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Kinetic Model for the Formation of Acrylamide during the Finish-Frying of Commercial French Fries

Jane K. Parker,[†] Dimitrios P. Balagiannis,[†] Jeremy Higley,[‡] Gordon Smith,[‡] Bronislaw L. Wedzicha,[§] and Donald S. Mottram*[,†]

[†]Department of Food and Nutritional Sciences, University of Reading, Whiteknights, Reading RG6 6AP, United Kingdom

[‡]ConAgra Foods, Six ConAgra Drive, Omaha, Nebraska 68102, United States

[§]School of Food Science and Nutrition, University of Leeds, Leeds LS2 9JT, United Kingdom

ABSTRACT: Acrylamide is formed from reducing sugars and asparagine during the preparation of French fries. The commercial preparation of French fries is a multistage process involving the preparation of frozen, par-fried potato strips for distribution to catering outlets, where they are finish-fried. The initial blanching, treatment in glucose solution, and par-frying steps are crucial because they determine the levels of precursors present at the beginning of the finish-frying process. To minimize the quantities of acrylamide in cooked fries, it is important to understand the impact of each stage on the formation of acrylamide. Acrylamide, amino acids, sugars, moisture, fat, and color were monitored at time intervals during the frying of potato strips that had been dipped in various concentrations of glucose and fructose during a typical pretreatment. A mathematical model based on the fundamental chemical reaction pathways of the finish-frying was developed, incorporating moisture and temperature gradients in the fries. This showed the contribution of both glucose and fructose to the generation of acrylamide and accurately predicted the acrylamide content of the final fries.

KEYWORDS: *acrylamide, asparagine, glucose, fructose, French fries, kinetic modeling, multiresponse modeling*

INTRODUCTION

Acrylamide is a probable carcinogen[1] formed during the high-temperature processing of asparagine-rich foods, such as potato, wheat, and rye products. Over the past 10 years, much effort has been devoted to understanding the formation pathway and developing mitigation strategies.[2,3] These include optimization of the frying time/temperature profile,[4] optimization of potato storage conditions,[5,6] and pretreatments, such as soaking and blanching[5,7] and altering glucose/fructose ratios,[8] as well as selective breeding programs.[9,10]

It is evident that acrylamide is formed during the reaction between asparagine and reducing sugars, and various mechanisms have been proposed.[11−16] The pathway involves the Maillard reaction; therefore, acrylamide formation is inextricably linked with the development of color and flavor. In the development of strategies to mitigate the formation of acrylamide in processed food products, it is necessary to be able to control and predict acrylamide levels while maintaining the desirable color and flavor of the product. Thus, an understanding of the kinetics of the reactions leading to acrylamide formation has considerable importance in the development of such strategies. Because of the complexity of the reaction, a mathematical approach, based on the fundamental chemical reaction pathways, is required to model the reaction kinetics. A very successful kinetic multiresponse model of the Maillard reaction was originally proposed by Wedzicha et al. in 1984.[17] Although initially developed for color formation in an aqueous model system, this model has subsequently been applied to the generation of acrylamide[15,18] and has been shown to hold for various simplified food matrices including potato doughs (e.g., from flake). Related models have been developed by De Vleeschouwer et al. to take into account different factors that can influence the reaction.[19] Various versions of the model have been used to probe the roles of sugars, moisture content, other amino acids, and pH.[20−24] All modeling studies to date have involved either aqueous solution or simple potato powder mixtures. The current challenge is to apply multiresponse modeling to the formation of acrylamide in a complex food matrix.

The kinetic modeling of acrylamide formation in a real food, such as French fries, is complicated by the shape of the fry and the fact that most of the color is formed at the surface. Given that color forms as a result of the Maillard reaction, it is likely that acrylamide formation also occurs predominantly at the surface, where the temperature is high and the moisture content low. Thus, consideration of the temperature and moisture profiles of the potato strip is necessary when the formation of acrylamide in French fries is modeled. Heat and mass transfer properties during the frying of a potato strip have been determined both experimentally and mathematically,[25−28] and Palazoglu has combined physical parameters with a simplified Maillard reaction scheme to predict acrylamide formation during the frying of raw potato strips.[29] The industrial preparation of French fries is further complicated because the potatoes undergo washing, cutting, blanching, heating, and treatment in dilute glucose solution, prior to finish-frying, which alter both the texture and the precursor concentrations

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of the fry.[5,7,30] Typically they are then partially fried (par-fried) for approximately 1 min before freezing and distribution to catering outlets, where they are finish-fried ready for consumption.

The aim of this study was to produce a kinetic model that would provide a better understanding of the underlying mechanism of acrylamide formation and accurately simulate the generation of acrylamide during the finish-frying of commercially preprocessed French fries.

EXPERIMENTAL PROCEDURES

Chemicals. Acrylamide, asparagine, sucrose, fructose, glucose, maltose monohydrate, and lactose monohydrate were obtained from Sigma-Aldrich (St. Louis, MO, USA); $^{13}C_3$-labeled acrylamide was from Isotec (St. Louis, MO, USA); *o*-phthalaldehyde and 9-fluorenylmethoxycarbonyl chloride were from Pierce (Rockford, IL, USA); norvaline, sarcosine, 3-mercaptopropionic acid, formic acid, and boric acid were from Acros (Pittsburgh, PA, USA); and sodium acetate, sodium phosphate monobasic monohydrate, glacial acetic acid, hydrochloric acid, and sodium hydroxide were from Fisher (Pittsburgh, PA, USA). HPLC grade methanol, acetonitrile, and reagent ethanol were obtained from J. T. Baker (Phillipsburg, NJ, USA). Deionized water (18MΩ) was prepared using an Elga Ultrapure Water System (Woodridge, IL, USA). Solutions containing acrylamide and $^{13}C_3$-labeled acrylamide were prepared and stored in "low actinic" glassware to minimize exposure to light.

Preparation of Potato Fries. Ranger Russet potatoes, 7.5−12.5 cm in length, from the 2011 harvest were stored at 7−9 °C before preparation as a ready-to-fry product typical of that used by catering outlets (shoestring fries). Raw potatoes were processed in 6−7 kg batches to yield 3−4 kg of partially fried, frozen French fries. The potatoes were washed and peeled and preheated in 54 °C water (∼150 L) for 30 min before they were cut into strips with a cross-section of 7.5 × 7.5 mm. The potato strips were blanched for 10 min in water heated to 74 °C (∼70 L) and then dipped in glucose (potato batch G) or fructose (potato batch F) solution at 70−75 °C for 30 s. Fresh blanch water was used after the processing of two batches of potatoes. They were dried at 60 °C to achieve about a 10% weight loss before par-frying in canola oil at 190 °C for 50 s. The strips were par-fried in ∼225 g batches in a 20.5 kW Frymaster deep fryer with an oil volume of ca. 30 L (model EH21721T; Shreveport, LA, USA). At this stage the fries were frozen at −23 °C, as occurs in commercial practice. Five glucose and five fructose concentrations were used in the dipping solutions: 0, 0.5, 1.0, 1.5, and 2.0% (w/v). Samples of potato strips were also taken after cutting and before par-frying and frozen for subsequent analysis.

