ACRYLAMIDE FORMATION IN DIFFERENT FOODS AND POTENTIAL STRATEGIES FOR REDUCTION

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Abstract: This paper summarizes the progress made to date on acrylamide research pertaining to analytical methods, mechanisms of formation, and mitigation research in the major food categories. Initial difficulties with the establishment of reliable analytical methods have today in most cases been overcome, but challenges still remain in terms of the needs to develop simple and rapid test methods. Several researchers have identified that the main pathway of formation of acrylamide in foods is linked to the Maillard reaction and in particular the amino acid asparagine. Decarboxylation of the resulting Schiff base is a key step, and the reaction product may either furnish acrylamide directly or via 3-aminopropionamide. An alternative proposal is that the corresponding decarboxylated Amadori compound may release acrylamide by a beta-elimination reaction. Many experimental trials have been conducted in different foods, and a number of possible measures identified to relatively lower the amounts of acrylamide in food. The validity of laboratory trials must, however, be assessed under actual food processing conditions. Some progress in relatively lowering acrylamide in certain food categories has been achieved, but can at this stage be considered marginal. However, any options that are chosen to reduce acrylamide must be technologically feasible and also not negatively impact the quality and safety of the final product.

Key words: Acrylamide, food, analysis, mechanisms of formation, mitigation

1. INTRODUCTION

The announcement by the Swedish National Food Authority in April 2002 of the presence of acrylamide predominantly in carbohydrate-rich foods

Chemistry and Safety of Acrylamid in Food, edited by
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(Tareke et al., 2002) sparked intensive investigations into acrylamide, encompassing the analysis, occurrence, chemistry, toxicology, and potential health risk of this contaminant in the human diet.

Several research groups have developed methods to quantify reliably acrylamide at relatively low levels in a large variety of different foodstuffs. Most of the methods published so far are based on either GC-MS or LC-MS techniques, with comparable performance of the two approaches. Based on the conclusions of a recent inter-laboratory trial (Wenzl et al., 2003) and the EC / JRC task force group held in April 2003 (Joint European Commission Workshop, 2003), many of the methods did not perform well in difficult matrices such as cocoa and coffee. Consequently, laboratories have adapted their methods to achieve the required precision and sensitivity for those foods in which gaps in the analytical science were initially identified. A fully validated method with adequate performance and that can be applied also to "difficult" matrices such as coffee and cocoa is briefly described in this report.

Several months after the Swedish announcement, a number of research groups simultaneously discovered that acrylamide is formed during the Maillard reaction, and the major reactants leading to the formation of acrylamide are sugars and the amino acid asparagine (Mottram et al., 2002, Sanders et al., 2002, Becalski et al., 2003, Stadler et al., 2002). Fundamental mechanistic studies published in 2003 have revealed a feasible route to acrylamide (Zyzak et al., 2003, Yaylayan et al., 2003), and a number of possible minor pathways have also been described (Stadler et al., 2003, Lingnert et al., 2002). This fundamental knowledge opens the way to concrete studies on kinetic modeling (formation over temperature/time, competitive reaction kinetics with amino acids and sugars), and identifying the rate limiting steps under actual food processing conditions. Measures could then be devised to attempt to reduce acrylamide in food products. This, however, necessitates extensive individual trials for each food category, and in most cases entails a combination of measures that on a case-by-case basis must be applied at the raw material stage, during storage, processing, and final preparation of the product in the home.

This report summarizes the current state of knowledge focusing on the analytical, mechanistic, and mitigation aspects. Furthermore, the progress and complexity of the research is highlighted in a number of different products, so indicating as well the challenges and constraints faced by industry in finding appropriate and practical solutions to this concern.

2. PROGRESS MADE TO DATE IN ANALYTICS

Only recently has a single method been reported that can achieve good sensitivity and selectivity of acrylamide in practically all of the relevant food matrices (Roach et al., 2003), and has been improved for coffee that was found particularly troublesome (Andrzejewski et al., 2004).

