CHEMICAL AND TANNIN COMPOSITION OF BROWSABLE SPECIES USED AS RUMINANT FEED SUPPLEMENTS IN THE VHEMBE DISTRICT OF SOUTH AFRICA

BY

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DECLARATION

I, Mahlako Kgabo Tryphina hereby declare that this dissertation was submitted in partial fulfillment of the requirements for the Masters of Science in Agriculture degree at the University of Venda hereby submitted by me has not previously been submitted for a degree at this or any other university, and in execution and that all reference material contained therein has been duly acknowledged.

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DEDICATION

To my Mom, Thamaris, brothers Solly, Janson and Douglas and sisters Esther, Rahab and Grace.

ABSTRACT

The chemical and tannin composition of *Acacia nilotica, Dichrostachy cinerea and Ziziphus mucronata* from high, medium and low rainfall areas were studied. Dry matter and ash, crude protein content, natural detergent fibre and acid detergent fibre were determined.. The macro elements were then determined by flame or hydride generation technique and micro using metrohm Ion Chromatography. The condensed tannins were determined using n-butanol-HCL colorimetric method The data were analyzed by analysis of variance for a 3 x 3 factorial in a completely randomised design using the General Linear Model (GLM) procedure of SPSS version 19. Averaged overall zones, *D. cinerea* had a higher ash content than the other two species. *Z. mucronata* had the highest CP content followed by *D. cinerea* and *A. nilotica*. *D. cinerea* had the highest NDF level followed by *A. nilotica* and then by *Z. mucronata.* Trees that grew in the medium rainfall zone had the highest CP level (153 g/kg DM) followed by those that grew in the low rainfall zone whilst those that grew in the high rainfall zone had the lowest CP content (101 g/kg DM) *A. nilotica* and *D. cinerea* did not differ in ADF and phosphorus but had higher levels than *Z. mucronata* ($P < 0.01$). *A. nilotica* had a high chlorine content (130 g/kg DM) than *the other tree* ($P < 0.01$). In addition the high and low rainfall trees had a high fluorine levels than the medium rainfall trees. *Z. mucronata* trees that grew in low rainfall areas had the highest total tannin content of 382 g/kg Mimosa tannin equivalent. The findings of this study showed that the three forage legumes species are high in condensed tannins but they are of good nutritive value and have potential for integration into ruminant livestock production in Limpopo Province of South Africa.

Keywords: Crude protein, Anti-nutritional, Minerals, Forage legumes, Agro-ecological

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LIST OF ABBREVATIONS

^0C – Degree Celsius

A. erioloba – Acacia erioloba

A. Karroo – Acacia Karroo

A. nilotica – Acacia nilotica

A. Sieberiana – Acacia Sieberiana

A. Tortilis – Acacia Tortilis

ADF – Acid Detergent Fibre

AOAC - Association of Official Agricultural Chemists

ARC – Agricultural Research Council

C. apiculatum - Chyrsocephalum apiculatum

C. molle – Chyrsocephalum molle

C. Zeyheri – Chyrsocephalum Zeyheri

Ca – Calcium

CL – Chlorine

CP – Crude Protein

CT – Condensed Tannins

D. cinerea – Dichrostachys cinerea

df – degrees of freedom

DM – Dry Matter

DMD – Dry Matter Digestibility

E – East

F – Flourine

FAO – Food and Agricultural Organization

G – Gram

GLM – General Linear Model

HT – Hydrolysable Tannins

K – Potassium

KG - Kilogram

M – Metres

MG – Milligrams

ML – Millilitres

MM – Millimetres

MT – Mimosa Tannins

N – Nitrogen

NDF – Neutral Detergent Fibre

NRC – National Research Council

NRF – National Research Foundation

NS – Non Significant

P – Phosphorus

P – probability

S – South

SANBI – South African National Biodiversity Institute

SCA – Supreme Court Appeal

SEM – Standard Error of Means

SPSS - Statistical Package for the Social Sciences

TBT – Total Bound Tannins

TCT – Total Condensed Tannins

TSCT – Total Soluble Condensed Tannins

Z. mucronata – *Ziziphus mucronata*

CHAPTER 1: INTRODUCTION

1.1Background

Fodder trees and shrubs have played a significant role in feeding domestic animals and they constitute a vital component in livestock productivity in the arid and semi-arid zones where about 52% of cattle, 57% of sheep, and 65% of goats are found (Petit, 2000;Petit and Diallo,2001; Ou'edraogo-Kone '*et al*., 2006). Trees and shrubs supply protein in harsh environmental conditions (Malaysian Agricultural Research and Development Institute,1990). They supply goats with the bulk of their nutritive requirements and complement the diet of cattle and sheep with vitamins and minerals, in addition to protein. These nutrients are deficient in bush straw during the dry season (von Kaufmann, 1986, Brenan,1983). Inadequate amount and low feed quality, particularly during the dry season, result in reduced livestock productivity in many tropical countries.

Many small-scale farmers in the Limpopo Province of South Africa keep livestock, particularly cattle, goats and sheep. Ruminant livestock productivity is low especially during the dry season, when there is a shortage of nutrients especially protein (Norton, 1994; Abdulrazak*et al.,*1996). Indigenous fodder trees offer potentially a suitable source of supplemental protein for ruminant livestock which would improve intake and digestibility of native pasture (Norton, 1994; Abdulrazak *et al*., 1996). Acacia trees form the third largest woody plant family in Southern Africa and are important ecological component of the bushveld vegetation that is prevalent in the Limpopo Province (Mokoboki*et al*., 2005).

1.2 Problem Statement

Animals suffer severe nutritional stresses in the dry season when the rangelands are of low nutritional value and usually in short supply of feed (Manyuchi and Ngongoni, 1993). Indigenous woody plants and shrubs occupy a large percentage of the ecosystem in Limpopo Province. Woody plants and shrubs can play a role in the feeding of animals during the dry season. The natural pasture which the animals feed on during dry seasons are low in protein content and high in fibre and lignin content which result in low feed intake (Baloyi, 2002). As a result of these adverse conditions, during the dry season animals lose weight, show poor body condition and have low milk yields, low conception rate and increased calf mortalities, all of which culminate into heavy economic loses to the smallholder farmers (Ngongoni*et al.,* 2006). Feed costs of protein-rich supplements are ever increasing and becoming scarce especially in the rural areas where the feed companies are few. As a result, livestock production becomes uneconomical to smallholder farmers who keep cattle, goats and sheep. Rural farmers make efforts to protect fodder trees and shrubs so that they can provide alternative protein source for livestock production. This can be realized by the increase in livestock production and reduction of feed costs (Jain and Baniwal, 1982; Joshi and Singh, 1990; Nicholison, 1992; Kabirizi, *et al.*, 2004; Miah, *et al.*, 2005).

1.3 Justification

The increasing demand and subsequent high costs of conventional feedstuffs has created the need for sustainable alternatives like indigenous natural feed resources. This search for alternative feed resource has, over the past few decades, rekindled research interest in the use of indigenous fodder trees and shrubs as sources of nutrients for ruminants as well as non-ruminants (D'Mello, 1992; Aletor and Omadara,1994). They are very important in communal livestock farming especially in winter and drought conditions when there is scarcity of herbage and animals lose weight due to nutritional imbalance in the little feed available (Preston and Leng, 1987; Mannetje, 1982). Animals suffer severe nutritional stress in the dry-season when rangelands are of low nutritional value and usually in short supply (Manyuchi and Ngongoni, 1993). The crude protein content of the veld grasses rapidly declines from 100g/kg in early summer (November) to 20g/kg in winter (May-June) while the fibre content increases progressively (Elliott and Fokkema, 1961).

Of over 5000 trees and shrubs listed as being suitable for feeding livestock in Africa, it has been suggested that only 80 are of real fodder value while five may be recorded as good (Okoli, *et al.,* 2003). This scenario underscores the lack of information on the values of many of those plants and the need to scientifically evaluate their nutritive value. The use of fodder trees and shrubs can supplement the quantity and quality of pasture for grazing livestock, despite their potential value in prevailing small farm systems (Devendra, 1983). These alternative feed merits increased research and development in the future because there is an inadequate knowledge on various aspects of their potential use (Devendra, 1990).

1.4 Objectives of the study

The overall objective of the study was to evaluate the nutritive value and anti-nutritional factors of indigenous trees and shrubs found in grazing areas of Vhembe district in the Limpopo Province, of South Africa.

The specific objective is to determine the chemical composition and tannin content of browsable fodder trees.

1.5 Hypothesis

Null hypothesis: Browsable species are high in crude protein and tannins.

CHAPTER 2: LITERATUREREVIEW

2.1 General introduction

In the past years, in academic and development forums on natural resources conservation, specialists have discussed the need for shifting from convectional livestock methods towards more sustainable production systems, capable of improving yields and quality (Mosquera-Losataet *al*., 2005). Fodder trees and shrubs have been recognized as valuable and accessible food sources, which contribute to solving some nutritional constraints for livestock during the dry season (Sanchez and Rosales, 1999; Dagang and Nair, 2003).

