FORMATION OF 5-HYDROXYMETHYLFURFURALDEHYDE IN LATVIAN WHOLE MEAL RYE BREAD DURING BAKING

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Abstract

During the baking heat exchange process, various chemical transformations take place and form new chemical compounds, and the final result is bread. Furans are some of these volatile aromatic compounds. They are formed in the heat exchange processes of food production- baking, heating, and roasting. One of these compounds is 5-hydroxymethylfurfuraldehyde (HMF), which is widely distributed in food products such as bread, coffee, honey, milk products, meat, fish, beer and others. Aromatic volatile compounds endow rye bread with its characteristic aroma and taste. The formation of furan and its derivatives in food products is not clearly understood. In 2004, the Europe Food Safety Authority started to collect data from EU member states of HMF content in food products. There is no such information available on Latvian foodstuffs. Specifically, in Latvia, there have been no investigations on HMF formation in rye bread during bake processing to date. The aim of this study was to investigate the dynamic of HMF formation in whole meal rye bread crumb and crust, as influenced by baking time. The bread was produced in a Latvian commercial bakery. The content of HMF in whole meal rye bread was determined by using liquid chromatography. The study showed that Latvian whole meal rye bread baked at the optimal time of 60 minutes contains 320.6 mg kg^{-1} HMF. Its content in rye bread crumb reached 22 mg kg^{-1}, which is substantially different from the HMF content of bread crust. During the prolonged baking time, the rate of HMF formation is no significant.

Key words: furan, HMF, rye bread, baking

Introduction

The Maillard reaction, first described by French scientist Louis-Camille Maillard in 1912, is comparable to a revolution in the food industry. For nearly 100 years food scientists have studied expressions of this reaction in food during thermal treatment, which leads to its characteristic colour, smell, taste, and structure. Also during the Maillard reaction (MR), aromatic compounds are formed which either enhance food flavours or reduce nutritional value, as happens during overly long-term food storage.

During the rye bread baking process, the Maillard reaction takes place, characteristically occurring in systems with intermediate moisture content, a temperature above 50 °C and a pH of 4–7, and caramelization also takes place during this process at temperatures above 100 °C (Ramirez-Jimenez et al., 2001).

5-hydroxymethylfurfuraldehyde (HMF) is one furan derivative, formed during the rye bread baking process as a result of the MR. HMF in food is a concern because, based on animal tests, it is considered possibly carcinogenic to humans by the International Agency for Research on Cancer (IARC, 1995). A hot debate arose around HMF a few decades ago and the compound`s toxicity, mutagenicity, and carcinogenicity are still controversial. The information contained in the scientific world is unequivocal. The threshold LD50 of HMF is 3.1 g kg^{-1} body weight in rats and dogs. There are studies at doses of 75 mg kg^{-1} which confirm HMF effects on animals, as well as researches that confound the assumption about the hazard of HMF (Michail et al., 2007).

The European Food Safety Authority (EFSA) founded a group to evaluate food safety, and during 2004–2009 collected data from EU Member States for furan and HMF content in food (EFSA, 2004). The scientific report lists 2908 furans containing samples from 20 different food groups (EFSA, 2009). There is no such information on Latvian foodstuffs. The EFSA working group, in summarizing findings obtained about the extent of HMF consumer exposure, origin, formation and toxicity, concluded, that the information does not represent a true picture of HMF in foodstuffs. Currently available data on food safety are insufficient to make evaluations or recommendations concerning potential changes in dietary regime. Further studies and data are required. Based on industry-provided data, the EU has

recommended a modified theoretical daily consumption (mTAMDI) of HMF, which is 1600 µg/person/day (EFSA, 2005)
Most Latvian consumers consider that the bread is the most important food in their diet after meat, dairy products and vegetables. Latvia is exceptionally rich in unique and consummate rye bread traditions. In Latvia rye bread is traditionally baked on the clay floor of a wood-fired oven. Such rye bread has a distinctive aroma, springy texture, and a slightly moist crumb with a fantastic taste. To date there have been no studies in Latvia on furan compounds and HMF formation during the baking of traditional rye bread. The Latvian Food Centre database has no information on the HMF content of any bread that consumers could access.
The aim of this study was to determine the formation of HMF in Latvian traditional whole grain rye bread during baking.

Material and Methods

The samples used in this experiment were obtained from rye whole grain flour (stock company 'Jelgavas Dzirnavas', Latvia). The dough was made with scald, natural starter, sugar, salt, malt and cumin.
The whole meal rye bread was baked in a Latvian commercial bakery under several conditions. Four 1kg loaves were formed and placed in the prebaking chamber for 1 minute at a temperature of 400 °C, and then the loaves were placed for baking in a chamber oven at a temperature of 250 °C. One loaf was used for measuring temperature during baking of loaf crust surface and crumb temperature inside the loaf by using thermo-couple. The start temperature of crumb was 37 °C and crust start temperature was 87 °C. Characteristics of the samples are given in Table 1.

