JOURNAL OF AGRICULTURAL AND FOOD CHEMISTRY

Article

pubs.acs.org/JAFC

Added versus *Accumulated* Sugars on Color Development and Acrylamide Formation in French-Fried Potato Strips

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ABSTRACT: *Added* (glucose addition) versus *accumulated* (in situ sugar development via cold-temperature storage) sugar treatments were investigated in relation to acrylamide formation within fried potato strips at standardized levels of finish-fried color (Agtron color scores ranged from 36 to 84). The *added* sugar treatment exhibited a relatively reduced rate of acrylamide formation and generally possessed a lower and less variable acrylamide content (61−1290 ng/g) than the *accumulated* sugar scheme (61−2191 ng/g). In a subsequent experiment, *added* fructose applied to strip surfaces via dipping prior to frying favored acrylamide formation over color development relative to *added* glucose, for which the reverse trend was observed. Thus, where acrylamide differences were noted between *added* and *accumulated* sugar treatments (given equivalent Agtron color scores), this result was likely aided by the relative higher fructose content in strips of the *accumulated* sugar scheme rather than simply a greater relative concentration of total reducing sugars.

KEYWORDS: *French fries, asparagine, Maillard browning, glucose, fructose*

INTRODUCTION

Potatoes are consumed almost exclusively following some form of heating such as baking, boiling, frying, or steaming, with deep frying being one of the most prevalent preparation methods utilized in both industry and the home.[1] Frying potatoes in an edible fat or oil above the boiling point of water facilitates rapid heat transfer and cooking, as well as development of desirable browning, texture, and flavor characteristics.[1] During frying, development of the desired sensorial properties (i.e., color, flavor, and aroma) within potato products occurs largely as a function of Maillard or nonenzymatic browning reactions.[2] The color of fried potatoes represents a key product attribute, since it is the first characteristic evaluated by consumers and is thus essential to product acceptance.[3]

Nevertheless, heating of foods such as potato chips, French fries, breads, or processed cereals at temperatures above 120 °C may induce formation of acrylamide.[4] Detection of acrylamide (37−4804 μg/kg) in a wide range of heated foodstuffs has raised concern over the safety of many high-temperature processed foods, since acrylamide is classified as a probable carcinogen in humans.[4−7] Consequently, the Food and Agriculture Organization (FAO) and World Health Organization (WHO), as well as the U.S. Food and Drug Administration (FDA), have recommended that food processors make significant efforts to reduce acrylamide levels in commonly consumed foodstuffs, especially potato chips and French fries.[8,9]

Acrylamide formation is promoted by the presence of the free amino acid, asparagine, as well as a carbonyl source, most commonly a reducing sugar.[10−13] In short, the α-amino group of asparagine reacts initially with a carbonyl group via a thermally induced condensation reaction, affecting the subsequent conversion of asparagine to acrylamide following decarboxylative deamination.[11] Alternatively, the Strecker (amine) degradation product of asparagine, 3-aminopriopionamide, may also be directly converted to acrylamide during high temperature heating.[14] Thus, acrylamide formation during high temperature heating of foods is impacted by both the concentrations of free asparagine and reducing sugars.[15] Asparagine is not only the most abundant free amino acid within the tuber[16,17] but is also more difficult to regulate than reducing sugars, making control of the latter much more practical for limiting acrylamide formation in potato products at present.[18]

Nevertheless, for processing French fries on an industrial scale, it is common practice to control or standardize finished product color by adding an appropriate amount of reducing sugar to blanched potato strips prior to frying.[19] Glucose, the most commonly utilized reducing sugar for such purposes, is imparted by spraying or dipping onto potato strip surfaces, where it is most likely to convey surface color to strips during frying via the Maillard reaction. Though the industrial practice of imparting glucose to strips prior to frying aids necessary color development, it also has potential to adversely impact acrylamide levels by localizing a key reactant at the potato strip surface, where it is exposed to the high temperatures during frying. Alternatively, there is existing commercial precedent for generating fried potato products (e.g., fresh-cut French fries) by relying solely on the intrinsic reducing sugar contents within the potato tissue to impart desired color to finish-fried products (as opposed to sugar addition). For experimental purposes, cold-temperature storage (3−4 °C) of tubers may be employed to promote in situ accumulation of reducing sugars (i.e.,

Received: May 14, 2012
Revised: July 30, 2012
Accepted: August 11, 2012
Published: August 11, 2012

sucrose accumulation and its subsequent conversion to reducing sugars by vacuolar acid invertase),[20] thus providing a range of potato intrinsic reducing sugar contents (i.e., *accumulated* sugar) and finish-fried product color intensities.[21] While *accumulated* sugars are not homogeneously distributed throughout the tuber,[22] often leading to uneven color development in fried potato products, they provide an interesting contrast to the conventional practice of glucose addition (i.e., *added* sugar), since sugar build-up occurs within the tissue itself as opposed to being strictly applied to strip surfaces. The direct comparison of *added* vs *accumulated* sugar color schemes has not been investigated in relation to acrylamide formation.

The primary research objective was to investigate relationships between color development and acrylamide formation occurring via high temperature reactions in fried potato strips as a function of *added* (i.e., glucose addition) versus *accumulated* (i.e., in situ sugar accumulation) sugar treatments.

MATERIALS AND METHODS

Materials. Russet Burbank potatoes were grown in commercially contracted fields in the Columbia Basin of Washington state. Following the 2009 harvest, potatoes were held at 10 °C for approximately three weeks for suberization, after which the holding temperature was gradually reduced to 8−9 °C to facilitate long-term storage. For various experiments comprising this study, potatoes were removed from storage and randomly segregated into experimental lots according to treatment as described in subsequent sections.

Acrylamide and asparagine were obtained from Sigma-Aldrich Chemical Corp. (St. Louis, MO, USA), while $^{13}C_3$-labeled acrylamide was obtained from Isotec (St. Louis, MO, USA). *ortho*-Phthalaldehyde and 9-fluorenylmethoxycarbonyl chloride were obtained from Pierce (Rockford, IL, USA). Norvaline, 3-mercaptopropionic acid, formic acid, and boric acid were obtained from Acros (Pittsburgh, PA, USA). HPLC grade methanol, acetonitrile, and reagent grade ethanol were obtained from J.T. Baker (Phillipsburg, NJ, USA). All other chemicals used were at minimum of analytical grade.