Livestock production constraints is mainly due to feed shortage, anti-nutritional factors and fodder scarcity. The scarcity is seasonal and during this long dry period fodder trees and shrubs are the major source of green fodder (Joshi, 1988). The utility of leaves, pods and edible twigs of shrubs and trees as animal feed is limited by the presence of anti-nutritional factors. These anti-nutritional factors in plants seems to be as a way of storing nutrient or as a means of defending their structure and reproductive elements (Harborne, 1989).

2.2 Livestock feed supply and its quality

Maize stovers are an abundantly available feed resource during the dry season. However, their low protein content and high fibre and lignin content limit their efficient utilization by ruminants due to limits set by the digestibility in maize stover (Harqueet *al*., 1986; Manyuchi *et al.,* 1997). The importance of tropical forage legumes (Topps, 1992) and browse legumes (Le Houerou, 1980) as sources of valuable protein for ruminant animal production in the tropics has been acknowledged (Norton, 1982). Supplementation of low quality roughages with forage legumes in ruminant diets has been shown to increase intake (Mosi and Butterworth, 1985) and digestibility (Minson and Milford, 1967).

The beneficial effect of forage legumes on animal performance have been demonstrated in ruminant grazing studies (Clatworthy, 1978). Leaves from indigenous trees are an important sources of nutrients for herbivores animal in South Africa (Mokoboki, 2005). According to McDonald and Ternouth (1979), commercial and small-holder farmers make extensive use of edible treessuch as *Colophospermum mopane* (Mopane, 198) and *Peltophorum africanum* (Weeping Wattle, 215) as livestock feed during the dry-period of the year. The leaves from these trees are usually rich in protein and their chemical composition tends tovary little within a season. The utilization and nutrients from many tree species are however limited by anti-nutritional factors.

2.3 Commercial protein-rich supplements

During the dry season,ruminants in the smallholder farming sector in Zimbabwe are generally supplemented with commercial high protein supplements such as cubes, blocks and concentrates with poor quality roughages such grain stover (Baloyi, 2002). Another way of increasing animal production on either veld or sown pastures is to introduce legumes which can grow and spread in summer and/or winter (Topps and Oliver, 1993). They produce herbage, seeds and pods which are relatively higher in protein, energy and minerals than that of grasses, particularly in late summer and in the dry season (Baloyi, 2002). The crude protein (CP) content of legumes is usually higher than that of the associated grasses at similar ages or stages of growth (Norton, 1982). It is usually in excess of that proposed as the minimum requirement for lactation (120 g/kg DM) and growth (113 g/kg DM) in ruminants Agricultural Research Council (1984).

2.4 The nutritive value of forages

The nutritive value of feed is measured by its ability to provide nutrients to an animal for maintenance and growth, in the absence of toxic factors, is a function of feed digestibility and voluntary feed consumption. It is now recognized that unlike grasses, there is often a little positive relationship between feed intake and digestibility; thus, the nutritive value of the browse only be an accurately determined by feeding trials (Norton, 1994). Legumes have become increasingly important as protein rich forages to supplement basal diets of either low protein grass or poor quality roughages such as maize stover for ruminant livestock. Although forage legumes have the potential to provide a valuable source of nutrients to animals, a major limiting factor to their use is the presence of anti-nutrients such as tannins and saponinswhich when ingested may cause deleterious effects ranging from loss of appetite to death (Baloyi, 2002).

Browse leaves and fruits are more valuable as supplement to low quality feeds than as the sole feed due to their high nutrient contents and limited supply. Protein supplements from plant and non-protein nitrogen sources are often used to improve animal performance. However, the use of browse sources by herbivores is restricted in many cases by defending mechanism related to high tannin contents in tree foliage (Silanikoveet *al*., 1994). The inverse relationship between high tannin level in forage and palatability, voluntary intake, digestibility and nitrogen retention in herbivores is well established (Silanikove *et al*., 1994; and Silanikoveet *al.*, 1996).

Papachristou and Nastasis, (1993a;b) Dumont *et al.* (1995) reported that both woody and herbaceous plants are considered important contributors to grazing animal nutrition. During summer, when herbaceous plants are dormant and herbage is of low quality and scarce, forage of woody vegetation, also known as browse (leaves and twigs) has to meet nutritional requirements of grazing animals (Le Houerou, 1993; Papanastasis, 1993a;b). Studies by Dini(1993), Plastis and Papanastasis (1993), Papachristou and Papanastatis (1994) and Papachristou (1997) suggested that certain deciduous woody species are most promising as animal feed during the dry season in terms of productivity, establishment and nutritive value. Tree fodder contains high levels of crude protein and minerals and many show high levels of digestibility. They are readily accepted by livestock and presumably because of their deep-root systems, they continue to produce well into the dry season. However, anti-nutritive factors can be a problem in some species (Patterson*et al.*, 1998). The palatable and good quality forage always deteriorates or disappears as a result of overgrazing.

However, high concentration of some minerals such as sodium chloride and selenium may be found in browse species (Underwood, 1981; Olsson and Welin-Berger, 1989), Vitamin A, B and C are present in varying concentrations in leaves, fruits and seeds of some trees and shrubs (Bergeret, 1986). Carotene content is usually high during early growth and declines rapidly with maturity, except in evergreen shrubs which tend to retain it for long periods (Ibrahim, 1981)

2.5 Anti-nutritional factors

Fodder tree leaves and fruits often contain anti-nutritional factors and the scope, nature and action of tannins, cynogens, saponins, alkaloids and non-protein amino acids have been extensively reviewed (Kumar and D'mello,1995). The hydrolysable tannins (esters of gallic acid), condensed tannins (proanthocyanidis) and other phenolic compounds (lignin, sterols,etc.) in browse species affect the nutritive value of leaves and fruits (Tanner *et al.*,1990).

Hydrolysable tannins are usually toxic but have effects over a short period of time. Condensed tannins, depending on nature and concentration may be beneficial or detrimental. Both tannins and high levels of phenolic compounds may depress intake through effects of feeding behaviour. Low intake and digestibility of browse are due to these deleterious substances which tends to reduce the nutritive value of the trees and sometimes can be toxic to animals (Mokoboki, 2006) However, the toxicity of tannins depends upon concentration in fodder and the rate at which forage is consumed (Storrs, 1992).

The presence of thorns, prickly leaves, volatile oils, suberised and highly lignified leaves, alkaloids and tannins ensure survival in these environments, but decrease the palatability and nutritive value of the foliage from these trees for animals (Otsyina and Mckell, 1984). Therefore, forage scarcity is prevalent and there is an urgent need for increase in feed resources in the arid zones of the country (Hopkins and Nicholson, 1999; Osman and Ghassaeli, 1997). Barry (1989) suggested low concentrations of condensed tannins (20 to 40 g/kg DM) had beneficial effects in protein protection in the rumen. Condensed tannin-protein complexes tend to be stable in the rumen pH (4-7). Therefore, they will escape degradation in the rumen but these complexes dissociate on exposure to the abomasal pH (2.5 - 3.5) and that of the small intestine (pH 8) (Barry and Manley, 1984; Waghorn *et al*., 1987;)

2.6 Mineral content

Tropical legumes generally maintain higher sulphur and calcium concentrations in plant tops than the companion tropical grasses provided no soil on environmental factors detrimental to sulphur availability. Sulphur deficiency, like protein, depresses microbial activities in the rumen (Whiteman, 1980). In addition, the legumes are more efficient at extracting calcium from soils low in calcium. Phosphorus concentrations are more variable, depending on the legume species (Cameron, 1988; Norton and Poppi, 1995) and have been reported to be generally low in the legumes provided phytates are low. However, the differences between levels of phosphorus in legumes and grasses growing in the same climate and soil are small.The phosphorus content of plant tissue declines with maturity. Legumes have higher calcium content than grasses generally. During the rainy season, animals depend on the natural pastures and forages to meet their nutrients requirements like essential minerals. The cold and dry seasons in Limpopo province is characterized by less and/or no vegetative growth. At this period ruminant livestock depend on crop residues and poor standinghay on the grazing lands to obtain their nutritional requirements (Aganga *et al*., 1998).

2.7 Climate and availability of livestock feed

Arid and semi-arid lands refer to those parts of the world where rainfall is insufficient or barely sufficient for satisfactory crop growth. Arid lands in the tropics refer to those areas receiving between 25-200mm annually and semi-arid lands refer to those areas with mean annual rainfall total of 200-800mm (Grove, 1977). Arid and semi-arid lands constitute about 50% of the total land area of the world. These areas are characterized by low and unreliable rainfall, low productivity, large human and livestock populations. Livestock production plays a vital role in production systems and in the livelihood of the people in the arid and semi-arid zones. Fodder trees and shrubs often called 'browse' are used as supplements to livestock especially during the dry season and fodder stress periods (LeHouerou, 1981; Otsyina and McKell, 1984; Dicko and Sikena, 1992, Lefroy*et al.,* 1992; Atta-krah, 1993). They also serve other useful purposes such as provision of food, drugs, firewood and building poles and recycling of nutrients. Not all types of fodder trees and shrubs perform well in each area. Different types of trees and shrubs are suited to different climates (depending on the temperature and the amount of rainfall), altitude and soil types.

