

An overview of nutritional and anti nutritional factors in green leafy vegetables

Abstract

Vegetables play important role in food and nutritional security. Particularly, green leafy vegetables are considered as exceptional source for vitamins, minerals and phenolic compounds. Mineral nutrients like iron and calcium are high in leafy vegetables than staple food grains. Also, leafy vegetables are the only natural sources of folic acid, which are considerably high in leaves of *Moringa oleifera* plants as compared to other leafy and non-leafy vegetables. This paper reviewed nutritional and anti nutritional factors in some important common green leafy vegetables. The type and composition of nutritional and anti nutritional factors vary among genera and species of different edible leafy vegetables plants. Anti nutritional factors are chemical compounds in plant tissues, which deter the absorption of nutrients in humans. Their effects can be direct or indirect and ranges from minor reaction to death. Major anti nutritional factors such as nitrates, phytates, tannins, oxalates and cyanogenic glycosides have been implicated in various health-related issues. Different processing methods such as cooking and blanching can reduce the contents of anti-nutritional factors. This paper also discussed in brief the various analytical methods for the determination of the various nutritional and anti-nutritional factors in some green leafy vegetables.

Keywords: green leafy vegetables, nutritional factors, anti nutritional factors

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Abbreviations: RUBISCO, ribulose-1,5-bisphosphate carboxylase/oxygenase; LNA, α -linolenic acid; WHO, world health organization; SDF, soluble dietary fiber; IDF, insoluble dietary fiber; USDA, us department of agriculture

Introduction

Generally, vegetables are widely designated as “protective foods” in human diet due to their varied health benefits attributable to the richness in vitamins, essential fatty acids, minerals, amino acids and dietary fiber¹ and various essential bioactive compounds.² These include health-promoting plant secondary metabolites composed of antioxidants and phenolic compounds. It is well acknowledged that to meet recommended daily allowance of nutrition, the World Health Organization (WHO) recommendation at least 400g of fruit and non-starchy vegetables (WHO, 2013) is used. The dietary guidelines for Americans recommend five servings of vegetables per day based on an intake of 2000 calories (HHS/USDA, 2015). It is also recommended that one of the five servings of vegetables should be green leafy vegetables. Nutritionists and dieticians are of the opinion that people should diversify their diets as no single vegetable meets all the nutritional requirements necessary for good health and wellbeing.³ Hence in today’s agri-food systems, more emphasize is given to crop diversification with the intention of fulfilling human nutritional requirements, and to reduce the pressure on cereal production.⁴ Globally, crop diversity and nutritional value of vegetable crops are of special significance for improving food and nutrition security.⁵ Plants are major sources of numerous bioactive compounds collectively termed as phytochemicals, which are reported to be key to good health.² Researchers have shown that the composition of phytochemicals is very distinctive and varies widely amongst plants.⁶ For instance, vegetables that belong to the *Alliaceae* family (e.g., onions, garlic, shallots, leek, Welsh onion and chives) are characterized by thiosulfides and flavonoids. Cruciferous

vegetables that belong to the *Brassicaceae* family (e.g., cabbage, cauliflower, kale and broccoli) contain high sources of glucosinolates, and those that belong to *Cucurbitaceae* family (e.g., squash, pumpkin, cucumber, melon and bitter gourd) are rich in carotenoids and tocopherols.² However, the compositions of these phytochemicals have been documented to be affected by factors such as genotypic characteristics, climatic conditions, edaphic factors and management practices.⁷⁻¹⁰ For instance, it was shown that for the same plant species, the mineral nutrients content and total phenolic compounds and antioxidants can be altered by the application of different sources of nutrients.⁷⁻¹¹

It is worth noting that while there are many groups of chemical compounds that have health benefits, others can be very toxic and fatal to humans when consumed. As such phytochemicals can be broadly classified as nutritional (e.g., essential fatty acids, proteins, vitamins, minerals and phenolic compounds) and anti-nutritional (e.g., oxalates, tannins, nitrate) chemical compounds.¹² It is important for consumers and researchers to understand the importance of these chemicals and their impacts on human health and available methods for their assessments. This paper aims at reviewing the common phytochemicals in green leafy vegetables grown in various geographic locations of the world. The health-promoting phytochemicals will be referred to as nutritional factors and the health-inhibiting or toxic phytochemicals will be referred to as anti-nutritional factors. The importance of these phytochemicals will be highlighted to reflect their beneficial or toxicity and/or inhibitory effects on human health and wellbeing. Additionally, a brief description will be made on methods for determining the compositions of these phytochemicals in green leafy vegetables.

