

Comparison of the sensory properties of fragrant and non-fragrant rice (Oryza sativa), focusing on the role of the popcorn-like aroma compound 2-acetyl-1-pyrroline

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1 Comparison of the sensory properties of fragrant and non-fragrant rice
2 (*Oryza sativa*), focusing on the role of the popcorn-like aroma
3 compound 2-acetyl-1-pyrroline

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14

15

16 **Abstract**

17 2-Acetyl-1-pyrroline (2-AP) has been widely reported as a key contributor to the popcorn-like
18 aroma of fragrant rice (*Oryza sativa*). To gain a greater understanding of its contribution to the
19 aroma in both fragrant and non-fragrant rice, sensory profiling was conducted with a trained
20 panel to examine the sensory properties of six boiled rice samples, three fragrant and three non-
21 fragrant varieties. The intensity of the popcorn note as an orthonasal odour, a retronasal flavour
22 and as an after-effect was significantly higher in fragrant rice than in non-fragrant rice.
23 However, panellists could not differentiate these popcorn attributes between the three different
24 fragrant rice varieties. 2-AP was extracted from the boiled rice samples by headspace solid-
25 phase microextraction and quantified by gas chromatography–mass spectrometry. 2-AP was
26 below the limits of quantitation in non-fragrant varieties; however, gas chromatography–
27 olfactometry of samples indicated the presence of 2-AP in both raw fragrant and non-fragrant
28 rice varieties.

29

30 **Highlights:**

- 31 • 2-Acetyl-1-pyrroline (2-AP) is a key discriminator of fragrant and non-fragrant rice.
- 32 • Trained panel could not separate fragrant rice varieties with 2-fold variation in 2-AP.
- 33 • Popcorn-like aroma and flavour of 2-AP could be perceived in non-fragrant rice.
- 34 • The odour perception of 2-acetyl-1-pyrroline fits Stevens' law.

35

36

37 **Keywords:** 2-acetyl-1-pyrroline; sensory evaluation; fragrant rice; GC–MS; HS-SPME; GC–
38 olfactometry

39 **1. Introduction**

40 Rice (*Oryza sativa*) provides energy for 25% of the world's population (FAO, 2002)
41 and in 2019–2020 contributed to 20.7% of worldwide total grain consumption (USDA, 2020).
42 It can be categorised into two types depending on its aroma: fragrant rice and non-fragrant rice.
43 According to the 2017 Rice Market Monitor report, non-fragrant long-grain and medium-grain
44 rice constitute the majority of world trade (79%). This report stated that the price of fragrant
45 rice was more than double that of high quality non-fragrant rice (FAO, 2017).

46 The aroma of fragrant rice was first evaluated analytically in the early 1980s and was
47 described as 'popcorn-like'. Perceived popcorn odour intensities in several fragrant rice
48 varieties were ranked, and 2-acetyl-1-pyrroline was considered as the most important
49 contributor to this odour (Buttery, Ling, Juliano, & Turnbaugh, 1983). This volatile compound
50 can contribute a popcorn-like aroma with a low detection threshold (0.02 ng/L in air; Schieberle,
51 1991). It was first identified in boiled fragrant rice (Buttery, Ling, & Juliano, 1982). This
52 compound is not only present in fragrant rice, it can also be detected in many different raw
53 food materials, such as hazelnuts, pandan leaf, and Manuka honey; in addition, 2-AP can also
54 be detected in some manufactured food products, such as popcorn, wheat bread crusts, and on
55 the surface of Mediterranean dried sausages, Parma ham and Italian-type salami, where it
56 contributes key odour characteristics (Wei, Handoko, Pather, Methven &, Elmore, 2017).

57 Sensory profiling, using techniques such as quantitative descriptive analysis (QDA), is
58 used to describe and quantify product attributes. Lexicons of rice descriptors have been
59 established in several studies, especially for fragrant rice (Goodwin et al., 1996; Piggott,
60 Morrison, & Clyne, 1991; Yau & Liu, 1999). The selection of descriptors depends on the
61 panellists' culture and their familiarity with the samples (Paule & Powers, 1989). Several
62 studies have indicated that the aroma contribution of 2-AP may be overemphasised in boiled

63 fragrant rice. Yang, Shewfelt, Lee, and Kays (2008) reported that popcorn-like note might not
64 be the only important attribute in boiled fragrant rice. In addition, Limpawattana, Yang, Kays,
65 and Shewfelt (2008) reported that there was no correlation between popcorn flavour and 2-AP
66 in boiled rice.

67 In this study, different boiled rice varieties were evaluated using quantitative
68 descriptive analysis (QDA). A lexicon was developed for both boiled fragrant and non-fragrant
69 rice varieties using a UK-based panel. This is the first time that a rice lexicon prepared by a
70 UK-based sensory panel has been reported. Differences in flavour and odour between fragrant
71 and non-fragrant rice were evaluated. In addition, 2-AP in boiled fragrant and non-fragrant rice
72 varieties was quantified using headspace solid-phase microextraction (HS-SPME) and gas
73 chromatography–mass spectrometry (GC–MS). The primary aim of this study was to determine
74 the strength of the relationship between perceived popcorn-like flavour and 2-AP content in
75 boiled fragrant and non-fragrant rice.

76 **2. Materials and Methods**

77 *2.1. Plant materials and chemicals*

78 Six varieties of milled (white) rice were obtained, including three fragrant rice varieties
79 (basmati and Thai jasmine from ASDA supermarket (Reading, UK), Sintanur from Indonesian
80 Centre for Rice Research) and three non-fragrant rice varieties (American long-grain from
81 ASDA supermarket (Reading), Arirang from Korea Foods Company Limited (Reading), and
82 Ciherang from Indonesian Centre for Rice Research). Only one batch of each rice variety was
83 collected for both GC–MS and QDA analysis, in order to limit the variation between batches.
84 Still mineral water (Harrogate Spring Water, Harrogate, UK) was used for sensory analysis and
85 HPLC-grade water (Fisher, Loughborough, UK) was used for chemical analysis. 2-AP and

86 partially deuterated 2-AP (2-AP- d_{2-5}) standards were used for 2-AP quantification (both 30,000
87 mg/kg in dichloromethane (DCM); aromaLAB GmbH, Planegg, Germany).

88 2.2. Quantitative descriptive analysis (QDA) in boiled rice

89 All rice samples (200 ± 1 g) were weighed and then boiled using 300 mL mineral water
90 in a rice cooker (0.8 L capacity; Lloytron PLC, Leigh, UK). To avoid cross contamination,
91 especially 2-AP in fragrant rice contaminating non-fragrant rice, each rice variety was cooked
92 in its own dedicated cooker. Rice samples were initially cooked with tap water. During
93 vocabulary development, panellists provided 'tap water' or 'kettle-like' attributes from
94 samples cooked in tap water. However, these attributes were absent in samples cooked in
95 mineral water. Subsequently, Harrogate Spring mineral water was used for rice boiling.
96 Cooking proceeded for 20 min before the rice cooker automatically turned to warm mode. The
97 samples were kept warm (>65 °C) in the rice cooker for 20 min before serving to panellists for
98 evaluation.

99 Sensory profiling using a quantitative descriptive analysis (QDA) approach was
100 conducted for six rice samples, using 11 trained, UK-based panellists, 10 female and one male.
101 The panellists had between 6 months and 10 years' experience of sensory analysis, were aged
102 between 30 and 60, and all screened and monitored for their sensory acuity. Seven QDA
103 sessions were conducted during the experiment: two sessions for vocabulary development, two
104 for training and three for scoring the samples. One batch of each type of rice was prepared for
105 each session.

