Range Mgmt. & Agroforestry 40 (1) : 143-149, 2019
ISSN 0971-2070

Seasonal alterations in anti-nutritional factors of common fodder trees of mid Himalayas

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Received: 15th January, 2018 Received: 12th April, 2019

Abstract

The present study was conducted on the foliages of eight different fodder trees of mid Himalayan region round the year at different intervals to assess the seasonal variations in anti-nutritional factors and to find the best lopping time for these fodder trees. *Albizia chinensis* exhibited significantly higher (P<0.05) level of total phenols (13.9%) during the month of July while it was minimum (P<0.05) during November in *Grewia oppsitifolia* (3.1%). In *Albizia chinensis* foliage, simple phenols were significantly higher (P<0.05) during the month of July (4.7%), while *Bambusa arundinacea* (1.1%) exhibited significantly lower (P<0.05) simple phenol content during the month of September. *Albizia chinensis* foliage had significantly higher (P<0.05) level of net tannins during May (9.3%), whereas *Grewia oppsitifolia* revealed significantly lower (P<0.05) net tannin content (1.4%) during the month of November. *Grewia oppsitifolia* foliage exhibited minimum condensed tannin (0.7%) during November, whereas higher level of condensed tannins was observed during July in *Albizia chinensis* (4.4%). The hydrolyzed tannins were higher (P<0.05) during September (5.1%) in *Albizia chinensis* foliage, while least content (0.7%) was recorded during November in *Grewia oppsitifolia*. It was observed that the level of different anti-nutrients decreases in advancing months of the year. The study revealed that *Grewia oppsitifolia* and *Bauhinia variegata* are important fodder trees of the region with low levels of anti-nutrients for long spell of the year and could constitute as supplementary fodder resource for mitigating the forage scarcity for the livestock during lean seasons of the year.

Keywords: Anti-nutrients, Fodder trees, Tannins, Tree foliage

Introduction

Tree foliage is known to be valuable alternative green forage source for grazing animals (Parissi *et al.,* 2018). As in many other regions of the world, farmers in the hill region of Indian Himalayas depend largely on tree fodder for sustaining their livestock especially during pronounced dry periods when other feed resources have been exhausted (Katoch, 2009; Verma *et al.,* 2015; Kaushal *et al.,* 2016; Sahoo *et al.,* 2016; Nag *et al.,* 2017; Katoch *et al.,* 2017). The leaves of fodder trees have potential for alleviating feed shortages and nutritional deficiencies for livestock production (Kamalak *et al.,* 2004; Nautiyal *et al.,* 2017; Nabi *et al.,* 2017). As compared to conventional forage grasses, tree foliage have good proportions of nutrients which keep intestinal microflora active for digesting cellulosic biomasses (Katoch *et al.*, 2012; Singh and Todaria, 2012). Moreover, they can also help to bridge over the gap between demand and supply of nutrients in available feeds. The tree foliage contributes adequate amount of vitamins, minerals and dietary nitrogen to complement deficiencies in the basal feed resources (Katoch *et al.,* 2013). Total fodder availability in Himalayan region has indicated a deficit of about 50% and a part of it is supplemented through tree foliage (Verma and Mishra, 1989). The quality of any forage material depends to a major extent on the presence or absence of anti-nutritional factors. Anti-nutrient factors are those substances generated in the natural feedstuffs by the normal metabolism of species, which exert effects contrary to optimum nutrition (Aganga and Tshwenyane, 2003; Allen and Segarra, 2001). The level of different anti-nutrient factors in forage varies appreciably with the seasonal and environmental conditions prevailing in that area. Differences in anti-nutrient contents also depend on individual plant species. Seasonal variations affect forage nutrient composition and this affect livestock performance (Aganga and Tshwenyane, 2004; Adesogan *et al.*, 2006). The constraints posed by dry season on continual availability of feeding materials in the mid Himalayan region, has been a matter of concern in livestock production. Dry season is a key factor that predisposes ruminants fed on standing hay and low protein forages and leads to consistent weight loss (Babayemi and Bamikole, 2006). Blakely and Blade (1994) asserted that the climate of an area affects the nutrient composition of the forage species growing in that area. Therefore, seasonal variations have been greatly emphasized as the main determinants of forage quality. The nutritional composition of common fodder trees has been also been reviewed earlier by Katoch (2009). Nutritional value of some fodder tree species has been extensively studied, but there is still scanty information of the effect of season on the concentrations of anti-nutrient components of forage trees commonly found in mid Himalayan region. Looking into the fodder deficit of this region where large numbers of fodder tree species are growing, it is important to study anti-nutritional composition in their foliage and optimum lopping time to supplement the fodder demand of animals in the hill region and to select a potentially suitable fodder tree species to be fed at a particular season.

