Anti-nutritional Factors in Food and Effect of Processing Authors: Shalini Gaur Rudra, Neha Saini, Kanika Walia

It is well known that plants generally contain anti-nutrients acquired from fertilizer and pesticides and several naturally-occurring chemicals. Some of these chemicals are known as "secondary metabolites" and they have been shown to be highly biologically active. These include saponins, tannins, flavonoids, alkaloids, trypsin (protease) inhibitors, oxalates, phytates, haemagluttinins (lectins), cyanogenic glycosides, cardiac glycosides, coumarins and gossypol. The list is inexhaustible.

Most of these secondary metabolites elicit very harmful biological responses, while some are widely applied in nutrition and as pharmacologically-active agents. The anti-nutritional factors (ANFs) may be defined as those substances generated in natural food stuffs by the normal metabolism of species and by different mechanisms (e.g. inactivation of some nutrients, diminution of the digestive process, or metabolic utilization of feed) which exert effects contrary to optimum nutrition. Besides, anti-nutritional factors may occur endogenously or may be formed during heat/ alkaline processing of proteins. Examples of important anti-nutritional factors formed during the heat/alkaline treatments of protein products include Maillard reaction products (MRP), oxidized forms of sulphur amino acids, D-amino acids and lysine-alanine (LAL, an unnatural nephrotoxic amino acid derivative).

Being an ANF is not an intrinsic characteristic of a compound but depends upon the digestive process of the ingesting animal. For example, trypsin inhibitors, which are ANFs for monogastric animals, do not exert adverse effects in ruminants because they are degraded in the rumen. Several considerations justify the continued surveillance, knowledge and future research on anti-nutritional factors/ toxic substances naturally present in plants used as foods and feedstuffs and ways of reducing them to safe level of consumption.

- The possibility that newly developed or exotic foods may contain natural toxicants must be taken into account in evaluating their usefulness. Also, introduction of new plant varieties into our diets may expose humans and animals to new toxic factors with unsuspected biological effects.
- 2. Improper processing of plant food like beans and pulses may expose humans and animals to high concentrations of these toxic factors. An example is the production of soymilk and its use as an alternative to cow's milk in infant formula. It is reported that soymilk, if

not properly processed and supplemented with iodine, causes goitre in infants.

- 3. These anti-nutritional factors are increasingly recognized to affect the overall nutritional value of foods and feeds.
- **4.** The plant breeder, in an attempt to develop higher-yielding or disease-resistant crop varieties must, at the same time, be alert to the possible production of undesirable components.
- **5.** Public health authorities and other food regulatory bodies need to be informed about the possible dangers related to the widespread, longstanding practices previously regarded as safe. For example, cassava and some legumes have been consumed in the tropics for a long time without apparent consideration for their high cyanide contents.

CLASSIFICATION OF INHERENT ANTI-NUTRITIONAL FACTORS

The anti-nutritional factors in plants may be classified on the basis of their chemical structure, the specific actions they bring about or their biosynthetic origin. Although this classification does not encompass all the known groups of anti-nutritional factors, it does present the list of those frequently found in human foods and animal feedstuffs. They are:

- (1) Proteins (lectins and protease inhibitors): sensitive to normal processing temperatures
- (2) Polyphenolic compounds (mainly condensed tannins), non-protein amino acids and galactomannan gums: stable or resistant to these temperatures
- (3) Enzyme inhibitors (trypsin, chymotrypsin inhibitors, plasmin-inhibitor; elastase inhibitors),
- (4) Haemaglutinnins (concanavalin A, ricin),
- (5) Plant enzymes (urease, lipoxygenase),
- (6) Cyanogenic glycosides (phaseolunatin, dhurrin, linamarin, lutaustralin),
- (7) Goitrogens (pro-goitrins and glucosinolates),
- (8) Oestrogens (flavones and genistein),
- (9) Saponins (soya sapogenin),
- (10) Gossypol from Gossypium species e.g. cotton,
- (11) Tannins (condensed and and hydrolysable tannins),
- (12) Amino acid analogues (BOAA, DAP, mimosine, N-methyl-1-alanine),

- (13) Alkaloids (solanine and chaconine),
- (14) Anti-metals (phytates and oxalates),
- (15) Anti-vitamins (anti-vitamins A, D, E and B12) and
- (15) Favism factors.

