European Journal of Nutrition & Food Safety

9(3): 192-209, 2019; Article no.EJNFS.2019.032
ISSN: 2347-5641

Reducing Acrylamide Exposure: A Review of the Application of Sulfur-Containing Compounds - A Caribbean Outlook

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Authors' contributions

This work was carried out in collaboration between both authors. Author DAA wrote the manuscript. Author GAB managed proofreading and editing of the document. Both authors read and approved the final manuscript.

Article Information

DOI: 10.9734/EJNFS/2019/v9i330058
Editor(s):
(1) Dr. R. C. Ranveer, Department of Meat, Poultry and Fish,PG Institute of Post – Harvest Management, India.
(2) Dr. Morten Poulsen, Head of research group, Div. of Toxicology and Risk Assessment, National Food Institute, Technical University of Denmark, Denmark.
Reviewers:
(1) K. Srinivasan, Dr. NTR University of Health Sciences, India.
(2) Ravi Bansal, Jiwaji University, India.
(3) César Leyva-Porras, Advanced Materials Research Center, México.
Complete Peer review History: http://www.sdiarticle3.com/review-history/48566

Review Article

Received 19 February 2019
Accepted 29 April 2019
Published 03 May 2019

ABSTRACT

Acrylamide, a known neurotoxin, reproductive toxin, genotoxin, probable carcinogen, hepatotoxin, and immunotoxin, has sparked intense curiosity due to its prominent presence in thermally processed, carbohydrate-rich foods. Acrylamide formation occurs via the Maillard reaction at temperatures ≥100ºC. Thorough investigations on acrylamide mitigation through the application of sulfur-containing compounds to raw materials, and during food processing have been conducted. Although prominent results in acrylamide reduction have been observed, limitations are considered. These limitations involve the social and economic challenges of a population, such as the Caribbean. This study seeks to answer just how effective the application of sulfur-containing compounds is in reducing acrylamide exposure, especially when this applies to a developing region.

Keywords: Acrylamide; potato; wheat; asparagine; sulfur-containing compounds.

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1. INTRODUCTION

Common to all carbohydrate-rich foods thermally processed at high temperatures, is the occurrence of the Maillard reaction [1,2], which is responsible for the formation of alluring flavors, and other compounds (Fig.1 (A)) [3]. A particular product of the Maillard reaction which has acquired keen scientific interest as a result of its toxic nature and threat to human safety, is called acrylamide (ACR) (2-propenamide). ACR can be physically identified as a white crystalline solid, void of odor and color with a melting point of 84.5°C and a boiling point of 136°C. It is formed in the lab by the hydration of acrylonitrile and is soluble in water, methanol, ethanol and acetone [4,5]. In addition, ACR is a monomer of polyacrylamide, which possesses several uses [2,4,6]. During food processing, acrylamide is primarily formed between the amino acid, asparagine, and reducing sugars, glucose or fructose [1,7,8] (Fig.1 (B)) [9,10].

