Modelling flavour formation in roasted malt substrates under controlled conditions of time and temperature

Hebe PARR[a], Irina BOLAT[b], David COOK[a‡]

[a] International Centre for Brewing Science, The University of Nottingham, Sutton Bonington Campus, Loughborough, Leicestershire, LE12 5RD, UK

[b] Boortmalt NV, Zandvoort 1, Haven 350, Bus 1, 2030 Antwerp, Belgium

hebe.parr@nottingham.ac.uk

irina.bolat@boortmalt.com

‡Corresponding Author:

david.cook@nottingham.ac.uk

+44 (0) 115 9516245

Abstract

Drum roasted products are used to impart colour, flavour and mouthfeel to beers. Here we designed a laboratory-scale roaster (100 g batch size) capable of precise time-temperature control and investigated the impacts of time, temperature and roasting substrate (barley, pale malt or germinated green malt) on formation of 20 key odour active aroma volatiles. Principal Components Analysis (PCA) of flavour volatile data across 37 laboratory roasted and 6 commercial roasted products generated a product flavour space depicting the relationship between roasting conditions and concentrations of these 20 compounds. Response surface models were produced for aroma compound concentrations across the design space of roasting times and temperatures for each substrate. These clearly illustrate the impacts of substrate moisture content and prior history (e.g. whether germinated or germinated and kilned) on flavour formation. In low moisture substrates a steep increase in associated heterocyclic aroma compound production was noted at process temperatures >180°C.

Keywords:

Roasted Malt Flavour; Gas Chromatography-Mass Spectrometry; Modelling Flavour Formation; Thermal Flavour Generation; Maillard Reaction

1. Introduction

Roasted or kilned speciality malts are used in brewing at low grist percentages to contribute desirable flavours, colours and mouthfeel to beers. The spectrum of flavours that are available from roasted products results from a number of contributing factors: the cereal and whether malted or unmalted, variety, malting parameters/degree of modification, and the thermal processing steps; namely kilning, stewing, and roasting (Coghe, Martens, D'Hollander, Dirinck, & Delvaux, 2004). Thermal processing steps have the greatest influence on the final flavour attributes of roasted malt products (Yahya, Linforth, & Cook, 2014).

Roasted malts can be separated intro three main categories, due to the roasting substrates that are used: colour malts, caramel/crystal malts, and roasted barley (Coghe et al., 2004; Gretenhart, 1997). The substrates are, respectively: pale malt, green malt, and raw barley. These raw materials are all taken from various stages of the malting process. Raw barley has not undergone malting. Green malt is the product of the steeping and germination of barley. The high moisture content of green malt (40 % - 45 %) provides the internal conditions to form it's characteristic 'glassy' endosperm under stewing and roasting as a result of amylolysis and proteolysis (Blenkinsop, 1991; Gruber, 2001; Vandecan, Daems, Schouppe, Saison, & Delvaux, 2011). Pale malt is the product of kilning green malt. As pale malt will have undergone thermal processing before roasting, it has a low moisture content, and retains some of the aromatics and flavour derived from the barley's natural sweetness (Gruber, 2001).

Previous studies of dark roasted speciality malts note the reliance of maltsters on monitoring the development of colour throughout the roasting process to indicate the extent of thermal flavour generation (Coghe, Gheeraert, Michiels, & Delvaux, 2006; Yahya et al., 2014). This approach neglects the significance of the roasting conditions on the extent of thermal flavour

generation reactions. In addition, the EBC (European Brewery Convention) colour of a malt can increase then decrease at the highest roasting temperatures (Vandecan et al., 2011). In commercial roasting operations, relying solely on colour data can result in batch to batch variation in the flavours the roasted product will impart to a product when used. Similarly, the colour of a roasted malt's husk does not necessarily indicate the colour of the endosperm within.

An investigation, studying the formation of flavour and colour of dark speciality malts by Vandecan et al. (2011) noted the importance of the moisture content of the malt during the roasting process. The malts in the study were processed up to 180 °C, which does not include the very highest temperatures used to commercially produce speciality malts (O'Shaughnessy, 2003). In commercial roasting operations, longer roasting times (up to 170 min in some cases) are employed to ensure the product temperature is as consistent as possible within the batch (O'Shaughnessy, 2003). The laboratory roasted products in the current study cover the full range of conditions employed to produce speciality malts, proportionate to the reduced batch size of a laboratory scale roaster.

The present study used Gas Chromatography-Mass Spectrometry (GC-MS) analysis to quantify and model the formation of 20 odour active compounds in roasted products produced from three different and commonly used roasting substrates (barley, green malt, and pale malt). In a prior study from our group (Yahya et al., 2014) we investigated flavour development in 3 commercial roasted products by sampling from roasting drums during their production; snap freezing samples in liquid nitrogen, and subsequently analysing the time-point samples for their flavour volatile profiles using Gas Chromatography. In the present study the objective was to model the formation of key roasted product flavour compounds across a range of process times, temperatures and initial moisture contents such that we could map the flavour space of potential roasted products prepared from the three basic substrates.

A key hypothesis of the present research was that such a flavour map, linking roasting conditions to the volatile flavour composition of products, might suggest conditions for the production of new products with novel flavours; notwithstanding that, a better understanding of flavour control through roasting operations should be attained. To do this we designed a laboratory scale roasting drum, featuring a cylindrical mesh cage which was rotated inside a Gas Chromatograph oven, used for precise time-temperature control. The objective here was to accurately control the roasting conditions in small batches (100 g) of substrate so that flavour formation could be accurately modelled relative to those conditions. It is acknowledged that further work would then be required to translate these findings to commercial roasting drum operations where bulk effects and differences in power input per tonne of substrate would impact on flavour formation. However, the present approach does enable a deeper understanding of how variation of the thermal processing conditions impacts on the formation of key groups of flavour compounds. The flavour volatile profiles of roasted malt products are complex. In this study we monitored the formation of 20 key compounds which were selected based on their known aroma impacts (from prior GC-Olfactometry studies (Parr, Bolat, Miller, Clegg, & Cook, 2018)) and which were representative of different thermal flavour generation chemistries – e.g. Maillard chemistry, Strecker degradation, caramelisation, lipid degradation.

2. Materials and Methods

2.1. Roasting Materials and Commercial Samples

All laboratory roasted products in this study were produced from the same batch of a winter variety of malting barley (Flagon) provided by Crisp Malt Ltd. The commercial roasted products investigated in this study were provided by Paul's Malt (Boortmalt) (Table 1, Figure 1).

2.2. Chemicals

Authentic analytical standards (>95% purity) were purchased to identify and quantify the 20 aroma compounds within the roasted samples. Suppliers of chemicals were as follows: Sigma Aldrich: 2-methylfuran, pentanal, hexanal, 1-methylpyrrole, pyrazine, 2-pentylfuran, 2,3-dimethylpyrazine, furfural, acetic acid, methyl-2-furoate, 5-methylfurfural, 2-acetyl-5-methylfuran, phenylacetaldehyde, 2-furanmethanol, 2-(5H)-furanone, furaneol, and hydroxymethylfurfural. Fisher Scientific: 2-n-pentylpyridine, maltol, and 2-formylpyrrole. Methanol (HPLC/ LC-MS grade) used for solvent extraction of volatile compounds was sourced from VWR International Ltd.

2.3. Laboratory Scale Roaster

A GC oven (Hewlett Packard (HP) 6890 Series GC System) was modified to accommodate a mini roasting vessel (drum dimensions: 8 cm diameter x 15 cm length. Mesh: 2×2 mm). The roasting substrate sample (~100 g batch size) was filled into the mesh drum and then attached to a rotating shaft via a push fit closure sealed with a heat resistant O-ring to secure the drum, while allowing easy release from the rotating component when roasting was complete. A barbecue rotisserie motor (GM012 model, BBQ Foukou, Korakas, Cyprus) was used to rotate the mesh drum (at 43 RPM) within the GC oven. The modification of the GC oven allowed accurate temperature control during roasting.