More often than not, a single plant may contain two or more toxic compounds, generally drawn from the two categories, which add to the difficulties of detoxification.

Trypsin Inhibitors

Many food products including legumes, cereals, potatoes and tomatoes contain inhibitors of enzymes such as trypsin, chymotrypsin, carboxypeptidases, elastase, and α -amylase. Protease inhibitors isolated from soyabean (the richest source of dietary trypsin inhibitors) fall into two main categories including the Kunitz inhibitor and the Bowman-Birk inhibitor. Inhibition by soyabean extracts of trypsin and chymotrypsin in rats and humans has been reported to be similar.

Levels of trypsin inhibitors in food and feed products

Data on trypsin inhibitor contents of some commonly used food and feed products are reviewed in Table 1. Soybeans are the most concentrated source of trypsin inhibitors among common food and feed products. Contents of tryspin inhibitors (mostly the Kunitz inhibitor) in soyabeans have been reported to vary from 8.6-48.2 mg/g sample or from 20.3-122.6 mg/g protein (Table 1). This large variation in contents of trypsin inhibitors could be due to differences in cultivars and, perhaps, to use of various methods of determination.

Compared to raw soybeans, peas, and other grain legumes, and properly processed soybean products contain considerably lower levels of trypsin inhibitors. Since the predominant trypsin inhibitors in soybeans are located mostly with the main storage proteins in the cotyledons, they tend to fractionate with the storage proteins when soybeans are processed into food ingredients. Therefore, when soybeans are defatted to produce raw flour, trypsin inhibitors become concentrated to levels of 28.0-65.8 mg/g sample or 57.8-131.6 mg/g protein.

Due to their proteic nature, trypsin inhibitors can be **inactivated** by heat processing including extrusion, infrared radiation, micronizing, boiling, autoclaving, steam processing or flaking or they can be removed by fractionation. The extent of heat inactivation of trypsin inhibitors depends upon a number of factors including the initial endogenous level, temperature, heating

time, particle size, moisture and perhaps crop species and cultivar. Most properly processed commercial soybean products for human consumption including concentrated soybean protein products such as concentrates (70% protein) and protein isolates (90% protein), soya-based infant formulas, soya milk and miso, have been subjected to sufficient heat treatment to inactivate up to 80% of the trypsin inhibitor activity in raw soya flour. Application of prolonged heating required to destroy all inhibitor activity would adversely affect protein digestibility and quality of the soybean products. For example, heating at 100°C for 10 min reduced trypsin inhibitor activity by about 80% and resulted in an optimal PER (Protein Efficiency Ratio) of about 3.1. However, prolonged heating to further reduce trypsin inhibitor activity resulted in a lower PER of about 2.9.

Table 1 Trypsin Inhibitor Content of Some Common Soya Protein and Other Legume Protein Products

Product	Trypsin inhibitor activity)	Trypsin inhibitor activity (mg/g)
Soyabeans, raw	16.7-27.2	34.7-122.6
Soyabeans, raw	48.2	-
Soyabeans, raw	8.62-18.21	20.3-51.1
Soya flour, raw	28.0-32.0	57.8
Soya flour, raw	52.1	104.2
Soya flour, raw	65.8	131.6*
Soyabeans, autoclaved	3.7-8.1	15.9-21.5
Soyabeans, boiled	0.9-4.0	2.2-11.7
Soya flour, roasted	62.6	-
Soya flour, autoclaved	4.2	-
Soya flour, toasted	3.2-7.9	-
Soya flour, toasted	7.9-9.4	-
Soya protein concentrate	5.4-7.3	8.4-11.2
Soya protein concentrate	6.3-13.7	-
Soya protein concentrate	4.4-7.3	6.8-11.2
Soya protein isolate	1.2-30.0	1.4-29.4
Soya-based infant formula	0.2-2.7	1.3-15.4
Soya tofu	0.6	9.2
Soya tofu	1.2-3.8	-
Soya milk	6.3	-
Soya sauce	0.3	3.3
Soya miso	4.1	22.9
Pea (various c-ultivars)	2.0-12.5	-
Pea, raw	-	11.9
Pea, boiled	-	2.1
Beans, indian indigeneous	-	13.5-62.3
Kidney bean	4.6	-
Jack bean, flour	-	2.3
Jack bean, prote0.1in isolate6.2	-	0.1
Velvet bean, flour	-	52.8
Velvet bean, protein isolate	-	6.2

