International Journal of Chemical Studies

P-ISSN: 2349–8528
E-ISSN: 2321–4902
IJCS 2019; 7(1): 2472-2479

Received: 09-11-2018
Accepted: 14-12-2018

Abhishek Thakur
Department of Food Science and Technology, Dr. YS Parmar University of Horticulture and Forestry, Nauni, Solan, Himachal Pradesh, India

Vishal Sharma
Department of Biotechnology, Dr. YS Parmar University of Horticulture and Forestry, Nauni, Solan. Himachal Pradesh, India

Aayushee Thakur
Department of Biotechnology, Dr. YS Parmar University of Horticulture and Forestry, Nauni, Solan. Himachal Pradesh, India

Correspondence
Vishal Sharma
Department of Biotechnology, Dr. YS Parmar University of Horticulture and Forestry, Nauni, Solan. Himachal Pradesh, India

An overview of anti-nutritional factors in food

Abhishek Thakur, Vishal Sharma and Aayushee Thakur

Abstract
Anti-nutritional factors are the compounds found in most food substances which are poisonous to humans or in some ways limit the availability of nutrient to the body. Anti-nutritional factors are present in different food substances in varying amounts, depending on the kind of food. Many anti-nutrients (oxalate, phytate, etc.) and toxic substances (cyanide, nitrate, phenols, etc.) are present in many plants and vegetables. Anti-nutrients in foods are responsible for deleterious effects related to the absorption of nutrients and micronutrients. However, some anti-nutrients may exert beneficial health effects at low concentrations. For example, phytic acid, lectins, tannins, saponins, amylase inhibitors and protease inhibitors have been shown to reduce the availability of nutrients and cause growth inhibition. However, when used at low levels phytate, lectin, tannins, amylase inhibitors and saponins have also been shown to reduce the blood glucose and insulin responses to starchy foods and/or the plasma cholesterol and triglyceride. Anti-nutrients are chemical substances which reduces the maximum utilization of nutrients especially proteins, vitamins, and minerals, thus preventing optimal exploitation of the nutrients present in a food and decreasing the nutritive value. Anti-nutrients can be divided into two groups: heat-stable group-phytic acid, tannins, alkaloids, saponins, non-protein amino acids etc. and heat labile group include lectins, cynogenic glycosides, protease inhibitors and Toxic amino acids etc. Due to the presence of several anti-nutritional factors, legumes possess low protein digestibility. Most of the toxic and anti-nutrient effects of these compounds in plants could be removed by several processing methods such as soaking, germination, boiling, autoclaving, fermentation, genetic manipulation and other processing methods, but extensive research is still needed to discover elimination methods for heat stable anti-nutrients present in various food without altering the nutritional value of food.

Keywords: Anti-nutritional, phytic acid, tannin, cynide

Introduction

Anti-nutritional factors are compounds which reduce the nutrient utilization and/or food intake of plants or plant products used as human foods. They play a vital role in determining the use of plants for humans. Plants evolved these substances to protect and prevent themselves from being eaten. However, if the diet is not varied, some of these toxins build up in the body to harmful levels. Some vitamins in food may be destroyed by anti-nutritional substances. These anti-nutritional factors must be inactivated or removed, if values of food substances are to be fully maintained. Plants which produce seeds rich in energy supplies (carbohydrates, lipids, proteins) usually accumulate potent chemical defence compounds. This also applies to grain legumes with comparably large and protein-rich seed which often contain substantial amounts of "anti-nutritive" factors (ANF), such as lectins, protease inhibitors, non-protein amino acids (NPAAs), alkaloids, cyanogenic glycosides, pyrimidine glycosides, saponins, tannins, isoflavones, oligo- saccharides, erucic acid, or phytates. Anti-nutritional factors are present in different food substances in varying amounts, depending on the kind of food, mode of its propagation, chemicals used in growing the crop as well as those chemicals used in storage and preservation of the food substances.

Many anti-nutrients (oxalate, phytate, etc.) and toxic substances (cyanide, nitrate, phenols, etc.) are present in many plants and vegetables. Cassava for example is known to contain high levels of cyanide, a respiratory poison (Ogbadoyi *et al.*, 2011). Consumption of vegetables in their fresh form which is believed to contain more micronutrients than processed vegetables becomes a major health concern because of the high levels of anti-nutrients and toxic substances that might be ingested with the associated health problems. Anti-nutrients or anti-nutritional factors may be defined as those substances generated in natural feedstuffs by the normal metabolism of species and by different mechanisms (for example inactivation of some nutrients, diminution of the digestive process or metabolic utilization of feed) which exerts effect contrary to optimum nutrition (Soetan and Oyewole, 2009). Being an anti-nutritional factor is not an intrinsic characteristic of a compound but depends upon the digestive process of the ingesting animal. Trypsin inhibitors, which are anti-nutritional factors for monogastric animals, do not exert adverse effects in ruminants because they are degraded in the rumen.

Legumes, including beans, occupy an important place in human nutrition as in many countries they are one of the staple food. Besides being a cheap source of valuable proteins, saccharides, and several micronutrients including minerals and vitamins, they are known as rich in dietary fibre and low in fat. The contribution of legumes in the daily diet has many beneficial physiological effects. It allows to prevent common metabolic diseases, such as diabetes mellitus, coronary heart disease (CHD) and cancer. Therefore, their consumption is supposed to have a positive correlation with reducing the CHD death (Krupa, 2008) [14]. Of particular interest are resistant starch, enzyme inhibitors, lectins and polyphenols, therefore their role as preventive agents in diets of persons suffering from metabolic disorders is gaining attention. The term "antinutritional compounds" (ANCs) will be used in regard to bioactive compounds of bean seeds.Some of these chemicals are known as ''secondary metabolites'' and they have been shown to be highly biologically active (Zenk, 1991) [37]. They include saponins, tannins, flavonoids, alkaloids, trypsin (protease) inhibitors, oxalates, phytates, haemagluttinins (lectins), cyanogenic glycosides, cardiac glycosides, coumarins and gossypol. The list is inexhaustible. Some of these plant chemicals have been shown to be deleterious to health or evidently advantageous to human and animal health if consumed at appropriate amounts. Most of these secondary metabolites elicit very harmful biological responses, while some are widely applied in nutrition and as pharmacologically-active agents (Soetan, 2008) [31]. The anti-nutritional factors 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 (Soetan and Oyewole, 2009).