2.4. Sample Preparation

2.4.1. Micromalting

Barley was micromalted using a Custom Lab Micromaltings K steep-germinator and kiln (Custom Laboratory Products, Keith, UK). The steep germinator housing four drums (500 g barley per drum) was used to produce the green malt and pale malt for this study. Malting was carried out under the following conditions: Steeping (16 °C) 7 h wet, 12 h dry, 8 h wet, 12 h dry, 4 h wet. Germination: 5 days at 16 °C. The drums were mechanically rotated every 10 minutes to prevent matting of rootlets. The green malt produced was then either refrigerated (0-5 °C) and roasted within the day, or kilned to produce pale malt. Kiln programming to produce pale malt was as follows: 55 °C for 12 h, followed by 65 °C for 6 h, then 85 °C for 2 h, and finally 95 °C for 2 h. The pale malt was then cooled to ambient temperature before removing rootlets. Samples were vacuum packed in foil-lined pouches, and stored at -80 °C for use within one month.

2.4.2. Production of laboratory roasted malts

Preliminary experiments were conducted using the laboratory scale roaster to determine the time ranges within which each substrate could be heated at temperatures between 100-230 °C to achieve representative roasted products. These ranges of time-temperature encompassed the realm of normal roasted products and also some additional extremes such that at the edges of design spaces some samples were not dried down to typical finishing moisture content or at the top end some samples bordered on the ‘burnt toast’ end of roasting.

Roasting parameters (isothermal in each case) for the roasting substrates were selected as follows:

- Pale malt and raw barley:
    - Time: 10, 15, 20, 25, and 30 min
    - Temperature: 100, 135, 165, 200, and 230 °C
- Green malt:
    - Time: 20, 28, 35, 43, and 50 min
    - Temperature: 135, 143, 150, 158, and 165 min

Green malt was first 'stewed' in a sealed glass bottle at 65 °C for 1 hour in a laboratory oven (Genlab Ltd., Cheshire, UK) before being transferred to the roasting drum and roasted under a series of time-temperature conditions within the above boundaries and as determined using experimental design software.

Design Expert (Version 11, StatEase, Minneapolis, USA) was used to create a 24 point D-optimal response surface design based on the above ranges of time and temperature for each of the three roasting substrates. Each substrate was then roasted using the 24 different combinations of time and temperature according to the D-optimal design. Run order was fully randomised within the design. After roasting, the products were immediately removed from the drum, and frozen in liquid nitrogen (-196 °C). At this stage, the frozen roasted green malt's rootlets were removed whilst brittle. The roasted products were vacuum packed, and stored at -80 °C prior to analysis.

2.4.3. Extraction of flavour volatiles

Flavour compounds from roasted barley and malt samples were extracted into methanol according to the method previously described by Yahya et al. (2014). A Buhler Miag disc mill (Uzwil, Swizerland) was used to produce a fine powder (0.2 mm) of each roasted product. Methanol (16 mL) containing an internal standard (5-nonanone, 5 μg/mL) was added to 8 g of sample in a sealable glass vial and mixed on a roller bed for 30 min, then transferred to a centrifuge tube by Pasteur pipette and centrifuged at 4000 *g* for 10 min. The supernatant

was then transferred to GC vials and stored at -80 °C prior to analysis. One flavour extract was prepared from each roasted sample.

2.5. Gas Chromatography – Mass spectrometry (GC-MS) Operating Conditions

The volatile compounds within the flavour extracts were separated using a Trace 1300 Gas Chromatograph (Thermo Scientific, Waltham, MA, USA), fitted with a ZB-Wax column (30 m × 0.25 mm ID × 1.0 μm film thickness; Phenomenex, Macclesfield, UK). The injector was operated in splitless mode (240 °C, 1 min), with helium carrier gas (18 psi). The oven temperature was programmed as follows: 40 °C for 1 min, then a temperature ramp at 4 °C/min to 220 °C, holding for 10 min. All GC effluent was analysed by the MS (Thermo Scientific, Waltham, MA, USA). The MS was run on selected ion methods (SIM) to identify the specific compounds of interest. m/z values monitored in each SIM are detailed in Methods 2.5.1 and 2.5.2. The selected ions were monitored for the corresponding time window in which the compound would elute from the column to prevent overburdening the method.

A guard column was used to prevent the impurities within the flavour extracts degrading the column itself, that would otherwise have resulted in reducing the accuracy of the peak areas recorded. The guard column and injector liner were changed after every 24 injections of samples to retain accuracy of data.

2.5.1. m/z values monitored in SIM 1

The m/z values monitored of the compounds of interest in SIM1: 2-methylfruan (81, 82), hexanal (56, 82), pyrazine (53, 80), 2,3-dimethylpyrazine (67, 108), furfural (95, 96), 2-n-pentylpyridine (93), methyl-2-furoate (95, 126), phenylacetaldehyde (91, 120), 2-(5H)-furanone (55, 84), furaneol (85, 128), hydroxymethylfurfural (97, 126).

2.5.2. m/z values monitored in SIM 2

The m/z values monitored of the compounds of interest in SIM2: pentanal (58, 86), 1-methylpyrrole (80, 81), 2-pentylfuran (81, 138), acetic acid (45, 60), 5-methylfurfural (109, 110), 2-acetyl-5-methylfuran (109, 124), 2-furanmethanol (97, 98), maltol (71, 126), 2-formylpyrrole (94, 95).

2.5.3. Peak Identification and Quantification by External Standards

Compounds were identified based upon three levels of validation: linear retention index (LRI) against alkanes (C8-C22) when compared to literary sources on the same WAX phase; LRI comparison with authentic standards when assessed under the same chromatographic conditions; and by EI-MS library matching. These methods of identification were carried out in addition to the previous identification of the 20 compounds' known aroma impact on the range of commercial roasted products from prior GC-Olfactometry studies (Parr et al., 2018). Authentic analytical standards of the 20 selected flavour volatiles were analysed by GC-MS at the following concentrations to give a calibration curve, from which concentrations could be calculated in the samples: 0.5 ppm, 1 ppm, 5 ppm, 10 ppm, 25 ppm. An internal standard (5-nonanone, 5 μg/mL) was used in each standard solution. Concentrations are reported as μg/g of roasted sample (as-is basis).

2.6. Moisture content determination

The moisture content of each of the 24 roasted samples for each substrate was determined according to EBC Analytica Method 4.2. Samples with moisture content <5% were considered to be 'finished' roasted products.

2.7. Data Modelling and Statistical Analysis

Following GC-MS analysis, the concentrations of each compound in each of the three substrates were modelled against the factors of time and temperature using the Design Expert software. Factors which were non-significant ($P > 0.05$) were removed from models until a significant model resulted with factors each of which were significant ($P < 0.05$), and the model R^2 was maximised. Interactions between factors were included in models where significant. Statistical details of the models of each compound in each roasting substrate are detailed in Table 2.

Principal Component Analysis (PCA) was carried out using XLSTAT software (Addinsoft, SARL, Paris) in order to depict the relationship between the concentrations of the 20 compounds over the range of roasted products' roasting time, temperature, and substrate.

3. Results and Discussion

The moisture content and analysed concentrations of each of the 20 odour active volatile compounds in the full set of laboratory roasted and commercial malt samples is reported in Table 1. To facilitate interpretation of this large amount of data we will first visualise the variation in the data set using PCA. We will then present response surface models showing the trends in volatile formation as a function of roasting time and temperature for five volatiles selected to be representative of different thermal flavour generation chemistries; namely: maltol, pyrazine, 2-acetyl-5-methylfuran, 2-pentylfuran and phenylacetaldehyde. Modelling data for all 20 compounds are summarised in Table 2 and indicate factors such as the fitted model significance, factor significance (time, temperature), model R^2 values and the degree of polynomial used to fit the data in each case.

3.1. Principal Component Analysis (PCA)

PCA was used to analyse the variation in concentrations of the 20 volatile compounds across 37 laboratory roaster prepared samples and the sample set of commercial roasted products. The number of laboratory roasted samples used for PCA analysis was narrowed down by including only those (n=37) which were deemed to be 'finished' products after roasting, i.e. had a moisture content of less than 5% w/w (Table 1).

Figure 1 shows the biplot of principal components 1 & 2. PC1 accounts for 37.05 % of the variation in the data set, whereas PC2 accounts for 28.80 % of the variation in the data set. PC1 mainly separates the samples according to roasting substrate, and the degree to which those substrates were roasted. Green malt samples load positively on PC1, as to a lesser extent do pale malt samples that were roasted for relatively short times at lower temperatures. Samples that project more negatively on PC1 have been roasted at higher temperatures, and for longer times. This trend is exhibited within each substrate group of the roasted samples.

PC2 is largely driven by the concentration of volatile compounds in the samples, which is why all of the volatile loading vectors project upwards in Figure 1. Samples plotted more positively on PC2 are more likely to have a higher concentration of the compounds investigated.