106 A consensus vocabulary was developed for appearance, odour, taste, flavour, mouthfeel,
107 and after-effects. After-effects included all attributes within the modalities of taste, flavour and
108 mouthfeel that remained after samples were swallowed (ASTM International, 2009). Attribute
109 definitions and references are given in **Table 1**. A pre-heated (120 °C for 20 min in the oven)

110 ceramic cup (50 mL) filled with boiled rice (20 g) covered by foil was served to panellists for
111 developing odour attributes and another 20-g sample was then served in the same manner for
112 developing all the other attributes. The scoring for each sample attribute was conducted in
113 individual booths in duplicate on separate days; samples were labelled with three-digit codes
114 and presented randomly in a balanced order. Data were collected using Compusense at-hand
115 software (Compusense, Guelph, Canada) using unstructured line scales (0–100), except for the
116 attribute “popcorn odour”, where a structured scale was used with anchors at positions defined
117 by the panel after sniffing various concentrations of the reference 2-AP.

118 References for ‘porridge’, ‘rice pudding’, ‘milky’ and ‘starchy water’ attributes were
119 provided (**Table 1**). The panellists were trained in recognition and scaling of popcorn odour,
120 using a series of dilutions of 2-AP standard. Five sniff strips (Sigma-Aldrich, St Louis, MO)
121 were wetted with four concentrations of 2-AP in dichloromethane (10, 100, 1000 and 5000
122 µg/kg) and a blank dichloromethane solution. After all solvent was evaporated using a nitrogen
123 stream, each strip was sealed in a 5-mL glass vial with screw lid. Each vial was only opened
124 once and sniffed by one panellist. The blank and the standard with highest 2-AP concentration
125 were first provided to each panellist for Nil and Extreme values on the 0–100 unstructured line
126 scale. The panellists were then was asked to sniff and score the other three concentrations of
127 2-AP on the same line scale. The average score for each 2-AP reference was added onto all 0–
128 100 line scales used to measure ‘popcorn’ odour in the rice sample scoring session. Five
129 concentrations of 2-AP standard (blank included) were also provided to panellists before
130 sample profiling (an individual set of standards was prepared for each panellist). Panellists
131 were asked to sniff the 2-AP standards in a separate room prior to the profiling session.

132 *2.3. 2-Acetyl-1-pyrroline quantification in boiled rice using solid-phase microextraction and*
133 *gas chromatography-mass spectrometry*

134 Rice (1.000 g \pm 0.001 g) and 1.5 mL HPLC-grade water were added to 20-mL SPME
135 glass vials. Vials were sealed with metal screw caps possessing PTFE-faced silicone septa. The
136 vials were heated in the oven of a Hewlett Packard 5890 gas chromatograph at 100 °C for 20
137 min and then cooled to room temperature. Finally, a 1.5-mL aliquot of 2-AP-*d*₂₋₅ aqueous
138 solution was added into the vials. The 2-AP-*d*₂₋₅ aqueous solution was prepared from 2-AP-*d*₂₋
139 ₅ in dichloromethane (100 μ g/kg); dichloromethane was evaporated by N₂ gas and replaced by
140 an equal amount of HPLC-grade water. During the dichloromethane evaporation, a proportion
141 of the 2-AP-*d*₂₋₅ could be lost due to the instability of this compound. Therefore, the 2-AP-*d*₂₋
142 ₅ aqueous solution was only prepared once in the whole experiment; it was used for all samples
143 and calibration standards, in order to avoid variation during aqueous solution preparation.

144 Headspace solid-phase microextraction (HS-SPME) followed by gas chromatography–
145 mass spectrometry (GC–MS) has been widely used in the aroma compound analysis of rice,
146 especially for 2-AP detection (Tulyathan, Srisupattarawanich, & Suwanagul, 2008; Bryant &
147 McClung, 2011; Mathure Jawali, Thengane, & Nadaf, 2014; Poonlaphdecha et al., 2016).
148 Believing that a higher extraction temperature can improve release of volatile compounds from
149 the food matrix, several studies have extracted 2-AP from rice using a high extraction
150 temperature (80 °C to 120 °C) (Grimm, Bergman, Delgado, & Bryant, 2001; Bryant &
151 McClung, 2011; Mathure et al., 2014; Poonlaphdecha et al., 2016). However, Hopper et al.
152 (2016) suggested the use of a lower extraction temperature; they indicated that 2-AP may be
153 generated at a high extraction temperature. Hence, to minimise 2-AP changes during extraction,
154 the HS-SPME method used in this paper was modified from that of Hopper et al. (2016). During
155 method development, a series of extraction times (30 min, 45 min, 60 min and 75 min) was
156 examined, in order to select a time that provided the highest signal-to-noise ratio of 2-AP in
157 the GC chromatogram; this occurred at 60 min, with no further increase at 75 min. Therefore,
158 60 min was subsequently used as the extraction time.

159 2-AP in boiled rice was extracted by an HS-SPME autosampler (GC Sampler 120;
160 Agilent, Santa Clara, CA), attached to a 6890 gas chromatograph with 5975 mass spectrometer
161 (Agilent). Each rice sample was incubated with agitation for 10 min at 40 °C, and then extracted
162 with a 1-cm divinylbenzene/CarboxenTM/polydimethylsiloxane (DVB/CAR/PDMS) SPME
163 fibre (Supelco, Bellefonte, PA) for 60 min at 40 °C with agitation.

164 After extraction, the SPME fibre was desorbed in the GC injection port at 250 °C for
165 20 min, in splitless mode, onto the front of a Zebron ZB-Wax column (30 m × 0.25 mm; 1 µm
166 film thickness; Phenomenex, Torrance, CA). The carrier gas was helium at a constant column
167 flow rate of 0.9 mL/min. The initial GC oven temperature was 40 °C held for 2 min, then
168 increased to 60 °C at the rate of 2 °C/min, at which point the rate was increased to 6 °C/min
169 and held for 35 min after the oven temperature reached 250 °C. Electron ionisation (EI) was
170 applied; ionisation energy was 70 eV, and the electron multiplier was set at 2824 V. Full scan
171 mode was used for analysis from m/z 30 to 280. Selected ion monitoring was also applied
172 (SIM/Scan mode); m/z 68, m/z 83 and m/z 111 were monitored for 2-AP; m/z 86 and m/z 114
173 were monitored for 2-AP- d_{2-5} . Dwell time of monitored ions was set at 100 ms/ion. A blank
174 sample was prepared from 1.5 mL 2-AP- d_{2-5} aqueous solution (100 µg/kg) with no rice and no
175 2-AP standard present in a 20-mL SPME vial, and it was run by GC-MS before calibration
176 standards and rice samples. Mass spectral fragments at m/z 68, m/z 83 and m/z 111 were absent
177 in 2-AP- d_{2-5} , which suggested that 2-AP- d_{2-5} is an ideal internal standard for 2-AP
178 quantification and m/z 86 and m/z 114 can be used to monitor 2-AP- d_{2-5} .

179 A matrix-matched calibration curve was established for accurate quantification of 2-
180 AP. Boiled American long-grain rice (non-fragrant rice) was used as the matrix for calibration
181 curves. Although a response for 2-AP in chromatograms was detected in all six rice samples
182 (trace levels of 2-AP were present in chromatograms of non-fragrant rice), American long-
183 grain rice gave the lowest response for 2-AP among all of the rice samples studied. A prepared

184 2-AP standard solution (5.5 mg/kg in dichloromethane) was used for this curve. American
185 long-grain rice (1 g) with 1.5 mL HPLC grade water was boiled in a 20-mL glass SPME vial
186 with lid in a GC oven at 100 °C for 20 min and then the vial was cooled to room temperature.
187 Four calibration standards (10 µg/kg, 50 µg/kg, 100 µg/kg, and 200 µg/kg) were prepared to
188 create a calibration curve for 2-AP. For each calibration standard, 100 µL 2-AP in DCM (0.1
189 mg/kg, 0.5 mg/kg, 1 mg/kg, 2 mg/kg) with 1.5 mL 2-AP-*d*₂₋₅ aqueous solution (the same
190 concentration as in the extracted rice samples) were then added into the boiled American long-
191 grain rice matrix and analysed by HS-SPME and GC-MS. The calibration curve formula
192 obtained from calibration standards was

$$193 \quad y = 0.0118x$$

194 where, *y* is (peak area of 2-AP)/(peak area of 2-AP-*d*₂₋₅) and *x* is the concentration of
195 2-AP. The *r*² value of the calibration curve was 0.9856; recoveries of calibration standards
196 containing 10 µg/kg, 50 µg/kg, 100 µg/kg, and 200 µg/kg of 2-AP were 175%, 108%, 76% and
197 104%, respectively. Therefore, when measuring 2-AP at a range between 50 µg/kg and 200
198 µg/kg, the calibration curve was acceptable.

