# Advances in the Understanding of the Chemical Reactions Responsible for Bread Flavour Quality

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**Abstract**: This work aimed at analysing bread extracts obtained under headspace and artificial mouth conditions in order to study the reactions responsible for bread flavour quality. Selected extraction conditions were first compared according to the odour and aroma representativeness of the bread extracts. The optimal conditions were then applied to extract volatiles of a conventional gluten bread formulation and an innovative gluten-free bread formulation. Results reveal that an extraction duration of 10 min in the artificial mouth was sufficient to obtain a representative aroma extract. Yet, 30 min of headspace extraction were necessary to obtain a representative odorant extract. The comparison between these both extracts shows that significantly higher quantities of bread compounds were extracted with artificial mouth. This innovative device could thus be used to better understand the mechanisms involved in the formation of those volatiles that are responsible for the perceived bread odour and aroma.

**Keywords**: gluten bread; gluten-free bread; headspace, artificial mouth; representativeness

## INTRODUCTION

Bread has been for ages a popular staple, consumed throughout the world. Bread aroma has been largely studied, and about 300 flavour compounds have been identified (Pozo-Bayón *et al.* 2006). Quantitatively, the most important groups are alcohols, aldehydes, esters, ketones, acids, pyrazines and pyrrolines, but there are also furans, hydrocarbons, and lactones. However, only a small part of these compounds plays a significant role in the final bread flavour (Grosch & Schieberle 1997). To study bread compounds, several techniques have been used, as solvent extraction (Schieberle & Grosch 1994) distillation (Frasse *et al.* 1992; Zehentbauer & Grosch 1998), or solid-phase microextraction (SPME) (Poinot *et al.* 2007). An extraction technique can be selected according to two different objectives: either by extracting the greatest quantity of food volatiles, or by producing extracts with sensory characteristics as close as possible to those of the corresponding food. The latter is named representativeness. With this aim, we have developed an "artificial mouth". It mimics the conditions that bread is subjected to in the human mouth, in order to extract those compounds that are implied in the perception of food aroma.

To answer consumer's health request, breads with added nutritional value are now expanding. For instance, gluten-free breads are designed for patients with coeliac disease. Complementary to their nutritional quality, these innovative bread formulations have to be characterised by a good flavour to be accepted by consumers.

This work aimed at analysing bread extracts obtained by headspace and artificial mouth. Conditions to obtain representative odour and aroma extracts were retained and applied to study volatiles from a conventional gluten bread formulation and an innovative gluten-free bread formulation. This work showed the advance allowed by the artificial mouth to understand bread flavour quality.

## MATERIAL AND METHODS

***Bread-making procedure.*** Gluten dough consisted in white wheat flour (T55) (100 g), yeast (5 g), salt (2 g), improvers (1 g) and water (58 g). Ingredients were mixed for 4 min and 20 s at 100 rpm and 6 min and 20 s at 200 rpm. Dough was rested (15 min), divided, rested (10 min), and flattened. It was then divided in pieces of 70 g which were proofed for 60 min at 35°C, 95% relative humidity. They were baked during 20 min at 230°C with 0.5 l of steam at start baking. For gluten-free formulation (Mezaize *et al.* 2009), rice flour (47.5 g), corn starch (29.1 g), corn flour (14.2 g), buckwheat flour (5 g), potato starch (4.2 g), guar gum (1.9 g) and salt (1.6 g) were blended at 46 rpm for 10 seconds. Yeast (5.1 g) was then incorporated and the ingredients were mixed again at 46 rpm for 10 s. Sunflower oil (5.9 g) and water (83.6 g) were added. The whole was mixed at 82 rpm for 2 min. Dough pieces of 70 g were placed in muffin-like pans. They were proofed for 50 min at 40°C, 95% relative humidity, baked at 200°C for 40 min with 0.5 l of steam at start baking.