Anti-nutritional factors in foods are responsible for the deleterious effects that are related to the absorption of nutrients and micronutrients which may interfere with the function of certain organs. Most of these anti-nutritional factors are present in foods of plant origin. Thus, the presence of cyanogenic glycosides, enzyme inhibitors, lectins, tannins, alkaloids, and saponins in foods may induce undesirable effects in humans if their consumption exceeds an upper limit. Certain harmful effects might also be due to the breakdown products of these compounds. However, some anti-nutritional factors as well as their breakdown products may possess beneficial health effects if present in small amounts. The mechanism through which the anti-nutritional and beneficial effects of food anti-nutritional factors are exerted is the same. Thus, manipulating processing conditions, in addition to removing certain unwanted compounds in foods, may be required to eliminate the deleterious effects of anti-nutritional factors and take advantage of their health benefits. Health risk factors associated with anti-nutritional factors include: lack of knowledge of the tolerance levels to these compounds in the human organism, little available information on the degree of variation of individual risks and little knowledge with respect to the influence of environmental factors on the detoxification capacity of the human organism.

Classification of the Anti-Nutritional Factors

Anti-nutritional factors also known as anti-nutrients which are poisonous substances that can be found in most food and able to limit the nutrient available to the body. Basically, anti-nutrients can be divided into two primary groups, which are heat-stable and heat labile group (Gemede and Ratta, 2014) [8]. Anti-nutrients with heat-stable property which resist and can be maintained at high temperature are Phytic acid, Condensed Tannins, Alkaloids, Saponins; whereas, for anti-nutrients that fall in heat-labile group that are sensitive to standard temperature and lost at high temperature are lectins, Cynogenic Glycosides, Protease inhibitors, and Toxic amino acids.

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.

According to Aletor (1993) [2], there are several anti-nutritional factors that are very significant in plants and are used for human foods and animal feeds. They are: (i) Enzyme inhibitors (trypsin and chymotrypsin inhibitors, plasmin inhibitors, elastase inhibitors), (ii) Haemaglutinnins, (iii) Plant enzymes (urease, lipoxygenase), (iv) Cyanogenic glycosides (phaseolunatin, dhurrin, linamarin, lutaustralin), (v) Goitrogens (pro-goitrins and glucosinolates), (vi) Oestrogens (flavones and genistein), (vii) Saponins (soya sapogenin), (viii) Gossypol from *Gossypium species* e.g. cotton, (ix) Tannins (condensed and hydrolysable tannins), (x) Amino acid analogues (BOAA, DAP, mimosine, N-methyl-1-alanine), (xi) Alkaloids (solanine and chaconine), (xii) Anti-metals (phytates and oxalates), (xiii) Anti-vitamins (anti-vitamins A, D, E and B12) and (xiv) Favism factors.

Table 1: Classification of endogenous anti-nutrients with in plant feedstuffs

Proteins	• Protease inhibitors >Trypsin inhibitors >Chemotrypsin • Hemagglutinins • Food allergens • Toxic amino acids
Glycosides	• Saponins • Cyanogens • Estrogens • Goitrogens
Phenols	• Gossypol • Tannins
Others	• Anti-minerals Phytic acid Oxalates • Anti-vitamins • Anti- enzymes

Source: Liener, 1980 [17].

Table 2: Adverse effects of some anti-nutrients

Anti-nutrients	Effects on body
Phytates	Reduce Ca and Fe absorption
Oxalates	Reduce Ca absorption, encourage kidney stone formation
Cyanide	Respiratory inhibitors
Lectins (Hem agglutinins)	Prevent absorption of digestive end products in the small intestine.
Protease inhibitors	Substances reduce protein digestion.
Phenol Compounds	They reduce bioavailability of some minerals (especially zinc). They may negatively affect pH mechanism, reduce protein digestion.

(***Source***: Gemede and Ratta, 2014) [8].

Protease inhibitors

Protease inhibitors are protein-based substances widely distributed within the plant kingdom, including the seeds of most cultivated legumes, which have the ability to inhibit the activity of proteolytic enzymes within the gastro-intestinal tract of animals. They are readily destroyed by heat; the degree of destruction or inactivation depending upon the temperature, duration of heating, particle size and moisture conditions. These substances reduce protein digestion. They decompose with heat. Therefore, when legumes are eaten raw or without being cooked properly, they upset digestive functions and cause diarrhea or excessive gas. Autoclave treatment or boiling also reduces the quantity of these substances. About 10-20% of the total active trypsin is found in human pancreatic juice. They bind proteases, which are resistant to digestion in the small intestine, and thus ensure their removal through excretion. The presence of trypsin inhibitors in the diet leads to the formation of irreversible condition known as enzyme-trypsin inhibitor complex. This causes a drop in intestine trypsin and a decrease in protein digestibility, leading to slower animal growth. Trypsin inhibitors are a unique class of proteins found in raw soybeans that inhibit protease enzymes in the digestive tract by forming indigestible complexes with dietary protein. These complexes are indigestible even in the presence of high amounts of digestive enzymes.

Amylase inhibitors

Amylase inhibitors are also known as starch blockers because they contain substances that prevent dietary starches from being absorbed by the body. Starch is a complex carbohydrates that cannot be absorbed unless they are first broken down by the digestive enzyme amylase and other secondary enzymes. Pigeon peas have been reported to contain amylase inhibitors. These inhibitors have been found to be active over a pH range of 4.5-9.5 and are heat labile (Marshall and Lauda, 2007) [18]. Amylase inhibitors inhibit bovine pancreatic amylase but fail to inhibit bacterial, fungal and endogenous amylase. Pigeon pea amylase inhibitors are synthesized during late seed development and also degraded during late germination. Amylase inhibitors are also very heat labile and have been reported as having hypoglycemic effects. However, instability of this inhibitor under the conditions of the gastrointestinal tract resulted in failure to reduce insulin responses and increase the caloric output of food by using them as starch blocker tablets (Giri and Kachole, 2004) [9].

