Effect of Additives and Color on Levels of Acrylamide in Chips and Crisps from Selected Solanum Tuberosum Tubers

Obed M. Nyabaro

[1]*Department of Chemistry, Kisii University, Kenya*

omainya@kisiiuniversity.ac.ke

https://doi.org/10.62049/jkncu.v5i2.321

Abstract

Acrylamide is a small and simple molecule that could be formed in heated foods via several different mechanisms, which may involve reactions of carbohydrates, proteins and amino acids, lipids and possibly other minor food components. It is a chemical substance formed by a reaction between amino acids and sugars (dissacharides) typically occurs when foods with high starch content such as potatoes, root vegetables and bread, are cooked at high temperatures (over 120°C) in a process of frying, roasting or baking that is capable of forming the Maillard reaction. A total of ten samples for analysis of levels of acrylamide due to use of additives and colour formation, were collected using stratified and simple random sampling techniques. They were collected from selected retail outlets and hotels from different regions within the county during three different times within a spell of three months. Analysis was done in triplicates. The presence and concentration were analysed using High Performance Liquid Chromatography (HPLC). Colour analysis was done using colour spectrophotometer, based on the three colour parameters; lightness, yellowness and redness, the lightest value was 66.31 nm, yellowness of 34 nm and 6.9 nm in the redness. The lower parameter values were experienced in chips samples where lightness was 36.31 nm, yellowness 3.3 nm and 26 nm for redness parameters. Samples for analysis on effect of additives and flavours on levels of acrylamide were collected from vendors that stocked the flavored chips and crisps. Five samples for each flavour and additive were analysed using HPLC. The lowest levels were found to be 0.87 ± 0.01 mg/kg in bhajia and the highest 1.83 ± 0.01 mg/kg in garlic and peppered chips while for crisps, salted and vinegar samples recorded concentrations of 17.2 mg/kg and 49.2 mg/kg respectively. Results from this research on effect of colour were still within the range of chips products and this could be due to the Maillard reaction that occurs during cooking and therefore, these additives would not reduce the reaction and acrylamide levels while additives in potato crisps and chips have an impact on the levels of acrylamide since these are added to the raw potatoes during soaking before they are cooked. These have an effect on the reducing sugars and asparagine that reacts as a result of temperature rise to produce acrylamide unlike the additives in potato chips that are added after cooking.

Keywords: Acrylamide, Dissacharides, Maillard, Chromatography, Melanoidins

Introduction

The preparation of potato chips at high temperature can lead to the formation of acrylamide [1]. The browning reactions that occur when meat is roasted or seared are complex and occur mostly by Maillard browning [2]. Temperature plays a major role in determining the levels of acrylamide (AA) in different cooking temperatures have been shown to have direct relationship with acrylamide with contributions from other chemical reactions, including the breakdown of the tetrapyrol rings of the muscle protein myoglobin.

The Maillard reaction has been described that upon gently heating sugars and amino acids in water will generate a yellow brown colour [3:4]. Maillard reaction has been well understood as a non-enzymatic reaction between reducing sugars and amino acids to generate the Maillard reaction products (MRPs) [4:5].

The application of food processing practices to generate MRPs can improve the oxidative stability of food products and preserve food from oxidation and microorganism contamination as well [5:6]. Maillard reaction is responsible for many flavors and colours in foods, such as coffee roasting, browning of various meats when seared or grilled, browning and umami taste in fried onions. It contributes to the darkened crust of baked foods, the golden-brown colour of potato chips and other crisps, of malted barley as found in malt beer and whiskey, and the colour and taste of dried and condensed milk, confection milk toffee, chocolate, roasted peanuts and black garlic.

Acrylamide Formation

6-Acetyl-2,3,4,5-tetrahydropyridine is responsible for the biscuit or cracker-like flavor present in baked goods such as bread, popcorn, and tortilla products. The structurally related compound 2-acetyl-1-pyrroline has a similar smell, and also occurs naturally without heating and gives varieties of cooked rice their typical smells. Both compounds have odor thresholds below 0.06 ng/l [7].

Caramelization is an entirely different process from Maillard browning, though the results of the two processes are sometimes similar to the naked eye (and taste buds). Caramelization may sometimes cause browning in the same foods in which the Maillard reaction occurs, but the two processes are distinct. They are both promoted by heating, but the Maillard reaction involves amino acids, as discussed above, whereas caramelization is simply the pyrolysis of certain sugars.

Temperature has been stated as important and key for producing MRPs. It has been recognized that significant increase of MRPs were obtained after an increase of temperature from 50 ºC to 60ºC [8], hence MRPs was temperature-dependent products. On the other hand, Maillard reaction relied on the pH of medium. It found out that increase in pH medium might enhance the reaction of Maillard [5]. Several factors in the reaction, which are reactants type and concentration, temperature, heating time, pH, and humidity [5 & 5] could not be disregarded. It is generally concluded that reactants and reaction conditions truly affect the result of final Maillard reaction products [10]. Scheme 1 represents Maillard reaction.

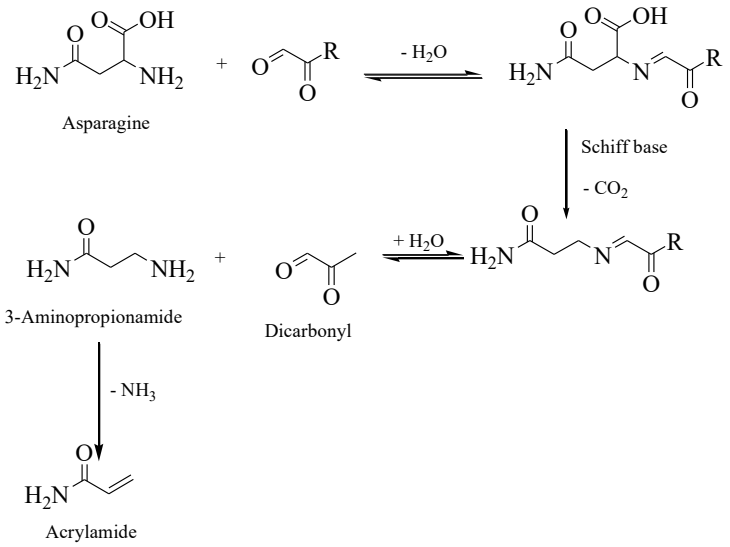

Figure 1: Maillard Reaction

Table 1 represents Acrylamide levels in cooked potato products by showing the minimum levels and maximum levels that are expected.