Materials and Methods

Experimental design and laboratory analysis: The study on the effect of seasonal variation in anti-nutritional factors was carried out in eight fodder trees prevalent in mid-hill region of Himachal Pradesh. Foliage of Kachnar (*Bauhinia variegata*), Deon (*Arratocarpus lakoocha*), Bamboo (*Bambusa arundinacea*), Khirak (*Celtis australis*), Toot (*Morus alba*), Siras *(Albizia lebbek),* Biul (*Grewia oppsitifolia*) and Oee (*Albizia chinensis*) were sampled periodically at one month interval during the month of May, July, September, November, January and March. The samples were air dried and ground in the Willey mill to pass through the mesh of 2 mm. They were packed in the air tight bags for analysis of anti-nutrients composition. Total phenolics, simple phenols were estimated using Folin-Ciocalteu reagent using tannic acid as a standard (Makkar, 2003) and net tannins were calculated by difference between total phenols and simple phenols. Total tannins content in tree leaves was estimated as per Makkar *et al.* (1993). Condensed tannins were determined by using Butanol-HCl (Porter *et al.,* 1986), whereas hydrolysable tannins were calculated by subtracting the condensed tannins from total tannins.

Statistical analysis: The data were analyzed statistically adopting the procedure described by Gomez and Gomez (1984). Wherever necessary, the percentage values were transformed to angular (arc sine) values, before carrying out the statistical analysis. The student's t-test was used to determine the significance level between anti-nutritional constituents in different fodder trees and lopping months, where P<0.05 was considered significant. The simple t-test was performed by using the following formula:

$$\text{Student's t-test} = \frac{X_d}{SE\,(X_d)} \quad \text{(at n - 1d.f.)}$$

where, X_d = mean-difference between two sets of related samples; SE (X_d)= Standard error of mean difference; n = Number of related samples

Results and Discussion

Variation in anti-nutrient contents of fodder trees during different months: The seasonal variations revealed significant impact on anti-nutritional constituents in different fodder trees under study (Table 1-5). The findings on the variations in the level of different anti-nutritional factors are discussed below.

Total phenolic content: During the month of May, foliage of *Bauhinia variegata* had significantly higher (P<0.05) level of total phenols (7.80%), thereafter, in the subsequent months from July to November there was a significant decline in the total phenolic content from 7.00 to 4.00% (Table 1). *Grewia oppositifolia* foliage exhibited significantly higher (P<0.05) level of total phenols during the month of May (4.80%) and July (5.60%). Significant reduction (P< 0.05) in total phenols was observed from September to November. From the month of May to July, *Albizia lebbek, Celtis australis, Morus alba* and *Albizia chinensis* foliage revealed significantly higher (P<0.05) level of total phenols. The similar trend for decrease in total phenolic content was observed in the month of November for *Albizia lebbek* and *Albizia chinensis* (7.00 and 8.60%, respectively) and during January for *Celtis australis* and *Morus alba* foliage (5.20 and 6.30%, respectively). During months of March and July, *Bambusa arundinacea* had significantly higher (P<0.05) level of total phenols (7.00 and 7.10%, respectively), however there was a significant reduction (P<0.05) in total phenolic content during the month of September and November (5.00 and 5.10%, respectively). A significantly lower (P<0.05) level of total phenols was observed in *Arrtocarpus lakoocha* foliage (7.30%) during November which subsequently increased from the month of January to March (9.20 to 9.30%). Rana *et al.* (2006) also observed highest concentration of total phenols in tree foliage during the month of May, whereas, lowest in the month of November. Amaral *et al.* (2010) observed a seasonal pattern consist of an increase of the total phenolic content from May to July.