Hemagglutinins (lectin)

They are proteins or glycoproteins. Lectin activity has been determined in more than 800 varieties of the legume family. 2-10% of the total protein legume seeds are lectins. One of their most important characteristics is that they prevent absorption of digestive end products in the small intestine. They enable the coagulation of red blood cells by affecting erythrocytes. Lectins possess some other interesting chemical and biological properties, some of which are as follows: they interact with specific blood groups; they perform various functions in mitotic division, demolish cancerous cells and have toxic effects in some animals. Since they bond with different sugar groups, their bonding with intestinal wall may exhibit variation depending on the type of sugar. If some types of beans are consumed raw, they may cause shock cramps. Besides these characteristics, lectins can easily disintegrate. Plant hemagglutinins are referred to as phytohemagglutinins (PHA). Lectins are carbohydrate binding proteins present in most plants, especially seeds like cereals, beans, etc., in tubers like potatoes and also in animals. Lectins selectively bind carbohydrates and importantly, the carbohydrate moieties of the glycoproteins that decorate the surface of most animal cells. Dietary lectins act as protein antigens which bind to surface glycoproteins (or glycolipids) on erythrocytes or lymphocytes (Sauvion *et al*., 2004) [27]. They function as both allergens and hemagglutinins and are present in small amounts in 30% of foods, more so in a whole-grain diet. The consumption of lectin-containing foods may lead to endogenous loss of nitrogen and protein utilization. The carbohydrates and proteins that are undigested and unabsorbed in the small intestines reach the colon where they are fermented by the bacterial flora to short-chain fatty acids and gases. These may in turn contribute to some of the gastrointestinal symptoms associated with the intake of raw beans or purified lectins. The lectin-induced disruption of the intestinal mucosa may allow entrance of the bacteria and their endotoxins to the blood stream and cause toxic response. Lectins may also be internalized directly and cause systemic effects such as increased protein catabolism and breakdown of stored fat and glycogen, and disturbance in mineral metabolism (Fereidoon, 2014) [7].

Allergens

They are substances that are generally found in nutrients. They cause allergic reactions that are specific to certain individuals. The level of harm done depends on the sensitivity level of individual’s body rather than the quantity of the substances taken with the food. Diarrhea and vomiting are symptoms of allergy. It is also argued that proteins with high molecular weight cause allergies (Perlman, 1980) [25]. Histamine and compounds of histamine derivatives act as antigens against allergens.

Toxic Amino acids

There are certain amino acids in legume plants that are not of protein nature and reduce nutritious value and cause toxic effects. These substances are commonly found in Lathyrus and broad beans. Dihydroxyphenyl alanine (DOPA) is the most common toxic amino acid found in legumes. Although these amino acids do not display a direct toxic effect, the plant firstly takes on a black color due to these substances, and then withers. Moreover, the nutritional value of plants that contain such amino acids (broad beans, Lathyrus) decreases substantially. Toxic amino acids are believed to combine causes of metabolic favism. When it is taken into account that pulses are sources of the highest quality vegetable proteins, the importance of studies on the toxicity mechanisms of toxic amino acids that have an unfavorable effect on the quality of this protein and the degree of their potential harm become obvious. Canavanine: another toxic amino acid found in the seeds of the legume sesbania (*Sesbania* spp.) and jack bean (*Canavalia* spp.) and acts as arginine antagonist. The toxicity of the seed proteins increasing with increasing dietary inclusion level, and being reduced with water extraction (canavanine being soluble in water).

Saponins

Saponins are a heterogeneous group of naturally occurring foam-producing triterpene or steroidal glycosides that occur in a wide range of plants, including pulses and oil seeds such as kidney bean, chickpea, soybean, groundnut, lupin and sunflower. Saponins are secondary compounds that are generally known as non-volatile, surface active compounds which are widely distributed in nature, occurring primarily in the plant kingdom. The name ‘saponin’ is derived from the Latin word sapo which means ‘soap’, because saponin molecules form soap-like foams when shaken with water. They are structurally diverse molecules that are chemically referred to as triterpene and steroid glycosides. They consist of nonpolar aglycones coupled with one or more monosaccharide moieties (Oleszek, 2002) [23]. This combination of polar and non-polar structural elements in their molecules explains their soap-like behaviour in aqueous solutions. The structural complexity of saponins results in a number of physical, chemical, and biological properties, which include sweetness and bitterness, foaming and emulsifying properties, pharmacological and medicinal properties, haemolytic properties, as well as antimicrobial, insecticidal, and molluscicidal activities (Sparg *et al*., 2004) [32]. Their general characteristics can be cited as follows: they give a bitter taste, foam when they are treated with various solutions and cause haemolysis in red blood cells. Since they reduce the surface tension of blood in cold-blooded animals, they have an extremely toxic effect. Saponins, in high concentrations, impart a bitter taste and astringency in dietary plants. The bitter taste of saponin is the major factor that limits its use. saponins were recognized as anti-nutrient constituents, due to their adverse effects such as for growth impairment and reduce their food intake due to the bitterness and throat-irritating activity of saponins (John *et al*., 2004) [13]. In addition, saponins were found to reduce the bioavailability of nutrients and decrease enzyme activity and it affects protein digestibility by inhibit various digestive enzymes such as trypsin and chymotrypsin (Simee, 2011) [30].