In generic terms, the upper right quadrant of Figure 1 represents volatile compounds which are maximised in green malt products, most typically as a result of Maillard chemistry. The diagonally opposite sector features samples which are the opposite of this – i.e. samples which are lowest in these Maillard products. Logically, these are roasted barley samples which had not been malted or stewed and thus contained particularly low concentrations of Maillard reaction precursors. The top left sector defines the heavily 'dry roasted' sector of products typified by black malt and chocolate malt commercial products. The volatile composition is typified by heterocyclic compounds such as pyrazines, substituted furans and pyrroles. In terms of the laboratory roaster samples the longest roasted samples of barley and pale malt at the highest temperatures (200-230 °C) tend to feature in this sector. The diagonally opposite sector (bottom right) features the lower temperature treated pale malt samples, which were much lower in their content of heterocyclics.

The clustering of samples according to their substrate type in Figure 1 clearly demonstrates the significance of substrate on roasted product flavour development. Whether or not the barley has been malted, its moisture content at the start of roasting, and whether or not a period of 'stewing' is utilised all exert a substantial influence over the product flavour characteristics. This is why it was important to include the three fundamental barley substrates in the present study. Whilst forming distinct clusters under less intense roasting conditions, roasted barley and pale malt samples locate in the upper left-hand sector of Figure 1 and become more similar to one another in their volatile composition as they are roasted at very high temperatures. Put simply, barley and pale malt have a more similar volatile

293 composition when pyrolyzed at higher temperatures and low moisture contents, but are
294 distinct from one another when more subtle roasting processes are applied. The latter
295 conditions enable the pale malts to generate and retain some characteristic Maillard pathway
296 intermediates and products, which is why those samples load positively on PC1.
297 Commercially available samples were analysed in this study to show where the lab roasted
298 samples fell within the commercial range of products. Proximity or co-location of samples on
299 Figure 1 means similarity in flavour composition, and samples projecting closely to specific
300 volatile loading vectors contain high levels of those particular compounds, whilst samples
301 positioned diametrically opposite a volatile compound contain the lowest levels. For
302 example, [RB, 230, 20] is plotted closely to 2,3-dimethylpyrazine, which indicates its
303 relatively high concentration (4.7 μg/g) in this sample. 2-furanmethanol is plotted closely to
304 medium crystal malt [MC] and caramalt [CA] commercial samples, which contained 553.5
305 μg/g and 403.7 μg/g respectively. In comparison, [RB, 230, 20] contained just 5.5 μg/g of 2-
306 furanmethanol.
307 It was noted that the commercially available crystal malt samples [CA] and [MC], were more
308 closely associated with higher concentrations of the green malt odour active compounds than
309 were the lab roasted green malt samples. In comparison to this, the highest roasting
310 temperatures of both pale malt ([PM, 230, 20] and [PM, 230, 30]) and raw barley ([RB, 23,
311 30] and [RB, 230, 20]) resulted in these samples being plotted outside the range of the
312 commercial samples of roasted pale malt and barley, in relation to having higher volatile
313 concentrations than the commercially available samples. These differences show a different
314 balance in volatile composition between the commercial drum roasted samples and the
315 laboratory roasted samples which doubtless reflect differences in the rates of heat transfer and
316 volatile stripping between the two techniques in addition to uncontrolled factors in the trial,
317 such as barley variety or the precise stewing conditions used for the green malt samples.

318 The biplot in Figure 1 represents a product 'flavour space' for commercial roasted products
319 generated from barley. Whilst the complexity of roasted product flavour should not be
320 underestimated, our approach of analysing the variation in 20 key odour active compounds as
321 a function of time and temperature maps the respective products according to similarity in
322 volatile composition and likewise suggests gaps where there currently are no commercial
323 products.
324
325 *3.2. Modelling Flavour Formation: Individual Models*
326 The concentration of a compound during roasting is a result of its rate of formation minus the
327 rate of its loss. Losses can be due to volatility, or through conversion to subsequent products
328 as a result of additional thermally induced reactions.
329 Of the 20 odour active compounds modelled in this study, we present full response surface
330 models for five compounds, chosen to be representative of particular thermal flavour
331 generation pathways; namely: maltol, pyrazine, 2-acetyl-5-methylfuran, 2-pentylfuran, and
332 phenylacetaldehyde. Differences in the generation of compounds across the three roasted
333 substrates will be examined. For the remaining 15 volatiles, model summary data are
334 presented in Table 2.
335 It is a visible feature of the response surface models (Figures 2-4) that the stewed roasted
336 green malt samples exhibit visibly different trends to the roasted raw barley and pale malt
337 samples. This is because green malt has higher levels of hydrolytic enzymes in the
338 endosperm. As a result of the additional stewing step, these enzymes continue to break down
339 starches and proteins. Consequently, there are dramatically different concentrations of
340 precursors to thermal flavour generation reactions in the stewed green malt which results in
341 higher concentrations of, for example, furanones (Mackie & Slaughter, 2000). In contrast, the
342 models shown in Figures 2 to 4 for the formation of the compounds in roasted raw barley and

pale malt are visibly similar in response surface shape, but with differences on the concentration axis.

3.2.1. Maltol

Maltol is formed in the intermediate stages of the Maillard reaction pathway (Vandecan et al., 2011). It has an oxygen containing heterocyclic structure, and is characterised by its sweet, jammy, baked aroma (Pittet, Rittersbacher, & Muralidhara, 1970; Scents, 2018c).

Roasted green malt samples contained the highest concentrations of maltol (from 226.1 μg/g to 972.0 μg/g), as compared with 24.7 μg/g to 175.1 μg/g for roasted pale malts and 5.1 μg/g to 100 μg/g for roasted barley samples (Figure 2a). With the 'dry roasted' (pale malt/ barley) samples it was evident that maximal levels of maltol were obtained in samples treated at the highest temperature for the longest time. This strongly suggests a pyrolytic route to maltol in addition to its production via classic Maillard chemistry; the model for raw barley clearly shows this effect at temperatures in excess of 200 °C and at longer process times. Under green malt processing conditions the model indicates that maltol formation was favoured by higher temperatures (165 °C) at the shortest process time (20 min) or for maximum concentration, lower temperature (135 °C) and the longest process time (50 min).

Maltol can be formed through a number of different pathways (e.g. from disaccharides, or from proline-amadori products) during thermal processing, which lead to its distinct concentrations in roasted products (Yaylayan & Mandeville, 1994). This is also influenced by the availability of precursors in the raw materials. Yahya et al. (2014) also showed that maltol concentrations in roasted products increased steeply during the late, high temperature-low moisture stage of roasting. This suggests, as noted here, that there are routes to maltol formation via pyrolysis in addition to Maillard reactions. An earlier study conducted by O'Shaughnessy (2003) monitored flavour formation in a range of three malts and barley in

368 commercial roasting operations. In chocolate malt, which is a highly coloured roasted pale
369 malt, the concentration of maltol increased over time, then decreased (O'Shaughnessy, 2003),
370 which is not in accordance with our studies. The maximum product temperature was 230 °C,
371 roasted for up to 97 min. The details of temperature ramping during the commercial
372 production of chocolate malt were not reported.
373
374 *3.2.2. Pyrazine*
375 Pyrazine is characterised by its pungent, roasted hazelnut, roasted barley, sweetcorn aroma
376 (Scents, 2018e). It is a nitrogen containing heterocyclic compound formed via the Maillard
377 pathways: the nitrogen coming from the amino group, and the carbon from the reducing
378 sugars that take part in the reaction pathway (Müller & Rappert, 2010). Pyrazine is typically
379 found in products that are processed to high temperatures (>180 °C) (Vandecan et al., 2011).
380 Pyrazine formation through thermally induced reactions has at least two major known
381 pathways. Firstly, the aminocarbonyl compounds produced via Strecker degradation of amino
382 acids can condense to form pyrazines. Secondly, small carbon fragments generated through
383 sugar degradation can react with ammonia generated from the pyrolysis of compounds such
384 as cysteine to produce the pyrazine ring structure. This second pathway is likely responsible
385 for the much higher production of pyrazine at 230 °C in roasted pale malt and roasted barley
386 (Figure 2b), whereas Strecker degradation reactions probably predominated in the roasted
387 green malt system where much lower levels of pyrazine were generated. Previous research
388 reported pyrazine concentration increased in speciality malts that were roasted to 180 °C
389 (Vandecan et al., 2011). The response surface models in Figure 2b show the marked increase
390 of pyrazine in roasted raw barley and pale malt after roasting temperatures exceed 200 °C
391 Roasted raw barley yielded the highest concentrations of pyrazine at the highest roasting
392 temperatures and times (Figure 6), particularly [RB, 230, 30] at 22.1 μg/g, whereas [PM, 230,

30] reached 5.6 μg/g. This supports the fact that the aroma descriptor 'roasted barley' is often assigned to pyrazine (Scents, 2018e).