Preparation of Fried Potato Strips. *Fried Potato Strip Processing.* For each treatment replicate, raw potatoes (ca. 4.5 kg) were processed as described below, yielding approximately 2.0−2.3 kg of partially fried, frozen potato strips. All potatoes (regardless of treatment) were washed, peeled, and heated in water at 54 °C for 30 min, after which tubers were cut into strips using a hydro-cutter equipped with a 7.52 mm × 7.52 mm knife block (GME International, Boise, ID, USA). Obtained potato strips were blanched (74 °C for 7 min), and then dipped (30 s) in an aqueous solution of 0.75% (w/v) sodium acid pyrophosphate (SAPP) heated to 74 °C. Blanched strips were dried in a forced air dryer at 60 °C (approximately 5−10 min) to achieve a 12−14% moisture loss by weight. A 20.5 kW Frymaster deep fryer (model EH21721T, Shreveport, LA, USA) was used for both partial- and finish-frying operations. Potato strips were immersed in oil only after the fryer had achieved the designated temperature, as indicated by the digital display on the fryer. The weight of potato strips (ca. 454 g per batch) entering the partial-frying operation was controlled to ensure consistent temperature exposure (partial-fried batches for a given treatment replicate were subsequently pooled). Partial-frying was conducted in a blend of canola and palm oil at 191 °C for 50 s, after which strips were blast frozen at −23 °C. Frozen strips were held in a conventional freezer at −18 °C until finish-fried (ca. 680 g) in canola oil at 177 °C for 150 s. At specific processing stages (following cutting, drying, and finish-frying steps), potato strips (ca. 454−680 g) of each experimental replicate were frozen (−18 °C) until further analyzed.

Accumulated versus Added Sugar Experiment. Tubers of the *accumulated* sugar treatment were held at 3−4 °C for 0, 7, 9, 14, 21, or 28 days of controlled storage to promote in situ sugar development, providing a range of tuber reducing sugar contents for study. Following storage, tubers were processed into fried potato strips as previously described.

In contrast, potatoes of the *added* sugar treatment were held at 8−9 °C over the same 28 day period to minimize sugar development. Tubers were processed into fried potato strips in the same manner as those of the *accumulated* sugar treatment, except that the SAPP dipping solution also included *added* glucose to promote desired color development of strips during finish-frying. The glucose concentration of the dipping solution was adjusted such that the color values of the *added* sugar finish-fried strips matched those of the *accumulated* sugar treatment for a given tuber storage interval (7, 9, 14, 21, or 28 days). Glucose concentrations of the dipping solutions were 0.00, 0.07, 0.20, 0.10, 0.65, and 0.60% (w/v) for the 7, 9, 12, 14, 21, and 28 day tuber storage intervals, respectively.

Added Fructose vs Glucose Experiment. Potatoes were held at 8−9 °C to minimize sugar development, and processed into fried potato strips as previously described, except that the dipping solution (in addition to SAPP) also included *added* sugar (0.3%, w/w) consisting of different ratios of glucose/fructose (100:0, 75:25, 50:50, 25:75, 0:100). Control potato strips were dipped, but in SAPP solution without *added* sugar.

Surface Color of Fried Potato Strips. The surface color of finish-fried potato strips was measured using an Agtron process analyzer (Agtron Incorporated, Reno, Nevada, USA). Finish-fried potato strips (≈ 400 g) were randomly selected from a given treatment population for analysis. The Agtron apparatus covers the range from black (Agtron score = 0) to white (Agtron score = 100) and was calibrated using appropriate reference tiles.

Moisture Content of Potato Strips. Moisture content of potato strips was measured via oven drying. Homogenized potato (≈ 3 g) was weighed accurately into aluminum pans and dried in a mechanical convection oven (Fisher Isotemp Oven, model 750F) at 125 °C to a constant weight (3−4 h).

Acrylamide Content of Fried Potato Strips. Acrylamide content of finish-fried potato strips was determined as described by Roach et al.[23] with modifications. Frozen, finish-fried potato strips (ca. 454 g) were homogenized in a Robot Coupe processor (Jackson, MS, USA). Homogenized potato (1.000 g; ± 0.100 g) was weighed into a centrifuge tube (50 mL), after which 0.1 mL of $^{13}C_3$-labeled acrylamide internal working standard (2000 ng/mL in 0.1%, w/v, formic acid) and 9.9 mL of deionized water were transferred to the tube. The resulting suspension was shaken (20 min at 400 rpm) on a mechanical shaker (MaxQ2000, Barnstead, Dubuque, IA, USA) and was subsequently centrifuged (3000*g*, 15 min). An aliquot of the clarified aqueous layer was collected for solid phase extraction (SPE) cleanup. An Oasis HLB SPE cartridge (6 mL, 200 mg) (Waters Corp., Milford, MA, USA) was conditioned with methanol (3 mL) followed by deionized water (3 mL). After conditioning, clarified tissue supernatant (1.5 mL) was loaded onto the Oasis HLB SPE cartridge, allowing supernatant to pass completely through the sorbent material by gravity. Additional deionized water (0.5 mL) was passed through the cartridge and the eluent was discarded. The cartridge was then eluted with deionized water (1.5 mL), with the resulting eluent collected in a test tube for further SPE cleanup. Next, the eluent (1.5 mL) obtained from the Oasis HLB SPE was loaded onto a BondElut Accucat SPE cartridge (3 mL, 200 mg) (Varian Inc., Chicago, IL, USA) that had been previously conditioned with methanol (3 mL) and deionized water (3 mL); the first 0.5 mL of eluent was allowed pass though the cartridge uncollected, after which the remainder of the eluted portion was collected and transferred to an amber autosampler vial for liquid chromatography-tandem mass spectrometry (LC-MS/MS) analysis.