Saponins are attracting considerable interest as a result of their beneficial effects in humans. Recent evidence suggests that saponins possess hypocholesterolemic, immunostimulatory, and anticarcinogenic properties (Oleszek, 2002) [23]. In addition, they reduce the risk of heart diseases in humans consuming a diet rich in food legumes containing saponins. On the other hand, due to their cholesterol-reducing effect, legumes are the most important sources of saponins. The fact that saponins can bond with cholesterol and therefore reduce absorption and that legumes contain saponins points to their importance for health. Saponin-rich foods are important in human diets to control plasma cholesterol, preventing peptic ulcer, Osteoporosis and to reduce the risk of heart disease.

Cynogens

A number of plant species produce hydrogen cyanide (HCN) from cyanogenic glycosides when they are consumed. These cyanogens are glycosides of a sugar, often glucose, which is combined with a cyanide containing aglycone. Cyanogenic glucosides are classified as phytoanticipins. Their general function in plants is dependent on activation by b-glucosidases to release toxic volatile HCN as well as a ketones or aldehydes to fend off herbivore and pathogen attack (Zagrobelny *et al*., 2004) [36]. Cyanogenic glycosides or cyanoglycosides account for approximately 90% of the wider group of plant toxins known as cyanogens. The key characteristic of these toxins is cyanogenesis, the formation of free hydrogen cyanide and is associated with cyanohydrins that have been stabilised by glycosylation (attachment of sugars) to form the cyanogenic glycosides. Hydrogen cyanide inactivates the enzyme cytochrome oxidase in the mitochondria of cells by binding to the Fe^{3+}/Fe^{2+} contained in the enzyme. This causes a decrease in the utilization of oxygen in the tissues. Cyanide causes an increase in blood glucose and lactic acid levels and a decrease in the ATP/ADP ratio indicating a shift from aerobic to anaerobic metabolism. Cyanogenic glucoside on hydrolysis yields toxic hydrocyanic acid (HCN). The cyanide ions inhibit several enzyme systems, depress growth through interference with certain essential amino acids and utilization of associated nutrients. They also cause acute toxicity, neuropathy and death (Osuntokun, 1972) [24].

Cyanide activates glycogenolysis and shunts glucose to the pentose phosphate pathway decreasing the rate of glycolysis and inhibiting the tricarboxylic acid cycle. Hydrogen cyanide will reduce the energy availability in all cells, but its effect will be most immediate on the respiratory system and heart. Cyanogenic glucosides are widely distributed in the plant kingdom and more than 2500 different plant species have been reported to contain cyanogenic glucosides including cassava (*Manihot esculenta*), linseed (*Linum usitatissmium*), various sorghums (*Sorghum spp.*) and white clover (*Trifolium repens*). Lesser quantities are found in the kernels of such plants as almonds (*Amygdalus communis*), apricots (Prunus armeniaca), peaches (*Prunus persica*), and apples (*Malus sylvestris*) (Zagrobelny, 2008) [35]. As cyanide is extremely toxic, one of the most obvious symptoms is death. In the body, cyanide acts by inhibiting cytochrome oxidase, the final step in electron transport, and thus blocks ATP synthesis. Prior to death, symptoms include faster and deeper respiration, a faster irregular and weaker pulse, salivation and frothing at the mouth, muscular spasms, dilation of the pupils, and bright red mucous membranes. The toxicity of a cyanogenic plant depends primarily on the potential concentration of hydrogen cyanide that may be released upon consumption. Upon consumption of a cyanogenic plant, β-glycosidase will be released during digestion and remain active until deactivated by the low pH of the stomach. This enzyme will hydrolyse the cyanogenic glycoside and release at least part of the potential hydrogen cyanide content of the plant (WHO, 2010) [34].

Phytoestrogens

Phytosterols are compounds exhibiting estrogenic activity and have been found in a wide variety of food plants and legumes, including wheat, rice, chick-pea, alfalfa, lupin, groundnut, linseed and soybean. For example, compounds exhibiting estrogenic activity in soybean have been identified as is of lavones, including genistein, daidzein, and coumestrol, of which genistein is the most prominent. However, although genistein is reported to be heat stable.

Goitrogens

Goitrogens are naturally occurring substances that can interfere with the function of the thyroid gland, have been found in legumes such as soybean and groundnut. Goitrogens get their name from the term ‘goiter’ which means the enlargement of the thyroid gland. They may act directly on the gland or indirectly by altering the regulatory mechanisms of the gland and peripheral metabolism and excretion thyroid glands. If the thyroid gland has difficulty synthesizing thyroid hormone, it may enlarge to compensate for this inadequate hormone production. They have been reported to inhibit the synthesis and secretion of the thyroid hormones. Since thyroid hormones play an important part in the control of body metabolism their deficiency results in reduced growth and reproductive performance. Goitrogenic effect have been effectively counteracted by iodine supplementation rather heat treatment (Akande *et al*., 2010) [1]. Soybean, a kind of oil seed and cruciferous vegetables contain glycosides called goitrogens, Consisting of sulphur, these glycosides cause the thyroid gland to grow by inhibiting the iodine intake of the thyroid gland. This toxic effect can be reduced with the addition of iodine to the diet (Nagaraj, 2009) [21].

Phenols

a) Gossypol

Gossypol is a naturally occurring polyphenolic compound present in the pigment glands of cotton seed (*Gossypium spp.*). The average gossypol content varying from 0.4-2.4% within glanded cotton seeds to less than 0.01% free gossypol within some low-gossypol cotton seed meals (Liener, 1980) [17]. Reduced lysine availability has been reported with cotton seed protein due to the ability of gossypol to bind with the reactive epsilon amino group of lysine during heat processing. The general symptoms of gossypol toxicity are depressed appetite, loss of weight, laboured breathing and cardiac irregularity. Death is usually associated with reduced oxygen-carrying capacity of the blood, haemolytic effects on erythrocytes and circulatory failure. Dietary gossypol also causes olive-green discolouration of yolks in eggs.

b) Tannins

Tannin is an astringent, bitter plant polyphenolic compound that either binds or precipitates proteins and various other organic compounds including amino acids and alkaloids. Tannins have molecular weights ranging from 500 to over 3000 daltons. Tannins also interfere with dietary iron absorption (Rao and Desothale, 1998) [26]. Food crops and legumes which have been reported to contain significant quantities of tannins include sorghum (containing up to 5% condensed tannins), faba bean, lima bean, sunflower seed meal (containing 1.2-2.7% chlorogenic acid) and rapeseed.