Our laboratory has recently reported a method by isotope-dilution liquid chromatography-electrospray ionization tandem mass spectrometry (LC-MS/MS) that achieves good precision, accuracy, and certainty of the analyte in a wide range of foodstuffs. However, acrylamide could not be quantified reliably in difficult matrices such as cocoa powder and coffee, mainly due to considerable loss of the analyte throughout the sample preparation steps (Riediker and Stadler, 2003). Improvements were thus made to the existing method (Figure 1), and sample pre-treatment essentially encompasses (a) protein precipitation with Carrez I and II solutions, (b) extraction of the analyte into ethyl acetate, and (c) solid phase extraction on a Multimode cartridge. This approach provided good performance in terms of linearity, accuracy and precision. Full validation was conducted in soluble chocolate powder, with good precision (Table 1).

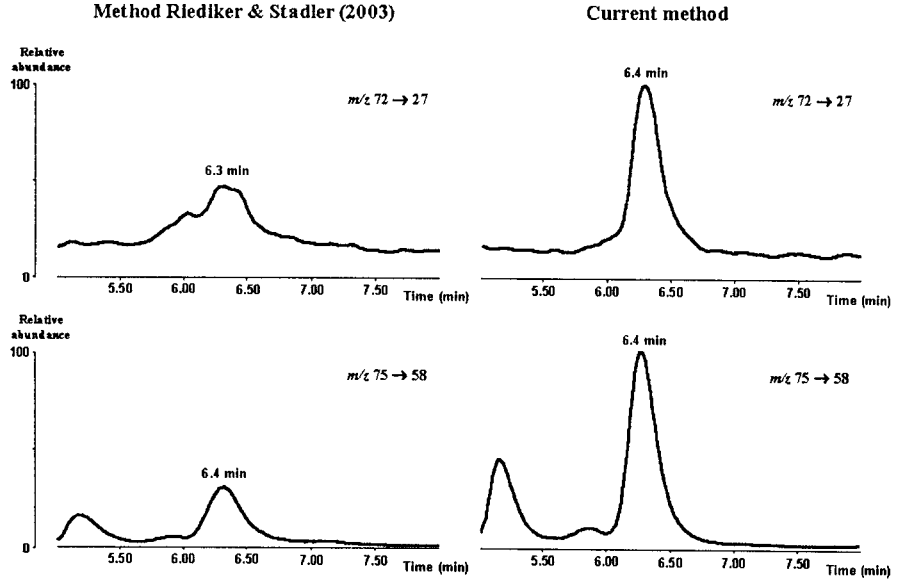

Figure 1. Improvement of the LC-MS/MS methods previously developed in our laboratory to determine acrylamide in "difficult" matrices. Shown is an excerpt of a LC-MS chromatogram of a cocoa powder analyzed using the initial method (left panels) and the improved method (right panels).

Table 1. Precision of the LC-MS/MS method for the quantification of acrylamide in soluble chocolate powder

Spiking level (µg/kg)	Precision (CV in%)	
	Repeatability	Intermediate precision
13	15.8	22.8
305	6.1	9.8
2504	5.4	7.7

The method achieves a limit of determination <10 µg/kg in all food matrices tested, and recovery (not corrected for loss of analyte by isotope dilution) of 43 – 51 % over three concentration ranges. The method was extended to the analysis of acrylamide in various foodstuffs such as mashed potatoes, crisp bread, and butter biscuit and cookies. Furthermore, the accuracy of the method is demonstrated by the results obtained in three inter-laboratory proficiency tests (Delatour et al., in press).

The availability of validated robust and reliable methods to determine acrylamide at low levels in foods are of paramount importance especially for accurate intake assessments. Our laboratory has conducted several hundred analyses of acrylamide on such matrices and can with confidence propose this method as a reference method using isotope dilution LC-MS /MS.

2.1 Stability of acrylamide in food products

Acrylamide may react with inherent food constituents, and we therefore re-analyzed selected dry food products after a certain period of their initial analysis (products kept in their original package at room temperature). As shown in Table 2, acrylamide is stable in certain foods (e.g. breakfast cereals) over prolonged storage periods of up to 12 months.