The LC-MS/MS system consisted of an Agilent 1200 Series HPLC system (Santa Clara, CA, USA) equipped with binary pumps, degasser, autosampler, and column heater, coupled to an Applied Biosystems Q-Trap 4000 MS/MS detector (Carlsbad, CA, USA) equipped with an electrospray ionization (ESI) interface. Both calibration standards and prepared samples (20 μL) were eluted on a Phenomenex Hydro-RP 80 Å analytical column (4 μm, 250 mm × 2 mm) (Torrance, CA, USA) maintained at 26 °C. The mobile phase consisted of 0.5%

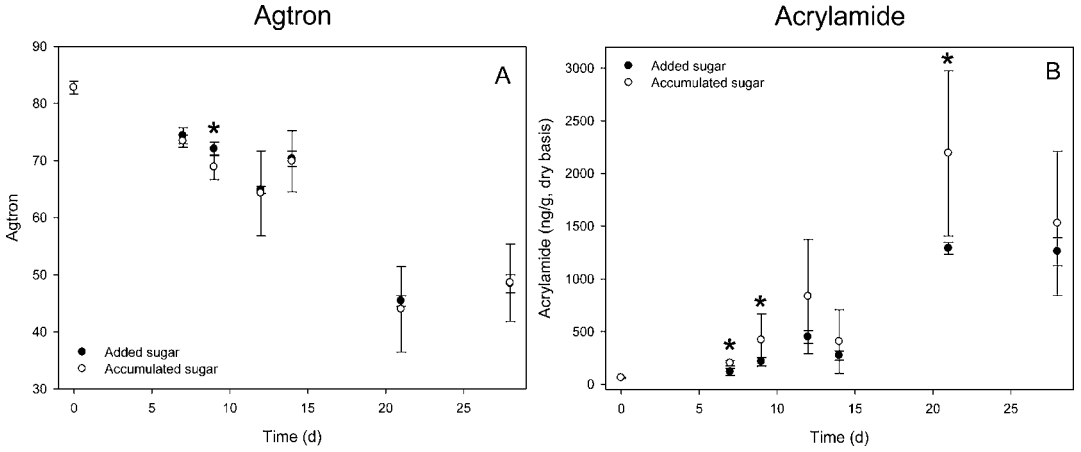

Figure 1. Agtron scores (A) and acrylamide contents (B) of finish-fried potato strips for the *accumulated* and *added* sugar color development schemes over 28 days of tuber storage. An asterisk above a set of data points denotes a significant difference between values of the *added* and *accumulated* sugar treatments for a particular tuber storage time interval ($p < 0.05$).

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 using the following parameters: ion spray voltage, 5500 V; desolvation temperature, 330 °C; curtain gas pressure, 15 psi nitrogen; declustering potential, 26 V; collision energy, 15 V; and dwell time, 50 ms, to monitor transitions 72 > 55 and 75 > 58 for analysis of acrylamide and $^{13}C_3$-labeled acrylamide, respectively.

For quantification, acrylamide and $^{13}C_3$-labeled acrylamide were combined and then further diluted with deionized water to prepare acrylamide calibration standards ranging from 0.5 to 500 ng/mL, each containing 20 ng/mL $^{13}C_3$-labeled acrylamide. Concentration of acrylamide was calculated as (ng/g) and then converted to dry basis for ease of analysis and comparison.

Free Asparagine Content of Potato Strips. Free asparagine content of potato strips was analyzed as described by Antoine et al.,[24] Bartolomeo and Maisano,[25] and Hodgin[26] with modifications. Homogenized potato (2−5 g, weighed to the nearest 0.01 g) was dispersed in 20 mM acetate buffer (30 mL, pH 4.25) and shaken for 30 min on a mechanical shaker. Extract was filtered through a 0.45 μm glass microfiber filter (Whatman, Piscataway, NJ, USA), after which filtrate (1 mL) was transferred to an autosampler vial containing norvaline internal standard solution (100 μL, 4.27 μmol/mL in 20 mM acetate buffer, pH 4.5).

Potato sample extracts were derivatized using *ortho*-phthalaldehyde (OPA) reagent (10 mg/mL OPA and 15 mg/mL mercaptopropionic acid in 400 mM borate buffer, pH 10.2) and Fmoc-Cl (2.5 mg/mL 9-fluoromethoxycarbonyl chloride in acetonitrile) in the autosampler immediately prior to HPLC injection using the following program: draw 5 μL borate buffer; draw 1 μL sample or standard; mix at max speed, 2×; wait 0.5 min; draw 0 μL of deionized water (needle wash); draw 1 μL of OPA; mix at max speed, 6×; draw 0 μL of deionized water (needle wash); draw 1 μL of Fmoc-Cl; mix at max speed, 6×; draw 0 μL of acetonitrile (needle wash); draw 32 μL of deionized water; mix at max speed, 2×; inject. Derivatized samples were eluted on an Agilent Zorbax Eclipse AAA column (3.5 μm, 150 mm × 3.0 mm) (Santa Clara, CA, USA) held at 40 °C using an Agilent 1100 series HPLC system (Santa Clara, CA, USA), equipped with quaternary pumps, autosampler, column heater, and a fluorescence detector. Mobile phase A was 40 mM NaH_2PO_4 (adjusted to pH 7.8 with 40% NaOH solution), while mobile phase B consisted of acetonitrile, methanol, and deionized water in a ratio of 45:45:10 (v/v/v), respectively. Separation of asparagine was accomplished at a flow rate of 0.9 mL/min 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); 22.3 min, back to 100% A, 22.3−30 min, 100% A (to equilibrate column).

For quantification, asparagine (5.78 μmol/mL in 20 mM acetate buffer, pH 4.25) was combined with norvaline internal standard and further diluted with 20 mM acetate buffer to prepare calibration standards ranging from 10 to 1250 nmol/mL, each containing 388 nmol/mL norvaline. Calibration standards were derivatized in the autosampler as previously described.

Sugar Contents of Potato Strips. Sugar contents of potato strips were analyzed according to AOAC Method 977.20[27] with modifications. Homogenized potato (0.5−5.0 g, weighed to the nearest 0.01 g) was transferred to a centrifuge tube (50 mL), after which extraction solution (25 mL) was added to the tube. The suspension was shaken (60 °C for 30 min) at a speed sufficient to keep the homogenate suspended and was subsequently centrifuged (3200*g*, 4 min). The resulting supernatant was passed through a 0.45 μm glass microfiber membrane filter (Whatman, Piscataway, NJ, USA), and the obtained filtrate was transferred to an autosampler vial. An Agilent 1200 series HPLC system (Santa Clara, CA, USA), equipped with a quaternary pump, autosampler, column heater, and Alltech evaporative light scattering detector (ELSD, model 3300, Lexington, KY, USA) was used for sugar analysis. Injected sample or calibration standard (2.5 μL) was eluted on an Alltech Prevail Carbohydrate ES column (5 μm, 250 mm × 4.6 mm, Lexington, KY, USA) at 30 °C. The mobile phase consisted of 75% acetonitrile/25% water; flow rate was maintained at 1.0 mL/min. Eluted sugars were detected by ELSD with a nebulizer gas flow of 1.5 mL/min and an evaporative temperature of 48 °C. Concentration (%, w/w) of glucose, fructose, and sucrose in samples was calculated via external calibration with authentic standards.