The anti-nutritional properties of tannins depend upon their chemical structure and dosage. Tannins are heat stable and they decreased protein digestibility in animals and humans, probably by either making protein partially unavailable or inhibiting digestive enzymes and increasing fecal nitrogen. Tannins are known to be present in food products and to inhibit the activities of trypsin, chemotrypsin, amylase and lipase, decrease the protein quality of foods and interfere with dietary iron absorption (Lumen and Salamat, 1980) [4].

There are two different groups of tannins: hydrolyzable tannins and condensed tannins. Condensed tannins are widely distributed in leguminous forages and seeds. Tannins may form a less digestive complex with dietary antagonize arginine and interfere with Ribonucleic Acid proteins and may bind and inhibit the endogenous protein, such as digestive enzymes (Kumar and Singh, 1984) [15]. Tannin-protein complexes involve both hydrogen-bonding and hydrophobic interactions. The precipitation of the protein-tannin complex depends upon pH, ionic strength and molecular size of tannins. Both the protein precipitation and incorporation of tannin phenolics into the precipitate increase with increase in molecular size of tannins. However, when the molecular weight exceeds 5,000 daltons, the tannins become insoluble and lose their protein precipitating capacity and degree of polymerization becomes imperative to assess the role of tannins in ruminant nutrition. Tannins have been found to interfere with digestion by displaying anti-amylase activity. Helsper *et al.* (1993) [10] reported that condensed tannins were responsible for the testa bound trypsin inhibitor activity of faba beans. Tannins also have the ability to complex with vitamin B (Liener, 1980) [17]. Other adverse nutritional effects of tannins have been reported to include intestinal damage, with iron absorption and the possibility of tannins producing a carcinogenic effect. Other toxic effects of tannins can be categorized as: depression of food intake, inhibition of digestive enzymes, increased excretion of endogenous protein, digestive tract malfunctions and toxicity of absorbed tannin or its metabolites.

c) Phytic acid

Phytate (also known as Inositol hexakisphosphate (InsP6)) is the salt form of phytic acid, are found in plants. Phytic acid causes the bioavailability of essential minerals to decrease and turn into insoluble compounds whose absorption and digestion is less in the small intestine (Desphande and Cheryan, 1984) [5]. Pulses are sources of dietary phytoid. When phytoid phosphor is not made use of, it is discharged with excretion. A way of preventing this is through the hydrolysis of phytoid phosphor; for this purpose, besides methods such as soaking, germinating, using food rich in vegetable endogen phytosis enzyme and storing, methods like cooking and performing autoclave where phytoid phosphor is demolished in the presence of heat can also be used. The studies that have been conducted demonstrated that phytoids reduce cholesterol level and protect against intestinal cancer of iron origin. Besides, phytoids exhibit characteristics of natural antioxidants also their benefits such as reducing lipid peroxidation. 62-73% and 46-73% of the total phosphorus within cereal grains and legume seeds being in form of organically bound phytin phosphorus, respectively (Matyka *et al*., 1993) [20]. Apart from the major part of the phosphorus contained within phytic acid being largely unavailable to animals (due to the absence of the enzyme phytase within the digestive tract of monogastric animals), phytic acid acts as a strong chelator, forming protein and mineral-phytic acid complexes; the net result being reduced protein and mineral bioavailability.

Phytic acid is present in considerable quantities within many of the major legumes and oilseeds. This includes soybean, rapeseed and cotton seed. As phytic acid accumulates in storage sites in seeds, other minerals apparently chelates to it forming the complex salt phytate. Whole soybeans have been reported to contain 1-2% phytic acids. The major part of the phosphorus contained within phytic acid are largely unavailable to animals due to the absence of the enzyme phytase within the digestive tract of monogastric animals. In the chicken there is a significant inverse relationship between phytic acid and the availability of calcium, magnesium, phosphorus and zinc in feedstuffs such as rapeseed, palm kernel seed, cotton seed and soybean meals. Phytic acid acts as a strong chelator, forming protein and mineral-phytic acid complexes; the net result being reduced protein and mineral bioavailability (Erdman, 1979) [6]. Phytic acid is reported to chelate metal ions such as calcium, magnesium, zinc, copper, iron and molybdenum to form insoluble complexes that are not readily absorbed from gastrointestinal tract. Phytic acid also inhibits the action of gastrointestinal tyrosinase, trypsin, pepsin, lipase and amylase. Erdman (1979) [6] stated that the greatest effect of phytic acid on human nutrition is its reduction of zinc bioavailability.

Dietary phytate may also have health benefits for Diabetes patients because it lowers the blood glucose response by reducing the rate of starch digestion and slowing gastric emptying. Likewise, phytate has also been shown to regulate Insulin secretion (Shamsuddin, 2002) [29]. It is believed that phytate reduces Blood clots, Cholesterol, and Triglycerides, and thus prevents Heart diseases. It is also suggested that it prevents renal stone development. It is used as a complexing agent for removal of traces of heavy metal ions (Selvam, 2002) [28]. Among the cooking treatments boiling appeared effective to reduce the phytate level, which could reduce as high as 20% of phytate.

Oxalates

Oxalates affects calcium and magnesium metabolism and react with proteins to form complexes which have an inhibitory effect in peptic digestion. Oxalic acid binds calcium and forms calcium oxalate which is insoluble. Calcium oxalate adversely affects the Ca absorption. Fatal human poisoning following the eating of large quantity of the leaves of certain plants i.e. rhubarb, known to contain relatively large amounts of oxalates. Cooking can reduce the soluble oxalate content of many common vegetables, but not the insoluble fraction, if the cooking water containing some of the leached soluble oxalate is discarded.