Nutritional factors in green leafy vegetables

Proteins: Proteins are large and complex molecules composed of various compositions of amino acids. Proteins play critical roles in

cellular functions, structure and regulations of metabolic activities in all living organisms. Hence, proteins have primary importance in the daily diets of consumers. Green leafy vegetables are the richest and cheapest sources of proteins. This is because of their ability to synthesize and accumulative amino acids with the help of abundant source of sunlight, water, oxygen and nitrogen which is readily available in the atmosphere.¹³ About 50% of total leaf cell protein is dominated by ribulose-1,5-bisphosphate carboxylase/oxygenase (RUBISCO), which can be found in leaf chloroplasts. It plays a vital role in the fixation of atmospheric carbon during photosynthesis.¹⁴ RUBISCO is a similar protein found in all green leafy vegetables with few changes in amino acid groups from species to species. Recent evidence showed that green leafy vegetables such as spinach (*Spinacia oleracea*), broccoli (*Brassica oleracea* var. *Italica*) and duckweed (*Lemna perpusilla*) provide all the essential amino acids that meet the FAO nutrition standards.¹⁵ Evidence showed that apart from lower methionine content, cassava (*Manihot esculenta*) leaves consumed as green leafy vegetable has amino acids profile comparable with pulse and dairy products.¹⁶ It has been found that some of the African leafy vegetables such as green leaves of septic weed (*Senna occidentalis*) and cassava both have 7 g protein/100 g of fresh weight. This is higher than that of exotic leafy vegetables such as *Brassica oleracea* subsp. Capitata with a protein content of 1 g/100 g of fresh weight.¹² However, protein content in African leafy vegetables cannot be compared with legume proteins such as white lupines (*Lupinus albus*), which has 11.5 g protein/100g of fresh weight.¹⁷ The amount of protein content in leafy vegetables can vary with farming practices and prevailing environmental conditions.¹⁸ Bioavailability of protein in leafy vegetables is typically influenced by thermal processing, which inactivates heat-labile anti-nutritional factors such as protease inhibitors, lectins, thiaminases and goitrogens but enhances digestibility of proteins and starch.¹⁹ Total protein is determined by evaluating total N using the Kjeldahl method and multiplying the N value by 6.25 as described by Association of the Official Analytical Chemists.²⁰

Dietary fiber: Dietary fiber is the constituent of plant cell wall (MacDougall, 1995). Dietary fiber is classified as soluble dietary fiber (SDF) and insoluble dietary fiber (IDF), which is collectively termed as total dietary fiber (TDF). Green leafy vegetables have been traditionally recognized as good sources of dietary fiber.²¹ Literature information showed that Indian Green leafy vegetables such as basella (*Basella rubra*), fenugreek (*Trigonella foenum graecum*), hibiscus (*Hibiscus cannabinus*), coriander (*Coriandrum sativum*), cabbage (*Brassica oleracea*) and spinach (*Spinacia oleracea*) are good sources of soluble dietary fiber content. Consumption of higher levels of vegetable fiber resulted in reduced risk of cardiovascular diseases and possibly, colon cancer.²² It was more significant in resolving the problem of constipation, diabetes, diverticulosis and obesity.^{23–25} The amount of total dietary fiber in green leafy vegetables can vary with different plant variety of the same species, agro-climatic conditions, stages of maturity and type and rate of fertilizer applications. Estimation of DF in GLV can be performed by using lyophilized powder and its fractions by enzymatic or gravimetric methods of AOAC.

