Effects of processing conditions on the formation of Acrylamide and 5-hydroxymethyl-2-furfural in cereal-based products

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Introduction

During heat treatment of foods, the Maillard reaction may occur when a reactive aldehyde - usually a reducing sugar such as glucose, fructose, maltose - and an amine -usually an amino acid, peptide or proteins – react (Nursten, 2002). The Maillard reaction results into compounds that contribute to the flavour, colour and aroma of many foods, but may also result into the formation of (potential) toxic compounds such as acrylamide (AA) and 5-hydroxymethyl-2-furfural (HMF). In wheat based products, like bread and bakery products, AA and HMF are considered the most important heat induced contaminants (Erbas *et al.*, 2012).

In 1994, the International Agency for Research on Cancer (IARC) classified AA as “potentially carcinogenic to humans” (IARC, 1994). In 2001, the Scientific Committee on Toxicity, Ecotoxicity, and the Environment reported that AA has toxic properties, including neurotoxicity, genotoxicity to both somatic and germ cells, carcinogenicity and reproductive toxicity (SCTEE 2001). Tardiff and co-workers (2010) estimated the tolerable daily intake (TDI) of AA for neurotoxicity to be 40 μg/kg-day and for cancer to be 2.6 μg/kg-day. Findings on the possible carcinogenicity of HMF are contradictory, as concluded in the literature review held by Abraham et al. (2012); overall evidence for carcinogenic potential of HMF was very limited. Other toxic effects were not observed, as based on a daily dose in the range of 80-100 mg/bw in animal experiments. Data on reproductive and developmental toxicity were not available. Given these uncertainties it is not possible to establish a TDI to date (Abraham *et al.*, 2012). According to a recent review of Capuano and Fogliano (2011) HMF can be converted in vivo into 5-sulfoxymethulfurfural (SMF) which is a genotoxic compound .

Food products that may have high concentrations of AA include cooked starchy foods such as potato crisps, French fries, home cooked potato products, biscuits, breakfast cereal and some non-starchy foods such as coffee (Capuano and Fogliano, 2011). Dietary exposure to AA may result from ingestion of high-carbohydrate food products such as potato crisps and chips, roasted cereals and breads (Keramat *et al.*, 2011). Dietary intake assessments of AA have been calculated for different countries. Based on these exposure assessment studies, summarized by Keramat and co-workers (2011), potato products account for around 50% and baking products and bread for about 20% of human exposure to AA. Another important food group for human exposure to AA includes coffee (Erbas *et al.*, 2012).High levels of HMF can be found in dried fruits (e.g. 25-2900 mg/kg), in particular of plums, coffee (e.g.100-4100 mg/kg), and caramel products. Cereals and bread may have elevated concentrations, due to variability in production and composition (Abraham *et al.*, 2012; Capuano and Fogliano, 2011).

In many food products, the formation of AA and HMF is directly related to the heat load applied to the food. Other processing conditions, such as water activity and the matrix, also influence the formation (and degradation) of AA and HMF. In order to mitigate AA and HMF in food products, the kinetic pathway for the formation of these two contaminants need to be known as well as effects of ingredients/precursors and processing conditions. However, to date, effects of processing conditions have mainly been studies in liquid model systems. Data of effects of processing on AA and/or HMF in cereal based dry model systems or dry real foods is limited. Effects may be different, though, in dry food systems as compared to liquid model systems.

This paper presents an overview of scientific knowledge on effects of precursors, and main processing conditions on the formation of AA and HMF in cereal derived food products, with a focus on type of sugars, moisture and time-temperature profiles.

Effects of precursors (reducing sugars)

AA in is formed predominantly from the amino group of asparagine and a carbonyl compound derived from reducing sugars (mainly glucose, fructose and maltose) in food. In cereal based dough used for bakery products, sugars seem to be the most important ingredients for AA formation as the level of free asparagine in wheat is relatively low (Gökmen *et al.*, 2007).

The impact of different types of sugars on the kinetics of AA formation and elimination using an asparagine-sugar model system under low moisture conditions (water activity of 0.92 at 4^0C) was studied by De Vleeschouwer et al. (2009). Evaluated sugars included: glucose (an aldose), fructose (a ketose) and sucrose (a disaccharide) at different temperatures (140^0C to 200^0C). AA concentrations were determined after different treatment times. Results showed that the model system with sucrose had a longer lag phase for AA formation than the model systems with glucose and fructose. This was explained by the authors by the fact that sucrose first needs to hydrolyze into glucose and fructose. This lag phase is shorter with higher treatment temperatures. Gökmen et al. (2007) evaluated different biscuit recipes with glucose or sucrose in a dry model system, and found glucose to result into higher AA concentrations than sucrose. The amounts of AA in the sucrose system were very low which, according to the authors, was due to limited hydrolysis of sucrose at the time-temperature used (11 minutes, 205^0C) in this baking experiment. Results of De Vleeschouwer et al. (2009) showed a higher AA concentration for the model system with fructose as compared to the model system with glucose, corresponding to results from earlier low moisture experiments (Robert *et al.*, 2005; Zhang *et al.*, 2007). For liquid model systems, opposite results have been reported, i.e., higher AA concentrations with glucose as compared to fructose (Claeys *et al.*, 2005). De Vleeschouwer et al. (2009) argue that-in liquid systems - glucose has a higher chemical reactivity than fructose due to the more reactive aldehyde group, explaining the higher AA concentrations. However, in dry systems, melting has to occur before the Maillard reaction can start, and the melting behavior of the precursors determines the rate of AA formation. Fructose has a lower melting point than glucose, explaining the higher AA concentrations when fructose was used in the low moisture model system.

