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Evaluation of Anti-nutritional Compounds in Selected Wild Plants Consumed by Ruminants in Pasturelands

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Abstract: *The present study was designed to determine the anti-nutritional compounds in the wild plants of Soone valley Punjab, Pakistan. A wide range of anti-nutritional factors exists in the wild plants of this area which are consumed as forages by the ruminants. Few anti-nutritional compounds i.e. oxalate, phytate, saponins and tannins were analyzed during the course of study. Oxalate content ranged from 0.4467% to 0.6267%. The highest oxalate content was found in Chenopodium album and lowest oxalate content was available in Mentha arvensis. The content of phytate ranged from 3.8167% to 4.9767% in all wild forages. The maximum amount was observed in Buxus papillosa and minimum amount was found in Ahadota vasica. Saponins ranged from 2.2700% to 3.7833%. The percentage of tannins varies from 1.3167% to 1.6300% in all plant species. The optimum value of tannins was found in Adatoda vasica and low value of tannins was investigated in the Mentha arvensis among the investigated plant species. Overall, the maximum factors observed in Buxus papillosa and lowest in Mentha arvensis plants. The values estimated in the present study are below the mark as compared to the toxic levels. Nutritional and health issues are developed due to the large consumption of monotypic wild edible plant parts during one meal. However, anti-nutritional compounds related to respective risks are less with the use of traditional methods.*

Keywords: *Anti-nutritional compounds, wild forages, oxalate, phytate, saponins, tannins*

1.Introduction

Anti-Nutritive Factors (ANF) are compounds which might be produced through more than one mechanisms and have an effect on the nutrient applications. Digestion and assimilation of nutrients in grazers is affected by these factors, hence the name anti-nutrient. The dangerous effects of anti-nutritive compounds are nullified in some ruminants by their decomposition in rumen of grazing animals [1]. About 700 species of wild plants are used to meet the food requirements of human beings globally [2]. Some of these wild plants have been domesticated due to their role in society and agriculture [3]. Wild plants contain a variety of chemicals some which interfere with the normal physiological activity of animals and human beings [4, 5]. Basic metabolism of living organisms are hugely affected by these elements [6] Animals feeding on such plants experience compromised immunity, reduction in growth and reproductive potential [7]. These ailments cause loss of weight in ruminants. Some animals have developed strategies for counteracting the damage caused by these chemicals [8]. Some of the major anti-nutritional compounds are saponins, tannins, phytic acids, oxalates, cynogenic glycosides, gossypol, goitrogens and lectins [9]. High cellulose and non-starch polysaccharides are also considered as anti-nutritional factors for monogastric animals [10]. This study aimed to assess the anti-nutritional compounds in the wild forages and their effects on grazers of the area.

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2.Materials and methods

Study area

The current sampling was carried out in Soone valley, also known as Soone Sakesar, located in Punjab Pakistan. Geographic parallel for immediate access to the region of Soone valley is given below.

Figure 1. Map of study area

Sample collection

Six important wild plants i.e. *Buxus papilosa, Adhatoda vaisca, Dodonaea viscosa, Chenopodium album, Mentha arvensis and Lactuca virosa* were identified and collected from four distinct sites (Nushera, khabeki, uchali, kufri) of Soone valley, District Khushab, Punjab, Pakistan. Six wild plants were compiled after collection from various location sites of Soone valley having promising potential forages. Three replicates of each plant were randomly collected from all selected sites of the area. Plant material particularly leaves were carefully collected using knives and diggers. Collected samples were stored in tagged paper bags. Plants were washed with distilled water to clear the impurities and dust particles. Washed samples were air dried and then oven dried for 4-7 days at 75^0C.

Table 1. List of collected wild plants with Scientific, Common and Family name

Scientific Name	Common Name	Family
Adhatoda vasica	Adusa	*Acanthaceae*
Buxus papillosa	Box wood	*Buxaceae*
Mentha arvensis	Wild mint	*Lamiaceae*
Lactuca virosa	Wild opium	*Asteraceae*
Chenopodium album	Bathu	*Amaranthaceae*
Dodonaea viscosa	Soapwood,hop wood	*Sapindaceae*

Plant Sample Identification

Identification of samples was done under expert supervision in Department of Botany, University of Sargodha.

Determination of Phytate

To investigate the phytic acid percentage in wild plants, method proposed by Wheeler and Ferrel [11] was applied. Standard curve of different Fe $(NO_3)_3$ concentrations was plotted along the readings on spectrophotometer. Phytate phosphorus was determined by the concentration of ferric ion assuming a molar ratio of 4:6 between iron and phosphorous.