The frozen par-fried material (∼225 g) was finish-fried in canola oil (in the same fryer as described above) at 165, 175, or 185 °C for times between 30 s and 5 min. A mechanical "hold-down" was used to keep the strips from floating during the frying process. Preliminary experiments were also carried out for extended frying times up to 15 min. At the end of the frying time, the fries were immediately submerged in liquid nitrogen for 40 s for rapid cooling. The cooked fries were then held in frozen storage and subsequently homogenized in a Robot Coupe processor (Jackson, MS, USA) in preparation for further analysis.

Measurement of Water and Total Fat Content. Oven-drying was used to determine the moisture content of the fries. Samples (∼2 g) were weighed accurately into predried and weighed aluminum oven-drying dishes. The samples were dried to constant weight at 125 °C in a fan oven (3−4 h).

Fat and fatty acids were extracted from the fries (2 g) by acid hydrolysis using 2 mL of ethanol and 10 mL of 8.3 M HCl on the basis of AOAC method 996.06. [31] Pyrogallic acid was added to minimize oxidative degradation of fatty acids during analysis and the triglyceride, triundecanoin ($C_{11:0}$), was added as internal standard. The fat was extracted into petroleum ether/diethyl ether (1:1) and then methylated to fatty acid methyl esters (FAMEs) using BF_3 in methanol. FAMEs were quantitatively measured by capillary gas chromatography against $C_{11:0}$ internal standard. Total fat was calculated as the sum of individual fatty acids expressed as triglyceride equivalents.

The moisture and fat contents of fries were used to calculate the concentrations of acrylamide and its precursors in fries on a dry, fat-free weight basis.

Color Measurement. The color of finish-fried strips was assessed with an Agtron E-30 Analyzer (Agtron Inc., Reno, NV, USA), which is designed specifically to evaluate color changes (degree of darkness) due to frying of French fries. The Agtron measures reflectance in two spectral modes: near-infrared and green. The ratio of the two is displayed as the Agtron reading.

Temperature Measurement in Fries during Cooking. Commercially processed par-fried frozen potato strips (7.5 × 7.5 mm) were thawed at ambient temperature to facilitate insertion of thermocouple wires into the ends of three strips (insertion depth was approximately 10−12 mm, with the tip of the wire as close to the center of the fry as reasonably achievable). The strips were then blast frozen at −23 °C. The frozen strips, with imbedded thermocouple wires, were combined with other frozen strips (0.22 kg) and fried in canola oil at 165, 175, or 185 °C for 5 min. The internal temperature of the strips was recorded every 2 s during frying using an Omega 4-Channel Datalogger Thermometer (model RDXL4SD) (Omega Engineering Inc., Stamford, CT, USA). With each batch, an additional thermocouple wire was wrapped around one strip to monitor the temperature just above the surface of the fry. A mechanical "hold-down" was used to keep the strips from floating during the frying process.

Acrylamide Analysis. The determination of acrylamide used the method reported by Higley et al.[30] and was based on that described by Roach et al.[32] Potato (1.0 g) was mixed with 9.9 mL of deionized water, and 0.1 mL of $^{13}C_3$-labeled acrylamide internal standard (2 μg/mL in 0.1% (w/v) formic acid) was added. After centrifuging, an aliquot (1.5 mL) of the aqueous layer was taken for cleanup by solid phase extraction (SPE) using Oasis HLB and BondElut Accucat SPE cartridges as described previously.[30] LC-MS/MS analysis of the extracts was carried out on an Aglient 1200 series HPLC system (Santa Clara, CA, USA) coupled to an Applied Biosystems Q-Trap 4000 MS/MS detector (Carlsbad, CA, USA). Samples (20 μL) were eluted on a Phenomenex Hydro-RP 80 A analytical column (4 μm, 250 mm × 2 mm, Torrance, CA, USA) at 26 °C. The mobile phase consisted of 0.5% methanol/0.1% acetic acid in deionized water, with a flow rate maintained at 0.2 mL/min. MS/MS was performed in positive ESI mode as described previously.[30] Transitions m/z 72 → 55 and 75 →> 58 were used in the identification and quantitation of acrylamide and $^{13}C_3$-labeled acrylamide, respectively.

Determination of Free Amino Acids. The analysis of free amino acids in the potato fries used the method described by Higley et al.[30] Analyses were carried out using an Agilent 1100 series HPLC system equipped with a robotic autosampler and a fluorescence detector. The amino acids were derivatized by *o*-phthalaldehyde (OPA) and 9-fluorenylmethoxycabonyl chloride (Fmoc-Cl) in the autosampler immediately before HPLC injection.

Homogenized potato (2−5 g) was dispersed in 30 mL of 20 mM acetate buffer (pH 4.25) and shaken for 30 min using a mechanical shaker. The extract was filtered through a 0.45 μm glass microfiber filter (Whatman, Piscataway, NJ, USA), and 1 mL filtrate was transferred into an autosampler vial containing 100 μL of internal standard solution (50 mg each of norvaline and sarcosine dissolved in 100 mL of 20 mM acetate buffer at pH 4.5). After derivatization in the robotic autosampler, the separation of the amino acids was accomplished at a flow rate of 0.9 mL/min using 40 mM NaH_2PO_4 (at pH 7.8) as mobile phase A and acetonitrile/methanol/deionized water in a ratio of 45:45:10 (v/v/v) as mobile phase B with the following elution profile: 0−1.9 min, isocratic at 100% A; 1.9−18.1 min, linear gradient to 57% B; 18.1−18.6 min, linear gradient to 100% B; 18.6−22.3 min, isocratic at 100% B (to wash column); 23.2−30 min, 100% A (to equilibrate column).

Table 1. Concentrations of Precursors and Acrylamide (Expressed as Fat-Free Dry Weight) at Three Stages during the Preprocessing

			concentration of glucose dip				
		raw potato	0%	0.5%	1%	1.5%	2%
moisture (%)	raw potato	78					
	pre-par-fry		78	77	77	76	77
	post-par-fry		63	63	62	61	63
glucose (mmol/kg dry wt)	raw potato	30					
	pre-par-fry		14	30	39	48	66
	post-par-fry		16	27	36	48	60
fructose (mmol/kg dry wt)	raw potato	23					
	pre-par-fry		11	13	13	12	16
	pos-tpar-fry		14	13	14	13	16
sucrose (mmol/kg dry wt)	raw potato	33					
	pre-par-fry		18	19	18	18	20
	post-par-fry		24	21	21	21	21
asparagine (mmol/kg dry wt)	raw potato	89					
	pre-par-fry		64	60	60	54	60
	post-par-fry		62	60	59	57	58
total amino acids (mmol/kg dry wt)	raw potato	239					
	pre-par-fry		177	171	172	146	163
	post-par-fry		166	168	162	155	156
acrylamide (μmol/kg dry wt)	raw potato	nd[a]					
	pre-par-fry		nd	nd	nd	nd	nd
	post-par-fry		1	4	5	8	6
fat content (g/100 g dry wt)	raw potato	0.2					
	pre-par-fry		0.6	0.6	0.8	0.8	0.6
	post-par-fry		21	21	22	20	20

[a]nd, not detected.