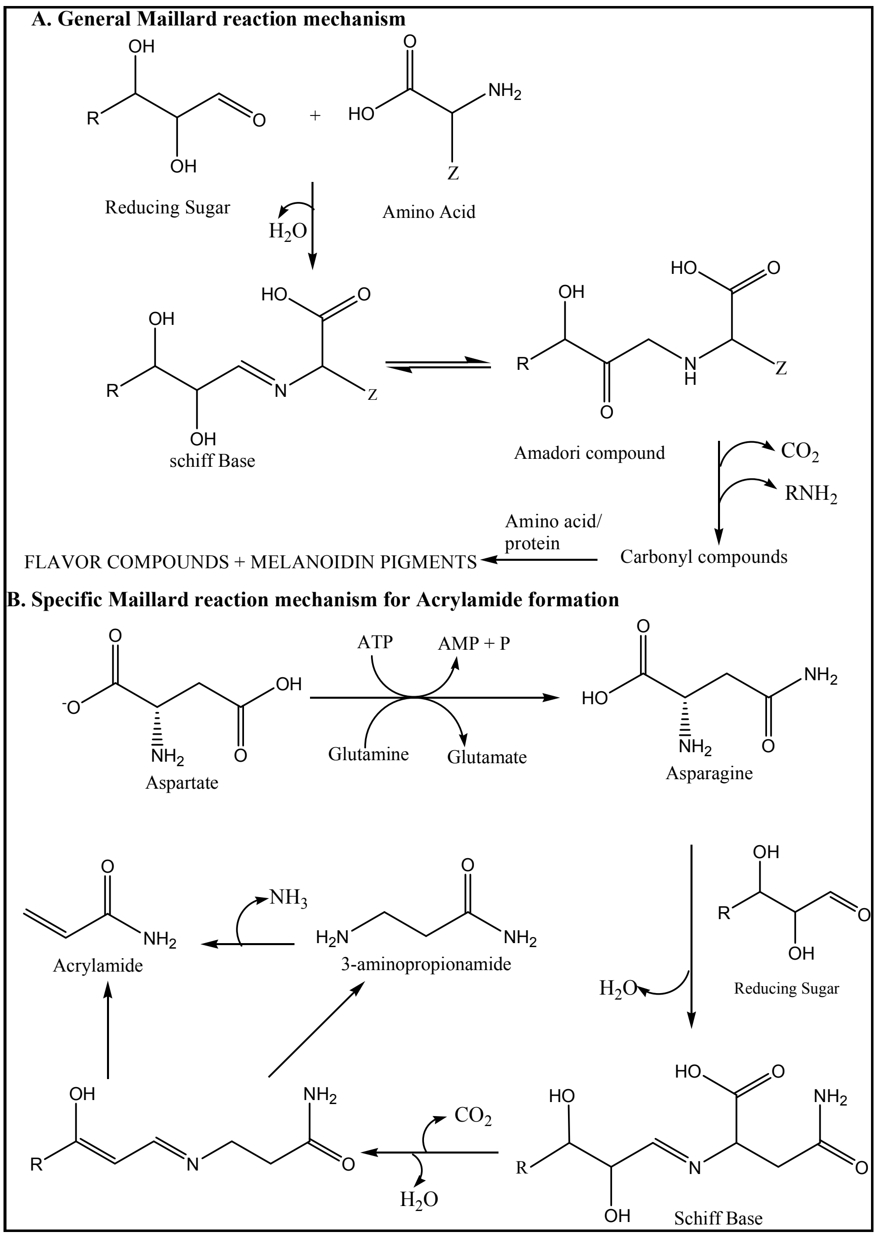

Fig. 1. Reaction mechanism of the Maillard reaction (A) general and (B) specific to the formation of acrylamide

Additional formation of ACR has occurred by the heating of asparagine alone [11], by the reaction between asparagine and other carbonyl sources [12,13] and through the oxidation of acrolein in the presence of asparagine and ammonia [14]. The formation of ACR has been documented at temperatures of ≥120ºC, and in some cases, below 100ºC [15].

ACR has maintained its popularity since its 2002 discovery at high concentrations in carbohydrate-rich foods [4]. Tareke's 2002 publication on the analysis of acrylamide during the heating of different foodstuffs resulted in a series of investigations on the analysis, sources, metabolism, toxicity, and mitigation of ACR [2]. The analysis of raw materials for food preparation has led to the determination of ACR-forming potential. The analysis of a wide range of processed foods has led to the detection of significant ACR levels. In raw materials, ACR occurrence is influenced by soil composition, farming regimes, crop cultivars, and harvest season. In prepared foods, ACR concentrations are influenced by treatment methods, processing conditions, and product formation [5]. Raw materials that have been heavily assessed for the occurrence of ACR are wheat grains and potatoes [16,17]. Large quantities of ACR can be found in processed foods such as: potato fries, cereals, biscuits/crackers, baked goods, and coffee [18-23]. A wide range of Caribbean-based foods including: banana chips, fried and roasted breadfruit (Artocarpus altilis), banana fritters, and fried dumplings (fried bake) showed ACR concentrations ranging from 65-3,640 µg/kg [24].

With the use of food consumption data from the Netherlands and USA, short term daily intakes were estimated ranging from 0.8 µg/kg bw per day to 3.0 µg/kg bw per day in the 95th percentile and extends to 6.0 µg/kg bw per day in the 98th percentile. Long term uncertainty estimates calculated on the basis of food consumption within developed countries indicate a range of 0.3-0.8 µg/kg bw per day [25]. An assessment of the consumption of foods containing high amounts of ACR is lacking for many developing regions such as the Caribbean. A comprehensive compilation of the food supply for the Caribbean region illustrates that refined carbohydrates, sugars, and fats are more prevalent than fruits and vegetables (Table 1) [26].

The categories of foods that are most prevalent for consumption (kcal/day), are known to contain and generate high levels of ACR; such foods include cereals (wheat and other grain products) and starchy root crops. The table indicates a significant imported supply of these products and other refined carbohydrates into the region. Additionally, a close analysis of data recorded from 1990-2010 showed that the share of dietary energy supply originating from cereals and roots and tubers have increased for the Caribbean. The findings suggest that these foods are becoming more preferred (Fig. 2) [27].

Food consumption assessment in the Caribbean could aid in providing information on the types of foods consumed by the population, the safety of the foods consumed, and the correlation between socio-economic and demographic factors, and food consumption [28].

Due to its distinct chemical structure, ACR is able to undergo a number of chemical reactions when it is absorbed by the body. Its reactivity with amino acids, thiols, hydroxyl groups, and DNA centers depends on the Michael addition (Fig. 3) [29].

Eighty-five percent of ingested ACR reacts with key cellular thiols forming mercapturic acid conjugates. These non-toxic metabolites are excreted from the body in urine. ACR undergoes alkylation reactions with thiols of proteins and adduct formation with hemoglobin (HB) pigments. About 15% of ingested ACR is made active by the cytochrome p-450 (CYP2E1) enzyme. The resulting metabolite is called Glycidamide (GA). GA may undergo: hydrolysis, conjugation with light, conjugation with HB forming GA-HB adducts, and may interact with DNA causing genetic mutations [30,31].

Former studies have correlated ACR's and GA's exposure to the following: neurotoxic effects on humans and rodents [32-34], carcinogenic effects on rodents [2,30,35,36], reproductive effects on rodents [37,38], and genotoxic effects on rodents and cells of humans [39-41]. Current studies have indicated ACR to be immunotoxic [42,43] and hepatotoxic [44,45]. Amidst the current progress made on the study of ACR toxicity [46-48], it is still classified as a group 2A probable carcinogen by the International Agency for Research on Cancer (IARC) due to insufficient epidemiological studies relating the induced carcinogenicity of ACR to human populations. However, research is presently ongoing to clarify the potential genotoxicity of ACR, and the mechanisms by which ACR may contribute to induction of carcinogenicity in rodents and humans [49-51].