Table 1

Characteristics of analyzed samples

Rye bread loaf	Baking time, min	Temperature of crumb, °C	Temperature of crust, °C
Nr. 1	30	94	134
Nr. 2	45	99	141
Nr. 3	60	100	149
Nr. 4	75	100	156

After baking the loaves were left to cool at room temperature for 12 hours. The loaves were cut into 1 cm thick slices. For analysis the crust was very carefully separated from the crumb by using a conventional knife. The crumb and the crust ratio of the samples was identified.
The study was conducted in the Scientific Laboratory of Natural Chemical Substances at the Department of Chemistry of the Latvia University of Agriculture in 2011.
For the HMF analysis, 5g of the homogenized sample was extracted in 50 ml of distilled water. The ten-minute extraction was carried out in an ultrasonic bath. After extraction the samples were centrifuged for 10 minutes at 6000 rpm (HERMLE Z 206A, Germany). The liquid phase of the sample was filtered through a 0.45 µm nylon filter, and was further used for HMF analysis by high performance liquid chromatography (HPLC), (CHIMADZU, Japan). The content of HMF in the samples was determined by the calibration curve method. Standard solutions of 20; 30; 40 mg l^{-1} were prepared of HMF (Sigma-Aldrich, Germany).
The filtrate of the sample (10µl) was injected into the HPLC, and analyzed by using (10/90) acetonitrile (CH3CN/H_2O) by flow rate of 1.3 ml min^{-1} as a mobile phase, and a Alltech C 18 column (4.6x250mm) connected to a UV/VIS detector (SPD-20A) set at 280 nm. Data were acquired and processed using Shimadzu LabSolutions software (Lesolution Version 1.21 SP1).

The colour of the samples was measured by Tristimulus colorimeter Color Tec/PCM (Accuracy Microsensors, Inc), reading the colour spectrum of the three coordinates, where L*is darkness, a*is redness, b*is yellowness. Statistical evaluation was performed using SPSS 14.0 for Windows. Significance was defined at $p \leq 0.05$ level.

Results and Discussion

A variety of structural, physiochemical, sensory changes and transformations take place during rye bread baking. The MR and caramelisation explain most of these changes, although it is difficult to characterize all the complexity of the compounds formed during the baking process. The HMF is one of the compounds formed during rye bread baking. The data obtained on HMF content in the rye bread crumb and crust at different baking times is shown in the Figure 1.

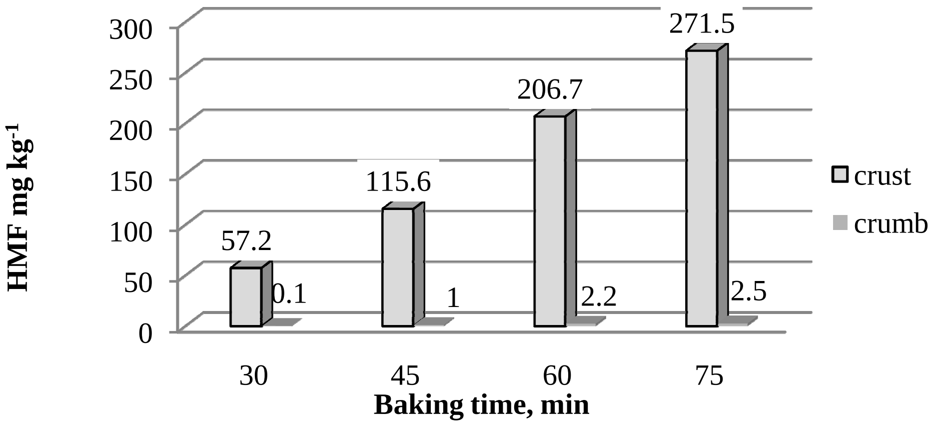

Figure 1.The HMF content in rye bread crumb and crust at different baking times

The section of bread loaf – crumb or crust – and baking time, are significant factors (p=0.001) which affect the formation of the HMF in bread. The HMF content of the rye bread crumb at the beginning of the baking process is 0.1 mg kg^{-1}. At optimal baking time – 60 min, – it increases to 2.2 mg kg^{-1}. Continuing baking for another 15 min, it reached 2.5 mg kg^{-1}. The temperature in the crumb increases from 94 ºC at 30 min of baking to 100 ºC at 75 min of baking. The HMF content of bread crust is 57.2 mg·kg^{-1} at an early stage of baking, and increase to 271.5 mg kg^{-1} at the end of baking. The HMF content of the samples at the beginning of baking differs significantly from the content at the end of baking, which can be explained by temperature effects.