Standards containing 18 amino acids (Asn, Gln, Arg, Ala, Gla, Val, Asp, Ser, Lys, Phe, Tyr, Thr, Ile, Met, Pro, His, Leu, and Gly) were prepared from an amino acid mix (Sigma-Aldrich) and diluted with 20 mM acetate buffer (pH 4.25) to give concentrations ranging from 10 to 1250 nmol/mL. These were mixed with the internal standard and derivatized as described above.

Determination of Sugars. Sugar contents of the potato fries were analyzed according to AOAC method 977.20,[31] with modifications as described by Higley et al.[30] Homogenized potato sample (0.5−5.0 g) was extracted with 25 mL of 1:1 water/ethanol and centrifuged at 3200*g* for 4 min. The supernatant was filtered through a 0.45 μm glass microfiber filter before HPLC analysis using an Agilent 1200 series HPLC system equipped with an Alltech evaporative light scattering detector (ELSD, model 3300; Lexington, KY, USA). The injected sample (2.5 μL) was eluted on an Alltech Prevail Carbohydrate ES column (5 μm, 250 mm × 4.6 mm) at 30 °C. The mobile phase consisted of 75% acetonitrile/25% water; the flow rate for the HPLC system was 1.0 mL/min. Eluted sugars were detected by ELSD with a nebulizer gas flow of 1.5 mL/min nitrogen and evaporative temperature of 48 °C.

Kinetic Modeling. Multiresponse modeling was performed using the Athena Visual Studio software package (Athena Visual Software Inc., Naperville, IL, USA).

RESULTS AND DISCUSSION

Changes Occurring during Preparation of Par-Fried Potato Strips. In a typical commercial French fry process, the raw potato strips are subjected to a series of heating, blanching, and glucose treatment (dipping) steps, which alter the composition and texture of the potato.[30] The impact of the initial preheating step is likely to be small, because the temperature is below the gelatinization temperature of potato starch and the potatoes are still whole. However, gelatinization and uptake of water occur during the blanching of the potato strips with leaching of key precursors from the potato strip.[5,30] The subsequent dipping process is fundamental to controlling the glucose concentrations in the potato, which is followed by a drying step resulting in an approximately 10% loss of moisture. The subsequent par-frying occurs at high temperature (190 °C) and, although, this is only for a short time, it will promote the early stages of the Maillard reaction.

Table 1 shows the impact of the pretreatment on the composition of potato strips that had been subjected to the standard pretreatment process and then dipped in different concentrations of glucose (0, 0.5, 1.0, 1.5, or 2%). It shows the concentration of precursors and acrylamide present at three key points during the pretreatment. For all five dip concentrations, the moisture content of the potato strips entering the par-frying process was approximately the same value as that of the raw potato, indicating that there was a 10% uptake of water during the blanching and dipping steps, to compensate for the 10%

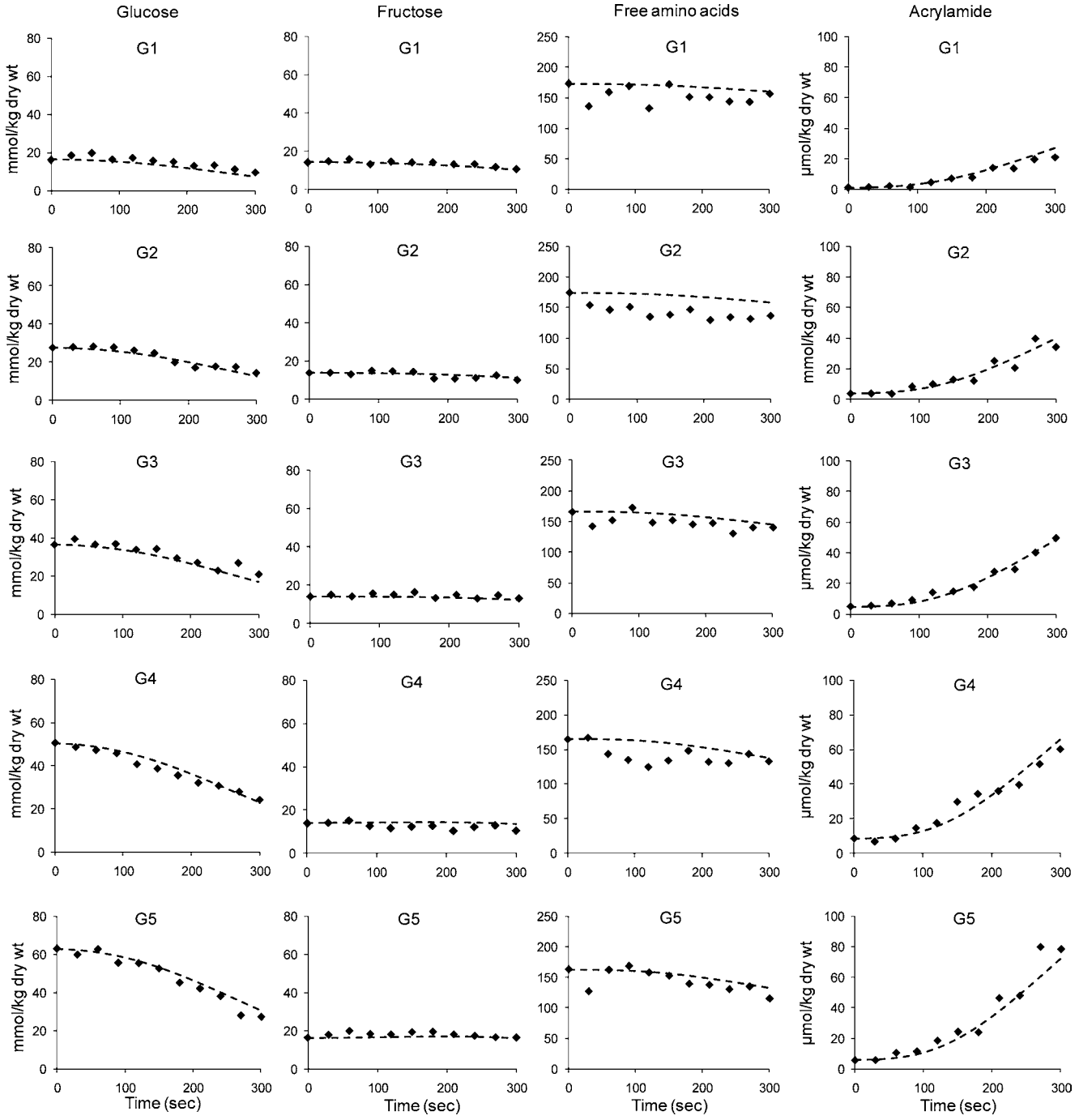

Figure 1. Glucose, fructose, total amino acids, and acrylamide concentrations as a function of time (0−5 min) during finish-frying of five batches of glucose-dipped potato strips at 165 °C (G1 = 0% dip, G2 = 0.5% dip, G3 = 1% dip, G4 = 1.5% dip, G5 = 2% dip). Symbols (◆) are the experimental data points for potato batch G, and the dashed line (- - -) shows the kinetic model derived from the combined data set using the rate equations shown in Figure 6 and the parameters shown in Table 2.

loss during the drying step. After par-frying (190 °C for 1 min), 15% of the moisture was lost, as steam.

