Chemical characterisation of shrubs browses from salt affected soils and Pothohar areas in Pakistan for their nutritional and anti-nutritional compounds

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Four species of shrubs browses comprising of \textit{Adhatoda vasica} \& \textit{Dodonaea viscosa} from the hilly areas and two species of \textit{Acacia ampliceps} \& \textit{Atriplex lentiformis} well adapted to the salt affected waste soils of the Punjab-province in Pakistan were randomly selected for their nutritional \& anti-nutritional compounds along with the mineral contents. The ground samples were analysed for chemical characterization for DM, OM, EE, CP, NDF, ADF, NFE, ash content and for anti-nutritional compounds such as total phenols, saponins, oxalates, condensed tannins and minerals. The OM\% (94.22 ± 1.36) was significantly (P<0.001) higher in \textit{Dodonaea viscosa} followed by \textit{Acacia ampliceps} (87.35 ± 1.36) and \textit{Adhatoda vasica} (83.97±1.36). Shrubs showed fair amount of CP 8.28 \%, 10.59 \%, 15.5 \% & 28.47 \% in \textit{D. viscosa}, \textit{A. lentiformis A. Ampliceps,} \& \textit{A. vesica}, respectively. The maximum (P<0.001) NDF \% (57.43± 0.39) was found in \textit{A. ampliceps} followed by \textit{A. lentiformis} (44.49 ± 0.39) and \textit{D. viscosa} (31.10 ± 0.39), respectively. The maximum values for the ADF \& ADL were in \textit{A. ampliceps} followed by \textit{A. lentiformis} and \textit{D. viscosa}, respectively. Minimum (P<0.001) levels of CT were found in \textit{A. lentiformis} (6.87± 0.72) followed by \textit{A. ampliceps} (9.32 ± 0.72). The total ash \% (21.98 ± 0.34) was found significantly (P<0.001) higher in \textit{Atriplex lentiformis} followed (16.03± 0.34) by \textit{Adhatoda vasica} (16.03 ± 0.34) and \textit{Acacia ampliceps} (12.65± 0.34), respectively. The maximum (P<0.001) condensed tannin (g/kg DM) values (30.9± 2.9), and total phenols (73.2± 1.0) along with saponins (16.4 ± 2.0) were found in \textit{D. Viscosa}. \textit{A. vasica} (9064±220) showed the maximum (P<0.001) values for Ca ppm, followed by \textit{D. Viscosa} (4397 ±220). \textit{A. lentiformis} depicted minimum (P<0.001) contents of the Ca (1028 ±220), P (174 ± 48), K (2248 ±205), Mg (726 ± 36), S (622±104), Fe (43.37 ±8.30), Mn (5.73 ± 3.95), Cu (0.98 ± 0.38) and Se (1.03 ±0.65) as compared to other shrub species tested. The tropical shrub browses may become potential supplements to traditional forages for ruminants specially during scarcity seasons.

Keywords: \textit{Adhatoda vasica, Dodonaea viscosa, Acacia ampliceps, Atriplex lentiformis}, total phenols, saponins, oxalates, condensed tannins, nutritional composition secondary compounds.

Novelty statement: Shrubs could be used as novel feed supplements in livestock production.
INTRODUCTION