Table 2. Time-dependent stability of acrylamide in various foodstuffs

Foodstuff	Interval (months)	Difference* (µg/kg)	% Change
Breakfast cereal	12	0	0
Soluble coffee powder	12	515	67
Roasted barley	9	40	15
Roasted coffee	7	56	28
Dried chicory	5	40	19
Roasted chicory	5	620	15
Cocoa	3	3	2

*Initial versus second analysis after the given interval.

On the other hand, loss of acrylamide was appreciable in coffee (roast and ground and soluble) and chicory (dried and roasted) after 5 – 12 months storage. Similar observations for coffee were also recently shown by

Andrzejewski et al. (2004), and it is possible that acrylamide interacts over time with inherent nucleophiles. Further evidence for the reactivity of acrylamide with coffee constituents was demonstrated in an experiment where roast and ground coffee was incubated for 36h at 60°C in closed or open jars. Acrylamide content was reduced by > 20% in the closed jars. The possibility of interaction of acrylamide with volatile nucleophiles (e.g. furfuryl-mercaptan) cannot be excluded and warrants further study.

3. MECHANISMS OF FORMATION

In 2003, several research groups reported more detailed chemical pathways to acrylamide (see Table 3 for a summary). Those published by Yaylayan et al. and Zyzak et al. can be considered the most likely routes by the Maillard reaction. Both groups have shown evidence for the importance of the Schiff base of asparagine, which corresponds to the dehydrated *N*-glucosyl compound. Decarboxylation of the Schiff base is a key step, and the reaction product may furnish acrylamide either directly or via 3-aminopropionamide (Zyzak et al., 2003).

Table 3. Examples of work published in 2003 on the mechanisms of acrylamide formation in foods

Postulated Mechanism(s)	Reference
Decarboxylation of the Schiff base via a oxazolidin-5-one intermediate, tautomer-ization to the decarboxylated Amadori product and subsequent beta-elimination	Yaylayan et al., 2003
Decarboxylation of the Schiff base, heterocyclic cleavage of the imine	Zyzak et al., 2003
Decarboxylation of the Schiff base, hydrolysis of the imine to afford to afford 3-aminopropionamide that subsequently deaminates	Zyzak et al., 2003
Acrylic acid + NH3 (ammonia from thermal degradation of amino acids) (only approx. 5% of the yield compared to asparagine)	Stadler et al., 2003
Acrolein (from triolein) + asparagine Acrolein + NH3 Acrylic acid + NH3 (amino dehydroxylation)	Yasuhara et al., 2003
Acrylic acid from 2-propenal and subsequent reaction of acrylic acid with NH3	Vattem and Shetty, 2003

Alternatively, the corresponding decarboxylated Amadori product, procured by tautomerization of the decarboxylated Schiff base, may release acrylamide by a beta-elimination reaction (Yaylayan et al., 2003). However, so far the key intermediates in food have not been characterized, and therefore the chemical reactions leading to acrylamide remain largely hypothetical.

To provide further evidence for the route supporting a β–elimination as the rate limiting step, we synthesized the decarboxylated Amadori products

and reacted these under low moisture conditions, measuring the formation and yield of the corresponding vinylogous or Strecker degradation products.

The corresponding vinylogous compounds were only generated if a β–proton was available, e.g. styrene from the decarboxylated Amadori compound of phenylalanine (Figure 2) (Blank et al., 2004). Therefore, it is suggested that this thermal pathway may be common to other amino acids resulting under certain conditions in their respective vinylogous reaction products (Stadler et al., 2003).