Table 1. Food Balance sheet for the Caribbean region (2013)

Caribbean + (Total) – 2013													**Food Balance Sheet**			
Item	**[a]Pop.**	**Domestic Supply**					**Domestic Utilization**						**Per Capita Supply**			
		[b]Prod.	**[c]Imp.**	**[d]Stock Var.**	**[e]Exp.**	**Total**	**Food**	**[f]Proc.**	**Feed**	**Seed**	**Losses**	**[g]Oth. Use**	**Total**		**[h]Prot.**	**Fat**
	(1000 persons)	**(1000 tonnes)**											**Kg/Yr**	**KCal/Day**	**g/Day**	
Population	37387															
Grand Total														2738	68.33	71.29
Vegetal Products														2352	42.56	45.17
Animal Products														386	25.76	26.11
Cereals - Excluding Beer		2047	5336	-99	97	7188	4047	172	2577	48	283	61	108.25	968	22.65	4.41
Wheat and products			2070	-22	73	1975	1493	0	368		84	31	39.93	304	8.54	1.39
Rice (Milled Equivalent)		1113	1001	9	20	2102	1860	42	99	22	70	8	49.74	492	9.67	1.07
Barley and products			135	-14	0	121	3	113	1		0	4	0.09	1	0.02	0
Maize and products		816	2104	-68	2	2850	634	16	2036	23	123	18	16.95	158	4.09	1.83
Rye and products			0	0	0	0	0		0			0	0.01	0	0	0
Starchy Roots		3795	131	0	78	3848	2949	0	426	14	437	72	78.88	199	2.38	0.44
Cassava and products		1133	3	0	0	1135	744	0	237	0	125	30	19.89	53	0.28	0.1
Potatoes and products		233	119	0	2	350	308		2	10	20	10	8.24	14	0.32	0.03
Sweet potatoes		1100	4	0	12	1092	871		41	0	151	29	23.3	60	0.56	0.15
Yams		987	0	0	8	979	768		107	1	101	1	20.54	53	0.94	0.11
Sugar Crops		23734	0	0	0	23734	45	21489	2080		120		1.22	1	0	0.01
Sugar (Raw Equivalent)		2227	355	76	1257	1401	1358	0	0		17	26	36.33	356		
Sweeteners, Other		0	44	1	8	37	29					8	0.78	5	0	
Oil crops		792	485	22	96	1204	402	296	309	1	19	176	10.76	58	2.89	4.62
Coconuts - Including Copra		748	6	-2	94	658	298	166			17	176	7.98	28	0.37	2.67
Vegetable Oils		107	544	-11	7	633	430	0				203	11.5	279	0.02	31.52
Soybean Oil		24	293	-12	4	301	227					75	6.07	147	0	16.65
Sunflower seed Oil			17	1		17	17					0	0.45	11		1.24
Coconut Oil		22	5	0	1	26	25					1	0.66	16	0	1.83
Oil crops Oil, Other		4	50	0	2	52	30	0				22	0.8	19	0.01	2.15
Vegetables		3583	223	0	78	3728	2778		595		350	5	74.29	50	2.12	0.43
Tomatoes and products		979	60		7	1032	934				97	1	24.98	15	0.6	0.19
Onions		187	30	0	0	217	196				21	0	5.24	6	0.19	0.03
Vegetables, Other		2417	133	0	71	2479	1648		595		232	4	44.07	30	1.34	0.21

Caribbean + (Total) – 2013																**Food Balance Sheet**
Item	**[a]Pop.**	**Domestic Supply**					**Domestic Utilization**						**Per Capita Supply**			
		[b]Prod.	**[c]Imp.**	**[d]Stock Var.**	**[e]Exp.**	**Total**	**Food**	**[f]Proc.**	**Feed**	**Seed**	**Losses**	**[g]Oth. Use**	**Total**		**[h]Prot.**	**Fat**
	(1000 persons)						**(1000 tonnes)**						**Kg/Yr**	**KCal/Day**	**g/Day**	
Fruits - Excluding Wine		6503	379	6	413	6475	5628	18	150		640	42	150.53	189	2	2.13
Bananas		1511	26	0	238	1299	1066		37		190	6	28.51	43	0.54	0.19
Plantains		1416	6	0	22	1400	1210		79		109	1	32.37	67	0.54	0.18
Pineapples and products		596	12	0	4	604	545				59	0	14.59	13	0.08	0.04
Fruits, Other		2274	132	-1	75	2329	2063		34		230	5	55.19	51	0.63	1.62
Stimulants		133	34	-15	66	86	83	0			1	1	2.23	6	0.4	0.36
Spices		23	11	0	3	32	32					0	0.84	8	0.25	0.33
Alcoholic Beverages		1227	405	3	311	1324	1104					223	29.54	73	0.23	
Meat		1089	651	0	14	1726	1668	0	1		0	57	44.61	202	14.66	15.36
Pulses		448	193	4	1	644	596		0	19	23	6	15.95	148	9.3	0.72

[a]Pop (population); [b]Prod (product.); [c]Imp (import); [d]Stock var (stock variable); [e]Exp (export); [f]Proc (process); [g]Oth.use (other use); [h]Prot (protein)