*Assuming a protein content of 50% in soya flour

(* Source: Gilani et al., 2012)

In the absence of regulatory upper safe limits of dietary trypsin inhibitors, there is no guarantee that each and every commercial product would be properly processed and, consequently, would contain minimal residual levels of trypsin inhibitors, which may vary greatly with the extent of heat and other processing conditions used in the preparation of soyabean products. For example, several commercial soya beverages have been reported to contain significant levels of residual trypsin inhibitor activity (up to 71 and 64% of that of whole soybeans) against bovine and human trypsin, respectively. Similarly, some soya-based infant formulas have been reported to retain up to 28% of the trypsin inhibitor activity. An outbreak of gastrointestinal illness in individuals who had consumed an unprocessed soya protein extender in tuna fish salad documented the fact that inadequately processed soya products can find their way into the human food chain.

Protein and amino acid digestibility and protein quality have been reported to be negatively affected in animal models by the presence of high levels of dietary trypsin inhibitors and other antinutritional factors from soyabeans, kidney beans and other grain legumes. Boiling (for 30 min) has been found to be more beneficial than autoclaving (for 20 min) in reducing levels of trypsin inhibitors ($2 \cdot 2 - 12 \cdot 7$ vs $15 \cdot 9 - 22 \cdot 5$ mg/g protein), and in improving true faecal protein digestibility (75–93 vs. 60–70%) and PER values (1.35–2.30 vs. 0.91–1.33).

Tannins

Tannins are naturally occurring water-soluble polyphenolic compounds with the ability to complex and precipitate proteins in aqueous solutions. Their molecular weights range from 0.5to 3 kDa, and they are present in various plant species including cereal grains and legume seeds. While tannins protect the grains against insects, birds and fungal attacks, this agronomic character is accompanied with reduced nutritional quality. It is well known that tannins are potential protein precipitants and they reduce protein and amino acid digestibility in animals fed tannin-containing cereals such as sorghum, and grain legumes such as field beans and fababean. It has been found that under optimal conditions, sorghum tannin is capable of binding and precipitating at least 12 times its own weight of protein.

Tannins are generally classified into hydrolysable and condensed tannins. The hydrolysable forms are readily hydrolyzed by acids, alkalis, and some enzymes, while the condensed tannins, mainly polymerized products of flavan-3-ol (catechin) and flavan-3,4-diol or mixture of these, are resistant to hydrolysis. The condensed tannins which are also referred to as flavolans or procyanidins, are the main polyphenols present in commonly consumed food products while the hydrolysable forms are present only in small amounts.

Among important food and feed products, sorghum, millet, various types of beans and peas may contain considerable amounts of tannins (up to 72 g/kg) (Table 2). Some forage crops such as browse legumes may contain even higher amounts of tannins (up to 111 g/kg).

Currently there is no accurate information available on dietary intake of polyphenols; only a few estimates are available which are 1g/day (US), 23mg/day (Dutch), and 28mg/day (Denmark).

Table 2 Polyphenolic content of different plant foods and beverages Food mg/100g dry matter

Cereals	22-102.60	
Legumes	34-1710	
Nuts	0.04-38	
Vegetables	6-2025 (mg/fresh matter)	
Fruits	2-1200 (mg/fresh matter)	
Beverages		
Tea	75-105 (mg/100mL)	
Red wine	1000-4000 (mg/L)	
White wine	200-300 (mg/L)	

*(adapted from Bravo, 1998)

In general, tannins are resistant to heat. Several technological treatments have been studied to reduce tannin contents of food and feed products including sorghum and fababean. These include dehulling, soaking in water or alkaline solutions, addition of chemicals with a high affinity for tannins such as polyvinyl pyrrolidone and polyethylene glycol or gelatin and germination. However, most of these processing treatments appeared to be rather laborious, expensive or ineffective. More recently, genetic manipulation has been successful in producing fababean cultivars containing highly reduced levels of tannin.