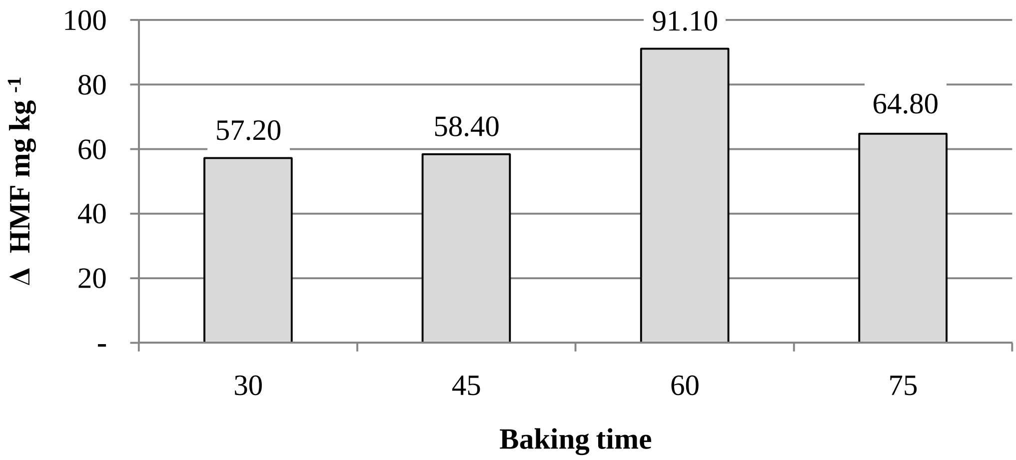

Figure 2.The HMF growth rate in the crust at different (30-45; 45-60, and 60-75 min) stages of baking

During baking, the water content on the surface of the loaf becomes less than in the middle and this, combined with the high temperature, is one of the factors that makes the crust different from the crumb. In these circumstances the MR takes place in the crumb, where HMF is a by-product, but in the crust, at a temperature of 134–156 ºC, sugar caramelisation takes place (Ramirez-Jimenez et al., 2001). Throughout the baking time the HMF content in bread crumb increases smoothly, but the amount is negligible compared to the amount in the crust. The HMF in the crust increases, but its growth rate at various stages of baking is different (Fig. 2.).

At an early stage of baking from 30 to 45 min the HMF growth rate was 3.9 mg min^{-1}. The highest growth rate of HMF in the crust was observed from 45 to 60 min, where it reached 6.1 mg $min^{-1,}$ but after 60 min it fell to 4.3 mg min^{-1}A similar kinetic was observed in a study on the cake baking process (Ait Ameur et al., 2008), where at the final stage of baking the growth of HMF decreased.

The colour L* a* b*measurements of loaf crumb and crust are summarized in Table 2.

Table 2

Colour parameters in crumb and crust at baking times

Baking time	Crumb			Crust		
	L*	a*	b*	L*	a*	b*
30 min	45.65±1.293	4.06±0.190	23.61±1.195	39.30±1.293	5.41±0.190	19.65±1.195
45 min	45.39±1.293	3.75±0.190	23.14±1.195	35.05±1.293	5.23±0.190	12.44±1.195
60 min	44.49±1.293	3.19±0.190	21.50±1.195	31.12±1.293	4.17±0.190	11.80±1.195
75 min	43.80±1.293	2.74±0.190	20.08±1.195	29.29±1.293	3.40±0.190	7.28±1.195

HMF, colour factors and temperature correlation were determined by correlation analysis. HMF is in a close negative correlation with the colour factor L*(r=-0.978), and factor b* (r=-0.955), and a close positive correlation with temperature (r=0.928). A close positive correlation between HMF and colour factor 100-L* has been observed in studies with biscuits and bread (Ramirez-Jimenez et al., 2001). The regression analysis showed that the HMF content can be predicted from L*factor and the temperature changes (HMF=1296.775 – 24.13 L* – 2.2 ·temperature). This regression model is significant (p=0.001), there is close correlation (r=0.986, R^2 =0.971).

During rye bread baking structural changes take place in the loaf, increasing the thickness of the crust. The ratio of the crust and crumb was determined for evaluation of HMF in bread (Fig. 3.).