Pakistan comprised of large number of livestock population with a contribution of more than half 56.3% in the agricultural value added and 11.8 % to national GDP (Anonym, 2015). Livestock are the mainstay of the socio-economic structure of the small land holders in the rural areas of the country (Sultan et al., 2007). Indigenous livestock farming had been dependent on seasonal fresh green biomass availability (cut & carry system) in an integrated system of agriculture. The land allocation for fodder production is continuously reducing due to the farmer’s preference for urgently needed cash crops. The pastures are also continuously converted into croplands leaving no land for livestock grazing (Pratt et al., 1997). Therefore, an acute shortage of green biomass has been observed both in irrigated as well as in mountainous and arid regions (Roy et al., 1989). Resultantly animals are facing severe deficiency both in energy and protein by 26% & 38%, respectively in Pakistan (Riaz et al., 2008). The lowered productivity of livestock in tropical regions has already been attributed to the inadequate and poor quality feed (Saleem et al., 2006). The waste or abandoned lands due to salinity, water logging or other soil hazards in addition to other un-commanded uphill ranges would become one of the potential land resources for range livestock production. Some drought resistant plants, shrubs and saltbushes are found adapted with sufficient green biomass production potential in the said conditions and may have potential to supplement the traditional fodders for range livestock (Ondiek et al., 2000). These multipurpose trees and shrubs vegetation had shown the potential to alleviate some of the feed shortages and nutritional deficiencies experienced particularly during fodder scarcity seasons observed permanently in a calendar year (Papachristou and Nastis, 1996; Salem et al., 2006, FAO, 1992; Tops, 1992). Trees & shrubs browse foliages may have higher levels of secondary metabolites i.e., alkaloids, saponins and tannins which are generally basic component of diets in range livestock (sheep, goats or camels) in the arid regions of the tropical countries. Several earlier workers from different parts of the world have revealed that trees and shrubs browses contained substantial amount of certain anti-nutritional compounds, mainly tannins in addition to other secondary metabolites which may reduce their nutritional value (Makkar and Becker, 1993; Salem, 2005; Reed, 1986; Aganga and Mosase, 2001; Dube et al., 2001). Salem et al., (2006) had suggested that the nutritional level of the tree browse foliages can be estimated by determining the levels of nutrients and secondary compounds (condensed tannins, total phenols, saponins, alkaloids and essential oils). The livestock industry in the world is also emphasizing on the investigation of plant browses as an alternative feed additives (European Union, 2003) for the substitution of chemical ionophores (Chalupa et al., 1980, Bergen and Bates, 1984). The present study was therefore designed for the chemical characterisation of shrub browses commonly found in Pakistan.

MATERIALS AND METHODS

Shrubs browse samples: The browse forage of the shrubs included an Australian Kiker (Acacia ampliceps) and saltbush (Atriplex lentiformis) was taken from the experiment field Pakaa Ana, Nuclear Institute for Agriculture and Biology (NIAB) Faisalabad-Pakistan. Faisalabad is situated at 31º- 26 º latitude and 73º- 60º longitude at 184 m elevation. Australian Kiker (Acacia ampliceps) and saltbush (Atriplex lentiformis). The other two shrubs included were Sanatha (Dodonaea viscosa) and Baker (Adhatoda vasica), from hilly areas of Pakistan situated at 457 to 610 m altitude with average rainfall 1143mm (Semi-arid) annually, having 24.4-35 ºC average temp in summer and 4.0 - 16.6 ºC in winter.

Nutrient analysis: The sun dried shrub browse samples of the bushes were further dried in a forced draught oven at 60 ºC and finally ground in a Wiley mill to pass a 1 mm sieve before the chemical analysis. The ground samples were analysed for dry matter (DM), organic matter (OM), ether extracts (EE) and ash (AOAC, 2006). Nitrogen was analysed by the Nitrogen analyzer: Leco Model FP- 528 and % CP was determined (crude protein% =% N*6.25). Samples were also analysed for NDF, ADF and ADL as by Van Soest et al., (1991). Nitrogen free extract was calculated by the equation; %NFE= 100-(%CP+ %CF + %EE+ % Ash).

Anti-nutrient compounds: The total phenols and condensed tannins were examined by the calorimetric method described by Makkar et al (1993) & Makkar (2003). Saponins were analysed by the method of Hiai et al (1976). Oxalic acid (mg)/100g was determined as described by Harinder et al. (2007) and original principles of AOAC (2006), and Baker (1952).

Mineral analysis: One g sample was placed into a Kjeldahl tube and 20 mL pure HNO3 were added for the determination of mineral contents. The samples were digested in Kjeldahl digestion chamber at 100 ºC and then digested samples were diluted to the original volume of 20 mL with water. The samples were filtered through Whatman filter papers no 541 and the concentrations of selected minerals were determined with inductively coupled plasma optical emission spectroscopy (ICP-OES) with Unicam 701 ICP-OES.

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Statistical analysis: All of the data were generated in duplicate for chemical analysis and for the secondary metabolites on dry matter basis was subjected to analysis of variance techniques (Steel and Torrie, 1984), by using General Linear Model of Mini Tab. Tuckey’s test was used for the comparison of the means at the significance (P<0.05) or (P<0.01) level.

