Dry matter production, intake and nutritive value of certain *Indigofera* species

by

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DECLARATION

I, Tlou Julius Tjelele, declare that this dissertation, for the degree M. Inst. Agrar.
(Animal Production) at the University of Pretoria, has not been submitted by me for a
degree at any other University.

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SUMMARY OF CONTENTS

ACKNOWLEDGEMENTS

ABSTRACT

UITTREKSEL

LIST OF TABLES

CHAPTER 1

1.	INTRODUCTION AND LITERATURE REVIEW	1
1.1	Description of Indigofera species	2
1.2	Chemical composition of <i>Indigofera</i> species	3
1.3	Factors affecting mineral contents of the plants	4
1.3.1	Soil pH	4
1.3.2	Stage of maturity	4
1.3.3	Climatic conditions	4
1.4	Environmental adaptation	4
1.5	Nutritive value	5
1.5.1	Factors which influence nutritive value	5
1.5.1.	Plant maturity	5
1.5.1.2	2 Environment	5
1.5.1.3	3 Genetic variation	6
1.5.2	Measurement of nutritive value	6
1.5.2.	1 Total collection	6
1.5.2.2	2 Marker techniques	6
1.5.3	Methods for predicting nutritive value	6
1.5.3.	Database selection	7
1.5.3.2	2 Development and evaluation of prediction equation	7
1.5.3.3	Biological procedure	7
1.5.3.4	Physical procedure	7
1.5.4	Anti-nutritive and toxic factors in forage tree legumes	8
1.5.4.1	Strategies for managing anti-nutritive factors	8

1.6	Voluntary feed intake	8
1.6.1	Intake of legumes	9
1.6.2	Factors which influence feed intake	10
1.6.2.1	Psychological factors	10
1.6.2.2	Physiological factors	10
1.6.2.3	Animal size	10
1.6.2.4	Physical factors	10
1.6.2.4.1	Plant structure	10
1.6.2.5	Dietary factors	11
1.6.2.5.1	Fibrous compounds	11
1.6.2.6	Environmental factors	11
1.6.2.6.1	Effects of heat	11
1.6.2.7	Effects of climate and season on forage quality	11
1.6.2.7.1	Temperature	11
1.6.2.7.2	Water	11
1.6.2.7.3	Soil	12
1.6.2.7.4	Defoliation and diseases	12
1.6.3	Utilization and beneficial effects of forage legumes	12
1.7	General description of Leucaena leucocephala	12
1.7.1	Environmental adaptation	13
1.7.2	Chemical composition	13
1.7.3	Herbage productivity	14
1.8	Limitations and problems	14
1.8.1	Non-toxic secondary plant compounds	14
1.8.2	Tannins	14
1.8.3	Lignin	15
1.8.4	Toxic-compounds in plants	16
1.8.5	Mimosine	16
1.8.6	Indospicine	16
1.8.7	Saponins	16
1.9	Hypothesis and objectives	17

CHAPTER 2

2.	MATERIALS AND METHODS	18
2.1	Introduction	18
2.1.1	Study location	18
2.1.2	Sample collection site	19
2.1.3	Sample preparation	19
2.2	Leaf: stem ratio	19
2.3	Chemical analysis	19
2.3.1	Dry matter determination	19
2.3.2	Ash determination	20
2.3.3	Neutral detergent fibre (NDF) determination	20
2.3.4	Nitrogen and crude protein determination	21
2.3.5	Organic matter (OM) determination	21
2.3.6	In vitro digestibility of organic matter (IVDOM) determination	21
2.4	Minerals	22
2.5	Voluntary intake trial	23
2.5.1	Feeding of experimental animals	23
2.5.1.1	Feed samples	24
2.5.1.2	Faeces samples	24
2.6	Statistical analysis	24
СНАРТ	TER 3	
3.	RESULTS AND DISCUSSION	25
3.1	Dry matter production	25
3.1.1	Leaf DM yield	25
3.1.2	Stem DM yield	26
3.1.3	Total DM yield	27
3.2	Leaf to stem ratio	28
3.3	Chemical composition	29
3.3.1	Ash concentration	29
3.3.1.1	Leaves	29
3.3.1.2	Edible components (leaves and fine stems)	30
3.3.2	Crude protein concentration	31

3.3.2.1	Leaves	31
3.3.2.2	Edible components (leaves and fine stems)	33
3.3.3	Neutral detergent fibre concentration	34
3.3.3.1	Leaves	34
3.3.3.2	Edible components (leaves and fine stems)	35
3.4	Digestibility	36
3.4.1	In vitro digestibility of organic matter (IVDOM)	36
3.4.1.1	Leaves	37
3.4.1.2	Edible components (leaves and fine stems)	37
3.5	Minerals	38
3.5.1	Macro elements	38
3.5.1.1	Calcium concentration	38
3.5.1.1.1	Leaves	39
3.5.1.1.2	Edible components (leaves and fine stems)	40
3.5.1.2	Phosphorus concentration	40
3.5.1.2.1	Leaves	40
3.5.1.2.2	Edible components (leaves and fine stems)	41
3.5.1.3	Magnesium concentration	42
3.5.1.3.1	Leaves	42
3.5.1.3.2	Edible components (leaves and fine stems)	43
3.5.2	Micro elements	43
3.5.2.1	Copper concentration	44
3.5.2.1.1	Leaves	44
3.5.2.1.2	Edible components (leaves and fine stems)	45
3.5.2.2	Zinc concentration	46
3.5.2.2.1	Leaves	46
3.5.2.2.2	Edible components (leaves and fine stems)	47
3.5.2.3	Manganese concentration	47
3.5.2.3.1	Leaves	48
3.5.2.3.2	Edible components (leaves and fine stems)	48
3.6	Voluntary feed intake and digestibility	48
3.7	Chemical composition of forage	50
3.7.1	Crude protein concentration	50
3.7.2	Neutral detergent fibre concentration	51

3.7.3	In vitro digestibility of organic matter	51
3.8	Intake and digestibility of lucerne, Indigofera species and	
	Leucaena leucocephala	51
3.8.1	Organic matter intake (OMI)	52
3.8.2	Digestible organic matter intake (DOMI)	53
3.8.3	Neutral detergent fibre intake (NDFI)	54
3.8.4	Organic matter digestibility (OMD)	55
3.8.5	Neutral detergent fibre digestibility (NDFD)	55
CHAI	PTER 4	
4.	GENERAL DISCUSSION	57
4.1	Dry matter production	57
4.2	Leaf to stem ratio	57
4.3	Chemical composition	58
4.3.1	In vitro digestibility of organic matter (IVDOM)	59
4.3.2	Minerals	59
4.3.2.1	Macro elements	59
4.3.2.2	2 Micro elements	59
4.4	Feed intake and digestibility	60
CHAI	PTER 5	
5.	SUMMARY, CONCLUSION AND RECOMMENDATIONS	61
5.1	Summary and conclusion	61
5.2	Recommendations	63
REFE	RENCES	64

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ABSTRACT

Dry matter production, intake and nutritive value of certain *Indigofera* species

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The objective of the study was to evaluate the dry matter production, intake and the nutritive value of *Indigofera* species. The dry matter yield, leaf:stem ratio, chemical composition, voluntary intake and digestibility of *Indigofera* species were determined. The leaves as well as the leaves and stems (<3mm) of five different *Indigofera* species (*I. amorphoides*, *I. cryptantha*, *I. costata*, *I. viciodes* and *I. arrecta*) were harvested. There was a greater total dry matter yield during autumn 2004 from *I. amorphoides*. However, no significant differences were obtained between all the species over the seasons.

There were significant differences between all the species in autumn with a lower proportion of leaves than in spring, except for *I. arrecta*, which had the same leaf: stem ratio in both seasons. During spring, *I. amorphoides* and *I. cryptantha* generally had a higher proportion of leaf material than other species. There were significant differences between all the species for the leaves as well as leaves and stems (<3mm) as a result of advancing maturity and decrease in leaf: stem ratio with respect to ash, crude protein (CP), neutral detergent fibre (NDF) concentration and *in vitro*

digestibility of organic matter (IVDOM). Despite a decrease in leaf: stem ratio, all the species had an adequate CP concentration for optimal animal production. All the minerals (macro and micro elements) found in this study, in both years, will satisfy the nutrient requirements of sheep. However, all mineral elements in this study appeared to decrease with ageing of the plants and decline in leaf: stem ratio, except for Mn concentration, which increased with ageing of the plants.

Lucerne, which was used during the intake study as a control, had a significantly higher organic matter intake (OMI) and digestible organic matter intake (DOMI) than *Indigofera* species and *Leucaena leucocephala*. However, there were no significant differences between *Indigofera* species and *L. leucocephala*. Intake levels in this study for *L. leucocephala* and *Indigofera* species would be insufficient for maintenance requirements of grazing sheep. The relatively lower IVDOM for *Indigofera* species and *L. leucocephala* compared to that of lucerne was because of a higher NDF concentration. Despite the relatively high NDF concentration, *Indigofera* species appeared to be a good fodder because of its high CP and Ca, P, Mg, Cu, Zn and Mn concentrations.

UITTREKSEL

Droë materiaal produksie, inname en voedingswaarde van sekere *Indigofera* species

deur

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Die doel van die studie was om die droë materiaal produksie, inname en die voedingswaarde van *Indigofera* spesies te ondersoek. Die droë material opbrengs, blaar:stam verhouding, chemiese samestelling en verteerbaarheid van *Indigofera* spesies is bepaal. Die blare sowel as die stamme (<3mm) van vyf verskillende *Indigofera* spesies (*I. amorphoides*, *I. cryptantha*, *I. costata*, *I. viciodes and I. arrecta*) is geoes. 'n Hoër totale droë material opbrengs is van *I. amorphoides* gedurende herfs 2004 geoes. Geen betekenisvolle verskille is egter tussen die spesies vir die verskillende seisoene aangeteken nie.

Daar was betekenisvolle verskille tussen al die spesies in herfs met 'n laer blaar verhouding as in die lente, uitgesonderd *I. arrecta* wat dieselfde blaar:stam verhouding in beide seisoene gehad het. Gedurende die lente het *I. amorphoides* en *I. cryptantha* oor die algemeen 'n hoër verhouding blaar material as die ander spesies getoon. Daar was betekenisvolle verskille tussen al die spesies vir die blare sowel as die blare en stamme (<3mm) weens volwasse wording en die afname in blaar:stam verhouding met verwysing na as, ruproteïen (RP), neutraal bestande vesel (NDF)

konsentrasie en *in vitro* verteerbaarheid van organiese materiaal (IVVOM). Ten spyte van 'n afname in blaar: stam verhouding het al die spesies voldoende RP konsentrasies vir optimale diereproduksie getoon. Beide die makro- en mikroelemente vir beide jare, sal aan die voedingsbehoeftes van skape voldoen. Alle minerale elemente wat in die studie geanaliseer is, se konsentrasie het verlaag soos die plante verouder het en soos die blaar:stam verhouding afgeneem het, behalwe vir die Mn- konsentrasie wat met veroudering verhoog het.

Medicago sativa, wat as 'n kontrole in die inname proef gebruik is, het 'n betekenisvolle hoër organiese material inname (OMI) en verteerbare organiese material inname (VOMI) as die Indigofera spesies en Leucaena leucocephala getoon. Daar was egter geen betekenisvolle verskille tussen die Indigofera spesies en L. leucocephala nie. Inname van L. leucocephala en die Indigofera spesies was onvoldoende vir onderhoud van skape. Die relatiewe laer IVVOM van die Indigofera spesies en L. leucocephala, in vergelyking met lusern, kan toegeskryf word aan die hoër NDF konsentrasies in eersgenoemde. Ten spyte van die relatiewe hoë NDF konsentrasie blyk dit asof die Indigofera spesies 'n goeie ruvoer is aangesien dit beskik oor hoë RP sowel as hoë Ca, P, Mg, Cu, Zn en Mn konsentrasies.

LIST OF TABLES

Table 1.1	Characterization of forage tree legume species	2
Table 1.2	Example of non-toxic plant compounds of tannins	
	present in forage and browse legumes	15
Table 2.1	Average temperature and rainfall for Hatfield	
	Experimental Farm	18
Table 3.1	The leaf DM yield (g/plot) of five Indigofera species	25
Table 3.2	The stem DM yield (g/plot) of five Indigofera species	26
Table 3.3	The total DM yield (g/plot) of five Indigofera species	27
Table 3.4	Leaf:stem ratio of five Indigofera species	28
Table 3.5	The ash concentration (%) of leaves and edible component	
	(leaves and fine stems) of five Indigofera species	29
Table 3.6	Variations in ash concentration with age (years) in	
	lucerne	30
Table 3.7	The crude protein concentration (%) of leaves and	
	edible component (leaves and fine stems) of five Indigofera	
	species	31
Table 3.8	Effect of stage of maturity on nutrient content	
	of lucerne	32
Table 3.9	The crude protein requirements of different	
	classes of ruminants	33
Table 3.10	The neutral detergent fibre concentration (%) of	
	leaves and edible component (leaves and fine stems) of five	
	Indigofera species	34
Table 3.11	Effect of stage of maturity on nutrient content	
	of lucerne forage	35
Table 3.12	The <i>in vitro</i> digestibility of organic matter (%) of	
	leaves and edible component (leaves and fine stems) of	
	five Indigofera species	36
Table 3.13	The calcium concentration (%) of leaves and edible component	(leaves
	and fine stems) of five <i>Indigofera</i> species	38

Table 3.14	Nutrient requirements based on NRC and ARC for	
	various ruminant species	39
Table 3.15	The phosphorus concentration (%) of leaves and edible component	
	(leaves and fine stems) of five Indigofera species	40
Table 3.16	Variation in mineral composition with age (days)	
	of Leucaena Leucocephala	41
Table 3.17	The magnesium concentration (%) of leaves and edible	
	component (leaves and fine stems) of five Indigofera species	42
Table 3.18	The copper concentration (mg/kg) of leaves and edible	
	component (leaves and fine stems) of five Indigofera species	44
Table 3.19	Threshold concentrations of macro-elements in forage for	
	ruminants	45
Table 3.20	The zinc concentration (mg/kg) of leaves and edible component	
	(leaves and fine stems) of five Indigofera species	46
Table 3.21	The manganese concentration (mg/kg) of leaves and edible	
	component (leaves and fine stems) of five Indigofera species	47
Table 3.22	Chemical composition of lucerne, Indigofera species and	
	Leucaena leucocephala	50
Table 3.23	Intake by sheep of lucerne, Indigofera species and	
	Leucaena leucocephala	52
Table 3.24	Digestibility of lucerne, Indigofera species and	
	Leucaena leucocephala utilized by sheep	55

CHAPTER 1

1. INTRODUCTION AND LITERATURE REVIEW

Poor nutrition is one of the major constraints of animal production in sub-Saharan Africa. This is because animals live predominantly on high fibre feeds, which are often deficient in nutrients (nitrogen, minerals etc.). Legumes have become more important for use as high quality forage for livestock, both in cultivated pastures and in naturally occurring associations. Tropical forage legumes are important in the nutrition of small ruminants (goats and sheep). Nutritionally they are 2-3 times richer in protein than cereal grains. There is an increasing interest in the use of leguminous trees as a source of high quality feed for grazing and as a supplement to improve the productivity of ruminants receiving poor quality roughages. Leguminous trees are usually long-lived and have low maintenance requirements and, therefore, enhance the sustainability of farming systems (Gutteridge and Shelton, 1994).

In semi-arid and arid environments, legumes are important because plant growth is limited by rainfall and inadequate feed supply represents the most critical constraints to animal production (Devendra, 1989). Tree and shrubs legumes have provided valuable forage for herbivores since the time of their domestication (Robinson, 1985). At least 75% of the shrubs and trees of Africa serves as browse plants and many of them are leguminous (Skerman, 1977).

The nutritional quality of tree legumes varies from excellent (*Leucaena leucocephala*) to quite poor (*Acacia* species). Poor quality can firstly be due to tannins, which reduce the digestibility of herbage and protein, and secondly due to phylloides (expanded and flattened leaf petioles) of some species, instead of pinnate or binnate leaves, which are very high in fibre and therefore of low digestibility (Gutteridge and Shelton, 1994).

Table 1.1 Characteristics of forage tree legume species (Brewbaker, 1986)

				Tolera	nce of				
Species	Form	Preferred env.	Drought	Water logging	Acid soils	Alkaline soils	Cold	СР	IVDOM
A. angustissima	Shrub	Humid/ tropics	Fair	Fair	Good	-	Fair	23	48
A.saligna	Shrub/ Tree	semiarid/ subtropics	Good	Fair	Good	Good	Good	12-16	40
I.species	Shrub	tropics/ subtropics	Good	Poor	Good	Good	Fair	-	-
L.leucocephala	Shrub/ tree	shrub/tree	Good	Poor	Good	Fair	Fair	15-28	55

1.1 Description of *Indigofera* species

Shrubby *Indigofera* species are up to 2m high, erect, branched:leaflets are in five to eight pairs and oblanceolate, about 1 to 1.5 cm long, smooth above and hairy beneath, while flowers are yellow in 6-12 flowered racemes. Pods are chestnut-brown when mature, 1.5 – 4 cm long, polished and six to eight seeded (Andrews, 1952). *Indigofera* species generally prefer light (sandy) and medium (loamy) soils, require well-drained soils and can grow in very alkaline soil (Liogier, 1990).

Indigofera spicata is a vigorous and potentially useful tropical legume but contains hepatotoxic amino acid (Indospicine), which interferes with the metabolism (Hutton, 1970). A number of other *Indigofera* species also contain indospicine and it is recommended that they should be fed with care and not constitutes more than 50% of the diet of cattle and should not be fed to pigs or poultry (Church, 1980). *Indigofera* species have low palatability during the rainy season, but are well browsed towards the end of the dry season when secondary shoots are also eaten readily.