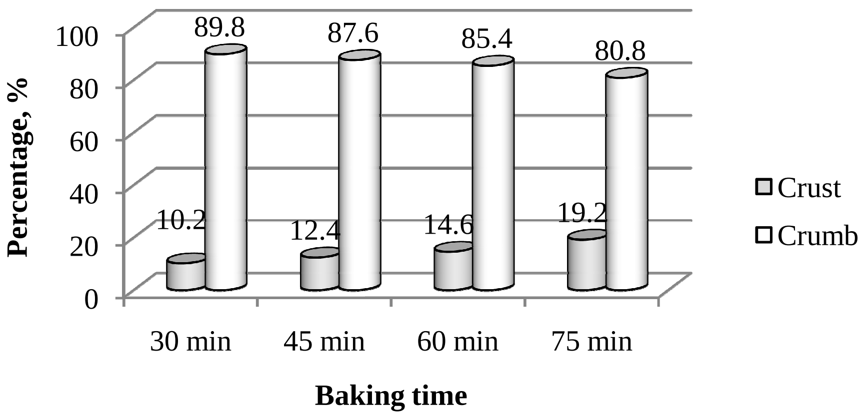

Figure 3. Crumb and crust ratio of bread at different time of baking

At the beginning of baking, the thickness of the crust is thinnest, and gradually becomes thicker with increased baking time while the crumb volume reduces. At the optimum baking time of 60 min bread crust is 14.6% and crumb is 85.4%.
The total content of HMF in the bread was calculated based on the HMF content in the crust and crumb as well as on crust and crumb ratio at various baking times (Fig. 4).

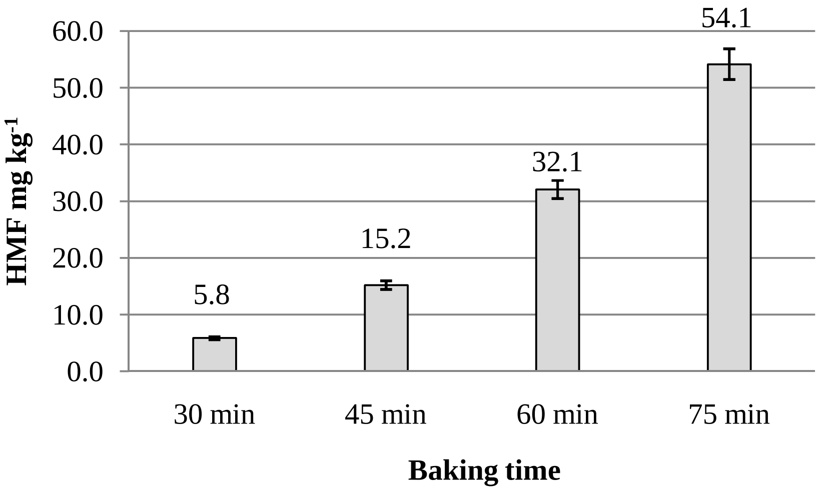

Figure 4. HMF content in the bread at various baking time

After studies and estimates the HMF content of 1 kg rye whole meal bread loaf at the optimal baking time (60 min) is 32.1 mg kg^{-1}. According to the literature, the content of the HMF in whole meal rye bread is reported to vary from 12.2 to 211.02 mg kg^{-1}(Bobere et al., 2009; Hiller B. et al.,2011). Latvian nutritionists recomended consuming 150–200 g of rye bread per day. In this case the exposure of HMF would be 4.82–6.42 mg per day. Spanish researches have calculated HMF exposure of 9.7 mg per day, of which 4.9 mg per day is ingested with coffee (Arribas-Lorenzo and Morales, 2010). Norwegian scientists have concluded,- that HMF is more actively assimilated into the human metabolism when consumed in coffee, dried fruits, and alcohol. Monosaccharides, disaccharides, and starch are not essential to the body`s metabolic process (Husoy et al., 2008). From this perspective, bread products are not a source of HMF potentially risky to human health. The issue of HMF in food is still not suffiiciently investigated, but it is necessary to look for ways to optimize HMF content in bread without losing its sensory properties. The FDA recommends that consumers eat a balanced diet, choosing a variety of foods that are low in trans fat and saturated fat, and rich in high-fiber grains, fruits, and vegetables.

Conclusions

1. The HMF content does not exceed 2.2 mg kg^{-1} in wholemeal rye bread crumb, but in the crust it was 57.2 mg kg^{-1} at the early stage of baking, and increased to 271.5 mg kg^{-1} at 75 min of baking.
2. Baking time and temperature have signiificant ($p=0.001$) impact on the HMF content in bread.
3. Wholemeal rye bread contains 32.1 mg kg^{-1} of HMF; prolonging baking time by 15 min, increases the content of HMF by 1.7 times.
4. The HMF is in close correlation ($r=0.986$) with the darkest shades of colors characterized by the factors L*. The greatest increase of HMF in the bread crust was found at 45 and 60 min of baking.

Acknowledgements

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