***Extraction of bread volatile compounds.*** A 75 µm Carboxen/Polydimethylsiloxane (CAR/PDMS) SPME fibre was used for both extraction methods. Headspace extracts were obtained with 6 g of homogenised crushed bread and 5 µl of internal standard (butanol) placed in a 125 ml sealed flask. The sample was stirred and placed at 35°C. After 5 min equilibrium, the SPME fibre was exposed to the headspace for 15, 30 or 60 minutes.

For artificial mouth extraction, 60 g of bread, cut into pieces were used with 30 ml of artificial saliva (Van Ruth *et al.* 1995) and 200 µl of internal standard (butanol). Artificial mouth container was maintained at 37°C. While bread samples were broken down by rotation and compression movements, helium gas carried away the volatiles released. This flow went to an opening in the cap where the SPME fibre was inserted. Extraction lasted for 2, 5 or 10 minutes.

***Bread extracts representativeness.*** This study was done following the recipes and procedures used in Poinot *et al.* (2007, 2009). Nine trained judges evaluated the representativeness of each extract. It consisted in scoring the odour proximity of headspace extracts with the original bread, and the aroma proximity of artificial mouth extracts with the original bread.

***GC-MS analyses.*** Gluten and gluten-free bread extracts were both obtained in triplicate by headspace for 30 min and by artificial mouth for 10 minutes. The identification and the quantification of volatiles were carried out by GC-MS/FID, following the procedure described by Poinot *et al.* (2009). Volatiles quantity was expressed in g/g butanol/g bread.

***Statistical methods.*** Analysis of Variance (ANOVA) and Least Significant Differences (LSD) tests with a confidence level of 95% were carried out on the representativeness scores. Principal Component Analysis (PCA), ANOVA and LSD (with a confidence level of 95%) were performed on the quantities of volatiles analysed by GC-FID.

## RESULTS AND DISCUSSION

As temperature and SPME fibre were similar in headspace and artificial mouth techniques, representativeness scores obtained by both techniques could be compared (Figure 1).

The extraction time required to obtain good representativeness scores was very different for

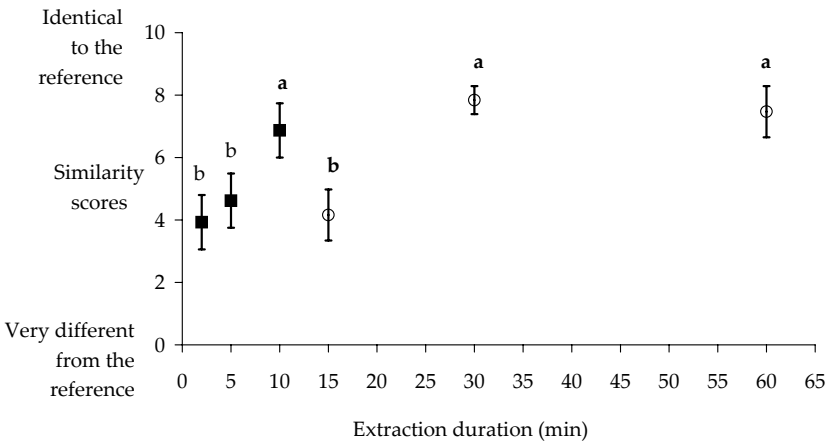


Figure 1. Evaluation of aroma similarity with the bread reference: mean scores given to artificial mouth extracts obtained for 2, 5 and 10 min, and to headspace extracts obtained for 15, 30, and 60 min; ■ artificial mouth extracts; ○ headspace extracts; a,b: LSD results