HMF is formed as an intermediate Maillard Reaction Product (MRP) upon heating at high temperatures and from dehydration of hexoses under mild acid conditions. Effects of sugars and temperatures on HMF formation in biscuit models were investigated in several studies. In the study of Gökmen et al. (2007) a linear relationship was seen between the amount of sugar in the biscuit recipe and HMF concentrations after baking at 205^0C for 11 minutes. HMF formation was higher with glucose in the recipe than with sucrose. A study of Ameur et al. (2007) indicated that biscuits prepared from sucrose generated less HMF than biscuits containing glucose and fructose at baking temperatures below 250^0C, whereas the reverse was observed at 300^0C. These results are in agreement with the conclusion of Gökmen et al. (2008) that using non-reducing sugars (sucrose) to produce biscuits can reduce HMF generation as compared to the use of reactive sugars such as glucose at temperatures below 210^0C.

Effects of processing conditions (time- temperature, moisture)

Processing conditions for preparation of cereal derived products, such as time-temperature treatment and moisture content, may influence AA and HMF formation.

Time-temperature

Generally, a temperature of 120˚C or higher is needed for AA formation, but it could be formed at lower temperatures in dry food systems (Keramat *et al.*, 2011). AA concentrations in food products are considered net amounts, i.e., the result of AA formation and disappearance. Prolonged heating at high temperatures as well as long storage may reduce AA levels.

In their experiments, De Vleeschouwer et al. (2009) showed clear effects of treatment time and temperature (120, 140, 160, 180, 180, 200˚C) on AA concentrations. AA increased with time, and the increase was higher at the higher temperatures. The initial increase in AA concentrations was followed by a decrease, which started earlier at the higher temperatures used. The increase and decrease is the net result of simultaneous formation and degradation of AA, and was independent of the type of sugar used. In the dry model system used by Gökmen et al. (2007), the decrease in AA levels was not seen; AA concentrations tended to reach a plateau after a certain time of heating. The plateau reached was higher and reached earlier with the higher treatment temperatures. Maximal treatment time in this case was much shorter (25 minutes) that with the experiments of De Vleeschouwer et al. (2009). Plateau values were also higher in biscuits containing glucose than those containing sucrose (Gökmen *et al.*, 2007). Using dry model systems, Capuano et al. (2008) studied the effects of time (up to 34 minutes) and temperatures (140, 160, 180^0C) on AA and HMF formation during toasting bread crisps. AA was formed at all times at 180^0C, until a final level of 262 µg/kg was reached after 25 minutes. At 160^0C, formation of AA showed a lag phase; it was only formed after 26 minutes and reached 161 µg/kg at the end of the treatment (34 minutes). In their study, Capuano et al. (2008) also did not see the degradation of AA with higher treatment time and temperature. They suggested differences between dry systems and systems containing residual water in that evaporation decreases the effective temperature.

In the experiments of Capuano et al. (2008), HMF concentration was also highly affected by toasting time and temperature. Levels increased with longer time and higher temperatures. Highest concentrations were found when the highest temperature (180^0C) and the longest heating time (25 minutes) were applied. At each temperature, formation of HMF followed a first order kinetic model. A study of Ameur et al. (2007) on model cookies showed that HMF concentrations in biscuits baked at 200^0C were 10-100 times less than those baked at the higher temperatures. In this study HMF formation also followed a first order kinetic model. Accordingly, using a biscuit model prepared from sucrose, baking temperatures below 200^0C prevent from an excessive decomposition of sucrose and HMF formation (Gökmen *et al.*, 2008).

Moisture content

During baking of bread and dough, the water content on the surface quickly decreases providing optimum conditions for the MRP formation and brown colouring. Inside the dough, the temperature is lower and the water activity remains relatively high. Therefore, in general the crumb is only weakly coloured with a low concentration of MRPs, whereas the crush is dark coloured and contains high concentrations of MRP. In experiments held by Açar and Gökmen (2009), no AA was found in thicker biscuits as compared to thinner ones, because of lower moisture conditions (<10%) in the thicker biscuits. In their experiments, Capuano et al. (2008) found only AA in the bread crust, probably because moisture in the crumb was too high to allow temperatures to reach values needed for AA formation. The found AA concentrations was inversely related to moisture content. In general, there seems to be an optimal range of water activity for AA formation, at about 0.4 – 0.8.

Water activity is also considered an important parameter for HMF production; the presence of too much water in the early stages of heating might inhibit the reaction. Results of the dry model system experiments of Capuano et al. (2008) showed HMF formation was strongly affected by moisture content, at each of the three treatment temperatures. In fact, HMF only starts to accumulate when a certain low moisture content has been reached. The authors argued the dependency of HMF formation from water content could be explained in that in the first phase of toasting, evaporative cooling keeps the temperature of the bread crisp not over 100˚C until most of the water has evaporated. The moisture content value at which HMF starts to form is higher with a higher temperature for toasting the bread slices. Using model biscuits, Ameur et al. (2007) showed that HMF formation started at water activity between 0.5 and 0.7, but also depending on baking temperatures and types of sugars used in the formulations.

Mitigation

From the above mentioned results, it can be concluded that, in general, using sucrose instead of reducing sugars (fructose, glucose) in biscuits can lower the formation of both AA and HMF using heating temperatures at or below 210^0C. Also, prolonged heating at lower temperatures may results in lower AA and HMF concentrations in the final cereal derived products. However, attempts to lower the concentrations of AA and HMF should also evaluate quality characteristics of the final products. Within the FP7 Prometheus project, experiments with baking biscuits have been performed recently (unpublished data). Biscuits with different recipes, used on industrial basis, were baked at various oven temperatures, including 180˚C, 190˚C and 200˚C for a duration of 15 minutes. Results of these experiments will be used to advise the bakery industry on processing conditions to limit AA and HMF levels while keeping acceptable product quality.

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