During dipping and drying (pre-par-fry), glucose levels increased linearly with the dip concentration. The glucose concentration in the raw potato was 30 mmol/kg, which subsequently increased to 66 mmol/kg after dipping in the 2% glucose dip (111 mmol/kg). Sucrose, fructose, glucose (in the instance when no glucose was added in the dipping solution), and free amino acid concentrations were significantly lower in the pre-par-fry potato, compared with the raw potato, due to the leaching out of these water-soluble compounds during the blanching steps. At the pre-par-fry stage, acrylamide was not detected, but low levels were formed in all five batches after the par-fry.

These results show that the pretreatment is extremely important for manipulating the concentrations of important precursors of acrylamide (and flavor/color), confirming recent observations on similar French fries.[30] This information enabled selection of appropriate parameters for the experimental designs.

Changes Occurring during Finish-Frying of Par-Fried Potato Strips. Portions of frozen par-fried potato strips, which

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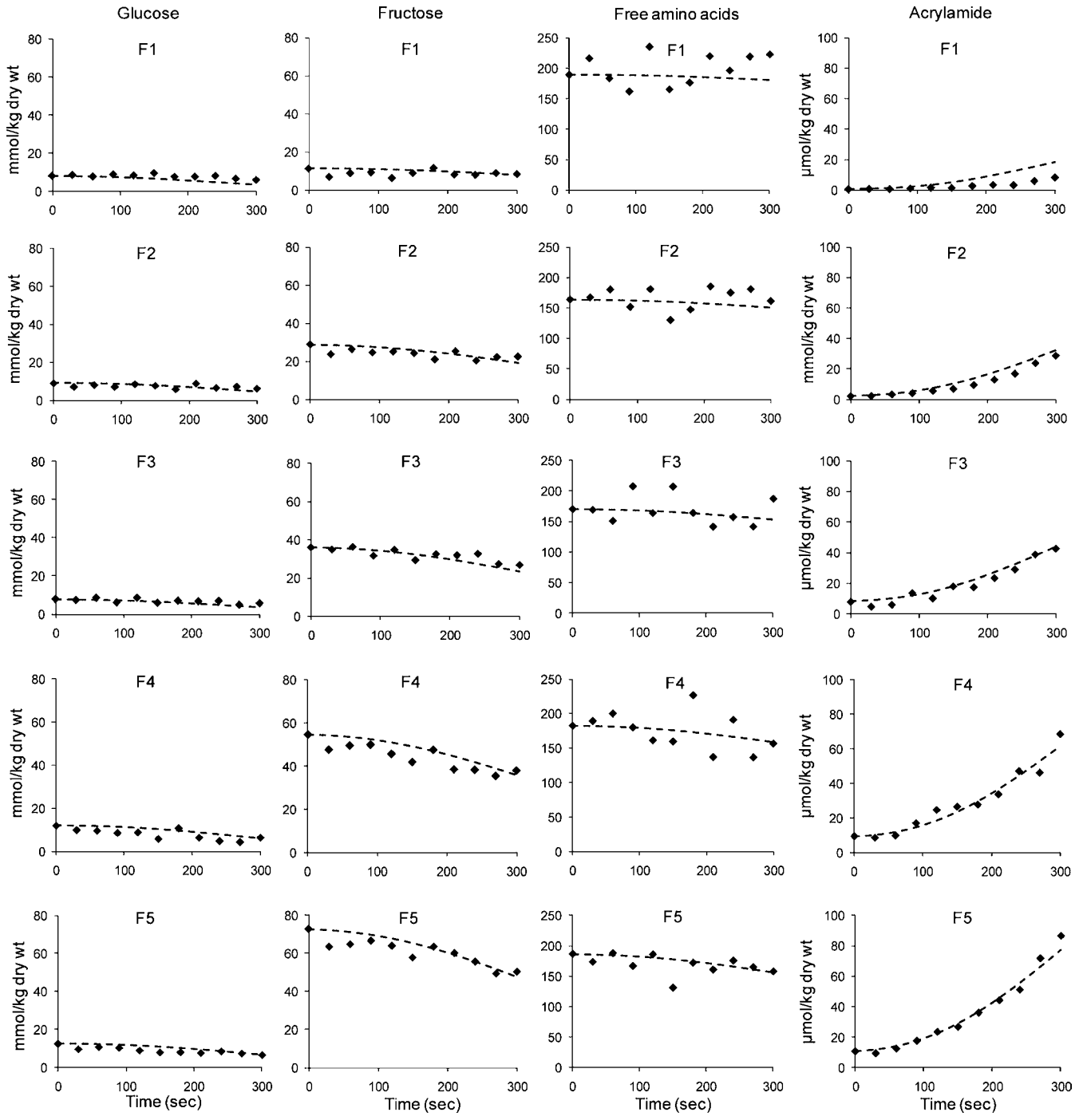

Figure 2. Glucose, fructose, total amino acids, and acrylamide concentrations as a function of time (0−5 min) during finish-frying of five batches of fructose-dipped potato strips at 165 °C (F1 = 0% dip, F2 = 0.5% dip, F3 = 1% dip, F4 = 1.5% dip, F5 = 2% dip). Symbols (◆) are the experimental data points for potato batch F, and the dashed line (- - -) shows the kinetic model derived from the combined data set using the rate equations shown in Figure 6 and the parameters shown in Table 2.

had been dipped in either glucose or fructose solutions ranging in concentration from 0 to 2%, were finish-fried at 165 °C for different times (30 s intervals up to 5 min). This time range was chosen after preliminary trials showed that, for times over 5 min, the fries were overcooked with a burnt appearance and a hard texture. In the glucose-dipped par-fried strips (G1−G5), initial glucose concentrations varied from 16 to 60 mmol/kg defatted dry weight compared with an initial concentration in the raw potato of 30 mmol/kg defatted dry weight (Table 1). The changes in the glucose, fructose, total amino acid, and acrylamide concentrations in the fries, as a function of cooking time (0−5 min), are shown in Figure 1. During the first minute there was very little loss of glucose but, over the next 4 min of frying, the glucose concentrations decreased by about 50%; the rate of loss of glucose increased as the strength of the glucose dip increased, and about 4 times more glucose was consumed in G5 compared with G1. Across this glucose series, the concentration of fructose at time zero was constant (15 mmol/kg), and this decreased over 5 min to about 10 mmol/kg. The formation of acrylamide had a lag phase during the first minute