In South Africa cattle, sheep and goats play an important role in the livestock industry. However these animals depend on the veld as their basic feed. South Africa is semi-arid,the temperature in Limpopo province varies between –2.5^{0}C and 40^{0}C, with an average evaporation of 1750 - 2500 mm per annum. The climate varies from unusual extremes of temperature and high humidity. The climate is extremely hot during summer (Matlebjane, 2009). These adverse conditions result in low productivity as animals lose weight due to shortage of protein-rich feeds which are expensive and unavailable (Ngongoni*et al.,* 2006), This results in reduced feed intake and digestibility, rates of passage of digesta and animal production or even deaths through under-nutrition (Manyuchi, andNgongoni 1993).

2.8 The importance of fodder trees and shrubs

Fodder trees and shrubs (browse) form part of the natural vegetation and are accessible to the majority of small holder farmers (Tedonkeng *et al.,* 2006b). Several recent reviews and studies in various parts of the world, have shown that browse trees and shrubs play very important role, mostly as supplements, in the nutrition of livestock in arid and Semi-arid lands of the world (Le Houerou, 1980; Otsyina and Mckell1984; Tothill*et al.*, 1989; Atta-krah, 1990; Lefroy *et al.,* 1992; Otsyina and Dzowela, 1995). Besides being used as feed, they have additional benefits when integrated into farming systems. These benefits include fuel, wood and timber, increased soil fertility (leguminous species) control of wind erosion, shade for man and livestock, folk medicines, etc (Le Houerou, 1980). Tedonkeng *et al.,*(2006a) observed that deficiencies in feed quality could be corrected by the addition of herbaceous legumes or multi-purpose trees to the basal diet.

The importance of browse legume trees and shrubs is also increasingly acknowledged throughout the world (Le Houerou, 1980). Their major value is that they provide protein,vitamins and mineral elements which are lacking in grasslands pastures during the dry season. They also provide standing feed reserves so that animals are able to survive critical periods of under nutrition (Baloyi, 2002). The digestibility dry matter (DMD), which is related to nutrient composition, varied widely among tree and shrub species. Skarpe and Bergstrom (1986), working in Botswana with Kalahari woody species has reported a range in digestibility from 38 to 78 %. The concentration of calcium and potassium is usually higher than that of other minerals; the average being around 1 to 1.5% for Southern African browse (Walker, 1980).

When farmers select trees for fodder production they should look for several characteristics, such as leaves and pods that contain a lot of proteins. Trees should produce many leaves and regrow easily after frequent grazing or browsing. The edible part of the tree should not contain toxins. The trees need to have a high palatability, which means that the animals like to eat them and can digest them well. The trees must preferably be tolerant to draught, pests and diseases and do not compete too much with other crops. For example, good fodder trees should form deep roots in order to avoid competition with shallow rooted crops for water (Rasheko, 1996; Roothaert and Karanja, 2000, van Tol, 2004;).

2.9 Description of the browsable species

2.9.1 Ziziphus Mucronata

This is widely known as buffalo thorn. Is found throughout South Africa; grows in areas dominated by thorny vegetationin both temperate and tropical climates. Its presenceindicates the present of water. *Ziziphus mucronata* are fast growing shrubs of small to medium-sized tree reaching a height of 10 m. It can survive in a variety of soil types, occurring in a wide variety of habitat. The leaves ovate to brodly ovate, 30-80 x 20-50 mm, glossy dark green above, lower surface slightly hairy or with coarse brownish hairs at least when young, margin finely toothed over upper two thirds: stipules, spinescent,one hooked the other straight or plants unarmed. Buffalo thorns have distinctive zigzag branch lets and hooked and straight thorns; these sharp little thorns of the buffalo trees are in pairs (Palmer and Pitman, 1972, 1973).

The leaves are browsed by game and stock. Widely used for magical and medicinal purposes and in traditional religious rituals. The juice from the tree is used by the San as a mixing agent for arrow poison obtained from the larvae of *diambhidia nigroornata*, a beetle that feeds on the leaves of *Commiphora Africana*(Braam and Piet van wyk, 1973). Flowers are in auxillary clusters, they are small and yellowish-green in colour. It bears fruits which are greenish before ripening and red-brown when ready; they are up to 30mm in diameter and shiny (Braam and Piet van wyk, 1997). The fruit is edible but not tasty. The main stem is green and hairy young. The bark is hard, reddish brown in colour roughly molted grey,cracked into small rectangular blocks. The wood is heavy and good for tool handles, spoons and general purpose timber when sunpapered smoothly.

2.9.2 Dichrostachys cinerea

Dichrostachys cinerea is a thorny, fast growing woody bush and semi-deciduous tree, native to Africa (Orwa *et al*., 2009; Göhl, 1982; Food and Agricultural Organization, 2011). It occurs in bushveld, often invasive and thicket forming, particularly in overgrazed areas, water land, road sides, fields and other disturbed areas. The species prefers clay-like soils, but found in all soil types (Global Invasive, 2008). They live for very long time and withstands cold. Shrubs or small tree is characterized by open round crown, often flat tapped, the intertwined or mattered canopy is heavy with fine feathery foliage (South African National BiodiversityInstitute, 2011; Orwa *et al*., 2009). They grow up to 6 metre in height, multi-stemmed and fast growing weedy bush (Palmer and Pitman, 1972). The leaves are bi-pinnate with 4-19 pairs of pinnae, 3-10cm long each carrying up to 41 pairs of grey-green leaflets per pinna. At the base of at least upper and basal pinnae pairs there is a very small petiolar gland (Braam and Piet van wyk, 1997).

These thorny trees havea bicoloured lovely flower which turns to grow larger and less hairy leaves and leaflets. They are hanging 40-60mm long pink and yellow bisexual flowers elongated pendulous spikes and sterile with pleasant odour and bisexual in October and January (Global Invasive, 2008). The thorns are very little, sharp and strong, generally grow up to 8 cm long. The bark is light brown to grey brown and pods in distinctive curled clusters (70-100 mm diameter) which fall without splitting (May and September) and yields a strong fibre (Palmer and Pitman, 1972). Sickle bush is a multipurpose tree and its pods and seeds are edible (Ecocrop, 2011). Sickle bush wood is heavy and termite-resistant and is thus used to make posts and fences. It can also be used for fuel since it makes no smoke and burns slowly (Orwa *et al*., 2009). The bark is fibrous and used for twine (Orwa *et al*., 2009).

Leaves and fruits are relished by wild and domestic ruminants (Aganga and Motshewa, 2007).Sickle bush is browsed during the dry season. The pods, twisted and borne in large bunch, are much relished by cattle and game (Göhl, 1982). The pods ripen during scarce periods. They are indehiscent and can be stored without problem (Mlambo *et al*., 2004), making sickle bush a valuable fodder in dry areas (FAO, 2011). The roots are used against snake bite for relief from pain. The crowded dark brown pods are very nutritious and are eaten by a wide variety of game and stock animals.

2.9.3 Acacia nilotica

Acacia nilotica is a small medium-sized tree with a somewhat flattened or surrounded crown; occurring in bushveld. Spines slender; usually deflexed backward leaves with 5-11 pairs of pinnae, hairless or sparsely covered by hairs; leaflets 12-36 pairs per pinna; or 2 petiole glands usually present, gland present at the base of the upper pinnae pairs. Flower in globose heads, golden yellow (Braam and Piet van wyk, 1997). The trees flowers irregularly but usually from June to September, with podding occurring between January and May (Khan, 1970). There is extensive leaf abscission between April and May, with refoliation occurring between May and August (Khan, 1970).

This corresponds to exfoliation in the latter part of the dry season, with subsequent refoliation at the onset of the wet season, where plant growth appears to be influenced by moisture availability, temperature affects flowering and fruiting (Khan, 1970). Carter and Cowan (1988) and Carter*et al.*, (1991) claims that *Acacia nilotica* flowers from March to June with ripe pods being produced from November to January.

Acacia nilotica has a wide tolerance for rainfall. It has been found in areas receiving less than 230 mm per year and areas receiving more than 1500 mm per year (Carter*et al.,* 1991; Fagg, 1992; Carter and Cowan, 1993). They are also reported to be intolerant of frost (Carter *et al.,* 1991) and Fagg (1992) reported that the plant is frost tender when young. It grows in areas where the mean monthly temperatures of the coldest month is 16^{0}C (Gupta, 1970 cited in Carter*et al.,*1991) and it can withstand temperatures up to 50^{0}C (Fagg, 1992; Adjers and Hadi, 1993).