Experimental Design and Statistical Analysis. All experiments consisted of six full treatment replications. For each replicate experimental unit of each treatment, all analyses (Agtron, acrylamide, asparagine, sugars, etc.) were conducted once. The normality of all experimental data was assessed using the Shapiro-Wilk test, which revealed that some data exhibited a non-normal distribution pattern. Thus, all data were analyzed using a nonparametric Kruskal−Wallis test ($\alpha < 0.05$), while Duncan's test was used to determine statistical differences among experimental mean values ($p < 0.05$). Contrasts were utilized to assess differences between slopes and equations of regression lines ($p < 0.05$), while Pearson's correlation analysis was used to examine relationships among experimental factors in relation to both acrylamide content and color development ($p < 0.05$). All statistical analyses were conducted using SAS version 9.1 for Windows (SAS Institute, Cary, NC, USA).

RESULTS AND DISCUSSION

Color Development within Fried Potato Strips. Color development within fried potato strips was managed via two

contrasting strategies, namely, by accumulation of reducing sugars within tubers during low temperature (3−4 °C) storage (i.e., *accumulated* sugar treatment) or by direct addition of glucose to potato strip surfaces prior to frying (i.e., *added* sugar treatment). To standardize color of the finish-fried potato strips across the two opposing treatments for a common tuber storage interval, an appropriate amount of glucose was applied to strip surfaces of the *added* sugar treatment prior to frying, necessary to match the color of finish-fried strips generated via the *accumulated* sugar treatment. Color of finish-fried strips was assessed using an Agtron abridged spectrophotometer, which accommodates measurement of foodstuffs of irregular geometry[28] and is commonly employed industrially as a quality control tool for fried potato products.[29] A higher Agtron score reflects a lighter surface color, with Agtron values in the range of 50−75 generally considered industrially acceptable.[30]

While in most instances the overall color scores for the two sets of fried potato strips were identical, those of the *added* sugar treatment possessed a much more uniform color distribution (both between strips as well as within a given strip) than those of the *accumulated* sugar strategy, most likely attributable to a more homogeneous distribution of glucose at strip surfaces. *Accumulated* sugar fried potato strips exhibited a patchy or mottled appearance, as well as an abundance of darkened (i.e., sugar) ends, both of which are generally considered commercial product defects. Sugar accumulation within cold-sweetened potatoes is not homogeneous, favoring build-up in the stem-ends of tubers.[22]

Figure 1A illustrates the array of color scores for finish-fried potato strips obtained via the two color development treatments across all storage times of the study. For the *accumulated* sugar scheme, Agtron scores, which ranged from 36 to 83, generally declined (i.e., color became darker) over 28 days of 3−4 °C storage, though not always in direct linear fashion. Patterns of sugar accumulation during cold storage are known to vary according to cultivar and are further reported to be nonlinear in nature.[31] Within these experiments, cold (3−4 °C) storage times of 7−15 days yielded finished-fried strips with a typical golden color, whereas extended lengths of cold storage (21 or 28 days) produced fried strips that were darker than normally targeted for quick serve restaurants. Nevertheless, Agtron values for *added* sugar finish-fried potato strips effectively mirrored those of the *accumulated* sugar scheme for common lengths storage (though in situ color development was generally more variable than that of glucose addition), making it possible to directly compare acrylamide levels within fried strips of the two treatments at essentially equivalent levels of color development.

Acrylamide Content of Potato Strips. Acrylamide contents for fried potato strips of this study ranged from 35 to 1250 ng/g on an "as consumed" or wet basis, and fell within typically encountered values reported for like products (<2310 ng/g).[32] From this point forward, acrylamide levels within fried potato strips are expressed and discussed on a dry weight basis to better facilitate scientific comparison.

Acrylamide contents of both *accumulated* and *added* sugar finish-fried strips (Figure 1B) were inversely proportional to their respective Agtron values (Figure 1A) and generally increased with increasing lengths of storage (i.e., acrylamide levels increased as color of fried strips became darker). This observation is in general agreement with previous reports.[3,33] Not only were acrylamide levels within fried strips of the *accumulated* (relative to the *added*) sugar treatment much more variable, they were also relatively higher at specific tuber storage intervals (Figure 1B).

To further analyze this latter observation, acrylamide levels were separately plotted against Agtron scores for each color development scheme (Figure 2). Regression lines for

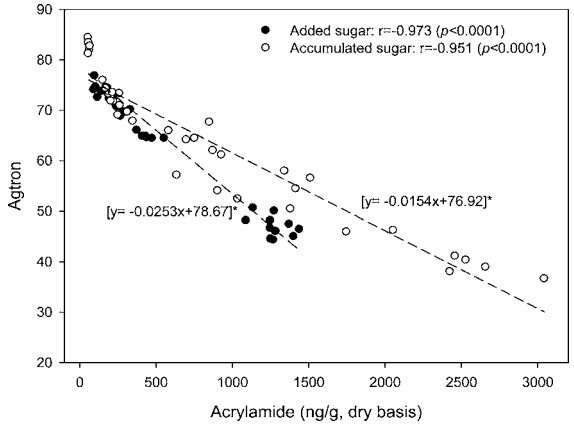

Figure 2. Correlations between Agtron scores and acrylamide contents of finish-fried potato strips according to *accumulated* and *added* sugar color development schemes. *Both the slope values and equations for the fitted regression lines of the *added* and *accumulated* sugar treatments were significantly different ($p < 0.0001$).

accumulated and *added* sugar treatments exhibited different slope values and were not parallel, providing evidence that acrylamide formation was slightly less for the *added* (relative to the *accumulated*) sugar treatment for same degree of color development. Thus, the two treatments exhibited different rates of acrylamide formation, the basis of which was further investigated.