Pyrazine can also be formed by heating serine or threonine in the absence of sugars (Hwang, Hartman, Rosen, & Ho, 1993).When forming pyrazine from reactions involving serine, it was found that pyrazine is formed to a higher concentration when heating under high temperature-short time conditions (300 °C for 7 min) as opposed to low temperature-long time conditions (120 °C for 4 hr) (Shu, 1999).

3.2.3. 2-acetyl-5-methylfuran

2-acetyl-5-methylfuran is characterised by its musty, nutty, hay-like, caramellic aroma (Scents, 2018a). It is an oxygen containing heterocyclic compound, known to be formed during the Maillard reaction (Nikolov & Yaylayan, 2011).

The response surface models for 2-acetyl-5-methylfuran (Figure 3a) indicate that this compound was formed at much higher concentrations in the dry roasted high temperature laboratory roasted samples. Models were remarkably similar when comparing pale malt and raw barley (Figure 3a), suggesting that the prior germination and kilning applied to pale malt had little influence on formation of this compound. The highest concentration of 2-acetyl-5-methylfuran in roasted raw barley was in [RB, 230, 30] at 4.0 μg/g, and in roasted pale malt sample [PM, 230, 30] at 3.6 μg/g.

In roasted stewed green malt, the concentration of 2-acetyl-5-methylfuran was notably lower than for the other two roasted raw materials, remaining below 1.0 μg/g in all roasted samples. Despite this, the individual factors of roasting time and temperature had a significant effect on the concentration of 2-acetyl-5-methylfuran in roasted stewed green malt ($p<0.0001$ and $p=0.0035$ respectively), as did the interaction between those two factors ($p=0.0034$).

417 The models presented in Figure 3a suggest that high roasting temperatures are required in
418 order to produce the highest levels of this compound; the green malt samples were not
419 finished at temperatures above 165°C, at which temperature concentrations in the product
420 plateaued at around 0.8 μg/g. In the roasted barley and pale malt models, 2-acetyl-5-
421 methylfuran production clearly increased steeply at process temperatures above 180 °C.
422

423 *3.2.4. 2-pentylfuran*

424 2-pentylfuran is another oxygen containing heterocyclic compound, with aroma attributes
425 including: fruity, green, earthy, beany, vegetal, and metallic (Scents, 2018b). While both
426 compounds examined in Figure 3 contain furan rings, 2-pentylfuran has a different origin to
427 that of 2-acetyl-5-methylfuran. While 2-acetyl-5methylfuran is a product of the Maillard
428 pathway, 2-pentylfuran can be formed by singlet oxygen from linoleic acid (Min, Yu, Yoo, &
429 Martin, 2005). Lipid oxidation is one of the many thermal flavour generation reactions that
430 occur during the roasting of malts and barley. The odour threshold of 2-pentylfuran is 6 ng/g
431 when found in the 'trapped' volatile headspace of dry popped corn (Buttery, Ling, & Stern,
432 1997). The concentration of 2-pentylfuran in the roasted malt and barley samples in this study
433 exceeded 6 ng/g, although the difference in sample volatile preparation should be noted. As
434 the lowest concentration of 2-pentylfuran is 0.746 μg/g in [PM, 100, 10], the aroma of 2-
435 pentlyfuran is likely to be detectable across the roasting parameters for all three roasting
436 substrates.

437 The most notable difference between Figures 3a and 3b is the higher concentration of 2-
438 pentlyfuran in roasted pale malt, with the highest concentration in [PM, 230, 30] at 12.4 μg/g.
439 Germination and kilning may have influenced the availability of linoleic acid as a precursor
440 to 2-penylfuran in the pale malt as a roasting substrate.
441

3.2.5. Phenylacetaldehyde

Phenylacetaldehyde has a floral, honey, green, cocoa, sweet aroma (Scents, 2018d). It is a Strecker aldehyde formed in thermally treated foodstuffs through the Strecker degradation of phenylalanine (Channell, Yahya, & Cook, 2010; Farmer, 1994; Rizzi, 1999; Smit, Engels, & Smit, 2009). Strecker degradation reactions require dicarbonyl compounds in addition to an amino acid. Small and reactive dicarbonyl compounds are generated from sugar degradation reactions, which may result from either Maillard chemistry or caramelisation reactions. The gross trends in phenylacetaldehyde production across the roasted substrates (Figure 4) indicate that much higher levels were generated in the roasted green malt products. This overall trend likely results from a combination of i) enhanced Maillard reactivity brought about by the stewing process in roasted green malt production and ii) the lower losses due to volatilisation of phenylacetaldehyde at the lower green malt finishing temperatures.

Figure 4 shows that the models for phenylacetaldehyde formation in roasted samples of raw barley and pale malt share similarities, but with differences in concentration. The highest concentrations were found in the samples roasted at the lowest temperatures ([PM, 100, 30] at 24 μg/g, for example). As the roasting temperature increased, the concentration of phenylacetaldehyde initially dropped ([PM, 200, 25] at 3.5 μg/g), then increased again at the very highest roasting temperatures ([PM, 230, 30] at 9.6 μg/g). This trend suggests that in very dry, high temperature roasted systems another pathway to phenylacetaldehyde might exist; for example via pyrolysis of phenylalanine as opposed to Strecker degradation. Naturally, this is not proven by the current experiments, but the shapes of the models for both raw barley and pale malt are consistent with there being a secondary route to the production of phenylacetaldehyde at high roasting temperatures. This trend was not seen in the roasted stewed green malt samples whereby treatments did not include a high enough roasting temperatures to exhibit the final increase of phenylacetaldehyde.

4. Conclusions

Understanding the formation of flavour during roasting is an essential step in product development and quality control, as this information may be used by maltsters to engineer roasted products with specific desirable characteristics.

Modelling key odour active compound formation over a range of roasting times, temperatures and substrates has developed better understanding of how the substrate and roasting conditions combine to generate the volatile aroma composition of roasted malt and barley products. This study also compared laboratory-roasted samples to commercial samples in terms of their concentrations of 20 key odour active compounds. The PCA plot of the resulting data (Figure1) depicts a 'flavour space' for roasted products produced from these three substrates, indicating how control of time, temperature and initial moisture content during roasting determined product volatile aroma characteristics. Compounds such as maltol, phenylacetaldehyde, 2-furanmethanol, HMF and acetic acid were formed at highest concentration in roasted green malt products, indicating greater formation via Maillard chemistry in the liquid phase and/or lower losses of volatiles at the more moderate green malt roasting temperatures. In contrast, odour-active compounds such as pyrazines, pyrroles, pyridines, 2-methylfuran, 2-pentylfuran, methyl-2-furoate and 2-acetyl-5-methylfuran were predominantly formed in the 'dry roasted' products, indicative of greater formation via Maillard chemistry in the solid phase or pyrolysis at higher temperatures and very low moisture contents.

The development of response surface models for the formation of each of the 20 compounds as a function of time and temperature in each roasting substrate clearly demonstrated the complexity of thermal flavour generation which results from factors such as there being multiple pathways to individual compounds which have different activation energies/ temperature ranges at which they become active. Furthermore, differences in compound

492 volatility and the potential for onwards thermal reactions in some cases further complicate the
493 form of models. Thus the predictive power of cubic models fitted to some compound
494 concentration data was still low and some models had a significant lack of fit, indicating that
495 the trends in data across the design space were too complex to accurately model without
496 'over-fitting' the data (e.g. pentanal, hexanal, 2-furanmethanol, 2-formylpyrrole,
497 hydroxymethylfurfural). Several of the models derived indicated that concentrations of
498 volatile compounds increased steeply at very high temperature (>180 °C) under low moisture
499 conditions. Since these conditions prevail at the end of pale malt or roasted barley production
500 this indicates how difficult precise flavour control is for these product-types. Traditionally
501 colour is used as the yardstick for process control, but brewers recognise that there can be
502 substantial differences in flavour attained from different batches of the same roasted product.
503 Arguably both hypotheses of the current study were confirmed. It is apparent from Figure 1
504 that roasting conditions could be manipulated to deliver flavour chemistries which either
505 extend the current product portfolio or which are substantially different to the existing
506 commercial products. Furthermore, enhanced understanding of the links between processing
507 conditions and flavour formation highlight the rapidly changing flavour profile which
508 prevails towards the end of commercial roasting operations. This highlights a need for better
509 process control systems, if roasted malts are to be controlled in terms of their flavour
510 properties as well as their colour.
511

512 # CRediT authorship contribution statement

513 **Hebe Parr**: PhD student. Conducted all research and formal analysis in this manuscript.
514 Writing – original draft.
515 **Irina Bolat**: Conceptualisation and input to design of investigation. Writing – review and
516 editing of manuscript.