Indigofera species contain the pigment indigo, which may become an important commercial crop in various tropical and sub-tropical areas, apart from its use as grazing

forage and high quality supplement for ruminants (Haude, 1997). Leaves of *Indigofera* species, alone or in combination with other ingredients, are also used in herbal medicine to treat fever, headache and acute cough (Liogier, 1990).

Indigofera arrecta originated from East Africa, and it is today found throughout the world 's tropical regions. Its dispersion is largely due to the growth of indigo production in the European colonies. It was taken to Indonesia where, during the 19th century it was widely grown. The same plant was later grown in India in comparison to indigenous indigo plants. Compared to other species, *Indigofera arrecta* contains better pigment quantities and became economically the most important indigo species in India.

1.2 Chemical composition of *Indigofera* species

An analysis of *Indigofera hirsuta* (Dougall and Bogdan, 1966) indicating a composition of 23.8% crude protein (CP), 2.0% ether extract (EE), 15.2% crude fibre (CF), 46.8% nitrogen free extract (NFE), 1.88% calcium (Ca) and 0.37% phosphorus (P), demonstrated its excellent quality. However, silage prepared of *Indigofera hirsuta* satisfied only the maintenance requirements of stock and was not well eaten by sheep (Catchpoole and Henzell, 1971). One of the valuable attributes of *Indigofera hirsuta* is that it produces prolific seed, namely 440 000 seeds/kg.

The calcium concentration of *Indigofera* species is relatively high and usually ranges from 0.5 to 2.0%, where normal ranges of 0.20 to over 0.30%, would satisfy animal requirements. The stems of leguminous plants became more fibrous with age and contain more (CF) and less (CP) than the younger stems. Little fibre has, however, been observed in the leaves of legumes and they remain nutritious and palatable at an advanced stage of growth, often throughout the dry season, when legumes are of particular value for late season grazing (Bartha, 1970).

1.3 Factors affecting mineral contents of the plants

Concentrations of mineral elements in forage are dependent upon the interaction of a number of factors, including soil pH, plant species, stage of maturity, and climatic conditions.

1.3.1 Soil pH

As the soil pH increases, the availability and the uptake of Mn, Zn, Cu by forages decrease, whereas the forage Mo and Se concentration increases (McDowell, 1985).

1.3.2 Stage of maturity

As the plants mature, mineral content declines due to a natural dilution process and the translocation of nutrients to the root system. In most circumstances P, K, Mg, Na, Cl, Cu, Co, Fe, Se, Zn and Mo decline as the plant matures (Ford *et al.*, 1979).

1.3.3 Climatic conditions

The temperature and rainfall all affect the rate of growth and the rate of transpiration and the latter factor has an influence on the amount of salts in solution brought in by the roots of the plant (Dougall and Bogdan, 1958).

1.4 Environmental adaptation

Indigofera spicata is distributed in tropical Africa, South Africa, Madagascar, Sri Lanka, Southern and South-eastern Asia and tropical America. It occurs in grasslands and rocky places, but mostly on wastelands and other disturbed habitats. *Indigofera spicata* is relatively drought resistant and can grow under moderate annual rainfall and on relatively poor soils. The plants are moderately specific in their rhizobium requirements and can be inoculated by a few strains of cowpea-type rhizobium (Henzell, 1962).

Introduced into cultivation, *Indigofera spicata* has yielded well, formed balanced mixtures with grasses and was reasonably grazed. The use under cultivation is, however, restricted by its toxicity to animals expressed in liver degeneration in cows and sheep, and especially in horses, while pregnant animals can abort (Hutton, 1970).

1.5 Nutritive value

It is important to understand the term nutritive value, which is also a key in this discussion. Nutritive value is a function of the feed intake and the efficiency of extraction of nutrients from the feed during digestion. Feeds of high nutritive value promote a high level of production (Eagan *et al.*, 1986). The nutritive value of feed is determined by the ability to provide the nutrients required by animals for maintenance, growth and reproduction. The nutritive value of browse legumes depends on the voluntary intake of feeds consumed and the extent to which the quantity of dry matter consumed by animals supplements dietary energy, proteins, minerals and vitamins. Much will depend on the actual quantity of feed eaten by the animal on a daily basis (Dougall *et al.*, 1964).

1.5.1 Factors which influence nutritive value

Different plant species differ inherently in their rate of reproductive development. This results not only in changes in chemical and anatomical characteristics, but also in proportion of plant parts e.g. leaf, stem and petiole. Management and environment can then play a significant role in affecting nutritive value.

1.5.1.1 Plant maturity

Advancing plant maturity is associated with lowering of nutritive value by virtue of a decrease in leafiness and a decrease in the leaf: stem ratio, changes in the composition of the cell wall (Akin *et al.*, 1977) and loss of cell contents during maturity (Ballard *et al.*, 1990). The loss of cell contents during maturation is a major factor contributing to the decline in nutritive value.

1.5.1.2 Environment

Temperature and light are the most important environmental factors that affect nutritive value. The temperature under which plants are grown has a direct effect on the concentration of chemical constituents, with genotype then determine exactly how different species change with increasing temperature. Higher temperature usually promotes the accumulation of structural material (cell wall material) and more rapid metabolic activity, which decrease the pool size of cell contents (Ford *et al.*, 1979).

1.5.1.3 Genetic variation

Plants have adapted to specific environments through evolution and those that have evolved under grazing have protective mechanisms against predatory attack (whether it be by animals or insects). Some of those mechanisms include lignification and secondary compounds, which will influence the nutritive value (Tabe *et al.*, 1993).

1.5.2 Measurement of nutritive value

The direct estimation of nutritive value involves, at least, the measurement of digestibility. The availability of digested nutrients and their efficiency of use by the animal may either be measured directly or, more usually, predicted from digestibility using standard equations derived from a large number of feeding trials (NRC, 1985).

1.5.2.1 Total collection

The usual method for direct measurement of digestibility is a total collection trial, in which animals are constrained and the entire amount of feed eaten and faeces voided are weighed and analysed, the difference being assumed to be digested (NRC, 1985; SCA, 1990).

1.5.2.2 Marker technique

When digestibility estimates of diet consumed by grazing animals are desired, total collection trials are difficult, and indirect methods, such as the use of a marker, are therefore recommended. This can be done either by the dosing of animals with markers to estimate both faecal output and intake, or by using markers, which are part of the herbage.

1.5.3 Methods for predicting nutritive value

Nutritive value can be predicted or estimated in terms of digestibility or the voluntary intake. Coleman *et al.* (1990) reviewed methods for predicting nutritive value, which included bioassay, chemical and structural characteristics as well as instrument-based methods such as the near-infrared reflectance spectroscopy (NIRS). There are two steps involved in predicting nutritive value, which are as follows:

1.5.3.1 Data-base selection

Selection of an appropriate sample data-base, with high quality reference data, is the most important part of the prediction process.

1.5.3.2 Development and evaluation of prediction equation

Weis (1993) proposed the use of theoretically based, rather than empirical relationships or models, to predict nutritive value.

1.5.3.3 Biological procedure

Three bioassay methods have been developed i.e. *in vitro* digestibility using rumen microorganisms (Tilley and Terry, 1963), *in vitro* digestibility using an enzyme preparation (McLeod and Minson, 1978) and the *in situ*, or nylon bag-technique (Ørskov and McDonald, 1979).

1.5.3.4 Physical procedure

Near-infrared reflectance spectroscopy

Noris *et al.* (1976) were the first to report the use of NIRS to estimate chemical composition and nutritive value of forages.

The nutritive value of feeds should be ranked on the basis of the following characteristics (Leng, 1986):

- voluntary consumption potential;
- potential digestibility and ability to support high rates of fermentative digestion;
- high rates of microbial protein synthesis in the rumen relative to volatile fatty acids;
- high rates of propionic synthesis relative to total volatile fatty acids synthesis; and
- ability to provide bypass nutrients for absorption from the small intestines.

Tree legumes must have both desirable agronomic characteristics and a high nutritive value to be useful as forage. The leaves and the stems may be used either as a complete feed or as a supplement to other feeds. In some species, a major limitation to the use of

one or more of the above-mentioned components is the presence of toxic and/or antinutritive factors (Norton, 1994).

1.5.4 Anti-nutritive and toxic factors in forage tree legumes

Leguminous trees and shrubs often have thorns, fibrous foliage and growth habits that protect the crown from defoliation. Certain anti-quality factors also affect animals and the nutritive value of forages (Norton *et al.*, 1992). The anti-nutritional effects present in some tree legumes are: reduction in voluntary intake; diminished digestibility of nutrients; adverse effects upon rumen metabolism and toxicity. Non- ruminants (e.g. pigs and poultry) are usually more susceptible to toxicity, as potential toxins may be denatured in the rumen (Duke, 1977).

1.5.1.1 Strategies for managing anti-nutritive factors

- a) Use supplements to overcome the anti-nutritive factor,
 - High concentrations of condensed tannins can lower the feeding value due to reduced availability of nutrients, especially proteins and lower cell wall digestion (Barry and Blaney, 1987).

b) Reduce access to the problem feed

 By reducing the proportion of the problem legume in the diet, adverse effects can be reduced (Wildin, 1985).

1.6 Voluntary feed intake

Rumination and fermentation are relatively slow processes and fibrous feeds may have to spend a longer time in the digestive tract. If feeds and their indigestible residues are retained for longer periods in the digestive tract, the animal's daily intake will be reduced. In ruminants there is a positive relationship between digestibility of feeds and their intake i.e. there will be an increase in intake, as the energy digestibility of feeds increase (Blaxter, 1961). Actually, intake is more closely related to the rate of digestion of diets than to digestibility, although the two measures are

often related to one another, i.e. feeds that digest rapidly and are of high digestibility and promote high intake (Campling and Lean, 1983).

1.6.1 Intake of legumes

Physical regulation of intake in ruminants is a major factor influencing intake of forage by its mechanism of retention time of dry matter in the rumen. Forages with a long retention time in the rumen have a lower intake than those with a shorter retention time (Thorton and Minson, 1973). The shorter retention of legume particles is related to leaf anatomy, resulting in disintegration into small round particles. This is distinct from the long needle-like particles of the vascular bundles, as generated from grass leaves, and also faster rate of digestion of legumes compared to grasses. The physical regulation of intake is expressed as a relationship between intake and digestibility but Laredo and Minson (1973) showed that forages of the same digestibility could have vastly different intakes.

The main chemical component of feeds that determines the rate of digestion is the neutral detergent fibre (NDF), which is a measure of cell wall content. There is a negative relationship between NDF content of feeds and the rate at which they are digested. One consequence of this relationship is that those feeds that are equal in digestibility, but differ in NDF (cell wall) content, have different intakes. The two families of pasture plants, grasses and legumes, provide an example. At equal digestibility, legumes contain less cell wall and are consumed in quantities about 20% greater than grasses (Forbes, 1986).

The digestibility of plant material in the rumen is related to the proportion and lignification of plant cell wall. Tree forages with a low NDF content (20-35%) usually have a higher digestibility, while species containing lignin often have a low digestibility. Stems have higher lignin content than leaves and are thus less digestible (Bamualim *et al.*, 1980).

1.6.2 Factors which influence food intake

1.6.2.1 Psychological factors

Psychological factors also play a role in determining the feeds which animals choose, and the amount which they can consume. Chesworth (1992) stated that sheep and goats kept in pens, would eat more when they can see more food that they can consume. They suggest a practical way of increasing food intake; if animals are fed in pens, the food bins should always have sufficient feed.

1.6.2.2 Physiological factors

Animals that are offered a diet that has a very low energy content will consume more in an attempt to compensate. There are areas of the brain, in and around the hypothalamus, that monitor the animal's physiological status by measuring the level of glucose, lipids and amino acids in the blood plasma. When animals eat, the level of these compounds in the blood rise and when they do, there is a growing feeling of satisfaction, such that the animal stops eating (Forbes, 1995).

1.6.2.3 Animal size

Food intake is generally determined by the metabolic size of the animal and it is proportional to the animal's metabolic body weight. A mature animal would eat a diet, which will provide only enough food to maintain body weight and condition, whereas an animal, which is growing, requires enough food to supply its needs, both for maintenance and for extra body tissues (Illius and Allen, 1994).

1.6.2.4 Physical factor

1.6.2.4.1 Plant structure

The content of fibrous cell walls is a major factor, since these structures are less soluble and take up more space than the cell contents. Forages contain a large proportion of their organic matter content (35-80%) as cell walls, which provide the structural integrity of the plant (Jung and Allen, 1995). Minson (1990) reported that legume forages have a greater DM digestibility than grasses.

1.6.2.5 Dietary factor

1.6.2.5.1 Fibrous compounds

The source of fibre has a great influence on the rate of digestion. As grasses and legume forages mature, the nitrogen content drops and digestibility of fibrous feeds decreases. The poor digestibility of fibrous feeds is reflected in very low intakes by livestock.

1.6.2.6 Environmental factor

1.6.2.6.1 Effects of heat

When ambient temperatures are high, food intake decreases dramatically. If humidity is high, food intake is also reduced. This is because of the fact that animals produce heat inside the rumen and within their bodies.

1.6.2.7 Effects of climate and season on forage quality

1.6.2.7.1 Temperature

Lower digestibility at higher temperature is the result of the combination of two main effects i.e. high environmental temperatures result in the increased lignification of plant cell wall and high temperatures also promote more rapid metabolic activity. This activity decreases protein and soluble carbohydrates and increases the structural cell wall components (Van Soest, 1994).

1.6.2.7.2 Water

Lack of water tend to retard plant development and thus to slow maturity with the result that digestibility is increased and dry matter yield is reduced. Various studies have shown that lack of water increases digestibility and irrigation tends to decrease it (Wilson, 1983; Evans and Wilson, 1984; Dias Filho *et al.*, 1991).

1.6.2.7.3 Soil

Plants grown on different soils offer a different balance of mineral elements, which influence their growth and composition. Soil effects can be viewed from two

points mainly: the accumulation in the plants of minerals and the influence of minerals in the plant on its organic matter yield, composition, and digestibility (Metson, 1978).

1.6.2.7.4 Defoliation and diseases

The physical loss of leaves, stems, or both represents a major stress that puts pressure on the plants to mobilize its reserves and put forth new leaves to restore its photosynthetic capability (Parsons *et al.*, 1988; Parsons and Penning, 1988).

1.6.3 Utilization and beneficial effects of forage legumes

There are a number of advantages concerning the use of leguminous forages (Devendra, 1988). These include:

- Provision of variety in the diet;
- Source of dietary nitrogen (N), energy, minerals and vitamins;
- Laxative influence on the alimentary system;
- Reduced cost of feeding.

1.7 General description of Leucaena leucocephala

With *Leucaena leucocephala*, for example, the forage provides a valuable source of protein, energy and sulphur for rumen bacteria. This genus includes about 50 species, which occur almost exclusively in tropical America. It originated from Mexico but spread by accidental introduction first to the Caribbean islands and then to other areas and now has a pan-tropical distribution. This plant is valued for: its ability to withstand repeated defoliation, high yields of foliage and its tolerance to low soil fertility and relatively low rainfall. Slow early growth and a risk of animal poisoning are weak points (Plucknett, 1970).

The toxic constituent in *Leucaena* is a non-protein amino acid, mimosine, which is an antimitotic and depilatory agent (Hegarty *et al.*, 1964). Mimosine occurs in all parts of the *Leucaena* plant, but in high concentrations particularly in the tips of actively growing shoots (8-12%) and young leaves (4-5%) (Lowry *et al.*, 1983). The effect only occurs if

Leucaena constitutes a high proportion of an animal's diet (>30%), for an extended period, and may be negated by innoculation with specific rumen bacteria.

1.7.1 Environmental adaptation

Leucaena leucocephala is tolerant of adverse moisture conditions, apparently because of its deep roots and can be grown at an annual rainfall ranging from 500 to 5000 mm. At a low rainfall range it responds well to irrigation. Well-drained soils are required for good growth and high yields and waterlogging or flooding are not tolerated. It can withstand a slight soil acidity (of up to pH=5.0) but grows much better in neutral or slightly alkaline soils. Leucaena leucocephala is more tolerant of a low phosphorus status of the soil than a number of other tropical legumes and this may be due to the presence of endotrophic mycorrhiza which has been found in the roots (Possingham et al., 1971).

Leucaena is a tropical species requiring warm temperatures (25-30°C) for optimum growth (Brewbeker et al., 1985). It is not tolerant of frost which causes shedding of the leaves (Isarasenee et al., 1984). It is well known for its high nutritional value and for the similarity of its chemical composition with that of lucerne. Tannins in the leaves, and especially in the stem of Leucaena, reduce the digestibility of the dry matter and protein. Digestibility and intake values for Leucaena range between 50-71% (Jones, 1979). The lower values were suggested by Jones (1969) to be associated with effects of Mimosine on intake when pure diets of Leucaena were fed.

1.7.2 Chemical composition

Crude protein, in the majority of references quoted by Hill (1971), range from 14 to 19% in dry matter for the whole herbage, but Oaks (1968) gave a wider range, 15 to 25%. The content of CF usually fluctuates from 33 to 38%, NFE from 35 to 44%, CP and CF contents in the leaves are given as 28.8 and 12.8%, respectively. CP contents vary with plant age, which in its turn depends on the frequency of utilization.

1.7.3 Herbage productivity

Dry matter productivity varies with soil fertility and rainfall, edible yields range from 3 to 30 tons dry matter/ha/year. Deep fertile soil receiving more than 1500mm of well-distributed rainfall produced the largest quantity of fodder. Yields of *Leucaena*, where the temperature limits the growth rates, may be 1,5 to 10 tons of edible fodder/ha/year (Brewbaker *et al.*, 1985).