Table 1. Seasonal variation in total phenols (%) in leaves of common fodder trees of mid Himalayas

Tree species	May	July	September	November	January	March	Mean
Bauhinia variegate	7.80[a]	7.00[d]	4.30[bd]	4.00[bd]	5.30	6.90	5.88
Grewia oppositifolia	4.80[ad]	5.60[ad]	3.20[bd]	3.10[bd]	3.70[d]	4.10[d]	4.08
Albizia lebbek	10.30[a]	9.20[a]	7.30	7.00[b]	7.50	8.20	8.25
Celtis australis	8.40[a]	9.60[a]	5.30	5.30	5.20[b]	6.30	6.68
Bambusa arundinacea	6.40[d]	7.10[ad]	5.00[b]	5.10[b]	5.70	7.00[a]	6.05
Morus alba	9.70[a]	10.30[a]	7.20	7.00	6.30[b]	7.90	8.07
Arrtocarpus lakoocha	8.90	8.00	8.10[c]	7.30[bc]	9.20[ac]	9.30[ac]	8.47
Albizia chinensis	13.60[ac]	13.90[ac]	9.20[c]	8.60[bc]	9.10[c]	10.30[c]	10.78
Mean	8.74	8.84	6.20	5.93	6.50	7.50	
S.E.	0.94	0.91	0.73	0.66	0.69	0.67	

* Superscripts 'a' and 'b' represent significantly higher and lower levels of total phenols during different months (n=6); whereas, 'c' and 'd' represent significantly higher and lower levels of total phenols among trees (n=8) in a particular month.

Table 2. Seasonal variation in simple phenols (%) in leaves of common fodder trees of mid Himalayas

Tree species	May	July	September	November	January	March	Mean
Bauhinia variegate	2.50[a]	2.30[d]	1.90	1.60[bd]	2.30	2.30	2.15
Grewia oppositifolia	1.90[d]	2.80[a]	1.60[d]	1.70	1.50[bd]	2.00	1.92
Albizia lebbek	2.80[a]	2.60[a]	2.00[b]	2.00[b]	2.00[b]	2.50	2.32
Celtis australis	2.30[a]	2.50[a]	1.80	1.90	1.50[bd]	1.90[d]	1.98
Bambusa arundinacea	1.80[ad]	1.90[ad]	1.10[bd]	1.20[bd]	1.50[d]	1.70[d]	1.53
Morus alba	2.30	3.50[ac]	2.60	2.50[c]	1.70[b]	2.10	2.45
Arrtocarpus lakoocha	3.40[ac]	3.10	2.90[c]	2.50[bc]	3.00[c]	3.00[c]	2.98
Albizia chinensis	4.30[c]	4.70[ac]	3.90[c]	2.70[bc]	2.50[bc]	3.90[c]	3.67
Mean	2.66	2.93	2.23	2.01	2.00	2.43	
S.E.	0.29	0.31	0.31	0.18	0.20	0.25	

*Superscripts 'a' and 'b' represent significantly higher and lower levels of simple phenols during different months (n=6); whereas, 'c' and 'd' represent significantly higher and lower levels of simple phenols among trees (n=8) in a particular month.

Simple or non-tannin phenols: The level of non-tannin phenols in *Bauhinia variegata* and *Arrtocarpus lakoocha* foliage was significantly high (P<0.05) during the month of May (2.50 and 3.40%, respectively) and significantly low (P<0.05) during the month of November (1.60 and 2.50%, respectively) (Table 2). The leaves of *Grewia oppositifolia* and *Morus alba* had significantly higher (P<0.05) level of simple phenols (2.80 and 3.50%, respectively) during the month of July. However, significant reduction (P<0.05) in simple phenolic content observed during the month of January (1.50 and 1.70%, respectively). *Albizia lebbek* and *Bambusa arundinacea* leaves had significantly high (P<0.05) simple phenolic content during the month of May and July. Significant reduction (P<0.05) in simple phenolic content was observed from September to January in *Albizzia lebbek* leaves, while it was from the month of September to November in *Bambusa arundinacea*. The foliage of *Celtis australis* also had significantly higher (P<0.05) level of simple phenols during the month of May and July, however, significantly lower (P<0.05) levels of non-tannin phenols was observed during the month of January (1.50%). In the month of July, *Albizia chinensis* foliage exhibited significantly higher (P<0.05) level of non-tannin phenols (4.70%), thereafter, there was a significant reduction in their content from November to January (2.70 to 2.50%).