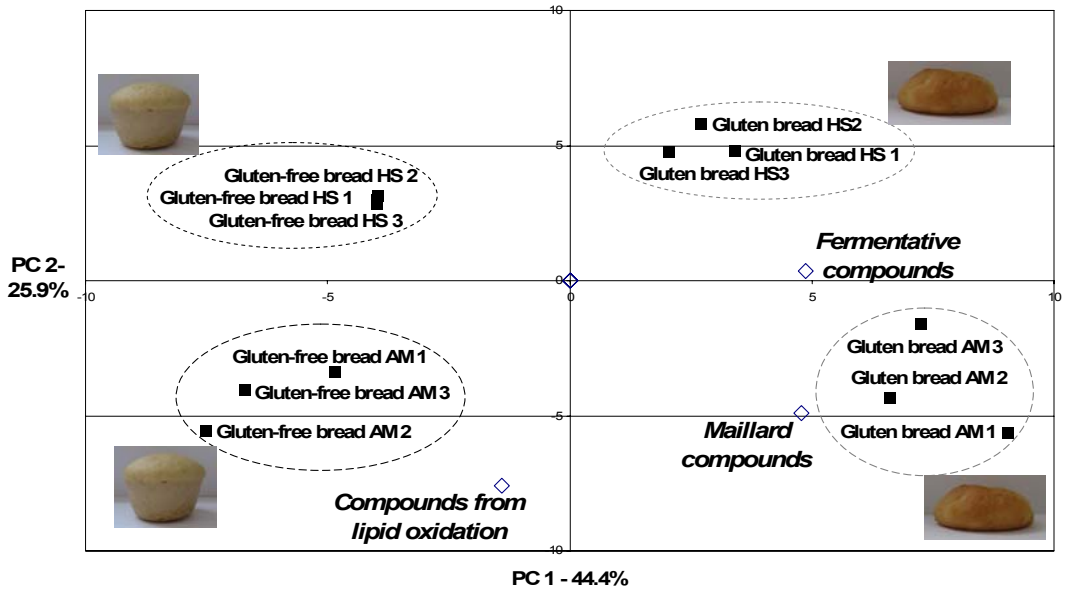


Figure 2. Bread extracts plot based on the quantitative volatile compositions in the two first dimensions; HS: headspace extract; AM: artificial mouth extract; 1/2/3: number of replicate

both techniques (respectively of 30 min for static headspace and 10 min for artificial mouth). Artificial mouth was thus much more efficient than headspace to extract bread volatiles.

Volatiles of gluten-free and gluten breads were then extracted under conditions leading to extracts with good representativeness: headspace for 30 min and artificial mouth for 10 min. The