Table 1: Acrylamide levels in cooked potato products

Acrylamide levels	**Minimum**	**Maximum (Mg/Kg)**
Potatoes (raw)	<10	<50
Potatoes (boiled)	<4	<50
Potato crisps (also called crisps)	<117	<4215
French fries (chips)	<59	<5200

Sources: [11, 12 & 13].

Process of Formation and Reduction of Acrylamide

Acrylamide is known to be a highly reactive molecule. It can react by ionic and by free-radical mechanisms and its presence, in free form, in food, was therefore unexpected. The observation of relatively high levels in certain foods rich in carbohydrates, and lower levels in protein-rich foods, may reflect the relative ease of formation in the former, or it may be due to volatilization or further reactions between acrylamide and food components in the latter [14 & 15].

The reactive carbonyl group of the sugar interacts with the nucleophilic amino group of the amino acid, and interesting but poorly characterized odor and flavour molecules result. This process accelerates in an alkaline environment because the amino groups do not neutralize. This reaction is the basis of the flavoring industry, since the type of amino acid determines the resulting flavour.

It all starts with a sugar and a protein/amino acid. These react and form a compound A reducing sugar (either an aldose or a ketose) reacts with an amine-group (an NH_2-group, from the protein) to form a so-called Heyns or Amadori compound. (R-NH_2) reacts with; If it's a sugar with an aldehyde group (-COOH) it will be an Amadori. If it's a sugar with a ketone group (-CO-) the Heyns compound will be formed. These molecules react further to form aromatic compounds. From a chemical structure perspective: most of these

molecules contain a ring in their structure which is formed in this stage. In the final step large complex molecules, melanoidins are formed. These will eventually give the product a brown colour. Scheme 2 represents the structure on the formation and reduction of acrylamide.

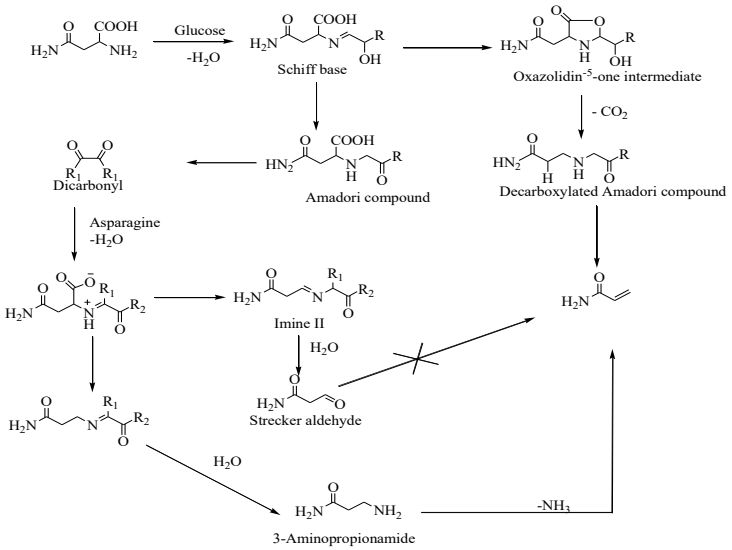

Figure 2: Structure on the formation and reduction of acrylamide [15].

Asparagine and reducing sugars are important determinants of acrylamide formation, it was demonstrated that relative reductions in acrylamide concentrations are possible by controlling the reducing sugar levels in potato cultivar. Asparagine presence in food with glucose or 2, 3-butane-dione (one of several dicarbonyl compounds formed in the Maillard reaction) causes significant amounts of acrylamide to form in dry products, but only trace amounts would form when asparagine is replaced with other amino acids. Heating asparagine on its own at 185°C does not produce acrylamide, confirming the requirement for the dicarbonyl reactant to be present and the Strecker degradation to occur [16]. However, replacing glucose with other carbohydrates (D -fructose, D -galactose concurrently with replacement of asparagine led to a significant amount of acrylamide to be formed.

Effect of Colour on Acrylamide Formation

Acrylamide (AA) formation depends on the system used in terms of constituents, such as oil oxidation and type, and processing conditions. Findings from model potato system suggested that lipid oxidation products do not affect AA formation [17,18,19 & 20]. However, studies carried out on baked cookies concluded that lipid oxidation products should be considered as an important factor in AA formation during baking of fat-rich products but only after prolonged heating time [21, 22,&23].

Colour is one of the most important food product characteristics, as it is the first quality parameter evaluated by the consumers. Recently, several researchers investigated the relation between the content of AA and surface colour, as evaluated by the standard CIE L*a*b* parameters or by computer vision. In cases, such as fried potatoes and cookies, there was a good correlation between the AA content and their colour [24, 25, 26 &27].

On the other hand, addition of amino acids, other than asparagine, is known to decrease AA formation and at the same time to promote flavour and colour generation. Specifically, many authors have reported an increased browning of their potato model system, bread and cereal products, when glycine was added [28, 29 & 30].

