Use of Ultrasound to Reduce Acrylamide Formation in French Fries

Fadwa Al-Taher[1], Lauren Jackson[2] and Jack Cappozzo[1]

[1]National Center for Food Safety and Technology, Illinois Institute of Technology
[2]National Center for Food Safety and Technology, Food and Drug Administration, CFSAN

ABSTRACT

Acrylamide is formed in high carbohydrate foods at elevated cooking or processing temperatures. There is interest in reducing acrylamide, a potential carcinogen, in fried potato products such as French fries. The objectives of this study were to (1) assess the effects of ultrasound-aided frying on acrylamide formation in French fries, and (2) evaluate the use of an ultrasound pretreatment on acrylamide generation in French fries prepared from asparaginase-treated potato slices. Frozen par-fried French fries were fried in corn oil at 177°C or 185°C (3-5 min) with and without sonication. Separate experiments measured acrylamide in French fries prepared from fresh potato slices pretreated with asparaginase (40°C for 10 min; 55°C for 3 min) with and without ultrasound. Although ultrasound during frying did not decrease acrylamide levels in French fries, it reduced the frying times needed to produce an acceptable product. Exposing raw potato strips to ultrasound with asparaginase at 55°C (767 ± 117 ng/g) was more effective than the enzyme treatment alone (1126 ± 89.0 ng/g) for reducing acrylamide levels in the slices during frying. Acrylamide levels in fried potato slices pretreated with sonication and asparaginase at 40°C (518 ± 58.5 ng/g) were similar compared to those exposed to enzyme alone (491.8 ± 47.0 ng/g). Ultrasound treatments may be useful in aiding asparaginase treatment of potato slices. However, more work is needed to optimize the process.

INTRODUCTION

High temperature processing generates acrylamide in carbohydrate-rich foods, such as potato products and baked goods. Acrylamide has been classified as a possible carcinogen in humans, so it is important to limit its formation in these products without compromising food quality and safety (1). Reducing processing time and/or temperature is an effective way to decrease acrylamide formation in foods. In the potato processing industries, it is possible to reduce acrylamide precursors (i.e. asparagine and reducing sugars) and therefore, acrylamide formation by blanching, soaking in water or acidic solutions (2) or treating raw potatoes with the enzyme, asparaginase (3). Asparaginase decreases acrylamide formation in bakery products by hydrolyzing asparagine to aspartic acid and ammonia and is added to the dough during kneading. Treating potato slices is more challenging as the solid nature of these whole-cut products limits enzyme-substrate contact (4). It is difficult for asparaginase to penetrate through the surface of the potato slices to access asparagine.

Ultrasound processing has many applications in the food industry. It has proved useful in the production of yoghurt, the cleaning of equipment surfaces, and the processing of meat products by reducing processing times and increasing efficiency (5). Ultrasound also has been used to aid in extraction and filtration and has been shown to increase enzyme activity (5). Our hypothesis is that use of ultrasound processing during frying could be used to reduce the acrylamide content of French fries by reducing frying time. Another goal was to investigate the use of ultrasound to enhance acrylamide mitigation in fried potato products by improving the ability of asparaginase to remove one of the acrylamide precursors, asparagine, from potato slices.

OBJECTIVES

1. Assess the effects of ultrasound-aided frying on acrylamide formation in French fries
2. Evaluate the use of an ultrasound pretreatment on acrylamide generation in French fries prepared from asparaginase-treated potato slices.

MATERIALS AND METHODS

Materials and Chemicals

Frozen par-fried French fries (9.5 x 9.5 mm straight cut, GloriFri, Simplot) and whole raw potatoes (Russet Burbank) were purchased from a local supermarket. The enzyme, asparaginase (3500 ASNU/g, Acrylaway® L), was obtained from Novozymes (Denmark). Unlabeled acrylamide and 1,2,3-$^{13}C_3$ acrylamide (internal standard) were purchased from Sigma-Aldrich (St. Louis, MO) and Cambridge Isotope (Andover, MA), respectively. Acetonitrile, n-hexane, formic acid and water were purchased from Fisher Scientific (Hanover Park, IL). All reagents were Optima or LC-MS grade.

Unlabeled acrylamide standard stock solution (1000 µg/mL) was prepared by dissolving 100 mg of unlabeled acrylamide in 100 mL acetonitrile. An internal standard stock solution (10 µg/mL) was made by diluting (1:100) the 1000 µg/mL labeled standard with acetonitrile. Stock solutions were stored at 4°C. All working standard solutions were prepared daily by serially diluting the stock solutions with acetonitrile.

Sample Preparation

Part I. Effect of sonication-aided frying on acrylamide levels in French fries.

