

ANTI-NUTRIENT PROFILE OF DIFFERENT CHENOPODIUM CULTIVARS LEAVES

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Abstract

Chenopodium album belongs to family Amaranthaceae commonly known as pigweed or goose foot or bathua (in Hindi) is a weedy plant found all over world and has gained renowned popularity these days due to its high nutritional composition especially amino acids. In the present study the leaves of four *Chenopodium album* cultivars were analysed for the anti-nutritional composition viz. tannins, simple phenols, total phenols, phytic acid, oxalates, flavonoids, alkaloids, Trypsin Inhibitor Activity (TIA), phytic acid and phytate phosphorus using standard methods. The results of present investigation showed that the maximum total phenol, simple phenol and tannin content were in *Ec*₁ cultivar i.e. 304.98, 101.007 and 203.91 mg GAE/100g respectively whereas minimum was present in *Ec*₂ cultivar i.e. 224.99, 72.50 and 152.49 mg GAE/100g respectively. The TIA content ranged between 0.11-0.17 TIU/mg whereas the saponin content ranged between 0.027-0.867g/100g. The highest TIA was present in *Ec*₂ cultivar whereas the saponin content was highest in *Ec*₁ cultivar. The highest phytic acid content was present in *Ec*₂ cultivar (268.33mg/100g) whereas lowest in *Ec*₁ cultivar (238.33mg/100g). Similarly the maximum phytate phosphorus content was present in *Ec*₂ cultivar and minimum in *Ec*₁ cultivar. The alkaloid content in *Chenopodium* cultivar leaves ranged between 1.27-1.67mg/100g whereas flavonoid content ranged between 220.0-406.67mg/100g respectively. The maximum oxalate content was there in *Ic*₁ cultivar (518.42mg/100g) and minimum in *Ec*₁ cultivar (394.19 mg/100g) respectively. Certain anti-nutrients among these besides being an anti-nutrient also exert positive health effect especially phenolic compounds, flavonoids, alkaloids thus can also utilized for pharmaceutical purpose.

Keywords: *Chenopodium album*, *Bathua*, anti-nutrients, underutilized plants, leafy vegetables

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1. INTRODUCTION

Green leafy vegetables play significant role in human nutrition especially as a source of vitamins, minerals and dietary fiber. The varieties of leafy vegetables are diverse, ranging from leaves of annuals and shrubs to tree leaves. However the utility of the leaves, pods and edible twigs of shrubs and trees as food is limited due to the presence of anti-nutritional factors. Anti-nutritional factors in plants seem to be as a way of storing nutrient or as a mean of defending their structure and reproductive elements (Harborne, 1989). The anti-nutritional factors which have been implicated in limiting the utilization of shrubs and tree forages include non protein amino acids, glycosides, alkaloids, triterpenes, oxalic acid and polyphenolics.

The genus *Chenopodium* comprises about 250 species which include herbaceous, suffrutescent and arborescent perennials, although most species are colonizing annuals. They have been contributed for centuries as leafy vegetables as well as an important grain crop for human and animal foodstuff due to high protein and a balanced amino-acid spectrum (Bhargava *et al.*, 2006). *Chenopodium album* also known as fat hen or lambs quarter or pigweed is a fast growing weedy annual plant in the genus *Chenopodium* and belongs to the family Amaranthaceae. The leaves and seeds of all the members of this genus are edible and are consumed in cooked form mainly in combination with other species. Like other green leafy vegetables the main problem in the consumption of the leaves of *Chenopodium album* is the presence of anti-nutritional factors. These factors may have

adverse effects on the health through inhibition of protein digestion, growth, iron and zinc absorption (Larsson *et al.* 1996). So, this paper reports the anti-nutritional composition of the leaves of various *Chenopodium album* cultivars.