199 *2.4. Gas chromatography–olfactometry of raw fragrant and non-fragrant rice extracts* 200 *prepared using solid-phase extraction*

201 Raw, milled Sintanur or Ciherang rice flour (10 g ± 0.01 g) was placed into a 50-mL
202 centrifuge tube and 35 mL HPLC-grade water were added. The tube was shaken for 20 min at
203 1700 rpm (Multi Reax; Heidolph, Schwabach, Germany), and then it was centrifuged at 7000
204 rpm (≈ 5100 *g*) and 15 °C for 15 min (Sigma 3K10 laboratory centrifuge; Sigma, Osterode,
205 Germany). A 20-mL aliquot of the supernatant was collected for solid-phase extraction (SPE).
206 The Isolute ENV+ cartridge (200 mg/6 mL; Biotage, Uppsala, Sweden) was firstly conditioned
207 with 10 mL methanol, then with 10 mL HPLC-grade water. Then 20 mL rice supernatant were

208 loaded onto the cartridge. After sample loading, the cartridge was washed with 10 mL HPLC-
209 grade water. The washed cartridge was dried under vacuum for 30 min. Finally, compounds
210 were eluted with 2 mL DCM. The DCM extract was then concentrated with a nitrogen stream
211 to around 100 μ L. This concentrated extract was transferred to a 200- μ L glass insert (Thermo
212 Scientific, Loughborough, UK) and then it was sealed in a 2-mL autosampler vial with metal
213 crimp-cap prior to gas chromatography–olfactometry (GC–O) analysis.

214 A Zebron ZB-Wax column (30 m \times 0.25 mm; 0.25 μ m film thickness; Phenomenex,
215 Torrance, CA) was used in this analysis. One microlitre of the extract was injected manually
216 in split mode (split ratio of 20:1) into the injection port of a Hewlett Packard 5890 Series II gas
217 chromatograph with olfactometer and flame ionisation detector (FID). The inlet temperature
218 was 250 $^{\circ}$ C and the carrier gas was helium at 6.2 psi constant pressure. The initial GC oven
219 temperature was 40 $^{\circ}$ C held for 2 min, then increased to 200 $^{\circ}$ C at the rate of 4 $^{\circ}$ C/min, at which
220 point the rate was increased to 15 $^{\circ}$ C/min and held for 15 min after the oven temperature
221 reached 250 $^{\circ}$ C. The eluting compounds were split between the FID and sniff port with a split
222 ratio of 1:1. Four trained sniffers were asked to sniff both Sintanur and Ciherang extracts in
223 duplicate. A timer was started at the beginning of sample injection. The sniffers were asked to
224 describe the odour they perceived, record the time point when they perceived the odour and
225 rate the intensity of the odour from 0 (nil) to 10 (extreme). An alkane standard (C5–C22) was
226 used to calculate linear retention index (LRI) values.

227 2.5. *Statistical analysis*

228 Sensory profiling data were collected by Compusense at-hand (version 8.8, Guelph,
229 Canada) and analysed using Senpaq (v4.2, 2008; Qi Statistics, Reading, UK). Two-way
230 ANOVA was used with sample fitted as a fixed effect and panellists as a random effect; effects
231 were tested against the sample by panellist interaction. Significant differences between samples

232 were assessed by Fisher's LSD pairwise comparison, and significance level was set at $p \leq 0.05$.
233 To compare fragrant and non-fragrant rice samples as two groups, Student's *t*-test was carried
234 out using XLSTAT software (2012, Addinsoft, Paris, France).

235 **3. Results and Discussion**

236 *3.1. Quantitative descriptive analysis (QDA) of boiled rice*

237 *3.1.1. 2-Acetyl-1-pyrroline reference standard training*

238 Panellists from different cultures and with different experiences can have use different
239 words to describe sensory attributes. Paule and Powers (1989) reported that descriptions of
240 fragrant rice aroma by different groups were different. Orientals or frequent rice consumers
241 described the predominant fragrant rice aroma as 'pandan-like'; however, non-Orientals or
242 infrequent rice consumers described it as popcorn-like. The fragrant rice aroma in this study
243 was initially described as 'popcorn-like', 'basmati-like' or 'jasmine rice-like' by 11 trained UK
244 based panellists. 'Popcorn-like' is the major descriptor for this aroma. Buttery et al (1982)
245 firstly described the aroma as 'popcorn-like' in fragrant rice and reported that it was contributed
246 by 2-acetyl-1-pyrroline. The popcorn-like aroma in boiled rice was described as 'a dry, dusty,
247 slightly toasted and slightly sweet aroma that can be specifically identified as popcorn' in the
248 lexicon developed by Kansas State Expert Sensory Panel (Goodwin et al., 1996).
249 Mahattanatawee and Rouseff (2014) described the fragrance in basmati, jasmine and Jasmati
250 varieties as 'cooked jasmine rice-like' using GC-O analysis. In the present study the
251 description of this aroma was finally unified to 'popcorn-like' with the unanimous consent of
252 all panellists. 2-AP standard was provided to panellists, to compare it with the fragrant odour
253 in boiled rice samples.

254 Panellists ($n = 11$) were asked to sniff five different concentrations of 2-AP standards
255 (blank (0), 10, 100, 1,000 and 5,000 $\mu\text{g}/\text{kg}$) and to score perceived intensity on an unstructured
256 line scale (0–100) for popcorn aroma training, as described in *Section 2.2*. A ranking test for
257 these five standards was conducted before training to ensure that all the panellists could
258 differentiate and rank 2-AP standards without difficulty. This ranking test suggested that 5- to
259 10-fold differences in 2-AP standards could be detected by a trained UK panel. The blank
260 standard was subsequently labelled as Nil and the 5,000 $\mu\text{g}/\text{kg}$ standard was labelled as
261 Extreme; these two standards were scored as 0 and 100 on the unstructured line scale. The other
262 three standards (10, 100, 1000 $\mu\text{g}/\text{kg}$) were labelled as ‘1’, ‘2’, ‘3’ from low to high
263 concentration and panellists ($n = 11$) were asked to sniff and rate these three references using
264 the line scale relative to the Nil and Extreme references. Results are shown in **Figure 1a**. Mean
265 scores were then used as anchors at 12, 40 and 75 on the 0–100 line scales for popcorn odour
266 in the subsequent sample rating tests.

267 According to Stevens’ law: “equal stimulus ratios result in equal sensation ratios rather
268 than equal sensation differences” and his psychophysical power law was proposed as

$$269 \quad R = kS^n$$

270 Therefore

$$271 \quad \log R = n \log S + \log k$$

272 where R is the response, k is a constant, S is the stimulus concentration, and n is the
273 modality-dependent exponent (Stone, Bleibaum, & Thomas, 2012). The log–log plot between
274 2-AP concentration and perceived popcorn odour intensity follows Stevens’ law and is shown
275 in **Figure 1b**; exponent n is 0.338, denoting a decelerating relationship, as expected for aroma
276 perception. This result indicates that with increasing 2-AP concentration, the perceived
277 popcorn-like odour intensity increases but to a less than proportional extent. Therefore, it may

278 be more difficult for panellists to notice changes of 2-AP concentration at higher concentrations
279 than at lower concentrations.