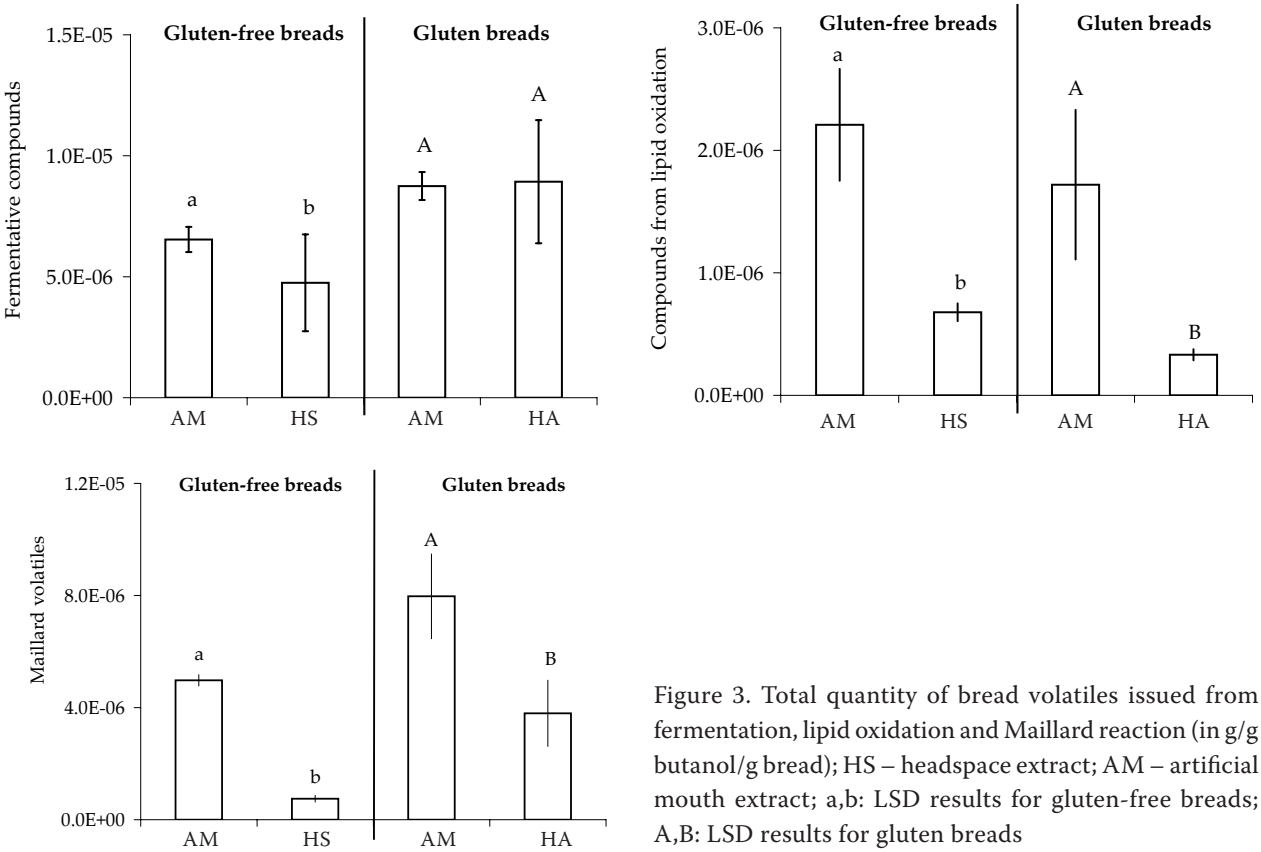


Figure 3. Total quantity of bread volatiles issued from fermentation, lipid oxidation and Maillard reaction (in g/g butanol/g bread); HS – headspace extract; AM – artificial mouth extract; a,b: LSD results for gluten-free breads; A,B: LSD results for gluten breads

obtained PCA is shown on Figure 2. Volatiles were gathered according to their origin: fermentation, lipid oxidation and Maillard reaction.

Gluten bread extracts were negatively correlated to gluten-free bread extracts according to the first axis (44.4% of variance). The second dimension (25.9% of variance) opposed headspace extracts to artificial mouth extracts. Gluten-free breads were characterised by high quantity of compounds from lipid oxidation leading to breads with more fatty and green notes. This could be due to the oil they contained. On the other hand, they contained lower quantity of fermentative compounds than gluten breads, which is quite surprising as both had similar amounts of yeast. Maillard volatiles were also in lower quantity in gluten-free breads than in the gluten ones. The latter could thus be probably characterised by more burnt and malty notes. This can be confirmed by the lighter crust colour of gluten-free breads compared to gluten breads. The absence of gluten and the addition of oil in the gluten-free formulation could be the reason for such results. Figure 3 also reveals that artificial mouth extracts contained significantly higher quantity of compounds from lipid oxidation and Maillard reaction than orthonasal extracts for the two types of bread. Fermentative compounds were also in higher quantity in artificial mouth extracts of gluten-free breads compared to the headspace extracts (Figure 3).

As more volatiles were extracted with artificial mouth, it could be thought that it would point out aromatic differences not shown by headspace techniques. These results highlight the potential of our artificial mouth to study foods which are relatively poor in flavour compounds. With this technique, the comparison of breads obtained with different formulations and processes would thus allow to understand the chemical reactions involved in the formation of food flavour quality.

## CONCLUSIONS

Results underline the interest of studying food volatiles with both orthonasal and retronasal ways of extraction. It could be used to better understand the chemical reactions involved in the formation of those volatile compounds responsible for the perceived odour and aroma.

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