Phytic acid

Phytic acid (myo-inositol hexaphosphoric acid) is a naturally occurring substance in the plant kingdom. This is mainly found in seeds, grains and nuts, where it functions as a source of mineral nutrients and inositol to be used during germination. Phytate accumulates primarily in the protein-rich aleuronic layers of monocotyledonous seeds, while it is distributed uniformly throughout the kernels in dicotyledonous seeds, including oilseeds and grain legumes (Table 3).

Phytate, with its abundance of negatively charged phosphate groups, is best known to chelate several nutritionally essential nutrients in the gastrointestinal tract of humans and animals, making them less bioavailable. Phytate interferes with zinc homeostasis and also negatively impacts on the bioavailability of other nutrients including proteins. It can negatively influence the activity of digestive enzymes such as carboxypeptidases and aminopeptidases, by the chelation of mineral cofactors or interaction with the protein (either enzyme or substrate). Binding of phytate to proteins may be direct (phytate: protein) or indirect (through cation

bridge). These complex interactions vary with pH, time, and relative concentrations. Due to its uneven distribution within grains, most of the phytate can be found in the germ of corn, the bran of wheat and the pericarp of rice. Processing treatments that remove or concentrate these portions can, therefore, result in dramatic changes in the phytate content of the finished product.

Product	Phytic acid (g/kg)	Phytic acid (g/kg	
		prtein)	
Rice, white, polished	3	-	
Rice, wild	22	-	
Rice polishing	55	377	
Wheat	6	53	
Wheat middlings	28	170	
Wheat, bran	34	-	
Sorghum	7	101	
Sunflower meal	27	89	
Corn	7	88	
Cottonseed meal	33	78	
Canola meal	26	72	
Soyabeans	26	-	
Soyabean meal	32-41	62-78	
Soyabean flours	10-20	20-37	
Soya protein isolates	10-20	11-22	
Soya milk	17	-	
Tofu	15-29	-	
Common bean (P. vulgarus), unextruded	8-11	459-578	
Common bean (P. vulgarus), extruded	7-8	351-423	
Chickpea	5-12	29-47	
Pigeon pea	7-17	29-72	
Moong bean	10-15	45-57	
Urad bean	13-15	46-54	
Lentils	7	27	
Cashew nuts	20	-	

(adapted from Gilani et al., 2012)

For example, rice polishings, wheat bran and middlings and defatted meals of oilseeds (soyabean, cottonseed sunflower and canola) contain substantial amounts of phytates (up to 5.5 per cent. In pulses (peas, lentils and beans), the greatest proportion of the phytate is found in edible cotyledons. Therefore, mechanical processing does not result in much reduction in phytate content. Moreover, phytate is relatively heat-stable, with only a small fraction being destroyed

during heat processing, while the phytase enzymes that might break down the phytate are heat labile. However, extrusion (with a central temperature and moisture content of 150 $^{\circ}$ C and 20 g/100 dry weight, respectively) of hard-to-cook common beans was reported to reduce phytate contents by about 20–30%.

Uricogenic nucleobases

Foods rich in nucleic acids, which elevate serum uric acid levels are restricted in the diets of hyperuricaemic individuals. The main food sources of nucleic acids are meats, leguminous seeds, some types of seafood, some vegetables and food yeasts. Among animal foods, organ meats are considered to be a rich source of nucleic acids. The use of new non-traditional protein sources such as inactive food yeasts may lead to increased consumption of nucleic acids and uricogenic bases. Inactive dried food yeasts, their autolysates and co-precipitates are increasingly used as functional ingredients in various foods and are also offered for sale as food supplements.

Due to their low cost, these protein-rich products have been commercially promoted for various nutritional applications. Various food yeasts may contain 8 to 12% nucleic acid and 0.7 to 1.0% adenine, depending upon the organism and the growth conditions used. The Protein- Calorie-Advisory Group (PAG) of the UN has recommended that a safe limit of nucleic acid from single cell protein products is 2 g/d and that the total nucleic acids from all dietary sources should not exceed 4 g/d for adults. The use of inactive dried food yeasts at low levels (3–5%) for functional purposes would not pose any problems if the yeasts contained not more than 8% nucleic acids. However, their use as a major protein source without significant reduction in their contents of nucleic acids, would have negative health effects including hyperuricaemia and inferior protein quality.