Colour may be used as an easily assessable and fast indicator of AA content by consumers. In this respect, the study aimed at checking the effect of AA levels on colour development. Colour development only begins when sufficient amount of drying has occurred in food and depends also on the drying rate and the heat transfer coefficient, during the different stages of baking. Surface browning is a common phenomenon for baked foods. As a generic visual feature, surface browning of thermally processed foods can be monitored by means of computer vision-based image analysis; thus, surface colour of baked potatoes was evaluated using this method.

Pearson correlations coefficients, between examined colour parameters and AA content, showed that AA is better correlated with browning ratio (R=0.89). Browning ratio was calculated from the segmented images of baked potatoes. Figure 1 shows visually how the potato chips get more red and darker as the browning ratio and AA content increase. The browning simply developed as a circle, which grew from the edge to the centre of potato discs, as the baking proceeded. It was observed that browning ratio more than 40 % resulted in dark brown almost burnt products, while a browning ratio of less than8 % resulted in an amount of AA less than 1000 g/kg, which is an indicative value that has been recently set for potato crisps by the European Commission [31]. Figure 3 represents the relationship between acrylamide content and browning Ratio.

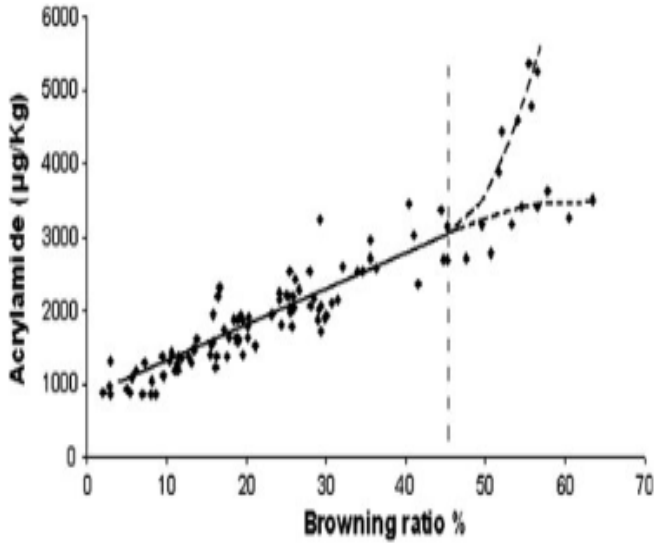

Figure 3: Relationship between Acrylamide content and Browning Ratio

Source: [18 & 32].

Acrylamide (AA) content, in the baked potato product, was correlated linear to browning, when its ratio ranged between 0 and 45 % Figure 3. After excess browning (45 %), AA formation and degradation occur simultaneously. Thus, there is no correlation (R2\0.1) between AA content and browning ratio. For

browning ratio less than 45 %, the curve slopes ranged between 47.099–51.585 and 55.873–63.864, when unheated and thermoxidised oils were used, respectively.

A linear correlation between AA content and colour has also been reported for frying potato products [32]. The potato variety, used for frying, had a strong effect over the total colour difference parameter (DE) of the potato slices. These decreased considerably, due to browning reactions that took place during frying; the higher the frying temperature, the faster the slices tended to get red. Similarly, a significant correlation was observed between the browning ratio and AA concentration in cookies [27].

Furthermore, at more severe processing conditions, the results are coincident with those found by other authors, who studied potato products and coffee. Taubert [33] reported a lack of correlation between surface browning and AA content, in potato material with high surface to volume ratio, due to net degradation of AA at long heating time, which was accompanied by a further increase in browning. Moreover, the results obtained by Gkomen [25] revealed that the dark-coloured coffee contains much lower amounts of AA than light coloured coffee, due to the exponential degradation at prolonged heating time at high temperatures.

The variables examined were browning, chromatic parameters a*, L* and b*, residual moisture and baking time. High statistical correlation coefficients were exhibited among browning ratio, moisture, chromatic component a*, time and the AA level in baked potato products, whereas CIE L* and CIE b*parameters did not show any significant correlation with AA content, as shown by Pearson correlation coefficients. Luminosity (L*) diminishes with baking time, since some parts of surface tend to get darker, as a result of browning during baking, whereas chromatic colour component b* does not change.

According to Gökmen and Senyuva, under these conditions, SLRA selected the browning ratio, as having the maximum correlation with AA formation. In such a correlation, the R^2 indicates how much of the variability in the outcome is accounted for by the individual predictors. The R^2 value was found to be 0.804 for the first model, which means browning accounts for 80.4 % of the variation in AA. The adjusted R^2 is very close to R^2, which indicates that our model generalizes ideally. It is concluded that the browning ratio may be considered as a reliable indicator of AA concentration in our system[25].

Materials and Methods

Sample Collection and Preparation

The study adopted an experimental design which involves quantitative data collection methods through laboratory analysis. The samples were prepared before analysis to avoid contamination and ease the analytical procedure on the instrument. A total of ten samples were collected in effect of colour on the levels of acrylamide and another ten samples obtained on the effect of additives on the levels of acrylamide, ready for analysis.

Potato chips and crisps samples were prepared for acrylamide analysis by weighing 1 g of crushed potato chips/crisps and mixing with 10 ml methanol on a wrist action shaker for 20 minutes to enable the sample to fully soak in the extract solution. The samples were refrigerated for 2 minutes for easier extraction of the oily top layer to avoid interferences. The supernatant was filtered through a 0.20 μm nylon syringe (silica based) filter and the filtrate discarded, and the residue stored for further cleanup and analysis.

The residue was conditioned through a carboPrep™ 200 SPE (silica based) tube using, 6 ml of the sample in 2 ml acetone and 2 ml methanol. The sample solution was allowed to pass through the tube by gravity and 0.5-1.0 ml water was run through the tube to wash the sample. A vacuum pump was used to dry excess water from tube for 1 minute followed by elution with 2 ml of acetone through gravity therefore ready for analysis in the instrument. Many sample extracts can be analysed directly, however, sample cleanup and solvent pre-concentration was essential.

