Nutritive Status of *Maytenus heterophylla* and *Acacia nilotica* Leaves Cultivated in Winter from Two Varied Landscapes

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**Abstract:** Winter season happens to be the period prone to drought and in most cases bedeviled with incidences of low-quality veld and a corresponding high rate of livestock mortality. The present research aimed to assess the nutritive worth of *Acacia nilotica* as well as *Maytenus heterophylla* leaves in the winter season from two varied landscapes (high landscape and low landscape). A randomized study design was employed with landscape and plant species as the key factors of a 100×100 m plot inside sites presenting blocks. Both nutritive and mineral components of the leaves were evaluated. In addition, condensed tannins, as well as phenolic compounds analysis were also carried out on the dry samples of the leaves. A significant (p<0.01) relationship existed between site and plant types with regard to DM, N and CP content. *Acacia nilotica* leave samples had greater values of N, DM as well as crude protein in high topography in comparison to *Maytenus heterophylla* leaves. No significant relationship (p>0.05) existed with NDF content for *A. nilotica* from the two sampled locations. *M. heterophylla* showed high content for NDF in the sites for low topography as compared to high topography. *Acacia nilotica* as well as *Maytenus heterophylla* species showed variations in the minor and major element content in relation to topography. Low landscape areas had higher values in most elements for both plant species as compared to those obtained from high topographic sites. Likewise, the low landscape area had higher mineral content than the high landscape area. Browse species and topography showed no significance with regards to Soluble Phenolics (SPh) as well as soluble condensed tannins of browse species. Significant interaction (p<0.05) existed between topographical location as well as browse species. Browse species from high topography showed higher soluble phenolics and soluble condensed tannins components in comparison to low topography. Our results revealed that leaves from low landscapes have better nutrient composition compared to leaves from high landscapes.

**Keywords:** Chemical Composition, Landscape, Dry Season, Browse Plants, Tannins

**Introduction**

Feed accounts for almost 70% of animal production (Oyegb et al., 2019). However, in order to tackle the rising challenge of lessening feed costs, financial insufficiency and rising food production as well as encourage food security in some parts of the world (semi-arid areas), scientists and farmers are now looking at the possibility of utilizing fodder trees to improve livestock (Idamokoro et al., 2016). According to Du Toit et al. (2012), seven categories of forage biomes form the source of conventional pasture for grazing range animals in Southern Africa. Within these biomes, there are several fodder trees such as *Acacia nilotica* and *Maytenus heterophylla* among others which are readily available and can be used as green nutritious browse trees to feed ruminant animals during prolonged dry seasons (Tefera et al., 2008). Conversely, these pastures are habitually deficient in adequate nutritive components to sustain livestock production (Mellado et al., 2005). According to Richardson and Hahn (2007); Mapiye (2009), the absence of quality feed is a main challenge to farm animal production in free ranging livestock production system in this region.

Mlambo et al. (2007) further reported that some specific nutritive contents such as protein are, specifically
a key limiting feed ingredient in livestock farming in semi-arid regions. The shortage of adequate nutrients in pastures may possibly be a result of the extremely fibrous nature of velds in semi-arid regions and the anti-nutritive compounds present in these tree species which might impact the diet and well-being of farm animals. Anti-nutritional components of worry are the high proportion of phenolic components including tannins in a variety of browse plants (Idamokoro et al., 2016). The phenolic compounds of different browse plants differ in terms of the different browse plants as well as their locations together with the aforementioned issues have restricted the prospect of farming in some arid locations (Gwaze et al., 2010).

The nutritional worth and yield of fodder trees also vary substantially with regard to species, altitude and soil (Shah et al., 2019). Investigation of the variations in these parameters is essential for plant breeders to recognize trees with better nutritive content as well as yield for livestock feed management. Knowledge of landscape (altitude/topography) factors affecting plant yield and nutritional quality is also needed to come up with local range management and fodder flow systems of farming and to enable rural pastoralists to utilize forage plants for feeding their livestock (Shah et al., 2019). The ability of farm animals to exploit forage and browse plants, in the long run, helps to boost food security as well as sustain the use of browse feed plants as feed; it also lowers rivalry between livestock and man use of the plant (Makkar, 2018).