280 *3.1.2. Boiled rice sensory attributes*

281 Thirty-seven attributes (covering appearance, mouthfeel, odour, taste, flavour, and
282 after-effects) were quantified in the six boiled rice samples; however, significant differences
283 between samples were only found in 8 attributes (**Table 2**). In physical modalities (appearance
284 and mouthfeel), significant differences between samples were found for cohesive mouthfeel (p
285 < 0.0001) and appearance attributes ($p < 0.0001$). The highest number of brown lines was
286 observed in American long-grain and Ciherang rice, and the lowest number of brown lines was
287 found in jasmine rice. Brown lines could not be observed on raw rice; they only appeared after
288 rice boiling and they were only found on the surface of the rice grain. Brown lines were not
289 present in every rice grain and this attribute was evaluated by how many grains with brown
290 lines could be observed in one sample portion (50 g). The rice manufacturers suggested that
291 the brown lines may be due to crack formation during rice postharvest processing or storage,
292 where perhaps incomplete drying or long-term storage may cause more brown lines to develop.
293 However, to our knowledge, this has not been reported in the literature.

294 After boiling, Arirang rice had the shortest rice grain and basmati rice had the longest
295 rice grain, while basmati rice also gave the thinnest grains. The physical attributes in boiled
296 rice, especially moisture content, stickiness and hardness are influenced by rice grain length
297 and their starch content. Arirang rice had the highest 'wet' score and basmati had the lowest.
298 Visible moisture differences may be caused by different water absorption abilities of the
299 different rice varieties. Water absorption of rice grain is dependent on surface area, amylose
300 and protein contents and gelatinisation temperature. Generally, long-grain varieties tend to
301 absorb more water than short-grain varieties (Bett-Garber, Champagne, Ingram, & McClung,

302 2007). Therefore, as the same amount of water was added to all samples for boiling in this
303 study, the shorter grain rice varieties (Arirang, Ciherang and Sintanur) appeared wetter than
304 the three longer grain varieties.

305 The ratio of amylose to amylopectin in rice grain can significantly influence stickiness
306 and hardness of boiled rice. Long-grain rice types (*indica*) usually contain more amylose and
307 less amylopectin and can be harder and less sticky. In contrast, short-grain rice types (*japonica*)
308 contain more amylopectin and less amylose; they are softer and stickier (Bao & Bergman,
309 2004). The stickiness of boiled rice is caused by leached amylose and amylopectin interacting
310 with each other, gelatinising and forming a coating on the surface of the grains (Bett-Garber et
311 al., 2007). The differences in starch composition in the different rice varieties were expressed
312 in their sensory attributes; high stickiness was expressed as lower grain separation appearance
313 and higher cohesive mouthfeel scores. Effort to chew reflected the hardness of boiled rice grain.
314 **Table 2** showed that basmati had highest grain separation and lowest cohesive mouthfeel.
315 Arirang rice had the highest score for cohesive mouthfeel. However, no significant difference
316 was found in this attribute between the six boiled rice varieties.

317 Of the 18 odour, taste and flavour attributes used to describe the boiled rice samples,
318 only popcorn odour differed significantly between the samples ($p = 0.028$). Only 2 samples
319 differed significantly for popcorn odour: the fragrant jasmine was significantly and
320 substantially higher in popcorn odour than the non-fragrant Ciherang (difference of 24 in odour
321 intensity rating score, $p = 0.002$ in multiple pairwise comparison post ANOVA; Tukey HSD).
322 The difference in popcorn flavour in mouth was not significant ($p = 0.13$), although the trend
323 was the same (jasmine highest and Ciherang lowest) with a mean difference of 12 in flavour
324 intensity rating score. Where popcorn was rated as an after-effect (flavour post-swallowing),
325 the trend ($p = 0.057$) was for the fragrant Sintanur and jasmine varieties to be rated higher than
326 the Ciherang.

327 When the six rice varieties were grouped into fragrant rice (jasmine, basmati and
328 Sintanur) and non-fragrant rice (American long-grain, Arirang and Ciherang), *t*-test results of
329 all of the odour and flavour-related attributes showed significant differences between fragrant
330 and non-fragrant rice types in popcorn odour ($p = 0.016$), popcorn flavour ($p = 0.026$) and
331 popcorn after-effect ($p = 0.019$), as shown in **Figure 2**. However, no differences were observed
332 in the other rice and cereal-related odour and flavour attributes. Yang et al. (2008) reported that
333 the popcorn-like note may not be the only important characteristic in boiled rice and other key
334 characteristics contributed by other volatile compounds could be found in boiled fragrant rice.
335 The results of this study concur with Yang et al., in that there were other aroma and flavour
336 attributes present in boiled rice. However, none of these additional odours or flavours (**Figure**
337 **2**) differentiated the fragrant and non-fragrant rice types.

338 As discussed earlier, the differences in popcorn attributes between all different rice
339 varieties were not obvious (Table 2). The significant difference in perceived popcorn odour
340 was driven by jasmine and Ciherang. However, panellists found it difficult to differentiate
341 popcorn odour in the other four boiled rice samples (basmati, Sintanur, American long-grain
342 and Arirang). Although jasmine and Sintanur tended to show higher perceived popcorn flavour
343 and after-effect than other samples, any differences between rice varieties were not significant
344 (Table 2). These results indicate that although the panellists could not differentiate individual
345 boiled rice varieties based on popcorn odour, flavour, or after-effect; fragrant and non-fragrant
346 rice samples could be distinguished as two separate groups based on all three of these
347 modalities.

348 Where the difference in popcorn odour between varieties was significant and any
349 differences between in-mouth popcorn flavour and popcorn as an aftertaste were not, this may
350 have been due to the use of the four reference anchors (2-AP standards) for training the
351 assessors. This may have helped panellists to improve their discrimination of different boiled

352 rice samples based on popcorn odour. However, the 2-AP standard training would have less
353 effect in improving the discrimination of popcorn retronasal flavour and after-effect because
354 the standards can only be sniffed; no standard levels of popcorn retronasal flavour and after-
355 effect were provided to panellists. The lack of flavour and aftertaste standards may have
356 resulted in higher variation between panellists in popcorn flavour and after-effect than in odour,
357 and hence resulted in a reduced likelihood of discrimination.

358 Popcorn was used as a reference material for ‘popcorn’ attributes in previous studies
359 (Limpawattana et al., 2008; Limpawattana & Shewfelt, 2010); it could have been used in the
360 training of ‘popcorn’ odour, flavour, and after-effect. However, other aromas present in
361 popcorn, such as ‘smoky’, may influence the understanding of ‘popcorn-like’ for panellists.
362 Schieberle (1991) suggested that not only ‘popcorn-like’, but also ‘fatty’, ‘coffee-like’ and
363 ‘spicy’ play important roles in the aroma of popcorn. In addition, intensities of ‘popcorn’
364 attributes cannot be controlled and adjusted in popcorn product during training.

365 *3.2. Quantification of 2-acetyl-1-pyrroline in boiled rice*

366 A matrix-matched calibration curve was established for 2-AP quantification in this
367 study. Rice itself should be the best matrix to build this curve, because the structure of food
368 including the starch content will significantly affect volatile compounds release from food
369 matrix. Increasing viscosity or gelatinisation of a food matrix can significantly decrease mass
370 transfer and therefore influence flavour release (Silva, Castro, & Delgadillo, 2002). It was
371 reported that release of aroma compounds is influenced by the amylose fraction in a
372 gelatinisation matrix; in contrast amylopectin is unlikely to form strong inclusion complexes
373 with aroma compounds (Silva et al., 2002).