David Cook: Funding acquisition, supervision of PhD, conceptualisation and input to design of study and writing – review and editing of manuscript.

Declaration of interests

☒ The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

☒The authors declare the following financial interests/personal relationships which may be considered as potential competing interests:

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Figures and Tables

Table 1 - Concentrations (μg/g) of 20 odour active compounds in laboratory roasted samples of raw barley, green malt, and pale malt and in six commerical roasted samples: amber malt (AM), caramalt (CA), medium crystal malt (MC), chocolate malt (CH), black malt (BL), and roasted barley (RB). Final moisture content of the roasted samples is reported (n=3).

	Temperature (°C)	Time (Min)	Final Moisture (% w/w)	2-methylfuran	Pentanal	Hexanal	1-methylpyrrole	Pyrazine	2-pentylfuran	2,3-dimethylpyrazine	Furfural	Acetic Acid	2-n-pentylpyridine	Methyl-2-furoate	5-methylfurfural	2-acetyl-5-methylfuran	Phenylacetaldehyde	2-furanmethanol	2-(5H)-furanone	Maltol	Furaneol	2-formylpyrrole	Hydroxymethylfurfural
				LRI ZB-WAX[A]																			
				894	1003	1107	1167	1239	1254	1373	1498	1534	1609	1615	1615	1656	1688	1697	1798	2019	2081	2081	2558
Raw Barley	135	25	4.41	0	0.0438	0.0839	0.0567	0.626	0.987	0.561	6.46	91.5	0	0.188	2.37	0.0554	6.93	2.46	1.76	6.50	1.86	2.50	85.7
	135	30	3.72	0	0.0410	0.0792	0.0530	0.576	1.07	0.519	6.06	95.0	0	0.129	2.41	0.0803	6.64	2.24	1.96	6.46	1.39	1.24	82.1
	165	10	4.69	0	0.0355	0.0759	0.0628	0.526	1.05	0.663	5.10	93.4	0	0.242	2.38	0.0891	6.23	2.49	3.17	7.21	2.43	2.67	73.5
	165	20	2.60	0	0.0335	0.0690	0.0594	0.564	1.13	1.32	6.76	115	0.140	0.261	2.77	0.112	2.79	3.48	3.37	7.62	4.36	3.17	72.7
	165	30	2.02	0.129	0.0357	0.0779	0.0561	1.00	1.22	2.02	13.1	130	0.201	0.241	4.13	0.144	1.87	5.25	3.36	7.02	4.14	4.34	71.9
	200	10	1.55	0.201	0.0325	0.0835	0.0547	1.96	1.06	2.84	23.5	137	0.433	0.384	6.40	0.247	2.84	8.29	4.04	8.82	5.73	4.88	93.9
	200	15	0.838	0.756	0.0424	0.0979	0.0418	3.08	1.09	2.37	76.7	148	0.276	0.996	27.4	0.336	1.92	16.8	9.25	11.6	5.79	19.1	164
	200	20	0.578	1.21	0.0512	0.106	0.0431	3.21	0.955	2.48	83.1	156	0.577	1.63	36.6	0.541	2.16	12.8	10.9	14.8	5.03	34.1	142
	200	25	0.307	1.88	0.0435	0.102	0.0351	3.34	1.46	2.84	80.1	127	1.33	2.16	26.5	0.757	1.72	9.05	7.34	13.7	3.08	37.9	104
	230	10	0.297	2.62	0.0415	0.104	0.0467	5.89	1.10	3.63	96.8	142	0.564	2.58	32.3	0.946	2.52	12.9	10.4	14.2	5.00	41.1	98.2
	230	20	0.117	7.45	0.0445	0.0888	0.101	9.84	1.99	4.73	78.5	135	4.47	7.93	24.3	3.02	6.49	5.52	12.9	46.0	3.34	38.5	56.1
	230	30	0.107	10.0	0.0472	0.103	0.176	21.5	3.09	6.84	84.3	119	11.4	10.1	37.6	3.79	8.79	4.58	12.8	93.8	3.82	22.2	89.1
Green	135	50	4.64	0.748	0.0665	0.0976	0.374	0.347	1.47	0.435	127	464	0	2.69	38.8	0.778	35.1	360	76.7	613	18.8	5.08	897

	143	43	4.06	0.625	0.0665	0.0879	0.272	0.393	1.32	0.537	123	406	0	2.97	33.8	0.715	23.7	265	71.2	513	16.5	4.39	839
	150	35	4.14	1.68	0.0776	0.0945	0.294	0.452	1.32	0.457	122	392	0	3.07	34.3	0.730	22.7	280	64.1	524	17.0	5.12	865
	150	50	2.33	0.598	0.0590	0.112	0.215	0.430	1.54	0.409	118	425	0	1.93	42.2	0.825	9.99	233	56.0	614	14.0	6.51	1006
	158	28	4.11	0.619	0.0707	0.0883	0.295	0.529	1.32	0.525	113	348	0	2.89	31.1	0.695	21.7	251	63.0	466	16.3	4.56	771
	158	43	2.02	0.606	0.0781	0.129	0.198	0.574	1.94	0.455	96.1	369	0	3.42	46.1	0.798	9.12	184	48.4	546	13.2	7.58	915
	165	35	2.07	0.671	0.115	0.134	0.255	0.605	2.15	0.620	114	451	0	3.34	62.0	1.08	11.4	217	53.5	656	14.1	8.72	945
	165	50	1.31	0.577	0.0535	0.129	0.111	1.01	1.62	1.36	174	233	0	4.24	49.0	0.791	10.9	75.2	47.0	295	8.57	6.07	747
Pale Malt	100	10	2.80	0.221	0.0435	0.129	0.161	1.73	0.746	2.32	26.0	255	0	1.14	10.9	0.181	20.4	12.0	20.0	27.4	8.02	3.27	349
	100	20	2.45	0.246	0.0649	0.132	0.180	1.96	0.875	1.76	26.7	235	0.199	0.486	9.88	0.192	23.1	10.2	19.9	30.0	7.44	4.16	318
	100	30	2.38	0.240	0.0682	0.141	0.170	1.76	0.856	1.85	29.4	253	0.0780	1.02	10.7	0.193	22.1	11.7	22.0	28.1	8.51	3.40	362
	135	10	2.23	0.253	0.0430	0.158	0.163	1.75	1.06	2.15	29.0	220	0.421	0.806	9.32	0.168	23.1	9.84	21.5	31.9	11.2	3.62	386
	135	15	1.99	0.235	0.0737	0.158	0.233	1.18	1.10	1.03	27.6	279	0.206	0.495	11.1	0.230	17.5	11.8	23.5	48.7	10.4	5.78	333
	135	25	1.60	0.263	0.0555	0.140	0.214	1.04	1.46	0.863	28.0	293	0	0.532	12.4	0.252	13.1	14.3	25.9	56.5	11.3	7.21	358
	135	30	1.39	0.235	0.0551	0.129	0.192	1.20	1.47	1.17	27.7	261	0.231	0.824	11.6	0.252	12.5	12.2	22.4	50.4	10.9	5.39	380
	165	10	0.779	0.379	0.0743	0.157	0.259	0.901	1.89	0.883	33.6	340	0.304	0.783	18.3	0.435	8.58	17.2	28.6	80.2	15.7	7.39	421
	165	20	0.674	0.424	0.0713	0.194	0.216	0.946	2.78	1.17	53.4	317	0.651	1.17	27.2	0.459	7.58	20.1	29.7	79.9	16.3	8.93	507
	165	30	0.225	0.545	0.0448	0.203	0.171	0.977	3.42	1.29	77.7	225	0.928	1.10	30.1	0.397	6.45	17.2	20.3	62.3	11.0	7.13	491
	200	10	0.301	0.998	0.0387	0.166	0.138	1.63	2.63	1.56	174	243	1.80	2.62	97.0	0.924	7.47	28.0	12.6	58.2	12.0	13.0	780
	200	15	0.119	3.08	0.0761	0.268	0.119	1.79	5.18	0.808	162	252	2.42	2.17	136	1.04	7.25	18.8	17.5	86.3	8.68	30.9	466
	200	20	0.023	4.73	0.198	0.431	0.111	2.21	6.47	1.02	166	268	5.60	2.89	148	1.28	9.76	14.1	22.2	107	9.34	43.4	458
	200	25	0.022	3.39	0.0679	0.263	0.0631	1.53	6.16	0.912	116	119	4.58	2.71	60.4	0.942	3.52	5.59	10.0	54.2	3.56	27.5	181
	230	10	0.077	5.79	0.0848	0.205	0.107	2.33	3.49	0.804	179	148	3.12	3.69	70.5	1.34	6.28	6.90	10.5	62.2	5.82	41.4	180