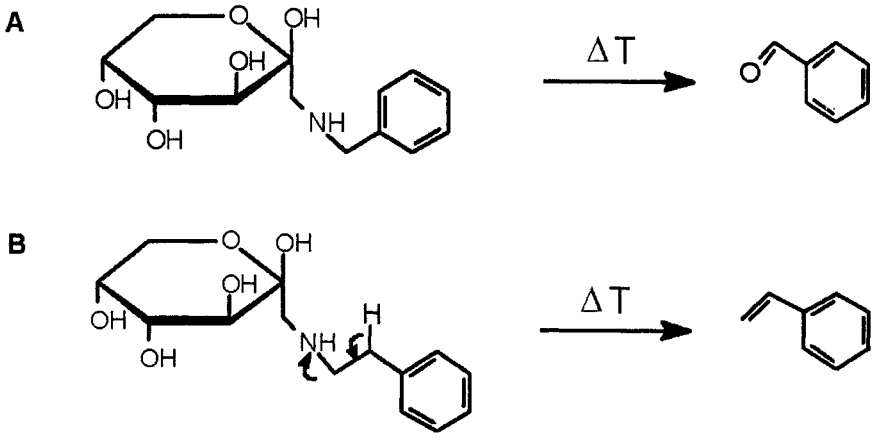

Figure 2. Formation of benzaldehyde (pathway A) and styrene (pathway B) via beta–elimination and Strecker-type degradation, respectively (Blank et al., 2004).

4. FORMATION IN DIFFERENT FOODS AND POTENTIAL MEASURES OF CONTROL

4.1 Potato products

Potato-based foods that are baked, fried, or cooked define a wide range of different products on which much investigative work has been done to date to determine the formation and potential control of acrylamide. Many possible avenues of reduction of acrylamide in potato products, in particular French fries, have been discussed in several recent reports (Biedermann et al., 2002a, 2002b; Noti et al., 2003; Jung et al., 2003;, Haase et al., 2003; Grob et al., 2003). These entail controlling the temperature of storage of the raw potato, selection of certain varieties, and modifying processing (frying) conditions. However, any modifications performed on the raw material constituents will inevitably impact the Maillard reaction and its products,

and concomitantly the organoleptic properties (taste and color) of the cooked food.

Table 4. Impact of different parameters and conditions on acrylamide formation in French fries

Study/Topic	Key Findings	Reference
Frying oil	Type of oil not important	
Max. frying temp. for finish frying	175°C	Taeymans et al., in press
Reduction of added sugar	Significant decrease in acrylamide	
Surface/volume ratio (SVR)	Lower SVR decreases acrylamide	
Variation between batches of prefabricated fries (deep frozen)	Considerable variation in acrylamide under standard frying conditions	Franke et al., 2003
Addition of citric acid	Optimal at 0.75%, decrease factor 2 (also considering loss sugars and Asn during the washing step); but strong impact on organoleptic quality	Biedermann et al., 2002a, Biedermann et al., 2002b
Stability of AA / Elimination reactions	Final amount of AA in potato products is dependent on the balance of formation and elimination	Gama-Baumgartner et al., 2004
Addition of citric acid (1 – 2%, w/w)	Significant reduction, impact on sensory at 2%	Jung et al., 2003
Moisture bound by adsorbents added to frying oil	Reduction of acrylamide by > 40%	Gertz and Klostermann, 2002, Gertz et al., 2003
Addition of citric acid to frying oil	No or only slight reduction	
Silicone added to frying oil	Slight increase in acrylamide formation	
Potato cultivar	Impact on acrylamide formation	
Field site	Impact on acrylamide formation	
Frying : load of the fries	Optimal conditions (10%, i.e. 100g/liter oil)	
Frying: size of the fries, larger fries	Slightly lower amounts of acrylamide (higher area/surface ratio)	Haase et al., 2003, Grob et al., 2003, Taeymans et al., in press
Oil type, additives, to improve heat transfer	No significant differences to the control	
Blanching, soaking	Reduction of acrylamide content	
Endpoint of frying	Recommended at max. 170°C (no general browning, but crispy and with adequate flavor)	

However, even though small scale and laboratory trials have shown that products such as French fries can be prepared with acrylamide amounts below 100 µg/kg (Grob et al., 2003), all these measures must be placed in the perspective of consumer acceptance, not forgetting those related to the supply chain management and logistics of harvesting, storage, and transport

of the raw potatoes. Table 4 summarizes the key findings in mitigation research around French fries.