374 It was discussed in *Section 3.3.2* that the starch composition was reflected in grain
375 separation, cohesiveness, and effort to chew. In “grain separation” attribute, basmati rice has a

376 significantly higher score than the other rice varieties; in “cohesiveness” attribute, Arirang rice
377 has a significantly higher score than American long-grain and basmati, while basmati has a
378 significantly lower score than jasmine, Sintanur and Arirang rice. In “effort to chew” attribute,
379 significant differences were not found among rice varieties (**Table 2**). More grain separation
380 and less cohesiveness are associated with harder texture, which is caused by a higher content
381 of amylose and a lower content of amylopectin; the converse is also true. Therefore, based on
382 textural properties, basmati should be the rice variety containing the most amylose and Arirang
383 the rice variety containing the least amylose, with intermediate values for the other four rice
384 varieties. Hence, the matrix for the calibration curve should be selected from jasmine, Sintanur,
385 Ciherang or American long-grain rice. 2-AP response was detected in the selected ion
386 chromatograms (m/z 68, m/z 83, and m/z 111) of all six boiled rice varieties, three non-fragrant
387 rice gave trace responses (lower than limit of quantification), and boiled American long-grain
388 rice gave the lowest response. Therefore, boiled American long-grain rice was arguably the
389 best choice as a matrix material for 2-AP calibration in this study.

390 Concentrations of 2-AP in the six boiled rice samples are shown in **Figure 3**. Significant
391 differences in 2-AP concentrations were found between the three boiled fragrant rice samples
392 ($p = 0.028$); jasmine rice contained most 2-AP (146 $\mu\text{g}/\text{kg}$), while the lowest 2-AP
393 concentration in a boiled fragrant rice was in Sintanur (80 $\mu\text{g}/\text{kg}$). As the most popular fragrant
394 rice on the UK market, basmati contained 113 $\mu\text{g}/\text{kg}$ of 2-AP, which would explain why it was
395 ranked in the middle of the three boiled fragrant rice varieties in this study for perceived
396 intensity of popcorn odour, even though the difference in intensities between the three was not
397 significant. Although a significant difference was found between the three fragrant rice samples
398 in 2-AP concentration, there was only a two-fold difference between jasmine and Sintanur rice.
399 The concentrations of the 2-AP standards used for popcorn odour training varied by 5 or 10
400 folds. Popcorn odour intensity of blank (0), 10, 100, 1000 and 5000 $\mu\text{g}/\text{kg}$ 2-AP reference

standards were ranked, and results showed that the trained panellists could differentiate and rank these samples in order of intensity with no difficulty. There was no evidence to show that a two-fold difference in 2-AP was great enough to be noticed by panellists, which might explain why there was no significant difference in popcorn odour, flavour or after-effect between the three fragrant rice samples. In addition, according to a log–log plot of 2-AP concentration and perceived popcorn odour intensity (**Figure 1b**), a decelerating relationship between 2-AP odour perception and 2-AP concentration may cause relatively more difficulty for panellists in discriminating higher 2-AP concentration samples and less difficulty in discriminating lower 2-AP concentration samples.

Limpawattana et al. (2008) reported that although 2-AP was the only contributor to popcorn-like note in boiled rice, there was no correlation between 2-AP concentration and perceived intensity of popcorn flavour. Their data showed that popcorn flavour had negative correlations with guaiacol and (*E,E*)-2,4-decadienal, which contributed smoky and fatty notes, respectively. They also reported that guaiacol was present in the popcorn they used for popcorn odour training, which might have affected the understanding of popcorn flavour. Guaiacol was not identified in the SPME extracts of any of the boiled rice samples in our study.

Yang et al. (2008) analysed 25 different odour-active compounds in five boiled fragrant rice samples and one boiled non-fragrant rice sample. They found that popcorn-like odour could be detected in both fragrant and non-fragrant rice varieties, and 2-AP was the only compound to contribute to this odour. Another study also evaluated 25 aroma-active compounds in fragrant and non-fragrant long-grain and medium-grain Italian rice. Again, 2-AP was the only compound contributing popcorn-like odour (Griglione et al., 2015).

Compounds other than 2-AP may contribute roasty or popcorn-like aroma in popcorn, such as 2-acetyltetrahydropyridine and 2-propionyl-1-pyrroline (Schieberle, 1991). 2-Acetyl-

425 2-thiazoline was reported to contribute to popcorn-like odour in boiled American-grown
426 jasmine-style long-grain rice (Mahattanatawee & Rouseff, 2014). This compound has a similar
427 aroma to 2-AP and is much more stable than 2-AP (Rey, Bel-Rhlid, & Juillerat, 2002). 2-
428 Acetyltetrahydropyridine, 2-propionyl-1-pyrroline and 2-acetyl-2-thiazoline were not detected
429 in SPME extracts of boiled rice samples in the present study.

430 2-AP was detected in some non-fragrant rice varieties in previous studies using
431 different extraction and quantification techniques; concentrations of 2-AP in non-fragrant rice
432 have been reported from 0.6 µg/kg to 24.7 µg/kg (Buttery et al., 1983; Buttery, Turnbaugh, &
433 Ling, 1988; Maraval et al., 2010). The lowest concentration of 2-AP standard that could be
434 quantified by GC-MS in our study was 5 µg/kg (see calibration curve preparation in *Section*
435 *3.2.3.*). Trace levels of key 2-AP ions (m/z 68, m/z 83 and m/z 111) were detected in samples
436 which contained less than 5 µg/kg 2-AP; these trace peaks could not be quantified. In our study,
437 2-AP levels in three non-fragrant rice varieties were lower than the limit of quantification (5
438 µg/kg), although peaks for the key ions of 2-AP could be observed (**Figure 3**).

439 *3.3. Detection of 2-acetyl-1-pyrroline in raw rice by GC-olfactometry*

440 The sensory evaluation results showed that popcorn-like aroma can be perceived in
441 boiled non-fragrant rice, although the intensity in non-fragrant rice is significantly lower than
442 in fragrant rice. The odour thresholds of 2-AP are 0.1 nL/L in water (Buttery et al., 1983) and
443 0.02 ng/L in air (Schieberle, 1991), levels which are much lower than the limit of quantification
444 (LOQ) of our method (5 µg/kg). Therefore, to confirm the presence of 2-AP or other popcorn-
445 like aroma contributors in non-fragrant rice samples, GC-olfactometry is likely to be a
446 technique with higher sensitivity than GC-MS.

447 The trial tests on raw and boiled fragrant and non-fragrant rice using SPME followed
448 by GC-O showed that popcorn note could not be perceived in raw or boiled non-fragrant rice

449 but could be perceived in raw and boiled fragrant rice; the compound which contributed this
450 popcorn-like note was identified as 2-AP based on its retention time. The SPME process that
451 was used for GC–O may not have been sensitive enough, since only 1 g of rice sample was
452 extracted, and only compounds in the rice headspace could be adsorbed. Moreover, only half
453 of the extract reached the GC–O sniffer port while the other half was split to the FID. In
454 addition, manual SPME instead of automatic SPME sampler was used with GC–O in our
455 laboratory, and this may also reduce the sensitivity of the analysis since agitation did not occur
456 during extraction when using manual SPME.

457 Therefore, the use of solid-phase extraction (SPE) as an extraction technique was
458 investigated. The aqueous extract from 10 g of rice sample could be loaded onto the SPE
459 cartridge and the dichloromethane used to elute the 2-AP could be concentrated to around 100
460 μL for analysis. In work carried out in our laboratory, gelatinisation of starch meant that the
461 supernatant from the centrifuged boiled rice/water solution could hardly pass through the SPE
462 sorbent. As Yoshihashi (2002) reported that 2-AP cannot be formed during rice boiling, it was
463 decided to extract uncooked rice. Therefore, raw milled Sintanur (fragrant) and Ciherang (non-
464 fragrant) were extracted and analysed by GC–O, to discover if popcorn aroma could be detected
465 in non-fragrant rice by GC–O.