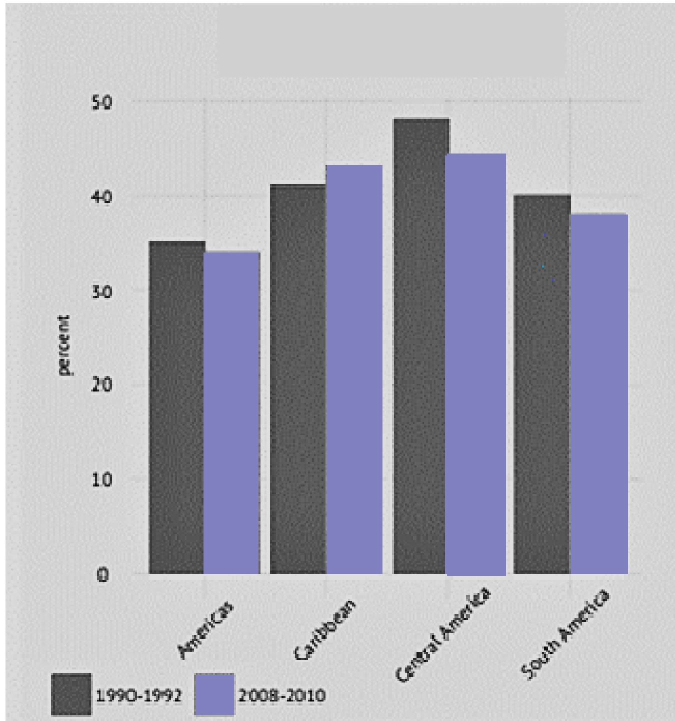

Fig. 2. Dietary energy supply originating from cereals, roots and tubers from 1990-2010

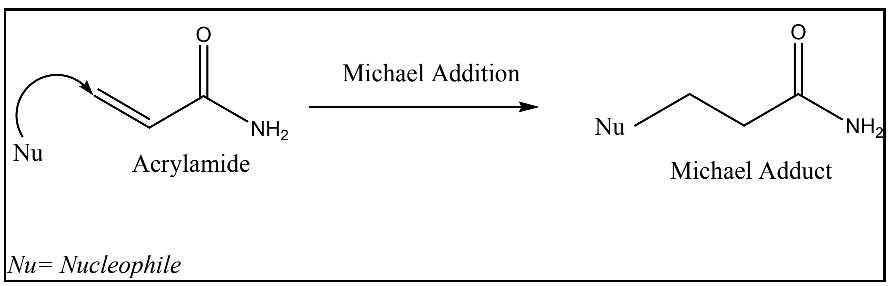

Fig. 3. Michael-Type addition involving Acrylamide

Given the span of ACR's toxicity and presence in relatively high quantities of various home-cooked and commercial products, its existence is alarming. Literature has presented thorough investigation on the utilization of amino acids and antioxidants on the mitigation of ACR levels and toxicity. Among these groups of additives, are compounds which contain a sulfur atom. A broad spectrum of sulfur-containing compounds involved in the mitigation of ACR include: thiols, thiolsulfinates, thioethers, allyl sulfides, sulfates, and in some instances, elemental sulfur. These compounds are characterized as: strong reducing agents, strong fatty acid hydroperoxide detoxifiers, excellent anti-browning agents, cellular detoxification activators, and oxygen radical scavengers. Although such characteristics render these compounds effective agents in the mitigation of ACR levels and toxicity, there are limitations to their utilization.

This review examines the outcomes of studies published between 2007-2018 and in certain instances, previous literatures. Emphasis is placed on the application of sulfur-containing compounds on the mitigation of ACR levels in raw materials and food processing, in conjunction with associated limitations. These limitations include the socio-economic challenges faced by the Caribbean region. Hopefully light is shed on avenues for further research in this area of study.

2. THE EFFECT OF SULFUR-CONTAINING COMPOUNDS ON ACRYLAMIDE-FORMING POTENTIAL IN RAW MATERIALS

In a report released in June 2015, the European Food Safety Authority explained the risk assessment of ACR in food. It was mentioned that fried potato products, coffee, biscuits, crackers, crisp bread, and soft bread contributed the most significantly to ACR exposure in humans. Apart from the storage and processing conditions of these foods, the raw materials from which they are made greatly impact ACR formation [52]. Common raw ingredients of these foods include wheat grains and potato tubers. Research has been conducted on the occurrence of ACR precursors (mainly glucose and asparagine) in these raw foods, and how their levels can be reduced.

2.1 Wheat

In cereal grains, asparagine makes up about 5% of the total free amino acids [53]. Recent studies have indicated asparagine levels as the limiting factor for ACR production in wheat grains [54]. In wheat, asparagine functions as a transport for nitrogen, a compound for storage, and serves as building blocks for the formation of various wheat proteins [54]. A specific mode of action involved in the endeavors of mitigating asparagine concentrations in wheat, is by understanding the correlation between sulfur, nitrogen, and asparagine content with ACR formation [55].

Elmore et al. [56] reported that sulfur-deficient wheat-flour contained greater levels of asparagine, producing ACR concentrations six times in excess of the amounts detected in sulfur-sufficient wheat-flour. Headspace analysis of sulfur-deprived heated flour showed: Strecker aldehydes, products of aldol condensations and alkylpyrazines. On the other hand, sulfur-sufficient flour demonstrated products of sugar degradations: thiophenes, and pyrroles. The products detected in sulfur-deprived flour were reactive intermediates of the Maillard reaction. In the final stage, these intermediates were converted into various colored pigments and flavor compounds and simultaneously, acrylamide was produced. In contrast, headspace analysis of the sulfur-sufficient flour suggested a lack therein of the Maillard reaction by the degradation of sugars, and the absence of reactive carbonyl intermediates. The authors further stated that apart from a correlation established between sulfur concentrations of the heated flour and the products indicated in the headspace, genetic factors also played an influential role.