Asparagine and Sugar Contents of Potato Strips. Free asparagine content within potato strips was monitored both after drying (following blanching/dipping, but prior to frying) and after finish-frying processing steps to assess its role in acrylamide development (Figure 3). Asparagine levels within potato strips varied minimally across treatments (*accumulated* vs *added* sugar) and lengths of tuber storage (Figure 3A), consistent with prior evidence that asparagine levels remain relatively constant over extended storage at 2−18 °C.[34] Free asparagine contents within strips of both the *accumulated* and *added* sugar strategies were significantly reduced ($p < 0.05$, statistical notation not shown) after finish-frying (Figure 3B), most likely due to asparagine consumption in thermal reactions (Maillard color, flavor, and aroma development, as well as acrylamide formation).[35−37] Very minimal variability in free asparagine was noted between the two treatments or across tuber storage times after finish-frying (Figure 3B), while free asparagine content (postdrying) did not correlate with acrylamide formation in finish-fried strips (data not shown). Because of the lack of variation in free asparagine levels observed over tuber storage (Figure 3A,B) and the observation that variations in free asparagine levels did not match patterns of acrylamide formation previously observed (Figure 1B), asparagine was not the limiting substrate for acrylamide formation.[18,38]

Sugar contents (glucose, fructose, and sucrose) were tracked within raw tubers, as well as within potato strips after drying (postblanch) and finish-frying processing steps (Figure 4). Within the raw tissue, cold storage of tubers within the

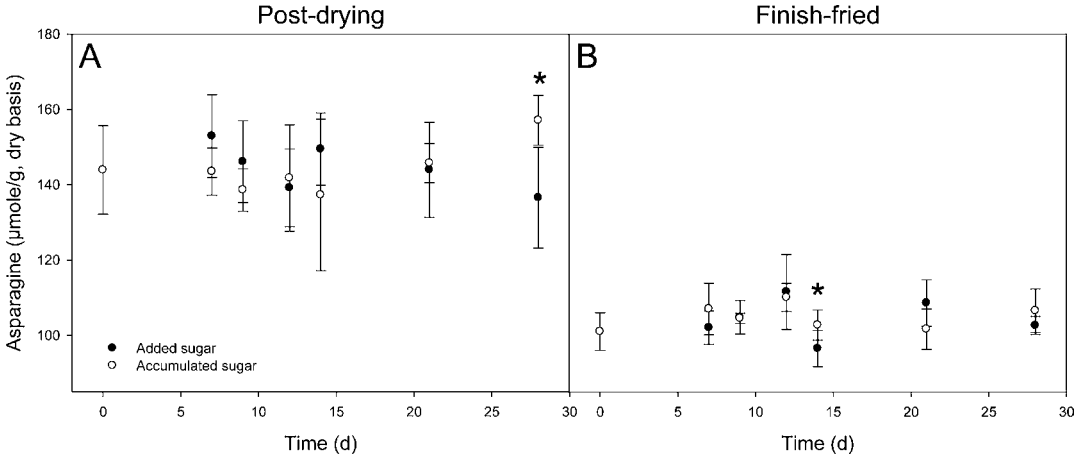

Figure 3. Asparagine contents within postdried (A) and finish-fried (B) potato strips for the *accumulated* and *added* sugar color development strategies over 28 days of tuber storage. An asterisk above a set of data points denotes a significant difference between values of the *added* and *accumulated* sugar treatments for a particular tuber storage time interval ($p < 0.05$).

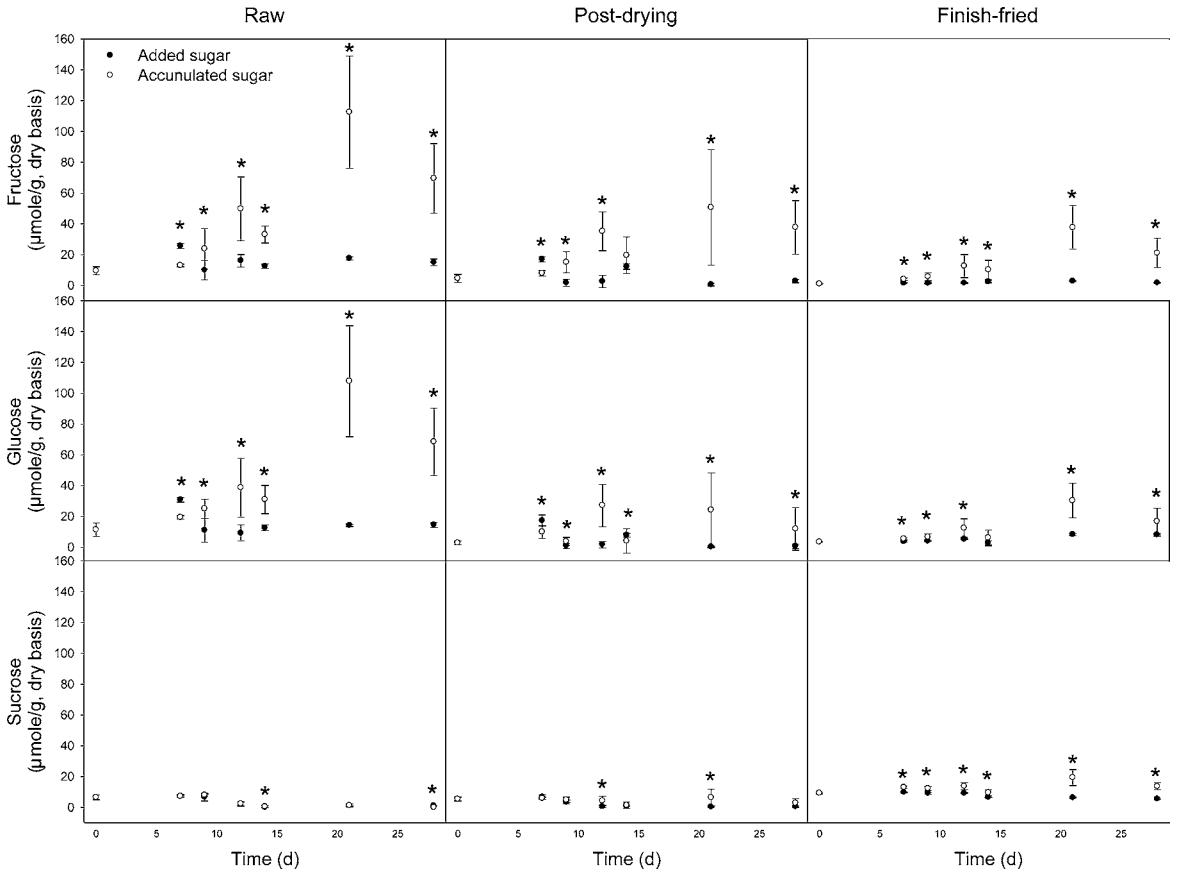

Figure 4. Sugar (fructose, glucose, and sucrose) concentrations within raw, postdried, and finish-fried potato strips for the *accumulated* and *added* sugar color development schemes over 28 days of tuber storage. An asterisk above a set of data points denotes a significant difference between values of the *added* and *accumulated* sugar treatments for a particular tuber storage time interval ($p < 0.05$).