RESULTS AND DISCUSSION

Nutrient analysis

The bushes samples were significantly (P<0.001) differed from each other. The total ash % was found significantly (P<0.001) higher (21.98 ± 0.34) in A. lentiformis followed by A. vasica (16.03 ± 0.34) and A. ampliceps (12.65± 0.34), respectively. The OM% (94.22 ± 1.36) was found significantly (P<0.001) higher in D. viscosa followed by A. ampliceps (87.35 ± 1.36) and A. vasica (83.97±1.36). The CP contents in all of the shrubs browses tested were in the range (8.09% - 28.47%). Maximum CP % was found in A. vasica (28.47 ± 0.8%) and significantly (P<0.001) varied from all other shrubs. A. lentiformis & A. ampliceps revealed fair amount of CP (10.59 % & 15.5 %, respectively). Only D. viscosa showed lower (8.28 ± 0.8 %) CP content and A. vasica browses contained > 20% CP content (28.47± 0.8%). The CP content in these shrubs was comparable to other traditional range grasses and herbs. Craig et al. (1991) also worked on the chemical composition as % age of DM of Acacia ampliceps as one of the acacia species tested from naturally saline areas and reported CP % (16.6 & 11.9), NDF (35 & 44), ADF (21 & 35) and ADL (11 & 20) content, respectively. The level of CP available in the shrubs would sufficient to support the maintenance of range livestock as the US National Academy of Sciences (1975) suggested that more than 8% CP is required for the maintenance of sheep. Milford and Haydock (1965) also indicated a minimal CP level 7.2% of DM for the maintenance of sheep. The results were also supported by Norton (1982). The maximum (P<0.001) NDF % (57.43± 0.39) was found in A. ampliceps followed by A. lentiformis (44.49± 0.39) and D. viscosa (31.10± 0.39), respectively. The maximum values for the ADF & ADL were in A. ampliceps followed by A. lentiformis and D.viscosa, respectively. All of the samples were significantly (P<0.001) differed from each other with respect to their chemical composition. Saleem et al. (2006) also analysed the chemical composition and secondary compounds of four species of the tree foliage and reported that the CP content ranged from 124 to 185 g/kg DM. The NDF (368-615), ADF (200-542) and ADL (101-192) and were comparable to values observed in the present study. The slight difference might be due to the stage and or age of plants in different climatic conditions. Chernery et al., (1993) also found that NDF, ADF and ADL increased with increasing maturity in tropical grasses. Wilson (1977) also analysed four species of shrubs and four tree species for NDF (34-62%), ADF (20-41%) & ADL (9.7-14.3%) on OM basis and reported that ash % (16.4- 29.5) was higher in shrubs than tree forage. Wilson (1977) conducted ash % (24.2 & 29.5) in two species of Atriplex (A. vasicaria & A. numularia). The nutrient levels determined in the present study were also comparable to Craig et al. (1991), LeHouerou (1980), Tops (1992), and Rubanza et al. (2003). Khanum et al., (2007) worked on the chemical composition of grasses, browse plants, cultivated fodders, salt tolerant plants, crop residues and concluded that CF, EE and NFE contents for roughages ranged from 13.8 to 35.1%, 1.0 to 4.8% and 29.5 to 62.9%, respectively. Hence the results for the chemical characterisation of shrubs foliages revealed that they could be supplemented in the feeding of livestock. Riaz et al., (2008), also revealed that poor roughages and crop residues may be supplemented to traditional fodders up to 30% level especially in fodder scarcity seasons, the findings of this study are also supported by Craig et al., (1991) who reported that the bushes like A. ampliceps from saline lands could be supplemented in other traditional forages at the maintenance level. However plenty of drinking water would be required for the bushes high in ash content just to excrete the salt load from the body.

Minerals profile

A.vasica (9064±220) showed the maximum (P<0.001) values for Ca ppm, followed by D. Viscosa (4397 ±220). A. lentiformis depicted minimum (P<0.001) contents of the Ca (1028 ±220), P (174 ± 48), K (2248 ±205), Mg (726 ± 36) and S (622±104) as compared to other shrub browse tested in this study. A. lentiformis also had significantly (P<0.001) lower values of Fe (43.37 ±8.30), Mn (5.73 ± 3.95), Cu (0.98 ± 0.38) and Se (1.03 ±0.65) as compared to other shrub species tested. Craig et al., (1991) also conducted the chemical composition of Acacia species from naturally saline areas and ash reported (10.8-14.6%) content comprising of P (0.17 & 0.10), Ca (1.43 & 2.13), and Na (0.92 & 0.96), and Mg (0.62 & 0.57) percent of DM having Ca: P ratio (8.4 & 21.3) in new shoots and mature phylloides of Acacia ampliceps. Riaz et al., (2008) also concluded that the halophytic plants showed higher concentrations of Na, K, Cl, Cu and Se, and lowered levels of Ca, P and Mg. The optimum level of mineral profile in these trees and shrub browses shows their nutritional importance for different categories of range livestock. Macro and micro mineral elements are considered valuable for keeping the livestock health, even deficiency of a single element cannot be masked over by the others (Abassi et al., 2009). The values of the mineral profile found in shrub browses tested in this research project were comparable to the previous studies and in agreement to those reported by Riaz et al., (2005) and Craig et al., (1991).
Table 1. Chemical composition (g/100g DM) of tropical shrub browses