The minimum requirements of ruminants for phosphorus (P) varies from 1.2 to 2.4g/kg feed dry matter, depending on the physiological function. Forage trees generally have high P concentrations (McMeniman and Little, 1974). Calcium (Ca) is closely associated with P metabolism in the formation of bones, and a Ca: P ratio of 2:1 is usually recommended for ruminant diets. Ca is rarely limiting in forage diets and the same is true for forage trees (Norton *et al.*, 1992). High concentration of oxalic acid in the leaves may, however, decrease the availability of Ca during digestion and affect Ca metabolism in sheep (Gartner and Hurwood, 1976).

1.8 Limitations and problems

1.8.1 Non-toxic secondary plants compounds

The non-toxic compounds limit the nutritive value of forages by lowering their digestibility and palatability (Van Soest, 1982). Higher concentrations (>20g/kg DM) of these compounds are required for negative effects and the primary site of activity is in the digestive tract or sensory organs associated with feeding behavior (Reed *et al.*, 2000).

1.8.2. Tannins

In particular, many tree legumes contain condensed tannins (CT). Tannins may have both positive and negative effects on feed quality for ruminants. Tannins are water-soluble phenolic compounds in plants with a molecular weight of >500 and with the ability to precipitate gelatin and other proteins from aqueous solution. In high concentrations they reduce intake and digestibility of proteins and carbohydrates, which will ultimately lead to a reduced animal performance.

Tannins can also increase the flow of protein compound through the rumen to the small intestines, thereby escaping microbial fermentation (McNeill *et al.*, 1998). Forages containing tannins can also protect animals against diseases caused by parasitic worms e.g. lambs grazing legume forage that contains tannins have a lower faecal parasitic egg count and worm burdens than lambs grazing *Medicago sativa*, which does not contain tannins (Reed, 1995).

Table 1.2. Example of non-toxic plant compounds of tannins present in forage and browse legumes

Pasture/browse Legumes	Predominant tannins	t Anima	al Nutritional effect		
Acacia aneura	CT^*	Sheep	Reduction in N digestibility, decreased		
			wool yield and growth (Prichard et al.,1988).		
A. cyanophylla	CT	Sheep	Reduced feed intake, negative N		
			digestibility, loss in weight (Reed et al., 1990).		
A. nilotica	CT	Sheep	low growth rate, reduced N and NDF		
			digestibility (Tanner et al., 1990).		
L. leucocephala	CT	Poultry	Poor N retention, low apparent ME		
			(D'Mello and Acamivic, 1989).		

^{*}CT means condensed tannins

1.8.3 Lignin

Plant stems contain more lignified structural tissue than leaves and as a result are much less digestible (Moore and Jung, 2001). It is known that forage lignin concentrations vary, depending on the environmental conditions, where warm temperatures tend to increase lignin concentration in tropical plants. Lignification tends to decrease under low light, because under limited light plant development is delayed (Reed, 1995). Lignin also reduces the nitrogen balance of the animals by increasing endogenous and microbial nitrogen loss in faeces (Woodward and Reed, 1995).

1.8.4 Toxic-compounds in plants

Plants contain a wide range of toxic compounds, which may affect animals. Animal species differ in their susceptibility to plant toxins. For example, browsers are less susceptible than grazers (Cheeke, 1995).

1.8.5 Mimosine

In ruminants, the deleterious effects of mimosine are diverse, including loss of hair and wool, organ damage and death in animals unadapted to *Leucaena leucocephala* forage or in those given intravenous or oral doses of the pure amino acid (Reis *et al.*, 1975). A solution to the mimosine problem could be the development of low mimosine cultivars. However, low mimosine types are found to be less productive and have poor vigour. The other approach is to feed leucaena mixed with other feeds. Hiremath (1981) suggested that the use of leucaena fodder might be restricted to 30% of the forage in the case of cattle and 50% for goats.

1.8.6 Indospicine

The toxic agent is 1-2-amino-6-amidinohexanoic acid, which was named indospicine (Hutton, 1970). Its toxicity to the animals is expressed in liver degeneration in cows and sheep, and especially in horses, and pregnant animals can abort (Hegarty and Pound, 1968).

1.8.7 Saponins

These are widely distributed in the plant kingdom and have a bitter taste and foaming properties (Agarwal and Rastogi, 1974). They have several negative effects that include poor growth, ruminal bloat, reduced feed intake and palatability, enzyme inhibition reduced nutrient absorption, antifungal activity that affects ruminal microbiology, rumen metabolism and ammonia binding properties (Cheeke, 1995).

1.9 Hypothesis and objectives

The main objective of this study is to evaluate the dry matter production, intake and the nutritive value of five *Indigofera* species, which are as follows; *I. arrecta*, *I. cryptantha*, *I. costata*, *I. amorphoides* and *I. viciodes*. This objective can be achieved through the analysis of chemical composition, *in vitro* digestibility and the determination of voluntary intake. The tropical legumes appear to be a richer source of protein and most minerals than grasses, and more usually legumes supplement grasses to improve the overall nutritive value of forage. Therefore it can be hypothesized that *Indigofera* species can be utilized as a supplement to grazing livestock (CP and minerals).

CHAPTER 2

2 MATERIALS AND METHODS

2.1 INTRODUCTION

Ten forage legume species, which were planted in January 2003, were as follows: Indigofera arrecta, I. amorphoides, I. viciodes, I. coerula, I. costata, I. trita, I. brevicalyx, I. vohemarensis, I. spicata and I. cryptantha. The plots were $3 \text{ m} \times 1.5 \text{ m}$ consisting of three plants rows with a 50 cm space between the rows.

2.1.1 Study location

The study was conducted at the University of Pretoria, Hatfield Experimental Farm in Pretoria, which is at an altitude of 1372 m. The area receives an average rainfall of 674 mm per annum. The average temperatures and rainfall data for the period January, February, March and April of 2003 and 2004 are presented in Table 2.1.

Table 2.1 Average temperature and rainfall for Hatfield Experimental Farm (Supplied by the Weather Bureau of South Africa)

	2003					
	January	February	March	April		
Min. Temperature (°C)	17.0	18.2	14.3	11.9		
Max. Temperature (°C)	28.9	28.6	28.2	25.8		
Rainfall (mm)	32.5	110.8	68.7	0		
	2004					
	January	February	March	April		
Min. Temperature (°C)	16.9	16.0	15.4	11.6		
Max. Temperature (°C)	26.3	25.6	22.2	24.7		
Rainfall (mm)	64.8	160.7	168.3	32.8		

2.1.2 Sample collection site

In the autumn of 2003 all the species were harvested at the same physiological stage. Five species were harvested in the autumn of 2004 i.e. *I. arrecta, I. cryptantha, I. costata, I. amorphoides and I. viciodes*. The third harvest was the spring re-growth after the winter of 2004, of the same species harvested in autumn 2004. The plants were harvested 10-15 cm from the ground. Only the leaves and stem with a diameter of <3 mm (edible material) were fed dry to sheep. Three plants from each of the five species were randomly harvested separately for chemical analysis.

The total dry matter accumulated for both *Indigofera* species and *Leucaena leucocephala* was 510-600kg which was used for an intake study using sheep. The sun dried plant material was fed to fifteen (15) Merino sheep, three times a day i.e. in the morning, midday, and in the afternoon. They were adapted for 10 days before the actual experiment of 7 days started and fed 2 kg DM/animal/day. The voluntary intake of *Indigofera* species and *Leucaena leucocephala* was then compared with that of *Medicago sativa*.

2.1.3 Sample preparation

All five plant species were dried and milled to pass through a 1 mm sieve size for chemical analysis. The following parameters were determined: dry matter (DM), ash, nitrogen (N), crude protein (CP), neutral detergent fibre (NDF), *in vitro* digestibility (IVDOM) and minerals (Ca, P, Mg, Cu, Zn and Mn).

2.2 Chemical analysis

2.2.1 Dry matter determination

A crucible was cleaned and dried in the oven for an hour. After an hour the crucible was removed and allowed to cool for at least half an hour in a dessicator. The crucible was then weighed to determine the dry mass. One gram (g) of the sample was then weighed in to the crucible. The sample and the crucible were dried for 18-24 hours at 100°C. The crucible and sample were then placed in a dessicator for half an hour to cool, before weighing (AOAC, 1990).

The dry matter % was calculated as follows:

DM % =
$$\frac{\text{Dry mass (g)}}{\text{Sample mass (g)}} \times 100$$

2.2.2 Ash determination

The crucible with dry sample was placed in a cold incinerating oven and then switched on at 600°C for four (4) hours. The oven was allowed to cool down for two hours and then placed in a dessicator to cool for another half an hour. The crucible and ash was then weighed. Ash % was calculated as follows:

Ash % =
$$\frac{\text{Ash mass (g)}}{\text{Sample mass (g)}} \times 100$$

2.2.3 Neutral detergent fibre determination

The NDF concentration was determined according to Robertson and Van Soest (1981) using the "tector fibertec system". A one gram sample was weighed in a filter crucible and placed in a hot extraction unit, and then a neutral detergent solution (NDS) was added into the crucible and boiled for an hour. Solution was removed by washing with hot distilled water. The residues were dried at 100°C and then cooled in a dessicator for half an hour and weighed. They were placed in a furnace at 600°C for three hours to be ashed. The oven was allowed to cool and the crucible with residue was placed in a dessicator to cool.

NDF was calculated as follows:

NDF % =
$$\frac{\text{W1 - W2}}{\text{W3}} \times 100$$

Where: W1= dry mass of sample after NDS extraction

W2= Mass of ash

W3= Sample mass

2.2.4. Leaf: stem ratio

Subsamples of all five *Indigofera* species which were harvested were used for this aspect. The leaves of each plant species were separated from the stem. The mass of each component was then determined and the dry matter content determined to establish the leaf:stem ratio. The leaf:stem ratio was calculated as follows:

2.2.5 Nitrogen and Crude protein determination

The nitrogen content of pasture sample was determined by macro Kjedahl method (AOAC, 1990), using a block digester and a Tecator kjeltec Model 1002.

$$CP \% = N\% \times 6.25.$$

2.2.6 Organic Matter (OM)

The organic matter concentration for calculating the *in vitro* digestibility was calculated as follows:

OM =
$$\frac{DM (g) - Ash (g)}{Sample mass} \times 100$$

2.2.7 *In vitro* digestibility of organic matter (IVDOM)

The *in vitro* technique requires rumen fluid which was obtained from rumen fistulated sheep fed with 100% lucerne. The method is based on Tilley and Terry (1963) as modified by Engels and Van der Merwe (1967) with 0.2 g samples being fermented anaerobically with rumen fluid, urea solution, artificial saliva mixture and carbon dioxide

for 48 hours at 39° C. The tubes were centrifuged at 2500 RPM for 15 minutes and then the clear liquid was decanted.

The dry matter residue was hydrolyzed with 20 ml of HCl and acid pepsin for a further 48 hours. After 48 hours, tubes were centrifuged at 2500 RPM for 15 minutes, then decanted, warm water added and centrifuged and clear liquid decanted as in stage one. The undigested residues were placed in the oven at 100° C for 18 hours. They were then cooled in a dessicator and weighed. The undigested residue was then placed in a furnace at 550° C for three (3) hours, cooled and weighed. A *Panicum maximum* with an IVDOM of 70-75 % was used as a standard.

IVDOM was calculated as follows:

2.3 Minerals

The following mineral contents in all the samples were analyzed: calcium (Ca), phosphorus (P), magnesium (Mg), copper (Cu), zinc (Zn), and manganese (Mn).

A 0.5g sample was weighed in duplicate and digested in a block digester at 240° C using the wet digestion technique. After the samples had been weighed, 25 ml of Nitric acid was added and tubes placed on a block digester for approximately 10- 15 minutes, and then taken off to cool for 5 minutes.

Then 10 ml of perchloric acid (HCLO₄) was added and placed back on the block for another 20 minutes, until it was clear. Allowed to cool and then diluted with 50 ml of distilled water, before being capped into the bottles. Calcium concentration was determined on a 5100PC Atomic Absorption Spectrophotometer, whereas magnesium, copper, zinc and manganese were determined on a GBC 905AA Atomic Absorption

Spectrophotometer. Phosphorus concentration was determined on a Technicon Auto Analyzer with the concentration determined from calibration curve.

The laboratory standards and controls with known concentration were also used in all the minerals analyzed to get accurate figures. Macro minerals were calculated as follows:

$$\% = \frac{\text{Reading ppm} \times \text{Initial Volume} \times \text{Dilution}}{\text{Sample mass} \times 10000}$$

Trace minerals were calculated as follows:

2.4 Voluntary intake trial

2.4.1 Feeding of experimental animals

A total of fifteen Döhne-merino sheep (wethers) were used. Before the trial, the animals were weighed and starved overnight. The animals were fed a mixture of five *Indigofera* species, which was compared with *Leucaena leucocephala* and lucerne as a control. They were fed *ad libitum*, three times a day at six-hour intervals i.e. 06H00, 12H00 and 18H00 in feed bins and had free access to fresh water. The animals were adapted for ten days in the metabolic house and during this period the voluntary intake was determined. Faichney (1992) suggested that the animals should to be maintained in a steady state by feeding continuously or at short regular intervals during the trial period for the most accurate results. During this period the orts were collected before the next feeding. After the adaptation period, the animals were fitted with faecal bags and kept individually in metabolic cages in the metabolic house for the actual experiment.

2.4.1.1 Feed sample

Feed samples were taken of the fresh feed offered daily and placed in a plastic bag and frozen. The orts were also taken before the next feeding and frozen for individual sheep. At the end of the experiment the sub-sample (10%) for individual animals was analyzed for DM, ash, and NDF.

2.4.1.2 Faeces sample

The faeces excreted daily by individual sheep was collected in faecal bags, weighed and a 10% grab sample then frozen in a plastic bag at -10°C. After the trial the faeces for individual sheep were mixed to obtain a representative sample. The initial dry matter content of each sheep was determined by drying 50g faeces sample at 100°C. The other 200g of faeces sample were dried at 60°C and ground through a 1mm sieve for laboratory analysis.

2.5 Statistical analysis

An analysis of variance with the GLM procedure (Statistical Analysis System, 2001) was used to determine the significant differences between different treatments and years for the balanced data. Means and standard deviation (SD) were also calculated. Significance of difference (5%) between means was determined by Bonferroni test (Samuels, 1989). The species and years interactions were also taken into account in the statistical analysis.

CHAPTER 3

3. Results and Discussion

3.1 Dry matter production

3.1.1 Leaf DM yield (g/plot)

The results of the leaf DM yield of all the plant species are presented in Table 3.1.

Table 3.1 The leaf DM yield (g/plot) of five Indigofera species

Species	2003	2004	2004
	Autumn	Autumn	Spring
I. amorphoides	194.8 ^a ₁ (± 116.1) [*]	152.2 ^a ₁ (± 89.6)	120.8 ^a (± 47.0)
I. cryptantha	$99.4^{ab}_{1} \ (\pm \ 29.0)$	86.3 ^{ab} ₁ (± 33.9)	89.3 ^a ₁ (± 29.2)
I. costata	24.5 ^b ₁ (± 9.8)	$23.3^{b}_{1}~(\pm~4.0)$	37.0 ^a ₁ (± 11.6)
I. viciodes	7.1 ^b ₁ (± 3.8)	27.2 ^b ₁ (± 15.7)	31.7 ^a ₁ (± 7.5)
I. arrecta	114.4 ^{ab} ₁ (± 27.7)	$89.3^{ab}_{1} (\pm 35.0)$	85.7 ^a ₁ (± 40.5)

^{a,b,c}Column means with common superscripts do not differ significantly (P>0.05)

There were significant differences during the autumn of 2003 in terms of available leaf dry matter yield between *I. amorphoides* and *I. costata* as well as *I. viciodes*. However, there were no significant differences found between *I. amorphoides*, *I. cryptantha* and *I. arrecta* as well as between *I. costata* and *I. viciodes*. During the autumn of 2004, significant differences were detected between *I. amorphoides* and *I. costata* as well as *I. viciodes*. However, no significant differences were found between *I. amorphoides*, *I. cryptantha* and *I. arrecta* as well as between *I. cryptantha*, *I. costata*, *I. viciodes* and *I. arrecta*. There were no significant differences during spring between the species. The leaf

 $^{^{1,2}}$ Row means with common subscript do not differ significantly (P> 0.05)

^{*}Standard deviation (SD)

DM yields in this study appeared to decrease with advancing maturity, environmental factors and cutting intervals, although not significant between the years (see Table 3.1). This was supported by Smith *et al.* (1992) who reported that the DM yield would increase due to the effect of environmental factors (temperature, rainfall), longer grazing or cutting intervals and advancing maturity. The proportion of inedible plant material will, however, also increase leading to a decline in forage quality.

3.1.2 Stem DM yield (g/plot)

The results of the stem DM yield of all the plant species are presented in Table 3.2.

Table 3.2 The stem DM yield (g/plot) of five *Indigofera* species

Species	2003	2004	2004
	Autumn	Autumn	Spring
I. amorphoides	127.5 ^a _{1,2} (± 79.9) *	206.9 ^a ₁ (± 124.6)	58.2 ^a ₂ (± 33.7)
I. cryptantha	45.6 ^{ab} ₁ (± 18.2)	$108.7^{ab}_{1}~(\pm~24.4)$	29.7 ^a ₁ (± 11.2)
I. costata	14.8 ^b ₁ (± 7.5)	79.0 ^b ₁ (± 11.4)	13.0 ^a ₁ (± 2.6)
I. viciodes	2.0 ^b ₁ (± 1.6)	43.7 ^b ₁ (± 12.0)	$10.0^{a}_{1} (\pm 2.2)$
I. arrecta	84.8 ^{ab} _{1,2} (± 30.2)	143.9 ^{ab} ₁ (± 44.3)	50.9 ^a ₂ (± 27.6)

a,b,c Column means with common superscripts do not differ significantly (P>0.05)

During the autumn of 2003, there were significant differences found between *I. amorphoides* and *I. costata* as well as *I. viciodes*. There were, however, no significant differences between *I. amorphoides*, *I. cryptantha* and *I. arrecta* as well as between *I. costata and I. viciodes*. There were significant differences detected during the autumn of 2004 between *I. amorphoides* and *I. costata* as well as *I. viciodes*. However, there were no significance differences between *I. cryptantha*, *I. costata*, *I. viciodes* and *I. arrecta*.