The pods are flat, smooth, black deeply constricted between each seed, the position of which is marked by a distinct raised pump on the valves, straight or slightly curved (Brenan, 1983). Sweet scentered when crushed, inde-hiscent, breaking up transverserly between seeds after falling. The wood is used for fencing posts and firewood; pods are eaten by game and stock, but are toxic to goats. Bark exudes an edible gum, and is used medicinally.

3.1 Site description

The experiment was conducted in three agro-ecological regions, high, medium and low rainfall areas represented by Mapila (22^0 54'40 "S and 30^0 03'10"E), Gogobole ($23^0$04'38"S and 29^0 46'26"E) and Davhana (23^0 12' 24"S and 30^0 27'34"E) villages respectively in the Vhembe district of South Africa. Figure 3.1 shows the villages in the three agro-ecological zones, namely Ha-Mapila, Davhana and Gogobole.

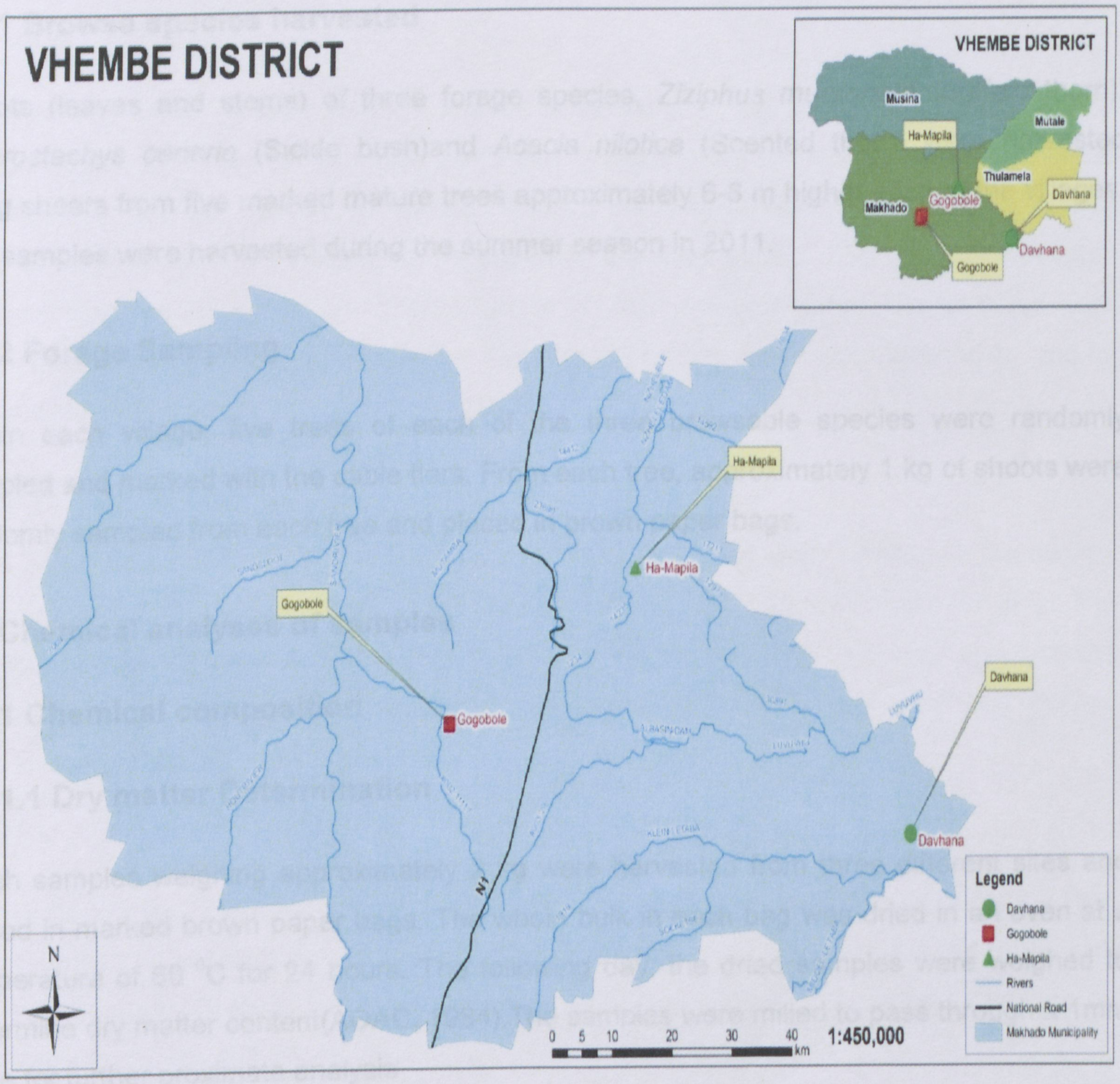

Figure 3.1: An illustration of the Vhembe district map and three different agro-ecological zones.

The rainfall in these zones starts from October and ends in March. The average annual rainfalls are 818.5mm, 689.25mm and 473mm for Mapila, Davhana and Gogobole respectively. The soils at Gogobole and Mapila are predominantly clay-loams, while at Davhana it is mostly sandy clay loam. *Acacia nilotica* was the dominant species in Gogobole and Mapila, while *Ziziphus mucronata* was the dominant species at Davhana.*Dichrostachys ceneria* was sparsely populated in all the villages.

3.2 Forages

3.2.1 Browse species harvested

Shoots (leaves and stems) of three forage species, *Ziziphus mucronata* (Buffalo thorn), *Dichrostachys ceneria* (Sickle bush)and *Acacia nilotica* (Scented thorn) were harvested using shears from five marked mature trees approximately 6-8 m highat each of the villages. The samples were harvested during the summer season in 2011.

3.2.2 Forage Sampling

Within each village, five trees of each of the three browsable species were randomly sampled and marked with the cable tiers. From each tree, approximately 1 kg of shoots were randomly sampled from each tree and placed in brown paper bags.

3.3 Chemical analyses of samples

3.3.1 Chemical composition

3.3.1.1 Dry matter Determination

Fresh samples weighing approximately 2 kg were harvested from three different sites and placed in marked brown paper bags. The whole bulk in each bag was dried in an oven at a temperature of 60 ^{0}C for 24 hours. The following day, the dried samples were weighed to determine dry matter content(AOAC, 1984).The samples were milled to pass through a 1mm sieve for further proximate analysis

3.3.1.2 Ash determination

Labelled crucibles plus about 2g ground dried sample were placed in a muffle furnace at 600 ^{0}C overnight. A metal thong was used to transfer hot crucibles to a desiccator in a cool room to cool for 1 hour; the masses were then recorded for ash determination (AOAC, 1984)

3.3.1.3 Crude Protein determination

Digestion

Duplicate 1 g of the ground forage samples were weighed on whatman filter and put in Kjeldahl flasks. Each set run with one blank and the laboratory standard forage sample was also run to gauge acceptability of the run. Boiling chips (2-3) were placed in a flask with sample in one kjeldahl packet and 30 ml sulphuric acid was added. The ventilation fan and water aspirator of the kjeldahl unit were turned on for fume extraction. The samples were digested for 2 hours and then flasks were then hours and the flasks were allowed to cool for 30 minutes.

Distillation

About 50 ml boric acid was put in 500 ml Erlenmeyer flasks for each sample 100ml sodium hydroxide and 250 ml water was added and turned on high burners for condensing. The burners were turned off and flasks were moved off the condenser tubes when the distillate reaches 200 ml. The distillation took about 30 minutes and water was shut off and kjeldahl flasks were washed when all done.

Titration

The distillate in the Erlenmeyer Flasks was titrated using 0.1 N sulphuric acids until the blue liquid returns to a purplish pink colour. The number of ml of sulphuric acid used on burette was then recorded. Calculations for crude protein (CP) values were calculated by multiplying the N value by factor of 6.25.

3.3.1.4 Neutral Detergent Fibre determination

Accurately 1.0 g of the ground samples were placed in the digestion vessel and 100 ml of natural detergent solution, 2 ml of dacahydronaphthalene and 0.5 g sodium sulphite were added. The mixture was heated to boil and the heat was reduced as boiling commenced to prevent foaming from the onset of boiling and allowed reflux for 60 minutes. The weighed sintered glass crucibles were placed on filtering apparatus. The samples and contents of flasks were transferred to crucibles. The contents were filtered in low vacuum initially and gradually the vacuum increased. Minimum of hot water were used to rinse samples into crucible. The vacuum was shut off and breaks of residues were washed with hot water followed by two washes with acetone. The crucibles were dried at 105 ^{0}C overnight and weighed. The residues are neutral detergent fibre (Robertson and van Soest (1981).

3.3.1.5 Acid Detergent Fibre Determination

Accurately 1.0 g of the sample was weighed into the reflux flask beaker. Cold acid detergent solution and 2 ml of dacahydronaphthalene were added to prevent foaming. The samples were allowed to heated to boil and reflux for 60 minutes from onset of boiling. Light suction initially was used to filter through tared crucible. The crucibles were washed twice with acetone and the filtered residues were dried in an oven at 105 ^{0}C overnight. The crucibles were transfered to cool in a desiccators and weight (Robertson and van Soest 1981).