Colour Determination

These were evaluated by a colour spectrophotometer as described by Abong [34] and measured with FM-722 high quality colour spectrophotometer using the CIE Lab L, a and b colour scale. The 'L' value which is the lightness parameter indicator for the degree of lightness of the sample was varied from 0 = black to 100 = white. The chromatic redness parameter, 'a' whose value, when tending to red colour means positive (+) and green colour negative (–). The yellowness chromatic parameter 'b corresponds to yellow colour when it is positive (+) and blue colour when it is negative (–). Each sample was measured in triplicates for precision.

Determination of the Effect of Additives in Potato Chips and Crisps in Acrylamide Levels

Different additives of both chips and crisps collected were randomly analysed using the standard extraction and preparation procedure, starting from their mean to determine their effect in acrylamide levels. They were randomly identified as represented in Table 2

Table 2: Types of Additives in Chips and Crisps Analysed

S/No.	Sample identification	Chips	Sample identification	Crisps
01	FA1	Bhajia	FA6	Salt and vinegar
02	FA2	Hot and spicy	FA7	Tomato crisps
03	FA 3	Tomato sauce	FA8	Sour cream and onion
04	FA4	Cheese and onion	FA9	Chilly lemon
05	FA5	Garlic and pepper	FA10	Babeque

Data Analysis

Both descriptive and inferential statistical tests including analysis of variance (ANOVA) and least significant difference test (LSD) for the variables were carried out using the Statistical Analysis System (SAS) version 9.1.3. Pearson correlation analysis and multiple regression analysis were also performed to determine relationships between acrylamide and colour parameters at $p < 0.05$.

Introduction and Preparation for Analysis

A 4-Digit Laboratory Weighing Balance Electronic Analytical Scale Four Decimal, model FA2004B from China was used for weighing. Detection and quantitation of acrylamide in chips and crisps were determined using High performance liquid chromatography (HPLC). Agilent 1100 (Waldbronn, Germany) HPLC system consisting of a quaternary pump with vacuum degasser, a DAD was used. Chromatographic separations were performed on an ODS-3 C^{18} column (250 mm × 4.6 mm, Intersil, Japan).

Sample concentrations were conducted on RE-2000 rotary evaporator (Shanghai Yarong Biochemical Apparatus Company, limited, Shanghai, China) and the solution thoroughly mixed using a Vortex mixer (Shanghai Qite Analytical Apparatus Company, limited, Shanghai, China). This was done before analysis.

A HL-2070 multi-function food processor (Shanghai Herine Electric Appliance Company limited, Shanghai, China) was used to pulverize and homogenize samples. Sample extractions were performed using HS2060A ultrasonic shaker (Kunshan Ultrasonic Instrument Company limited, Kunshan, Jiangsu, China).

Centrifugal separation was carried out by using A Refrigerated Centrifuge (Biofuge stratos, Germany). While the residue was conditioned through a carboPrep™ 200 SPE made in Germany, Filtered using 0.2 µm micro-filters both made in the USA. A 60 ml separatory funnels, 50-ml round-bottom flasks, brown glass tubes, autosampler vials, a stainless steel pan with 6 litre capacity, 24.3 cm inner diameter and 0.5 cm thickness stainless steel lid with hermetic property and an electrical heater (model thermal, HK3-2, Germany) while, Accu-BOND Si (6 ml, 500 mg) solid-phase extraction (SPE) cartridges were supplied by Agilent Technologies (Santa Clara, CA) were used.

All solvents and chemicals used in the analysis procedure were of analytical grade. They were purchased from Merck (Darmstadt, Germany). Analytical water grade was used since the instruments were highly sensitive and to avoid breakdown after calibration for the HPLC instrument, analytical grade of HPLC water was used.

Acrylamide (standard) (99%) and $^{2}H_{3}$ -labeled acrylamide (isotopic purity 98%) was purchased from Sigma-Aldrich (St. Louis, MO) and Cambridge Isotope Laboratories (Andover, MA), respectively. Methanol (HPLC-grade) was supplied by Merck (Darmstadt, Germany). Water was purified with a Milli-Q system (Millipore, Bedford).

Standards and reagents stock solution of acrylamide (1 mg/ml) and $^{2}H_{3}$ –labeled acrylamide (0.1 mg/ml) were prepared by dissolving suitable amount of the compounds in water. These solutions were then appropriately diluted with water to prepare working standards at 10 and 4 µg/ml, respectively. All stock solutions and working standards were kept at 4°C for a month.

Potato chips and crisps samples were prepared for acrylamide analysis by weighing 1 g of crushed potato chips/crisps and mixing with 10 ml methanol on a wrist action shaker for 20 minutes to enable the sample to fully soak in the extract solution. The samples were refrigerated for 2 minutes for easier extraction of the oily top layer to avoid interferences. The supernatant was filtered through a 0.20 μm nylon syringe (silica based) filter and the filtrate discarded, and the residue stored for further cleanup and analysis.

The residue was conditioned through a carboPrep™ 200 SPE (silica based) tube using, 6 ml of the sample in 2 ml acetone and 2 ml methanol. The sample solution was allowed to pass through the tube by gravity and 0.5-1.0 ml water was run through the tube to wash the sample. A vacuum pump was used to dry excess water from tube for 1 minute followed by elution with 2 ml of acetone through gravity therefore ready for analysis in the instrument. Many sample extracts can be analysed directly, however, sample cleanup and solvent pre-concentration was essential.

Blank solutions (blank chromatograms) that were done every after change of the blank solutions that were used in sample preparations, different dilutions of the standards varying from 30 ppm, 20 ppm, 10 ppm, 5

ppm, 2.5 ppm and 1 ppm. Increase in dilution led to a better peak area. A dilution of 20 ppm was found appropriate for use while preparing standards. Figure 4 represents 20ppm dilution standard.