	230	20	0.045	11.2	0.102	0.345	0.152	4.34	9.96	0.904	225	188	14.8	9.99	108	3.22	11.3	7.67	17.7	138	5.83	44.0	196
	230	30	0.103	8.57	0.0742	0.336	0.124	5.14	11.0	1.56	177	114	22.8	10.5	86.9	3.45	11.5	3.25	8.53	157	5.21	21.4	212
Commercial	AM		1.67	0.300	0.640	0.520	0.360	1.44	2.86	0.880	62.8	366	0.780	1.00	14.1	0.300	25.6	60.1	15.1	177	6.94	4.80	672
	CA		5.06	0.620	0.960	1.26	0.240	1.02	3.28	1.02	281	377	12.8	3.62	34.6	0.600	56.5	404	46.2	730	9.20	4.00	1944
	MC		2.84	0.940	1.10	0.340	0.200	0.580	1.40	0.600	359	565	0	2.70	43.7	0.900	30.6	554	61.1	1021	14.2	5.72	2423
	CH		2.98	0.120	0.440	0.440	0.120	7.98	0.800	2.36	146	128	11.6	14.7	26.1	2.16	12.1	3.74	16.5	246	3.30	28.4	230
	BL		1.97	1.44	0.380	0.240	0.120	8.00	5.44	3.10	200	107	17.2	14.6	45.4	2.58	11.4	5.36	17.4	293	4.44	28.9	350
	RB		2.12	0.220	0.580	2.04	0.260	27.0	2.58	5.12	156	186	5.46	17.5	45.3	2.40	12.3	5.98	14.1	427	4.26	32.3	237

604 [A] Linear retention index against alkanes (C8-C22) on a ZB-WAX column.

Table 2 – Model fit data for the predictive models for compound concentration in roasted raw barley (n=24), green malt (n=24), and pale malt (n=24). Factor A (roasting temperature (°C)) and Factor B (roasting time (Min)). Table 2a: Compounds 1-10, Table 2b: Compounds 11-20)

2a

		2-methylfuran	Pentanal	Hexanal	1-methylpyrrole	Pyrazine	2-pentylfuran	2,3-dimethylpyrazine	Furfural	Acetic Acid	2-n-pentylpyridine
Raw Barley	Model	Cubic	Quadratic	Quadratic	Quadratic	Cubic	Cubic	Quadratic	Quadratic	Cubic	Cubic
	Model p Value	<0.0001±	0.0017±	0.0012^	0.0003±	<0.0001^	<0.0001±	<0.0001±	<0.0001^	<0.0001±	<0.0001±
	Model R^2	**0.991**	**0.581**	**0.475**	**0.656**	**0.992**	**0.968**	**0.973**	**0.872**	**0.947**	**0.940**
	A p Value	0.2671	0.0368	0.7562	0.0154	0.2383	0.3116	<0.0001	<0.0001	<0.0001	0.8214
	B p Value	0.3939	0.7774	-	0.0237	0.5233	0.1641	<0.0001	-	0.3312	0.9817
	AB p Value	<0.0001	-	-	0.0038	<0.0001	<0.0001	<0.0001	-	0.0377	<0.0001
	A^2 p Value	<0.0001	0.0002	0.0003	0.0692	<0.0001	0.0043	<0.0001	0.0002	0.1315	<0.0001
	B^2 p Value	-	0.0180	-	-	0.0155	-	-	-	0.4463	-
	A^2B p Value	<0.0001	-	-	-	<0.0001	0.0007	-	-	0.0007	0.0013
	AB^2 p Value	-	-	-	-	0.0017	-	-	-	-	-
	A^3 p Value	<0.0001	-	-	-	0.0012	0.0029	-	-	<0.0001	0.0365
	B^3 p Value	-	-	-	-	-	-	-	-	0.0378	-
Green Malt	Model	Mean	Linear	Cubic	2Fl	Quadratic	2Fl	Quadratic	Linear	Quadratic	NF
	Model p Value	-	0.0008±	<0.0001±	<0.0001±	<0.0001±	<0.0001±	0.0124±	0.004±	0.0002±	-
	Model R^2	**0**	**0.526**	**0.851**	**0.778**	**0.934**	**0.784**	**0.445**	**0.346**	**0.714**	**-**
	A p Value	-	0.0069	0.0036	<0.0001	0.2748	0.0001	0.2124	-	0.0053	-
	B p Value	-	0.0016	<0.0001	0.0002	<0.0001	<0.0001	0.0260	0.0040	0.2869	-
	AB p Value	-	-	0.0684	0.0012	<0.0001	0.0341	-	-	<0.0001	-
	A^2 p Value	-	-	0.4294	-	0.0284	-	0.0251	-	-	-
	B^2 p Value	-	-	-	-	0.0039	-	-	-	-	-
	A^2B p Value	-	-	-	-	-	-	-	-	-	-
	AB^2 p Value	-	-	-	-	-	-	-	-	-	-
	A^3 p Value	-	-	0.0136	-	-	-	-	-	-	-
	B^3 p Value	-	-	-	-	-	-	-	-	-	-
Pale Malt	Model	Cubic	Linear	Quadratic	Quadratic	Cubic	Cubic	Quadratic	Quadratic	Quadratic	Cubic
	Model p Value	<0.0001±	0.0322±	<0.0001±	0.006^	<0.0001^	<0.0001±	0.0008±	<0.0001^	0.0002±	<0.0001^
	Model R^2	**0.979**	**0.210**	**0.873**	**0.411**	**0.963**	**0.991**	**0.598**	**0.917**	**0.583**	**0.982**
	A p Value	0.0029	0.0322	<0.0001	0.0185	0.6930	<0.0001	0.0001	<0.0001	0.0019	0.0231

	B p Value	0.0071	-	0.0046	-	0.2566	0.0040	-	-	-	0.6045
	AB p Value	0.0010	-	0.0064	-	<0.0001	<0.0001	-	-	-	<0.0001
	A^2 p Value	<0.0001	-	0.0337	0.0218	<0.0001	<0.0001	-	0.0003	0.0020	<0.0001
	B^2 p Value	0.0112	-	0.0810	-	-	0.0140	-	-	-	-
	A^2B p Value	-	-	-	-	0.0006	<0.0001	-	-	-	<0.0001
	AB^2 p Value	0.0010	-	-	-	-	0.0066	-	-	-	-
	A^3 p Value	0.0009	-	-	-	0.0148	-	-	-	-	0.0076
	B^3 p Value	-	-	-	-	-	-	-	-	-	-