466 The results from the GC–O analysis (four assessors analysing each rice extract in
467 duplicate) showed that popcorn-like odour was only perceived over an LRI range between 1330
468 and 1347 in both raw Sintanur and Ciherang rice. The LRI value of 2-AP on the same stationary
469 phase (Zebron ZB-Wax) when used in the GC–MS analysis was 1333. Therefore, it seems
470 likely that 2-AP was the sole contributor to perceived popcorn-like odour in both raw fragrant
471 and non-fragrant rice. Sniffers rated aroma intensity from 0 (nil) to 10 (extreme) when
472 compounds eluted from the GC column. Average perceived 2-AP intensity in Sintanur was
473 7.00 ± 0.50 and in Ciherang was 3.88 ± 0.93 ; Student's *t*-test showed that 2-AP intensity in

474 Sintanur was significantly higher than in Ciherang ($p = 0.0001$). In addition, all the sniffers
475 scored popcorn intensity higher for Sintanur rice than Ciherang rice.

476 The concentration of 2-AP in boiled fragrant rice was at least 15-fold higher than that
477 in boiled non-fragrant rice (based on the LOQ of 2-AP obtained using SPME with GC–MS) in
478 our study; however, its perceived odour intensity in raw fragrant rice by GC–O was only two
479 times higher than in raw non-fragrant rice. As discussed in *Section 3.1*, the odour perception
480 of 2-AP fits Steven’s law and shows a decelerating relationship with increasing concentration.
481 Since the detection threshold of 2-AP is 0.02 ng/L in air (Schieberle, 1991), which is much
482 lower than the LOQ of 2-AP, the difference in 2-AP perceived intensity between fragrant and
483 non-fragrant rice is somewhat less than the difference in 2-AP concentration.

484 Although 2-AP in non-fragrant rice could not be quantified by GC–MS in our
485 laboratory, GC–O provided clear evidence that a low concentration of 2-AP was present in raw
486 Ciherang non-fragrant rice. Based on the sensory profiling of boiled non-fragrant rice, it can
487 be concluded that 2-AP can also contribute popcorn-like odour to non-fragrant rice.

488 Mutation of the gene *badh2* is regarded as the key reason for 2-AP generation in
489 fragrant rice (Bradbury, Fitzgerald, Henry, Jin, & Waters, 2005, Fitzgerald, McCouch, & Hall,
490 2009). Due to the loss of function of the enzyme BADH2 caused by mutated *badh2*, the
491 metabolite GABA is dehydrated to 1-pyrroline in fragrant rice (rather than forming γ -
492 aminobutyric acid (GABA) through BADH2 catalysis) and then acetylated to 2-AP (Bradbury,
493 Gillies, Brusheet, Waters, & Henry, 2008). However, as a positive correlation was found
494 between 2-AP and the amino acid metabolite 1-pyrroline-5-carboxylate (P5C) in fragrant rice,
495 a BADH2-independent pathway was proposed by Huang et al. (2008). Ornithine, glutamic acid,
496 and proline can form P5C through amino acid metabolism; P5C could be degraded to 1-
497 pyrroline then acetylated to 2-AP, or P5C could react with methylglyoxal to generate 2-AP

498 directly. In the study of Huang et al. (2008), 2-AP was not detected in non-fragrant rice samples
499 using GC–FID, as this technique is not sensitive enough to detect 2-AP in non-fragrant rice.
500 Hence a correlation between P5C and 2-AP in non-fragrant rice was not reported. However,
501 the presence of P5C and methylglyoxal was noted, which could generate a small amount of 2-
502 AP during non-fragrant rice growth.

503 While generation of 2-AP during growth of non-fragrant rice may occur, its formation
504 post-harvest appears unlikely. Several studies have reported 2-AP losses in fragrant rice when
505 it is dried and stored under a variety of conditions (Wongpornchai, Dumri, Jongkaewwattana,
506 & Sirri, 2004; Widjaja, Craske & Wootton, 1996a,b). 2-AP formation was reported at 100 °C
507 in a proline + methylglyoxal model system in phosphate buffer (Hofmann & Schieberle, 1998),
508 suggesting that boiling may generate 2-AP in rice. However, as stated earlier, Yoshihashi (2002)
509 measured 2-AP in fragrant rice after heating at 90 °C without water, and boiling with water for
510 8, 10, 12, and 14 min, and concluded that 2-AP could not be generated during rice cooking.

511 Detection of 2-AP is a limitation of the current study; only traces of 2-AP were detected
512 in non-fragrant rice by GC-MS. GC with quadrupole-time-of-flight mass spectrometry could
513 provide higher sensitivity and resolution than single quadrupole MS. For example, a problem
514 in analysis of 2-AP by GC-MS is the coelution of 2-AP with 6-methyl-5-hepten-2-one on a
515 polar GC column, both compounds having a number of fragment ions in common. However,
516 these fragment ions with the same unit mass have different molecular formulae and would be
517 readily separated under high resolution conditions, leading to an increase in the signal-to-noise
518 ratio for 2-AP (Wei et al., 2017). The use of chemical ionisation (CI) rather than electron
519 ionisation mass spectrometry could also improve detection sensitivity, as the former is a softer
520 ionisation technique, producing a strong $M + 1$ ion at m/z 112 (Maraval et al., 2010).

521 **4.4. Conclusions**

522 This study emphasised that 2-AP and popcorn-like attributes given by 2-AP (odour,
523 flavour, and after-effect) are the most important discriminators between fragrant and non-
524 fragrant boiled rice. Sensory profiling showed that significant differences were observed in
525 popcorn odour, flavour, and after-effect when fragrant and non-fragrant rice samples were
526 compared as two groups. 2-AP quantification concluded that significant differences in 2-AP
527 concentration between the three fragrant rice types were too small to cause differences in their
528 perceived popcorn-like aroma. Trace levels of 2-AP were found in non-fragrant rice by GC-
529 MS, and its presence in non-fragrant rice was confirmed by GC-O, but levels were lower than
530 the limit of quantification by GC-MS. At least 15 times higher levels of 2-AP were found in
531 fragrant rice than non-fragrant rice (based on the LOQ of 2-AP by GC-MS).

532 Our study emphasised that 2-AP is the most important aroma contributor in fragrant
533 rice and confirmed that 2-AP and its popcorn-like aroma is the discriminator for fragrant and
534 non-fragrant rice. However, the popcorn-like aroma of 2-AP can also be perceived in non-
535 fragrant rice, although below the level of detection of the GC-MS used in this work.