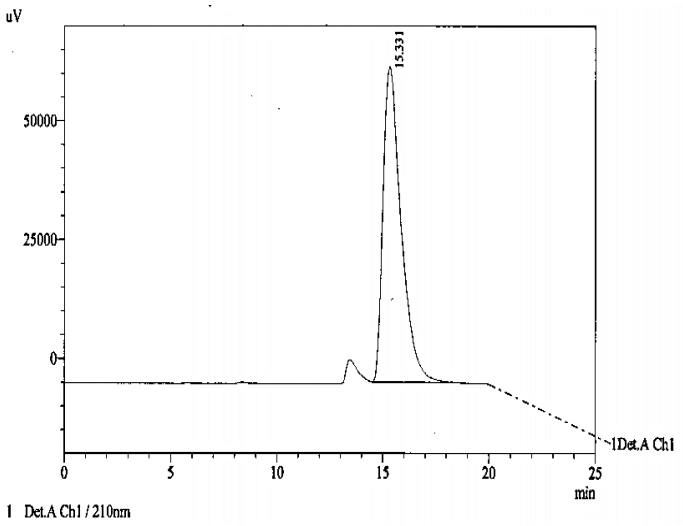

Figure 4: 20ppm sample standard

Further, application of the results gave a graph with the best of fit which resulted in a quadratic equation that gave a guideline towards determination of individual concentrations of the samples that were analysed.

Results

Colour Determination

Colour of the Crisp Samples

The colour of the crisp samples was quantified by means of a nondestructive technique based on digital image analysis [35]. Each crisp sample was snapped and the images stored and analyzed using ADOBE Photoshop, 7.0. The colour was transposed into L* (lightness), a* (redness), b* (yellowness) colour coordinates according to CIE system. The results are tabulated in Table 3.

Table 3: Colour of Crisp Samples

Sample	L* (nm)	a* (nm)	b* (nm)
Sample 1	66.31	6.9	34
Sample 2	43.23	3.2	26
Sample 3	61.51	-1.9	18
Sample 4	55.27	2.8	25
Sample 5	56.36	-0.8	31
Sample 6	60.12	3.9	24
Sample 7	57.57	4.1	20
Sample 8	52.78	6.3	18
Sample 9	53.31	5.5	30
Sample 10	59.37	-1.9	22

Samples collected and analysed were light coloured with L>50 apart from that of sample 2 which was the dullest with an L value of 43.23 nm indicating extreme browning. Sample 1 was the lightest with an L value of 66.31 nm. The least yellow was sample 3 with a* value of 18 nm while the highest value of yellowness parameter was obtained for sample 1 with b* of 34 nm. Most of the samples tended to the redness as most of the a* values were >0.0 indicating relatively high degree of browning, especially for samples 1, 4 and 7. The positive value of the yellowness parameter b* is an indication that all the samples in the study area tended towards yellow. Figure 5 shows a graphical comparison of the L, a and b for each of the samples.

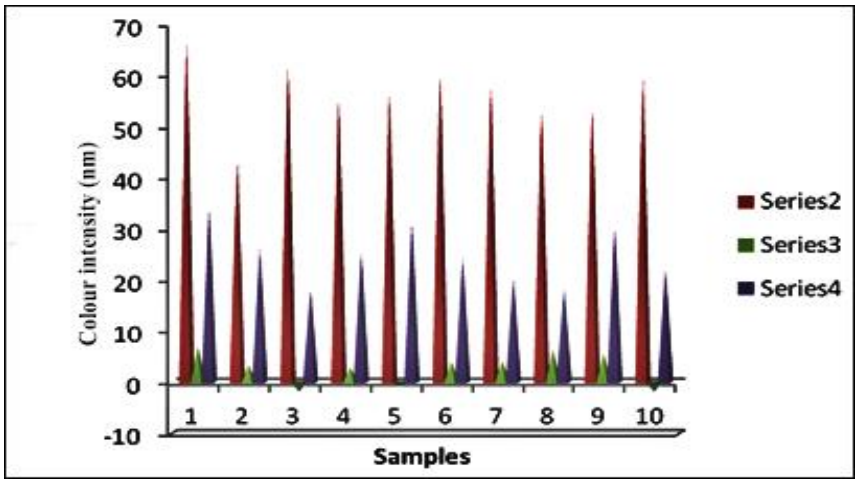

Figure 5: Comparison of the L, a* and b* for each of the crisp samples

Key: *Series 2 represent L*nm(Red), Series 3 represent a* nm (Green), Series 4 represent b* nm (purple)*

The negative values in Sample 3, 4, and sample 10 with values of -1.9 nm, - 0.8 nm and -1.9 nm respectively, could be due to undercooking of the crisps since they appeared to have had a lighter colour concentration, while, sample 1 had the highest redness value which attributes it to high levels of acrylamide.

These results match with those of Viklund [36] who analyzed the colour of potato crisps purchased from vendors in Lund, Sweden. Their findings indicate that for most of the samples, L>50, implying light colour. In addition, most of a* values of the samples were >0.5 indicating relatively high degree of browning. They attributed this to high levels of acrylamide in the samples. In contrary, Ogolla [37] in the test of acrylamide in crisps worked in Nairobi, Kenya, found out that most of the samples tended towards green as the redness parameter veered towards the negative indicating low degree of browning. This could be attributed to the probable low temperature cooking method used to cook crisps by most of the vendors resulting in low concentrations of acrylamide.

Cooking colour is an important quality characteristic in processed potato products such as potato crisps and potato chips. The light brown colour preferred by consumers is often produced when reducing sugar concentrations in tubers are low [25]. Colour development only begins when sufficient amount of frying has occurred in potato chips and depends also on the drying rate and the heat transfer coefficient during the different stages of cooking.