Net tannin content: Tannins in tree fodders form the most common detrimental factors that bind and precipitate the proteins and make complex bonds with starch, cellulose and minerals affecting the digestibility. Tannins also affect the palatability of feed and foliage by forming complexes with salivary glycoproteins. Tannins have been claimed to exert adverse effect on the digestibility of dietary protein and to a lesser extent, that of available carbohydrate and lipid (Mosely and Griffiths, 1979). *Bauhinia variegata*, *Grewia oppositifolia*, *Celtis australis* and *Morus alba* foliage had significantly higher (P<0.05) amount of net tannins during the month of May to July. A significant reduction (P<0.05) in net tannin content observed from September to November except *Morus alba,* where there was consistent reduction in tannin content up to month of January (Table 3). The foliage of *Albizia lebbek* had significantly higher (P<0.05) levels of net tannins during the month of May (7.50%), thereafter a significant reduction (P<0.05) in the net tannin content was observed during the month of November (5.00%). In the month of July, *Bambusa arundinacea* foliage revealed significantly higher (P<0.05) level of net tannins (5.20%), thereafter level of net tannins reduced to 3.90% during September and November. Foliage of *Arrtocarpus lakoocha* clipped during the month of January and March showed significantly higher (P<0.05) level of net tannins (6.20 and 6.30%, respectively). Salaj and Karmutak (1995) also reported such increase in net tannin content during winter season in *Abies alba* foliage. It was observed that *Arrtocarpus lakoocha* had significantly lower (P<0.05) level of net tannin during months of July and November. Significantly higher (P<0.05) level of net tannins observed in *Albizia chinensis* foliage during the month of May and July (9.30 and 9.20%, respectively) thereafter, net tannin content decreased significantly (P<0.05) during the month of September (5.30%). Various species of tree leaves responded differently to seasonal changes as some species showed an increase in tannin content in summer and some showed an increase in winter (Vaithiyanathan and Singh, 1989). Gupta *et al*. (1992) reported an increase in tannin content in summer season.

Condensed tannin content: The presence of higher levels of condensed tannins in the leaves is toxic to the animal as they affect the mucosa of the digestive tract, which affects decrease in absorption of methionine and lysine amino acids. Decreased methionine availability could increase the toxicity of other plant compounds such as cyanogenic glycosides, because methionine is involved in the detoxification of cyanide (Reed, 1995). *Bauhinia variegata* foliage lopped during the month of March, revealed significantly higher (P<0.05) amount of condensed tannins (1.40%) which reduced significantly in preceding month of January (0.80%) (Table 4). *Grewia oppositifolia* foliage harvested during the month of May and July had significantly higher level (P<0.05) of condensed tannins (1.70 and 1.50%, respectively). Afterwards, there was a significant reduction (P<0.05) in condensed tannins content during the November (0.70%). The leaves of *Albizia lebbek* revealed significantly higher (P<0.05) level of condensed tannins content (2.80 and 2.50%) during months of May and July, respectively, however significantly lower level (P<0.05) of condensed tannins was observed during the month of March (1.30%). Significantly higher (P<0.05) level of condensed tannins was observed in foliage of *Celtis australis* during the month of May and July (3.60 and

Table 3. Seasonal variation in net tannins (%) in leaves of common fodder trees of mid Himalayas