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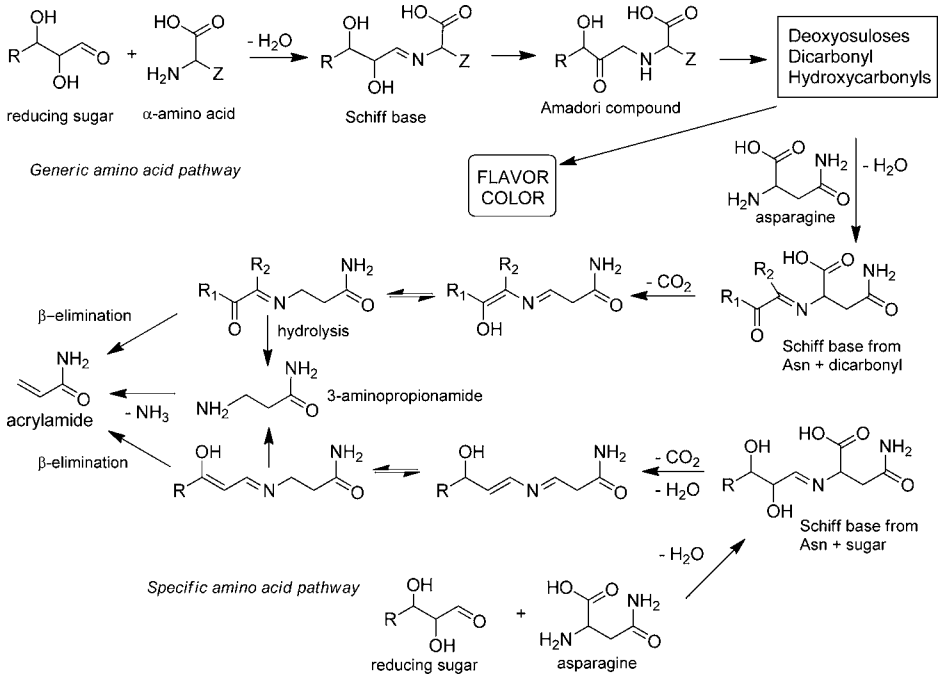

Figure 3. Postulated chemical mechanisms for the formation of acrylamide from a reducing sugar and asparagine.

and, thereafter, was formed more quickly as the dip strength increased. The final concentration was 4 times higher in G5 compared with G1.

A similar pattern was observed in a different batch of potatoes that had been pretreated with fructose solutions ranging from 0 to 2%, series (F1−F5), instead of glucose (Figure 2). The levels of glucose in these samples remained low and showed a modest decrease of about 5 mmol/kg over the heating time, similar to the decrease shown by fructose in the glucose treatment. As the fructose dip strength increased, the loss of fructose increased and the rate of acrylamide formation increased. For each dip strength, the loss of fructose in the fructose series was less than the glucose loss in the corresponding glucose series, but the impact on acrylamide formation was similar. It is clear from these data that both glucose and fructose promote the formation of acrylamide. There was a linear relationship between the acrylamide concentrations in the final fry (after 5 min) and the dip concentration for both glucose (R^2 = 0.995) and fructose (R^2 = 0.995). These results show that the kinetic modeling must take the pretreatment into account, but that the appropriate starting point for the model is at the start of the finish frying, not the raw potato.

Chemical Mechanism. The formation pathway of acrylamide from asparagine has been postulated by a number of authors.[13,15,16,33−35] It has been shown that reactive dicarbonyl intermediates generated during the Maillard reaction can be involved.[11,15] The initial step in this pathway is the reaction between a reducing sugar and any amino acid to give a Schiff base, which rearranges to give an Amadori rearrangement product (ARP) or a Heyns rearrangement product (HRP). These dehydrate and fragment, regenerating the free amino acid and forming a group of highly reactive deoxyosulose, dicarbonyl, and hydroxycarbonyl compounds (Figure 3, generic amino acid pathway). These intermediates undergo a classical Strecker degradation with any amino acid to form flavor and color but, when the amino acid is asparagine, a series of reactions leads to the formation of acrylamide. However, Zyzak proposed that acrylamide could be formed directly from the Schiff base of glucose and asparagine (specific amino acid route), bypassing both the ARP and the fragmentation of the sugar.[13] In low-moisture systems, Stadler demonstrated the contribution of hydroxycarbonyl intermediates to this route.[14] It is likely that all of these pathways contribute to the formation of acrylamide and that the dominant pathway is determined by the specific composition of the system and the processing conditions employed. In foods, asparagine is just part of the amino acid pool, and the other amino acids, some of which are expected to be more reactive than asparagine (e.g., glutamine, arginine, and lysine), will generate the precursors required for the generic amino acid pathway and, thus, potentially play a significant role in acrylamide formation. These chemical pathways form the basis for the kinetic modeling.

Kinetic Model. To develop a kinetic model for the formation of acrylamide, it is important to identify the control points in Figure 3. Leong and Wedzicha postulated a kinetic model for the development of color in a simple aqueous model based on two stable intermediates and the rate-limiting steps.[36] In this system, they demonstrated that the first intermediate comprised a group of compounds that included 1- and 3-deoxyhexosuloses and postulated that the second intermediate was a group of highly reactive dicarbonyl compounds. This kinetic mechanism, comprising two intermediates, was subsequently shown to hold for the generation of acrylamide in model systems and simple food matrices,[15] although the identities of the intermediates were less clear. De Vleeschouwer[19] modified this model, on the basis of the chemical mechanism proposed by Zyzak,[13] by postulating that the formation of acrylamide proceeds via just one intermediate, which they proposed was the Schiff base of asparagine and the reducing sugar. This is analogous to the specific amino acid pathway. The fit of their data with this model was excellent for the aqueous model systems containing just asparagine and reducing sugar but, in mixed amino acid systems and in potato

powder, where the presence of other amino acids can promote the generic amino acid pathway, the fit was less good.[20,23]

The starting point for our kinetic modeling is shown in Figure 4, where both specific and generic amino acid pathways

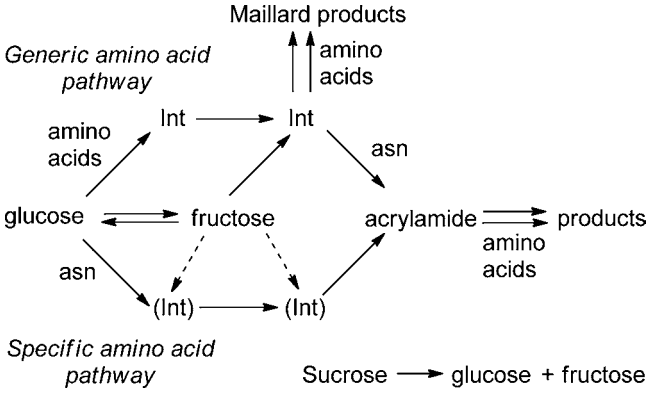

Figure 4. Postulated kinetic mechanisms for the formation of acrylamide from a reducing sugar and asparagine showing all pathways tested in the model.

are postulated. Unlike the De Vleeschouwer model, there is no assumption, at this stage, that the two pathways have common intermediates, nor is there any assumption about the number of intermediates in each pathway. This model postulates the degradation of acrylamide (both with and without the participation of amino acids) and the formation of amino acid-containing Maillard products. It includes the breakdown of sucrose to glucose and fructose and also the interconversion of glucose and fructose. Fructose may also break down without the participation of an amino acid.[37] All of these pathways, in different combinations, were used to construct and test a variety of kinetic models.