Colour of the Chips Samples

A total of ten potato chips samples were made, intensely brown coloured with HU>40 apart from those of samples 1 and 4. The presence of colour in food samples does not necessarily indicate toxicity. This is because colour-causing substances in most foods are harmless. Consequently, there are no set primary maximum permissible levels (P-MPL) of colour in foods. However, the Secondary Maximum Contaminant Level (S-MCL) by FDA [38 & 39] for colour concentration in foods is 20 HU mainly for aesthetic purposes [39]. L* (lightness), a* (redness), b* (yellowness) colour coordinates according to CIE system. The results are tabulated in Table 4

These samples had intense brown colour. This was attributed to the high cooking temperature while excessive browning of the starchy food products was found to increase at higher temperatures. This occurs as acrylamide compound is formed and broken down to glycidamide. Samples 1 and 4 had comparatively low brownness attributed to the method of cooking used in these two regions. These vendors mostly use charcoal jikos unlike the rest who use electric and gas cookers in cooking the potato chips.

In perfect conditions, the crisp samples should have been yellow in colour and this could be attributed to the fingerling and rose gold potato species commonly planted in this region which produce intensely yellowish potatoes. Table 4 shows the concentrations of colour in the samples.

Table 4: Colour concentrations of chips samples

Sample	L* (nm)	a* (nm)	b* (nm)
Sample 1	36.31	3.3	24
Sample 2	33.23	3.2	16
Sample 3	21.51	1.2	18
Sample 4	25.27	1.8	15
Sample 5	26.36	-1.8	21
Sample 6	20.80	2.9	24
Sample 7	27.25	2.1	19
Sample 8	22.14	1.3	18
Sample 9	33.33	2.5	20
Sample 10	19.37	-1.9	12

The current research findings are similar with those reported by [32] on the colour changes and acrylamide formation in potato chips samples from Santiago and Chile. Their findings showed that for both control and blanched potato chips, browning decreased dramatically as the frying temperature decreased from 180^0 to 120^0C. Additionally, there is a linear correlation between the non-enzymatic browning of potato chips and their acrylamide content for the range of the oil temperatures tested. Mestdagh [23] also observed a linear relationship between acrylamide content and Maillard browning potato chips, whereby their findings proved that there existed an exponential correlation between acrylamide formation and surface browning of the potato chips.

This relationship, however, appeared to be dependent upon the glucose/fructose ratio of the raw material. An excess of fructose compared to glucose stimulated acrylamide formation to a higher extent than Maillard browning. The opposite effect was established with an excess of glucose. The same results were reported

by Pedreschi [40] who noticed a high correlation (R^2 of 0.854) between browning and acrylamide content of potato chips. However, they discovered that browning reduced drastically when the potato strips were immersed in citric acid solution of 10 g/kg before frying.

Correlation Between Acrylamide Content and Colour

The results of correlation between acrylamide content in the samples and colouration are shown in Table 5.

Table 5: Correlation between acrylamide content and colouration

Parameters	Acrylamide	Colour
Acrylamide	1.000	0.410
Colour	0.410	1.000

The results indicate a positive relationship between the acrylamide contents and the colour content where R = 0.410. The positive correlation is attributed to browning of the starchy food products which increased at higher temperatures and also that acrylamide formation increased as the cooking temperatures increased. These findings agree with those of Rommens [41] who reported that browning of processed potato chips correlated positively with acrylamide levels such that the higher the browning the higher the acrylamide levels.[37] also found out that the brown colouration of the potato chips increased exponentially with the level of acrylamide and also the cooking temperature.

Effect of Additives/Flavors on Acrylamide Formation

Table 6 represents acrylamide levels in different additives for chips and crisps

Table 6: Acrylamide levels in chips additives

Additive	Peak 1	Peak 2	Peak3	Mean Peak	Conc.(Mg/Kg).
Bhajia	15395	15115	15213	15241	0.8723
Hot and spicy	20166	19943	19889	19999.33	1.1446
Tomato sauce	21080	22410	23873	22454.33	1.2851
Cheese and onion	26863	25149	26138	26050	1.4909
Garlic and pepper	31750	32254	32197	32067.163	1.8353

There was a relatively low amount of acrylamide in mayonnaise additives (0.8723 mg/kg) compared to the other flavours in this study, this could be due to undercooking of the sample or the sample being served before cooking well. While that of salt and vinegar in crisps had low acrylamide levels (17.5350 mg/kg), this could be due to presoaking of the crisp samples that led to reduction of sugar levels that aid in Maillard reaction leading to reduction of acrylamide. The statistical analysis of the data was done and represented in Table 7.

Table 7: Descriptive Statistics

	N	Minimum	Maximum	Mean	Std. Deviation
Peak 1	5	15395.0	31750.0	23050.800	6344.8079
Peak 2	5	15115.0	32254.0	22974.200	6366.2776
Peak 3	5	15213.0	32197.0	23462.000	6411.7558
Grand mean				**23162.33**	

The results show that there was a slight change in acrylamide levels compared to the earlier results. For instance these results were still within the range of chips products, and this could be due to the Maillard reaction that occurs during cooking and therefore, these additives would not reduce the reaction and acrylamide levels. Figure 6 represents the levels of acrylamide in different flavors in crisps.