4.1.1 Impact of Raw Material Variability

The experimental trials that have been conducted so far on acrylamide in potatoes have shown that the major determinants of acrylamide formation are reducing sugars (mainly glucose and fructose) as well as the (free) amino acid asparagine. The content of sugar (glucose/fructose) in the raw potato is well correlated ($r^2 = 0.85$) to the amount of acrylamide formed upon heating (Biedermann et al., 2003b). A wide range of potatoes were analyzed for free amino acids and sugars (glucose, fructose and sucrose). As already documented in several reports, among the various free amino acids measured, the content of free asparagine was the highest. Widely varying concentrations of asparagine, glucose, fructose and sucrose were observed, as also shown recently by Amrein et al. (2003).

This variability may be one important explanation for the difference in the amounts of acrylamide that may be formed in the products during processing. The reasons for this large spread in asparagine and reducing sugars is probably due to multiple factors, such as potato cultivar, farming systems, field site, fertilization, pesticide/herbicide application, time of harvest, storage time and temperature. Clearly more studies will need to be conducted in the future to enable a better understanding of how these many factors may affect the variability of raw material composition.

4.2 Bread and Bakery wares

Only a few reports on the formation of acrylamide in bread and bakery products have been published (Taeymans et al., in press, Springer et al., 2003, Amrein et al., 2004, Surdyk et al., 2004). In crisp bread, acrylamide concentration could be reduced by decreasing the average longitudinal oven baking temperature and increasing the baking time (Taeymans et al., in press). A similar empirical trial approach has been applied to biscuits. Acrylamide is not present in uncooked dough, but the acrylamide level rises rapidly with time. Temperature and cooking time are closely related in the baking process, as is final moisture content, which in some trials has been shown to be inversely proportional to the acrylamide content in the final product. Acrylamide formation has also been studied with regard to ingredients and formulations. The addition of whole wheat flour and bran to biscuit formulas tended to increase acrylamide in comparison with plain counterparts. Reducing the amount of the raising agent ammonium bicarbonate in formulas lowered acrylamide in plain flour matrices. The

addition of lactic acid also lowered acrylamide content in plain flour matrices (Taeymans et al., in press). Experiments are ongoing to determine the relative impact of baking temperature, baking time, final moisture content and biscuit thickness on acrylamide formation. An important observation in general is the large batch-to-batch variation in acrylamide content in biscuits manufactured under industrial conditions and sampled from the line.

A recent study on gingerbread demonstrated that acrylamide is formed evenly over the whole baking process (Amrein et al., 2004). The acrylamide concentration could be considerably lowered when replacing ammonium hydrogencarbonate as raising agent. The same study showed that acidulants such as citric acid also contributed to decreasing the acrylamide content in the final product. In yeast-leavened wheat bread, acrylamide is mainly formed in the crust (99%), and progressively increases with temperature showing a good correlation to color (Surdyk et al., 2004).

4.3 Breakfast Cereals

A marked feature of breakfast cereals is that they possess a clear product identity. They are produced by different and distinct processes that essentially entail a cooking and toasting step. The Maillard reaction develops flavors and color in both the cooking and the toasting steps of the process. Results of trials to date show that most (> 90%) of the acrylamide present in cereals is formed in the toasting step (Taeymans et al., in press). Cereals are characterized by a wide range of acrylamide values observed within and between batches of the same product, processed under the same conditions. This variability creates a difficulty for experimental design and to date no modification to process has had a beneficial effect as large as this variation. Hence, an understanding of what is driving this within-batch and between-batch variation is needed (Taeymans et al., in press).

In model systems, acrylamide begins to form at temperatures >120°C (Stadler et al., 2002). Experimental studies on a wheat biscuit cereal have shown that acrylamide is present in both the surface (270 μg/kg) and cooler centre (60-80 °C, 128 μg/kg) of the biscuit. When biscuits are toasted to the lowest degree compatible with edibility the acrylamide concentration is increased by 15 to 45%. When biscuits are toasted to a near burnt state the acrylamide concentration is decreased by 40 to 50%. Similar results have been seen for several other forms of cereal and during flash frying of potatoes (Taeymans et al., in press), suggesting a balance between formation and elimination with the latter being more rapid at higher temperature.

However, if the conditions of cooking the cereal are varied and the toasting kept constant, then variations in the acrylamide content of the cereal

may be seen. This infers that a precursor formed during the cooking stage may be procured in variable amount and converted to acrylamide at toasting. The wet cooking stage may offer at least as much potential for control of acrylamide content, and as emphasized in the "Mechanisms of Formation" section, it would be important to determine the key intermediates/precursors during food processing.