Furthermore, Curtis et al. [57] showed the assessment of different strains of wheat grains, having differing concentrations of free asparagine. These strains were analyzed under sulfur-sufficient and sulfur-deficient conditions. All strains displayed asparagine accumulation under sulfur-deprived conditions; some to a greater extent than others. All strains showed asparagine mitigation under sulfur-sufficient conditions. Additionally, after milling, baking, and drying, fractions of the grain samples were analyzed for ACR. A quadratic correlation of R^2=0.9945 was obtained between the two variables, asparagine concentration and ACR levels. ACR levels in relation to sulfur content in the wheat grains showed a negative correlation. These results suggest that an inverse relationship exists between the presence of sulfur and ACR levels in wheat grains. It was also observed that genetic factors do not affect the ACR mitigating potential presented by sulfur-sufficient conditions, but rather may implicate the potential of the plant to accumulate ACR precursors, namely asparagine.

The success of sulfur-sufficient conditions in mitigating asparagine can be explained by the dependence of asparagine concentrations in plants on the sulfur to nitrogen ratio, as opposed to the nitrogen level only. Sulfur-deficient conditions resulted in a reduction of prolamins from 51.0% to 27.0% in plants and consequently, an increase in aspartate and asparagine from 5.7% and 5.3% to 19.2% and 18.5%, respectively [58]. Prolamins are a group of storage proteins in plants comprised of cysteine and methionine, and can be found mainly in the seeds of cereal grains. It has been suggested that the accumulation of asparagine results from an attempt to provide alternative nitrogen storage when prolamin production is marred [58]. Sulfur fertilizers can be added to soil or surface of plants including wheat, in the form of elemental sulfur, ferrous sulfate, or aluminum sulfate to aid in counteracting the accumulation of asparagine [59].

A study conducted on rye grains, a grain closely related to Triticum (Wheat genus), showed that free asparagine increased by 70% in sulfur-deficient conditions. However, nitrogen application resulted in a three-fold increase in free asparagine levels. Additionally, there was a statistically significant association observed between nitrogen and sulfur especially under deprived conditions of both elements [60]. These results are similar to outcomes obtained from former investigations conducted on barley [58] and wheat grains [61]. These studies confirm that the effect of sulfur application on asparagine reduction is transparent among various types of grains, and is consequently a better application towards ACR mitigation than nitrogen fertilizers.

2.2 Potatoes

Although potatoes are usually grown in the absence of sulfur, its positive effects towards crop composition and consequently, food safety cannot be left un-noticed. When different varieties of potatoes were grown in pots, sulfur application resulted in a decrease in free asparagine and reducing sugars [62]. However, in field trials, sulfur displayed no significant effects on asparagine levels [63]. The discrepancies between the relationship of sulfur application and asparagine levels observed, may suggest the presence of other factors that influence asparagine accumulation. More research is needed to establish the relationship between sulfur application and asparagine levels, especially in relation to environmental conditions. A similar inverse relationship between sulfur application and reducing sugars in the pot samples, was observed in the field trials [63]. This suggests that sulfur application was effective in mitigating ACR-forming potential in potatoes by reducing glucose levels. Although beneficial towards improving food safety, glucose reduction may affect the palatability of potatoes [64] as potato flavor is dependent on glucose levels.

Nitrogen application, however, is beneficial to potato cultivation as it promotes vegetative growth, increases size and yield, and canopy senesce. Despite its benefits, nitrogen application poses a threat to safety by increasing the ACR-forming potential of various types and varieties of potatoes through asparagine accumulation. The effect on glucose levels was not clearly defined across the various types and varieties [63]. It was concluded that nitrogen application on potatoes can influence ACR formation, depending on the types and varieties [63].

The significance of the ratio of asparagine to total free amino acids in evaluating the formation of ACR in potatoes was emphasized in the Acrylamide Toolbox (2017) [65]. It was stated that the direct relationship between reducing sugar levels and ACR levels is inconsistent. The relative ACR exposure in potato varieties can be indicated more precisely by consideration of the asparagine to free amino acid ratio. The application of both sulfur and nitrogen fertilizers can impose conflicting effects on the asparagine to free amino acids ratio based on potato variety, as such, no optimum fertilizer ratio has been established [65]. Current leads being explored in the area of potato and potato tubers include the development of varieties with lower asparagine content, and the effects of storage and fertilizer regimes on asparagine levels. Further investigation on the mitigating potential of sulfur-based fertilizers on asparagine levels in potatoes is needed in aiding with the precise assessment of its effectiveness toward mitigating ACR formation.

The investigation of sulfur and nitrogen fertilizers and their effects towards asparagine levels should be extended to other crops besides wheat or cereal grains, and potatoes. Demands for gluten-free (lacking prolamin) products have elevated in recent times [66-68]. Competitive gluten-free flour on the market include: cassava, coconut, and breadfruit. Further insight could be obtained in terms of utilizing sulfur compounds in the cultivation of these raw materials to reduce the ACR-forming potential.

2.3 Limitations

2.3.1 Environmental conditions and genetic factors

The quest of sulfur-containing compounds to mitigate ACR-forming potential is limited by the natural accumulation of asparagine by plants when they are subjected to stressful conditions. These conditions include: drought and salt stress, the effect of soil contaminated with heavy or toxic metals, and the attack of plants by pathogens [69-72]. Not all stresses can be avoided or identified by farmers until it may be too late. Also, a farmer may induce plant stress indirectly by attempting to prevent it. Muttucumaru et al. [73] showed that irrigation may promote ACR formation in potatoes. It was suggested that farmers irrigate only if necessary as water availability can influence the amino acid and glucose concentrations in potato tubers.