<table>
<thead>
<tr>
<th>Composition (%)</th>
<th>A. vasica</th>
<th>D. viscosa</th>
<th>A. ampliceps</th>
<th>A. lentiformis</th>
</tr>
</thead>
<tbody>
<tr>
<td>DM</td>
<td>91.8±0.16</td>
<td>92.24±0.16</td>
<td>93.66±0.16</td>
<td>95.46±0.16</td>
</tr>
<tr>
<td>OM</td>
<td>83.97±1.36</td>
<td>94.22±1.36</td>
<td>87.35±1.36</td>
<td>78.02±1.36</td>
</tr>
<tr>
<td>ASH</td>
<td>16.03±0.34</td>
<td>5.78±0.34</td>
<td>12.65±0.34</td>
<td>21.98±0.34</td>
</tr>
<tr>
<td>CP</td>
<td>28.47±0.8</td>
<td>08.28±0.8</td>
<td>15.5±0.8</td>
<td>10.59±0.8</td>
</tr>
<tr>
<td>NDF</td>
<td>23.13±0.4</td>
<td>31.10±0.4</td>
<td>57.43±0.4</td>
<td>44.49±0.4</td>
</tr>
<tr>
<td>ADF</td>
<td>15.66±1.3</td>
<td>21.04±1.3</td>
<td>49.32±1.3</td>
<td>21.45±1.3</td>
</tr>
<tr>
<td>ADL</td>
<td>10.93±1.3</td>
<td>11.93±1.3</td>
<td>31.99±1.3</td>
<td>13.36±1.3</td>
</tr>
<tr>
<td>NFE</td>
<td>31.09±1.2</td>
<td>53.13±1.2</td>
<td>13.02±1.2</td>
<td>22.01±1.2</td>
</tr>
<tr>
<td>EE</td>
<td>1.28±0.56</td>
<td>1.71±0.56</td>
<td>1.40±0.56</td>
<td>0.93±0.56</td>
</tr>
</tbody>
</table>

Mean ± standard deviation. Values in same rows, sharing same letters differ non-significantly (P>0.001)

Table 2. Minerals (ppm) profile in the tropical shrub browses

<table>
<thead>
<tr>
<th>Minerals profile (ppm)</th>
<th>A. vasica</th>
<th>D. viscosa</th>
<th>A. ampliceps</th>
<th>A. lentiformis</th>
</tr>
</thead>
<tbody>
<tr>
<td>Calcium (Ca)</td>
<td>9064±220</td>
<td>4397±220</td>
<td>3551±220</td>
<td>1028±220</td>
</tr>
<tr>
<td>Phosphorus (P)</td>
<td>576±48</td>
<td>593±48</td>
<td>533±48</td>
<td>174±48</td>
</tr>
<tr>
<td>Sodium (Na)</td>
<td>334±202</td>
<td>1570±202</td>
<td>5474±202</td>
<td>6754±202</td>
</tr>
<tr>
<td>Potassium (K)</td>
<td>6254±205</td>
<td>8248±205</td>
<td>4097±205</td>
<td>2248±205</td>
</tr>
<tr>
<td>Magnesium (Mg)</td>
<td>2715±36</td>
<td>1166±36</td>
<td>2270±36</td>
<td>726±36</td>
</tr>
<tr>
<td>Sulfur (S)</td>
<td>1119±104</td>
<td>1360±104</td>
<td>3758±104</td>
<td>622±104</td>
</tr>
<tr>
<td>Iron (Fe)</td>
<td>59.40±8.30</td>
<td>69.44±8.30</td>
<td>104.25±8.30</td>
<td>40.37±8.30</td>
</tr>
<tr>
<td>Manganese (Mn)</td>
<td>14.24±3.95</td>
<td>20.44±3.95</td>
<td>11.47±3.95</td>
<td>5.73±3.95</td>
</tr>
<tr>
<td>Zinc (Zn)</td>
<td>9.05±1.62</td>
<td>23.52±1.62</td>
<td>6.71±1.62</td>
<td>5.7±1.62</td>
</tr>
<tr>
<td>Copper (Cu)</td>
<td>6.73±0.38</td>
<td>5.12±0.38</td>
<td>2.77±0.38</td>
<td>0.98±0.38</td>
</tr>
<tr>
<td>Selenium (Se)</td>
<td>2.50±0.65</td>
<td>3.17±0.65</td>
<td>2.03±0.65</td>
<td>1.03±0.65</td>
</tr>
</tbody>
</table>