2. MATERIAL AND METHODS

2.1 Procurement of Materials

The four cultivars of *Chenopodium album* seeds (two indigenous and two exotic) viz. IC 415477 (Ic₁), IC 107299 (Ic₂), EC 507742 (Ec₁) and EC 507733 (Ec₂) were procured from National Bureau of Plant Genetic Resources (NBPGR), Regional Station, Shimla (H.P.) India and the leaves were planted in different concrete pots during the month of December and were and were brought to the laboratory after harvesting from those pots during the month of March. In the laboratory the leaves were sorted manually and washed with double distilled water to remove adhering dirt and dust, the leaves were dried at 40±5°C and were grounded in a mixer and stored in airtight containers for further analysis. The entire chemicals used in the present investigation were of analytical grade. The various methods followed for the determination of various anti-nutrients are given below.

2.2 Anti-nutritional Analysis: The tannins were determined by the method given by Makkar *et al.* (1993) with the determination of total and simple phenols using folin ciocalteau reagent and absorbance was read at 725nm using tannic acid (0.1mg/ml) for standard curve. Trypsin inhibitor activity was estimated by the modified method of Ray and Rao (1971). The extraction was carried out using 25ml of 0.05M phosphate buffer (pH 7.0) and the TCA soluble proteins were determined by the method of Lowry *et al.* (1951). Whereas, the saponin content in leaves samples was determined by the method given by Obadoni and Ochuko (2001). The phytic acid was determined following Haugh and Lantzech (1983) method. Alkaloids in the leaves samples was estimated by the method given by

Harborne (1973) with little modification by Obadoni and Ochuko [7]. The flavonoid in the sample of various *Chenopodium* cultivars leaves was determined by the method given by Boham and Kocipia (1994) using 80% aqueous methanol. However, the method of Abaza *et al.* (1968) was followed to determine oxalate content.

3. RESULTS AND DISCUSSION

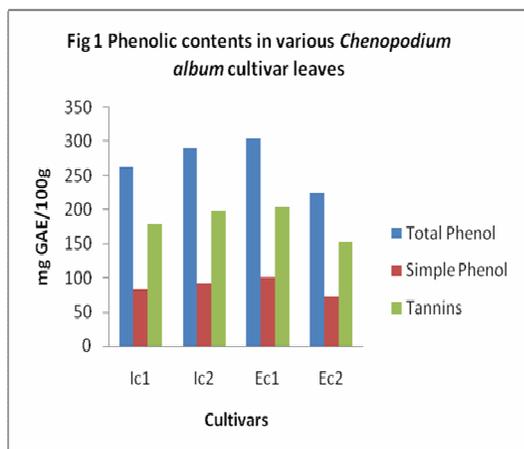
3.1 Total Phenols

Fig 1 shows the total phenol content present in leaves of various *Chenopodium* cultivars. It is clear that significantly higher total phenol was present in leaves of Ec₁ cultivar (304.98 mg GAE/100g) and minimum in leaves of Ec₂ cultivar (224.99mg GAE/100g). Kaur and Kapoor (2002) reported 253.5 mg GAE/100g total phenol content in the leaves of *Chenopodium album*. Phenols exhibit antioxidant potential (Awika *et al.* 2003) due to their redox properties which allow them to act as reducing agents, hydrogen donors and single oxygen quenchers (Chang *et al.* 2001). Plant phenolics are biosynthesized following different routes, the shikimic acid pathway being the most biosynthetic route involved. This pathway as reported by Rivero *et al.* (2001) is thought to be an acclimatization mechanism of plant external stress (Temperature, injury, infection etc.) the cold having the highest phenylalanine ammonia-lyase activating effect and the highest peroxidase and polyphenol oxidase inhibiting effect. The result of present investigation are in accordance with other workers however slight variation in result might be due to differences in varieties or species, agro-climatic conditions and extraction or analytical procedure applied to determine the content.