3.3.2 Minerals determination

The forage legumes were ashed in a CNW Model SXL muffle furnance at the temperature of 600^{0}C overnight. The ashedmaterials werethen digested in 1M nitric acid. Each digested material was then made to a volume of a 100ml volumetric flask, and introduced to the autosampler for analysis of calcium (Ca), phosphorus (P), potassium (K), chlorine (CL) and fluorine (FL).

3.3.2.1 Calcium, Phosphorus and Potassium determination

The varian spectrophotometer instrument (model AA 20) equipped with a graphite tube atomizer (model GTA 110) with a programmable sample dispenser that diluted standards to the required concentration was used. Calibration of Ca, P and K elements was done using the merck standards. The three macro elements were then determined by flame or hydride generation technique (AOAC, 2000).

3.3.2.2 Chlorine and Flourine determination

Calibration of chlorine and fluorine trace elements was done using deionized water and merck standards to prepare samples. Each sample was then injected into the sample vials.

The two elements were then determined using metrohm Ion Chromatography (Model 850 Professional compact), (AOAC, 2000)

3.4 Analysis of Condensed Tannins (Proanthocyanidin)

3.4.1 Extraction ofthe CondensedTannins

The condensed tannins were determined using n-butanol-HCL colorimetric method (Reed, 1982). Aqueous acetone (70:30 v/v; acetone/water) was used to extract the condensed tannins before analysis by the n-butanol-HCL colorimetric method (Reed, 1982).

Duplicates of 200mg of each ground sample was put into separate 10-ml screw cap test tubes.10 ml aqueous acetone was added to each sample and the test tubes were shaken by hands and allowed to stand for 24hours at room temperature with occasional swirling (Makkar, 1995). The supertanant extracts were decanted off into storage bottles. The residues were freeze dried (Edwards Freeze Dryer, Super Monodulyo, Edwards Calibration, Eastbourne, UK) for 24 hours to allow analysis of total insoluble tannins. The analysis was done on the same day of extraction.

3.4.2 Determination of Condensed tannins using the n-butanol-HCL method

Condensed tannins were assayed as proanthocyanidins by the n-butanol-HCL (95:5; v/v) (Reed *et al.*,1982) method as modified by Makkar (1995). The analysis was carried out separately on the supertanants of aqueous acetone (70:30 v/v) extract (extractable proanthocyanidins), their freeze-dried extracted residues after extraction (Insoluble or bound proanthocyanidins) The extracts (0.5 ml or 10 ± 2mg) of freeze dried residue, after proanthocyanidins extraction, or NDF residues (Makkar and Singh, 1991) was put into 15ml Pyrex tubes. The n-butanol-HCL reagent (95:5v/v) was added to give levels of 3ml for the extracts and 6 ml for dried residues. An initial run was first carried out to establish the appropriate dilutions to be used for the extracts, and the amount of butanol-HCL reagent was added to freeze-driedresidues. The trial runs were necessary in order to avoid absorbency readings higher than 0.6, above which the relationship between concentration and absorbency may be non-linear.

The tubes were capped and transferred to a water bath that had been pre-heated and maintained at 100^{0}C. After one hour the tubes were removed, cooled and the absorbency was read at 550 nm using Beckman Coulter DU530 UV vis Spectrophotometer for life sciences.

3.4.3 Preparation of the standard curve

Stock solution of Mimosa (MT) tannins (1.0 mg/ml) were prepared in aqueous acetone (70:30 v/v) by adding 100 mg of mimosa powder (Mimosa Extract Company (Pty) Ltd, Pietermaritzburg, South Africa) to 100ml aqueous acetone over 24 hours. From the stock solutions, working standards (0.0, 0.2, 0.4, 0.6, 0.8 and 1.0 MT mg/ml) were prepared by separately diluting with aqueous acetone. The working standards were then treated in a similar way to the sample extracts, and their absorbencies (A_{550}nm) were used to plot the standard curves of absorbency against concentration and the corresponding regression equations was obtained. The concentration of the proanthocyanidins in the extracts and residues were estimated from the regression equation.

3.5 Statistical methods

The data were analyzed by analysis of variance for a 3 x 3 factorial in a completely randomised design using the General Linear Model (GLM) procedure of SPSS version 19 (2011). Three agro-ecological zones and three species were studied. The statistical model used to analyze the data was:

$$Y_{ijk} = \mu + S_i + Z_j + (SZ)_{ij} + e_{ijk}$$

where Y_{ijk} is the k^{th} sample of the i^{th} speciesin the j^{th} zones

μ = overall mean

Z_j = effect of the i^{th} species

S_i = effect of the j^{th} zone

$(SZ)_{ij}$ = effect of the interaction between the i^{th} species and the j^{th} zone.

E_{ijk} = Residual

Tukey's w procedure (Steel and Torrie,1981) was used to compare treatment means.

CHAPTER 4: RESULTS

4.1 Chemical composition

Dry matter (DM), ash, crude protein (CP), neutral detergent fibre (NDF) and acid detergent fibre (ADF) of *Acacia nilotica*, *Dichrostachy cinerea* and *Ziziphus mucronata* forage legume species harvested from different agro-ecological zones are summarized in Table 4.1. Ash, CP, NDF and ADF contents were significantly influenced ($P < 0.01$) by forage species. Averaged over all zones all the three(*A. nilotica, D. cinerea and Z. mucronata*) species had similar DM contents but *D. cinerea* had a higher ash content than *A. nilotica* and *Z. mucronata* ($P<0.05$) which did not differ from one another ($P> 0.05$) *Z. mucronata* had a higher CP content(151g/kg DM) than *A. nilotica*(109g/kg DM)and *D. cinerea*(127g/kg DM).The *D. cinerea* had a higher ash content than *A. nilotica* ($P< 0.05$).The*D. cinerea* had the highest level (548 g/kg DM) of NDF followed by *A. nilotica*(476 g/kg DM) and *Z. mucronata*(409g/kg DM)had the lowest level of NDF ($P< 0.05$). *A. nilotica* and *D. cinerea* did not differ in ADF content and both had on average about 5% more ADF than Z. mucronata.

The agro- ecological zones did not have any effect on ash ADF content ($P < 0.05$) but the trees that grew in the high rainfall zone had the highest DM content followed by those that grew in the low rainfall zone whilst the trees that grew in the medium rainfall zone had the lowest content of DM ($P< 0.05$). Trees that grew in the medium rainfall zone had the highest CP level ($P < 0.05$) and those that grew in the high rainfall zone had the lowest ($P<0.05$). The NDF contents were not significantly ($P > 0.05$) different between trees in the medium and low rainfall zones but these trees had a significantly lower NDF content ($P < 0.05$) than those that grew in the high rainfall zone.

The ash and ADF contents ($P < 0.05$ and 0.01 respectively) were influenced by an interaction between species and zones. The zone did not have any influence on the ash contents of *A. nilotica* and *Z. mucronata* but for *D. cinerea* trees that grew in the low zones had a significantly ($P < 0.05$) high ash content than those that grew in the high and medium zones. Zone did not have any effects ($P > 0.05$) on the ADF contents of *A. nilotica* and *D. cinerea* but for *Z. mucronata* trees that grew in the low rainfall zones had significantly ($P < 0.05$) lower ADF contents than those that grew in the high rainfall zones.

Table 4.1. Means and standard errors for dry matter, ash, crude protein, acid detergent fibre and neutral detergent fibre of *Acacia nilotica, Dichrostachys cinerea and Ziziphus mucronata* species found in different agro-ecological zones

Forage species	Zones	[1]DM (g/kg)	Ash (g/kgDM)	[2]CP (g/kgDM)	[3]NDF (g/kgDM)	[4]ADF (g/kgDM)
[5]A. nilotica	High	639^{abc}	46.8^{b}	81.6^{f}	514	525^{a}
	Medium	560^{bc}	41.4^{b}	137^{bc}	422	496^{a}
	Low	677^{abc}	57.1^{b}	109^{de}	492	559^{a}
[6]D. cinerea	High	706^{abc}	63.4^{b}	98.1^{ef}	612	561^{a}
	Medium	535^{c}	72.1^{b}	151^{ab}	517	486^{a}
	Low	726^{ab}	92.5^{a}	130^{bcd}	515	478^{a}
[7]Z. mucronata	High	769^{a}	56.0^{b}	123^{bcd}	414	415^{b}
	Medium	503^{d}	49.9^{b}	170^{a}	375	465^{a}
	Low	591^{bc}	43.6^{b}	157^{a}	438	508^{a}
[8]SEM		367	6.61	24.7	31.3	21.1
		Forage species means				
A. nilotica		625	48.4^{b}	109^{c}	476^{b}	527^{a}
D. cinerea		656	76.0^{a}	127^{b}	548^{a}	508^{a}
Z. mucronata		621	49.8^{b}	151^{a}	409^{c}	463^{b}
SEM		212	3.82	3.03	18.1	12.2
		Forage zone means				
	High	705^{a}	55.4	101^{c}	513^{a}	501^{a}
	Medium	533^{b}	54.5	153^{a}	438^{b}	482^{a}
	Low	665^{a}	64.4	131^{b}	482^{ab}	515^{a}
SEM		212	3.82	3.03	18.1	12.2
Species (S)		NS	**	**	**	**
Zone (Z)		**	NS	**	NS	NS
S x Z		NS	*	NS	NS	**

[1]DM = Dry matter; [2]CP = Crude protein; [3]ADF = Acid detergent fibre; [4]NDF = Neutral detergent fibre; [5]*A. nilotica* = *Acacia nilotica;* [6]*D. cinerea* = *Dichrostachy cinerea;* [7]*Z. mucronata* = *Ziziphus mucronata*; [8]SEM = Standard error of means. *P <0.05; **P < 0.01. NS = non significant (P > 0.05).