^{1,2}Row means with common subscript do not differ significantly (P> 0.05)

^{*}Standard deviation (SD)

During spring of 2004, there were no significant differences found between the species. There were significant differences detected between autumn and spring of 2004 for *I. amorphoides* and *I. arrecta*. Evans and Rotar (1987) reported that climate, soil types, maturity and management practices (such as fertilizer use, height and cutting interval as well as intercropping) may affect the DM yield.

3.1.3 Total DM yield (g/plot)

The results of the total DM yield of all the plant species are presented in Table 3.3

Table 3.3 The total DM yield (g/plot) of five *Indigofera* species

Species	2003	2004	2004
_	Autumn	Autumn	Spring
I. amorphoides	$322.3^{a}_{1,2} (\pm 194.4)^{*}$	359.1 ^a ₁ (± 213.3)	179.0 ^a ₂ (± 80.0)
I. cryptantha	145.0 ^{ab} ₁ (± 47.1)	195.1 ^{ab} ₁ (± 58.1)	119.0 ^a ₁ (± 40.4)
I. costata	39.3 ^b ₁ (± 17.3)	102.2 ^b ₁ (± 10.1)	50.0 ^a ₁ (± 10.0)
I. viciodes	9.1 ^b ₁ (± 5.4)	70.9 ^b ₁ (± 19.7)	41.3 ^a ₁ (± 9.6)
I. arrecta	199.2 ^{ab} (± 57.9)	230.9 ^{ab} ₁ (± 79.1)	136.7 ^a ₁ (± 68.0)

a,b,c Column means with common superscripts do not differ significantly (P>0.05)

In autumn of 2003, there were significant differences found in terms of total dry matter yield between *I. amorphoides* and *I. costata* as well as *I. viciodes*. There were, however, no significant differences between *I. amorphoides*, *I. cryptantha* and *I. arrecta* as well as between *I. cryptantha*, *I. costata*, *I. viciodes and I. arrecta*. There was a significant difference during autumn 2004 between *I. amorphoides* and *I. costata* as well as *I. viciodes*. However, no significant differences were detected between *I. amorphoides*, *I. cryptantha* and *I. arrecta* as well as between *I. cryptantha*, *I. costata*, *I. viciodes* and *I. arrecta*.

 $^{^{1,2}}$ Row means with common subscript do not differ significantly (P> 0.05)

^{*}Standard deviation (SD)

There were no significant differences found during the spring of 2004 in terms of the total dry matter yield between all the species. There was, however, a significant difference between the autumn and spring of 2004 for *I. amorphoides*. Van Soest (1982) reported that as the forage matures there is an increase in dry matter yield leading to a decline in digestible dry matter.

3.2 Leaf to stem ratio

The results of the leaf to stem ratio of all the species are presented in Table 3.4

Table 3.4 Leaf:stem ratio of leaves and stems of five *Indigofera* species

Species	2004 Autumn	2004 Spring	
I. amorphoides	47:53 ^a ₁	59:41 ^a ₂	
I. cryptantha	41:59 ^a ₁	59:41 ^a ₂	
I. costata	41:59 ^a ₁	57:43 ^a ₂	
I. viciodes	43:57 ^a ₁	57:43 ^a ₂	
I. arrecta	52:48 ^a ₁	52:48 ^a ₁	

^{a,b,c}Column means with common superscripts do not differ significantly (P>0.05)

There were no significant differences found within the seasons for all the plant species. However, there was a significant difference between the seasons for all species except for *I. arrecta*, which showed no significant difference. There were lower leaf:stem ratios during the autumn of 2004, which resulted in a decrease in CP content, IVDOM and an increase in NDF concentrations (see Tables 3.7; 3.12 and 3.10). Therefore, a leaf to stem ratio is a good indicator of forage quality. Crowder and Chheda (1982) reported that the decline in forage quality with maturity is primarily due to the increasing lignification of the stem and an increasing proportion of the stem compared to leaf. Legume quality is affected by leaf:stem ratio. Shehu *et al.*(2001) reported that the leaf: stem ratio in legumes is valuable because the leaves are metabolic organs and the quality of stems are largely affected by their structural function. It is important to note that both maturation

 $^{^{1,2}}$ Row means with common subscript do not differ significantly (P> 0.05)

and ambient temperature will affect various parts of the same plant differently (Buxton *et al.*, 1995).

3.3 Chemical composition

Samples from the five species (*Indigofera arrecta, I. cryptantha, I. costata, I. viciodes, and I. amorphoides*) were collected in autumn and spring. The results of the chemical composition are presented below.

3.3.1 Ash concentration

Table 3.5 The ash concentration (%) of leaves and edible components (leaves & fine stems) of five *Indigofera* species

Species	Leav	res	Edible (leaves	& fine stems)
	2003	2004	2004	2004
	Autumn	Autumn	Autumn	Spring
I. amorphoides	$13.4^{a}_{1}(\pm 0.27)^{*}$	$5.8^{a}_{2}(\pm 0.26)$	5.1 ^a ₂ (± 0.38)	$6.6^{ab}_{1} (\pm 0.06)$
I. cryptantha	9.1 ^b ₁ (± 0.38)	5.5° ₂ (± 0.24)	4.5 ^a ₂ (± 0.18)	8.2 ^a ₁ (± 1.11)
l costata	13.4 ^a ₁ (± 3.81)	5.0 ^a ₂ (± 0.19)	4.1 ^a ₂ (± 0.10)	6.8 ^{ab} ₁ (± 0.35)
I. viciodes	9.6 ^b ₁ (± 0.11)	$7.0^{a}_{2}(\pm 0.13)$	4.5 ^a ₂ (± 1.45)	6.2 ^b ₁ (± 0.66)
I. arrecta	12.2 ^{ab} ₁ (± 1.23)	5.9 ^a ₂ (± 1.19)	4.5 ^a ₂ (± 0.65)	$7.5^{ab}_{1} \ (\pm \ 0.31)$

a,b,c Column means with common superscripts do not differ significantly (P>0.05)

3.3.1.1 Leaves

There were significant differences in the ash concentration of leaves during the autumn of 2003 between *I. amorphoides* and *I. cryptantha* as well as *I. viciodes*, and between *I. costata* and *I. cryptantha* as well as *I. viciodes*. However, there were no significant differences found between *I. arrecta* and all the other species as well as between *I.*

 $^{^{1,2}}$ Row means with common subscript do not differ significantly (P> 0.05)

^{*}Standard deviation (SD)

amorphoides and *I. costata*. In the autumn of 2004, no significant differences were found between the different species. There were significant differences between years in all species. There was a dramatic decrease in ash concentration during the autumn of 2004 in all the species. Thomas and Thomas (1985) as well as McDonald *et al.* (2002) reported that as the plant grows, the ash concentrations decrease (see Table 3.6). This is probably the reason for a lower ash concentration in 2004 as compared to 2003. The ash concentrations reported in this study during 2003 are in close agreement with those reported by Haafat and Hassani (1966) for lucerne (12.6%) and Van Rensburg (1968) and Everist (1969) of 9.86% for *L. leucocephala* and 11.78% for *I. arrecta*. Ahn *et al.* (1989) and Goodchild (1990) reported ash concentrations of 4.8% for *Acacia aneura* and 5.7% for *L. leucocephala* (which are also representative of fodder trees in the tropics and subtropics) compares well with the results obtained during 2004 in this study.

Table.3.6 Variations in ash concentration with forage age (years) in lucerne (Thomas and Thomas 1985; McDonald *et al.*, 2002)

Legume	Forage age	Ash %
Medicago sativa	1	12.6
	2	11.6
	3	10.8

3.3.1.2 Edible components (leaves and fine stems)

The ash concentration in the edible components of all the species in the autumn of 2004 showed no significant differences. There was, however, a significant difference in the spring of 2004 between *I. cryptantha* and *I. viciodes*. However, there were no significant differences found between *I. amorphoides* and all other species as well as between *I. cryptantha*, *I. costata* and *I. arrecta*. There were significant differences between the two seasons in the edible component of all species. The lower ash concentration in autumn compared to spring is probably due to a decrease in leaf/stem ratio (Table 3.4). Shehu *et*

al. (2001) reported that the quality of legume forage is negatively affected by an increase in the proportion of stems.

3.3.2 Crude protein concentration

Table 3.7 The crude protein concentration (%) of leaves and edible components (leaves and fine stems) of five *Indigofera* species

Species	Leaves		Edible (leaves	& fine stems)
	2003	2004	2004	2004
	Autumn	Autumn	Autumn	Spring
I. amorphoides	$26.6^{ab}_{1}(\pm 3.03)^{*}$	22.3 ^b ₂ (± 2.37)	$13.7^{a}_{2}(\pm 2.37)$	$22.8^{a}_{1} \ (\pm \ 0.96)$
I. cryptantha	29.7 ^a ₁ (± 0.67)	24.4 ^b ₂ (± 1.19)	8.10 ^a ₂ (± 1.19)	28.7 ^a ₁ (± 0.84)
l costata	22.6 ^b ₂ (± 0.31	31.1 ^a ₁ (± 3.51)	12.7° ₂ (± 3.51)	26.2 ^a ₁ (± 7.76)
I. viciodes	25.5 ^{ab} ₂ (± 3.71)	29.1 ^{ab} ₁ (± 4.26)	12.9 ^a ₂ (± 4.26)	23.6 ^a ₁ (± 5.68)
I. arrecta	$25.3^{ab}_{1}(\pm 3.78)$	24.6 ^b ₁ (± 8.91)	18.2 ^a ₁ (± 8.91)	26.1 ^a ₁ (± 3.25)

a,b,c Column means with common superscripts do not differ significantly (P>0.05)

3.3.2.1 Leaves

There were significant differences found in the CP of leaves during the autumn of 2003 between *I. cryptantha* with the highest CP and *I. costata*. However, no significant differences were detected between *I. amorphoides* and all other species. During autumn of 2004, there were significant differences between *I. costata* and *I. amorphoides*; however, there were no significant differences between *I. amorphoides*, *I. cryptantha*, *I. viciodes* and *I. arrecta*. There were, however, significant differences between the two years for all the species, except *I. arrecta*. The CP concentrations of all species in this study obtained during 2003 compares well with the CP concentrations reported by

^{1,2}Row means with common subscript do not differ significantly (P> 0.05

^{*}Standard deviation (SD)

Robertson (1988) and Ahn et al. (1989) of 26.7% for L. leucocephala and 22.5% for Acacia angustissima.

Jones (1969), reported that lucerne plants may have 18% CP, but if the leaves and the stems were separated and analyzed, the leaves will have 26% CP, while the stems might have 11% CP. Van Soest (1982) stated that as plants mature, crude protein decreases, fibre increases and digestibility declines (see Table 3.8). This is in close agreement with the CP concentration obtained during 2003 in this study. As forages mature, there is a point at which the accumulation of digestible DM declines despite increasing forage DM yields.

Table 3.8 Effect of stage of maturity on the nutrient content of lucerne (Van Soest, 1982)

Stage of maturity	Crude protein (%DM)	Neutral detergent fibre (%DM)
Vegetative	22	41
Bud	20	44
Early bloom	17	48
Mid bloom	16	50
Full bloom	15	52
Mature	13	55

The CP results in this study are similar to those reported by Karachi (1997) for *Lablab purpureus* (25%). Due to the high CP concentrations, farmers may use homegrown forages, such as *Indigofera*, lucerne and *L. leuecocephala*, to provide supplemental protein to grazing livestock (Phillips *et al.*, 2002). The CP concentration of all the plant species recorded in both years will fulfill the CP requirements of cows and ewes for different functions (Table 3.9).

Table 3.9 The crude protein requirements of different classes of ruminants (NRC, 2001)

Classes of ruminants	CP (%)
Beef cows (maintenance)	9.2
Beef cows (early lactation)	9.6
Mature ewes (maintenance)	9.5
Mature ewes (lactating)	13.3

3.3.2.2 Edible components (leaves and fine stems)

There were no significant differences in the CP of edible components identified between the species during autumn and spring (Table 3.7). However, there was a significant difference between the two seasons for all species, except *I. arrecta*, which showed no significant differences. The CP concentration of all the plant species in both seasons fell within the general range of protein concentration of 12-30% in browse plants species (Gupta and Pradhan, 1975; McDonald and Ternouth, 1975; Bamualim, 1981; Minson, 1990; Rittner and Reed, 1992).

The decline in CP concentration during autumn is probably due to a decrease in leaf: stem ratio (Table 3.4). Shehu *et al.* (2001) reported that legume quality is affected by leaf:stem ratio. Evans (2002) reported a range 12.7 to 14.1% CP for the whole plant, which compares well with the CP concentration obtained during autumn in this study. Khamseekhiew *et al.* (2001) stated that the CP concentration of edible material (leaves & small stems) of *L. leucocephala* ranged from 14-30% CP, which is in close agreement with the CP concentration in this study during spring.

3.3.3 Neutral detergent fibre concentration

The results of NDF concentration determinations are presented in Table 3.10.

Table 3.10 The neutral detergent fibre (%) of leaves and edible components (leaves & fine stems) of five *Indigofera* species

Species	Leaves		Edible (leaves	s & fine stems)
	2003	2004	2004	2004
	Autumn	Autumn	Autumn	Spring
I. amorphoides	$18.9^{a}_{2}(\pm\ 2.05)^{*}$	40.2 ^b ₁ (± 0.10)	$62.5^{a}_{1}(\pm 3.52)$	33.0° ₂ (± 1.56)
I. cryptantha	22.2 ^a ₂ (± 0.40)	$45.7^{ab}_{1}(\pm 0.40)$	65.4 ^a ₁ (± 3.12)	35.1 ^a ₂ (± 1.90)
I costata	22.5° ₂ (± 5.05)	50.4 ^a ₁ (± 0.72)	62.2 ^a ₁ (± 4.03)	34.7° ₂ (± 6.60)
I. viciodes	25.5 ^a ₂ (± 6.30)	42.2 ^b ₁ (± 0.16)	60.7 ^a ₁ (± 7.45)	36.5° ₂ (± 3.83)
I. arrecta	24.2 ^a ₂ (± 1.97)	46.5 ^{ab} ₁ (± 0.60)	59.5 ^a ₁ (± 6.32)	32.8 ^a ₂ (± 4.60)

a,b,c Column means with common superscripts do not differ significantly (P>0.05)

3.3.3.1 Leaves

There were no significant differences in the NDF of leaves detected in the autumn of 2003 between all the plant species. However, in the autumn of 2004 there were significant differences found between *I. costata* and *I. amorphoides* as well as *I. viciodes*. No significant differences were detected between *I. amorphoides*, *I. cryptantha*, *I. viciodes* and *I. arrecta* as well as between *I. cryptantha*, *I. costata* and *I. arrecta*. There were significant differences between the two years for all the species. An increase in the NDF concentrations in 2004 was probably due to the ageing of the plants. Van Soest (1982) reported that the quality of foliage decreases with advancing maturity (Table 3.11). The decreased IVDOM in the autumn of 2004 (Table 3.12), as plants matured, is similar to that reporting an increase in NDF concentration being associated with a decrease in digestibility (Van Soest, 1982).

 $^{^{1,2}}$ Row means with common subscript do not differ significantly (P> 0.05)

^{*}Standard deviation (SD)

Tree foliage with low NDF concentrations (20-35%) is usually of high digestibility and species with high lignin are often of low digestibility (Bamualim *et al.*, 1980; NRC, 2001). Goodchild (1990) reported an NDF concentration of 30% for *L. leucocephala*, which is slightly higher than the NDF concentrations obtained during 2003 and lower than those in 2004. Fodder trees and shrubs have relatively high concentrations of crude protein, minerals and NDF (Wilson, 1977; Ibrahim, 1981). This is particularly in agreement with the results obtained in this study and emphasizes their value as dry season feeds for grazing livestock.

3.3.3.2 Edible components (leaves and fine stems)

The NDF concentrations of the edible components of all species investigated within the two seasons showed no significant differences. However, there were significant differences between the two seasons. The high NDF concentrations (59.50-65.43%) in autumn are probably due to a decrease in the leaf:stem ratio (Table 3.4). The stems have higher NDF concentrations than leaves, which is due to the higher concentrations of fibre and lignin (Karachi, 1997).

The quality of stems is largely determined by their structural function, which results in an increase in NDF concentrations (Shehu *et al.*, 2001). The average NDF of the whole plant for *L. leucocephala* is 34.5%, which is comparable to the NDF concentrations obtained during spring in this study (Murphy and Colucci, 1999). The NDF concentrations obtained during autumn in this study are in close agreement with 60.30% of *Albizia chinensis* reported by Robertson (1988). It is very important to note that total NDF concentration of forage is a dominant factor in determining forage quality. Forages that contain 40% NDF or less are generally of higher digestibility than forages that contains 60% NDF (Hoffman *et al.*, 2001). The results found during spring in this study agree fully with the results by NRC (2001), that a low NDF concentration (<35%) results in higher digestibility.