Modeling of the Maillard reaction, to date, has been applied to homogeneous matrices in which the heating-up times are small compared with the total reaction time, and the reaction takes place uniformly throughout the system. In the frying of potato strips, however, the temperature of the outside approaches that of the frying oil almost instantaneously, but the heat transfer into the center of the potato is slow. Furthermore, the center maintains a relatively high moisture content throughout the frying process (up to 5 min), and the internal temperature is therefore unlikely to rise above 100 °C. Thus, when the reaction is modeled in potato strips, the temperature profile within the potato strip must be taken into account.

The temperature profile of an infinite slab of raw potato has been calculated from first principles by Ni et al., who modeled heat and mass transfer under typical frying conditions.[25] Their model is based on the mass and energy balance within a multiphase porous matrix and takes into account physical parameters such as surface evaporation, moisture diffusion, heat transfer within the oil, convective flow, and diffusion within the strip. This temperature profile, in conjunction with the temperature data obtained from the temperature probes placed in the center and on the surface of the potato strip, was used to derive an expression (eq 1) that approximated the temperature of the potato strip, T (°C), as a function of time t (s) and distance from the surface of the strip x (mm), where T_{ref} is the frying temperature measured just above the surface of the fry.

$$T = T_{ref} - \frac{780x}{(-4.96 \times 10^{-5}t^2 + 9.81 \times 10^{-2}t + 1.1657)} \quad (1)$$

Another critical factor in determining the rate of the Maillard reaction and, therefore, the rate of acrylamide formation is the moisture content. The surface of the potato strip dries out very rapidly, whereas the interior of the strip is still moist at the end of the frying time. It is, therefore, important to consider the change in moisture of the potato strips with distance and time. This was approximated, in a similar manner to the equation for the temperature profile, using the theoretical relationship calculated by Ni et al.[25] to derive an expression for moisture (M, g/100 g) as a function of distance (x) and time (t) (eq 2), where M_{ref} is the bulk moisture at $t = 0$. This was adapted to fit the par-fried potato strips by incorporating experimentally determined bulk moisture contents to solve for A and B (A = 57.7 ± 1.4; B = 11.2; error could not be determined).

$$M = M_{ref} - \frac{100t}{A\ e^{Bx}} \quad (2)$$

Kinetic models were based on differential equations that apply to the steps in the relevant kinetic scheme. As an example, the simple bimolecular loss of glucose (eq 3) postulated in the first step of all models is used here to illustrate how the temperature and moisture profiles were incorporated into each differential equation.

$$\frac{d[Glu]}{dt} = k[Glu][AAs] \quad (3)$$

Equation 3 was reparametrized using the Arrhenius equation to express the change in k with temperature (k_{ref} = rate constant at the frying temperature). The expression for moisture (M) was incorporated into eq 3 by multiplying both [Glu] and [AAs] by $(1 - M)$, effectively converting the concentrations based on dry weight to aqueous concentrations. Finally, integration, with respect to distance, of those expressions that vary throughout the strip (i.e., moisture and temperature) gives eq 4, which, when combined with eqs 1 and 2, gives an expression for the change in bulk glucose concentration as a function of time.

$$\frac{d[Glu]}{dt} = k_{ref}[Glu][AAs] \int (1 - M)^2\ e^{-E_a/R(\frac{1}{T_{ref}} - \frac{1}{T})}\ dx \quad (4)$$

The model was compiled using the complete set of differential equations, each incorporating the temperature and moisture profiles as described above. These equations were coded for the Athena multiresponse modeling software. The integral was evaluated numerically with respect to distance between 0 and 3.75 mm using a Simpson approximation (n = 51) within the Athena code. Initial values for the model parameters were taken from the literature[15] and preliminary models. The optimal values of the parameters were determined by the software.

At this stage in the development of the model(s), a number of assumptions were necessary;

- The control points in the reaction were adequately described by Figure 4.
- The mass and heat transfer properties of a par-fried potato strip were similar to those of a raw potato slab.
- The temperature of the surface of the strip was the same as that of the oil.

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- There were mass and heat transfers from four sides of the strip; corner effects were ignored.
- The precursors were uniformly distributed throughout the potato strip during pretreatment, and there was no redistribution of precursors during the finish-frying the strip.
- One universal value for the activation energy (E_a) could be applied to all kinetic steps of the reaction. This was set to 100 kJ/mol.

Kinetic models involving one or more combinations of the pathways shown in Figure 4 were considered in the first instance. Early iterations of the model suggested that the hydrolysis of sucrose was not contributing to the reaction during the 5 min frying time. This is consistent with the fact that there was no decrease in sucrose over that time period, and it was only after 5 min when sucrose started to degrade. This step was removed from subsequent iterations. Likewise, rate constants for the degradation of acrylamide were consistently returned as zero, so this step was removed from the model. Under these conditions, the loss of acrylamide was insignificant compared to the formation.

Other iterations were carried out that involved intermediates from both the generic and specific amino acid pathways (Figure 4). A kinetic model based on the specific amino acid pathway involving two steps and just one intermediate did not fit the experimental data, confirming the need for a three-step kinetic pathway. When a kinetic mechanism based on two parallel three-step pathways was constructed, a good fit was achieved, but the rate constant for the formation or loss of glucose via the specific amino acid pathway was indeterminate. In other words, the model again confirmed the three-step nature of the process, but could not calculate the split between the generic and the specific amino acid pathways. The best results were obtained using the one three-step pathway and just two intermediates. This effectively combines the two pathways, and the intermediates are a combination of those derived from both the specific and generic amino acid pathways. Two scenarios were tested: one in which the second intermediate was a group of transient intermediates that reacted very quickly compared to the other steps, and a second in which the reactions of the second intermediate were assigned rate constants allowing for the accumulation of the intermediate. The former produced the better fit.