Anti-vitamin factors

Raw kidney beans are believed to contain an antagonist to vitamin E as evidenced by liver necrosis in rats and muscular dystrophy and low concentration of plasma tocopherol in chicks. Anti-vitamin E has also been noted in isolated soya protein, which is suspected to be α -tocopherol oxidase. Unheated soyabean flour has been found to be deficient not only in Vitamin B12, but it also contains a heat-labile factor that increases the requirement for vitamin B12. **Alkaloids** are also reported to cause alteration of normal foetal developments resulting in foetal malformation in ewes. These are caused by teratogenic alkaloids. Glycoalkaloids are reported to

cause haemolysis and toxicity to humans. Some plant alkaloids are reported to cause infertility. **Saponins** are characterised by a bitter taste and foaming properties. Erythrocytes lyses in saponin solution and so these compounds are toxic when injected intravenously. The anti-nutritional effects of saponins have been mainly studied using alfalfa saponins. Ricin occurs in castor beans (*Ricinus communis*) which have been reported to cause poisoning in all classes of livestock.

PROCESSING TO REDUCE ANFS

Castor bean meal can be detoxified by autoclaving at 20 psi for 60 min for incorporation in diets. Gamma irradiation treatment with its radiolytic effects can destroy glucosinolate molecules. Glucosinolate content was reduced to 40, 70 and 89% at irradiation dose levels of 10, 20 and 30 kGy, respectively. Other investigators reported that anti nutritional factors, such as protease inhibitors, α -amylase inhibitors, phytohamagglutinins, oligosaccharids and tannin were significantly inactivated by gamma irradiation.

Moth Bean (*Vigna aconitifolia*) was given different treatments including soaking, sprouting and cooking and the changes in the level of the anti-nutritional factors were estimated. Soaking the seeds in plain water and mineral salt solution for 12 h decreased phytic acid to the maximum (46–50%) whereas sprouting for 60 h had the most pronounced saponin lowering effect (46%). The processing methods involving heat treatment almost eliminated trypsin inhibitor activity while soaking and germination partly removed the metal chelating activity.

El-Adawy (2002) reported the nutritional composition and anti-nutritional factors of chickpeas (*Cicer arietinum L.*) undergoing different cooking methods and germination. Germination was less effective than cooking treatments in reducing trypsin inhibitor, hemagglutinin activity, tannins and saponins; it was more effective in reducing phytic acid, stachyose and raffinose. Cooking treatments and germination decreased the concentrations of lysine, tryptophan, total aromatic and sulfur-containing amino acids. However, cooked and germinated chickpeas were still higher in lysine, isoleucine and total aromatic amino acid contents than the FAO/WHO reference. The losses in B-vitamins and minerals in chickpeas cooked by microwaving were smaller than in those cooked by boiling and autoclaving. Germination resulted in greater retention of all minerals and B-vitamins compared to cooking

treatments. In vitro protein digestibility, protein efficiency ratio and essential amino acid index

were improved by all treatments. Based on these results, microwave cooking appears to be the best alternative for legume preparation in households and restaurants.

Mosha et al. (1995) reported the effect of blanching on the content of anti-nutritional factors in selected vegetables. Levels of both tannic acid and phytic acid were significantly (p<0.05) reduced by conventional and microwave blanching methods while oxalic acid levels were not significantly (p>0.05) reduced in most of the treatments by either of the blanching methods. In general, they recommended blanching as an effective method for reducing the anti-nutritional factors in green vegetables; however, further investigation on the heating times for both conventional and microwave blanching methods has been suggested.

Alonso et al. (2000) reported the effects of extrusion and traditional processing methods on antinutrients and *in vitro* digestibility of protein and starch in faba and kidney beans. De-hulling significantly increased protein content and greatly reduced condensed tannin and polyphenol levels in both legumes. Extrusion was the best method to abolish trypsin, chymotrypsin, α amylase inhibitors and haemagglutinating activity without modifying protein content. Furthermore, this thermal treatment was most effective in improving protein and starch digestibilities when compared with dehulling, soaking and germination. Roasting and autoclaving has been reported to decrease phytic acid in dry bean, chick pea and black

gram, cowpea and black bean. Cooking or autoclaving of *D.lablab* seeds reduced the tannin contents by 70 and 60%, respectively. Generally, adequate heat processing inactivates the trypsin and chymotrypsin. Heat stable compounds in cereals and legumes such as tannins and hydrates are easily removed after germination and fermentation.