Mean ± standard deviation. Values in same rows, sharing same letters differ non-significantly (P>0.001)

Table 3. Secondary compounds (g/kg DM) of some trees & shrubs browse foliage

<table>
<thead>
<tr>
<th>Secondary compounds (g/kg DM)</th>
<th>A. vasica</th>
<th>D. viscosa</th>
<th>A. ampliceps</th>
<th>A. lentiformis</th>
</tr>
</thead>
<tbody>
<tr>
<td>C. tannins</td>
<td>09.126±2.93</td>
<td>30.90±2.93</td>
<td>18.12±2.93</td>
<td>04.18±2.93</td>
</tr>
<tr>
<td>T. phenols</td>
<td>21.55±1.03</td>
<td>73.21±1.03</td>
<td>09.32±1.03</td>
<td>06.87±1.03</td>
</tr>
<tr>
<td>Saponins</td>
<td>04.50±1.98</td>
<td>16.40±1.98</td>
<td>08.85±1.98</td>
<td>04.18±2.93</td>
</tr>
<tr>
<td>Oxalates</td>
<td>0.29±0.12</td>
<td>0.61±0.12</td>
<td>0.55±0.12</td>
<td>1.02±0.12</td>
</tr>
</tbody>
</table>

Mean ± standard deviation. Values in same rows, sharing same letters differ non-significantly (P>0.001)

Secondary metabolites

The compositions of secondary compounds are also presented in Table 3. Minimum (P<0.001) levels of total phenols were found in A. lentiformis (6.87±1.03) followed by A. ampliceps (6.87±1.03). The maximum (P<0.001) condensed tannin (30.9±2.9), and total phenols (73.12±1.03) along with saponins (16.40±1.98) were found in D. viscosa. Minimum (P<0.001) CT (4.18±2.93) and total phenols (6.87±1.03) were found in A. lentiformis. Craig et al., (1991) also found TP, CT and SAP values 44.3, 31.6 & 8.3 g/kg DM in the composition of E. camaldulensis & 102.3, 68.1 & 14.6 g/kg/DM in Acacia ampliceps from naturally saline areas, respectively. Saleem et al., (2006) also observed the TP, CT, and SAP within the range of 29.0-102.3, 20.8-68.1 & 3.0-14.6 g/kg DM, respectively in four tree foliages i.e., C. speciosa, C. fistula, S. molle and E. camaldulensis.
The values for TP, CT & SAP 6.00-103.1, 4.18-55.02 & 1.30-86.35 g/kg DM obtained in the present study are in agreement with the mentioned earlier workers. These tropical shrub browse have higher levels of secondary compounds as compared to certain other traditional forages might be due to their defensive mechanism against pest and predators, high environmental temperature and drought stress (Mangan, 1988). Shayou and Uden (1999) and Abdulrazak et al. (2000), reported similar findings (high TP & CT level) in some East African browses. High polyphenolic compounds were also reported in semi-arid of north Egypt (Saleem, 2005) and arid regions of Sudan (Fadel Elseed et al., 2002) and in agreement with the present study. Rubanza et al. (2003) revealed that TP and CT were between 65-237 and 6-74 g/kg DM, respectively in some tree foliages studied. The differences in values of the secondary compounds are attributed to different assays and assay standards in addition to certain variation in chemical composition of polyphenolics among foliages (Makkar and Becker, 1993); Pino et al., 2005). Secondary compounds are reported as the main property of plant genotypic factors controlling physiological synthesis and accumulation of secondary compounds in drought resistant plants (Okuda et al., 1993; Kelman et al., 1997).

CONCLUSIONS

The tropical shrub browses may become potential supplements to traditional forages for ruminants maintained especially during scarcity seasons in Pakistan.

REFERENCES


