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The aroma profile of wheat bread crumb influenced by yeast concentration and fermentation temperature

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ABSTRACT

The consumers of today have an increasing interest in high quality bread with appealing aroma. The scope of this work is to investigate how aroma in wheat bread crumb is influenced by different fermentation conditions: amount of yeast (20, 40 and 60 g/kg flour) and fermentation temperature (5, 15 and 35 °C). Dough samples were fermented to equal height and baked, and the aroma compounds from the bread were extracted by dynamic headspace extraction and analyzed by gas chromatography–mass spectrometry. Quantification of the aroma compounds was performed by multiple headspace extraction. The most aroma active compounds identified were 3-methylbutanal, (E)-2-nonenal, 3-methyl-1-butanol, and 2,3-butanedione. Increasing the yeast concentration was found to increase formation of the majority of the compounds formed from the yeast metabolism, with 2,3-butanedione and phenylacetaldehyde as the most aroma active compounds. High fermentation temperature (15 and 35 °C) increased formation of many lipid oxidation compounds, with hexanal and heptanal having the highest odor activity values. Low fermentation temperature (5 °C) was found to increase formation of the three esters ethyl acetate, ethyl hexanoate, and ethyl octanoate, with ethyl hexanoate having the highest odor activity value. The odor activity values of the esters were generally low.

1. Introduction

Today, industrially produced wheat bread is often made from dough with a high concentration of yeast and the dough is fermented at high temperature in order to decrease the production time (Cauvain, 1998a). A short fermentation time results in an economical benefit and a continuous flow on the production line. However, it is generally believed that a longer fermentation time results in bread with a more pleasant aroma, although only few studies have been done in this area (Hironaka, 1986; Maeda et al., 2009). Aroma development in bread crumb has been found to be dependent on yeast concentration, mixing stage and fermentation time (Frasse et al., 1992; Gassenmeier & Schieberle, 1995; Maeda et al., 2009; Richard-Molard, Nago, & Drapron, 1979; Schieberle & Grosch, 1991). Fermentation temperature was found to have a significant effect on the crust aroma of baguettes (Zehentbauer & Grosch, 1998). The influence of fermentation temperature on the crumb aroma has only been investigated for the two fermentation products 3-methyl-1-butanol and 2-phenylethanol (Gassenmeier & Schieberle, 1995). Aroma compounds identified in fermented bread crumb are mainly derived from the metabolism of yeast and from the oxidation of flour lipids (Frasse et al., 1992; Schieberle & Grosch, 1991), whereas the aroma compounds in the crust originates from Maillard reactions occurring at high temperatures and low water activity between reducing sugars and amino acids (Purlis, 2010). Dynamic headspace extraction (DHE) followed by gas chromatography–mass spectrometry (GC–MS) has proven to be a sensitive and fast method to analyze the aroma compounds in bread or bread-like systems (Christensen, Leitao, Petersen, Jespersen, & Engelsen, 2009; Luning, Roozen, Moëst, & Posthumus, 1991). Quantification of aroma compounds by multiple headspace extraction (MHE) developed by Kolb and Pospisil (1977) is an effective quantification method, since it is independent of the food matrix. MHE has recently been successfully performed within food systems (Carrillo & Tena, 2006; Soria, Martinez-Castro, & Sanz, 2007). Numerous papers have elucidated the effects of yeast and fermentation temperature on aroma development during alcoholic fermentation in wine and beer production (Molina, Swiegers, Varela, Pretorius, & Agosin, 2007; Saerens, Verbelen, Vanbeneden, Thevelein, & Delvaux, 2008;

Abbreviations: AACC, American Association of Cereal Chemists; ANOVA, analysis of variance; DHE, dynamic headspace extraction; GC–MS, gas chromatography–mass spectrometry; ICC, International Association for Cereal Science and Technology; MHE, multiple headspace extraction; PCA, principal component analysis; PLS, partial least squares; OAV, odor activity value; OT, odor threshold.

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Tominac et al., 2008; Trelea, Titica, & Corrieu, 2004). However, knowledge within aroma development in bread fermented at different yeast concentrations and fermentation temperatures is lacking. Information on how these different fermentation conditions influence the aroma in bread crumb is of interest for the bread industry in order to improve the aroma of the bread products. The purpose of this research paper is therefore to investigate how different yeast concentrations (20, 40 and 60 g/kg flour) and dough fermentation temperatures (5, 15 and 35 °C) influence the aroma profile of wheat bread crumb. The aroma compounds were extracted by DHE followed by GC–MS analysis, and the quantification of the identified aroma compounds was performed by MHE.

2. Material and methods

2.1. Flour

Wheat flour (Reform) was supplied by Lantmännen Mills A/S (Vejle, Denmark). Moisture content of the flour was measured the day of baking (HOH-express, Pfeuffer) and varied from 11.7 to 11.9%. The gluten content was 28.9% (wet gluten) and the gluten index was 95 (Glutomatic 2100, Perten) according to the American Association of Cereal Chemists (AACC) international approved method no. 38-12 (AACC, 1995). The following Farinograph dough parameters were measured: the dough development time, 3 min; the dough stability, $1^3/_4$ min; the degree of softening, 20 BU; and the water uptake was 54.8% (Farinograph DCE 330, Brabender) according to AACC international approved method no. 54-21 (AACC, 1995). The falling number was 299 s (Falling number 1500, Perten) according to ICC standard method no. 107/1 (ICC, 1995).