During drought seasons (e.g., winter), the nutritional quality and shortage of veld results in the scarcity of livestock feeds (Idamokoro et al., 2016). Conversely, as a result of the scarcity of forages, feeding a single type of crop residue to livestock causes poor productivity of most animals in tropical nations (Babayemi et al., 2004). Limited resource farmers who rear livestock often depend/rely on fodder or natural/domestic waste to maintain their animals (Idamokoro et al., 2017). Some commonly grown fodder trees including Artocarpus lakoocha, Thysanolaena maxima, Ficus roxburghii and Bauhinia purpurea are used as maintenance ration to sustain their livestock (Shah et al., 2019). Fodders are known to provide certain needed nutrients in the feeds of livestock during the dry period when protein as well as nutrient deficiency are expected to happen (Bhatt and Verma, 2002; Singh et al., 2022).

Acacia nilotica and Maytenus heterophylla are two ecological species widely overlooked as browse plants for livestock in the Ciskei region of South Africa. The present study seeks to assess the anti-nutritional and nutritive value of A. nilotica as well as M. heterophylla leaf samples from two varied landscape terrains in the winter season. This season is usually characterized by dryness (winter season) and it was chosen for this study because this is the period when there is a shortage of pasture with little or no nutritive value for livestock to utilize in the present study region. The findings from the study may be helpful to both animal feed nutritionists and rural pastoralists in the choice of what and where to graze their livestock during this season of drought and food shortage.

Materials and Methods

Study Site and Harvesting Methods

Leaves (fresh) from twenty (20) randomly carefully nominated specific trees, each from Maytenus heterophylla and Acacia nilotica tree species, were collected by hand in high and low topographic sites from the former Nkonkobe municipality (Alice) located 120 km East in the eastern coastline. The sites of the study lie between 26.9 East as well as 32.8 South as well as positioned 520 m above sea level. The vegetative features are described as false thornveld. Meanwhile, the climatic condition is split into autumn, winter, spring, as well as summer seasons. A mean rainfall (480 mm) is habitually experienced annually in the summer whereas the mean temperature ranges between 19.2 and 27.6°C. The region's soil types are typically mudstone-derived as well as shale. Meanwhile, the vegetative features look like Savannah vegetation. The main vegetation/plant density of the location includes Cympopogon plurinodis, Themeda triandra, Cynodon dactylon, Sporobolus fimbratus, Digitaria eriantha, Sporobolus africanus, Aristida congeta, as well as Eragrostis (Idamokoro et al., 2019). The study conformed to the institution's animal research ethics board standards for conducting research and an ethical clearance certificate was obtained to carry out the research with reference number: GAJ011SVEL01.

Study Treatments and Experimental Design

The experimental approach (factorial) adopted for this study is a block design (randomized) which was utilized with the topography (landscape) as well as the tree species used as the key factors and the 100×100 m plots within the topography (landscapes) representing the blocks which were replicated. Fresh leaves obtained from twenty (20) arbitrarily chosen individual trees, each from Maytenus heterophylla and Acacia nilotica browse plant were obtained from the selected (2) plots manually (using hands). Individuals of the 40 collected samples were kept differently in labeled paper carrier bags. Labeling of the carrier bags was carried out in line with the site of harvest, the tree numbers as well as the tree species. The samples were worked upon in the laboratory located at the livestock and pasture department at the University of Fort Hare. The browse species were oven-dried to a standard temperature of 60°C (Babu et al., 2018). After that, the leaves were grounded (using the Polymix PX-MFC 90 D) before the chemical contents were determined aside from the phenolic constituent This was done after the leaf samples were firstly milled and then allowed to go

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through a sieve (with a 1 mm diameter size). Samples (ground) of leaves were retained in labeled tubes until when they were analyzed for their nutritional content.

**Measurements of Leave Samples**

**Proximate Components**

The Dry Matter (DM) content, total nitrogen, ADF, NDF as well as the Organic Matter (OM) component were estimated using standard approved method and equipment (macro-Kjeldahl technique (AOAC, 1995; ANKOM\textsuperscript{2000} Fibre analyser; ANKOM Technology, New York) (Soest et al. (1991)).