Oxalate detection

For the determination of oxalates in the collected samples, titration method [12] was used. 1 g powdered samples was heated on a water bath with 30 mL of 0.25 N HCl for 1 hour. The sample was filtered after cooling and 10 mL of filtrate was added to a centrifugation tube and kept for 10 min at 5^{o}C. 2 mL of saturated acetic acid solution of anhydrous sodium acetate was added and kept the tube undisturbed overnight for precipitation of oxalates. Mixture was centrifuged at 5000 rpm and supernatant was drained. The precipitates were again dissolved in 5 mL 0.25 N HCl and the procedure was repeated for re-precipitation of oxalates. After washing with ammonia-alcohol mixture, precipitates were dried at 100^{o}C for 30 min. For measurement of oxalates, precipitates were dissolved in 5 mL of 2 N H_2SO_4 and the solution was titrated against 0.02 N $KMnO_4$. Quantification was done by the given formula:

Total Oxalates (%) = titration volume (mL) x 1.8001

Tannin detection

Preparations of standard curves

A mixture was prepared by adding 1 mL standard solution, 1 mL Na_2CO_3 and 0.5 mL of Folin-Denis reagent. Distilled water was added to make a final volume of 10 mL. The solution was kept undisturbed for 30 min and absorbance was read at 760 nm [13, 14].

Extraction of tannins

1 g of finely ground sample was mixed with 75 mL distilled water in a conical flask for heating. After removing from heat, the sample was centrifuged at 2000 rpm for 20 min. The supernatant was collected and diluted to make a final volume of 100 mL. 1 mL of this solution was added to 75 mL water, 10 mL Na_2CO_3 and 5 mL Folin-Denis reagent and shaken well. The absorbance of mixture was read at 700 nm.

Determination of Saponins

The capacity of sample for creating foam is considered as an indicator of presence of saponins. 50% aqueous methanolic extract of the sample was agitated strenuously in the test tube. Constant foaming on the surface of solution indicates the occurrence of saponins [15].

Statistical analysis

The SPSS (Statistical Package for the Social Sciences) software version 20 was used for statistical anlysis. The one-way analysis of variance (ANOVA) was accomplished between assorted samples. The important means of possibility levels at 0.001, 0.01 and 0.05 were investigated [16]

3.Results and discussions

Oxalates

All sampled plants exhibited highly significant ($P<0.001$) variations among oxalate concentrations (Table 2). Oxalate values in wild plants ranged from 0.44 to 0.62% (Figure 2). Currently investigated oxalate percentages were higher than those reported by Amata and Iwelu [15] and Owolabi *et al.* [17] and lower when compared with the values suggested by Khan *et al.* [18]. Phytate, polyphenol and

oxalate are commonly affected by the accessibility of nutrients substances. The complexes of oxalate formed with bivalent ions like Ca^{2+}, Zn^{2+}, Mg^{2+} and Fe^{2+} ions are inaccessible owing to their insolubility [19].

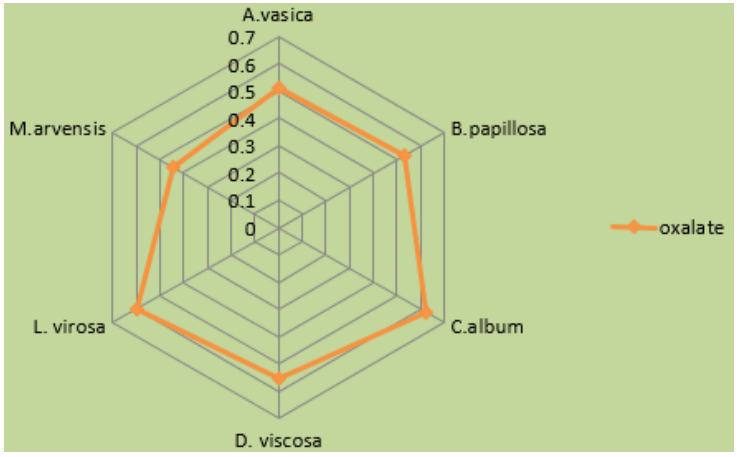

Figure 2. Oxalate level fluctuations in different wild plant species

Table 2. Analysis of variance for anti-nutrients of different wild plant species

SOV	Df	Oxalate	Phytate	Saponin	Tannin
Plants	5	0.01253***	0.63124***	0.63124***	0.04822***
Error	12	0.00157	0.08393	0.08393	0.00211

***=significant at 0.001 levels

Phytate

Phytate concentrations in wild plants were significantly (P<0.001) variant as unveiled by the analysis of variance (Table 2). The range of phytate concentration was from 3.8167% to 4.9767% (Figure 3). Phytate values reported in the current study were lower than the values reported by Okon and Akpanyoung [20]. On the contrary, values obtained by this investigation were higher in comparison with the values of Ogbe and Affiku [21] and Prohp et al. [22]. Phytate is the chief phosphorus storage compound in plants. The ability of animals to intake minerals is inversely related to the concentration of phytate in plants. As the phytate cotent increases, mineral uptake declines [23]. Phytate causes increase in the mineral deficiency in digestive tract of animals by binding with the mineral ions such as iron, magnesium, calcium and zinc, and hinders the digestion of protein by the formation of phytate-protein complexes [24].

