

ATTEMPT TO REDUCE ACRYLAMIDE CONTENT IN ROASTED CHICORY

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ABSTRACT

The aim of this study was to reduce the formation of acrylamide during roasting of chicory roots by soaking the fresh roots in a solution of calcium chloride, by the use of different temperature and time of roasting of dried roots, as well as by the addition of the enzyme (asparaginase) during roasting of dried roots.

It was shown, that with increasing roasting temperature of chicory roots from 100 - 175 ° C the acrylamide content also increased, while at a temperature of 210 ° C the growth was inhibited. Increasing roasting time from 10 - 25 minutes resulted in an increased acrylamide content. Soaking the roots in the CaCl₂ solution for 20 minutes reduced the formation of acrylamide during the roasting approximately by 40%, similarly as the application of asparaginase to the dried roots during the roasting process.

Keywords: Chicory root, roasting, acrylamide

INTRODUCTION

Before the advent of natural coffee in Europe, back in the old days there were consumed drinks prepared from cereals or root products containing carbohydrates. However, the actual development of chicory coffee manufacturing could be assigned/ to Napoleonic wars period (Hrankowski, 1976).

Grain coffee is obtained by mixing a roasted and-grounded plant material, mainly wheat, barley and dried roots of chicory and sugar beet. The main objective of this technology is to obtain a product possessing the taste and aroma similar to natural coffee. It is possible due to the chemical transformations occurring during raw materials roasting, and by proper composition of the individual components. As a result of chemical transformation, a compounds providing the raw material with suitable flavours, aromatics, colorants, and nutritional properties are created (Hrankowski, 1976).

The greatest changes take part in carbohydrates fraction - flavouring agents are formed due to their decomposition at high temperature (Hrankowski, 1976). Also an important role is played by proteins contained in the roasted material. Under the influence of high temperature proteins undergoes the decomposition, probably up to amino acids. The brews from roasted cereals contain histamine, which causes the secretion of gastric juice and thus has a positive effect on the digestive system (Pazola *et al.*, 1970)

During the heat treatment of foods (frying, roasting), there are undergoing many chemical transformations initiated by reaction between amino acids (mainly asparagine) and carbonyl compounds. The products of this reaction are condensed glucosylamines- an initial products of Maillard reaction, which play a very important role in the acrylamide formation. These compounds are unstable and quickly undergo Amadori rearrangement. Amadori compounds could be subjected to a further degradation reactions into the highly reactive compounds. This process is very complex, and high-molecular substances are created, which are responsible for flavour, colour and odour of food products. They are mainly high molecular weight polymers and copolymers known as melanoids, as well as such compounds as furfural, acrolein and acrylamide (Stadler *et al.*, 2002; Żyżelewicz *et al.*, 2010).

So far, it was not established the level of acrylamide harmful to the human body. It has been proven, that this compound is toxic to nerve tissue; the neurotoxic effect has been demonstrated on experimental animals, and confirmed by observations in humans. Studies have shown, that acrylamide can act carcinogenic and promote the possibilities of cancer. This compound can reduce glutathione content in the body, which is a natural antioxidant, scavenging the free radicals. Free radicals damage cells, causing tissue aging and cancer development (Klaunig, 2008; Czerwińska, 2011).

One strategy for reducing acrylamide content in food is therefore the selection of proper varieties of plant material containing a small amount of acrylamide precursors (Capuano *et al.*, 2009). Another possibility to reduce the content of this compound, is the selection of appropriate methods of materials processing, such as blanching and soaking of potato chips, soaking the material in a solution of divalent metal salt or use of the enzyme asparaginase (Friedman and Levin, 2008; Rachwał and Nebesny, 2012). Using proper additives can also prevent the formation of acrylamide. These include: acidifying agents, amino acids (glycine and cysteine), flavonoids and polyphenols (Claus *et al.* 2007; Hedegaad *et al.* 2007; Ciesarova *et al.* 2008). The selection of an appropriate parameters of technological treatment, i.e. pH, time, temperature and humidity can also have a significant effect on the formation of acrylamide (Friedman, 2003; Friedman and Levin, 2008; Żyżelewicz *et al.*, 2010; Rachwał and Nebesny, 2012).

Therefore the aim of this study was to reduce the formation of acrylamide during roasting of chicory roots by soaking them in a solution containing divalent cations, by the use of different roasting regimes (temperature and time) of dried roots, and by use of the enzyme asparaginase during roasting of dried roots.