The implications of environmental factors on asparagine accumulation can be further understood by considering a plant's genetic composition. Asparagine synthetase was investigated thoroughly in wheat, and four asparagine synthetase genes were observed: TaASN1, TaASN2, TaASN3, and TaASN4 [74-76]. The most influential gene in wheat is TaASN2. Nevertheless, it is the TaASN1 gene that is expressed in response to saline, drought, and sulfur and nitrogen deficiencies [60,77]. In potato tubers, two genes were identified, StASN1 (found in high concentrations in the tuber), and StASN2 (found throughout the entire plant) [78]. So far, a detailed model network has been constructed on asparagine metabolism, illustrating established relationships between stress response and many genes vital for asparagine metabolism in wheat plants [79]. While this model network aids in confirming the accuracy of existing relationships, there is room for the exploration of novel relationships. Further applications may involve its use in the genetic engineering of cultivars of wheat grains. Asparagine synthesizing genes can perhaps be modified to resist stimulation from certain environmental conditions. Such a model network is lacking for potato crops.

2.3.2 Crop yield

The use of sulfur fertilizers is common in the cultivation of cereal grains, particularly wheat, as it promotes grain and protein yield [80]. On the other hand, sulfur-based applications are limited in potato tubers. Although they are able to mitigate ACR-forming potential, they add no established benefit to crop yield [63,81]. Farmers may not view the purchase of sulfur fertilizers as value for money. Creating educational programs that will enable farmers to understand the correlation between acrylamide-forming potential and the use of certain fertilizers, or introducing incentives for the purchase of sulfur fertilizers may aid in generating a positive reception towards their use. Moreover, the investigation of an appropriate sulfur to nitrogen ratio fertilizer application that can result in a considerable reduction of asparagine concentrations and efficient crop yield, may create a win-win outcome for both the public and the farmers. However, a clear and precise relationship between sulfur and asparagine levels in potato tubers first needs to be established.

2.3.3 Potato palatability

In a chapter written by Jansky [64], it was indicated that sucrose and reducing sugars were the key determinants in potato flavor. Additionally, flavors obtained from baked potato and potato chips were considered to be resultant to the occurrence of pyrazines which are products of the Maillard reaction [1,82]. Studies have shown that the application of sulfur during potato cultivation is advantageous in reducing ACR formation by lowering glucose [63] and asparagine levels [62], but may negatively affect the palatability of potatoes. A reduction in glucose levels means a reduction in the occurrence of the Maillard reaction. This leads to a reduction in the generation of flavor compounds and hence, the palatability of potato and potato products.

2.3.4 Agricultural implications in the Caribbean region

Wheat grains are not cultivated in the Caribbean region due to the lack of an environment conducive to the biology of the crop; one which does not favor heavy rainfall and tropical conditions [83]. Nevertheless, wheat makes its way to the Caribbean mostly in the form of cereal. The diet of developing regions consists of 27% cereal and 3% protein requirements. Additionally, many people prefer wheat flour as opposed to traditional flour from staple crops such as cassava, simply because of the certain status attached to it [84]. Wheat grain based products such as: bread and wheat flour which are the most prevalent in the Caribbean, are also the products which contribute the most significantly to ACR exposure [19,23,52]. Although research concerning the application of sulfur in the cultivation of wheat grains has been increasing, there is no guarantee that the wheat products imported into the Caribbean region were produced from wheat grains cultivated under sulfur-sufficient conditions.

Besides the high influx of processed wheat into the Caribbean, traditional products of cassava such as: cassava flour and farine (dried, unprocessed cassava) are still widespread due to affordability [85]. Other popular gluten free products on the market include: breadfruit, banana, and coconut flour [66,86,87]. There is no existing research on the levels of ACR precursors in the raw materials of these products nor has the effects of sulfur or nitrogen application on the levels of ACR precursors in these raw materials been investigated. It is imperative that raw materials cultivated in the Caribbean be assessed for the levels of ACR precursors. This will encourage more stringent measures on farming regimes and the type of food products generated within the Caribbean. Even if a suitable farming regime was obtained to reduce levels of ACR precursors in raw materials, it would be challenging to implement such a policy across the region. Currently, there is a lack of agricultural policies governing agricultural practices or production for a sustainable development. Instead, a grave dependence of the economy on agriculture has resulted in a short-term vision of economic growth rather than a long term plan for sustainability. The lack of agricultural policies has resulted in a limited expansion of agricultural production for many Caribbean countries in terms of achieving a standard by which the quality of foods produced can be assessed. Recently, the Caribbean Single Market Economy was established to benefit the Caribbean functioning as a single unit to operate in trade with the rest of the world. However, there is still a long way to go [88].