For cereal as for other model systems those spiked with asparagine generate more acrylamide than controls. In terms of the real process knowledge of the "normal" range of free asparagine content for cereals is important. Further investigations are underway to assess the variability of the amount of free asparagine in different wheat varieties.

4.4 Coffee

Compared to the many other fried, roasted and baked food products, roast and ground coffee has been reported to contain relatively low concentrations (170 – 351 μg/kg on a powder basis) of acrylamide (Friedman, 2003). There are no significant differences in acrylamide concentrations in caffeinated versus decaffeinated coffees. Roast and ground coffee is not consumed as such, but prepared as a beverage. Coffee is prepared by the addition of hot water and subsequent filtration. Hence, calculation of the acrylamide content per cup is an important term of exposure levels (Andrzejewski et al., 2004).

Coffee is typically roasted at temperatures in the range of 220 – 250°C, and the roasting time and speed of roast have an important impact on the sensorial properties (aroma/taste). These are carefully fine tuned to a characteristic profile leading to a clear identity of the coffee product.

Experiments have shown that acrylamide is degraded/eliminated during roasting, and the profile of acrylamide formation during the roasting of coffee reflects this effect very clearly (Taeymans et al., in press). In coffee, acrylamide is formed at the beginning of the roasting step, and toward the end of the roasting cycle a loss of acrylamide seems to dominate. Therefore, light roasted coffees may contain relatively higher amounts of acrylamide than very dark roasted beans. The temperature *per se*, however, does not show a significant difference in the formation of acrylamide. Towards the commercial roasting (color) range, the acrylamide level was reduced by a factor of approximately 10 compared to the highest level recorded during the complete roasting cycle (Taeymans et al., in press). However, higher roasting as a potential option to reduce acrylamide could generate other undesirable compounds and negatively impact the taste/aroma of the product. Consequently, no practical solutions are today at hand that would

reduce acrylamide levels and concomitantly retain the quality characteristics of coffee, since the roasting step cannot be fundamentally changed.

5. CONCLUSION

Over the past two years, researchers from academia, industry, and national authorities/enforcement laboratories, have gained increasing insight in understanding the presence, formation and potential risk to public health posed by the unexpected discovery of acrylamide in some foods. Major progress has been made in the analytical methodology, with good performance of the methods as judged by inter-laboratory trials. The next steps have been defined and entail the development of rapid and cheaper methods to determine acrylamide with adequate accuracy and precision. A further important task is to prepare certified reference materials that can be used as quality control samples by laboratories. The EC-JRC/IRMM has commenced such a study, choosing the most active laboratories to participate in this exercise.

Several reports have been published on the mechanisms of formation of acrylamide, and there is overall consensus on the key role of asparagine Schiff base in the reaction. Concrete evidence of certain intermediates is, however, so far lacking in foods. More than 20 research papers have been published so far on mitigation research in different foods using mainly experimental and pilot-scale conditions. These have provided avenues that may be pursued to reduce acrylamide levels, and as highlighted in this report industry has achieved moderate success in some selected products. However, since acrylamide formation is directly linked to the desired Maillard reaction that generates important flavor and aroma compounds, any measures taken must assess the impact on overall quality and consumer acceptance.

Finally, a concern that needs to be addressed is the lack of knowledge about the effects of final preparation in food service and domestic situations on acrylamide formation. As recommended by WHO/FAO, SCF, and the U.S. FDA, people should not change their dietary habits and continue to eat a balanced diet rich in fruit and vegetables and moderate their consumption of fried and fatty foods.

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REFERENCES

Amrein, T.M., Bachmann, S., Noti, A., Biedermann, M., Barbosa, M.F., Biedermann-Brem, S., Grob, K., Keiser, A., Realini, P., Escher, F., and Amado, R., 2003, Potential of acrylamide formation, sugars, and free asparagine in potatoes: a comparison of cultivars and farming systems, *J. Agric. Food Chem.* **51**: 5556-5560.