[abcdef] within the section of each column, means carrying the same superscripts are not different at P<0.05.High, Medium, Low = Rainfall areas.

4. 2. Mineral composition

The means and standard errors for calcium (Ca), phosphorus (P), potassium (K), chlorine (Cl) and fluorine (Fl)*A. nilotica, D. cinerea and Z. mucronata* of forage legume species harvested from different agro-ecological zones are shown in Table 4.2. Tree legume species affected P and Cl contents at ($P < 0.05$) and ($P < 0.01$), respectively. Agro-ecological zones influenced the content of Fl ($P < 0.05$). *A. nilotica* and *D. cinerea* had higher P (21.6 g/kg DM, 31.8 g/kg DM)levels than Z. mucronata (2.75 g/kg DM).TheCl content of *D. cinerea* and *Z. mucronata* than *A. nilotica* (*P < 0.05)*. There were also an interaction between species and zones for Fl. For *D. cinerea* and *Z. mucronata* the Fl content were the same in all the rainfall zones but for *A. nilotica* trees that grew in the high and medium rainfall zones had higher Fl than those that grew in the low rainfall zones.

4.3 Condensed tannin composition

The standard curve for Mimosa tannins used to express quantity of condensed tannins in the browse shoots are shown in Figure 4.1. The results of the analyses of variance, means and standard errors for total soluble condensed tannins (TSCT), total bound tannins (TBT) and total condensed tannins (TCT) expressed as g/kg Mimosa tannins are summarized in Table 4.3. The values for TCT content are illustrated in Figure 4.3.of *A. nilotica, D. cinerea* and *Z. mucronata* harvested from three agro-ecological zones. Tree species affected TST, TBT and TCT ($P < 0.01$) but zone did not ($P > 0.05$). The *Z. mucronata* had a significantly higher total soluble condensed tannins content ($P < 0.01$) than the other two species. The highest total soluble tannins, total bound tannins and total tannins values ($P < 0.05$) were found in *Z. mucronata* followed by *A. nilotica* and then *D. cinerea*

For total soluble condensed tannins and total bound tannins *A. nilotica* had the highest content ($P < 0.01$) in the low rainfall zone followed by the high rainfall zone and the medium zone for *D. cinerea* trees that grew in the high rainfall zone had higher total soluble tannins and total bound tannins level ($P < 0.01$) than those that grew in the medium and low zones; trees that grew in latter two zones did not differ from one another($P < 0.05$). For *Z. mucronata* the trees that grew in the medium rainfall zone had higher ($P < 0.01$) levels of total soluble condensed tannins and total bound tannins than those that grew in the high and low rainfall zones which did not differ from each other.

Table 4.2. Means and standard errors for potassium, calcium, phosphorus, chlorine and fluorine of *Acacia nilotica, Dichrostachys cinerea and Ziziphus mucronata* species found in different agro-ecological zones

Forage species	Zones	[1]Ca (g/kgDM)	[2]P (g/kgDM)	[3]K (g/kgDM)	[4]CL (g/kgDM)	[5]Fl (g/kgDM)
[6]A. nilotica	High	0.02	6.00	13.4^{b}	88.1^{b}	5.31^{a}
	Medium	0.21	26.8	54.1^{a}	93.9^{b}	8.88^{a}
	Low	0.02	38.2	45.9^{a}	208.4^{a}	0.00^{b}
[7]D. cinerea	High	0.02	24.4	44.7^{a}	92.3^{b}	0.00^{b}
	Medium	0.10	17.0	52.8^{a}	13.7^{b}	1.94^{b}
	Low	0.09	54.0	26.4^{a}	40.1^{b}	2.41^{b}
[8]Z. mucronata	High	0.02	2.31	62.3^{a}	16.0^{b}	1.19^{b}
	Medium	0.02	8.26	45.6^{a}	0.00^{b}	0.00^{b}
	Low	0.02	0.00	34.1^{a}	11.7^{b}	2.96^{b}
[9]SEM		0.08	11.7	10.2	147.0	0.83
			Forage species means			
A. nilotica		0.08	21.6^{a}	37.8	130.1^{a}	1.77
D. cinerea		0.07	31.8^{a}	41.3	48.9^{b}	0.80
Z. mucronata		0.02	2.75^{b}	47.3	3.89^{b}	0.99
SEM		0.04	6.75	5.89	62.8	0.48
			Forage zone means			
	High	0.02	8.14	40.1	60.1	1.77^{a}
	Medium	0.11	17.3	50.8	35.9	0.00^{b}
	Low	0.04	30.7	35.5	87.0	1.79^{a}
SEM		0.04	6.75	5.89	62.8	0.48
Species (S)		NS	*	NS	**	NS
Zone (Z)		NS	NS	NS	NS	*
S x Z		NS	NS	NS	NS	**

[1]K = Potassium; [2]Ca = Calcium; [3]P = Phosphorus; [4]Cl = Chlorine; [5]Fl = Fluorine; [6]*A. nilotica* = *Acacianilotica*, [7]*D. cinerea* = *Dichrostachycinerea*, [8]*Z. mucronata* = *Ziziphus mucronata*, [9]SEM = standard error of means *P < 0.05; **P < 0.01. NS = non significant (P > 0.05).

[abcdef] within the section of a column, means carrying the same superscripts are not different at P<0.05.High, Medium, Low = Rainfall areas.

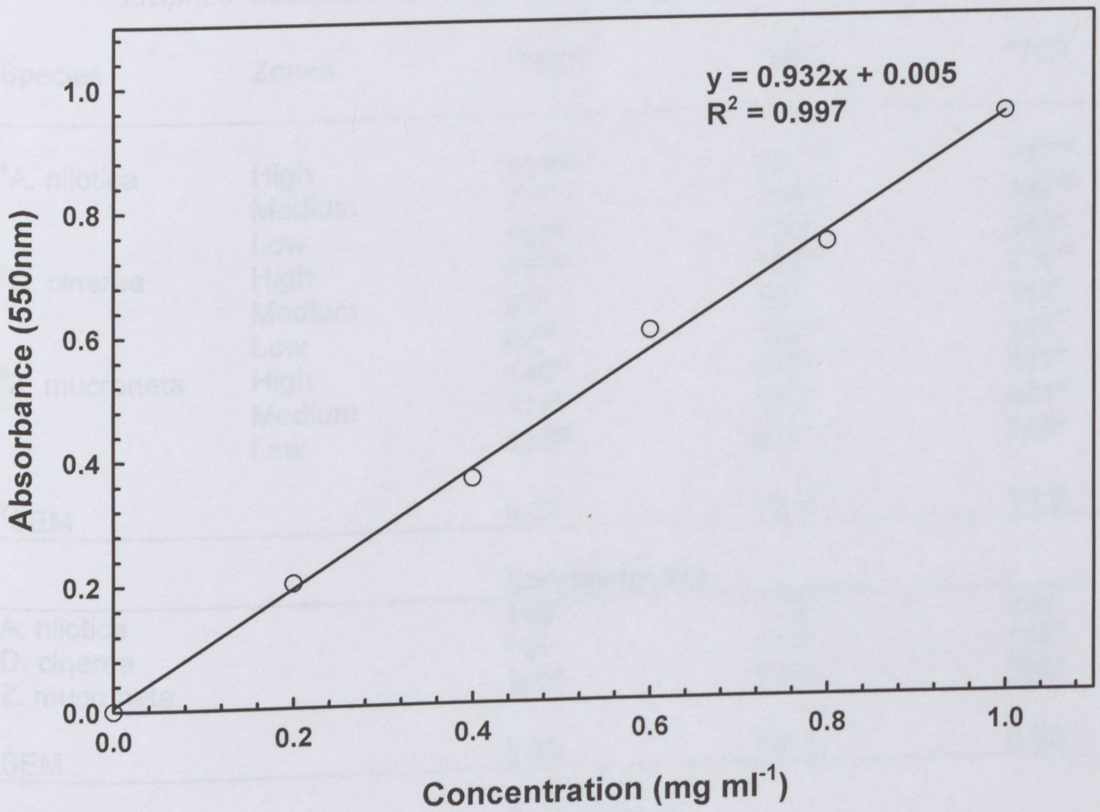

Figure 4.2 Standard regression curve of the Mimosa tannin for the n-butanol-HCl test

Table 4.3. Means and standard errors for total soluble tannins, total bound tannins and total tannins (g/kg Mimosa (MT) equivalent of *Acacia nilotica, Dichrostachys cinereaand Ziziphus mucronata* species found in different agro-ecological zones.