Figure 5 shows the kinetic mechanism that produced the best fit, and the set of differential equations derived from this model

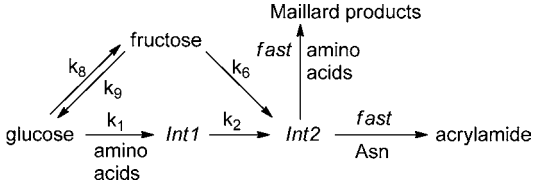

Figure 5. Postulated kinetic mechanisms for the formation of acrylamide from a reducing sugar and asparagine showing a simplified model used for the final iteration of the model.

are shown in Figure 6. The first two equations show the rate of change of glucose and fructose concentrations, respectively, taking into account the interconversion between the two. Int2 is formed from both glucose and fructose. The route from glucose is bimolecular, involving the participation of an amino acid via another group of intermediates (Int1), whereas fructose

$$\frac{d[Glu]}{dt} = -k_1[Glu][AAs] - k_8[Glu] + k_9[Fru]$$

$$\frac{d[Fru]}{dt} = -k_6[Fru] + k_8[Glu] - k_9[Fru]$$

$$\frac{d[AAs]}{dt} = -(k_2[Int1] + k_6[Fru])$$

$$\frac{d[Acr]}{dt} = (k_2[Int1] + k_6[Fru])R_{Asn}F_{Asn}$$

$$\frac{d[Int1]}{dt} = k_1[Glu][AAs] - k_2[Int1]$$

Figure 6. Rate equations derived from the kinetic mechanism (Figure 5) where concentrations are mmol/kg defatted dry wt, [Glu] = glucose, [Fru] = fructose, [AAs] = total free amino acids, [Acr] = acrylamide, [Int1] = intermediate 1; k_1, k_2, k_6, k_8, k_9 = rate constants (see Table 2 for units); R_{Asn} and F_{Asn} = ratios defined in the text.

can also undergo a unimolecular reaction to such intermediates without the involvement of amino acids. This is reflected in Figure 6, where the expression for the rate of change of glucose involves a term for amino acids and the expression for the rate of change of fructose does not.

Although the amino acids are involved in the first stage of the reaction, there is no net loss in this step, because they are regenerated from Int1. The expression for the change in amino acid concentration takes into consideration only the loss of amino acids during the formation of Maillard products from Int2. Because Int2 is a transient intermediate, which does not build up to any extent, the rate of the formation of Maillard products is equal to the rate of formation of Int2, which is formed from both glucose (via Int1) and fructose (directly), as shown by the equation expressing the loss of amino acids in Figure 6.

The reaction of Int2 is considered to be very fast, so individual rate constants for the subsequent reactions are not kinetically important and the products are formed in proportion to the amount of secondary reactants. For acrylamide formation, the rate depends on the molar ratio of asparagine to total free amino acids (R_{Asn}). This ratio varies with the batch and variety of potato, but is constant throughout the frying time. This parameter was determined experimentally for both batches of potatoes, showing good agreement between potato batch G and potato batch F (R_{Asn} = 0.37 and 0.38, repsectively; R^2 = 0.94 in both cases), allowing the use of one fixed value of R_{Asn} in the final combined model (R_{Asn} = 0.37, R^2 = 0.96). However, not all of the asparagine present reacts to form acrylamide, and some of it forms other Maillard products. The fraction that is converted to acrylamide (F_{Asn}) is included in the model as a variable.

The data from potato batches G and F finish-fried at 165 °C were combined with data obtained at 175 and 185 °C to build the final model. The software was able to achieve a solution for all parameters (Table 2). The rate constant k_1, associated with the disappearance of glucose, was 7.6×10^{-4} mol^{-1} kg s^{-1} and was calculated with some confidence (95% HPD confidence interval was 12%), although the rate constant for the next step, k_2, had more error associated with it (58%). This is expected because the concentration of neither Int1 nor Int2 was measured. The rate constant k_6, associated with the disappearance of fructose, was 1.8×10^{-2} s^{-1} (95% confidence interval is 11%). Once the amino acid concentration term is accounted for in the bimolecular loss of glucose, the initial rate

Table 2. Parameter Estimates from Athena Visual Studio for the Final Kinetic Model (Proposed in Figure 5), Generated from Data for All Glucose- and Fructose-Dipped Potato Strips Fried at 165, 175, and 185 °C

parameter	units	optimal estimate × 10^3	95% confidence interval × 10^3
k_1	$mmol^{-1}$ kg s^{-1}	0.76	±0.1 (12%)
k_2	s^{-1}	400	±230 (58%)
k_6	s^{-1}	18	±2 (11%)
k_8	s^{-1}	7.0	±1.7 (25%)
k_9	s^{-1}	1.1	±1.7 (152%)
F_{Asn}		5.9	±0.4 (7%)

for the loss of glucose is about double that of fructose but, in cultivars where the total free amino acid content is low, this ratio is expected to decrease. This demonstrates that fructose is involved in generating significant quantities of acrylamide. The rate constants for the interconversion of glucose and fructose were of a similar order of magnitude, but there was a much greater uncertainty in the conversion of fructose to glucose. F_{Asn} represents the fraction of the overall loss of asparagine that is converted to acrylamide. The value of F_{Asn} was returned as 5.9 × 10^{-3} and indicates that only 0.6% of the asparagine consumed during the process ends up as acrylamide, the rest being involved in the formation of other Maillard products or other degradation products.

Observed experimental values compared with values fitted by the model are shown, over the time course of the finish-frying, in Figures 1 and 2. Visual inspection shows that there is an excellent fit for glucose, fructose, acrylamide, and free amino acids, although there is more variation associated with the amino acid data. Figure 7 shows plots of observed against predicted values for acrylamide, glucose, and fructose for all batches (G1−G5 and F1−F5) compared with the line of perfect fit. The correlation is good in each case with slopes of 1 ± 0.02 and the points scattered randomly about the line of perfect fit. The correlation for free amino acids is less good as the model tends to underestimate the loss of amino acids, suggesting additional loss of amino acids via another route and provides scope for further work.

Another iteration of the model based on the data obtained at a frying temperature of 165 °C was used to predict the outcome of a batch of fries, of known starting composition, finish-fried at 175 and 185 °C. These are shown in Figure 8. The fact that these experimental data fit well to the predicted values shows that the model is robust for typical frying temperatures and that under all such conditions one universal activation energy of 100 kJ/mol can be used. Further validation is required using potatoes of different amino acid concentrations, different asparagine contents, and different Asn/AAs ratios.