accumulated sugar treatment resulted in elevated fructose and glucose contents over the course of the 28 day study. In contrast, tuber sugar levels managed via the *added* sugar scheme varied minimally over the 28 day storage period due to the lack of a cold temperature-induced sweetening effect. These same overall trends generally remained true for potato strips processed from tubers of the two color development treatments (*accumulated* vs *added* sugar) after drying (postblanch) and finish-frying processing steps. Thus, blanching itself did not entirely eliminate differences in reducing sugar patterns or contents within processed strips of the two color development schemes. In fact, fructose and glucose concentrations within potato strips of the *added* sugar treatment were generally lower than those obtained via the *accumulated* sugar scheme (postdrying stage), even though glucose had been incorporated onto *added* sugar strips postblanch for color development purposes. In comparing sugar levels within raw and processed potato strips of the *added* sugar scheme at the postdrying

dx.doi.org/10.1021/jf302114s | J. Agric. Food Chem. 2012, 60, 8763−8771

Table 1. Fructose, Glucose, and Sucrose Contents[a,b] for Potato Strips of the *Added* Sugar Treatment before (Raw State) and after Blanching/Dipping/Drying (Postdrying)[c] Processes

		storage time (days)						
		0	7	9	12	14	21	28
fructose	raw	9.61 ± 2.54a	25.74 ± 1.77a	9.99 ± 6.23a	16.06 ± 4.03a	12.54 ± 1.36a	17.58 ± 1.00a	14.94 ± 2.20a
	postdrying	4.56 ± 2.59b	17.08 ± 1.71b	1.80 ± 2.28b	2.60 ± 3.88b	12.31 ± 1.83a	0.59 ± 0.92b	2.90 ± 1.10b
glucose	raw	11.40 ± 4.26a	30.91 ± 1.59a	11.04 ± 7.67a	9.15 ± 5.18a	12.48 ± 1.88a	14.21 ± 0.75a	14.59 ± 1.74a
	postdrying	2.73 ± 1.27b	17.22 ± 3.71b	0.86 ± 1.64b	1.64 ± 2.02b	7.80 ± 1.29b	0.22 ± 0.53b	0.62 ± 1.52b
sucrose	raw	6.51 ± 1.51a	7.39 ± 0.92a	6.71 ± 2.51a	2.24 ± 1.04a	0.00 ± 0.00b	1.09 ± 0.07a	1.18 ± 0.04a
	postdrying	5.45 ± 1.22a	6.78 ± 1.47a	3.52 ± 1.32b	0.56 ± 00.73b	0.80 ± 0.07a	0.34 ± 0.38b	0.52 ± 0.40b

[a]Values within a given sugar category and column sharing a common letter are not significantly different ($p < 0.05$). [b]Values are reported as μmol/g, dry basis. [c]Potato strips were blanched (74 °C for 7 min), dipped in a solution containing SAPP (0.75%, w/v) and glucose (0.00−0.60%, w/v, to achieve target color), and dried at 60 °C for 5−10 min to achieve a 12−14% moisture loss.

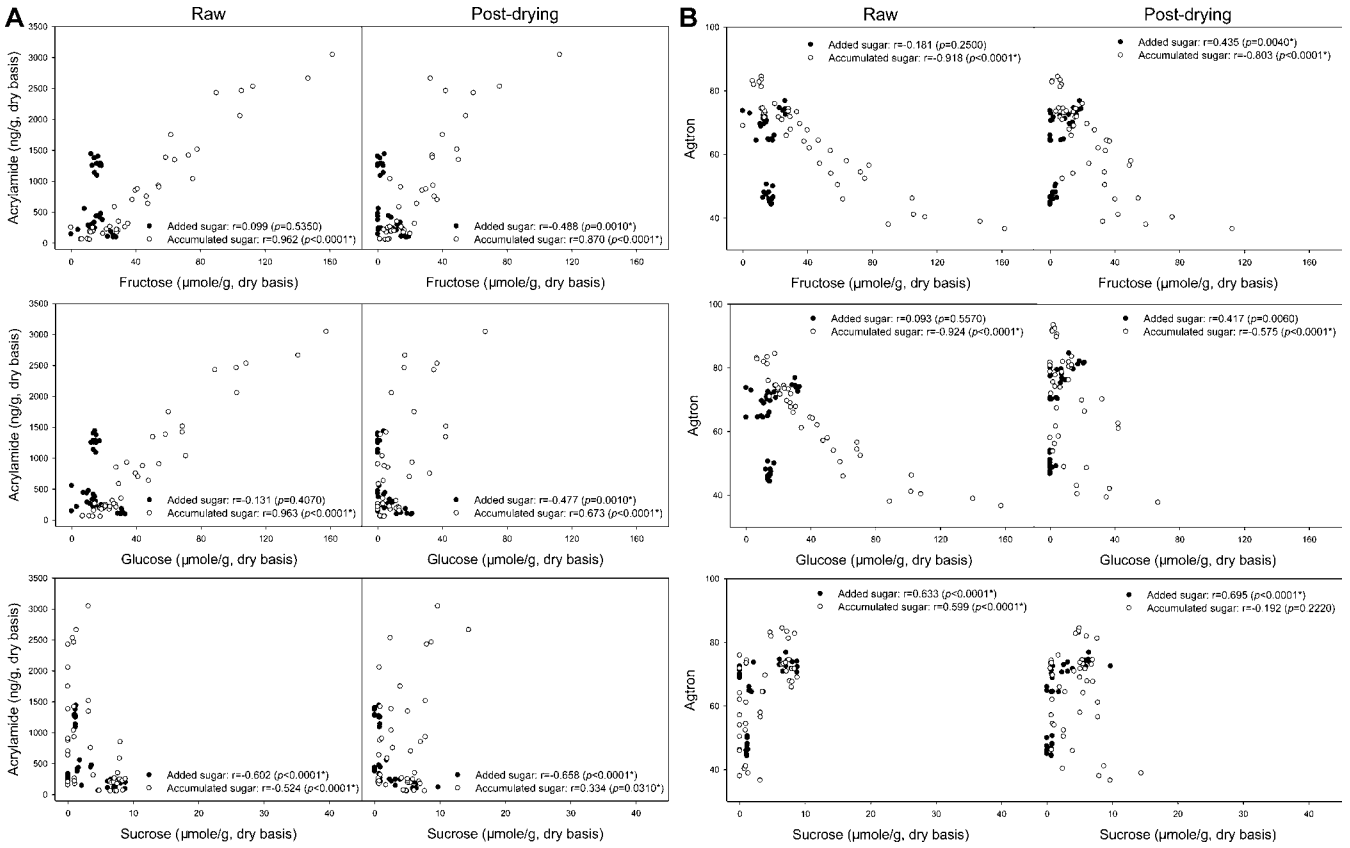

Figure 5. Correlations between sugar (fructose, glucose, and sucrose) concentrations and acrylamide contents (A), and Agtron scores (B), for raw and postdried potato strips of the *accumulated* and *added* sugar color development schemes.

processing stage (after blanching and dipping steps), more glucose and fructose were generally leached from potato strips during blanching[39] than were added back by dipping, resulting in a net loss of glucose from strips (Table 1).