Important Antinutritional Factors Formed During the Processing of Foods

Maillard Reaction Products (MRP)

MRP are widely consumed by this population, mostly as a result of their high intake of fast foods and snacks. It was concluded that the consumption of a diet rich in MRP negatively affects protein digestibility and utilization. Excessive intake of MRP during adolescence, however these studies warrant further investigation. The metabolism and anti-nutritional effects of MRP have been reviewed mainly in rats. Limited human studies relate to the effects of MRP on protein digestibility and utilization Different processing conditions used in the preparation of infant formulas such as sterilization, spray-drying, and treatment at ultra-high temperatures were reported to cause strong protein-protein and protein-lipid interactions in processed milk. These interactions were more pronounced in conventionally (in-can) sterilized than in spray-dried (powdered) and UHT (ultra heat treated) products demonstrating their temperature dependency. Analysis of raw materials, intermediate and end products revealed that in-can sterilization caused irreversible denaturation of proteins. In vitro analysis demonstrated significantly lower protein digestibility of in-can sterilized infant formulas compared to their spraydried and UHT counterparts. The amount of "available lysine" was lower in sterilized form. Similarly, values for true digestibility of protein, lysine, and protein quality (measure as relative PER) in liquid concentrates were up to 21% lower than those in powders.

D-amino acids and lysinoalanine (LAL)

The presence of D-amino acids in proteins leads to impaired protein and amino acid digestibility and nutritional quality. Although some insects, worms and marine invertebrates contain substantial quantities of D-amino acids, such organisms are not major components of the human diet. However, in communities where marine shellfish is an important source of food, the antinutritional implications of D-amino acids must be taken into account. Milk, meat and various grains do not contain substantial quantities of D-amino acids. However, during the course of preparation for consumption, processing treatments are applied which may give rise to racemization. The influence of processing treatments on racemization of amino acids was investigated by determining D-aspartic acid concentrations. In this study, raw milk contained the lowest level of D-aspartic acid (1.48 %) but quantities of D-aspartic acid increased with the extent of processing. For example, fat-free milk powder, kefir, evaporated milk, yoghurt and milk-based infant formulas contained 2.15, 2.44, 2.49, 3.12 and 4.95% of D-aspartic acid, respectively. A D-aspartic acid content of 31% was found in casein heated at 230 °C for 20 min. Similarly, high ratios of D-aspartic acid have been reported in textured soya protein and bacon (up to 13 %), alkaline/heat treated soya protein (up to 27.7 %), and alkaline/heat treated zein (up to 40.2 %) In mature cheeses, D-aspartic acid, D-alanine and D-glutamic acid were reported to be up to 68 %, suggesting that foods which have undergone microbial fermentation would contain substantial quantities of D-amino acids. In general, the L-AA do not racemize rapidly unless exposed to high temperatures.

LAL-amino acid contents of some common food and feed Products

LAL (an unnatural amino acid derivative) is formed when proteins are subjected to an alkaline treatment. It is formed mainly by the addition of an e-amino group of lysine residue to the double bond of a dehydro-alanine residue that has been generated by the b-elimination reaction of cystine, phosphoserine, or glycoserine residue. The amount of LAL formed in processed protein products is dependent upon temperature, concentration of alkali, time of exposure to alkali, type of protein, and type of cations in the solution. Heat treatment under non-alkaline conditions was also reported to form LAL in a variety of proteins. Although protein-containing food and feed products are commonly subjected to alkaline/ heat treatments, information on the amounts of LAL found in food products that are part of the every day diet are limited.