Potato is a staple crop grown widely across the Caribbean region in contrast to wheat grain. Although it grows best at around 20ºC, it can be cultivated under a wide range of climate conditions. To combat the effects of periodic droughts, and the attack of pests and diseases, fertilizers are employed during cultivation. Caribbean farmers mainly employ the use of nitrogen fertilizers [89]. According to World Fertilizer Trend and Outlook 2018, the Caribbean and Latin America were listed as the third highest consumers of nitrogen fertilizers worldwide. The high usage of nitrogen fertilizers in the Caribbean may be due to the ease of accessibility, cost, and production. In the region, nitrogen fertilizers are most prevalent as ammonium salts [89] which are produced in Trinidad and Tobago [90]. Trinidad and Tobago has an abundant source of natural gas, which is essential for ammonia production [89]. Sulfur fertilizers on the other hand, are produced in the USA and may not be readily available to Caribbean farmers. Sourcing from within the Caribbean is cheaper than sourcing internationally [91].

Agriculture accounts for 16% of employment within the Caribbean, specifically: 30% in Guyana, 25% in Dominica, 20% in St. Lucia, and 18% in Jamaica. Any decline in agriculture affects the economic and social stability especially in rural areas, as most of the agriculture occurs in rural areas. If crop yields were to decline, the poverty rate of various Caribbean islands would increase [88,92]. For this reason, nitrogen fertilizers are heavily relied upon for the associated benefits toward tuber size and yield. Sulfur application becomes even less appealing, having no established benefit to crop yield. ACR precursors could therefore be more prevalent in potato tubers cultivated in the Caribbean region.

3. THE EFFECTS OF SULFUR-CONTAINING COMPOUNDS ON ACRYLAMIDE MITIGATION IN FOOD PROCESSING

The application of sulfur-containing compounds on the mitigation of ACR levels is assessed during food processing under food preparation conditions and/or in various food matrices. Ismial et al. [93] assessed the effects of different soaking treatments on the ACR levels in potato slices, in comparison to the maximum permissible level set by the World Health Organization (WHO). The slices were soaked in tap water for 15 minutes, distilled water for 60 minutes, acidic solutions, salt solutions, amino acid solutions, and phenolic solutions for 60 minutes at room temperature. Frying at 190±5ºC was followed by immediate analysis of ACR levels. L-cysteine, and L-glycine (0.05 M) were among the most effective in reducing ACR concentrations, showing significant reductions of 84.74% and 84.94%, respectively.

Similar solutions were used in an assessment of the blanching treatment on potato slices. The treatment was carried out at 65ºC, for 5 minutes. $MgCl_2$ (0.1 M) and L-cysteine (0.05 M) were the most effective in ameliorating ACR levels at 97.97% and 97.17%, respectively [93]. Blanching and soaking treatments were effective in decreasing ACR levels in potato chips by 60% due to the leaching of the glucose content [94,95]. Cysteine was effective in the enhancement of ACR reduction by preventing the Maillard reaction through the replacement of asparagine, [96] and by reacting with ACR in a Michael-type addition [97]. On the contrary, other additives like $MgCl_2$ dries out glucose, preventing its participation in the Maillard reaction [98]. The discrepancies in the mitigating ability of cysteine between the soaking and blanching treatments may be attributed to the higher temperature conditions administered during blanching. Blanching involves scalding the raw material through boiling, whereas, soaking occurs at room temperature. Additionally, Ismial et al. [93]

illustrated the effects of soaking and blanching on the quality, taste, texture, appearance, odor, and color of the potato slices. In both treatments, cysteine (0.1 M) showed a relatively poor rating, whereas, (0.05 M) showed a relatively fair rating. The other additives as listed above, were rated as relatively good.

Casado et al. [99] analyzed the ACR mitigating potential of various additives in ripened olives, using an alkali-treated olive juice heated at 120ºC for 30 minutes. Among the various salts, amino acids, and antioxidants used, L-cysteine, L-arginine, and sodium bisulfite showed the strongest mitigating ability. The taste and ACR reduction of black, ripened olives were assessed by additional compounds of N-Acetyl-L-cysteine, reduced glutathione, methionine, tea, oregano, rosemary, and garlic. The thiols: cysteine, N-acetyl-L-cysteine, and glutathione were effective in ACR reduction, but affected the savor. On the other hand, sodium bisulfite was effective and the savor unaffected. Arginine, along with garlic showed results that were promising. Sodium bisulfite could be an excellent additive to food as it showed effective acrylamide mitigating potential, without any effect on food quality. However, the presence of sulfites in food is questionable, as it has been linked to certain health issues [100,101]. Further research is needed in the recognition of suitable additives that are effective in reducing ACR levels without affecting the health of consumers or food quality. Other investigations illustrated the effects of absence and presence of garlic in a low moisture system containing 1.2 mmol of both glucose and asparagine. A 0.05g (mass fraction) of garlic was added to the system and heated at 200ºC for six minutes. A generation of 674.0 nmol of ACR occurred in the absence of garlic. However, this amount was quickly reduced by 43% in the presence of garlic [102]. Garlic is known to contain biologically active sulfur-containing compounds, such as allicin [103].

The mitigation effects of 10 amino acids including cysteine and methionine, were investigated on 10 µmol ACR in a reaction model system. Investigations occurred after heating at 160ºC for 15 minutes. At natural pH and an adjusted pH of 7, cysteine displayed mitigating effects of 94.4% and 94.8%, respectively. Asparagine showed the lowest mitigating effect at natural pH, and glutamine, at pH 7. Methionine, although not as successful as cysteine, showed mitigating effects from a value of: 10.28±0.23 µmol to 7.92±0.35 µmol ACR (natural pH), and from 10.11±0.21 µmol to 7.24±0.06 µmol ACR (pH 7) [104]. In another study observing the ACR mitigation during a heat treatment of canned coffee, cysteine and dithiothreitol gave positive results. Cysteine showed 95% reduction with heat treatment at 121ºC for 6 minutes. However, cysteine in combination with dithiothreitol was unsuccessful [105]. The combination of both reactants may have resulted in the oxidation of the sulfhydryl groups of cysteine and dithiothreitol forming disulfide bonds, thus lowering the availability of sulfur atoms to participate in Michael-type reactions with ACR.