Vitamins: Green leafy vegetables are abundant sources for β -carotene. In leaves, vitamin A is present in the form of provitamin A carotenoids such as β -carotene (ca. 25-30%), α -carotene, γ -carotene, β -cryptoxanthin and non-provitamin A carotenoids lutein (ca. 45%), violaxanthin (ca. 15%) and neoxanthin (ca. 15%),²⁶ the content of

vitamin A is expressed in retinol equivalents (RE) with one (1) RE being equivalent to 6 μ g of β -carotene and 12 μ g of the other pro vitamin carotenoids. The recommended daily allowance (RDA) for vitamin A is also expressed in RE but recently, the U.S. Institute of Medicine has replaced RE with the term “retinol activity equivalent” (RAE; IOM, 2001). The Institute of Medicine dietary reference intake recommended 900 and 700 μ g RAE of vitamin A for an adult male and female, respectively.²⁷ Increasing the consumption of green leafy vegetables that are widely available in developing countries help in combating prevailing vitamin A deficiency in regions where pharmaceutical supplements and vitamin A fortified foods are limited.²⁸ Processing techniques such as cooking, boiling, and steaming have significant influence on availability of carotenoids in green leafy vegetables.²⁹ A study conducted on 30 commonly used green leafy vegetables for nutritional purposes confirm the presence of good amount of lutein³⁰ and richness in various vitamins. For instance, any species of Amaranths are excellent source of vitamin C.³¹ Furthermore, comparisons can be made on vitamins distribution among seeds and leafy plants. For instance, data obtained from USDA national nutrient database¹⁵ clearly shows that vitamin A and K1 (Phylloquinone) in leafy vegetables are exceptionally high for kale and spinach than the cereals and pulses. In addition, cereals and pulses are devoid of vitamin C whereas, leafy vegetables are the potential sources of vitamin C with good amount in kale (1014mg/100g). Vitamin E in duck weed (45.7mg/100g) is high among many common leafy vegetables and seeds. However, vitamin B5 in cereals and pulses are higher than spinach and kale but not in duck weed (2.1mg/100g). Folate is a water-soluble compound, which belongs to vitamin B group. Deficiency of folic acid has severe metabolic and clinical consequences.³² Plants are the major source of foliates for humans especially, green leafy vegetables.³³ Cereal grains and tuber based staple diet are very low in foliate, which can be improved by the addition of green leafy vegetables. Leaves of colocasia (*Colocasia esculenta*) are used as an excellent source of foliate in the Indian diet. However, it is noticed that about 50% or more of foliate in food is found to be destroyed during cooking. This is mainly attributed to prolonged heating of vegetables in a large volume of water. Hence, it is advisable to consume the water used during the cooking of vegetables.³⁴ A research study reported that 5-formyl-5,6,7,8-tetrahydrofolic acid (502.1 μ g/100g dry weight) and 5,6,7,8-tetrahydrofolic acid (223.9 μ g/100g dry weight) are the dominant forms of foliate present in *Moringa oleifera*. *Moringais* known to be a significant source of foliate with higher bioavailability compared to other vegetables.³⁵ High-performance liquid chromatography i.e. HPLC–DAD–MS/MS based method developed by Santos et al.³⁶ allows a simple and sequential extraction and monitoring of several free forms of water soluble vitamins (vitamins C, B1, B2, B3, B5, B6 and B9) and fat-soluble vitamins (pro-vitamin A and vitamin E) present in raw green leafy vegetables Table 1.

Minerals: WHO (1996) stated that the “overall malnutrition must no longer be considered without reference to micronutrient status, as the two are inextricably linked. Attempting to improve protein-energy status without addressing micronutrient deficiencies will not result in optimal growth and function”.³⁷ Metal ions are important to the normal functioning and wellbeing of humankind as they serve as cofactors in enzymatic reactions, and maintain protein structures. Iron deficiency in women and children lead to the development of anemia.³⁸ Zinc deficiency results in impaired gastrointestinal and immune functions.³⁹ A comparative mineral profile for seeds and green leaves are presented in Table 2. The data shows that green

leafy vegetables are good sources of mineral nutrients. For instance, spinach has highest amount of calcium (1036mg/100g), magnesium (827mg/100g), iron (28.4mg/100g) and sodium (827mg/100g) whereas duck weed is high in zinc (15mg/100g). However, soy seed also has considerable amount of calcium (195mg/100g), iron (6mg/100g), magnesium (407mg/100g), phosphorus (469mg/100g) and potassium (2387mg/100g), sodium (12.3mg/100g) and zinc (3.7mg/100g) when compared to all the seeds but comparatively higher in the leafy vegetables. The absorption of mineral nutrients