A salt formed from oxalic acid is known as an Oxalate: for example, Calcium oxalate, which has been found to be widely distributed in plants. Strong bonds are formed between oxalic acid, and various other minerals, such as Calcium, Magnesium, Sodium, and Potassium. This chemical combination results in the formation of oxalate salts. Some oxalate salts, such as sodium and potassium, are soluble, whereas calcium oxalate salts are basically insoluble. The insoluble calcium oxalate has the tendency to precipitate (or solidify) in the Kidneys or in the Urinary tract, thus forming sharp-edged calcium oxalate crystals when the levels are high enough. These crystals play a role to the formation of kidney stones formation in the urinary tract when the acid is excreted in the urine (Noonan and Savage, 1999) [22].

Higher content of oxalate can bind to calcium present in food, thereby rendering calcium unavailable for normal physiological, and biochemical role such as the maintenance of strong bone, teeth, cofactor in enzymatic reaction, nerve impulse transmission, and as clotting factor in the blood. Though lost of calcium leads to degeneration of bones, teeth, and impairment of blood clotting process. When oxalic acid is consumed, it irritates the lining of the gut, and can prove fatal in large doses. If food with excessive amounts of oxalic acid is consumed regularly, nutritional deficiencies are likely to occur, as well as severe irritation to the lining of the gut. In ruminants oxalic acid is of only minor significance as an anti-nutritive factor since ruminal micro-flora can readily metabolize soluble oxalates, and to a lesser extent even insoluble Ca oxalate. The values of oxalate changes as a result of processing. Soaking and cooking of foodstuffs high in oxalate will reduce the oxalate content by leaching. Boiling may cause considerable skin rupture, and facilitate the leakage of soluble oxalate into cooking water; this may be the possible reason to observed high reduction in oxalate level upon boiling (Bhandari, and Kawabata, 2004) [3]. It is reported that boiling affects the highest reduction in oxalate. Calcium oxalate is insoluble at a neutral or alkaline pH, but freely dissolves in acid. Oxalate can be found as soluble and insoluble forms in plants. Soluble salts are formed when oxalate binds with potassium, sodium and magnesium (magnesium oxalate is less soluble than the potassium and sodium salts) while insoluble salts are produced when the oxalate binds with calcium and iron.

Anti-vitamin factors

Anti-vitamin factors are a wide variety of compounds exhibiting anti-vitamin activity have been isolated from plants, including 1) anti-vitamin A factor present in soybeans, which destroys carotene and is not readily destroyed by heat, 2) anti-vitamin D factor present in soybeans, which interferes with calcium and phosphorus absorption in chicks, and is destroyed by autoclavinlg, 3) anti-vitamin E factor present in kidney beans, soybeans, alfalfa and field pea, causing liver necrosis and muscular dystrophy in chicks and lambs, and is destroyed by autoclaving, 4) anti-vitamin K factor present in sweet clover, 5) anti-thiamine factor called thiaminase present in cottonseed, linseed, mung bean, and mustard seed, 6) anti-niacin factor present in sorghum, 7) anti-pyridoxine factor present in linseed, which is destroyed by water extraction and autoclaving, and 8) anti-vitamin B 12 factor present in raw soybeans (Hill, 2003) [11].

Alkaloids

Alkaloids are one of the largest groups of chemical compounds synthesised by plants and generally found as salts of plant acids such as oxalic, malic, tartaric or citric acid. Alkaloids are small organic molecules, common to about 15 to 20 per cent of all vascular plants, usually comprising several carbon rings with side chains, one or more of the carbon atoms being replaced by a nitrogen. They are synthesized by plants from amino acids. Decarboxylation of amino acids produces amines which react with amine oxides to form aldehydes. The characteristic heterocyclic ring in alkaloids is formed from Mannich-type condensation from aldehyde and amine groups. Tubers of the common potato (*Solanum tuberosum*) have a natural content of the two toxic and bitter glycoalkaloids (GA) a-solanine and a-chaconine. The levels are normally low and without adverse effects on food safety and culinary quality. However, consumption of potato tubers with unusual1y high GA contents (300-800 mg kg-I) has occasionally been associated with acute poisoning, including gastro- intestinal and neurological disturbances, in man. Tuber GA levels are inheritable and can vary considerably between different species. Environmental factors experienced by tubers during germination, growth, harvest and storage may affect GA levels further (Jadhav *et al*., 2009) [12]. Alkaloids are considered to be anti-nutrients because of their action on the nervous system, disrupting or inappropriately augmenting electrochemical transmission. For instance, consumption of high tropane alkaloids will cause rapid heartbeat, paralysis and in fatal case, lead to death. Uptake of high dose of tryptamine alkaloids will lead to staggering gate and death. Indeed, the physiological effects of alkaloids have on humans are very evident. Cholinesterase is greatly inhibited by glycoalkaloids, which also cause symptoms of neurological disorder.

Alkaloids cause gastrointestinal and neurological disorders (Aletor, 1993) [2]. The glycoalkaloids, solanine and chaconine present in potato and *Solanum* spp. are haemolytically active and toxic to fungi and humans. Some of the toxicological manifestations of potato glycoalkaloids involve gastro-intestinal upsets and neurological disorders, especially in doses in excess of 20 mg/100 g sample. Coumarins, which are constituents of forage, have been associated with the so-called bleeding disease in cattle consuming spoiled or putrid sweet clover. Alkaline pH conditions generally enhance absorption of glycoalkaloids, where binding with sterols in cell membranes causes extra disruption. Nicotine, caffeine, quinine and strychnine are well-known examples of alkaloids. As an illustration, lower dose of alkaloids mediate important pharmacological activities, such as analgesic, reducing blood pressure, killing tumour cells, stimulating circulation and respiration (Simee, 2011) [30]. Some of the more well-known alkaloids include nicotine (tobaccao), cocaine (leaves of coca plant), quinine (cinchona bark), morphine (dried latex of opium poppy) and solanine (unripe potatoes and potato sprouts).