3.4 Digestibility

3.4.1 IVDOM

The results of IVDOM analyses are presented in Table 3.12

Table 3.12 The *In vitro* digestibility of organic matter (%) of leaves and edible components (leaves & fine stems) of five *Indigofera* species

	Leaves		Edible (leaves & fine stems)	
Species	2003	2004	2004	2004
I. amorphoides	Autumn $71.7^{a}_{1}(\pm 4.00)^{*}$	Autumn 59.8 ^{bc} ₂ (± 1.85)	Autumn 56.8 ^a ₁ (± 3.94)	Spring 63.2 ^a ₁ (± 2.64)
I. cryptantha	70.7 ^a ₁ (± 2.88)	56.8 ^{bc} ₂ (± 2.31)	50.7 ^a ₂ (± 2.46)	72.2 ^a ₁ (± 2.60)
I. costata	65.5 ^a ₁ (± 1.21)	55.8° ₂ (± 1.38)	52.1 ^a ₂ (± 7.92)	67.7 ^a ₁ (± 3.60)
I. viciodes	65.5 ^a ₁ (± 3.96)	66.6° ₁ (± 1.85)	52.5 ^a ₂ (± 3.78)	67.1 ^a ₁ (± 7.22)
I. arrecta	$70.2^{a}_{1}(\pm 3.05)$	63.1 ^{ab} ₂ (± 1.21)	53.5 ^a ₂ (± 3.71)	65.5 ^a ₁ (± 8.23)

 $^{^{}a,b,c}$ Column means with common superscripts do not differ significantly (P>0.05)

3.4.1.1 Leaves

The *in vitro* digestibility of the organic matter of all species in the autumn of 2003 showed no significant differences. There were, however, significant differences in the autumn of 2004 between *I. costata* and *I. viciodes* as well as *I. costata* and *I. arrecta*. There were significant differences between the two years for all the species, except *I. viciodes*. As the plants mature, there is an increase in the proportion of fibre in the herbage, which has a strong influence on digestibility (McDonald *et al.*, 2002). As plants mature, IVDOM declines. Similar results were obtained by Forwood *et al.* (1988) and Relling *et al.* (2001).

 $^{^{1,2}}$ Row means with common subscript do not differ significantly (P> 0.05

^{*}Standard deviation (SD)

The IVDOM of all the species, obtained in both years, falls within the general range of tropical browse plants of 36-69% (Milford and Minson, 1968). The results of IVDOM of leaves obtained in this study in the autumn of 2004 for *I. amorphoides*, *I. cryptantha* and *I. costata* are in close agreement with IVDOM reported by Lukhele and Van Ryssen (2002) of 55.9% for *Compretum molle*. Karachi (1997) reported that the IVDOM of the leaves of *L. purpureus* was 64.4%, which is in close agreement with the IVDOM reported in the autumn of 2003 in this study. Bulo *et al.* (1985) found that the IVDOM of leaves of shrubs and tree legumes varied from 36 to 63.4%.

3.4.1.2 Edible components (leaves and fine stems)

There were no significant differences found between all species for both seasons. However, there were significant differences between the two seasons for all species, with a higher IVDOM in spring compared to autumn, except for *I. amorphoides*, which showed no significant difference (56.80% and 63.15%). An increase in the IVDOM in spring was a result of a higher leaf:stem ratio (Table 3.4). The results obtained during autumn in this study compared well with the IVDOM reported by Lukhele and Van Ryssen (2002) of 52.6 to 54.3% for *Colophospermum mopane*. The decline in IVDOM in the autumn of 2004 is probably due to a decrease in leaf:stem ratio as a result of advancing maturity (Table 3.4).

3.5 Minerals

Livestock producers generally provide mineral supplements to meet the dietary requirements of their animals. As a matter of fact, it is known that deficiencies in certain minerals can cause health problems e.g. low Ca intake may, or will, cause thin and brittle bones. Therefore, it is important to understand the knowledge of mineral requirements of forage plants and grazing animals. A good nutrition programme not only meets the animal's needs, but also does so at minimal cost. This emphasis on cost is essential since cost/return analysis of livestock feed costs represent approximately 50-70% of the total cost, and feed costs are one of the few areas in which producers can make significant changes (Meissner *et al.*, 1995).

3.5.1 Macro elements

3.5.1.1 Calcium concentration

The results of calcium concentration of all the species are presented in Table 3.13.

Table 3.13 The calcium concentrations (%) in leaves and edible components (leaves and fine stems) of five *Indigofera* species

Species Lea			Edible (leaves a	nd fine stems)
•	2003	2004	2004	2004
	Autumn	Autumn	Autumn	Spring
I. amorphoides	$3.87^{ab}_{1}(\pm 0.45)^{*}$	$1.79^{a}_{2}(\pm 0.02)$	$1.03^{a}_{2}(\pm 0.13)$	$2.12^{a}_{1}(\pm 0.13)$
I. cryptantha	2.66 ^b ₁ (±0.07)	1.34 ^a ₂ (±0.05)	1.20 ^a ₂ (±0.25)	1.82 ^a ₁ (± 0.13)
I. costata	4.52 ^a ₁ (±1.52)	0.22 ^a ₂ (±0.03)	0.99 ^a ₂ (±0.06)	1.73 ^a ₁ (± 0.13)
I. viciodes	3.22 ^{ab} ₁ (±0.12)	1.44 ^a ₂ (±0.04)	1.38 ^a ₁ (±0.68)	1.61 ^a ₁ (± 0.13)
I. arrecta	3.79 ^{ab} ₁ (±0.63)	0.97 ^a ₂ (±0.03)	1.20 ^a ₂ (±0.32)	1.96 ^a ₁ (± 0.13)

^{a,b,c}Column means with common superscripts do not differ significantly (P>0.05)

3.5.1.1.1 Leaves

There was a significant difference in the Ca concentration in autumn of 2003 between *I. cryptantha* and *I. costata*, although no significant difference was detected among *I. amorphoides*, I. *cryptantha*, *I. viciodes* and *I. arrecta*. In the autumn of 2004, no significant differences were found between the different species. However, there were significant differences between the two years for all the species. The decrease in Ca concentrations during 2004 was probably due to an age effect, as reported by Ibrahim (1981) that Ca concentrations decrease with advancing maturity.

 $^{^{1,2}}$ Row means with common subscript do not differ significantly (P> 0.05)

^{*}Standard deviation (SD)

The Ca concentrations reported in this study in the autumn of 2004, are in close agreement with those reported by Dougall and Bogdan (1966), Van Rensburg (1968) and Everist (1969) of 1.88% for *I. hirsuta*, 2.52% for *I. arrecta* and 1.09% for *Acacia cana*. The concentrations of Ca of all the species included in this study will meet the Ca required by beef cows during lactation (0.18-0.27%), as recommended by NRC (1996). An inadequate intake of Ca may cause weakened bones, slow growth and low milk production. In a number of tropical countries (e.g. South Africa, Argentina, Brazil and Senegal) death from botulism as a result of bone chewing has been reported (McDowell, 1992). The Ca concentrations of the *Indigofera* species used in this study will satisfy the Ca requirements of ruminants (Table 3.14).

The nutrient requirements for various ruminant species are presented in Table 3.14.

Table 3.14 Nutrient requirements based on NRC and ARC for various ruminant species (McDowell, 1992 & 1997)

Elements	Requirements of ruminants (%)	Critical level based on ruminant needs (%)
Ca	0.18-0.82	0.3
Mg	0.1-0.2	0.2
P	0.18-0.48	0.25

3.3.1.1.2 Edible components (leaves and fine stems)

The Ca concentrations of the plant species investigated, within seasons, showed no significant differences. However, there was a significant difference between two seasons for all species, except *I. viciodes*. The higher Ca concentrations in spring were probably due to a higher leaf:stem ratio (Table 3.4). McMeniman and Little (1974) reported that forage tree leaves generally have higher Ca and P concentrations than stems.

3.5.1.2 Phosphorus concentration

The results of phosphorus analyses are presented in Table 3.15.

Table 3.15 The phosphorus concentrations (%) in leaves and edible components (leaves and fine stems) of five *Indigofera* species

Species	Leaves		Edible (leaves a	and fine stems)
•	2003	2004	2004	2004
	Autumn	Autumn	Autumn	Spring
I. amorphoides	$0.26^{ab}_{1}(\pm 0.08)^{*}$	$0.26^{ab}_{1}(\pm 0.01)$	$0.11^{a}_{2}(\pm 0.03)$	$0.24^{a}_{1}(\pm 0.03)$
I. cryptantha	$0.33^{a}_{1}(\pm 0.01)$	0.19 ^b ₂ (± 0.01)	0.10 ^a ₂ (± 0.04)	$0.29^{a}_{1}(\pm 0.05)$
I. costata	$0.23^{b}_{1}(\pm 0.05)$	$0.25^{ab}_{1}(\pm 0.01)$	0.10 ^a ₂ (± 0.01)	$0.27^{a}_{1}(\pm 0.09)$
I. viciodes	0.30 ^{ab} ₁ (± 0.02)	0.28 ^a ₁ (± 0.01)	0.13 ^a ₂ (± 0.01)	0.21 ^a ₁ (± 0.02)
I. arrecta	0.28 ^{ab} ₁ (± 0.02)	$0.19^{b}_{2}(\pm 0.01)$	0.15 ^a ₂ (± 0.07)	0.23 ^a ₁ (± 0.02)

 $^{^{}a,b,c}$ Column means with common superscripts do not differ significantly (P>0.05)

3.5.1.2.1 Leaves

There was a significant difference in autumn of 2003 of P concentration in leaves between *I. cryptantha* and *I. costata* however, no significant differences were obtained between *I. amorphoides* and all other species. In the autumn of 2004, there was a slight decrease in P concentrations with significant differences between *I. viciodes* and *I. cryptantha* as well as *I. arrecta*. There were significant differences between the two years, except for *I. amorphoides*, *I. costata* and *I. viciodes*.

The decrease in the autumn of 2004 was most probably due to maturity. Kabaija and Smith (1989) reported that P concentrations decline with maturity (Table 3.16). The P concentrations of all the plant species in this study during both years compared well with P concentrations reported by Van Rensburg (1968) of 0.29% for *I. arrecta*. The

^{1,2}Row means with common subscript do not differ significantly (P> 0.05)

^{*}Standard deviation (SD)

concentrations of mineral elements in the plants are dependant upon several factors e.g. stage of maturity and plant species (McDowell, 1992).

The variation in mineral composition with forage age in *Leucaena leucocephala* is presented in Table 3.16.

Table 3.16 Variation in mineral composition of forage with age (days) in *Leucaena leucocephala* (Kabaija and Smith, 1989)

Forage age (days)	P (%)	Mg (%)
21	0.12	0.42
42	0.13	0.25
63	0.10	0.25
84	0.10	0.24

3.5.1.2.2 Edible components (leaves and fine stems)

There were no significant differences in the P concentrations of edible components among all species for both seasons. However, there was a significant difference between the two seasons for all species, with an increase in P concentrations during spring, due mainly to a higher leaf:stem ratio (Table 3.4). The results obtained in the autumn of 2004 are in close agreement with the findings reported by Kabaija and Smith (1989) of 0.10%-0.13% for *L. leucocephala* while the P concentrations obtained in spring would fulfill the P requirements of ruminants (Table 3.14).

3.5.1.3 Magnesium concentrations

The results of magnesium concentrations are presented in Table 3.17.

Table 3.17 The magnesium concentrations (%) in leaves and edible components (leaves and fine stems) of five *Indigofera* species

Species	Species Leaves		Edible (leaves and fin	
-	2003	2004	2004	2004
	Autumn	Autumn	Autumn	Spring
I. amorphoides	$1.07^{a}_{1}(\pm 0.14)^{*}$	$0.44^{b}_{2}(\pm 0.01)$	$0.50^{a}_{1}(\pm 0.08)$	$0.45^{a}_{1}(\pm 0.02)$
I. cryptantha	0.39 ^c ₁ (± 0.08)	0.32 ^{bc} ₁ (± 0.02)	0.21 ^b ₂ (± 0.08)	0.61 ^a ₁ (± 0.06)
I. costata	0.46 ^c ₁ (± 0.05)	0.41 ^b ₁ (± 0.02)	0.19 ^b ₂ (± 0.04)	0.48 ^a ₁ (± 0.23)
I. viciodes	0.52 ^{bc} ₂ (± 0.03)	0.65 ^a ₁ (± 0.04)	$0.29^{ab}_{2}(\pm 0.04)$	0.47 ^a ₁ (± 0.08)
I. arrecta	0.65 ^b ₁ (± 0.01)	0.21 ^c ₂ (± 0.02)	0.24 ^b ₂ (± 0.03)	0.47 ^a ₁ (± 0.07)

^{a,b,c}Column means with common superscripts do not differ significantly (P>0.05)

3.5.1.3.1 Leaves

There were significant differences in the Mg concentration of leaves during the autumn of 2003 between *I. amorphoides* and all other species as well as between *I. arrecta* and *I. cryptantha* and *I. costata*. No significant differences were, however, detected between *I. cryptantha*, *I. costata* and *I. viciodes* as well as between *I. viciodes* and *I. arrecta*. In the autumn of 2004, there were significant differences between *I. viciodes* and all other species as well as between *I. arrecta* and *I. amorphoides* and *I. costata*. However, no significant differences were found between *I. amorphoides*, *I. cryptantha* and *I. costata* as well as between *I. cryptantha* and *I. arrecta*. There were also significant differences between two years for all species, except for *I. cryptantha* and *I. costata*. There was a decrease in Mg concentration in 2004 in *I. amorphoides*, *I. viciodes* and *I. arrecta*, most probably due to advancing maturity. Kabaija and Smith (1989) reported that Mg

 $^{^{1,2}}$ Row means with common subscript do not differ significantly (P> 0.05)

^{*}Standard deviation (SD)

concentrations decreased with ageing (Table 3.16). The results reported by Kabaija and Smith (1989), of 0.24%- 0.42% for *L. leucocephala*, compare well with the Mg concentrations reported in this study. The Mg concentration of all the species found in this study will fulfill the Mg requirements of ruminants (Table 3.14).

3.5.1.3.2 Edible components (leaves and fine stems)

There were significant differences in the autumn of 2004 between *I. amorphoides* and *I. cryptantha*, *I. costata*, as well as *I. arrecta* however, no significant differences were found between *I. cryptantha*, *I. costata*, *I. viciodes* as well as *I. arrecta* and between *I. amorphoides* and *I. viciodes*. During the spring of 2004, no significant differences were found between all the species. There were significant differences between the two seasons for all the species, except for *I. amorphoides*. There was a marked increase for all species except for *I. amorphoides* in Mg concentration in the spring of 2004 due to an increase in leaf:stem ratio (Table 3.4). Marten *et al.* (1988) reported that leaves may have two to three times the Mg concentration of stems. The Mg concentrations of all the species in this study will satisfy the Mg required by beef cows during lactation of 0.17-0.20% (NRC, 1996).

3.5.2 Micro elements

McDowell (1997) stated that undernutrition is one of the most important limitations to grazing livestock production. Many classes of livestock are mostly dependent for all their nutrients on the quality of forage available to them, either in the form of grazing, or as conserved hay or silage.

3.5.2.1 Copper concentration

The results of copper analyses are presented in Table 3.18.

Table 3.18 The copper concentrations (mg/kg) in leaves and edible components (leaves and fine stems) of five *Indigofera* species

Species	Leav	ves	Edible (leaves	and fine stems)
•	2003	2004	2004	2004
	Autumn	Autumn	Autumn	Spring
I. amorphoides	$11.8^{a}_{1}(\pm 1.76)^{*}$	$8.8^{a}_{1}(\pm 1.36)$	9.1 ^a ₁ (± 1.15)	10.4 ^a ₁ (± 1.59)
I. cryptantha	10.9 ^a ₁ (± 0.99)	10.8 ^a ₁ (± 0.56)	9.1 ^a ₁ (± 1.44)	10.1 ^a ₁ (± 1.11)
l. costata	13.3 ^a ₁ (±2.77)	9.5 ^a ₂ (± 0.80)	10.2 ^a ₁ (± 1.53)	11.1 ^a ₁ (± 2.27)
I. viciodes	15.3 ^a ₁ (± 3.40)	10.2 ^a ₂ (± 0.64)	9.2 ^a ₁ (± 5.83)	11.8 ^a ₁ (± 1.93)
I. arrecta	13.7° ₁ (± 3.82)	9.0°2(± 0.62)	11.0 ^a ₁ (± 1.88)	9.6 ^a ₁ (± 1.15)

^{a,b,c}Column means with common superscripts do not differ significantly (P>0.05)

3.5.2.1.1 Leaves

There was no significant difference in the Cu concentration of leaves between all the species for both years. However, there were significant differences between the two years for *I. costata*, *I. viciodes* and *I. arrecta*. The Cu concentrations of all the species found in this study were above the general requirements of 6.00mg/kg for grazing animals (MacPherson, 2000) (Table 3.19). McDonald and Wilson (1980) stated that maturity leads to a decrease in Cu content of forage because of a decline in the proportion of leaf present and a drop of the Cu content of the stem. The recommended Cu concentration for beef cattle is 10mg/kg and it is also important for normal red blood cell formation (NRC, 1996).

^{1,2}Row means with common subscript do not differ significantly (P>0.05)

^{*}Standard deviation (SD)

The threshold concentration of micro-elements in forage for ruminants is presented in Table 3.19.

Table 3.19 Threshold concentration of micro-elements in forage for ruminants (MacPherson, 2000)

Minerals	Cattle	Sheep
Cu (mg/kg)		
Desirable	>10.0	>5.0
Marginal	>10.0	>5.0
Deficient	<10.0	< 5.0
Zn (mg/kg)		
Desirable	50	50
Marginal	20-40	30-50
Deficient	<20.0	<30.0
Mn (mg/kg)		
Desirable	25	25

3.5.2.1.2 Edible components (leaves and fine stems)

The Cu concentrations of all the plant species, within and between the two seasons, showed no significant differences. The results obtained in this study during spring will satisfy the Cu requirements of sheep (Table 3.19).

3.5.2.2 Zinc concentrations

The results of Zinc analyses are presented in Table 3.20.

