounds and their attributes identified in fresh sample and dried chilli samples.

**Figure S1.** HPLC chromatograms of standard of capsaicin (a); capsaicin in fresh chilli (b); capsaicin in hot-air dried chilli (c) and capsaicin in sun-dried chilli (d).