2.2. Yeast

Commercial pressed baker's yeast (*Saccharomyces cerevisiae*, Malteserkors from Lallemand, De Danske Gærfabrikker, Grenå, Denmark) was used without washing the yeast cells. The dry matter was 28%. The cell count was $1.44*10^{13}$ yeast cells/g yeast and $2.0*10^9$ lactic acid bacteria/g yeast. Preliminary tests showed that the yeast cell density was the same in the dough immediately after mixing of the dough and in the end of the fermentation period. The yeast was taken from the same batch to decrease the risk of different cell count of yeast and different contaminant bacteria.

2.3. Bread making

300 g of Flour (adjusted to 14% moisture content), 190 mL water (30 °C), 4 g saccharose, 4 g NaCl and 6, 12 and 18 g yeast (corresponding to 20, 40 and 60 g yeast/kg flour), respectively were mixed in a baking machine (FAB-100 from Funai) for 19 min. Two pieces of 235 g dough was each transferred to 1 L beaker glass. The beaker glasses were sealed with aluminum foil and the doughs were left for fermentation at 5, 15 or 35 °C, respectively in an incubator. The fermentations were terminated when the doughs reached 8 cm in total height. Preliminary tests were made at each yeast level and fermentation temperature to find the right fermentation time as the dough height was monitored by a web-camera. The fermentation times are presented in Table 1. The dough was baked at 130 °C for 31 min in a convection oven with steam (Conmatic line, Hounö) to a center temperature of 98 °C. The relatively low baking temperature and steam during baking were chosen to decrease crust formation. The bread was cooled for 15 min and was then released from the beaker glass and further cooled at room temperature for 1 h on a grate. 1 cm crust was quickly removed and samples of 15 g of bread crumb were cut into 4–6 pieces and packed in tin foil surrounded by a plastic bag.

Table 1
Fermentation times for the dough to reach 8 cm in height according to fermentation temperature and yeast concentration.

Fermentation temperature (°C)	Yeast concentration (g/kg flour)		
	20	40	60
5	21 h	3 h 30 min	3 h
15	3 h	1 h	35 min
35	50 min	25 min	15 min

Approximately six samples could be taken from each bread. The samples were frozen at −18 °C until aroma analysis within few weeks. A preliminary test was made for comparison of the aroma profile of frozen and fresh bread samples. The test showed no significant differences between the treatments (data not shown), therefore freezing of the samples were done for practical reasons.

2.4. Standards

1-Penten-3-ol, 3-methyl-1-butanol, 3-methyl-3-buten-1-ol, 1-pentanol, 2-penten-1-ol, 1-hexanol, 3-hexenol, 2-ethyl-1-hexanol, 1-octanol, 3-nonen-1-ol, 2-phenylethanol, 1-dodecanol, 3-methylbutanal, heptanal, octanal, nonanal, decanal, benzaldehyde, (E)-2-nonenal, phenylacetaldehyde, 2-heptanone, 3-hydroxy-2-butanone, 2-octanone, 3-octen-2-one, ethyl acetate, ethyl octanoate, gamma-nonalactone, acetic acid, butanoic acid, hexanoic acid, 2-pentylfuran and trimethylpyrazine were purchased from Sigma–Aldrich (Gillingham, U.K.). 2-Methyl-1-propanol, 1-octen-3-ol, hexanal, 2,3-butanedione, 2-methylpropanoic acid and pentanoic acid were purchased from Fluka (Buchs, Switzerland). 1-Propanol, 1-heptanol, 2-furancarboxaldehyde, ethyl 3-methylbutanoate and ethyl hexanoate were purchased from Merck (Darmstadt, Germany). 1-Butanol was purchased from Ferak (Berlin, Germany) and phenylethyl acetate was purchased from ICN Pharmaceuticals, Inc (Costa Mesa, U.S.).

2.5. Dynamic headspace extraction (DHE)

15 g of Frozen bread crumb in pieces of 4–6 was placed in a 500 mL glass flask (7.5 cm in diameter). A trap containing Tenax-TA (200 mg) was attached to the sealed flask. The flask containing the sample was immersed in a laboratory water bath and held at 40 °C. The sample was tempered for 10 min before purging with nitrogen. One sample set (nine sample combinations in triplicates according to Table 1) was purged with nitrogen (50 mL/min) for 5 min to extract the very volatile aroma compounds. Another sample set (nine sample combinations in triplicates according to Table 1) was purged with nitrogen (150 mL/min) for 60 min to extract the less volatile compounds. Tenax-TA traps purged for 60 min were furthermore dry-purged directly with nitrogen (50 mL/min) for 10 min to remove excess water. Sealed Tenax-TA traps were kept from 1 to 3 days at 5 °C before analysis by gas chromatography–mass spectrometry.

2.6. Multiple headspace extraction (MHE)

To determine the total amount of the aroma compounds in the bread samples seven consecutive dynamic headspace extractions were performed from the same sample (the sample containing 40 g yeast/kg flour fermented at 15 °C). The same DHE procedures as described above were used, including two combinations of flow and time. All extractions were performed in triplicates. To determine the relationship between peak areas and absolute amounts of