Table 3.20 The zinc concentrations (mg/kg) in leaves and edible components (leaves and fine stems) of five *Indigofera* species

Species	Leav	/es	Edible (leaves	and fine stems)
•	2003	2004	2004	2004
-	Autumn	Autumn	Autumn	Spring
I. amorphoides	48.4 ^a ₁ (±15.54) [*]	30.3 ^a ₂ (±0.85)	31.1 ^a ₂ (±2.21)	51.8 ^a ₁ (± 4.06)
I. cryptantha	50.2 ^a ₁ (±15.99)	50.9 ^a ₁ (±6.93)	51.8 ^a ₁ (±13.75)	53.1 ^a ₁ (± 4.88)
I. costata	35.0 ^a ₁ (±8.32)	27.1 ^a ₁ (±0.30)	27.1 ^a ₂ (±9.88)	51.4 ^a ₁ (± 9.05)
I. viciodes	47.4 ^a ₁ (±8.74)	39.4 ^a ₁ (±0.06)	49.2 ^a ₁ (±13.33)	42.2 ^a ₁ (± 9.20)
I. arrecta	45.4 ^a ₁ (±4.87)	48.6 ^a ₁ (±16.33)	41.8 ^a ₁ (±21.02)	47.4 ^a ₁ (± 4.48)

^{a,b,c}Column means with common superscripts do not differ significantly (P>0.05)

3.5.2.2.1 Leaves

There were no significant differences in the Zn concentration of leaves between all the species for each year. There were, however, significant differences between two years for *I. amorphoides*. The Zn concentrations in all the species meet the requirement of ruminants and it is important for normal development and functioning of the immune system (MacPherson, 2000). The recommended Zn requirement in beef cattle is 30mg/kg, which is present in sufficient concentrations in all species for both years, except *I. costata* during 2004.

^{1,2}Row means with common subscript do not differ significantly (P>0.05)

^{*}Standard deviation (SD)

3.5.2.2.2 Edible components (leaves and fine stems)

In the autumn/spring of 2004, there were no significant differences among all species. However, the Zn concentration showed a significant difference between the two seasons for *I. amorphoides* and *I. costat*a. The Zn concentration of all species, in both seasons, will fulfill the requirements of ruminants (Table 3.19).

3.5.2.3 Manganese concentrations

The results of manganese analyses are presented in Table 3.21.

Table 3.21 The manganese concentrations (mg/kg) in leaves and edible components (leaves and stems) of five *Indigofera* species

Species	Leav	es	Edible (leaves and f	ine stems)
	2003	2004	2004	2004
	Autumn	Autumn	Autumn	Spring
I. amorphoides	$148.0^{b}_{2}(\pm 9.90)^{*}$	281.3 ^a ₁ (± 13.46)	143.8 ^a ₁ (±29.53)	125.8 ^b ₁ (±10.7)
I. cryptantha	137.4 ^b ₂ (±11.52)	279.8 ^a ₁ (± 2.51)	139.3 ^a ₁ (±33.91)	169.6 ^b ₁ (±43.2)
I. costata	153.1 ^b ₂ (±28.54)	210.6 ^b ₁ (± 3.76)	164.9 ^a ₁ (±23.24)	214.8 ^{ab} ₁ (±107.2)
I. viciodes	142.5 ^b ₂ (±1.20)	213.2 ^b ₁ (± 3.78)	117.1 ^a ₁ (± 5.83)	218.9 ^{ab} ₁ (±66.5)
I. arrecta	186.0 ^a ₂ (±13.97)	227.3 ^b ₁ (± 9.11)	165.4 ^a ₂ (±24.58)	345.7 ^a ₁ (±144.0)

^{a,b,c}Column means with common superscripts do not differ significantly (P>0.05)

^{1,2}Row means with common subscript do not differ significantly (P>0.05)

^{*}Standard deviation (SD)

3.5.2.3.1 Leaves

During the autumn of 2003, there were significant differences between *I. arrecta* and all other species in terms of Mn concentration in the leaves. In the autumn of 2004, *I. amorphoides* and *I. cryptantha* contained significantly more Mn concentrations than *I. costata*, *I. viciodes* and *I. arrecta*. However, there were no significant differences between *I. amorphoides* and *I. cryptantha* as well as between *I. costata*, *I. viciodes* and *I. arrecta*. Significant differences were found between the two years for all the species. The Mn concentration of leaves for these two years differs from the results reported by Beeson and MacDonald (1951) who stated that Mn concentrations were found not to change consistently with advancing maturity. The Mn concentration in forages is usually present in excess of the requirements of ruminants (Minson, 1990). This is in agreement with our results, with the highest Mn concentrations of 281mg/kg. Mn is important in cattle reproduction because it is required for normal oestrus and ovulation in cows and for normal libido and spermatogenesis in bulls. The Mn concentrations of all the plant species in both years will fulfill the Mn requirements of ruminants (Table 3.19).

3.5.2.3.2 Edible components (leaves and fine stems)

The Mn concentration of all species in the autumn of 2004 did not differ significantly. There were significant differences found in spring of 2004 between *I. arrecta* and *I. amorphoides* as well as *I. cryptantha*, however, no significant differences were detected between *I. amorphoides*, *I. cryptantha*, *I. costata* and *I. viciodes* as well as between *I. costata*, *I. viciodes* and *I. arrecta*. There was a significant difference between the two seasons for *I. arrecta*. The findings in this study agree with those reported by Minson (1990), that Mn concentration in forages is usually present in excess. MacPherson (2000) stated that the absorption of manganese by livestock appears to be poor and it is adversely affected by high concentrations of Ca and P. Wedekind and Baker (1990) reported, however, that an excess of P appears to be a greater inhibitor of dietery Mn than the Ca concentration. The Mn concentrations of edible components for all the species in this study will meet the requirements of ruminants (Table 3.19).

3.6 Voluntary feed intake and digestibility

The prediction of intake is important because feed costs may account for 70%, or more, of the total costs (Meissner *et al.*, 1995). Intake is more closely related to the rate of digestion of diets than the digestibility, although the two are generally related to one another. Feeds that are digested rapidly and are also of high digestibility, promote high intake. The feed intake of animals determines the amount of nutrients available for production above that required for maintenance (McDonald *et al.* 2002). Illius (1998) has suggested that intake is probably the most important variable determining animal performance and voluntary intake is generally correlated with the amount of nutrients that can be extracted from a feed i.e. digestibility. Intake of feed is related to feed quality, species of the animal, its status, energy demand and even its sex. A growing animal consumes relatively more feed than a mature one, and pregnant or lactating female consumes even more (Fox *et al.*, 1990; Robbins, 1993).

Van Soest (1982) reported that there is a greater variation in intake amongst animals than variation in digestibility and intake is, therefore, a more important factor affecting production, than digestibility. The quantity of dry matter voluntarily eaten by an animal is the most important factor controlling the productive value of a feed. Therefore, if animals consume only a small quantity of a tropical legume, the production of meat, or milk, will be low, no matter how high the protein or mineral content of each unit of feed (Milford and Minson, 1968).

The physical regulation of intake in ruminants is thought to be the major factor influencing the intake of forages, by the mechanism of retention time in the rumen. Forages with a long retention time in the rumen have a lower intake than those with a shorter retention time (Thorton and Minson, 1973). This physical regulation of intake is often expressed as a relationship between intake and digestibility, but Laredo and Minson (1973) showed that forages of the same digestibility could have different intakes.

For forages, digestibility is determined by features of the plant, but potential digestibility and hence potential intake may not be achieved due to the interactions between feeds and

the animal itself (Gill and Romney, 1994). A major factor, which could enhance intake of forages, is a lower cell wall content. This is a major reason for the advantages of legumes over grasses and immature forages over those of greater maturity (Buxton *et al.*, 1995).

Large quantities of forages can have an effect on DMI, because of the amount of fibre present and the digestibility of fibre. There are differences in the digestibility and rate of digestion for different forages species. However, intake is considered to be more important than digestibility in influencing DMI from forages (Mertens, 1992).

Environmental effects on forage quality

The environmental conditions where the plant is grown have an effect on the quality of forage, but the effects are not as great as those of increasing maturity. Temperature is one of the factors, which has a great effect. A rise in temperature reduces the leaf: stem ratio, which generally reduces forage digestion because of the lower digestibility of the stems (Buxton *et al.*, 1995). Buxton *et al.* (1995) stated that for each 1°C increase in temperature the digestibility of forages would decrease by 3 to 7%. Therefore, forages grown in cooler regions are of a higher quality than forages grown in warm climates.

3.7 Chemical composition of forages

The results of the chemical composition of the feeds (hand cut samples) used in the intake trial are presented in Table. 3.22.

Table 3.22 Chemical composition of lucerne, *Indigofera* spp and *L. leucocephala*

Parameters	Lucerne	Indigofera spp	L. leucocephala
CP (%)	$20.4 ^{\text{a}} (\pm \ 0.30)^{^{\star}}$	$14.9^{b}(\pm~0.95)$	$21.4^{a}(\pm 1.03)$
NDF (%)	43.8 °(± 1.27)	$64.6^{a}(\pm\ 2.04)$	47.9 ^b (± 2.25)
IVDOM (%)	67.7 ^a (± 1.35)	53.3 ^b (± 2.16)	46.3 °(± 1.60)

^{a,b}Row means with common superscripts do not differ significantly (P>0.05)

^{*}Standard deviation (SD)

3.7.1 Crude protein concentrations

There was a significant difference in the CP concentration between lucerne and *Indigofera* species as well as between *L. leucocephala* and *Indigofera* species. There was, however, no significance difference between lucerne and *L. leucocephala*. Jones (1979) reported that leucaena is well known for its high nutritional value and for the similarity of its chemical composition with lucerne. This is evident from the CP concentrations in this study (Table 3.22).

The CP concentrations of the three feeds used in this study are sufficient for optimal livestock production. This is supported by Leng (1990) who stated that less than 8% CP cannot sustain optimal livestock production and recommended N supplementation of such forages to obtain an optimal level of animal production. Evans (2002) reported a wide range between 12.7-14.1% for *Lablab purpureus* for the whole plant, which compares well with the results obtained in this study for *Indigofera* species (14.92%). The CP concentrations of *L. leucocephala* obtained in this study were in close agreement with the value of 20.9% CP reported by Tudsri *et al.* (2002). Duke (1983) reported a CP concentration of 20.4% of lucerne, which is similar to the results for lucerne (20.4%) in this study.

3.7.2 Neutral detergent fibre concentrations

There are significant differences between lucerne, *Indigofera* species and *L. leucocephala* of NDF. Meissner *et al.* (1989) reported an NDF concentration of 50.9% for sainfoin, which is lower than the results for *Indigofera* species obtained in this study. The NDF concentration of 42.4% for *Lablab purpureus* (Aganga and Autlwetse, 2000) is similar to the results obtained with lucerne, but lower than *L. leucocephala* in this study. NRC (2001) reported NDF values of lucerne, which ranged from 35-45%. This compares well with the results for lucerne (43.8%) in this study. Meissner *et al.* (1991) stated that intake is generally limited where NDF levels exceed 55 to 60% of dry matter, as was the case with the *Indigofera* species.

3.7.3 *In vitro* digestibility of organic matter

There are significant differences between lucerne, *Indigofera* species and *L. leucocephala*. Wilman and Asiedu (1983) reported an *in vitro* digestibility of organic matter of 67.2% for lucerne, which correspond with the results for lucerne obtained in this study (Table 3.22). Kruger (1991) reported that IVDOM of *Leucaena leucocephala* ranged from 46 to 63%. This correlates with the recordings of IVDOM for leucaena and *Indigofera* species (Table 3.22). The IVDOM values for lucerne reported by Meissner *et al.* (1989), which ranged from 59.2% to 68.7%, also correspond well with the results (67.7%) in this study. The IVDOM figures of lucerne, *Indigofera* species and *L. leucocephala* obtained in this study fall within the general range of tropical browse plants of 36-69% reported by Milford and Minson (1968).

3.8 Intake and digestibility of lucerne, Indigofera spp and L. leucocephala

Organic matter intake (OMI), digestible organic matter (DOMI) and neutral detergent fibre intake (NDFI) of lucerne, *Indigofera* species and *L. leucocephala* are presented in Table 3.23.

Table 3.23 Intake by sheep of lucerne, *Indigofera* species and *L. leucocephala*

Parameters	Lucerne	<i>Indigofera</i> spp	L. leucocephala
Initial Ave. Weight (kg)	61.0	70.5	56.5
OMI (g/d)	1414.5 ^a (± 45.4) [*]	1194.6 ^b (± 152.2)	1205.7 ^b (± 70.5)
DOMI (g/kg W ^{0.75})	44.9 ^a (± 9.5)	26.6 ^b (± 2.6)	28.6 ^b (± 5.8)
NDFI (g/d)	679.4 ^b (± 37.3)	803.8 ^a (± 94.9)	626.9 ^b (± 30.6)

^{a,b}Row means with common superscripts do not differ significantly (P>0.05)

^{*}Standard deviation (SD)

3.8.1 Organic matter intake

There were significant differences between the OMI of lucerne and that of *Indigofera* species and *L. leucocephala*. No significant differences were, however, found between *Indigofera* spp and *L. leucocephala*. There is a general trend for voluntary intake in sheep to increase with an increasing digestibility of dry matter (Milford and Minson, 1968). This is most probably the reason for an increase in the organic matter intake for lucerne as compared to *Indigofera* species and *L. leucocephala*. A number of authors have shown that ruminants decrease their intake of feeds in response to ingestion of toxins (Provenza *et al.*, 1990; Thompson and Stuedemann, 1993).

The foliage of *L. leucocephala* contains the toxic amino acid mimosine, which may reach levels of up to 12%. In the rumen this is converted to DHP (3 hydroxy-4-(1H)-pyridone), which causes goitre, loss of appetite, hair loss and loss of weight (Lowry, 1987). This was, therefore, the probable reason for a decrease in intake (OMI) of *L. leucocephala*. The lower intake of leucaena was reported by Jones (1979) to be associated with the effects of mimosine when pure diets of *L. leucocephala* were fed. Van Soest (1982) reported that intake also declines with increasing ADF and NDF concentrations in the forage and digestibility declines with increasing lignin content of the forage. This statement agrees with the lower OMI of *Indigofera* species compared to lucerne and *L. leucocephala* due to it having the highest NDF concentration (64.6%) in this study.

The higher neutral detergent fibre and lower crude protein concentrations in these *Indigofera* species (Table 3.17), compared to that of lucerne and *L. leucocephala*, could have affected the organic matter intake of *Indigofera* species (Table 3.23). This is supported by Nocek and Russell (1988), who reported that excess neutral detergent fibre often limits intake because of physical fill in the rumen. Roux and Meissner (1984) stated that the feed intake of forages is controlled by physical constraints, primarily the rumen fill and the rate of removal of digesta from the rumen, while Milford and Minson (1966) reported that the minimum crude protein requirements of the microbial population in the rumen are 7% for animal grazing tropical pastures.

3.8.2 Digestible organic matter intake (DOMI)

The amount of feed consumed by animals during the intake trial in this study is expressed as organic matter intake per day. However, in this case it is also expressed as digestible organic matter intake (DOMI) per kg metabolic livemass of the animals DOMI g/kg $W^{0.75}$ /day.

While there were significant differences in DOMI obtained between lucerne and the other two forages, there were no significant differences between *L. leucocephala* and the *Indigofera* species. Engels (1972) reported that the maintenance requirement for grazing sheep is 33.5 g DOMI/kg W^{0.75}/d. We have to account that animals in this study were fed in metabolic cages, which could have an influence on voluntary intake of animals. Nsahlai *et al.* (1997) reported that the DOMI requirements for stall fed animals are 28.2 g DOMI/kg W^{0.75}/d. The results obtained in this study, for *L. leucocephala* and lucerne, indicate that these forages will be able to supply the maintenance requirements of sheep (Table 3.23). Under this experimental circumstance *Indigofera* species did not fulfill the maintenance requirements of sheep. The lower DOMI for *Indigofera* species in this study is associated with higher NDF concentrations (Table 3.7). This was supported by Berg and Hill (1989), who reported that intake, declines with an increase in NDF concentration.

3.8.3 Neutral detergent fibre intake

There was a significant difference in NDF intake detected between lucerne and *Indigofera* species as well as between *Indigofera* species and *L. leucocephala*. No significant difference was, however, found between lucerne and *L. leucocephala*. Van Soest (1987) reported that the use of NDF intake within forages is an indicator of forage quality. Quality is closely linked to animal performance. However, great variation exists amongst forage types, which must be considered. Ruiz *et al.* (1995) stated that a measure of NDF digestibility would explain the differences in fibre quality. A measure of NDF intake could explain the indigestible and slowly digestible portion of the diet that occupies space in the digestive tract and thus lower intake.

The organic matter digestibility (OMD) and neutral detergent fibre digestibility (NDFD) are presented in Table 3.24.

Table 3.24 Digestibility of lucerne, *Indigofera* species and *L. leucocephala* utilized by

snee	9		
Parameters	Lucerne	<i>Indigofera</i> spp.	L. leucocephala
OMD (%)	$67.1^{a}(\pm 3.4)^{*}$	$63.7^{a}(\pm 2.3)$	56.4 ^b (± 2.2)
NDFD (%)	44.4 ^b (± 6.8)	55.5 ^a (± 1.1)	41.0 ^b (± 3.4)

a,bRow means with common superscripts do not differ significantly (P>0.05)

3.8.4 Organic matter digestibility

There were significant differences in organic matter digestibility (OMD) between lucerne and *L. leucocephala* as well as between *Indigofera* species and *L. leucocephala*. No significant difference was, however, found between lucerne and *Indigofera* species. McDonald *et al.* (2002) reported that feeds, that are digested rapidly and are of a high digestibility, promote high intakes. This corresponds with the OMI and OMD for lucerne in this study (Table 3.23 and Table 3.24). NRC (2001) reported a 60% OMD for lucerne forage, which is lower than the OMD of lucerne in this study.