**Mineral Composition**

The present study analyzed the mineral content at the Cedara agricultural training institute of South Africa by utilizing the dry ashing micro as well as the macro minerals techniques with the use of the provided guidelines as given by the Agri-Laboratory Association of Southern Africa (Agrilasa, 1998).

**Phenolic Determination and Condensed Tannins**

**Extraction of Plant Samples**

A 500 mg dried sample was used with 75 mL of 70% methanol following approved laboratory standards.

**Phenolic Determination**

This was determined by using the Folin-Ciocalteu’s technique spectrophotometrically using an approved standard. The experiment was conducted in triplicate. After that, the phenol component was extrapolated from the gallic acid calibration graph formula; \( y = 0.0102x + 0.1861 \). R\(^2\) = 0.9994. The unit was presented as mg Gallic Acid Equivalent (GAE)/g from the equation CV/m.

From the given formula (i.e., CV/m) “C” represents the concentration as obtained from the calibration curve equation in mg/mL. In addition, “V” represents the volume of the extract that was utilized in the assay in mL. Finally, “m” represents the mass of the extract utilized in the assay in gram (g).

**Proanthocyanidin Determination**

0.5 mL of the pulverized plant extract with varied concentrations of the standard catechin (0.1-1 mg/mL) was used to determine the proanthocyanidin (condensed tannin) using the approved standard spectrophotometrically. The experiment was conducted in triplicate. Condensed tannin constituent was expressed using the calibration curve formula: \( y = 0.0009x \), R\(^2\) = 0.997 and was calculated as mg Catechin Equivalent (CE)/g using the formula, CV/m.

**Data Statistics**

Data were analyzed in a 2x2 design (factorial) with 2 varieties and 2 species using a software package, version 9.1.3. The GLM procedure as well as the least significant difference was employed for analysis. The significant level was kept at 5%.

The statistical model is given below:

\[
y_{abc} = \mu + S_a + GE_b + (S \times GE)_{ab} + E_{abc}
\]

where:

- \( Y_{abc} \) = Dependent variable
- \( M \) = Mean group
- \( S_a \) = Effect of browse plants
- \( GE_b \) = Impact of growth surrounding
- \( (S \times GE)_{ab} \) = Interaction impact between browse species and growth surroundings
- \( E_{abc} \) = Random error linked to observation abc

**Results**

**Proximate Analysis**

Chemical contents (cellulose, hemicellulose, OM, DM, ASH, ADF, NDF, N, ADL) of *Acacia nilotica* and *Maytenus heterophylla* species were presented on a DM basis and are shown in Table 1. Plot as well as browse species showed no significance (p>0.05) on all the measured chemical contents. Significant differences existed (p<0.01) between plot as well as browse species with regard to N, DM as well as CP components of leaf samples. In addition, the DM content from the two browse species is within the range of 839.50 and 855.67 g/kg. From Table 1, the result showed that the leaves from *Acacia nilotica* were higher in the amount of DM, N and CP in comparison to *Maytenus heterophylla* in the high landscape terrain. The CP constituent ranged from 8.72-22.64%. Conversely, the nitrogen component from the two species in the different landscapes ranged from 1.40-3.62%. Our result showed no significant difference (p>0.05) recorded in the NDF of leaves from *A. nilotica* in both the high and low topography. *M. heterophylla* showed high NDF in the low topography compared to high topography. The NDF content of leaves ranged from 36.81-44.62%. *M. heterophylla* had higher ash content than those from *A. nilotica*. The ash content from the two browse species Spanned from 61.29-142.43 g/kg. *A. nilotica* had greater OM content in high topography in comparison to *M. heterophylla*. The organic matter content of the plant species spanned from 716.73-820.55 g/kg. No significant (p>0.05) difference in the ADL of the two species from the two topographies. The ADL % of the two species ranged from 25.01-32.64. Furthermore, no significant difference (p>0.05) in cellulose and hemicellulose from the two species harvested from the different topographies. The cellulose % of the leaves spanned from 15.83-22.93 and the hemicellulose percentage spanned from 11.80-13.66%.
The effect of browse species and topography/landscape on major minerals is presented in Table 2. The two (2) browse species showed a major difference in the macro element constituent with relation to topography in both the low and the high landscapes. In the low topographic areas, most of these elements had higher chemical contents for both species than those from the high topographic locations. In the high landscape area, leaves from Acacia nilotica had higher potassium and phosphorus compared to those from the low topographic areas while Maytenus heterophylla had higher sodium contents in the high topographic location compared to the low landscape terrain. In the low landscape area, a significant difference (p<0.05) in calcium was noticed, with the Maytenus heterophylla having a higher Ca content than those in the high landscape.