536

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540

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652

653 **Figure captions**

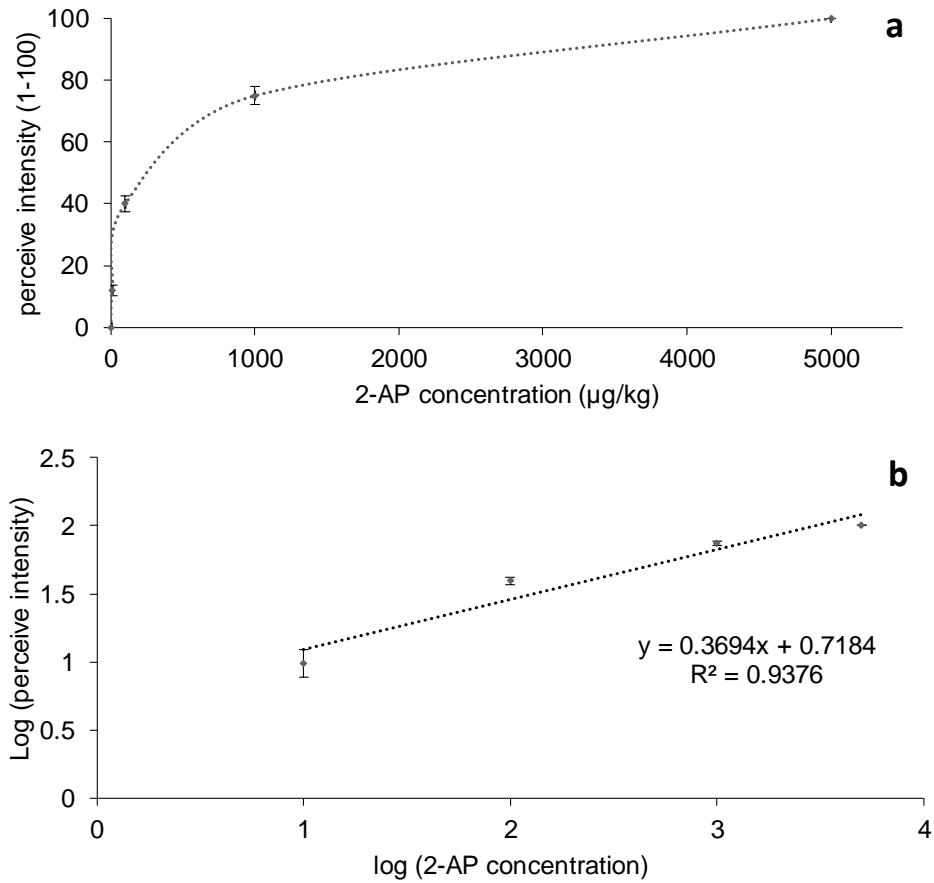
654 **Figure 1:** (a) Mean perceived intensity of odour of 2-acetyl-1-pyrroline (2-AP) standard
655 references (0, 10, 100, 1000 and 5000 $\mu\text{g}/\text{kg}$) assessed by 11 panellists; (b) log
656 stimulus *vs* log response plot of perceived intensities of odour of 2-AP standard
657 references (10, 100, 1000 and 5000 $\mu\text{g}/\text{kg}$) from 11 panellists. Error bars represent
658 standard error of the mean.

659 **Figure 2:** Perceived intensities of odour, taste, and flavour-related attributes for fragrant and
660 non-fragrant rice types. The numbers above the bars indicate the probability that the
661 samples are significantly different ($p < 0.05$; Student's *t*-test). Error bars represent
662 standard error of the mean.

663 **Figure 3:** 2-AP concentrations in six boiled rice samples. Bars not sharing a common letter
664 are significantly different ($p < 0.05$). Error bar represents standard deviations. 'trace':
665 concentration lower than 5 $\mu\text{g}/\text{kg}$.

666

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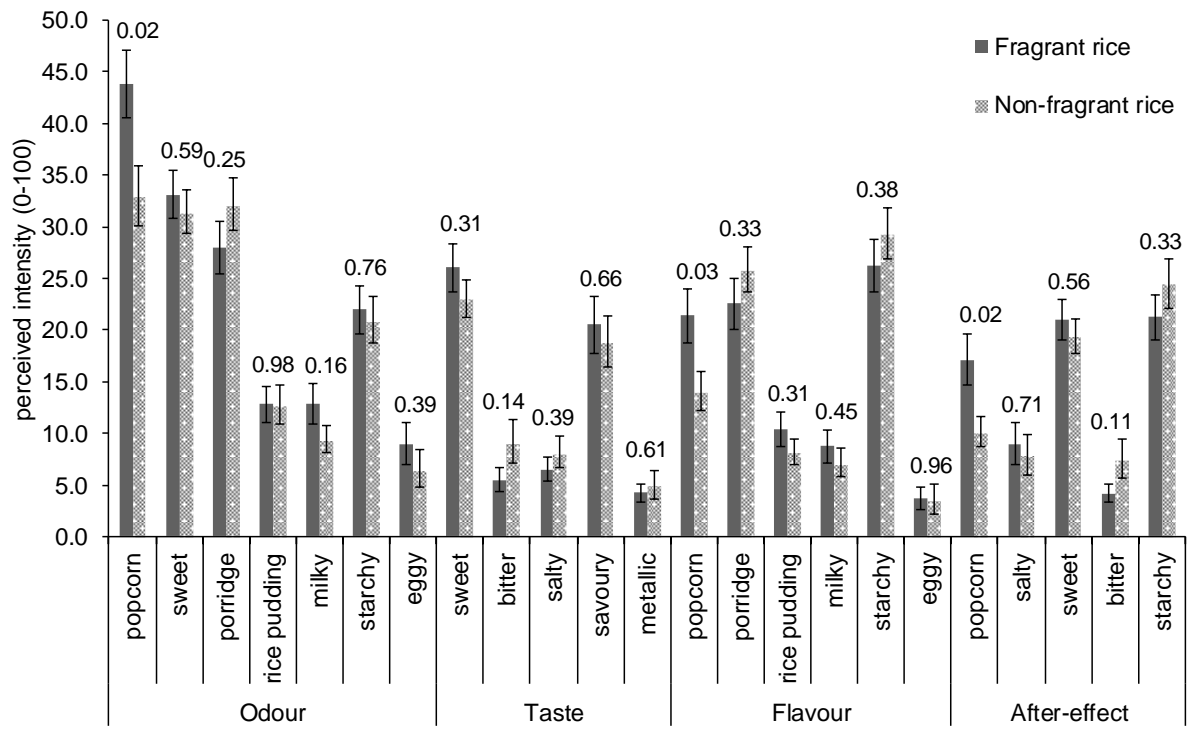
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670 **Figure 1:** (a) Mean perceived intensity of odour of 2-acetyl-1-pyrroline (2-AP) standard references
 671 (0, 10, 100, 1000 and 5000 µg/kg) assessed by 11 panellists; (b) log stimulus vs log response plot of
 672 perceived intensities of odour of 2-AP standard references (10, 100, 1000 and 5000 µg/kg) from 11
 673 panellists. Error bars represent standard error of the mean.

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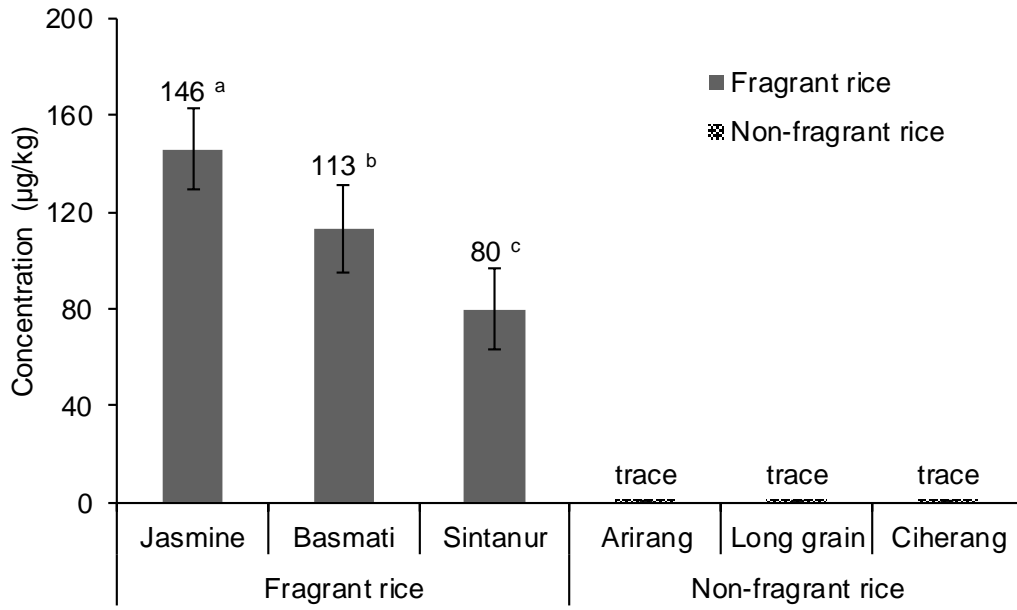
679 **Figure 2:** Perceived intensities of odour, taste and flavour-related attributes for fragrant and non-
 680 fragrant rice types. The numbers above the bars indicate the probability that the samples are
 681 significantly different ($p < 0.05$; Student's t -test). Error bars represent standard error of the mean.