Anti-enzymes

Several compounds exhibiting anti-enzyme activity, in addition to the Protease inhibitors, have been isolated from plants, including 1) the cholinesterase inhibitor solanine present in green potatoes, which is heat stable and water soluble, 2) amylase inhibitors present in wheat, oats and rye, and 3) an arginase inhibitor present in sunflower seeds, which is a derivative of chlorogenic acid (Liener, 1980) [17].

Table 3: Anti-nutritional factors present in some plant feedstuffs

Feedstuffs	Antinutritional Factors
Soybean meal	Trypsin inhibitors (glycinin), lectins, oligosaccharides, phytic acid.
Peas	Lectins, tannins, oligosacharides.
Rapeseed meal	Glucosinolinates, tannins, phenolic acids.
Lupin	Alkaloids.
Sunflower meal	Tannins.
Wheat	Phytic acid, polyphenols, tannins, saponins.
Barley	β- glucans.
Some wild edible plants (bauhinia recemosa etc.)	Oxalate, phytate and tannins

***Source*:** Liener, 1980 [17]

Table 4: Anti-nutritional factors present in some pulses

Pulses/Legume	Botanical Name	Anti-Nutritional Factors
Broad/faba bean	*Vicia faba*	Protease inhibitors, Phytic acid, Phytohaemagglutinins, tannins
Chick pea/Bengal gram	*Cicer arietinum*	Protease inhibitors, Cyanogens, Phytic acid, saponins, Estrogens.
Cow pea	*Vigna unguiculata*	Protease inhibitors, Phytic acid, Phytohaemagglutinins.
Kidney bean	*Phaseolus vulgaris*	Protease inhibitors, Phytic acid, Phytohaemagglutinins, cyanogens, Saponins, Anti-vitamin E factors, Amylase inhibitor.
Hyacinth bean	*Dolichus lablab*	Protease inhibitors, Cyanogens, Phytohaemagglutinins.
Mung bean/ green gram	*Phaseolus aureus*	Protease inhibitors, Phytic acid, Saponins, Anti-thiamine factors.
Field pea	*Pisum sativum*	Protease inhibitors, cyanogens, Phytohaemagglutinins, Phytic acid, Saponins, Anti-vitamin E factors.
Pigeon pea/red gram	*Cajanus cajan*	Protease inhibitors, cyanogens, Phytohaemagglutinins, Phytic acid.
Black gram	*Phaseolus mungo*	Protease inhibitors (Trypsin, Chemotrypsin), phytic acid.

***Source*:** Tacon, 1992 [33].

Table 5: Anti-nutritional factors present in oilseeds

Oilseeds	Botanical Name	Anti-Nutritional Factors
Groundnut	*Arachis hypogaea*	Protease inhibitors, Phytic acid, Phytohaemagglutinins, Saponins, Estrogenic factors.
Rapeseed	*Brassica campestris napus*	Protease inhibitors, Glucosinolates, Phytic acid, tannins.
Indian mustard	*Brassica juncea*	Protease inhibitors, Glucosinolates, Anti-thiamine factor.
Cottonseed	*Gossypium spp.*	Phytic acid, estrogenic factors, Gossypol, Anti-vitamin E factor.
Linseed	*Linum usitatissimum*	Cyanogens, Phytic acid, Estrogenc factor, Anti-thiamine factor, Anti-pyridoxine factor.
Soybean	*Glycine max*	Protease inhibitors, Glucosinolates, Phytohaemagglutinins, Phytic acid, Saponins, Estrogenic factors, Anti-vitamin E factor, Anti-vitamin A factor, Anti-vitamin D factor, Anti-vitamin B_{12} factor, Allergens.
Sesame	*Sesamum indicum*	Phytic acid.

***Source*:** Hill, 2003 [11].

Table 6: Removal of anti-nutritional factors by different methods

Physical Processing	Working
Autoclaving, pressure cooking, steaming	Heating at ultrahigh temperatures (>100 °C).
Blanching	Mild boiling (75 °C-95 °C) to inactivate endogenous enzymes
Ordinary cooking	Usually preceded by soaking, de-hulling, germination and fermentation
Extrusion	High temperature short time (HTST) processing
Roasting	Dry heating at 120 °C-250 °C
Soaking	Exposure to water and salt solutions
Processing chemical	Treatment with thiols, sulphite, Cu-salts (± ascorbic acid)

***Source*:** Price *et al*., 1987; Shahidi, 1997

Conclusion

All processing treatments were effective in reduction of anti-nutritional factors, however, pressure cooking was found to be the best for removal of ANFs. The processing treatments have significant effect on chemical composition, mineral content and free fatty acid and protein digestibilty. Blanching and cooking resulted in significant reduction in oxalic acid; only blanching resulted in significant reduction in phytic acid and polyphenol contents. The anti-nutrients are more susceptible to moist heat than dry heat. Germination and fermentation are potential processes to improve mineral extractability and decrease anti-nutrients which binds to mineral and reduce their availability. In some pulses, soaking before cooking is the most traditional, economic and appropriate method to reduce anti-nutrients. Today, a deeper knowledge of the chemical structure of the anti-nutrients involved can help to devise technological strategies to process legume seeds in order to obtain toxin free products. Most of the toxic and anti-nutrient effects of these compounds in food could be removed by several processing methods such as soaking, germination, boiling, autoclaving, fermentation, genetic manipulation and other processing methods, but extensive research is still needed to discover elimination methods for heat stable anti-nutrients present in various food without altering the nutritional value of food.