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2b

		Methyl-2-furoate	**5-methylfurfural**	**2-acetyl-5-methylfuran**	**Phenylacetaldehyde**	**2-furanmethanol**	**2-(5H)-furanone**	**Maltol**	**Furaneol**	**2-formylpyrrole**	**Hydroxymethylfurfural**
Raw Barley	Model	Cubic	Quadratic	Cubic	Cubic	Cubic	Quadratic	Cubic	Cubic	Quadratic	Cubic
	Model p Value	<0.0001±	<0.0001^	<0.0001^	<0.0001^	<0.0001^	<0.0001±	<0.0001±	<0.0001±	<0.0001^	>0.05^
	Model R^2	**0.986**	**0.840**	**0.986**	**0.966**	**0.751**	**0.940**	**0.985**	**0.786**	**0.778**	**0.383**
	A p Value	0.2190	<0.0001	0.4927	<0.0001	0.0002	<0.0001	0.8329	<0.0001	<0.0001	0.0164
	B p Value	0.5526	-	0.9021	<0.0001	0.0785	0.0465	0.8954	-	0.4496	-
	AB p Value	<0.0001	-	<0.0001	<0.0001	0.0193	-	<0.0001	-	-	-
	A^2 p Value	<0.0001	0.0006	<0.0001	<0.0001	0.9920	<0.0001	<0.0001	0.0221	0.0036	-
	B^2 p Value	-	-	-	-	-	0.0342	0.3208	-	-	
	A^2B p Value	<0.0001	-	<0.0001	<0.0001	-	-	<0.0001	-	-	-
	AB^2 p Value	-	-	-	-	-	-	0.0419	-	-	-
	A^3 p Value	<0.0001	-	<0.0001	<0.0001	0.0071	-	0.0044	0.0041	-	0.0299
	B^3 p Value	-	-	-	-	-	-	-	-	-	-
Green Malt	Model	Linear	Linear	Quadratic	Cubic	Cubic	Quadratic	Quadratic	Cubic	Linear	Quadratic
	Model p Value	>0.05±	0.0003±	<0.0001±	<0.0001±	<0.0001±	0.0005±	<0.0001±	<0.0001±	<0.0001±	0.0003±
	Model R^2	**0.127**	**0.568**	**0.856**	**0.969**	**0.926**	**0.674**	**0.774**	**0.883**	**0.747**	**0.695**
	A p Value	-	0.0624	0.0035	<0.0001	0.2945	0.0260	0.1520	0.6015	0.0003	0.9439
	B p Value	-	0.0001	<0.0001	<0.0001	0.4678	0.1145	0.0021	0.0681	<0.0001	0.0001
	AB p Value	-	-	0.0034	0.0013	<0.0001	0.0001	<0.0001	<0.0001	-	0.0274
	A^2 p Value	-	-	-	0.0018	0.5713	-	0.0199	0.8802	-	-
	B^2 p Value	-	-	-	0.0646	0.0061	-	-	0.0440	-	0.0379
	A^2B p Value	-	-	-	-	-	-	-	-	-	-

	AB^2 p Value	-	-	-	0.0030	<0.0001	-	-	0.0013	-	-
	A^3 p Value	-	-	-	-	0.0107	-	-	0.0494	-	-
	B^3 p Value	-	-	-	-	-	-	-	-	-	-
Pale Malt	Model	Cubic	Cubic	Cubic	Cubic	Quadratic	Quadratic	Cubic	Quadratic	Quadratic	Cubic
	Model p Value	<0.0001^	<0.0001^	<0.0001±	<0.0001±	0.0044±	0.0002±	<0.0001±	<0.0001±	<0.0001±	0.0027^
	Model R^2	**0.959**	**0.868**	**0.949**	**0.928**	**0.436**	**0.586**	**0.862**	**0.629**	**0.892**	**0.536**
	A p Value	0.0150	<0.0001	<0.0001	<0.0001	0.3153	0.0014	<0.0001	0.0682	<0.0001	0.0926
	B p Value	0.8615	0.0442	0.9135	0.0030	-	-	0.9895	-	0.2048	-
	AB p Value	0.0009	-	0.0001	0.3395	-	-	0.0015	-	0.0363	-
	A^2 p Value	<0.0001	0.1520	<0.0001	<0.0001	0.0017	0.0024	0.5946	<0.0001	0.0009	0.0012
	B^2 p Value	0.1413	0.5607	-	-	-	-	-	-	0.0379	-
	A^2B p Value	0.0258	-	0.0140	0.0056	-	-	0.0314	-	-	-
	AB^2 p Value	0.0059	-	-	-	-	-	-	-	-	-
	A^3 p Value	0.0265	0.0080	-	0.0111	-	-	-	-	-	0.0355
	B^3 p Value	-	0.0386	-	-	-	-	-	-	-	-

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612 ± Non-significant lack of fit for model.

613 ^ Significant lack of fit for model.

Figure 1 – PCA bi-plot of 20 odour active compounds and the concentrations in the roasted samples. Lab roasted samples are pale malt (PM), green malt (GM), and raw barley (RB) followed by roasting temperature (°C), and roasting time (min). Commercial roasted malt samples are: Roasted Barley [RB], Black malt [BL], Chocolate Malt [CH], Medium Crystal Malt [MC], Caramalt [CA], and Amber Malt [AM]. Samples plotted are <5% moisture.

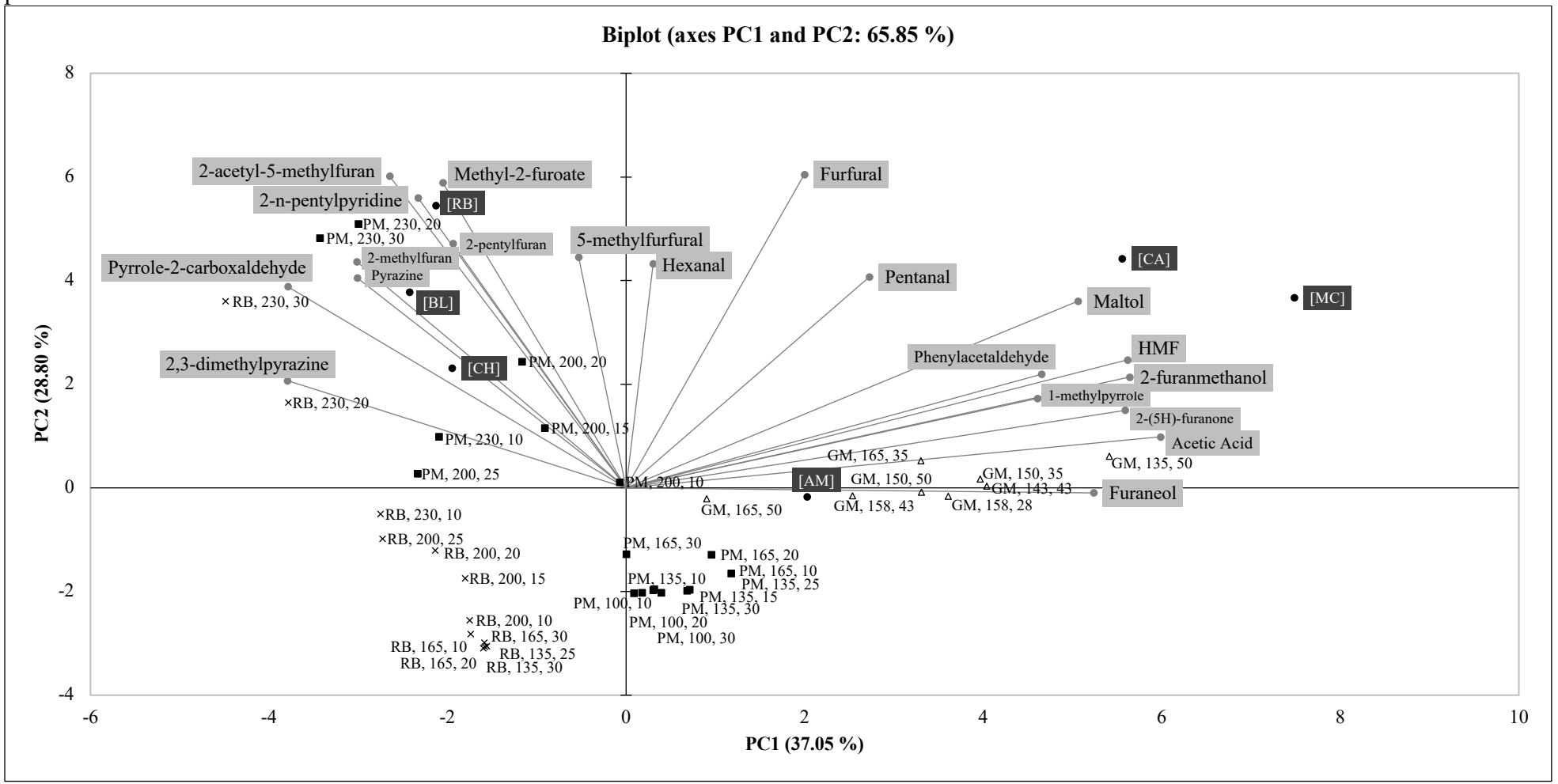

Figure 2 – The concentrations of a) maltol (µg/g) and b) pyrazine (µg/g) modelled as a function of roasting time (min) and temperature (°C) for three different substrates: raw barley, green malt, and pale malt.