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688 **Figure 3:** 2-AP concentrations in six boiled rice samples. Bars not sharing a common letter are
 689 significantly different ($p < 0.05$). Error bar represents standard deviations. 'trace': concentration lower
 690 than 5 µg/kg.

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Table 1: Consensus vocabulary for boiled rice developed by 11 trained UK panellists during sensory profiling.

attributes	definition	reference	anchors
<i>appearance</i>			
brown lines	extent of brown lines on the surface of rice grains		nil to extreme
wet	moistness of rice grain		dry to wet
yellow	colour of rice grain		white to yellow
uniform	shape of rice grain		irregular to regular
separated grain	separation between rice grains after cooking		unseparated to separated
length	length of rice grain		short to long
thickness	thickness of rice grain		thin to thick
<i>mouthfeel</i>			
smooth	smoothness of the sample on chewing		nil to extreme
effort to chew	springiness of the sample on chewing		nil to extreme
drying	mouth drying		nil to extreme
cohesive	stickiness of rice grain		nil to extreme
watery	how moist the sample felt in the mouth		nil to extreme
<i>odour</i>			
popcorn	aroma of popcorn	Five sniff stripes wetted in a blank solution and four levels of 2-acetyl-1-pyrroline standard (10, 100, 1000 and 5000 µg/kg) and placed in sniff bottles	nil to extreme, standards were given as three anchors at 0, 12, 40, 75 and 100 along the line scale
sweet	aroma of Demerara sugar	Demerara sugar	nil to extreme
porridge	aroma of cooked oat porridge	Quaker wholegrain rolled oats porridge (Quaker, UK)	nil to extreme
rice pudding	aroma of rice pudding	Ambrosia original tinned rice pudding (Ambrosia, UK)	nil to extreme
milky	aroma of uncooked milk	pasteurised Tesco skim milk (Tesco, UK)	nil to extreme
starchy water	aroma of starch water from boiled non-fragrant rice	cold starchy water collected from boiled non-fragrant rice	nil to extreme
eggy	aroma of boiled egg		nil to extreme

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<i>taste</i>			
sweet	elicited by sucrose		nil to extreme
bitter	elicited by caffeine		nil to extreme
salty	elicited by sodium chloride		nil to extreme
savoury	brothy or meaty		nil to extreme
metallic	metal-like		nil to extreme
 <i>flavour</i>			
popcorn	flavour of popcorn		nil to extreme
porridge	flavour of oat porridge	Quaker wholegrain rolled oats porridge (Quaker, UK)	nil to extreme
rice pudding	flavour of rice pudding	Ambrosia original tinned rice pudding (Ambrosia, UK)	nil to extreme
milky	flavour of uncooked milk	pasteurised Tesco skim milk (Tesco, UK)	nil to extreme
starchy water	flavour of starch water from boiled non-fragrant rice	cold starchy water collected from boiled non-fragrant rice	nil to extreme
eggy	flavour of boiled egg		nil to extreme
 <i>after-effect</i>			
popcorn	residual popcorn odour and flavour in mouth after swallowing		nil to extreme
salty	residual saltiness in mouth after swallowing		nil to extreme
sweet	residual sweetness in mouth after swallowing		nil to extreme
bitter	residual bitterness in mouth after swallowing		nil to extreme
drying	mouth drying after swallowing		nil to extreme
residue	particulates left in mouth after swallowing		nil to extreme
starchy water	starchy water flavour in mouth after swallowing		nil to extreme

698 **Table 2:** Mean value and significance of sensory attributes for six boiled rice types. Where values in a
699 row do not share the same letter, they are significantly different ($p < 0.05$, Fishers LSD)

attributes	Mean value of perceived intensity (0–100)						effect of rice type (p -value)
	fragrant rice			non-fragrant rice			
	Jasmine	Basmati	Sintanur	Arirang	American long-grain	Ciherang	
appearance							
wet	28.9 ^{ab}	5.86 ^c	30.3 ^{ab}	35.2 ^a	15.4 ^{bc}	28.9 ^{ab}	< 0.0001
yellow	14.8	16.2	20.7	24.3	13.7	14.6	0.203
brown lines	1.99 ^c	4.50 ^{abc}	2.93 ^{bc}	10.0 ^{ab}	10.6 ^a	11.4 ^a	< 0.0001
uniform	64.4 ^a	71.4 ^a	57.5 ^{ab}	56.3 ^{ab}	41.9 ^b	55.5 ^{ab}	< 0.0001
separated grain	47.8 ^{ab}	56.5 ^a	41.5 ^{ab}	31.5 ^b	31.4 ^b	36.9 ^b	< 0.0001
length	56.6 ^{ab}	71.9 ^a	38.1 ^c	37.7 ^c	46.1 ^{bc}	49.1 ^{bc}	< 0.0001
thickness	51.5 ^a	29.7 ^b	55.7 ^a	65.4 ^a	51.9 ^a	53.0 ^a	< 0.0001
odour							
popcorn	49.1 ^a	43.2 ^{ab}	39.1 ^{ab}	32.2 ^{ab}	42.0 ^{ab}	24.9 ^b	0.028
sweet	39.3	32.1	28.0	31.1	34.8	28.3	0.297
porridge	31.8	26.6	25.5	35.6	30.2	30.7	0.647
rice pudding	16.8	9.90	11.9	14.5	13.7	10.1	0.609
milky	16.1	12.0	10.6	11.0	10.0	7.34	0.479
starchy water	21.3	22.0	22.6	16.0	19.2	27.8	0.442
eggy	9.32	10.1	7.42	2.72	6.30	10.8	0.572
taste							
sweet	27.5	23.8	27.0	24.9	23.2	21.0	0.819
bitter	8.92	3.15	4.44	8.12	10.8	8.70	0.472
salty	7.88	4.79	6.91	8.88	6.94	8.70	0.843
savoury	19.0	23.3	19.3	22.3	15.6	18.7	0.875
metallic	4.20	3.29	5.19	4.15	6.57	4.49	0.909
flavour							
popcorn	24.5	16.2	23.5	17.3	12.8	12.2	0.134
porridge	22.7	22.8	22.2	30.8	24.0	22.8	0.691
rice pudding	13.5	6.51	11.2	11.0	5.76	7.98	0.235
milky	12.8	4.15	9.27	7.85	7.66	5.94	0.254
starchy water	25.7	23.0	30.0	32.6	23.1	32.4	0.402
eggy	7.68	1.46	1.98	2.49	1.89	6.45	0.173
mouthfeel							
smooth	50.7	48.9	52.7	46.0	38.6	44.5	0.316
effort to chew	38.3	45.7	36.6	43.8	47.0	41.3	0.489
drying	33.7	36.8	32.3	31.9	34.5	34.9	0.981
cohesive	44.1 ^{ab}	22.4 ^c	44.6 ^{ab}	54.8 ^a	28.1 ^{bc}	39.6 ^{abc}	< 0.0001
watery	12.2	4.55	10.5	12.3	8.09	11.0	0.376
after-effect							
popcorn	18.1	11.3	22.1	12.2	10.7	7.70	0.057
salty	7.84	11.2	7.98	11.4	7.85	4.48	0.765
sweet	22.6	18.8	21.6	20.1	17.8	20.5	0.912
bitter	5.77	3.49	3.35	6.99	8.43	7.18	0.664
drying	27.8	31.8	28.0	23.7	29.9	32.9	0.766
residue	27.8	16.8	28.4	26.0	22.8	24.6	0.664
starchy water	23.0	20.2	20.7	27.0	19.6	26.8	0.639