MATERIAL AND METHODS

The research material included: 15 pieces of fresh chicory roots (about 7 kg) and fresh roots soaked in CaCl₂ solution. Also producer of grain coffee provided the material consisting of already prepared extracts from the chicory roots roasted with and without addition of asparaginase, and powdered flakes of chicory roots roasted without and with the addition of the enzyme (asparaginase). The amount of enzyme added, and the conditions of treatment (drying and roasting) are confidential.

Impregnation of chicory roots in a solution of 3% calcium chloride was carried out according to a method based on the patent EP 2 160 948 A1. Chicory roots were soaked at room temperature (about 22 °C) in 3% solution calcium chloride for 20 minutes. Then roots were chopped into flakes (approx. 1 cm thickness) and dried at 70 °C for 3 hours. Next, the dried flakes were roasted in MIWE Condo baking oven at 150 °C for 40 minutes. Control sample was also performed.

Dried chicory roots were roasted under the following conditions:

- at 100 °C for a period of 10, 15, 20 and 25 minutes
- at 150 °C for a period of 10, 15, 20 and 25 minutes
- at 175 °C for a period of 10, 15, 20 and 25 minutes
- at 210 °C for a period of 10, 15, 20 and 25 minutes

In all examined samples acrylamide content was determined in high-performance liquid chromatography HPLC/UV (Paleologos and Kontominas, 2005). Knauer chromatographic system consisted of: Smartline pump 100, the valve K-6, ion exchange column Eurokat H +, thermostat, and a UV detector Smartline 2500. Chromatographic separation was performed isocratic, eluting with a mixture acetonitrile and 0.01 M sulfuric acid (VI) in proportions by volume of 2:8. The volume of injected material was 20 ml. Detection was at 200 nm, at 40°C, at a flow rate of 0.6 cm³ / min

All results were subjected to one-way analysis of variance (ANOVA) by means of Statistica 10.0. software. The significance of differences were calculated using Duncan test at $\alpha = 0.05$. Results are presented as the mean of three replicates and standard deviation ($\bar{x} \pm SD$).

RESULTS AND DISCUSSION

Natural coffee is generally drunk in order to temporary improve the psychophysical fitness and well-being, and such result is attained by the main component of the brew - caffeine. But it should be also taken into consideration that approximately 700 other chemical compounds were also identified, whose consumption could not be neutral for body, and more importantly - their role and significance were not fully understood. Also in roasted coffee beans there were detected a large amounts of a potentially carcinogenic compound – acrylamide, especially in instant coffee. In recent years, due to the acrylamide presence, a great interest was paid to a coffee substitute manufactured from roasted wheat, sugar beet and chicory roots (Orzel and Biernat, 2011).

Therefore, the aim of this research was to reduce the formation of acrylamide during roasting of chicory roots, one of the most important raw material in the production of grain coffee, responsible for its flavour.

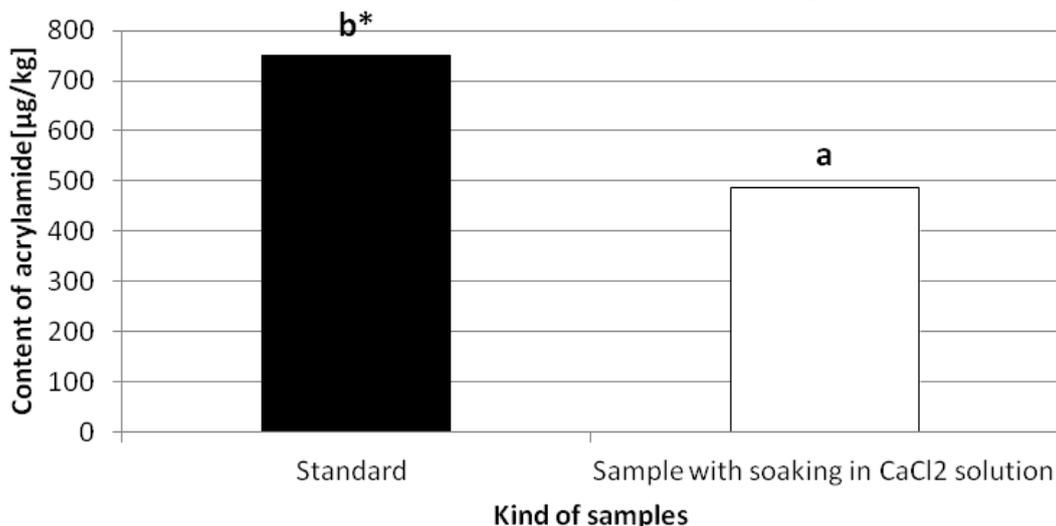


Figure 1 The content of acrylamide in dried, roasted flakes of chicory roots subjected to soaking in CaCl₂ solution or without soaking (standard sample)