3.2 Simple Phenols

Simple phenols consist of a singly substituted phenolic ring with either alcoholic, aldehydic or carboxylic acid groups. As is evident from the Fig 1 significantly higher simple phenol content was present in Ec₁ cultivars leaves. The variation in various cultivars might be due to genetic factors and varietal dependent variability. A broad range of phenolic compounds occur in the food products especially those coming from the plant material, in which they contribute to the organoleptic properties i.e. astringency, beer hazes, specific colouration and off-flavour. Moreover the

effects of the dietary phenolic compounds are of great interest due to their anti-oxidative, cardiovascular protective, anti-allergic, anti-inflammatory, antiviral and anti-carcinogenic activities (Landbo and Meyer, 2001). The results of present investigation suggested that phenolics are an important constituent of this plant and their pharmacological effects could be attributed to the presence of these valuable constituents.



3.3 Tannins

As is clear from Fig 1 significantly highest tannin content was present in Ec₁ cultivar and lowest in Ec₂ cultivar. The variation in tannin content in different cultivars might be due to varietal and genetic variability. Tannins are basically polyphenolic compounds having complex mixture and are present in many plants. They form complexes with proteins, starches and digestive enzymes thereby reducing the nutritional value of foods (Serrano *et al.* 2009) and causing growth depression. They also interfere with protein absorption and reduce iron availability. Kumar and Kumar (2008) reported that the leaves of *Chenopodium album* contained various anti-nutrients like alkaloids, flavonoids, glycosides, saponins and tannins. Besides exhibiting negative effects tannin also provide some health benefits including antioxidant and radical scavenging properties, anti-carcinogenic, anti-bacterial and anti-enzymatic effects (Vattem *et al.* 2005) thus has gained popularity in today's scenario.

3.4 Trypsin Inhibitor Activity (TIA)

Data in Table 1 reveals the TIA present in leaves of various *Chenopodium* cultivars. It is clear from the data maximum TIA content was present in leaves of Ec₂ cultivar (0.17 TIU/mg) and minimum in Ic₂ cultivar

(0.11 TIU/mg). Gupta and Wagle (1988) found 0.91 TIU/mg TIA content in leaves of *Chenopodium album*. TIA is an anti-nutritional factor present in plant foods which is responsible for reduced protein digestibility in human system. Leafy vegetables contained significant level of trypsin and chymotrypsin inhibitors that impairs the utilization of proteins and the amino acids present (Glew *et al.*, 2005) by interacting with proteolytic enzymes rendering them unavailable for protein digestion. The results of present investigation are lower than that of Gupta and Wagle which might be due to varietal differences, season, agro-climatic condition and agro-technical procedures. In addition the comparison of results from different studies can be difficult due to variability in the experimental conditions amongst the method used (Stratil *et al.* 2006).

3.5 Saponin

As is evident from the Table 1 the highest saponin content was present in leaves of Ec₁ cultivar followed by leaves of Ic₁ and Ic₂ cultivar and lowest in leaves of Ec₂ cultivar. Saponins are nitrogen free glycosides each consisting of a sapogenin and a sugar. The sapogenin may be a steroid or a triterpene and the sugar is generally glucose, galactose, pentose or methyl pentose. They impart bitter taste and tend to foam in aqueous solution. Saponins are poorly absorbed and most of their effects are attributable to their hydrophilic/ hydrophobic asymmetry and consequently their capacity to reduce interfacial tension (Champ, 2002). Gupta and Wagle (1988) reported 0.90 per cent saponin content in leaves of *Chenopodium album*. This variation in result from other worker might be due to different varieties which lead to genetic variability, time of harvest, stage of harvest, analytical procedure applied and climatic conditions. Bhargava *et al.* [2] revealed that saponin content in leaves of quinoa vary in different growing stages, low saponin content was found in branching stage and higher in blooming stage. From the nutritional or pharmacological point of view saponins could also have some value. They can increase membrane permeability thus enabling use for increased food intake at the intestinal level or even for drug assimilation (Gee *et al.* 1993). Stuardo and San Martin (2008) reported antifungal activity of *Chenopodium* saponins due to its capacity to associate with steroids of