References

1. Akande K, Doma U, Adamu H. Major Antinutrients Found in Plant Protein Sources: Their Effect on

Nutrition. Pakistan Journal of Nutrition. 2010; 9(8):827-832.
2. Aletor VA. Allelochemicals in plant foods and feeding Stuffs. Part I. Nutritional, and Physiopathological aspects in animal production. Vet. Human Toxicol. 1993; 35(1):57-67.
3. Bhandari MR, Kawabata J. Cooking effects on oxalate, phytate, trypsin, cyanide and amylase inhibitors of wild yam tubers of Nepal. Journal of Food Composition and Analysis. 2004; 19:524-530.
4. De Lumen BO, Salamat LA. Trypsin inhibitor activity in winged beans (*Psophocarpus tetragnolobus*) and the posible role of tannins. J Agric. Food Chem. 1980; 28:533-536.
5. Desphande S, Cheryan M. Effect of phytic acid, divalent cations and their interactions on alfa-amilase activity. J Food Sci. 1984; 49:516-519.
6. Erdman JW. Oilseed phytates: Nutritional implications. J Am. Oil Chemist Soc. 1979; 56:736-741.
7. Fereidoon S. Beneficial Health Effects and Drawbacks of Anti-nutrients and Phytochemicals in Foods. Appl Microbiol Bio technol. 2014; 97:45-55.
8. Gemede HF, Ratta N. Antinutritional factors in plant foods: Potential health benefits and adverse effects. Global Advanced Research Journal of Food Science and Technology. 2014; 3(4):103-117.
9. Giri AP, Kachole MS. Amylase inhibitors of pigeon pea (*Cajanus cajan*) seeds. Phytochemistry. 2004; 47:197-202.
10. Helsper JP, Hoogendijk JM, Norel AV, Burger-meyer K. Antinutritional factors in faba beans (*Vicia faba* L.) as affected by breeding toward the absence of condensed tannins. J Agric. Food Chem. 1993; 41:1058-1061.
11. Hill T. Plant Antinutritional Factors. Sumarski List. 2003; 109:325-328.
12. Jadhav SJ, Sharma RP, Salunke DK. Naturally occurring toxic alkaloids in foods. CRC Crit Rev Toxic. 2009; 9:21-104.
13. John Shi, Konesh Arunasalam, David Yeung, Yukio Kakuda, Gauri Mittal, Yueming Jian. Saponins from Edible Legumes: Chemistry, Processing, and Health Benefits. J Med Food. 2004; 7(1):67-78.
14. Krupa U. Main nutritional and antinutritional compounds of bean seeds- A REWIEV. Polish journal of food and nutrition sciences. 2008; 58(2):149-155.
15. Kumar R, Singh M. Tannins: Their adverse role in ruminant nutrition. J Agric. Food Chem. 1984; 32:447-453.
16. Kumar R, Nataraju S, Jayaprehash C, Sabapathy SN, Bawa AS. Effect of heat treatment on the stability of oxalic acid in selected plant foods. The Indian Journal of Nutrition and Dietetics. 2006; 43:337-340.
17. Liener IE. Toxic constituents of plant food Stufls. Second edition, New York and London, Academic Press, 1980, 502.
18. Marshall JJ, Lauda CM. Purification and properties of *Phaseolus vulgaris*. J Biol. Chem. 2007; 250:8030-8037.
19. Massey LK, Palmer RG, Horner HT. Oxalate content of soybean seeds (*Glycine* max: Leguminosae), Soya foods, and other edible legumes. Journal of Agriculture and Food Chemistry. 2001; 49:4262-4266.
20. Matyka S, Bogusz G, Korol W. Phytate contents in cereal grains, legume and rape seeds. Biuletyn Informacyjny Przemyslu Paszowedo (Poland). 1993; 32:37-43.
21. Nagaraj G. Oil Seeds- Properties, Processing, Products and Procedures. New India Publishing Agency, New Delhi, 2009, 601.
22. Noonan SC, Savage GP. Oxalic acid and its effects on humans. Asia pacific Journal of Clinical Nutrition. 1999; 8:64-74.
23. Oleszek WA. Chromatographic determination of plant saponins. J Chromatogr. A. 2002; 967:147-162.
24. Osuntokun BO. Cassava diet and cyanide metabolism in Wistar rats. Brit. J Nutr. 1972; 24:797-805.
25. Perlman F. Allergens. Toxic constitutes of plant foodstuffs. Academic Pres, New York, 1980, 295-327.
26. Rao P, Desothale YG. Tannin content of pulses varietal difference and effects of germination and cooking. J Sci. food Agric. 1998; 33:1013-1015.
27. Sauvion N, Nardon C, Febvay G, Gatehouse AM, Rahbe Y. Binding of the insecticidal lectin concanavalin A in pea aphid, *Acyrthosiphon pisum* (Harris) and induced effects on the structure of midgut epithelial cells. J Insect Physiol. 2004; 50:1137-1150.
28. Selvam R. Calcium oxalate stone disease: Role of lipid peroxidation and antioxidants. Urological Research. 2002; 30(1):35-47.
29. Shamsuddin AM. Anti-cancer function of phytic acid. International Journal of Food Science and Technology. 2002; 37(7):769-782.
30. Simee W. Isolation and determination of antinutritional compounds from root to shells of peanut (*Arachis Hypogaea*). J Disper. Sci. Technol. 2011; 28:341-347.
31. Soetan KO. Pharmacological and other beneficial effects of anti-nutriional factors in plants-A Review. Afr. J Biotechnol. 2008; 7(25):4713- 4721.
32. Sparg SG, Light ME, Van Staden J. Biological activities and distribution of plant saponins. J Ethnopharmacol. 2004; 94:219-243.
33. Tacon AGJ. Nutritional fish pathology: morphological signs of nutrient deficiency and toxicity in farmed fish. FAO Fisheries Technical Paper No. 330, FAO, Rome, 1992, 75.
34. World Health Organization. Toxicological evaluation of certain food additives and naturally occurring toxicants. WHO Food Additive Series: 30. World Health Organization, Geneva, 2010.
35. Zagrobelny M, Bak S, Moller BL. Cyanogenesis in plants and arthropods. Phytochemistry. 2008; 69:1457-1468.
36. Zagrobelny M, Bak S, Rasmussen AV, Jorgensen B, Naumann CM, Moller BL. Cyanogenic glucosides and plant insect interactions. Phytochemistry. 2004; 65:293-306.
37. Zenk HM. chasing the enzymes of secondary metabolism: Plant cell cultures as a pot of goal. Phytochemistry. 1991; 30(12):3861-3863.