3.1 Limitations

3.1.1 Unpleasant food taste

The employment of cysteine to foods in a quest to mitigate ACR is limited by its negative impact on food taste, aroma, and texture [93,99,106,107]. However, methionine addition as a food flavorant seems promising [108,109]. Studies conducted on methionine's impact towards food quality are still inadequate, and so, further exploration is needed on its use as an ingredient in food preparation; this may result in new approaches toward ACR mitigation.

3.1.2 Reactivity

Studies have shown that the ACR mitigating potential of some sulfur compounds are greater than others. A reason for this outcome may be due to their differences in chemical reactivity. Variation in chemical reactivity has been correlated to size and intramolecular forces of attraction. In a study conducted by Bent et al. [110], the chemical reactivity of thiols was compared. Measurements were conducted by investigation of the loss of the sulfhydryl group, thus forming the thiolate anion in the presence of ACR over time. Using a tris/HCl buffer system, cysteine displayed a greater chemical reactivity than glutathione (GSH) and captopril, respectively, over the pH range of 7.10-9.10. The results suggested that cysteine's performance was due to its small size and lower thiol dissociation constant. However, GSH is larger than captopril but displayed faster chemical reactivity. Therefore, size could not explain the chemical reactivity between the two. It was observed that significant intramolecular hydrogen bonding was the determinant of the stabilization of the transition state species, ACR-

$SR^{\neq}$, of the two thiols. The magnitude of intramolecular hydrogen bonding in GSH was greater than was observed in captopril which increased the nucleophilicity of the S^- of GSH and hence, its reaction with ACR.

Methionine's lower ACR mitigating potential in comparison to cysteine, can be correlated to its tendency to behave as an ACR precursor, producing substantial amounts of acrolein in the presence of appropriate reagents [14,108]. Further studies are needed to investigate the chemical reactivity of cysteine and methionine with ACR. This may eventually shed light on the prospect of using methionine as an ingredient during food processing.

3.1.3 Cultural methods of food preparation and food importation in the Caribbean

In the Caribbean, the most prevalent means by which sulfur application can mitigate ACR exposure may be in the preparation of foods. Multiple Caribbean cuisines are prepared with seasonings of vegetables and spices. These culinary influences originated from the Caribbean's history of ethnic and cultural diversity from: Africa, France, Spain, India, the Netherlands, and the indigenous Amerindians. The most common seasonings used which contain sulfur compounds include: garlic, onions, chives, curry, black pepper, and tomatoes. These are added to both home-cooked and commercial products before processing.

However, there are several challenges to consider. The Caribbean's cultural and ethnic diversity not only influences the ingredients in food preparation, but the methods by which foods are prepared. Many common dishes are prepared by baking, frying or roasting. These dishes include: roasted and fried bake, roti, banana fritters, roast corn, roasted and fried breadfruit, cassava bread, baked cassava, and baked potato [111,112]. These methods of cooking occur at high temperatures. Acrylamide formation is increased in response to an increase in temperature within the range of 120ºC-180ºC [113]. This is highlighted in Bent's work [114] where it was seen that breadfruit roasted and then fried, and raw breadfruit chipped then fried, produced more acrylamide as the temperature rose from 150ºC-200ºC.

Additionally, the Caribbean's market has shifted from a local production of fruits, vegetables, root crops, and tubers to a massive importation of processed foods, wheat and maize products, meat, and dairy. Food importation in several Caribbean countries has surmounted a total of US $4 billion, and this is expected to increase by the year 2020 to values of US $8-10 billion [115]. This may result in an increase in ACR exposure. The use of seasonings which contain a wide spectrum of sulfur compounds that may be effective in mitigating ACR levels during food processing, encounters two counteracting effects: a) popular or cultural methods of cooking which subjects food to high temperatures, a condition favorable for ACR formation; b) poor food security, which results in a high influx of imported processed foods, maize and wheat products which adds to ACR exposure in the Caribbean. Considering the lack of awareness in the region on ACR exposure, a threat to safety may subsist, that may be too severe to fathom.

4. CONCLUSION

This review examined the application and limitations of sulfur-containing compounds on the mitigation of ACR exposure in raw materials and food processing. It is evident that a wide range of sulfur-containing compounds are successful in the reduction of ACR, but the success is accompanied by few challenges worthy of consideration. In raw materials, the success of sulfur-containing compounds in mitigating ACR formation is abased by: the plants' genetic makeup to accumulate asparagine in response to stressful conditions, the palatability of the final product, and the non-beneficial use of sulfur fertilizers to potato yield. In food processing, the success of sulfur-containing compounds is restricted by the negative impacts of cysteine concentrations towards food quality, and factors that affect the reactivity of sulfur-containing compounds with ACR. In addition, a Caribbean outlook was included, highlighting the social and economic factors of a developing region in the context of applying sulfur-containing compounds towards ACR mitigation. This review seeks to contribute towards the global improvement in ACR reduction, thus improving food safety and security.

ACKNOWLEDGEMENTS

Funding for this review was provided by the Department of Chemistry, and School of Graduate Studies & Research, The University of the West Indies, St. Augustine Campus, Trinidad and Tobago.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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