fungal membranes causing damage to its integrity and pore formation

3.6 Phytic Acid

Phytic acid (hexa phosphate of inositol) is a complex class of naturally occurring anti-nutritional factor present in plant foods and is a major phosphorus storage compound in green leafy vegetables. It affects the digestion and absorption of minerals. The data on phytic acid content present in leaves of various *Chenopodium* cultivars is given in Table 1. The phytic acid content in different *Chenopodium* cultivars ranged from 238.33-268.33 mg/100g. A significant ($P \leq 0.05$) difference was there in phytic acid content when different cultivars were compared with each other. Yadav and Sehgal (2003) reported 234.50 mg/100g phytic acid content in *bathua* (*C album*) leaves. The results of present investigation are slightly higher than that of other worker might be due to variability in the experimental conditions amongst the method used, variation in varieties, environmental conditions and genetic makeup of the crops. High dietary phytate content is reported to cause growth reduction affect food value by binding and making mineral ions unavailable to the consumer affect the homeostasis of zinc and iron, inhibit enzymatic digestion of proteins by forming complexes with enzyme protein (Marfo *et al.* 1990). Phytic acid also exhibit certain beneficial effects due to its metal chelating abilities the lower inositol's involvement in signaling pathways and to their phosphate donor/acceptor capabilities. Lower inositol phosphates can act as antioxidants by inhibiting iron mediated oxidative reactions, enhancing immunity by increasing natural cell function and activity and stimulating bacterial killing by neutrophils (Bohn *et al.* 2008).

3.7 Phytate Phosphorus

Phytate phosphorus is relatively unavailable phosphorus that exists in plants as calcium, magnesium and potassium salt of phytic acid. Data in Table 1 reveals the phytate phosphorus content present in leaves of *Chenopodium* cultivars. It is clear from the data maximum phytate phosphorus content was found in leaves of Ec_2 cultivar (75.62mg/100g) and minimum in Ec_1 cultivar (67.16mg/100g). A significant ($P \leq 0.05$) difference was observed in

phytate phosphorus content when different cultivars were compared with each other. The variation in different cultivars might be due to the varietal and genetic differences. According to Lopez *et al.* (2002) phytate is the main storage compound in plants and can account for 80 per cent of total phosphorus in form of phytate phosphorus. The various processing techniques like soaking, germination, cooking, blanching etc. help in reduction of phytate phosphorus content and thus increase the availability of phosphorus.

Table 1 Anti-nutritional factors of *Chenopodium album* (*Bathua*) Cultivars Leaves

Parameters /Cultivars	Ic ₁	Ic ₂	Ec ₁	Ec ₂	$p \leq 0.05$
TIA (TIU/mg)	0.12	0.11	0.12	0.17	0.01
Saponin (g/100g)	0.04	0.033	0.867	0.027	0.056
Phytic Acid (mg/100g)	245.0	256.6	238.3	268.3	3.33
Phytate Phosphorus (mg/100g)	69.0	72.32	67.16	75.62	0.94
Alkaloids (mg/100g)	1.33	1.27	1.67	1.53	0.69
Flavonoids (mg/100g)	220.0	343.3	406.6	263.3	21.05
Oxalates (mg/100g)	518.0	477.0	394.1	423.8	9.51

Each value is average of three replications

3.8 Alkaloid

Alkaloids are a class of naturally occurring organic nitrogen containing bases. Most common alkaloids include morphine, strychnine, quinine, ephedrine and nicotine. The bitterness of some plants might be due to the presence of alkaloids which affect the consumer acceptability of product. These compounds also possess medicinal properties. Data in Table 1 shows the alkaloid content present in Ic_1 , Ic_2 , Ec_1 and Ec_2 cultivar leaves as 1.33, 1.27, 1.67 and 1.53 mg/100g respectively. A non-significant difference was there in alkaloid content when various *Chenopodium* cultivars were compared with each other. Adedapo *et al.* (2011) reported the alkaloid content in leaves of *C album* as 1.8 mg/100g. The results of present investigation are slightly lower than that reported by

Adedapo et al. which might be due to varietal and agro-climatic variations.