Instances where statistical differences in sugar (Figure 4) and acrylamide (Figure 1B) contents between *accumulated* sugar and *added* sugar treatments after drying or finish-frying could not be discriminated were largely attributable to the highly variable nature of sugar and acrylamide levels within *accumulated* sugar strips. In short, the higher relative reducing sugar contents of dried potato strips of the *accumulated* sugar compared to those of the *added* sugar strategy could explain in part their slightly higher acrylamide concentrations after frying. Further, the shapes of the fructose and glucose curves (across the tuber storage period) at the raw, postdrying, and postfrying processing stages (Figure 4) appeared to most closely mirror that observed for acrylamide formation (Figure 1B).

***Accumulated* versus *Added* Sugar on Acrylamide Formation and Color Development.** To gain further insight into acrylamide formation and color development, the range of fructose, glucose, and sucrose contents encountered within raw tubers, as well as potato strips at the postdrying processing stage just prior to frying, were plotted against the acrylamide contents and Agtron scores of finished-fried strips (Figure 5, panels A and B, respectively). In the case of the *accumulated* sugar treatment, contents of glucose and fructose, but not sucrose, exhibited strong, positive correlations with acrylamide formation at both raw and postdrying stages of processing (Figure 5A, white data points). The highest levels of acrylamide were observed in finish-fried strips of the *accumulated* sugar treatment, which strips possessed the highest

levels of reducing sugars. Thus, high reducing sugar content prior to frying was associated with elevated levels of acrylamide. Sucrose, a nonreducing sugar, does not directly contribute to acrylamide formation. While it may undergo hydrolysis during high temperature frying to yield reducing sugars capable of reacting to form acrylamide,[40] no direct evidence of this phenomenon was observed in our experiments. Similar, yet inverse, correlations were observed between Agtron scores of finish-fried strips and sugar contents for raw and dried potato strips of the *accumulated* sugar treatment (Figure 5B), with sucrose content exhibiting weak correlation for reasons previously discussed.

In contrast, the *added* (relative to the *accumulated*) sugar treatment did not exhibit strong correlations between the reducing sugar contents of raw or processed (postdried) strips and the acrylamide contents of finished-fried potato strips (Figure 5A, black data points). The approximate ranges of fructose and glucose contents within both raw and dried strips of the *added* sugar treatment (0−40 and 0−20 μmol/g, respectively) were much narrower than those of the *accumulated* sugar treatment (0−160 and 0−120 μmol/g, respectively) (Figure 5A). Potato strips of the *added* sugar treatment were submerged in glucose solution for only a very brief time period (30 s), minimizing the extent of glucose diffusion into the potato tissue matrix and effectively limiting most of the *added* glucose to strip surfaces. The majority of acrylamide formation and color development occurs at or near fried strip surfaces, where the highest temperature is achieved during deep frying.[41] Consequently, a relatively high proportion of the *added* glucose was converted to Maillard products and acrylamide, resulting in an almost vertical arrangement of data points along the *y*-axis due to the narrow range of glucose concentrations within strips (Figure 5A). Similar, yet inverse, trends were observed between reducing sugar concentrations of dried strips and Agtron scores of finish-fried strips (Figure 5B).

For the *added* sugar treatment, two separate groupings of data points were observed within plotted correlations (Figures 2 and 5A,B), constituting two distinct levels of acrylamide and color development. Upon closer analysis, data points of the higher acrylamide (or lower Agtron score) grouping coincided with tuber storage times of 21 and 28 days, which potato strips were dipped in higher glucose concentrations (0.65 and 0.60%, w/v, respectively) to match the color of fried strips of the *accumulated* sugar scheme at corresponding lengths of tuber storage. Conversely, the lower acrylamide (or higher Agtron score) grouping corresponded to the shortest tuber storage times (7, 9, 12, and 14 days), which strips were dipped in lower concentrations of glucose (0, 0.07, 0.1, and 0.20%, w/v, respectively). Though only minimal variability in measured glucose concentrations was observed among *added* sugar potato strips (postdrying) of the various storage times (Figure 4), it is evident, based on both observed color and acrylamide levels (Figure 1), that actual differences in glucose concentrations at strip surfaces among the storage times existed. Though the vertical arrangement of acrylamide data points was observed for all plots (fructose, glucose, or sucrose vs acrylamide) (Figure 5A), this effect was almost certainly attributable to *added* glucose. This notion is substantiated by plotting acrylamide levels and/or Agtron scores in relation to the glucose concentrations of the dipping solutions (Figure 6). Though concentrations of reducing sugars within potato strips of the *accumulated* sugar treatment were generally higher than those of the *added* sugar scheme (Figure 4), they were also more

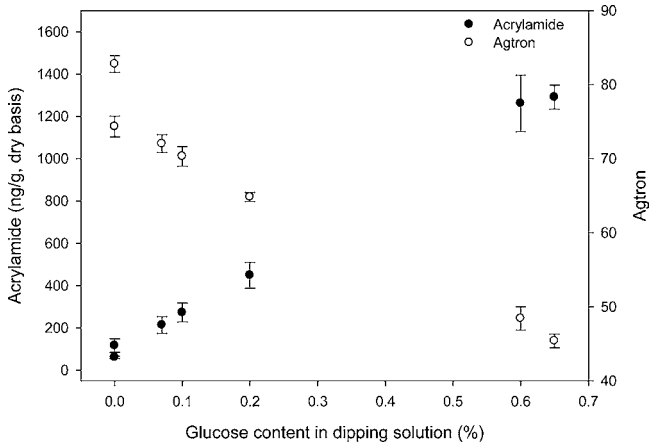

Figure 6. Acrylamide contents and Agtron scores of finish-fried potato strips versus glucose concentration of the dipping solution for the *added* sugar color development treatment.

likely to be distributed throughout the tissue (as opposed to surface-located), resulting in relatively less effective conversion to both acrylamide and Maillard browning products. For the *added* sugar treatment, both color development and acrylamide contents of finish-fried strips were almost exclusively derived from reaction with *added* glucose, while for the *accumulated* sugar scheme, both fructose and glucose likely participated in these reactions. Thus, it was not clear whether the relatively higher acrylamide levels observed within finish-fried strips of the *accumulated* sugar scheme were due to the higher relative concentrations of total reducing sugars (Figure 4) or differing proportions of reducing sugars present at strip surfaces.