Three hundred gram batches of frozen par-fried French fries were finish fried (Waring Commercial Deep Fryer, Torrington, CT) in four liters of corn oil (Mazola; ACH food companies, Memphis, TN) for 3, 4, and 5 min at 177°C or 185°C. A 400W sonicator probe (Hielscher Ultrasound Technology; Teltow, Germany) was then set up in the deep fryer (Figure 1) to sonicate the oil while separate batches of French fries were fried at the same times and temperatures as the non-sonicated controls. Control and sonication frying experiments were performed in triplicate for each time/temperature combination. Surface color measurements were taken for each fried sample with a Hunter Labscan XE reflectance colorimeter (Reston,VA).

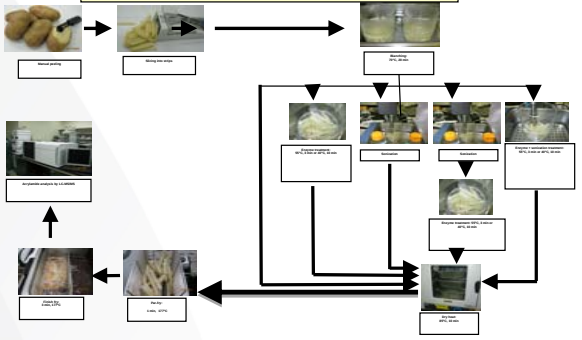
Figure 1. Ultrasound-aided frying of par-fried frozen French fries.

Part II. Effect of washing pretreatments with and without sonication on acrylamide formation in potato strips.

Three bags of potatoes were purchased and the potatoes were randomized. The potatoes were peeled and then sliced into strips (9mm x 9mm). A blanching step was used for all pre-treatments and consisted of immersing the strips in hot (70°C) distilled water for 20 min. The strips were then dried in a convection oven at 85°C for 10 min. Potato strips treated with this series of processing steps served as our control. Other treatments involving the use of asparaginase and/or sonication were added after the blanching step. The potato strips were immersed in a 7500 ASNU/L asparaginase solution at two different conditions: 55°C for 3 min or 40°C for 10 min (ratio of potato to enzyme solution 1:2). A sonication (5 min) step was added 1) after the blanching step (no enzyme treatment), (2) after the blanching step, and before the enzyme was added, and (3) after blanching and during enzyme treatment.

After the treatments, the potato strips were dried in a convection oven at 85°C for 10 min, and then par-fried in 4 liters of corn oil at 177°C for 1 min. Par-fried potatoes were frozen and stored at -20°C in plastic bags for two days before final frying. Final frying consisted of frying the par-fried slices at 177°C for 3 min. Figure 2 illustrates the treatments used to study the effect of sonication and/or enzyme treatment on acrylamide levels in French fries. Each experiment was conducted four times. Surface color measurements were taken for each fried sample with a Hunter Labscan XE reflectance colorimeter (Reston,VA).

Figure 2. Enzyme and ultrasound pretreatment of fresh potato strips



Extraction of Acrylamide

French fries were homogenized using a Power Pro™ II Food Processor (Black & Decker, Towson, MD). A QuEChERS extraction method developed by Mastovska and Lehotay (6) was used for quantifying acrylamide in potato samples. One g of French fries was weighed into a 50 mL centrifuge tube obtained from Agilent's SampliQ QuEChERS Extraction kit (Agilent Technologies; Santa Rosa, CA). The internal standard ($^{13}C_3$-acrylamide) was added at 0.5 µg/g. Hexane (5 mL) was added, and then the tube was vortexed. Water (10 mL) and acetonitrile (10 mL) were then added followed by the Agilent SampliQ QuEChERS extraction salt mixture for acrylamide, which contained 4 g of anhydrous magnesium sulfate ($MgSO_4$) and 0.5 g of sodium chloride. The sample tubes were vigorously shaken for 1 min, and then centrifuged at 5000 rpm for 5 min. The hexane layer was discarded and 1 mL of the acetonitrile extract was transferred to a 2 mL Agilent SampliQ QuEChERS AOAC Dispersive SPE tube containing 50 mg of primary secondary amine (PSA) and 150 mg anhydrous $MgSO_4$. The tubes were vortexed for 30 sec and then centrifuged at 5000 rpm for 1 min. The supernatant was then placed in an autosampler vial for LC-MS/MS analysis. All samples were analyzed extracted and analyzed for acrylamide content four times.

LC-MS/MS Analysis

Analysis was performed on an Agilent 1200 Series HPLC equipped with an ESI 6460 triple quadrupole mass spectrometer (Agilent Technologies). Separations were performed on an Atlantis reversed phase C_{18} column, 2.1 mm x 150 mm, 3 µm film thickness (Waters Corp.; Milford, MA, USA) at 35°C. The elution mode was isocratic using 2.5% methanol/ 97.5% of 0.1% formic acid as the mobile phase flowing at a rate of 0.2 mL/min. Data was processed by Mass Hunter software.