is adversely affected by the presence of inhibitors like oxalate and phytates and many other anti nutritional factors.⁴⁰ Minerals have greater stability during food processing as compared to vitamins and proteins.⁴¹ But mineral profiles in green leafy vegetables are highly dependent on application of external synthetic fertilizer or organic soil amendments. Minerals in leafy vegetables can be determined by atomic absorption spectrophotometry (AAS) method,⁴² inductively coupled plasma optical emission spectroscopy (ICP-OES).³¹

Table 1 Vitamin compositions for some seed and leafy vegetables^a

Vitamins concentrations ^b	Seed					Leaf		
	Wheat	Corn	Rice	Soy	Lentil	Kale	Spinach	Duck-Weed
Vitamin A, IU	9	167	0	114	68	130,000	85,500	77,900
Vitamin B1, mg	0.4	0.2	0.2	0.6	0.6	0.9	0.9	1.1
Vitamin B2, mg	0.2	0.1	0	1.1	1.3	0.9	1.8	2.8
Vitamin B5, mg	1	0.5	1.5	1.5	0.4	0.9	0.9	2.1
Vitamin B6, mg	0.3	0.3	0.8	0.5	0.5	2.5	1.8	1
Vitamin C, mg	0	0	0	0	2	1014	256	94
Vitamin E, mg	0.8	0.3	0.2	1.8	ND	9.3	18.2	45.7
Vitamin K1, mg	1.9	0.2	0	67	ND	6900	4400	51

^aSource: (Edelman and Colt, 2016). "Derived from the USDA National Nutrient Database (<http://nutritiondata.self.com>) for: wheat (*Triticum aestivum*) flour, whole grain; corn (*Zea mays*) flour, whole grain, yellow; rice (*Oryza sativa*) flour, white, un enriched; soy (*Glycine max*) flour, full-fat, raw; chick pea (*Cicer arietinum*), mature seeds, raw; lentil (*Lens culinaris*), pink, raw; spinach (*Spinacia oleracea*), raw; broccoli (*Brassica oleracea* var. *italica*), raw; kale (*Brassica oleracea* var. *sabellica*), raw. Data for duckweed (*Lemnoideae*) determined by Eurofins USA for a local Israeli isolate of dried, raw, *Wolffia* sp"; b values are per 100g sample. All samples are normalized at 10% moisture; ND, indicates not determine.

Table 2 Mineral compositions of some seed and green leafy vegetables^a

Mineral concentration ^b	Seed					Leaf		
	Wheat	Corn	Rice	Soy	Lentil	Kale	Spinach	Duck-weed
Calcium (mg)	34	6.4	10	195	34	846	1036	607
Iron (mg)	3.8	2.2	0.4	6	6.4	8.3	28.4	25.7
Magnesium (mg)	120	85	36	407	46	265	827	231
Phosphorus (mg)	332	250	100	469	276	519	513	1741
Potassium (mg)	405	289	78	2387	664	2769	5840	5319
Sodium (mg)	3.1	4.6	0	12.3	5.9	214	827	132
Zinc (mg)	3	1.6	0.8	3.7	3.2	3.2	5.5	15

^aSource: (Edelman and Colt, 2016). "Derived from the USDA National Nutrient Database (<http://nutritiondata.self.com>) for: Wheat flour, whole grain; corn flour, whole grain, yellow; rice flour, white, un enriched; soy flour, full-fat, raw; chick pea, mature seeds, raw; lentil, pink, raw; spinach, raw; broccoli, raw; kale, raw. Data for duckweed determined by Eurofins USA for a local Israeli isolate of dried, raw, *Wolffia* sp"; b values are per 100g sample. All samples are normalized to 10% moisture.