* Different letters indicate statistically significant differences at a significance level $\alpha = 0.05$

The level of acrylamide in roasted chicory flakes is presented on Fig. 1. The content of acrylamide in soaked flakes was significantly lower than in the standard sample. Soaking for 20 minutes in a 3% solution of calcium chloride reduced the acrylamide content by approximately 40%. Results presented in EP 160948A1 2 patents demonstrated, that soaking in the calcium chloride led to more rapid colour development during roasting of chicory roots, depending on the concentration of CaCl₂. With increase of calcium chloride concentration in the impregnating solution it is possible to decrease the roasting time. It was also found, that the level of acrylamide decreased with increasing calcium concentration in the impregnating solution. Thus, 2% concentration of CaCl₂ after two hours of impregnation resulted in limiting the acrylamide content by 97% (EP 2 160948A1; Stadler and Scholz, 2004).

It is surprising that at moderately high roasting temperature, which was applied in this work (i.e. 150 °C) and a 40-minute roasting time, a product was obtained characterized by more desirable darker colour, when the chicory roots were previously soaked in CaCl₂. Reference sample (without soaking) roasted under the same conditions, showed a much lighter colour, and a significantly higher content of acrylamide (Fig. 1). The conclusion is, that there is not always a

connection between a darker colour and higher acrylamide content. A similar trend was also observed by other authors in studies conducted on oats flour (Zięć *et al.*, 2011).

It is also important that, the impregnation of chicory roots by divalent cations do not affect the organoleptic properties of the final product, namely the grain coffee (EP 2 160948A1; Delatour *et al.*, 2004; Ariseto *et al.*, 2008).

Fig. 2 presents the results of acrylamide content in flakes of dried chicory roots roasted at 100, 150, 175 and 210 °C, for 10, 15, 20 and 25 minutes. It was observed that with increasing temperature and time of roasting acrylamide content was significantly increased. Similar results were obtained by Zięć and Gambuś (2012) examining the content of acrylamide in biscuits baked at different times (8 - 20 min.). They observed a progressive increase in the level of acrylamide with increased baking time, at the same temperature.

As can be observed from the data presented in Figure 2, the exposure time at a given temperature, influenced in different ways the content of acrylamide in the roasted chicory samples. At 100 °C, with increasing time from 10 to 25 min, the acrylamide content increased about 3 - fold; at 150 °C - about 2.5 - fold; at 175 °C - about 4 - fold and at 210 °C about 3 - fold (Fig. 2).