Acrylamide was identified by multi-reaction monitoring (MRM) in positive electrospray ionization mode (ESI+) with jet stream technology. Three different fragment ion transitions were monitored for both acrylamide (m/z 72 → 72, m/z 72 → 55 and m/z 72 → 27) and the internal standard, $^{13}C_3$-acrylamide, (m/z 75→ 75, m/z 75 → 58 and m/z 75 → 29). The fragment voltage was 50V and the collision energy voltage was 0V and 16V for the acrylamide product ions having m/z of 55 and 27, respectively. For the acrylamide internal standard, the fragmentor voltage was 50V and the collision energy voltage was 0V and 8V for the internal standard product ions having m/z of 58 and 29, respectively. The electrospray source had the following settings (with nitrogen): capillary voltage 4kV; nozzle voltage 500V; source temperature 325°C at 5 L/min; drying gas temperature 350°C at 11 L/min with nebulizer pressure at 35 psi.

Statistical Analysis

Statistical analysis was performed with one-way ANOVA followed by Tukey's multiple comparison test at p<0.05 using Minitab® (State College, PA) statistical software.

RESULTS AND DISCUSSION

Part I. Effect of ultrasound-aided frying on acrylamide levels in French fries:

- As the fries were fried for longer periods of time and at higher temperatures, the "L" component of color (a measure of the white/black component of color) decreased while the "a" values (degree of redness) increased for both non-sonicated and sonicated fries (data not shown).
- Under equal frying conditions (time/temperature), use of ultrasound during frying increased acrylamide levels in French fries (Figure 3).

Part II. Effect of washing pretreatments with and without sonication on acrylamide formation in fried potato strips:

- There was no significant differences (p>0.05) in the surface color (determined visually and with a colorimeter) of the finish-fried potato strips exposed to the different washing treatments (data not shown).
- Treating raw potato strips with ultrasound + asparaginase at 55°C was more effective at reducing acrylamide levels (767 ± 117 ng/g) in finish-fried potato strips than exposing the strips to asparaginase alone (1126 ± 89.0 ng/g) at 55°C (Figure 4).
- Acrylamide levels in finish-fried potato strips pre-treated with ultrasound + asparaginase at 40°C (518 ± 58.5 ng/g) were similar compared to those exposed to the enzyme alone (491.8 ± 47.0 ng/g) at 40°C (Figure 4).

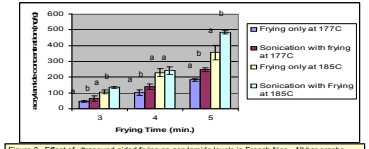


Figure 3. Effect of ultrasound-aided frying on acrylamide levels in French fries. All bar graphs represent the average ± standard deviation of four trials. Bar graphs with different letters for the same temperature and time are significantly different (p<0.05).

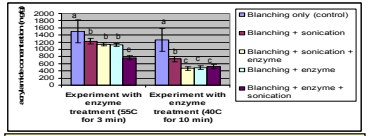


Figure 4. Effect of washing pretreatments with and without sonication on acrylamide formation in finish-fried potato strips. All bar graphs represent the average ± standard deviation of four trials. Bars graphs with different letters for the same set of experiments are significantly different (p<0.05).

CONCLUSIONS

- Although ultrasound during frying did not reduce acrylamide levels in French fries, it may aid in reducing frying times needed to produce an acceptable product.
- Ultrasound treatments may be useful in aiding asparaginase treatment of potato slices to cause greater reductions in acrylamide levels during finish frying.
- It will be beneficial to study enzyme activity at different temperatures and determine what conditions (time, temperature, concentration) asparaginase and sonication can be used to further reduce acrylamide levels in French fries.

REFERENCES

1. Tareke, E., Rydberg, P., Karlsson, P., Eriksson, S., and Tornqvist, M. Analysis of acrylamide, a carcinogen formed in heated foodstuffs. *J Agric Food Chem* **2002,** 50, 4998-5006.
2. Pedreschi, F., Kaack, K., and Granby, K. Acrylamide content and colour development in fried potato strips. *Food Research International* **2006,** 39, 40-46.
3. Pedreschi, F., Kaack, K., and Granby, K. The effect of asparaginase on acrylamide formation in French fries. *Food Chem* **2008**, *109*, 386-392.
4. Hendriksen, H.V., Kornbrust, B.A., Ostergaard, P.R., and Stringer, M.A. Evaluating the potential for enzymatic acrylamide mitigation in a range of food products using an asparaginase from *Aspergillus oryzae*. *J. Agric Food Chem* **2009**, *57*, 4168-4176.
5. Mason, T.J., Paniwnyk, L., and Lorimer, J.P. The uses of ultrasound in food technology. *Ultrasonics Sonochemistry*, **1996**, *3*, S253-S260.
6. Mastovska, K. and Lehotay, S.J. Rapid sample preparation method for LC-MS/MS or GC-MS analysis of acrylamide in various food matrices. *J. Agric. Food Chem*, **2006**, *54*, 7001-7008.

National Center for Food Safety and Technology
Advancing Innovations in Food Safety and Nutrition Science www.iit.edu/ncfst

FDA CFSAN

ILLINOIS INSTITUTE OF TECHNOLOGY