Joyce *et al.* (1973) reported an OMD of 62.5% for lucerne, which is slightly lower than 67.1% obtained in this study (Table 3.24). Skerman (1970) reported an OMD for *L. leucocephala* of 65%, which is much higher than the OMD recorded in this study. Leucaena is well known for its nutritional value and for the similarity of its chemical composition to that of lucerne. Jones (1979) reported that the organic matter digestibility for *L. leucocephala* ranged from 50 to 71%. The OMD of lucerne, *Indigofera* species and *L. leucocephala* found in this study, falls within that range. McManus *et al.* (1985) stated that tannins in the leaves and especially in the stems of *L. leucocephala*, reduce

^{*}Standard deviation (SD)

digestibility. This is most probably true for the OMD of *L. leucocephala* reported in this study.

Tannins are secondary metabolites with a high capacity to form complexes with protein. These complexes are stable at a normal rumen pH and remain undegraded in the rumen resulting in a reduced protein availability and thus limiting animal production (McManus *et al.*, 1985). McDonald *et al.* (2002) reported that the lower the NDF concentration, the higher the digestibility. The relatively lower NDF concentration obtained in this study for lucerne (43.8%) and *L. leucocephala* (47.9%) resulted in a higher organic matter intake than with the *Indigofera* species, which had a higher NDF concentration of 64.6% (Table 3.22).

3.8.5 Neutral detergent fibre digestibility

There were significant differences in neutral detergent fibre digestibility (NDFD) between lucerne and *Indigofera* species as well as between *Indigofera* species and *L. leucocephala*. There was, however, no significant difference between lucerne and *L. leucocephalala*. The NDF digestibility values of lucerne, *Indigofera* species and *L. leucocephala*, obtained in this study are lower than the NDF digestibility of 60% for lucerne reported by Oba and Allen (1999). The primary factor that influences NDF digestibility within a species is maturity, or the stage at which the forage was harvested.

When cell and stem diameter increases and heavily lignified xylem tissues develop, NDF digestibility decreases (Hoffman *et al.*, 2001). This is probably the reason for the low NDF digestibility reported for *L. leucocephala* in this study. Hoffman *et al.* (2001) reported that lactating dairy cows would increase their dry matter intake and produce more milk when fed forages that have a higher NDF digestibility. The NDF digestibility is an important factor affecting feed intake and production. Oba and Allen (1999) reported that one unit increase of digestibility is associated with 0.17 kg increase in dry matter intake.

CHAPTER 4

4. GENERAL DISCUSSION

The objective of this study was to evaluate the dry matter production, intake and the nutritive value of *Indigofera* species. This was done by analyzing the chemical composition (ash, crude protein and neutral detergent fibre), *in vitro* digestibility of organic matter and minerals (Ca, P, Mg, Cu, Zn and Mn). The dry matter yields of all five plant species were measured as well as the leaf:stem ratio. The voluntary intake trial which was conducted, compared *Indigofera* species with *L. leucocephala* and lucerne as a control.

4.1 Dry matter production

There were higher leaf DM yields in autumn 2003 than in autumn 2004 and spring, however, *I. amorphoides* appeared to have higher leaf DM yields in both years (Table 3.1). As shown in Table 3.2 and Table 3.3, *I. amorphoides* and *I. arrecta* had the highest DM yields in both years.

4.2 Leaf to stem ratio

There were significant differences in leaf to stem ratio between the seasons (autumn and spring) during 2004 (Table 3.4). In the autumn, all the *Indigofera* species, except for *I. arrecta*, had a lower leaf:stem ratio, with the proportion of stem increasing with ageing of the plants. The highest leaf:stem ratio for *I. arrecta* (52:48) was coupled with a crude protein concentration of 18.24% followed by 13.71% obtained during autumn (2004) and the lowest NDF concentration of 59.50% (see Table 3.4 and Table 3.10). Akin *et al.* (1977) and Ballard *et al.* (1990) reported that advancing maturity is associated with a declining nutritive value as a result of a decrease in leafiness and an increase in proportion of stem material.

The lower CP concentration in the edible component during autumn of 2004, had a positive correlation with a decrease in leaf:stem ratio (Table 3.4). However, in the spring of 2004 there was an increase in the leaf:stem ratio of 59:41, 59:41, 57:43, 57:43 and

52:48 for *I. amorphoides*, *I. cryptantha*, *I. costata*, *I. viciodes* and *I. arrecta* respectively (Table 3.4). A higher proportion of leaf to stem ratio in the spring, as illustrated in Table 3.4, was correlated with a higher Zn concentration in *I. amorphoides* and *I. crypatantha*. Dougall and Bogdan (1958) reported that as the plant matures mineral contents (Cu, Zn and Fe) decline.

4.3 Chemical composition

In the autumn of 2004, there was a marked lower ash concentration in all the species as a result of advancing maturity of the plants. *I. viciodes* had the highest ash concentration. A high ash concentration was found in the edible component during the spring of 2004 of all the species, when compared to autumn of the same year. This was because of a higher leaf to stem ratio (Table 3.4). *I. amorphoides* and *I. cryptantha* had the highest ash concentrations in autumn and spring of 2004 respectively. In the autumn of 2003 and 2004 (leaves), *I. cryptantha* and *I. costata* had the highest CP concentration (Table 3.7).

There was, however, a dramatic increase in CP concentrations in the spring of 2004 in all the species, with the highest being in *I. cryptantha* (28.74%). Most importantly, all the species in this study have more than 8% CP, which was regarded by Leng (1997) as the optimum level for maintenance requirements for mature ewes. The CP concentrations in all the species in this study were relatively high. It is known that CP concentration is positively related with quality (high protein forages are generally high quality forages). Livestock fed *Indigofera* species will not, therefore, require protein supplements. This firstly reduces the feed costs, since most of the protein supplements are purchased, and secondly there will be an increase in production.

The lowest NDF concentration in the leaves of *I. amorphoides* was found in both years with 18.9% and 40.2% (Table 3.10). The lower NDF concentration in edible components during spring of 2004 is most likely the result of lower lignification and a higher leaf:stem ratio (Table 3.4).

4.3.1 *In vitro* digestibility of organic matter (IVDOM)

The IVDOM of leaves in the autumn of 2003 was the highest in *I. amorphoides*, while in autumn of 2004 it was for *I. viciodes*. A marked increase in the *in vitro* digestibility of organic matter of edible components of *I. cryptantha* in the spring of 2004 is ascribed to the higher proportion of leaf material (Table 3.4).

4.3.2 Minerals

4.3.2.1 Macro elements

The concentrations of Ca, P and Mg recorded in the *Indigofera* species, in this study, will satisfy the nutrient requirements of animals in both seasons. There was a marked decrease in Ca concentration in leaves of all the species from 2003 to 2004 (Table 3.13). However, *I. amorphoides* had the highest Ca concentration in both 2003 and 2004. As shown in Table 3.13, there was a marked increase in Ca concentration of the edible component of *I. amorphoides* during spring. The P concentration in leaves of *I. cryptantha* and *I. viciodes* during 2003 and 2004 appeared to have higher values than the other species (Table 3.15).

4.3.2.2 Micro elements

The micro-element concentrations in all the species will fulfill the micro-mineral requirements of sheep. During autumn of 2003, the Cu concentration in leaves of *I. viciodes* appeared to be higher than the other species. There was, however, a decrease in Cu concentrations in autumn of 2004 in all the species. During the autumn and spring of 2004 Cu concentration in the edible component did not differ significantly within and between the species (Table 3.18).

The Zn concentration in *I. cryptantha* was consistently the highest for both leaves and edible components (Table 3.20). Unlike the Cu and Zn concentrations, the Mn concentrations showed an increase with age in the autumn of 2004 (leaves) and the spring of 2004 (edible components) in this study. During 2003 (leaves), *I. arrecta* had the highest Mn concentration value, whereas *I. amorphoides* proved to be good forage. In the autumn and spring of 2004, the edible component of *I. arrecta* appeared to have the highest Mn concentrations (Table 3.21).

4.4 Feed intake and digestibility

Lucerne had significantly higher OMI and DOMI than *Indigofera* species and *L. leucocephala*. However, *L. leucocephala* appeared to be slightly better than *Indigofera* species though there were significant differences (Table 3.23). *Indigofera* species was found to have a higher NDFI than lucerne or *L. leucocephala*, while, lucerne did not differ significantly from *L. leucocephala*. Lucerne appeared to be higher in OMD than both *L. leucocephala* and *Indigofera* species, with no significant differences being found between lucerne and *Indigofera* species. However, *Indigofera* species had a higher NDFD than the other forages.

CHAPTER 5

Summary, Conclusions and Recommendations

5.1 Summary and conclusions

The following parameters were employed in this study: DM yield, leaf:stem ratio, chemical composition of the different *Indigofera* species (ash, CP, NDF, IVDOM and minerals), as well as voluntary intake and digestibility in comparison with *Medicago sativa* and *Leucaena leucocephala*. The highest dry matter yields were obtained in the autumn of 2004 from *I. amorphoides*. The leaves as well as the edible components were harvested over two years in different seasons. The lower leaf:stem ratio observed in autumn compared to spring is reflected in a decline in ash, CP, IVDOM and an increase in NDF concentration. It has been repeatedly emphasized in the literature that forage quality is affected by a decrease in the proportion of leaves and an increase in the proportion of stemmy fractions.

It was noted in this study, that there was a decline in chemical composition with advancing maturity and with an increase in the proportion of stem. It was stated in the literature that ageing of the plant has a negative effect on the nutritive value of forages. Despite the decrease in leaf:stem ratio with advancing maturity, *Indigofera* species maintained a fairly high forage quality. This is supported by the fact that all the species investigated in this study were above the minimum requirements of CP concentration (8%) as reported by Leng (1997).

Despite the high NDF concentration of all the species in the edible components during spring, *Indigofera* species could not be regarded as a poor feed. This is mainly because of the relatively low NDF concentration recorded in autumn of 2004 in the leaves and spring of 2004 in the edible component. Hoffman *et al.* (2001) reported that forages, which contain 40% NDF or less are generally of good quality. In spite of the advancing maturity of the plants and a decrease in leaf:stem ratio, the IVDOM of all the species in this study fall within the general range of tropical browse plants as noted in the literature.

The concentration of minerals (macro-elements and micro-elements) for all the species, for leaves as well as for the edible components, indicated levels that are adequate for ruminant feeding requirements and also proves *Indigofera* species to be a relatively good quality forage. This had been noted from the literature.

The OMI obtained in this study for *L. leucocephala* and *Indigofera* species appeared to be lower than that of lucerne, most probably due to the relatively higher NDF concentration. This is supported by many references from the literature, which reported that intake declines with an increasing NDF concentration.

The DOMI for the *Indigofera* species and *L. leucocephala* was lower that of lucerne, as a result of an increased NDF concentration. As a result, the intake of *Indigofera* species and *L. leucocephala* were below the minimum maintenance requirements of 33.5 g DOMI/kg W^{0.75}/d for grazing sheep as reported by Engels (1972). However, lucerne and *L. leucocephala* will supply the maintenance requirements of stall fed sheep. This is supported by Nsahlai *et al.* (1997) who reported that the DOMI requirements for stall fed animals are 28.2 g DOMI/kg W^{0.75}/d.

Dado and Allen (1996) reported that NDF concentration is a good indicator for organic matter digestibility. This suggests that the relatively higher NDF concentration obtained in this study of *Indigofera* species and *L. leucocephala* is probably the reason for a lower OMD compared to that of lucerne, which had a lower NDF concentration.

The relatively lower NDF digestibility of *L. leucocephala* compared to that of lucerne found in this study is positively related to the lower intake. This is supported by Oba and Allen (1999) who stated that NDF digestibility is an important factor affecting feed intake in livestock. Based on the facts that have been presented, and despite the lower DOMI required for maintenance, it can be concluded that *Indigofera* species produce a fairly good quality forage, which can be used by farmers for feeding animals during drought seasons. This could also minimize the purchase of protein supplements.

5.2 Recommendations

The following recommendations are made, based on the results obtained in this study:

- 1. It was noted in this study that the chemical composition of different *Indigofera* species deteriorated with advancing maturity of the plant. Therefore, it is recommended that the productive value of *Indigofera* species as feed for sheep will be improved through proper management such as utilization of the herbage whilst is still immature (harvesting before it matures). This will increase the leaf: stem ratio since it was the reason for the decline in chemical composition.
- 2. Reid *et al.* (1988) reported that acid detergent fibre (ADF) is the best indicator of organic matter intake. It is, therefore, recommended that the analysis of ADF be considered in future.
- 3. It was reported from the literature that *Indigofera* species often contain toxic Indospicine. The effect of Indospicine should be examined, to determine whether it has any effect on intake and the performance of livestock. All the *Indigofera* species in this study were found to have relatively high CP concentrations. Therefore it is also recommended that the production potential of different *Indigofera* species should be evaluated using criteria such as wool growth and quality, weight gain and milk production.

REFERENCES

AGANGA, A.A. & AUTLWETSE, M.N., 2000. Utilization of sorghum forage, millet forage, veldt grass and buffel grass by Tswana sheep and goats when fed *Lablab* purpureus as protein supplement. *Asian-Aust. J. Anim. Sci.* 13: 1035-1188.

AGARWAL, S.K. & RASTOGI, R.P., 1974. Triterpenoid saponins and their genius. *Phyto-Chem.*, 13: 2623-2645.

AHN, J.H., ROBERTSON, B.M., ELLIOT, R., GUTTERIDGE, R.C. & FORD, C.W., 1989. Quality assessment of tropical browse legumes: Tannin and protein degradation. *Anim. Feed Sci. Technol.* 27: 147-156.

AKIN, D.E., ROBINSON, E.L., BARTON, F.E. & HIMMELSBACH, D.S., 1977. Changes with maturity in anatomy, histochemistry, chemistry and tissue digestibility of bermudagrass plant parts. *J. Agric. Food Chem.*, 25: 179-186.

ALLEN, M. & OBA, M., 1996. Fiber digestibility of forages. 7th Minnesota Nutrition Conference Protiva Tech. Symp. Ext. Special Programs, Bloomington, MN. Univ. Minnesota, St Paul. pp. 151-171.

ANDREWS, F.W., 1952. The flowering plants of the Anglo-Egyptian Sudan. J. Bungel and Co., Scotland.

A.O.A.C., 1990. Official methods of analyses (15th ed.). Association of Official Analytical Chemist., Virginia: U.S.A.

BALLARD, R.A., SIMPSON, R.J. & PEARCE, G.R., 1990. Losses of the digestible components of annual ryegrass (*Lolium rigidium* Gaudin) during senescence. *Aust. J. Agric. Res.* 41: 719-731.

BAMUALIM, A., JONES, R.J. & MURRAY, R.M., 1980. Nutritive value of tropical browse legumes in the dry season. *Proc. Aust. Soc. Anim. Prod.* 13: 229-232.

BAMUALIM, A., 1981. Nutritive value of some tropical browse species in the wet and dry season. M.Sc. dissertation. James Cook. University of Queensland. No. 167.

BARRY, T.N. & BLANEY, B.J., 1987. Secondary compounds of forage. Eds. Hacker, J.B. & Ternouth, J.H., Nutrition of Herbivores, Academic Press, Australia, pp. 91-119.

BARTHA, R., 1970. Fodder plants of the Sahel Zone of Africa, Weltforum Verlag, Munchen.

BERG, C.C. & HILL, R.R., 1989. Maturity effect on yield and quality of spring harvested orchard grass forage. *Crop Sci.* 29: 944-948.

BLAXTER, K.L., WAINMAN, F.W. & WILSON, R.S., 1961. The regulation of food intake by sheep. *Anim. Prod.* 3: 51-61.

BRANSBY, D.I., 1989. Composition in the design and conduct of grazing experiments. In: Grazing research: design, methodology and analysis. CSSA special publication no.16.

BREWBAKER, J.L., HEDGE, N., HUTTON, E.N., JONES, R.J., LOWRY, J.B., MOOG, F. & VAN DEN BELDT, R., 1985. Leucaena – Forage Production and Use. NFTA, Hawaii. pp. 39.

BREWBAKER, J.L., 1986. Nitrogen fixing trees for fodder and browse in Africa. In: Alley farming in humid and sub-humid tropics. Eds. Kang, B.T. & Raymonds, L. Proceedings of a workshop held at Ibadan, Nigeria, 10-14 March 1986. IDRC, Ottawa, pp. 55-70.

BULO, D., WARREN, B.E. & IVORY, D.A., 1985. Nutritive value assessment of grass and legume species Balai Penelitian Ternak, Ciawi, Indonesia. Annual Report- Forage Research Project. pp. 40-41.

BUXTON, D.R., MERTENS, D.R. & MOORE, K.J., 1995. Forage quality for ruminants: Plant and animal considerations. *Prod. Anim. Sci.* 11: 121.

CAMPLING, R.C. & LEAN, I.J., 1983. Food characteristics that limit voluntary intake. In: Nutritional Physiology of Farm Animals. Eds. Rook, J.A.F. & Thomas, P.C., Longman, London.

CATCHPOOLE, V.R. & HENZELL, E.F., 1971. Silage and silage making from tropical herbage species, *Herbage Abstr.* 41: 213-221.

CHEEKE, P.R., 1995. Biological effects of feed and forage saponins and their impact on animal production. Symposium on saponins: Chemistry and biological activity, *American Chemical Society*, Chicago, August 22-25.

CHESWORTH, J., 1992. The tropical agriculturalist. CTA. Wageningen. The Netherlands. pp.170.

CHURCH, D.C., 1980. Digestive physiology and nutrition of ruminants. Vol. 3, Practical nutrition. 2nd edition Prentice- Hall, New Jersey.

COLEMAN, S.W., LIPPKE, H.G. & GILL, M., 1990. Estimating the nutritive potential of forages. In: Nutritional Ecology of Herbivores. Eds. Jung, H.G. & Fahey, G.C., *Proc. 5th Int. Symp. Nutr. Herb. American Soc. Anim. Sci*, Illinois, pp. 647-695.