Tree species	May	July	September	November	January	March	Mean
Bauhinia variegate	5.30^{a}	4.70^{a}	2.40^{bd}	2.40^{bd}	3.00^{d}	4.60	3.73
Grewia oppositifolia	2.90^{ad}	2.80^{ad}	1.60^{bd}	1.40^{bd}	2.20^{d}	2.10^{d}	2.17
Albizia lebbek	7.50^{ac}	6.60	5.30^{c}	5.00^{bc}	5.50^{c}	5.70	5.93
Celtis australis	6.10^{a}	7.10^{a}	3.50^{b}	3.40^{bc}	3.70	4.40	4.70
Bambusa arundinacea	4.60^{d}	5.20^{a}	3.90^{b}	3.90^{b}	4.20	5.30	4.52
Morus alba	7.40^{ac}	6.80^{a}	4.60^{b}	4.50^{b}	4.60^{b}	5.80	5.62
Arrtocarpus lakoocha	5.50	4.90^{b}	5.20^{c}	4.80^{b}	6.20^{ac}	6.30^{ac}	5.48
Albizia chinensis	9.30^{ac}	9.20^{ac}	5.30^{bc}	5.90^{c}	6.60^{c}	6.40^{c}	7.12
Mean	6.08	5.91	3.98	3.91	4.50	5.08	
S.E.	0.70	0.68	0.50	0.52	0.54	0.50	

*Superscripts 'a' and 'b' represent significantly higher and lower levels of net tannin during different months (n=6); whereas, 'c' and 'd' represent significantly higher and lower levels of net tannin among trees (n=8) in a particular month.

Table 4. Seasonal variation in condensed tannins (%) in leaves of common fodder trees of mid Himalayas

Tree species	May	July	September	November	January	March	Mean
Bauhinia variegate	1.20^{d}	1.10^{d}	1.00^{d}	1.00^{d}	0.80^{bd}	1.40^{ad}	1.08
Grewia oppositifolia	1.70^{ad}	1.50^{ad}	0.90^{d}	0.70^{bd}	0.90^{d}	1.00^{d}	1.12
Albizia lebbek	2.80^{a}	2.50^{a}	2.00^{c}	1.90^{c}	2.00^{c}	1.30^{bd}	2.08
Celtis australis	3.60^{ac}	3.70^{ac}	2.30^{c}	2.10^{c}	1.60^{b}	1.90	2.53
Bambusa arundinacea	1.80^{d}	1.90	1.70^{b}	1.70^{b}	1.90	2.10^{a}	1.85
Morus alba	3.20^{a}	3.50^{ac}	1.30	1.20^{b}	1.10^{bd}	1.70	2.00
Arrtocarpus lakoocha	2.60^{a}	2.40	1.90^{b}	1.90^{bc}	2.30^{c}	2.20^{c}	2.22
Albizia chinensis	4.20^{ac}	4.40^{ac}	2.10^{c}	1.90^{bc}	1.90^{b}	2.60^{c}	2.85
Mean	2.64	2.63	1.65	1.55	1.56	1.78	
S.E.	0.36	0.41	0.19	0.18	0.20	0.19	

3.70%, respectively). Following the month of July, leaves of *Celtis australis* had significantly lower (P<0.05) level of condensed tannins. *Bambusa arundinacea* foliage showed significantly higher (P<0.05) level of condensed tannins during the month of March (2.10%) however, during September and November, the condensed tannins content reduced significantly (P<0.05). In the month of May, *Arrtocarpus lakoocha* foliage had higher level of condensed tannins (2.60%) thereafter, significant reduction (P<0.05) in condensed tannin content (1.90%) was observed in the month of September and November. *Morus alba* and *Albizia chinensis* had significantly higher (P<0.05) level of condensed tannins during the month of May and July (3.20 and 3.50%, 4.20 and 4.40%, respectively), which decreased significantly during the month of November and January. Rana *et al*. (2006) also reported that condensed tannins start accumulating in tree leaves during winter season and reached to the maximum level in April and May. Subba *et al*. (1996) observed higher tannin content, during spring season in fodder trees. During winter season, the condensation of tannins is a contributing process to plant defense system in order to avoid any injury from biotic and abiotic stress (Salaj and Karmutak, 1995).