A major driver of French fry quality is appearance (i.e., color, length, and cut quality), so it is important to compare the development of color with the formation of acrylamide. The standard color measure in the industry uses an Agtron spectrophotometer to give an empirical value for reflectance. Figure 9 shows a plot of reflectance (Agtron) versus acrylamide formation for both the glucose-dipped and fructose-dipped batches of fries. This shows clearly that, for a given color, the fructose-dipped fries contain more acrylamide than the glucose-dipped fries, particularly in the shaded region where the color of the fries is generally acceptable to the consumer. Thus, the process of dipping in glucose, which replaces fructose removed during blanching, decreases the acrylamide potential compared with the unblanched strip. Mestdagh et al.[8] reported that the ratio of fructose to glucose affected both color and acrylamide levels in fried potato strips, with a relatively higher fructose concentration favoring acrylamide concentration. Recently, this observation was confirmed in a study of the acrylamide and color formation in French fries prepared from potato strips that had been dipped in sugar solutions with variable fructose/glucose ratios.[30]

In this paper, a model has been constructed that predicts accurately the concentration of acrylamide in French fries from the composition of the par-fried potato strip and the frying time and temperature. An understanding of how these parameters affect acrylamide formation is crucial in the development of

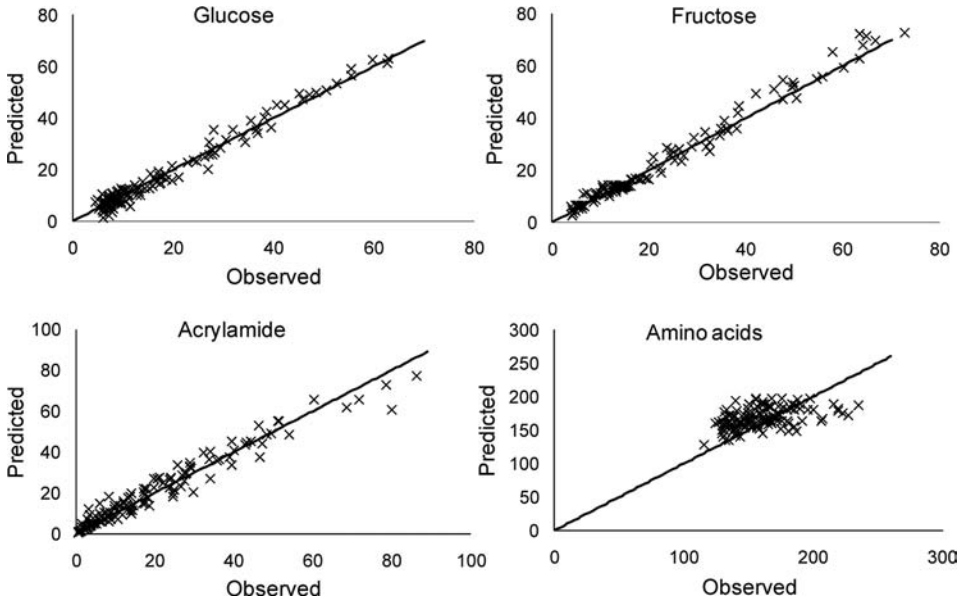

Figure 7. Predicted against observed values for all batches of fries (G1−G5 and F1−F5) compared with the line of perfect fit ($y = 1$). Units: glucose, fructose, amino acids, mmol/kg fat-free dry weight; acrylamide, μmol/kg fat-free dry weight.

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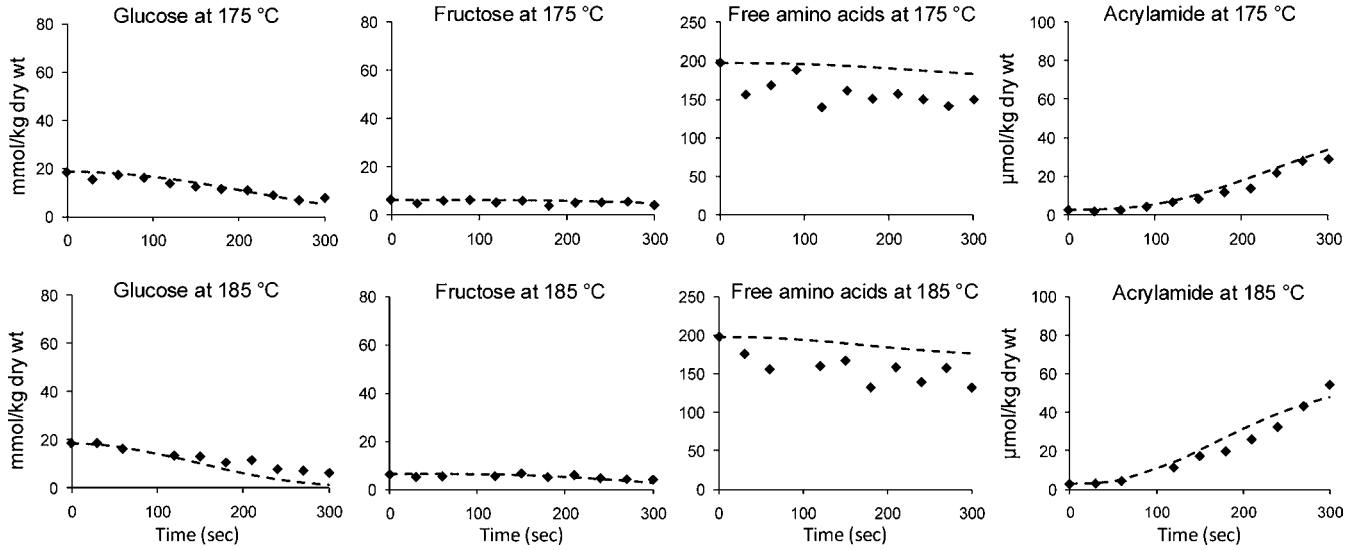

Figure 8. Comparison of experimental values for glucose, fructose, acrylamide, and total free amino acids with values predicted from the kinetic model for fries dipped in 0.5% glucose and cooked at 175 and 185 °C. Symbol (◆) represents experimental values, and the dashed line (- - -) represents the predicted values.

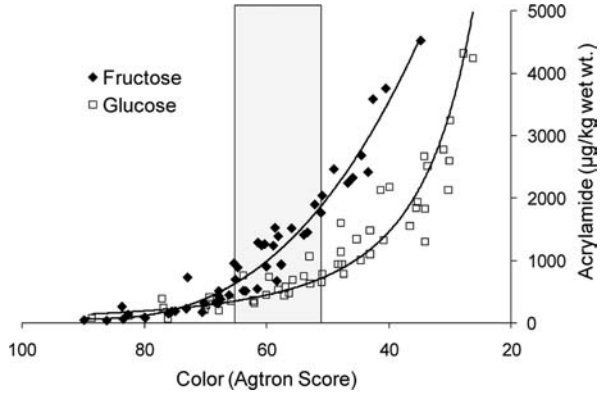

Figure 9. Correlation between color (reflectance) and acrylamide concentration (ppb wet wt). Data shown as open symbols were obtained from the series G1-G5, and series F1–F5 is depicted by solid symbols. The shaded area represents the region where French fries have a color that is generally accepted by the consumer.

mitigation strategies. The fructose/glucose ratio is one parameter that can have a profound effect on acrylamide formation. Although the ratio in a raw potato is typically 0.8, this value can be altered during the pretreatment (blanching and dipping) and provides another means of control. Implicit in the model is that acrylamide formation increases with Asn/AAs ratio, which is important for those processes that alter the ratio either by enzymatic modification or crop selection.

AUTHOR INFORMATION

Corresponding Author

*Phone: +44 118 378 8700. Fax: +44 118 378 7708. E-mail: d.s.mottram@reading.ac.uk.

Notes

The authors declare no competing financial interest.

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