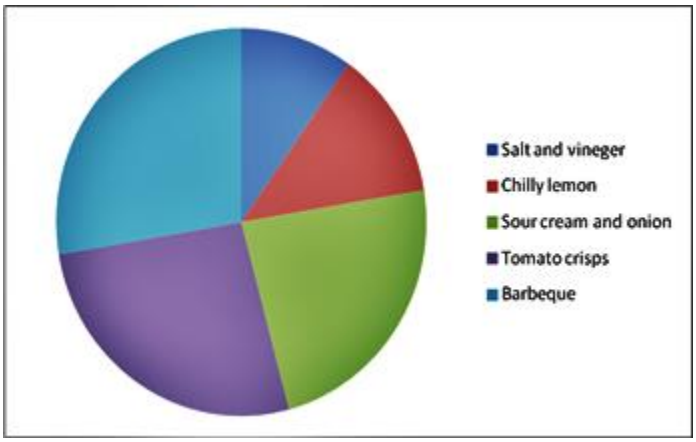

Figure 6

Figure 4 Levels of acrylamide in different flavours in crisps

In potato crisps there was a clear effect in acrylamide levels. Salt and vinegar samples were found with the lowest levels compared to the other samples. This could be due to the presoaking of the samples with vinegar and salt before they are cooked since this reduced the sugars that are involved in the Maillard reaction, and this is also supported by Di [42] who reported that presoaking had an effect on acrylamide levels.

Generally, additives in potato crisps have an impact on the levels of acrylamide since these are added to the raw potatoes during soaking before they are cooked. These have an effect on the reducing sugars and asparagine that reacts as a result of temperature rise to produce acrylamide unlike the additives in potato chips that are added after cooking.

Some studies have identified a number of other ingredients that reduced acrylamide formation in laboratory studies in potato chips or other potato products, including plant extracts, hydrocolloids, vitamins, antioxidants, and spices. The efficacy of these compounds in finished food products is not clear. For new ingredients, it is important to consider such factors as impact on sensorial quality, nutritional quality, regulatory status, and potential formation of byproducts. Table 8 represents statistical analysis of data on different flavours of crisps.

Table 8: Statistical Analysis

	N	Minimum	Maximum	Mean	Std. Deviation
Peak 1	5	56798.0	77190.0	67086.800	8595.9915
Peak 2	5	55475.00	76897.00	66653.6000	9142.73555
Peak 3	5	56129.00	77585.00	67464.2000	8870.27991
Grand mean				156228.5	

Generally, additives in potato crisps have an impact on the levels of acrylamide since these are added to the raw potatoes during soaking before they are cooked. These have an effect on the reducing sugars and asparagine that reacts as a result of temperature rise to produce acrylamide unlike the additives in potato chips that are added after cooking while colour has an effect on the levels of acrylamide based on the results obtained in this research where the acrylamide levels were high in dark or overcooked potato products.

References

Alvarenga, C. J. S., Santos, R. V., Vieira, L. C., Lima, B. A. F., & Mancini, L. H. (2014). Meso-Neoproterozoic isotope stratigraphy on carbonates platforms in the Brasilia Belt of Brazil. *Precambrian Research*, 251, 164–180.

Amrein, T. M., Schönbächler, B., Rohner, F., Lukac, H., Schneider, H., Keiser, A., Escher, F., & Amadò, R. (2004). Potential for acrylamide formation in potatoes: Data from the 2003 harvest. *European Food Technology*, 219, 572–578.

Arribas-Lorenzo, G., Fogliano, V., & Morales, F. J. (2009). Acrylamide formation in a cookie system as influenced by the oil phenol profile and degree of oxidation. *European Food Restoration Technology*, 229, 63–72.

Bastos, A. D. S., Chimimba, C. T., von-Maltitz, E., Kirsten, F., & Belmain, S. (2011). Identification of rodent species that play a role in disease transmission to humans in South Africa. *Proceedings of the South African Society for Veterinary Epidemiology and Preventive Medicine*, 78–83.

Becalski, A., Lau, B. P. Y., Lewis, D., Seaman, S. W., Hayward, S., Sahagian, M., & Leclerc, Y. (2004). Acrylamide in French chips: Influence of free amino acids and sugars. *Journal of Agricultural and Food Chemistry*, 52(12), 3801–3806.

Brathen, E., Kita, A., Knutsen, S. H., & Wicklund, T. (2005). Addition of glycine reduces the content of acrylamide in cereal and potato products. *Journal of Agriculture Food Chemistry*, 53(8), 3259–3264.

Capuano, E., Oliviero, T., Acar, O. C., Gokmen, V., & Fogliano, V. (2010). Lipid oxidation promotes acrylamide formation in fat-rich model systems. *Food Restoration Technology*, 143, 1021–1026.

Codex Alimentarius Commission. (2004). Discussion paper on acrylamide. Joint FAO/WHO food standard programme. *Codex Committee on Food Additives and Contaminants*, Thirty-sixth Session, Rotterdam, The Netherlands, 22–26 March 2004.

Commission recommendation of 10/01/2011 on investigations into the levels of acrylamide in food.

Delgado-Andrade, C., Rufián-Henares, J. A., & Morales, F. J. (2004). Hydroxymethylfurfural in commercial biscuits marketed in Spain. *Journal of Food and Nutrition Research*, 48, 14–19.

Di, R., Kim, J., Martin, M. N., & Leustek, T. (2003). Enhancement of the primary flavor compound methional in potato by increasing the level of soluble methionine. *Journal of Agriculture Food Chemistry*, 51, 5695–5702.

European Food Safety Authority. (2015). Scientific Opinion on acrylamide in food. *EFSA Journal*, 13(6), 4104. https://doi.org/10.2903/j.efsa.2015.4104

Fink, G. R., Nygren, E., & Sarma, K. (2006). Acrylamide in food: A critical review. *Food and Chemical Toxicology*, 44(9), 1282–1289. https://doi.org/10.1016/j.fct.2006.03.013

Food and Drug Administration (FDA). (2003). Detection and quantitation of acrylamide in foods. *European Journal of Nutrition*, 39(2), 150–157.

Gökmen, V., Acar, O. C., Arribas-Lorenzo, G., & Morales, F. J. (2008). Investigating the correlation between acrylamide content and browning ratio of model cookies. *Journal of Food Engineering*, 87, 380–385.