Species	Zones	[1]TSCT	[2]TBT	[3]TCT
[4]A. nilotica	High	100^{cb}	127^{dcb}	227^{cd}
	Medium	77^{dc}	104^{dc}	182^{de}
	Low	123^{b}	160^{b}	283^{c}
[5]D. cinerea	High	95^{dcb}	143^{cb}	238^{cd}
	Medium	53^{d}	90^{d}	143^{f}
	Low	62^{dc}	102^{dc}	163^{ef}
[6]Z. mucronata	High	145^{a}	222^{a}	367^{b}
	Medium	178^{a}	253^{a}	431^{a}
	Low	136^{ab}	212^{a}	348^{b}
[7]SEM		9.22	10.3	10.9
		Species means		
A. nilotica		100^{b}	130^{b}	230^{b}
D. cinerea		70^{c}	112^{b}	182^{c}
Z. mucronata		153^{a}	229^{a}	382^{a}
SEM		5.32	5.93	6.30
		Zone means		
	High	114	164	277
	Medium	103	149	252
	Low	107	158	265
SEM		5.32	5.93	6.30
Species (S)		**	**	**
Zones (Z)		NS	NS	NS
S x Z		**	**	**

[1]TSCT=Total soluble condensed tannins; [2]TBT=total bound tannins; [3]TCT.=.Total condensed tannins; [4]*A. nilotica = Acacianilotica;*[5]*D. cinerea = Dichrostachy cinerea,*[6]*Z. mucronata = Ziziphus mucronata,*[7]SEM = standard error of means *P < 0.05; **P < 0.01. NS = non significant (P > 0.05).

[abcdef] within the section of a column, means carrying the same superscripts are not different at P<0.05.High, Medium, Low = Rainfall areas.

For *A. nilotica* trees that grew in the low rainfall zoneshad the highest (283 g/kg DM) total tannins level (P < 0.01) followed by those that grew in the high rainfall and then followed by those (227 g/kg) thatin the medium rainfall zones. For *D. cinerea* trees that grew in the high rainfall zones had the higher (238 g/kg DM) total tannins value (P < 0.01) than those that grew in the other two zones and the trees that grew in the low rainfall zone had a higher level than those that grew in the medium rainfall zone. For *Z. mucronata* trees that grew in the medium zone had higher143 g/kg DM) level of total tannins (P < 0.01) than those that grew in the high and low rainfall zones. The two latter groups however did not differ from each other (P < 0.01)

CHAPTER 5: DISCUSSION

The differences among the chemical, mineral and tannin composition of *A. nilotica*, *D. cinerea* and *Z. mucronata* forage leaves could be due to differences in agro-ecological zones (climate, rainfall, season and temperature), plant (age and leaf size), and soil (status, pH and nutrients), (Ibrahim *et al.,* 1988, Thapa *et al.,* 1997, Topps and Olivier, 1993). Plant maturity and soil nutrients have an influence on chemical, mineral and tannins composition although the soil type data of the areas are slightly similar (Department of Agriculture., 2004). The significant differences in ash, CP, ADF and NDF contents at ($P < 0.01$) by forage species when compared across all three zoneswere comparable to findings of Ibrahim et al., (1988), Topps and Olivier (1993) and Teferedegne (2000) and who reported that environmental differences influence the chemical composition of forages grown in different areas and harvested at the same age of maturity. They attributed the differences to soil and environmental factors.

The high NDF and ADF contents of the forages could be attributed to lignifications and low fibre content which result in high digestibility (Topps,1997). Variation in ADF among different forages might be due to the influence of genotype and growth environment (Tilley and Terry, 1963., Matizha,*et al* 1997, Mupangwa,*et al.*, 1997, Valante, *et al.,* 2000). Rainfall is the factor which most clearly determines the distribution of plant communities in South Africa, as well as the potential productivity of these communities (Tainton and Hardy, 1999). Low levels of the detergent fibres are associated with high voluntary DM intakes in ruminants consuming all forage diets (Mokoboki *et al.,* 2005). The current study showed that NDF levels varied between 408 and 548 g/kg DM among the different species and these variations in NDF levels in browse have also been observed in other studies (Reed *et al.*, 1990; Tanner *et al.*, 1990; Kaitho *et al.*, 1997; Larbi *et al.*, 1998; Maasdorp *et al.*, 1999; Hove *et al.*, 2001). The higher NDF levels recorded from this study could be due to plant maturity and environmental factors in the foliage of all species used.

The differences among the species for CP content for Z. mucronata (range 123 – 157 g/kg DM), *D. cinerea* (range 98 – 151g/kg DM) and the lowest *A. nilotica* (range from 82 – 137 g/kg DM) in all three zones may be due to influence of genotype (Tilley and Terry, 1963, Matizha *et al.,* 1997, Mupangwa *et al.*, 1997, and growth environment (Valente *et al.,* 2000). Mero and Uden (1998) reported that forages with high CP content tend to have high intake. The *A. nilotica*forage has a potential as the browse sources for ruminants due to its appreciable CP content (Abdulrazak et al., 2000) and *D. cinerea* which is relatively high in CP content may be an alternative source of indirect microbial protein (Choongo *et al.,* 2008, Feedipedia, 2011;). Crude protein is important for ensuring the survival and development of ruminants and CP content should be high in the diet to meet the requirements of animals for different physiological functions (Matlebjane *et al.*, 2009).

Compared with the tropical mature grasses, browse appears to be richer in crude protein and minerals than in subtropics (Le Houerou, 1980). Devendra (1993) reviewed the significance of shrub and tree fodders as sustainable feed resource and affirmed that fodder trees and shrubs had potential as sustainable protein supplement source during the dry season. The same author reported that when fodder trees and shrubs are used as supplements the optimum dietary level of tannins should be about 30-50% of the ration on a DM basis. Rittner and Reed (1992), Makkar and Becker (1998) and Njidda *et al.* (2009) also indicated that most tropical browse species are high in CP and can be used to supplement poor quality roughages to increase productivity of ruminant livestock in tropical regions. Hove *et al.*, 2001; Fondevila *et al.*, 2002; Baloyi *et al.*, 2008 reported that livestock productivity is constrained by the shortage of good quality feed, especially during the dry season when the grasses have low CP and while the fibre content increases progressively (Elliott and Fokkema, 1961).

Tree fodders maintain higher protein and mineral contents during growth than do grasses, which decline rapidlyin quality with progress to maturity (Aganga and Tshwenyane 2003). The low protein content and the high fibre and lignin content of veld grasses result in reduced voluntary feed intake. The crude protein contents findings of the three forage species observed in different agro-ecological zoneswere sufficient for high producing dairy animals, Meissner, 1997 reported that all the forage species contained more than 130 to 140 g CP/kg DM, are required for high producing dairy animals

The forage species had more CP content range than the range of 26.3 -153 g/kg DM reported by Minson (1990), Skerman and Riveros (1990), Barnes *et al.*, (1995) and Mwilawa *et al.*, (1998). However Aganga *et al.*,(1998),using (*A. Karroo, A. erioloba, A. nilotica, A. sieberiana, A. tortilis* and *Leucaena leucocephala*),Ngwa *et al.*, (2000), using *D.cinerea and A.nilotica,) (Lonchocarpus capassa, Ziziphus mucronata, Sclerocarya birrea, Kirkia acuminata* and *Rhus lancea seeds),and* Aganga and Mosase(2001)using*(Combretum species, C. apiculatum, C. molle* and *C. zeyheri*, as well as that of *Colophospermum mopane* reported CP content range from 88 – 193.2 g/kg DM for speciesgrown in sub-tropical areas which significantly differs from range of the three forage species that are used in this study. The CP levels in the present study were above the minimum requirements for maintenance of between 70 and 80 g /kg CP DM for ruminants (Meissner, 1997).