Essential fatty acids: Omega-3 fatty acids are important for normal growth and development, and play vital role in the prevention and treatment of coronary artery diseases, hypertension, diabetes, arthritis, cancer and other inflammatory and autoimmune disorders.⁴³ α -linolenic acid, the precursor of omega-3 fatty acid has been found in green leafy vegetables with beneficial effects on health.⁴⁴ It is known that essential fatty acids help to control various chronic diseases.⁴⁴ Evidence shows that omega-3 fatty acids is relatively high in wild plants than in cultivated vegetables.^{45,46} For instance, the US Department of Agriculture (USDA) and other studies have reported

that purslane (*Portulaca oleracea*), a wild plant and weed in most cases, is a good non-aquatic source of α -linolenic acid with 4 mg/g fresh weight. The same study also reported the presence of α -linolenic acid in other leafy green vegetables such as 1.7 mg/g in spinach, 1.1mg/g in mustard greens, 0.7 mg/g in red leaf lettuce and 0.6 mg/g in butter crunch lettuce.⁴⁴ Fatty acids in leaves can be determined by gas chromatography.¹

Anti nutritional factors in green leafy vegetables

Undesirable chemical substances referred to as anti nutrients are

abundant in both cultivated and wild plant species.⁴⁷ These anti-nutrients are also referred to as “Allelochemicals”.⁴⁸ The quantity and the distribution of these chemical compounds vary with plant genera and species. According to Cheek and Shull,⁴⁹ being an anti nutritional factor is not an intrinsic characteristic of a compound but depends on the digestive process of the ingesting animal. The level of adversity is largely dependent on the diet pattern and method of processing involved before the consumption of the specific plant food. The individual compounds and their impact on human diet are described below. Some of the anti nutrients commonly found in leafy vegetables are nitrates, oxalates, tannins, phytates and cyanogenic glycosides.

Nitrate: Nitrate is one of the important and natural compounds found in vegetables, and is responsible for characterizing vegetable quality. Typically, nitrate concentration in leafy vegetables is found higher than other groups of vegetables such as root and fruits vegetables.⁵⁰ Agronomic (practices such as the amount, timing and form of N fertilizer, and environmental and genetic factors can significantly influence the levels of nitrate in raw green leafy vegetables.⁵¹ A study conducted on 10 leafy vegetables harvested at two different light intensities (i.e. 200 and 400 $\mu\text{mol m}^{-2} \text{s}^{-1}$) showed higher nitrate accumulation when harvested at low light intensity $\leq 200 \mu\text{mol m}^{-2} \text{s}^{-1}$.⁵² Amount of nitrate content present per serving of any vegetable is non-toxic but its metabolites and bi products such as nitrite, nitric oxide and N-nitros compounds are the main compounds of health concern.⁵³ Dietary intake of total nitrate undergoes enterosalivary circulation, which is then converted into nitrites by oral bacteria and salivary enzymes. This conversion rate is about 5-7% in healthy adults but considerably higher in infants and patients with gastroenteritis.⁵⁴ The presence of nitrates in infants’ causes very serious disease called methemoglobinemia or ‘blue baby syndrome’ leading to suffocation and death.⁵⁵ The World Health Organization has set the acceptable daily intake of nitrate at 3.7 mg/kg body weight and nitrite at 0.06 mg/kg body weight.⁵⁶ In addition, the European Commission Regulation number 1881/2006 (EC, 2006) established the thresholds of nitrates in spinach at 2500-3500 mg/kg fresh weight, lettuce at 3000-5000 mg/kg fresh weight, and lettuce type ‘Iceberg’ at 2000-2500 mg/kg fresh weight.⁵² Analytical methods for the quantification of nitrates and nitrites present in food is the colorimetric method,⁵⁷ photometric,⁵⁸ potentiometric assay,⁵⁹ Capillary electrophoresis,⁶⁰ HPLC⁶¹ with UV/DAD detection⁶² or post-column derivatisation.⁶³ It can also be determined using IC coupled with mass spectrometry⁶⁴ and flow injection analysis.⁶⁵