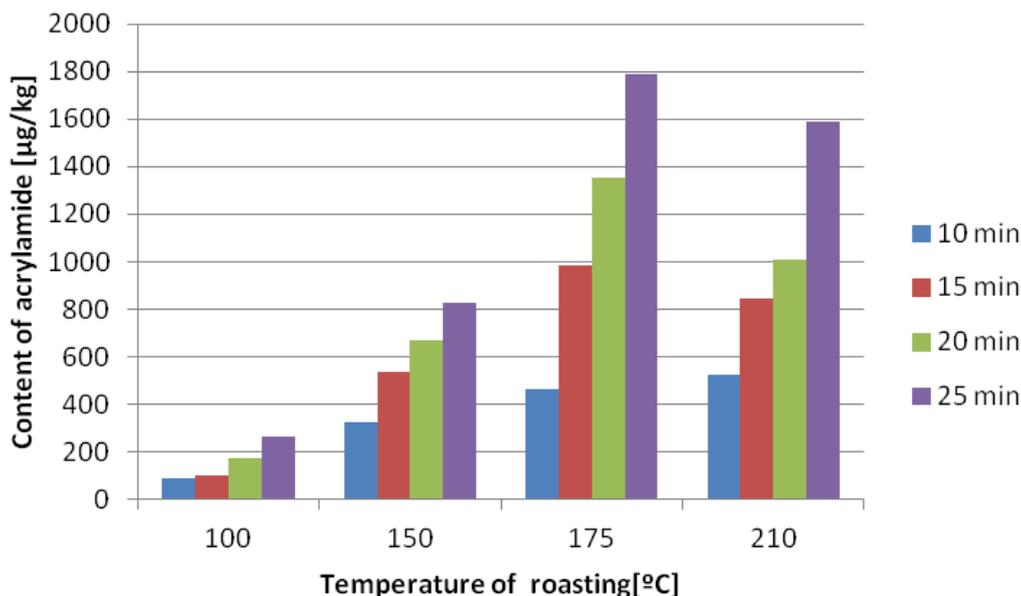


Figure 2 The content of acrylamide in the flakes of chicory roots roasted at different times and different temperatures

On the other hand, comparing the contents of acrylamide at the same time of roasting, but at different temperatures, it clearly shows a significant increase in the content of this compound, with increasing temperature from 100 to 210 °C (Fig. 2). The largest increase in acrylamide content was observed in chicory roots roasted at temperature of 175 °C for 25 minutes. It is worth emphasizing, that even at higher temperatures (210 °C) it was not observed an expected increase in the content of acrylamide, even after 25 minutes of roasting.

Inhibition of acrylamide level growth at high temperature can be explained by the thermal decomposition of this compound to simpler compounds such as acrolein, triglycerol or acrylic acid (Bräthen and Knutsen, 2005).

As it is derived from the literature, the use of asparaginase enzyme can significantly reduce the acrylamide content in the finished product (Lisińska et al., 2007; Kuilman and Wilms, 2007; Pedreschi et al., 2008; Orzel and Biernat, 2011). Table 1 presents the results concerning acrylamide content in extracts and powdered roots, roasted without and with asparaginase. It was observed a significant reduction in acrylamide content in samples treated with enzyme – in case of extract from chicory roots the content of this compound was reduced by 64%, and in the case of powdered roots only by 10% (Table 1) when compared with samples treated without enzyme (standard).

Table 1 The content of acrylamide in extracts and powdered roots, roasted without and with asparaginase enzyme

Kind of extract	Acrylamide content [µg/kg]	Kind of powder	Acrylamide content [µg/kg]
Standard 1*	665,9 b** ± 3,8	Standard 2	636,1 b ± 3,1
L1	429,4 a ± 2,1	L2	567,3 a ± 1,8

* Standard 1 Standard sample, extract of roots roasted without asparaginase; Standard 2 - standard sample, powdered roots roasted without asparaginase enzyme; L1 - extract of roots roasted with asparaginase; L2 - powdered roots roasted with asparaginase enzyme

** Different letters in the columns indicate statistically significant differences at a significance level $\alpha = 0.05$

Perdreschi et al. (2008) studied the content of acrylamide in French fries without and with addition of the asparaginase. In French fries blanched in a solution of the enzyme prior to frying, it was observed a significant reduction in acrylamide content. Asparaginase decomposes asparagine, the substrate of acrylamide formation in Maillard reaction, into ammonia and aspartic acid (Lisińska et al., 2007; Kuilman and Wilms, 2007; Pedreschi et al., 2008; Orzel and Biernat, 2011).

It can be summarized, that methods applied in present research to decrease the acrylamide content in roasted chicory roots, proved to be effective and recommended to be implemented in the industrial production of grain coffee.

CONCLUSION

Impregnation of chicory roots with 3% solution of calcium chloride for 20 minutes prior to the roasting process effectively and significantly reduced the acrylamide content in roasted flakes, as opposed to the flakes roasted at the same conditions but without soaking. As the temperature of chicory roasting raised from 100 - 175 °C the content of acrylamide significantly increased at the same exposure/roasting time, both at the period of 10, 15, 20 and 25 minutes. It was observed the successive increase of acrylamide content with increasing roasting time that was stopped at temperature of 210 °C. The determination of acrylamide content, both in the extracts and in the powders of roasted chicory roots obtained with and without the application of asparaginase enzyme, showed a significant reduction of this compound when enzyme was applied, especially in extracts (approx. 60%) when compared to reference sample. Applied in this work methods of acrylamide content reduction in roasted chicory roots, proved to be effective and recommendable to implement in the industrial production of grain coffee.

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