All animal and plant tissues contain widely varying amounts and proportions of mineral elements, which largely remain as oxides, carbonates, phosphates and sulfates in the ash after ignition of organic matter. Minerals such as calcium and phosphorus can form structural components of body organs and tissues, While dietary The Ca content concentrations of 2-6 g/kg, with higher requirements for lactation have been variously recommended for cattle and sheep National Research Council (National Research Council, 1978, 1984, 1985: ARC, 1980), the findings of Sykes and Field (1972) suggest that levels of 2.6 g/kg are adequate in most circumstances. The results indicate that problems of Ca deficiency would be expected. Ca is the most abundant mineral in the body, 98% Ca is found in the bones and teeth (Rick, 2007). Calcium is normally one of the primary limiting factors in the diets of sheep and goat and hence need to be provided as supplement. 99% of the Ca and 80% of the P in the entire body are found in skeleton and teeth (McDonald*et al* 2002). McDowell *et al.,* 2005; According to NRC (1975) calcium and phosphorus requirements for pregnant ewes are 0.21-0.27 % and 0.20-0.25 %, respectively,Fodder legumes are an important source of mineral such as sulphur, calcium and copper.

The Ca and P contents (ranging from 0.2-2.1% and 0.6-5.4 respectively) in this study were found to be higher than the NRC estimates for pregnant ewes. All the tree species have almost the same level of calcium. *Z. mucronata* in low rainfall areas did not have P this can be the result of the plant age. Maturity of the plant increases the Lignin content of the plant and decreases the plant nutrients. *Acacia nilotica* trees that grew in low rainfall zone had highest chlorine content than all three forage species and this can be a result of soil status, minerals did not leach from the soil. Underwood (1981) considered a dietary P level of 1.7 g/kg to be marginal for grazing animals, in essential agreement with work of Little (1980, 1985) which indicated that a figure of 1.4 g/kg should be regarded as minimal for growing cattle. For animals grazing on range lands, these three forage species would be best alternative source of phosphorus.

Masters and Feels (1990) reported that during autumn and summer, pasture contained adequate potassium (6-22 g/kg) for sheep, the highest concentrations being recorded in summer. The K contents of the forage species were found to be higher than the records made by Masters and Feels (1990). The K in the present study ranged (from 13 to 62 g/kg DM). Green growing forages are excellent sources of K (Rick, 2007). Supreme Court Appeal, (1990) suggested that the K requirement for sheep is 5 gram per kg feed. On the other hand, McDowell (1997) recommended 0.5-1.0 % potassium (on dry basis) as a requirement for ruminants. In relation to the essential mineral elements, the level of K in all the forages was above the level of 8 g/kg recommended for grazing animals (Underwood, 1981). However, it has been suggested (McDowell, 1985) that weaned calves and high producing dairy cows under stress, such as heat stress, may require K level above 10 g/kg.

The nutritive value of forages for livestock feeding depends on the balance between the nutritive components of the plants, the digestibility of such nutrients, the metabolism of absorbed nutrients and the quantity of nutrients ingested by the animal (Seoane *et al*., 1981). The quantity and quality of feed which an animal consume is probably the most important factor that affects productivity and profitability of a livestock enterprise. In general, ruminant livestock production is dependent on the use of natural pasture (veld) as the basic feed (Baloyi, 2002). Chemical analyses help to determine the nutritive value and have been suggested as alternative ways to determine quality of feedstuffs (Van Soest, 1983). Tannins are naturally occurring plant phenols with a molecular weight of between 500 and 3000 and have their ability to precipitate protein and other compounds (Baloyi et al.,2008).

Tannins have toxic effect on ruminants especially during winter when the animals browse the forage species. It is, however, more important from a nutritional perspective to consider tannins in terms of CT and HT tannins (Mueller-Harvey, 2001). Condensed tannins are polymers of flavonol units most frequently linked at C4 - C8 or C4 – C6. They interfere with intake and digestion of the feeds in which they occur (Dube *et al*., 2001) at high levels, but at very low levels most have beneficial effects (Foo *et al*., 1996). The ideal CT concentration for ruminant animal nutrition has been suggested to be in the range 20 to 40 g/kg DM, based on the butanol-HCl method (Barry and Duncan, 1984). The CT range was from 53-178 g/kg DM and this could be due to either plant age or genotype. Variations in chemical composition among species of *Acacia* foliages may be partly due to genotypic factors that control accumulation of forage nutrients. Accumulation of nutrients in plants is a property of species (Minson,1990), and varies among species and genera.

The *Ziziphus mucronata*species are rich in total condensed tannins. Due to minerals being unleashed from the soil,*Acacia* that grew low rainfall zones had highest content of total soluble and soluble tannins. Tannins in forage legumes have both negative and positive effects on the nutritive value of fodder (Mueller- Harvey and McAllan, 1992; Paterson, 1993; Reed, 1995). Barry (1989) suggested low concentrations of condensed tannins (20 to 40 g/kg DM) had beneficial effects in protein protection in the rumen. Tannins in high concentrations of 45 to 60 g/kg reduce voluntary feed intake, digestibility of proteins and carbohydrates, and animal performance (Barry and Duncan, 1984; Paterson, 1993). On the other hand, tannins in low to moderate concentrations (30 to 40 g/kg) prevent bloat and increase the flow of non-ammonia nitrogen and essential amino acids from the rumen (Barry and Manley, 1984, Waghorn, 1987) to the intestine.

Condensed tannins are high in all forage species. The high levels of tannins in plant parts may bind protein and, at high levels, suppress animal production. The CT tannins in this present study were found to be more in bound form and this correlates with Mokoboki, (2006). Baloyi, (2007) reported that more CT exist in bound form in NDF compared to ADF and that NDF represents not only hemicellulose, cellulose, lignin and silica fractions but also proanthocyanidins in the leaves of tree species, and this can be regarded as the environmental differences which influence the chemical composition of forages grown in different areas and harvested at the same age of maturity.

CHAPTER 6: CONCLUSION AND RECOMMENDATIONS

6.1 Conclusion

The current study showed that browse leaves and stems harvested from *A. nilotica, Z. mucronata* and *D. cinerea* could be effective feed supplements for ruminants fed low quality forages since they have high CP content compared to the natural pasture grasses and could provide part of the solution to shortage of protein especially in rural areas where the veld is the only source of feeds for ruminants. High levels of condensed and bound tannins could limit optimal utilization of nutrients in *Z. mucronata* due to their toxicity. A large proportion of the total CT in *Z. mucronata* was bound to protein compared to the condensed tannins. The values of browse species grazed by livestock in Limpopo Province of South Africa identified were associated with high levels of crude protein and varying amount of tannins.

6.2 Recommendations

- There is a need to investigate possible presence, composition and chemical structure on anti-nutritional factors (tannins) in *A. nilotica, D. cinerea* and *Z. mucronata* forage legumes harvested at different stages of maturity.
- Three forage legumes could provide valuable source of browse if managed well, based on the current study it is proposed that the farmers must help the nature conservation scheme to protect the forage legume species against late veld fires during the latter half of the dry season when it should be utilized and to ensure that the will be enough feed (protein) during the dry season.

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Appendix 1. Mean squares of analysis of variable for dry matter, ash, crude protein, acid detergent fibre and neutral detergent fibre of *Acacia nilotica, Dichrostachys cinerea and Ziziphus mucronata* species found in different agro-ecological zones.

Source	df	[1]DM	ASH	[2]CP	[3]ADF	[4]NDF
Species (S)	2	494174.56	3330.21**	5395.88**	14062.04**	72807.42**
Zones (Z)	2	6090619.67**	412.86	7991.70**	3854.20	17813.65*
S x Z	4	1700675.98	583.71	81.17	10961.28**	5226.60
Error	32	674767.62	218.59	137.28	2222.80	4900.60

[1]DM = Dry matter; [2] CP = Crude protein; [3]ADF = Acid detergent fibre; [4] NDF = Neutral detergent fibre.

*P < 0.05; **P < 0.01.

Appendix 2. Mean squares of variables for total soluble tannins, total bound tannins and total tannins of *Acacia nilotica, Dichrostachys cinerea and Ziziphus mucronata* species found in different agro-ecological zones.

Source	df	[1]TST	[2]TBT	[3]TT
Species (S)	2	26076.93**	57908.27**	160972.31**
Zones (Z)	2	359.92	606.58	1879.16
S x Z	4	3462.80**	4491.19**	15729.01**
Error	32	205.40	252.10	898.24

[1]TST = Total Soluble Tannins, [2]TBT = Total Bound Tannins, [3] TT = Total Tannins.

*P < 0.05; **P < 0.01.

Appendix 3. Mean squares of analysis of variable for, calcium, phosphorus, potassium, chlorine and fluorine of *Acacia nilotica, Dichrostachy cinerea and Ziziphus mucronata* species found in different agro-ecological zones.

Source	df	[1]Ca	[2]p	[3]K	[4]Cl	[5]Fl
Species (S)	2	0.02	3230.74*	289.95	46332.50**	2.89
Zones (Z)	2	0.03	1549.78	886.55	9410.96	14.62*
S x Z	4	0.02	868.58	1058.61	8240.20	17.00**
Error	32	0.03	683.65	519.77	4849.14	3.45

[1]K = Potassium; [2]Ca= Calcium; [3]P = Phosphorus; [4]Cl = Chlorine; [5]Fl = Fluorine.

*P < 0.05; **P < 0.01.