3.9 Flavonoids

Flavonoids are a group of polyphenolic compounds naturally present in most edible fruits and vegetable plants. They are secondary plant metabolite and are a natural antioxidant which helps to maintain the body's health and to protect against diseases. They also exhibit anti-nutritional properties due to their metal chelating properties. However epidemiological and clinical studies have provided evidence of a potential role for flavonoids in lowering the risk of coronary heart diseases prevention, cardiovascular diseases, alzheimers disease, neurodegenerative diseases, diabetes, osteoporosis and lung cancer (Lampila *et al.*, 2009) through antioxidative action and / or the modulation of several protein functions. However nutritionally flavonoid is considered as an anti-nutrient as its polyphenol structure is a metal chelator therefore capable of binding iron and facilitating its removal from the body. A glance of data in Table 1 reveals the flavonoid content present in leaves of *Chenopodium* cultivars. As is clear from the table highest flavonoid content was present in Ec₁ cultivar (406.67 mg/100g) and lowest in Ic₁ cultivar (220.0 mg/100g). A significant ($P \leq 0.05$) difference was observed in flavonoid content when Ic₁, Ic₂, Ec₁ and Ec₂ cultivars were compared with each other. Rahiminejad and Gornall (2004) reported that the leaves of *Chenopodium album* contain flavonoids in higher amounts mainly the 3-O-glycosides of quercetin, kaempferol and isohamnetin. Adedapo *et al.* found 182 mg/100g flavonoid content in *C. album* leaves. The difference in flavonoid content might be due to genetic variability, varietal differences, agro-climatic variation, soil topography and analytical method.

3.10 Oxalates

Oxalates is a dicarboxylic acid and is found in the form of soluble salts of potassium and sodium and as insoluble salts of calcium, magnesium and iron in algae, fungi, lichens, ferns and higher plants. Insoluble oxalate is excreted in the feces while the soluble oxalate is absorbed by the body. Soluble oxalate forms strong

chelates with dietary calcium, rendering it unavailable for absorption and assimilation. As is evident from the Table 1 the significantly higher oxalate content was present in Ic₁ cultivar (518.42 mg/100g) and lower in Ec₁ cultivar leaves (394.19 mg/100g). According to Guil *et al.* (1996) the leaves of *Chenopodium album* (goosefoot) contained oxalic acid with a range value of 360-2000 mg/100g. The results are at par with other worker however the slight differences in degree of accumulation of oxalates might be related to species, the plant part, age of plant and the agro-climatic conditions. However, high dietary intake of soluble oxalate can lead to the formation of kidney stones. A diet high in oxalates may require supplementation of divalent minerals to prevent deficiencies. Addition of a source of calcium to vegetables containing high levels of soluble oxalate has been shown to reduce the intestinal available oxalate content in such food (Radek and Savage, 2008).

4. CONCLUSION

The results of the present study revealed that the leaves of *Chenopodium album* contained total phenol, simple phenol and total phenol which act as an anti-nutrient by binding minerals but besides that they also exert antioxidant potential. The trypsin inhibitor activity was also present in lesser amount in different *Chenopodium album* cultivars thus making more of the proteins available to the body. The saponin and phytic acid content was also present in lesser amount. The presence of flavonoids and alkaloids were also shown in the leaves. They also exhibit antioxidant potential besides being an anti-nutrient as reported by other workers. However further studies are required in this context to study the effect of various processing techniques on these anti-nutrients. These investigations would provide a more complete picture of the nutritional significance of *Chenopodium album*.

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