Hydrolysable tannin content: During the month of May and July, leaves of *Bauhinia variegata* had significantly high (P< 0.05) level of hydrolysable tannins (4.10 and 3.60%, respectively) (Table 5), which reduced to 1.40% during month of September and November. *Grewia oppositifolia* foliage revealed significantly higher (P< 0.05) levels of hydrolysable tannins (1.30%) during July and January, whereas significant reduction (up to 0.70%) observed during September and November. *Albizia lebbek* foliage harvested during the month of May and March revealed significantly high (P<0.05) levels of hydrolysable tannins (4.70 and 4.40%, respectively), whereas significantly lower (P<0.05) level was observed during the months of September and November. During July, leaves of *Celtis australis* exhibited significantly higher (P<0.05) amount of hydrolysable tannins (3.40%) however, significant reduction (P<0.05) in hydrolysable tannins content recorded in September and November (1.20 and 1.40%, respectively). The foliage of *Bambusa arundinacea* revealed significantly higher (P<0.05) level of hydrolysable tannins during the month of July and March (3.30 and 3.20%, respectively), whereas, significantly lower (P<0.05) level (2.20%) of hydrolysable tannins was observed in the months of September and November. *Morus alba* revealed significantly higher (P<0.05) levels of hydrolysable tannins during the month of May and March (4.20 and 4.10%, respectively), while lower levels of hydrolysable tannins were observed from July to November. *Arrtocarpus lakoocha* foliage revealed significantly higher (P<0.05) levels of hydrolysable tannins (3.90 and 4.10%) during month of January and March. Hydrolysable tannins reduced significantly (P<0.05) during month of July (2.50%). In *Albizia chinensis*, the highest level of hydrolysable tannins was observed in the month of May (5.10%) whereas, lowest level (2.20%) was recorded during month of September. The pattern of hydrolysable tannins showed that concentration was high during summer and autumn season. The results of the study confirmed the earlier finding of Gupta *et al*. (1992) who reported high level of hydrolysable tannins in tree leaves during summer season.

Table 5. Seasonal variation in hydrolysable tannins (%) in the leaves of common fodder trees of mid Himalayas

Tree species	May	July	September	November	January	March	Mean
Bauhinia variegate	4.10^{a}	3.60^{a}	1.40^{bd}	1.40^{bd}	2.20	3.20	2.65
Grewia oppositifolia	1.20^{d}	1.30^{ad}	0.70^{bd}	0.70^{bd}	1.30^{ad}	1.10^{d}	1.05
Albizia lebbek	4.70^{ac}	4.10^{c}	3.30^{bc}	3.10^{bc}	3.50	4.40^{ac}	3.85
Celtis australis	2.50	3.40^{a}	1.20^{bd}	1.30^{bd}	2.10^{d}	2.50	2.17
Bambusa arundinacea	2.80	3.30^{a}	2.20^{b}	2.20^{b}	2.30	3.20^{a}	2.67
Morus alba	4.20^{a}	3.30^{b}	3.30^{bc}	3.30^{bc}	3.50	4.10^{ac}	3.62
Arrtocarpus lakoocha	2.90	2.50^{b}	3.30^{c}	2.90	3.90^{ac}	4.10^{ac}	3.27
Albizia chinensis	5.10^{ac}	4.80^{c}	2.20^{b}	4.00^{c}	4.70^{c}	3.80	4.10
Mean	3.44	3.29	2.20	2.36	2.94	3.30	
S.E.	0.46	0.37	0.37	0.41	0.40	0.38	

*Superscripts 'a' and 'b' represent significantly higher and lower levels of hydrolysable tannin during different months (n=6); whereas, 'c' and 'd' represent significantly higher and lower levels of hydrolysable tannin among trees (n=8) in a particular month.

Conclusion

The results of study revealed that seasonal variations exist in the anti-nutrient contents among fodder trees of mid hill Himalayan region. Foliages from *Grewia oppsitifolia* and *Bauhinia variegata* had significantly lower levels of anti-nutrients thereby; the foliages from these trees could serve as an excellent alternate feed resource for livestock. Lopping of these trees from the month of September to December provide better quality foliage for livestock feeding. *Grewia oppsitifolia* may be lopped preferably in the month of November, while October is good lopping month for *Bauhinia variegata.* The study also pointed that during the month of September and November, *Morus alba* had low levels of anti-nutrients, therefore, *Morus alba* could be lopped between September to November for feeding livestock. The other fodder trees including *Arratocarpus lakoocha, Bambusa arundinacea, Celtis australis* and *Albizia lebbek* could be lopped between the months of September to December for obtaining fodder with low level of anti-nutrients. The best time for loping *Albizia chinensis* for fodder is the month of November. Any delay in lopping beyond these periods may cause appreciable gain in anti-nutrient concentration, which could have adverse effect on the health of livestock.

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