Gökmen, V., Palazoğlu, T. K., & Şenyuva, H. Z. (2006). Relation between the acrylamide formation and time–temperature history of surface and core regions of French fries. *Journal of Food Engineering*, 77(4), 972–976.

Gökmen, V., & Süğüt, I. (2007). A noncontact computer vision based analysis of colour in foods. *Journal of Food Engineering*, 3(5).

Harrison, T. J., & Dake, G. R. (2005). An expeditious, high-yielding construction of the food aroma compounds 6-acetyl-1,2,3,4-tetrahydropyridine and 2-acetyl-1-pyrroline. *The Journal of Organic Chemistry*, 70(26), 10872–10874.

Hwang, C. S., Sukalo, M., Batygin, O., Addor, M. C., Brunner, H., Aytes, A. P., Mayerle, J., Song, H. K., Varshavsky, A., & Zenker, M. (2011). Ubiquitin ligases of the N-end rule pathway: Assessment of mutations in UBR1 that cause the Johanson-Blizzard syndrome. *Journal Research Support*.

Lambert, F., Delmonte, B., Petit, J., Robert, B., Matthias, K., Patrik, R., Hutterli, M. A., Stocker, T. F., Ruth, U., Steffensen, J. P., & Maggi, V. (2008). Dust record from the EPICA Dome C ice core, Antarctica, covering 0 to 800 kyr BP. *PANGAEA*, 1, 23–24.

McGee, H. (2004). *On Food and Cooking: The Science and Lore of the Kitchen*. New York: Scribner. 3, 778–779.

Mendoza, F., & Aguilera, J. M. (2004). Application of image analysis for classification of ripening bananas. *Journal of Food Science*, 69, 471–477.

Mestdagh, F., De Wilde, T., Castelein, P., Németh, O., Van Peteghem, C., & De Meulenaer, B. (2008). Impact of the reducing sugars on the relationship between acrylamide and Maillard browning in French fries. *European Food Research and Technology*, 227(1), 69–76.

Mills, C., Mottram, D. S., Wedzicha, B. L., & Wiley, H. (2009). Acrylamide. In Stadler, R. H., & Lineback, D. R. (Eds.), *Process-induced food toxicants* (pp. 23–50).

Mottram, D. S., Wedzicha, B. L., & Dodson, A. T. (2002). Food chemistry: Acrylamide is formed in the Maillard reaction. *Nature*, 419(6906), 448–449.

Nyabaro, O. M., Wanjau, T., & Kituyi, L. (2019). Effect of temperature on acrylamide in potato (Solanum tuberosum) chips. *International Journal of Innovative Research and Development*, 8. https://doi.org/10.24940/ijird/2019/v8/i3/FEB19001

Nyabaro, O. M., Kituyi, L., Wanjau, T., & Okemwa, E. (2020). Analysis of tuber maturity on levels of acrylamide in selected Solanum tuberosum products (chips and crisps). *International Journal of Scientific and Research Publications*, 10(5).

Obed, M., Nyabaro, O., & Kituyi, L., & Wanjau, T. (2019). Analysis of levels of acrylamide in (Solanum tuberosum) potato chips and crisps. *International Journal of Advanced Research*, 7, 321–327. https://doi.org/10.21474/IJAR01/8330

Ogolla, J. A. (2013). Acrylamide contamination in commercial potato crisps in Kenya: Levels of intake and effects of processing parameters in local cultivars (Master's thesis, University of Nairobi), 19–96.

Pedreschi, F., Kaack, K., Granby, K., & Troncoso, E. (2007). Acrylamide reduction under different pre-treatments in French chips. *Journal of Food Engineering*, 79(4), 1287–1294.

Pedreschi, F., Moyano, P., Kaack, K., & Granby, K. (2005). Colour changes and acrylamide formation in fried potato slices. *Food Research International*, 38(1), 1–9.

Petersen, B. J., & Tran, N. (2005). Exposure to acrylamide: Placing exposure in context. *Advanced Experimental Medical Biology*, 561, 63–76.

Phisut, N., & Jirapon, B. (2013). Characteristics and antioxidant activity of Maillard reaction products derived from chitosan-sugar solution. *International Food Research Journal*, 20(3), 1077–1085.

Rommens, C. M., Yan, H., Swords, K., Richael, C., & Ye, J. (2008). Low-acrylamide French chips and potato chips. *Plant Biotechnology Journal*, 6(8), 843–853.

Şenyuva, H. Z., & Gökmen, V. (2005). Study of acrylamide in coffee using an improved liquid chromatography mass spectrometry method: Investigation of colour changes and acrylamide formation in coffee during roasting. *Food Additives and Contaminants*, 22, 214–220.

Sun, L., Zhang, F., Yong, W., Chen, S., Yang, M. L., Ling, Y., Chu, X., & Lin, J. M. (2010). Potential sources of carcinogenic heterocyclic amines in Chinese mutton shashlik. *Food Chemistry*, 123, 647–652.

Tareke, E., Rydberg, P., Karlsson, P., Eriksson, S., & Törnqvist, M. (2002). Analysis of acrylamide, a carcinogen formed in heated foodstuffs. *Journal of Agricultural and Food Chemistry*, 50(17), 4998–5006.

Taubert, D., Harlfinger, S., Henkes, L., Berkels, R., & Schömig, E. (2004). Influence of processing parameters on acrylamide formation during frying of potatoes. *Journal of Agricultural and Food Chemistry*, 52(9), 2735–2739.

Viklund, G., Mendoza, F., Sjöholm, I., & Skog, K. (2007). An experimental set-up for studying acrylamide formation in potato crisps. *LWT - Food Science and Technology*, 40(6), 1066–1071.

Williams, J. S. E. (2005). Influence of variety and processing conditions on acrylamide levels in fried potato crisps. *Food Chemistry*, 90, 875–881