Oxalates: Oxalic acid $[(\text{COOH})_2]$ in combination with its salts or minerals form oxalates. Oxalic acid is present in the cell sap of many of the green leafy vegetable.⁶⁶ Depending on plant species, oxalates can occur as insoluble salts of calcium, magnesium and iron, and soluble salts of potassium and sodium or as a combination of these two forms.⁶⁷ However, oxalic acid does not interfere with zinc absorption and metabolism.⁶⁸ Insoluble oxalates are excreted in feces. Whereas, soluble oxalates affect the human body by forming a strong chelate with dietary calcium and other minerals rendering the complex unavailable for absorption and assimilation. This insoluble calcium oxalate in the crystal form is stored in the kidney causing serious health-related problem called kidney stone.⁶⁹ The adverse effect of calcium absorption is higher when the ratio of oxalate: calcium is more than 9:4.⁶⁷ Besides dietary intake, oxalates are also formed in the human body as the by-product of ascorbic acid and glyoxylate metabolism.⁷⁰ The distribution of oxalic acid is uneven in plants and varies among species. Generally, the amount of oxalate is high in

leaves followed by seeds and less in stems.⁷¹ Hence, consumption of leafy vegetables has more of a concern when there is a risk of high oxalic acid concentration. Reports show leaves of amaranth, rhubarb (*Rheum rhabarbarum*), spinach and beet (*Beta vulgaris*) have more oxalate levels than in their stems.⁶⁷ The various methods employed in the determination of oxalate in plant leaf tissues are enzymatic analysis.⁷²⁻⁷⁴ But this method of analysis in plants interferes with the carbohydrates in the plant tissue.⁷⁵ Other methods are HPLC,^{76,77} titration,⁷⁸ capillary and gas chromatography.⁷⁹

Tannins: The word tannin was coined by Seguin (1976) to mean substances present in vegetable extracts and are responsible for converting animal skin into leather products. Vegetable tannins are polyphenols with relatively high molecular weight (up to 20,000 Da) and can form complexes with carbohydrates and proteins in aqueous solutions.⁸⁰ Plant tannins are present in plant bark, wood, fruits, fruit pods, leaves, roots and plant galls.^{81,82} Gupta & Haslam⁸³ further explained that vegetable tannins are normal metabolic products and are not the products of in vitro transformation by chemical or other means. Freudenberg⁸⁴ classified tannins into two groups based on the structural types, namely; hydrolysable tannins and non-hydrolysable or condensed tannins. This is the most widely accepted classification. Of the two groups, condensed tannins are abundantly found in higher plants (e.g., proanthocyanidins). The hydrolysable tannins consist of glucose surrounded by phenolic acids and are readily hydrolyzed by acids, bases and certain enzymes. These condensed tannins are oligomers, which resist hydrolysis.⁸⁵ Vegetable tannins are water soluble phenolics compounds that are widely distributed in the plant kingdom. Tannins can produce undesirable effects in food when present in considerably high amounts. Tannins have ability to form a complex with proteins through hydrogen bonding and covalent linkages. This results in the precipitation of proteins to make it unavailable for absorption.⁸⁵ Incidence of esophagus cancer is linked with dietary tannins.^{86,87} No method has been found completely satisfactory due to the complex nature of phenolic compounds. However, Vanillin assay,⁸⁸ HPLC coupled with MS⁸⁹ and UV spectrometers⁹⁰ are some of the methods for determination of tannins.

Phytic acid: Phytic acid (i.e. myo-inositol hexaphosphoric acid) is a natural substance that acts as a major storage of phosphorus in all leafy vegetables.⁹¹ Phytic acid is found in plant tissues as salts of cations such as potassium, magnesium and calcium. Significant number of negatively charged phosphate groups in phytic acid chelate essential mineral nutrients in human body making it less available for absorption.⁹² Phytates has negative impact on the activity of digestive enzymes and act through chelation of mineral cofactors or interaction with protein. For instance, phytate interferes with zinc homeostasis. Protein binding by phytate can be direct (phytate: protein) or indirect (through a cation bridge).⁹³ Processing techniques such as boiling and cooking have no effect in reducing the level of phytic acid as the phytate is relatively heat stable.⁹⁴ However, evidence showed that although food preparation techniques help in reducing the phytic acid level, the most effective methods are lactic acid fermentation and soaking in acid medium.⁹⁵ The main method for phytic acid determination in food samples was developed by Wade & Morgan⁹⁶ after whom various methods were developed. Current methods found in the literature are complexometric titration,⁹⁷ enzymatic reaction,⁹⁸ potentiometric⁹⁹ and ionic chromatography.¹⁰⁰