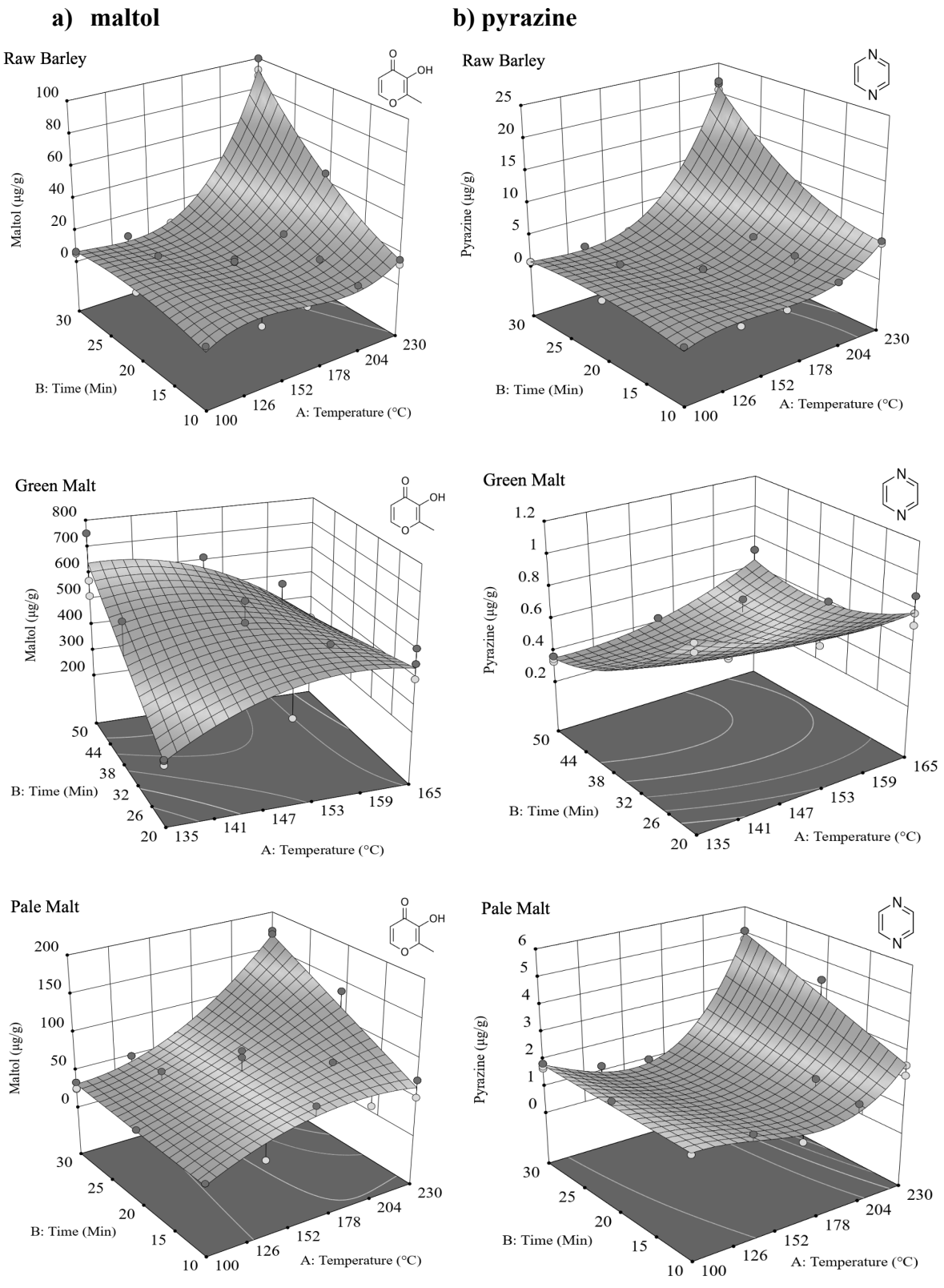

Figure 3 - The concentrations of a) 2-acteyl-5-methylfuran (µg/g) and b) 2-pentylfuran (µg/g) modelled as a function of roasting time (min) and temperature (°C) for three different substrates: raw barley, green malt, and pale malt.

a) 2-acetyl-5-methylfuran **b) 2-pentylfuran**

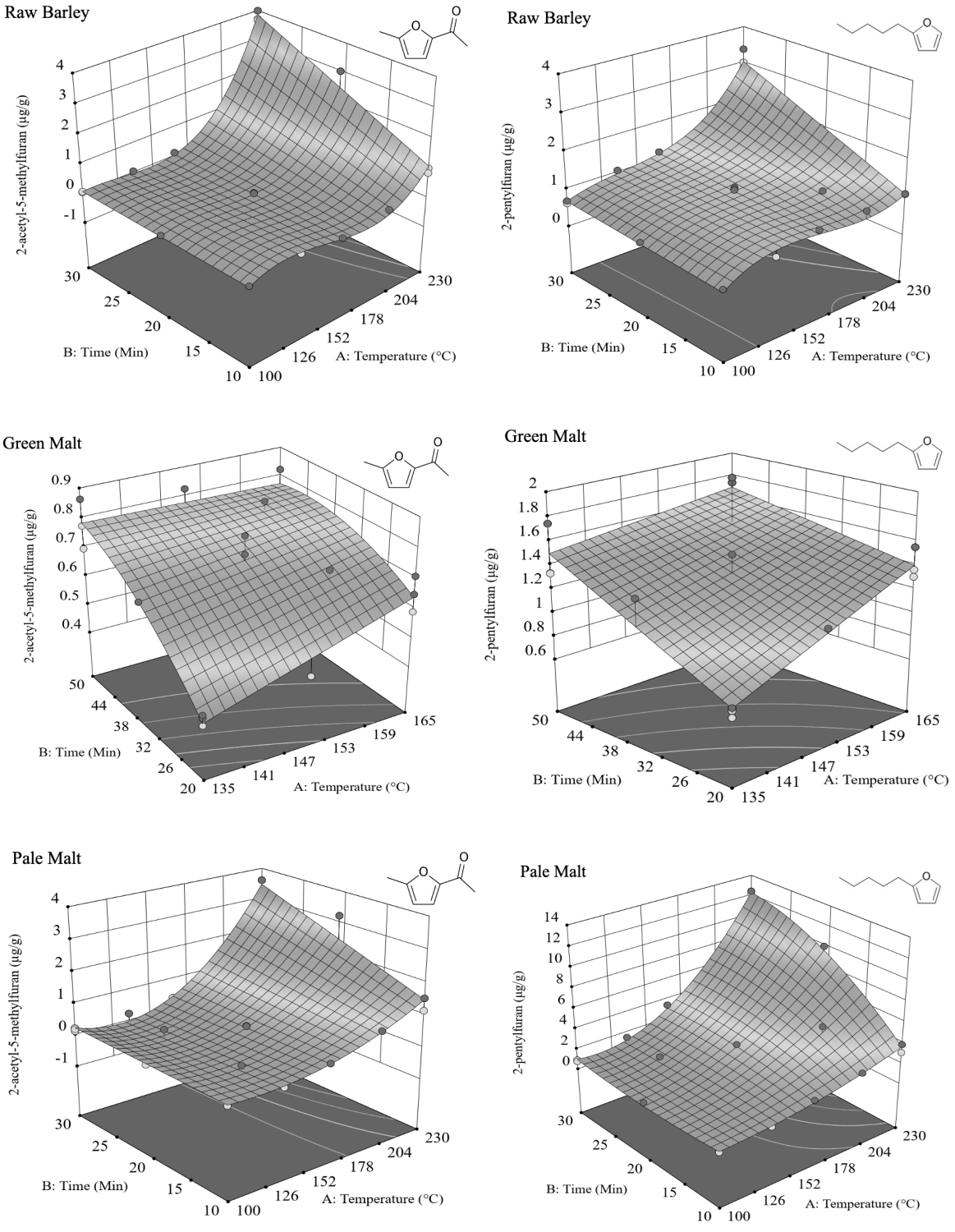

Figure 4 – The concentrations of phenylacetaldehyde (μg/g) modelled as a function of roasting time (min) and temperature (°C) for three different substrates: raw barley, green malt, and pale malt.