Effect of Fructose to Glucose Ratio on Acrylamide Formation and Color Development. Mestdagh et al.[28] reported that the ratio of fructose to glucose impacted both color and acrylamide levels of fried potato strips, with a relative higher fructose concentration favoring acrylamide concentration. To verify the variable effects of fructose and glucose on acrylamide formation and color development according to defined processing conditions utilized within this study, potato strips were dipped (30 s) in a 0.30% (w/w) total sugar solution comprised of variable fructose to glucose ratios (100:0, 75:25, 50:50, 25:75, 0:100). Agtron scores for finish-fried potato strips of all *added* sugar treatments (62−71) fell within an acceptable range typical of fries for quick serve restaurants,[30] contributing validity to the experiment. A higher proportion of fructose (relative to glucose) within strips favored higher acrylamide formation and higher Agtron scores (i.e., less color development) after finish-frying (Figure 7), whereas the reverse relationship was observed for strips containing higher relative proportions of glucose. In relating these findings to prior experiments contrasting *accumulated* versus *added* sugar color development strategies, a relatively lower concentration of glucose (compared to fructose) would be required at strip surfaces to achieve equivalent levels of color development, with the possible added benefit of relatively lower acrylamide levels. Given that both fructose and glucose present at strip surfaces are likely to be similarly consumed in Maillard/acrylamide reaction pathways during frying, the tendency for the *accumulated* (relative to the *added*) sugar treatment to generate higher levels of acrylamide (given equivalent levels of color development) is likely aided by its higher fructose content

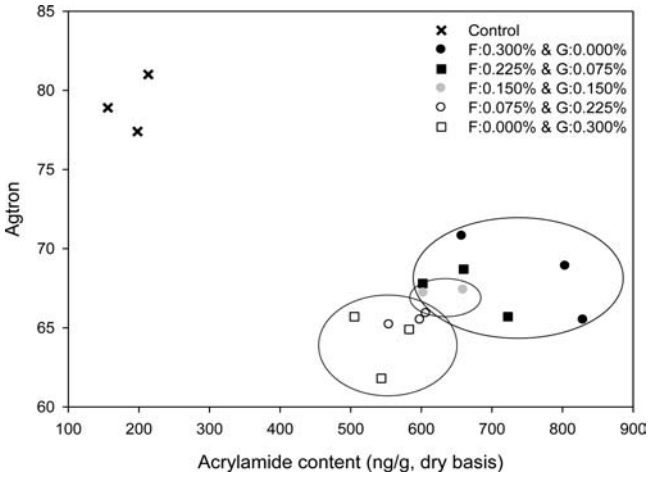

Figure 7. Agtron color scores versus acrylamide contents for finish-fried potato strips dipped in sugar solutions (0.3%, w/w) comprised of varied fructose (F)/glucose (G) ratios. (Control strips were dipped in water.)

rather than simply its overall greater concentration of total reducing sugars.

In summary, in contrasting the effects of *added* and *accumulated* sugars on acrylamide formation, fried potato strips of the *added* sugar treatment generally exhibited lower levels of acrylamide (for similar levels of color development), despite the fact that *added* glucose was most likely localized at strip surfaces (as a function of dipping), where thermal reactions are most favored to occur. Conversely, for the *accumulated* sugar treatment, it can be theorized that both fructose and glucose were likely present at strip surfaces in essentially equimolar proportions, as both sugars are derived directly from sucrose via cold temperature-induced sweetening. In noted dipping experiments contrasting the effects of *added* glucose and fructose on color development and acrylamide formation, glucose contributed more to color and less to acrylamide formation, whereas the reverse phenomenon was observed for fructose. Consequently, in instances where differences in acrylamide content were observed between *added* and *accumulated* sugar treatments, these differences likely had more to do with the type (fructose versus glucose), rather than strictly the total amount, of reducing sugars available for reaction at strip surfaces. This scenario is further substantiated by the observation that *added* and *accumulated* sugar treatments exhibited two significantly different rates of acrylamide formation, even though fried potato strips of the two methods had been standardized to yield similar levels of color development.

While it is clear that the lack of *added* sugar (i.e., glucose addition), coupled with practices that minimize inherent sugar accumulation in tubers, would further reduce acrylamide formation in commercial fried potato strips, the resulting product is unlikely to meet consumer expectations due to an unacceptably light color. Thus, efforts to minimize reducing sugar formation in tubers, coupled with optimization of blanching conditions (to leach away *accumulated* sugars) and controlled augmentation with glucose (to achieve target color), likely result in less acrylamide formation, as compared to the alternative of allowing reducing sugars inherently present within the tuber to drive color development. Such practices, in addition to efforts to acclimate consumers to lighter colored products, could help to reduce consumer exposure to acrylamide in relation to fried potato products.

Aside from absolute differences in acrylamide levels, fried potato strips produced via the *added* (relative to the *accumulated*) sugar treatment exhibited much less variation in both color and acrylamide levels, affording a much greater ability to accurately predict and control these parameters within a large-scale industrial process. The ability to accurately monitor and predict acrylamide levels within fried potato products at the industrial level not only provides a reliable benchmark for estimation of population exposure to acrylamide but also supports continuing efforts to understand and mitigate acrylamide levels in food.

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Notes

The authors declare the following competing financial interest(s): Authors Gordon Smith and Jeremy Higley are employed by ConAgra Foods, which is a manufacturer of frozen french fries.

ACKNOWLEDGMENTS

The authors gratefully acknowledge the significant contributions of Greg Hoffman, Bob Hill, Indarpal Singh, and Matt Hetzler to experimental execution and technical analyses.

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