Cyanogenic glycosides: Cyanogenic glycosides (CN) are derivatives of amino acids, and a group of secondary metabolites present in

more than 2500 plant species.¹⁰¹ Chemically, cyanogenic glycosides are described as the glycosides of α -hydroxynitriles. Amygdalin is the first isolated cyanogenic glycosides as a product of hydrolysis from seeds of bitter almond (*Amygdalus communis*) by Pierre Jean Robiquet and Antoine-Francois Boutron-Chalardin 1802.¹⁰² Most plants are known to produce small quantity of cyanide along with ethylene production.¹⁰³ Cyanogenic glycosides have received much research consideration because of the known poisoning nature of hydrogen cyanide (HCN). Its presence in economically important crops such as cassava, durian (*Durio zibethinus*), white clover (*Trifolium repens*) and many other cultivated plants species has been of great concern and research attention.¹⁰¹ Cassava is the staple food in many African and Latin American countries¹⁰⁴ where not only the root tubers but the leaves are also eaten as green leafy vegetable.¹² It is therefore, common to report on cassava poisoning in many of these countries. For instance, acute cassava poisoning has been reported in Nigeria since the late 1980s.¹⁰⁵ Kwashiorkor is the most serious problem prevailing in areas where cassava is largely consumed as staple food due to the presence of cyanogenic glycosides that hinders protein absorption in humans.¹⁰¹ According to Kamalu,¹⁰⁶ inhibition of Na-K-ATPase is caused by the absorption of linamarin in cassava diet. This results in electrolyte imbalance with K depletion. Some of the methods to determine cyanogenic glycosides are gas-liquid chromatography,¹⁰⁷ ultra-high-performance liquid chromatography¹⁰⁸ coupled with a mass spectrometer, a reversed-phase high-performance liquid chromatography¹⁰⁹ and HPLC.¹¹⁰

Processing methods to reduce anti nutrients

Heat treatment is the most effective method to reduce the anti-nutrition factors present in green leafy vegetables so far.¹¹¹ Cooking and blanching helps in the removal of anti nutrients through rupturing the plant cell wall followed by leaching out of soluble compounds into the blanching medium. The levels of phytic acid and oxalic acid can be effectively reduced by cooking and blanching methods.¹¹² However, this practice could also lead to the leaching out of nutritional elements.¹¹³ A study conducted on leaves of amaranth (*Amaranthus tricolor*), bathua (*Chenopodium album*), fenugreek (*Trigonella foenum grecum*) and spinach (*Spinacia oleracia*) for reducing anti nutrients reported blanching alone between 10 and 15 minutes resulted in significant reduction in phytic acid. Oxalic acid content was reduced by both blanching and cooking whereas drying and storage did not significantly affect anti-nutrition factors in green leafy vegetables.¹¹⁴⁻¹¹⁶

Conclusion

Green leafy vegetables provide vital nutrients required for human health and wellbeing. These include amino acids, vitamins, essential fatty acids, minerals and dietary fiber. It also has significant socioeconomic benefits. For instance, farmers in the tropics and subtropics, mostly women, grow and harvest green leafy vegetables to supplement household income. In rural areas, traditional leafy vegetables play important role as nutritional source, and it is available all-year. Green leafy vegetables are usually considered as the cheapest source of food for vitamins and micronutrients supplementation to combat nutrients deficiencies. It is also used as herbal and medicinal plants in various cultural and traditional settings for many different ailments. The presence of anti nutritional factors such as nitrates, oxalates, phytates, cyanogenic glycosides and tannins in green leafy vegetables can affect micronutrients absorption and thus, make the latter unavailable. Thermal processing of leafy vegetables through

boiling, cooking and blanching before consumption help in reducing the level of anti nutrients. There is a scope for research to identify and explore the potential of edible leaves of indigenous and under-exploited plants for use as food and medicine, and inclusion in mainstream agri-food systems. Research is also needed to explore different varieties and possibility of adopting agronomic practices that will reduce the concentration and effect of anti-nutritional factors in green leafy vegetables and enhance their nutritive value.

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Conflict of interest

The author declares no conflict of interest.

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