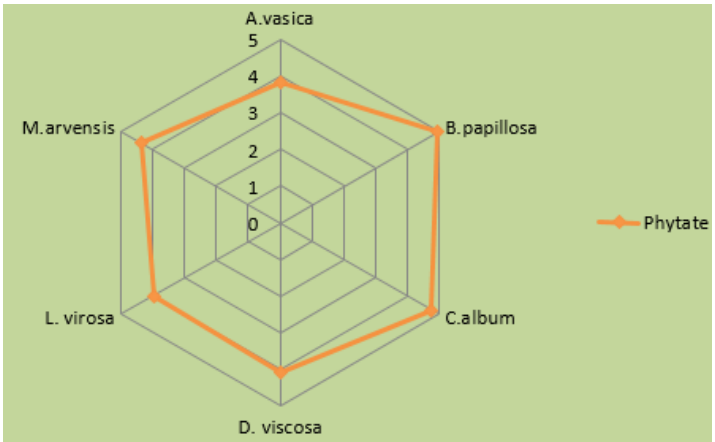

Figure 3. Phytate level fluctuations in different wild plant species

Saponins

Analysis of variance showed highly significant (P<0.001) variations among saponin content of the wild forages (Table 2). Saponins ranged from 2.2700% to 3.7833% in the given samples (Figure 4). The present work reported lower values of saponins in the wild plants samples than the values (7.22%) reported by Uadia et al. [25], while these values were higher than those suggested by Owolabi *et al.* [17] and Ezeabara et al. [26]. Saponins have been found to be potentially useful for the treatment of hypercholesterolemia which indicated that saponin might be acting by interfering with intestinal absorption of cholesterol, thus have antidiabetic effects [27]. Precipitation, coagulating red blood cells, cholesterol binding, formation of foams in aqueous solution and hemolytic activity is reduced by the feed rich in saponins [28]. High content of saponins in food plants impart bitter taste and astringent properties, thus less preferred for intake by grazers. High quantity of saponins in diet causes many acute disorders such as reduction in bioaccessibility of nutrients and enzyme activity. Digestion of proteins is also impaired by higher levels of saponins by inhibition of various digestive enzymes like trypsin and chymotrypsin [29].

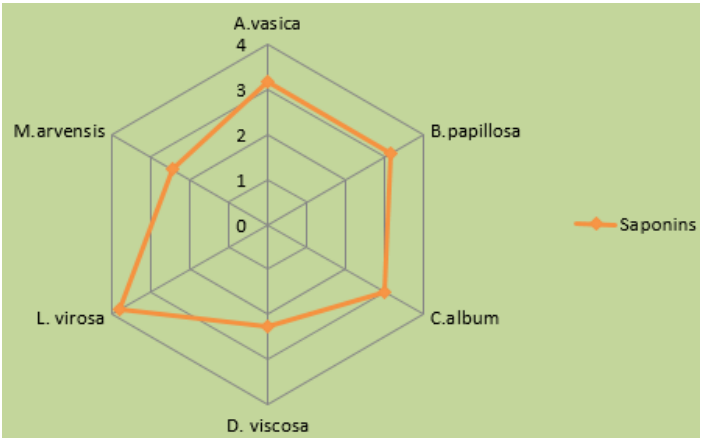

Figure 4. Saponin level fluctuations in different wild plant species

Tannins

Tannin shows highly significant (P<0.001) variation among all plant species as revealed by analysis of variance. The values of tannins were between the range of 1.3167% to 1.6300% (Figure 5). The current research suggested that tannin values were lower in wild plant samples than the findings of Uadia et al. [25] and Friday et al. [30] and lower when compared with the findings of Khan et al. [18]. Tannin is composed of polyphenolic compounds and has astringent quality [31]. Tannin rich diet can lead to intestinal digestion depression in animals [32] as it interferes with dietary iron absorption and adversely affected the quality of proteins in food. Protein metabolism is affected by tannins owing to their capacity to bind with various enzymes like chymotrypsin, trypsin, lipase and amylase [33].

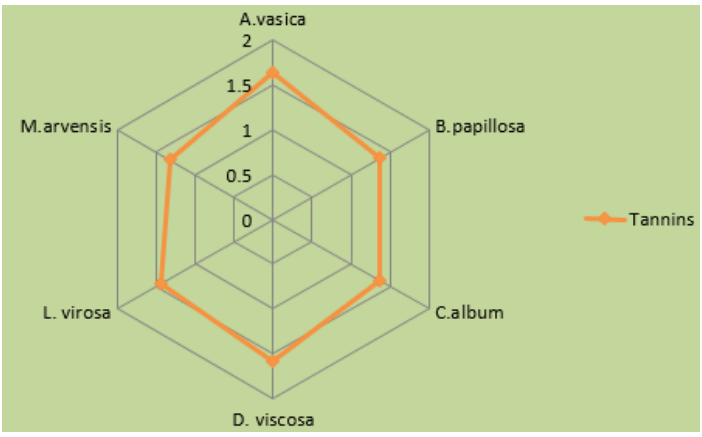

Figure 5. Tannins level fluctuations in different wild plant species

4.Conclusions

The evaluation of anti-nutritional components of the wild plants forages depicted the occurrence of a wide range of these compounds in the sampled plants. Most of the sampled plants are replete with anti-nutritional compounds. Although variations occur in their concentration, yet all of them are present in these forages in fair amount. As the ruminants of study area feed on these plants, higher values of anti-nutrients can lead to toxicities and impairments in grazers thus leading to reduction in their productivity.

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