The K (0.77-1.61%), Na (877.16-1735.98 mg/kg), Ca (1.25-3.71%) and P (0.12-0.22%) contents of forages had no significant (p>0.05) differences with respect to variations in the plant species. Meanwhile, the Mg content (0.20-0.22%) had significant (p<0.05) differences with regard to variations in browse plant species and topography. With regards to species, Maytenus heterophylla had higher Mg levels (1.08%) than the Acacia nilotica in both topographic locations. In addition, high Mg contents were observed for Maytenus heterophylla and Acacia nilotica respectively in the low topography in comparison to high topography.

### Mineral Composition

#### Macro Minerals

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#### Micro Minerals

The Micro-mineral (including Fe, Mn, Cu, Zinc and Al) from Maytenus heterophylla as well as Acacia nilotica
from the two different topographies are in Table 3. The Fe content of tree species ranged from 88.60-283.80 mg/kg. Leaves from low topography were higher in iron (Fe) content (185 mg/kg) compared to the ones from high topography (109 mg/kg). In the low topography, A. nilotica had higher (p<0.05) Fe content in comparison to M. heterophylla. The Mn content of tree species ranged from 30.60-96.80 mg/kg. Leaves from low topography had a higher Mn content (76.1 mg/kg) than those from high topography (35.9 mg/kg).

The Cu content ranged from 24.47-9.76 mg/kg. Leaves from low topography had higher Cu content (185 mg/kg) compared to the ones from high topography (109 mg/kg). In the low topography, A. nilotica had higher (p<0.05) Fe content in comparison to M. heterophylla.

The Mn content of tree species ranged from 30.60-96.80 mg/kg. Leaves from low topography had a higher Mn content (76.1 mg/kg) than those from high topography (35.9 mg/kg).

The Cu content ranged from 24.47-9.76 mg/kg. Leaves from low topography had higher Cu content (185 mg/kg) compared to the ones from high topography (109 mg/kg). In the low topography, A. nilotica had higher (p<0.05) Fe content in comparison to M. heterophylla.

Soluble phenolic content and Soluble condensed tannins.

The phenolic contents from M. heterophylla as well as A. nilotica are shown in Figs 1-2. The browse species as well as topography showed no significant (p>0.05) impact on SPh as well as SCTs. A significant (p<0.05) relationship between species and topography was recorded. Leaves from high topography had higher SPh and SCTs in comparison to low topography. The SPh content spanned from 28.77-44.97 mg/GAE/g while the SCTs content spanned from 44.84-98.32, respectively.

<table>
<thead>
<tr>
<th>Species</th>
<th>Landscape</th>
<th>Fe (mg/kg)</th>
<th>Mn (mg/kg)</th>
<th>Cu (mg/kg)</th>
<th>Zn (mg/kg)</th>
<th>Al (%)</th>
</tr>
</thead>
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<tr>
<td>A. nilotica</td>
<td>HLS</td>
<td>115.40a</td>
<td>41.20b</td>
<td>6.54b</td>
<td>33.00b</td>
<td>070.60a</td>
</tr>
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<td></td>
<td>LLS</td>
<td>283.80b</td>
<td>96.80b</td>
<td>9.76b</td>
<td>12.20b</td>
<td>258.80b</td>
</tr>
<tr>
<td>M. heterophylla</td>
<td>HLS</td>
<td>102.60ab</td>
<td>30.60ab</td>
<td>4.74ab</td>
<td>23.20ab</td>
<td>104.20ab</td>
</tr>
<tr>
<td></td>
<td>LLS</td>
<td>088.60ac</td>
<td>55.40ac</td>
<td>5.50ac</td>
<td>18.20ac</td>
<td>077.80ac</td>
</tr>
<tr>
<td>SEM</td>
<td>S</td>
<td>019.98</td>
<td>08.53</td>
<td>0.81</td>
<td>01.02</td>
<td>010.47</td>
</tr>
<tr>
<td></td>
<td>LS</td>
<td>019.98</td>
<td>08.53</td>
<td>0.81</td>
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<tr>
<td></td>
<td>S × LS</td>
<td>028.26</td>
<td>12.06</td>
<td>1.14</td>
<td>01.44</td>
<td>014.81</td>
</tr>
</tbody>
</table>