Amrein, T. M. Schoenbaechler, B., Escher, F., and Amado, R., 2004, Acrylamide in gingerbread: critical factors for formation and possible ways for reduction, *J. Agric. Food Chem.* **52**: 4282-4288.

Andrzejewski, D., Roach, J.A.G., Gay, M.L., and Musser, S.M., 2004, Analysis of coffee for the presence of acrylamide by LC-MS/MS, *J. Agric. Food Chem.* **52**: 1996-2002.

Becalski, A, Lau, B. P-Y, Lewis, D and Seaman, S., 2003, Acrylamide in foods: Occurrence, sources and modelling, *J. Agric. Food Chem.* **51**: 802-808.

Biedermann, M. Biedermann-Brem, S. Noti, A. and Grob, K., 2002a, Methods for determining the potential of acrylamide formation and its elimination in raw materials for food preparation, such as potatoes, *Mitt. Lebensm. Hyg.* **93:** 653-667.

Biedermann, M., Noti, A., Biedermann-Brem, S., Mozzetti, V. and Grob, K., 2002b, Experiments on acrylamide formation and possibilities to decrease the potential of acrylamide formation in potatoes, *Mitt. Lebensm. Hyg.* **93**: 668-687.

Blank, I., Robert, F., Vuataz, G., Pollien, P., Saucy, F., and Stadler, R. H., 2004, A new insight into the formation of acrylamide via the early Maillard reaction. Abstracts of Papers, 227th ACS National Meeting, Anaheim, CA, United States, March 28-April 1.

Delatour, T., Perisset, A., Goldmann, T., Riediker, S., and Stadler, R.H. Improved sample preparation to determine acrylamide in difficult matrices such as chocolate powder, cocoa, coffee, and surrogates by liquid chromatography tandem mass spectroscopy. *J. Agric. Food Chem.* **52**: 4625-4631.

Franke, K., Kreyenmeier, F., and Reimerdes, E. H., 2003, Acrylamide - taking a comprehensive look at the events surrounding its formation is critical! *Lebensmitteltechnik*; **35 (3)**: 60-62

Friedman, M., 2003, Chemistry, biochemistry, and safety of acrylamide, *J. Agric. Food Chem.* **51**: 4504-4526.

Gama-Baumgartner, F., Grob, K., and Biedermann, M., 2004, Citric acid to reduce acrylamide formation in French fries and roasted potatoes? *Mitt. Lebensm. Hyg.* **95(1)**: 110-117.

Gertz, C., Klostermann, S., and Kochhar, S.P., 2003, Deep frying: the role of water from food being fried and acrylamide formation, *Lipides*, 2003, **10 (4)**: 297-303.

Gertz, C., and Klostermann, S., 2002, Analysis of acrylamide and mechanisms of its formation in deep-fried products, *Eur. J. Lipid Sci. Technol.* **104**: 762-771.

Grob, K., Biedermann, M., Biedermann-Brem, S., Noti, A., Imhof, D., Amrein, T., Pfefferle, A., and Bazzocco, D., 2003, French fries with less than 100 µg/kg acrylamide. A collaboration between cooks and analysts. *Eur. Food Res. Technol.* **217**: 185-194.

Haase, N.U., Matthaeus, B., and Vosmann, K., 2003, Acrylamide formation in foodstuffs – minimizing strategies for potato crisps. *Deutsche Lebensm. Rund.* **99**: 87-90.

Joint European Commission Workshop "Analytical methods for acrylamide determination in food", Joint European Commission (DG Sanco & DG JRC)/Swedish National Food Administration, 28-29th April, 2003.

Jung, M.Y., Choi, D.S., and Ju, J.W., 2003, A novel technique for limitation of acrylamide formation in fried and baked corn chips and french fries, *J. Food. Sci.* **68**: 1287-1290.

Lingnert, H., Grivas, S., Jagerstad, M., Skog, K., Tornqvist, M., and Aman, P., 2002, Acrylamide in food: mechanisms of formation and influencing factors during heating of food, *Scand. J. Nutr.* **46 (4)**: 159-172.