Antinutritional effects of protein-bound D-amino acids and LAL

Protein-bound D-amino acids formed during processing, especially at alkaline pH, may have adverse effects on protein digestibility and the quality and safety of processed foods. When absorbed, D-amino acids may be made utilizable by the action of racemases or epimerases or D-amino acid oxidases. The amino acid oxidase system (which varies in amount and specificity of oxidases in different animal species) may become saturated when foods containing high concentrations of D-amino acids are consumed. Proteins containing D-amino acids can be hydrolyzed at peptide bonds containing L-amino acids. However, the hydrolysis rates may be slower than those for corresponding unprocessed proteins. These changes adversely affect the nutritional quality and safety of foods by generating biologically non-utilizable forms of amino acids, creating D-D, D-L, and L-D peptide bonds partly or fully inaccessible to proteolytic enzymes. Moreover, these racemized proteins may compete with proteins that do not have racemized amino acids for the active site of digestive proteinases in the gut and thus adversely affect the biological utilization of the un-racemized proteins. The slower absorption of free and peptide-bound D- compared with L-amino acids may result in decreased protein digestibility.

Formation of Pyrolytic and Thermal Decomposition Products

The fluids at the surface of food are rich in amino acids, sugars and fats. High temperatures

decompose these compounds into smaller, more reactive molecules that combine to form stable compounds by pyrolysis. Many of these reactions are responsible to the characteristic colour, flavors and aromas associated with the food. Cooking temperatures above 200°C can promote formation of compounds that have been shown to be mutagenic and carcinogenic, such as polycyclic aromatic hydrocarbons and heterocyclic amines. The kind and amount of pyrolysis products formed depends on the parent compounds and the temperature. Modification of cooking techniques can help reduce their formation.

Acrylamide formation in starchy foods which are oven-baked or fried foods is of concern to the health fraternity. Owing to its low molecular weight and polarity it is readily incorporated and distributed in thymus, liver, kidney, heart, brain of animals and humans and (it along with its derivative glycidamide) has a potential to react with DNA leading to point mutations and cancer. Formation of glycidamide is a critical step for the genotoxic effects of acrylamide and its derivatives. Indian population is also at substantial risk as our diets also contain many starchy fried foods including chips from potato, tapioca, banana, jackfruit, tikkis, puris, bhature, paranthas. Although studies for determining the level of trans fats in Indian diets are lacking, we also need to gear up in reducing these levels as such a scare can hamper our trade.

Effects of acrylamide on health is mainly carcinogenicity as levels required for causing neurotoxicity are impractical from human consumption point of view. Clear evidence of carcinogenic and genotoxic effects of acrylamide and glycidamide have been found in in-vitro and in-vivo trials.

The Confederation of Food and Drink Industries of the EU, even launched an acrylamide reduction toolbox to offer the most efficient solutions to assist the affected industries. The toolbox is a combination of suggestions for changes in raw materials, processes and recipes, with importance placed on the fact that the brand-specific customer acceptance of the final products should not be impacted. Several factors such as sugar contents in food matrixes; processing conditions (eg heating times and oil type); heat processing methods (e.g. frying after a microwave pre-cooking step); pH modification (e.g. use of citric acid);and the use of protein hydrolysates, divalent cations (e.g. Ca^{2+}), hydrogencarbonates (e.g. NaHCO₃), and antioxidants (e.g. tocopherol), are known to influence the formation of acrylamide in heat processed foods.

Strategies for acrylamide reduction in potato products

- Low reducing sugar potato varieties: Limiting factors for acrylamide formation in poatao products are reducing sugars. Hence, the selection of potato varieties low in reducing sugars offers a promising strategy for acrylamide reduction. Furthermore, increased nitrogen fertilization increased free asparagine, but lowered reducing sugars and therefore acrylamide.
- **Di and trivalent cations:** The addition of di and trivalent cations has been proposed in the patent literature to reduce acrylamide in manufactured potato products
- Vacuum Frying: The application of vacuum frying might also offer an opportunity to reduce acrylamide, since lower temperatures can be applied without altering the sensory properties of the crisps. By lowering the frying temperature of potato chips from 185° to165°C, it is possible to reduce the acrylamide formation by half.
- **Flavonoids:** Acrylamide levels were significantly reduced by addition of a flavonoid containing blend of spices.
- The addition of fish meat, nearly pure protein, significantly reduced acrylamide which is probably due to the reaction of previously formed acrylamide with the SH and amino groups of proteins.
- Acids: Addition of consumable acids such as citric acid is a very simple but efficient method to minimize acrylamide in bakery products which was due to a lower pH.

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