Significance:
- S = Species
- LS = Landscape
- S × LS = Species × Landscape

Means with the same column having different superscripts were significantly different (p<0.05); SEM = Standard error of the mean; Significance level: ** = Significant at p<0.01; * = Significant at p<0.05; NS = Not significant at p>0.05. Sp = Species; LS = Landscape; HLS = High Landscape; LLS = Low Landscape.
Discussion

The nutritional worth of browse species is affected immensely based on several environmental factors including the stage of harvest, climate change, season, soil nutrition and sometimes the topography/altitude (Bakshi and Wadhwa, 2012). Animal diets of more nutritional worth usually boost a high level of livestock efficiency (Makkar, 2018). The nutritional worth of diets is evaluated by how they are able to give needed nutrients to livestock for their growth, maintenance, as well as reproduction. Operational stratagems that enhance the utilization of diet by livestock depend on the knowledge of the nutritional components that constitute their feed as well as the interactions of the chemical contents to other factors such as plant species type, terrain of plants and season of harvest among others (Barnes et al., 2007).

Our study estimated the ingredient contents of *M. heterophylla* as well as *A. nilotica* browse species to determine the nutrient value of these plants and their significance to livestock in the current study area. *M. heterophylla* species was observed to show lesser DM, N and CP in comparison to *A. nilotica* harvested in the high landscape, which indicates their unreliability as a dietary resource of fiber and protein required by livestock in comparison to *A. nilotica*. Bakshi et al. (2011) in their study revealed a high DM and CP in some selected leaves from trees grown in high landscapes. Meanwhile, the lower CP content of *M. heterophylla* is a testament that protein is one of the limiting nutrients found in browse plants for livestock in the region (Idamokoro et al., 2016). In addition, given that *A. nilotica* leaves possess a higher dietary fiber and protein than *M. heterophylla*, it is safe to recommend that they may be utilized by farmers to graze livestock during drought season.

The study by Norton (1994), revealed that high NDF constituent (35% >) in animal feed lowers digestibility in comparison to the ones whose NDF content is less than 35%. However, the NDF components of the two species were greater in comparison to the one (36.81- 48.47%) revealed by Norton (1994), which shows that the digestibility of the studied browse plants by livestock is low. The high fiber constituent in *A. nilotica* and *M. heterophylla* could be expounded from their high phenolic components. In line with the findings of Barnes et al. (2007), it was reported that a high level of secondary chemical substance results in increased fiber contents in the plant. *A. nilotica* and *M. heterophylla* showed greater tannins, which possibly is the reason for their high NDF content. However, the tannins from leaves of tree species reported in another study were lower than the present study (Norton, 1994). The reason for the variation could be a result of several factors including the soil nutrients, the time of harvest of the plants and probably the human activities taking place in different study locations (Elbasiony et al., 2022). However, the result of our finding (tannin content) is similar to the study by Reed (1986) in line with tannin content.