Mottram, D S, Wedzicha, B I, and Dodson, A T. 2002, Acrylamide is formed in the Maillard reaction, *Nature* **419**: 448.

Noti, A., Biedermann-Brem, S., Biedermann, M., Grob, K., Albisser, P., and Realini, P., 2003, Storage of potatoes at low temperature should be avoided to prevent increased acrylamide during frying or roasting, *Mitt. Lebensm. Hyg.* **94**:167-180.

Roach, J.A.G.; Andrzejewski, D.; Gay, M.L.; Nortrup, D.; Musser, S.M. 2003, Rugged LC-MS/MS survey analysis for acrylamide in foods. *J. Agric. Food Chem.*, **51**: 7547-7554.

Riediker, S., and Stadler, R.H., 2003, Analysis of Acrylamide in Food Using Isotope-Dilution Liquid Chromatography Coupled with Electrospray Ionization Tandem Mass Spectrometry, *J. Chrom. A.* **1020**: 121-130.

Sanders, R. A., Zyzak, D. V., Stojanovic, M., Tallmadge, D. H., Eberhart, B. L., and Ewald, D. K., 2002, An LC/MS acrylamide method and it's use in investigating the role of Asparagine. Presentation at the Annual AOAC International Meeting, Los Angeles, CA, September 22 – 26.

Springer, M., Fischer, T., Lehrack, A., and Freund, W., 2003, Acrylamide formation in baked products, *Getreide, Mehl und Brot*, **57(5)**: 274-278.

Stadler, R. H., Blank, I., Varga, N., Robert, F., Hau, J., Guy, P. A., Robert, M-C. and Riediker, S. Acrylamide from Maillard reaction products. *Nature* **419**, 449 (2002).

Stadler, R.H., Verzegnassi, L., Varga, N., Grigirov, M., Studer, A., Riediker, S., and Schilter, B., 2003, Formation of vinylogous compounds in model Maillard reaction systems. *Chem. Res. Tox.* **16**: 1242-1250.

Surdyk, N., Rosen, J., Andersson, R., and Aaman, P., 2004, Effects of Asparagine, Fructose, and Baking Conditions on Acrylamide Content in Yeast-Leavened Wheat Bread, *J. Agric. Food Chem.* **52(7)**: 2047-2051.

Tareke, E. P., Rydberg, P., Karlsson, S., Erikson, M., and Törnqvist, M., 2002, Analysis of acrylamide, a carcinogen formed in heated foodstuffs, *J. Agric. Food Chem.* **50**: 4998-5006.

Taeymans, D., Ashby, P., Blank, I., Gondé, P., van Eijck, P., Lalljie, S., Lingnert, H., Lindblom, M., Matissek, R., Müller, D., O'Brien, J., Thompson, S., Studer, A., Silvani, D., Tallmadge, D., Whitmore, T., Wood, J., and Stadler, R.H., 2004, A review of acrylamide : an industry perspective on research, analysis, formation and control, *Crit. Rev. Food Sci & Nutr.*, in press.

Vattem, D.A., and Shetty, K., 2003, Acrylamide in food: a model for mechanism of formation and its reduction, *Inn. Food Sci. and Emerging Technol.* **4**: 331-338.

Wenzl, T., de la Calle, B., and Anklam, E., 2003, Analytical methods for the determination of acrylamide in food products : a review, *Food Addit. Contam.* **20**: 885-902.

Yasuhara, A., Tanaka, Y., Hengel, M., and Shibamoto, T., 2003, Gas chromatographic investigation of acrylamide formation in browning model systems, *J. Agric. Food Chem.* **51**: 3999-4003.

Yaylayan, V.A., Wnorowski, A., and Locas, C.P., 2003, Why asparagine needs carbohydrates to generate acrylamide, *J. Agric. Food Chem.* **51**: 1753-1757.

Zyzak, D., Sanders, R.A., Stojanovic. M., Tallmadge, D., Eberhart, B.L., Ewald, D.K., Gruber, D.C., Morsch, T.R., Strothers, M.A., Rizzi, G.P., Villagran, M.D., 2003, Acrylamide formation mechanism in heated foods, *J. Agric. Food Chem.* **51**: 4782-4787.