High levels of phenolics may be caused by plants' adaptive methods to specific locations through evolution as well as the ones that may have evolved during grazing have protective strategies against attacks from predators (whether by insects or animals in the range). Some of the protective strategies (mechanisms) include lignification as well as secondary compounds which may negatively affect the dietary fiber of plants (Tabe et al., 1993; Moore and Jung, 2001). From the present study, the species and topography do not show a significant (p>0.05) impact on SPh and SCTs. Furthermore, a significant (p<0.05) interaction existed between species and topography. Leaves from high topography had higher SPh and SCTs in comparison to low topography (Figs 1-2). The reason for the variation in the SPh and SCTs contents may be due to the fact that the trees in the high altitude may have experienced more environmental stress than those in the low altitude. According to Ngwa et al. (2000), trees thriving in stressful conditions are likely to secrete more tannins which is their means of defense mechanism against browsing. Conversely, the SCTs for the high landscape are comparable to browse species grown in high altitudes in India (Bakshi and Wadh, 2012; Bakshi et al., 2011). Leaves from *M. heterophylla* had high ash content which may signify that the plants are in their early maturing stage. McDonald et al. (2002) revealed that as a plant germinates, the concentration of ash decreases. Again, Dobrowolska et al. (2011) reported that different growth stages in plants influence the presence of ash in them.

Leaves of *A. nilotica* from low topography had greater Fe, Mn, Cu, Mg and Al in comparison to the ones from high topography. The variations may have resulted from the leaching of nutrients in the study area. According to Thakur and Kumar (2020), micronutrients such as boron as well as manganese are vulnerable to leaching in some soils resulting in a lack of availability of the nutrients to be available for plant use. No significant difference in phosphorus was revealed in this study for all locations because P is immobile in soils as a result of adsorption as well as precipitation to mineral surfaces (Lehmann et al., 2003). In line with our findings, there was variation in the chemical components of browse plants with those growing at a low altitude having a higher elemental component as compared to those growing at a high altitude (Shah et al., 2019). However, Singh et al. (2010; 2022) reported that the nutritive values (chemical components) of high altitudinal browse trees were higher than those from the low altitude. Some of the factors that may be responsible for this variation in findings may be due to the season of harvesting of the plants.
(Navale et al., 2022) and geographical structure, i.e., elevation/topography/altitude (Shah et al., 2019; Elbasiouny et al., 2022). Other possible factors that may cause variation between browse plants grown on low landscapes and high landscapes include daylight, type of soil, precipitation, organic matter contents and temperature among others (Shah et al., 2019).

The Ca content present in the leaves of A. nilotica and M. heterophylla is comparable to those found in tree leaves grown in high topography of a previous study (Bakshi and Wadhwa, 2012). However, when compared to NRC (2001) recommendations (2.0%) for a balanced ration for cattle (dairy), calcium in M. heterophylla as well as A. nilotica (in low landscape) were greater than the requirements (Table 2), which could be beneficial for high yielding livestock during early stages of lactation. Conversely, the average concentration of Ca in A. nilotica and M. heterophylla leaves was found beyond tolerance limits (Bakshi and Wadhwa, 2012). Likewise, both the A. nilotica and M. heterophylla leaves in the present study were deficient in phosphorus and the majority of the microminerals when compared to the recommendation of the Research Council (NRC, 2001). It is therefore recommended that livestock in the study area should be provided with quality mineral mixture, to evade the element dearth in them.

Study Limitations

Regardless of the findings from our study, it is vital to acknowledge some limitations that are related to the current study. One such limitation is that the study did not consider the effects of seasonal variations on the nutritive status constituents in studied fodder. The study did not also consider the age/stage of harvest of leaves that were analyzed.

Conclusion

Leaf samples of A. nilotica and M. Heterophylla may be utilized by livestock as promising protein resources for livestock consuming poor nutritive grasses with minimal protein constituents, especially in the dry season, due to the fact that their CP constituent was higher than 15%, a nutritive value that cannot be provided by grasses for ruminants' use. Leaves from high topography showed high phenolic constituent, which is a confirmation that varied growth locations impact the chemical constituents of browse species. Furthermore, the two plot locations had related chemical components for several of the analyzed minerals even though those from low topography were more in comparison to those from high topography, which indicates that with respect to mineral nutrition, browse species from low topography may have more minerals for foraging livestock which is an additional benefit to grazing animals in the low topography than those in the high topography. Even though fiber is seen to be a non-limiting ingredient for livestock, the two analyzed browse species had varying prospects to be utilized as dietary fiber.

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Author’s Contributions

All authors equally contributed in this study.

Ethics

The study conformed to the animal research ethics board standards for conducting research and the study received an ethical clearance of GAJ011SVEL01.

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