CROWDER, L.V. & CHHEDA, H.R., 1982. Tropical grassland husbandry. Longman; London and New York.

DADO, R.G. & ALLEN, M.S., 1996. Enhanced intake and production of cows offered ensiled alfalfa with higher digestibility. *J. Dairy Sci.* 79: 418-428.

DEVENDRA, C., 1988. Forage supplements: Nutritional significance and utilization for draught, meat and milk production in buffaloes. Proceedings Second World Buffalo Congress, Indian Council of Agricultural Research, New Delhi, Vol. 2 pp. 409-423.

DEVENDRA, C., 1989. The nutrition of and feeding strategies for sheep in Asia: In: *Proc. Sheep Prod.* in *Asia.* Philippines Council for Agriculture Research and Development Resource. Eds. Devendra, C. & Faylon, P.S. Book Series No. 80, pp. 21-42.

DIAS FILHO, M.B., CRSI, M., CUSSATO, S. & PINHEIRO C., 1991. *In vitro* organic matter digestibility and crude protein content of *Panicum maximum* jacq. cv. *tobiata* under water stress. *Pesqui. Agropecu. Bras.* 26(10): 1725-1729.

D'MELLO, J.P.F. & ACAMOVIC, T., 1989. *Leucaena leucocephala* in poultry nutrition. *Anim. Feed Sci. Technol.* 26: 1-28.

DOUGALL, H.W, DRYSDALE, V.M. & GLOVER, P.E., 1964. The chemical composition of Kenya browse and pasture forage. *E. Afr. Wildlife J.* 2: 85-125.

DOUGALL, H.W. & BODGAN, A.V., 1958. The chemical composition of the grasses of Kenya. *E. Afr. Agric. J.* 25: 17-23.

DOUGALL, H.W. & BODGAN, A.V., 1966. The chemical composition of some leguminous plants grown in the herbage nursery at Kitale, Kenya, *E. Afr. Agric. J.* 32. No.1: 45-49.

DUKE, J.A., 1977. Phytotoxin tables. CRC Critical Reviews in Toxicology 5: 189-237.

DUKE, J.A., 1983. Handbook of Energy Crops. httpwww.hort.purdue.edu/newcrop/duke-energy/medicago-sativa. Html#. cultivation.

EAGAN, A.R., WANAPAT, M., DOYLE, P.T., DIXON, R.M. & PEARCE, G.R., 1986. Production limitation of intake, digestibility and of passage. In: Forage in Southeast Asia and South Pacific Agriculture. Eds. Blair, G.J., Ivory, D.A. & Evans, T.R., ACIAR Proceedings No.12 Canberra, pp. 104-110.

ENGELS, E.A.N. & VAN DER MERWE, F.J., 1967. Application of an *in vitro* technique to South African forages with special reference to the effect to certain factors on the result. *S. Afr. J. Agric. Sci.* 10, 983.

ENGELS, E.A.N., 1972. A study of the nutritive value of natural and sown pastures in the Central Orange Free State with special reference to the energy requirements of sheep. PhD. Thesis, University of Stellenbosch.

EVANS, D.O. & ROTAR, P.P., 1987. Productivity of *Sesbania* species. *Trop. Agric*. (Trinidad) 64: 193-200.

EVANS, T.R. & WILSON, J.R., 1984. Some responses of grasses to water stress and their implications for herbage quality and animal liveweight gain. In: The Impact of Climate on Grasses Production and Quality. Eds. H. Riley & A.O. Skjelvag, Norwegian State *Agric Res*. St. As, Norway.

EVANS, D.O., 2002. Sustainable Agriculture in Hawaii. Green Manure: Legume. Lablab. http://www.2.ctahr.Hawaii.edu/sustainag/sustainableAg/Greenmanure/lablab.asp.

EVERIST, S.L., 1969. Use of fodder trees and shrubs. Queensland Department of Primary Industry, Advisory leaflet: No. 1024.

FACHNEY, G.J., 1992. Application of the double-marker method for measuring digesta kinetics to rumen sampling in sheep following a dose of the markers or the end of their continous infusion. *Aust. J. Agric. Res.* 28: 1055.

FORBES, J.M., 1986. The voluntary food intake of farm animals. Butterworths, London.

FORBES, J.M., 1995. Voluntary food intake and diet selection in farm animals. CAB International, Wallingford, UK.

FORD, C.W., MORRISON, I.M. & WILSON, J.R., 1979. Temperature effects on lignin, hemicellulose and cellulose in tropical and temperate grasses. *Aust. J. Agric. Res.* 47: 453-464.

FORWOOD, J.R., MATCHES, A.G. & NELSON, C.J., 1988. Forage yield, non-structural carbohydrates levels, and quality trends of Caucasian Bluestem. *Agron. J.* 80: 135-139.

FOX, D.G., SNIFFEN, C.J., O'CONNOR, J.D., RUSSELL, J.B. & VAN SOEST, P.J., 1990. The Cornell net carbohydrates and protein system for evaluating cattle diets. Search Agriculture. Cornell University. *Agric. Exp.* Stn. No. 34. Ithaca. N.Y.

GARTNER, R.J.W. & HURWOOD, I.S., 1976. The tannin and oxalic contents of *Acacia aneura* and their possible effects on sulphur and calcium availability. *Aust. Vet. J.* 52: 194-196.

GILL, M. & ROMNEY, D., 1994. The relationship between the control of meal size and the control of daily intake in ruminants. *Livestock Prod. Sci.* 39: 13-18.

GOODCHILD, A.V., 1990. Use of leguminous foliage to supplement low quality roughage for ruminants. PhD thesis, The University of Queensland.

GUPTA, P.C. & PRADHAN, K., 1975. A note on the comparative nutritive value of legume and non-legume forage. *Indian J. Anim. Sci.* 45: 290-291.

GUTTERIDGE, R.C. & SHELTON, H.M., 1994. Forage Tree legume in tropical agriculture. CAB. International. Wallingford. Oxon, UK.

HAAFAT, M.A. & HASSANI, M., 1966. Paper presented to the Arab Scientific Congress, Baghdad.

HAUDE, M.E., 1997. Identification and classification of colorants used during Mexico's early Colonial period. Book and Paper Group Annual Vol. 16. The American Institute of Conservation, pp 16-05.

HEANEY, D.P., PRITCHARD, G.I. & PIGDEN, W.J., 1968. Variability in *ad libitum* forage intake by sheep. *J. Anim. Sci.* 27: 159-164.

HEGARTY, M.P. & POUND, A.W., 1968. Indospicine, a new hepatotoxic amino acid from *Indigofera spicata*. *Nature*, 217: 354-355.

HEGARTY, M.P., SCHINCKEL, P.G. & COURT, R.D., 1964. Reaction of sheep to the consumption of *Leucaena glauca* and to its toxic principle Mimosine. *Aust. J. Agric. Res.* 15: 135-167.

HENZELL, E.F., 1962. Nitrogen fixation and transfer by some tropical and temperate pastures legumes. *Aust. J. Exp. Agric. Anim. Husb.* 2: 132-140.

HILL, G.D., 1971. *Leucaena leucocephala* for pastures in the tropics, *Herbage Abstr.* 41: 111-119.

HIREMATH, N.B., 1981. Subabool (*Leucaena leucocephala*)- A wonder plant. *Indian Dairyman*, 33: 351-356.

HOFFMAN, P.C., SHAVER, R.D., COMBS, D.K, UNDERSANDER, D.J., BAUMAN, L.M. & SEEGER, T.K., 2001. Focus on forage. Vol. 3: No. 10.

HUTTON, E.M., 1970. Tropical pastures. Adv. Agron. 22: 1-73.

IBRAHIM, K.M., 1981. Shrubs for fodder production. In: Advances in Food Producing System for Arid and Semi-arid Lands. Academic Press Inc. pp.601-642.

ILLIUS, A.W. & ALLEN, M.S., 1994. Assessing forage quality using integrated models of intake and digestion by ruminants. In: Forage Quality, Evaluation and Utilization. Eds. Fahey, G.C., Collins, M., Mertens, D.R. & Moser, L.E., *Am. Soc. of Agron., Crop Sci. Soc. of Am., Soil Sci. Soc. of Am.*, Madison, Wisconsin, pp. 869-890.

ILLIUS, A.W., 1998. Advalues and retreats in specifying the constraints on intake in grazing ruminants. In: *Proc.* of XVIII Int. *Grassl Congress*. Vol. III. Eds. Buchanan-Smith, J.G., Bailey & McCoughy Association Management Centre Calgary. pp.39-44.

ISARASENEE, A, SHELTON, H.M. & JONES, R.M., 1984. Accumulation of edible forage of *Leucaena leucocephala* cv. Peru over late summer and autumn for use as dry season feed. Leucaena Research Report 5, 3-4.

JONES, R.J., 1969. A note on the *in vitro* digestibility of two tropical legumes-Phaseolus atro-purpureus and Desmodium intortum. J. Aust. Int. Agric. Sci. 35:62.

JONES, R.J., 1979. The value of *Leucaena leucocephala* as a feed for ruminants in the tropics. *World Anim. Review* 31: 13-23.

JOYCE, J.P., BRUNSWICK, L. & PARKER, J., 1973. Feeding value of lucerne. *Proc. N.Z Soc. Anim. Prod.* 32: 54.

JUNG, H.G. & ALLEN, M.S., 1995. Characteristics of plant cell walls affecting intake and digestibility of forage by ruminants. *J. Anim. Sci.* 73: 2774-2790.

KABAIJA, E. & SMITH, O.B., 1989. Influence of season and age of regrowth on the mineral profile of *Gliricia sepium* and *Leucaena leucocephala*. *Trop. Agric*. 66: 125-128.

KARACHI, M., 1997. Growth and nutritive value of *Lablab purpureas* accessions in semi-arid Kenya. *Trop. Grassl.* 31: 214-218.

KHAMSEEKHIEW, B., LIANG, J.B., WONG, C.C. & JALAN, Z.A., 2001. Ruminal and intestinal digestibility of some tropical legume forages. *Asian-Aust. J. Anim. Sci.* 14: 321-325.

KRUGER, A.J., 1991. *Leucaena leucocephala* and *Desmanthus virgatus*: Useful species for the production of forage protein by subsistence farmers. In: *African Agroforestry*: Emphasis on Southern Africa. Ed. Koen J.H., Environmental forum report of Forest Biome Group. pp. 57-68.

LAREDO, M.A. & MINSON, D.J., 1973. The voluntary intake, digestibility and retention time by sheep of leaf and stem fractions of five grasses. *Aust. J. Agric. Res.* 24: 875-888.

LENG, R.A., 1986. Determining the nutritive value of forage. ACIAR Proceedings No. 12, Canberra, pp. 111-123.

LENG, R.A., 1990. Factors affecting the utilization of "poor quality" forage by ruminants particularly under tropical conditions. *Nutr. Res. Rev.* 3: 277-303.

LENG, R.A., 1997. Tree foliage in ruminant nutrition. Animal Production and Health paper No. 139, FAO, Rome.

LIOGIER, H.A., 1990. Plantes Medicinales de Puerto Reis Caribe. Inc San Juan, PR. pp. 566.

LOWRY, J.B., MARYANTO, N. & TANGENDJAJA, B., 1983. Antolysis of Mimosine to 3-hydroxy-4 (1H) Pyridone in green tissue of *Leucaena leucocephala*. *J. feed Sci. Agric*. 34: 529-533.

LOWRY, B., 1987. The leucaena story- a biological solution to a toxicity problem- An account of one man's belief. *Aust. Sci. Magazine* 4: 53-58.

LUKHELE, M.S. & VAN RYSSEN, J.B.J., 2002. The chemical composition and potential nutritive value of the foliage of four subtropical tree species in Southern Africa for ruminants. *S.A. J. Anim. Sci.* 33. No. 2 pp.132-141.

MacPHERSON, A., 2000. Trace-minerals status of forages. In: Forage Evaluation in Ruminant Nutrition. Eds. By Givens, D.I., Owen, E., Oxford, R.F.E. & Omed, H.M. CAB International, pp. 345-371.

MARTEN, G.C., BUXTON, D.R. & BARNES, R.F., 1988. Feeding value (Forage Quality). In: Alfalfa and Alfalfa Improvement. Monograph No. 29. *Am. Soc. Agron.*, Madison, WI 53711.

McDONALD, W.J.F. & TERNOUTH, J.H., 1979. Laboratory analysis of the nutritive value of western Queensland browse feeds. *Aust. J. Husb.* 18: 344.

McDONALD, P., EDWARDS, R.A. & GREENNAGH, J.F.D., 2002. Animal Nutrition. 6th ed. Longman, New York.

McDONALD, R.C. & WILSON, K.R., 1980. Dry matter yield, digestibilities, mineral levels and cattle growth rates on green fed oats at different stages of development. *N. Z J. Exp. Agric.* 8: 105-109.

McDOWELL, L.R., 1985. Nutrition of grazing ruminants in warm climates. Academic Press, San Diego, pp. 524.

McDOWELL, L.R., 1992. Minerals in animal and human nutrition. Academic Press. San Diego, pp. 524.

McDOWELL, L.R., 1997. Minerals for grazing ruminants in tropical regions, 3rd edn. University of Florida, Gainevine, Florida, USA, pp. 81.

McLEOD, M.N. & MINSON, D.J., 1978. The accuracy of the pepsin-cellulose technique for estimating the dry matter digestibility *in vivo* of grasses and legumes. *Anim. Feed Sci. Tech*nol. 3: 277-287.

McMANUS, J., DAVIS, K. BEARS, J., GEOFFRY, S., LILLEY, T. & HASLAM, E., 1985. Polyphenol interactions. Part 1. Introduction; some observation on the reversible complexation of Polyphenol with proteins and polysaccharides. *J. Chem. Soc.* Perkin Transcript II, 1429-1437.

McMENIMAN, N.P. & LITTLE, D.A., 1974. Studies on the supplementary feeding of sheep consuming mulga (*Acacia aneura*) 1. The provision of phosphorus and molasses supplements under grazing conditions. *Aust. J. Exp. Agric. Anim. Husb.* 14:316-321.

McNEILL, D.M., OSBORNE, N., KOMOLONG, M. & NANKERVIS, D., 1998. Condensed tannins in the genus *Leucaena* and their nutritional significance. In: Leucaena – Adaptation, Quality and Farming System. Eds. Shelton, H.M., Gutteridge, R.C., Mullen, B.F. & Brey, R.A., Proceedings of Workshop held in Hanoi, Vietnam. ACIAR Proceedings No. 86, pp. 205-214.

MEISSNER, H.H., VAN NIEKERK, W.A., SPREETH, E.B. & KOSTER, H.H., 1989. Voluntary intake of several planted pastures by sheep and an assessment of NDF and IVDOM as possible predictors of intake. *J.Grassl. Soc.S.A.* 6: 156.

MEISSNER, H.H., KOSTER, H.H., NIEWOUDT, S.H. & COERTZE, R.J., 1991. Effect of energy supplementation on intake and digestion of early and mid-season rye grass and Panicum/Smuts finger hay and on *in sacco* disappearance of various forage species. *S. Afr. J. Anim. Sci.* 21: 33.

MEISSNER, H.H., SMUTS, M. & COERTZE, R.J., 1995. Characterization and efficiency of fast growing feedlot steers fed different dietary energy concentrations. *J. Anim. Sci.* 73:931.

MERTENS, D.R., 1992. Non-structural and structural carbohydrates. Large Dairy Herd Management. ADSA, Champaign, I.L 9Van horn and Wilcox, (ed).

METSON, A.J., 1978. Seasonal variation in chemical composition of pasture. I. Calcium, magnesium, potassium, sodium and phosphorus. *J. Agric.Res.* 21: 341-353.

MILFORD, R. & MINSON, D.J., 1966. The feeding value of tropical pastures. In: Tropical Pastures. Ed by: Davies, W. and Skidmore, C.L. Faber and Faber, London.

MILFORD, R. & MINSON, D.J., 1968. The effect of age and method of hay making on the digestibility and voluntary intake in forage legumes. In: Tropical Forage Legumes. Eds. Skerman, P.J., Cameroon, D.G. & Riveros, F., FAO plant production and protection series, No.2 Rome, pp. 185-193.

MINSON, D.J., 1990. Forage in ruminant nutrition. Academic Press California.

MOORE, K.J. & H.G. JUNG., 2001. Lignin and fibre digestion. *J. Range Man.* 54:420-430.

MURPHY, A.M & COLUCCI, P.E., 1999, A tropical forage solution to poor quality ruminant diets: A review of *Lablab purpureus*. *Livestock Res. Rural Dev.* (11) 2.

NOCEK, J.E. & RUSSELL, J.B., 1988. Protein and energy as an integrated system. Relationship of ruminal protein and carbohydrate availability to microbial synthesis and milk production. *J. Dairy Sci.* 71: 2070-2107.

NORIS, K.H., BARNES, R.F., MOORE, J.E. & SHENK, J.S., 1976. Predicting forage quality by infrared reflectance spectroscopy. *J. Anim. Sci.* 43: 889-897.

NORTON, B.W., 1994. Anti-nutritive and toxic factors in forage tree legumes. In: Forage Tree Legumes in Tropical Agriculture Eds: R.C Gutteridge & H.M Shelton. CAB International; Oxford, UK. Pp. 202-215

NORTON, B.W., KAMAU, F.K. & ROSEVEAR, R., 1992. The nutritional value of some tree legumes as supplements and sole feed for goats. *Proc. of the sixth A.A.A.P Anim. Sci. Cong*, 23-28 November 1992. Volume III, pp. 151.

NRC., 1985. Nutrients requirements of sheep, 6th edn. National Academy Press. Washington, D.C., USA.

NRC., 1996. Nutrient requirements of beef cattle (7th ed). National Academy Press, Washington DC., USA.

NRC., 2001. Nutrient requirements of dairy cattle. 7th rev. ed. Natl. Acad. Sci., Washington D.C., USA.

NSAHLAI, I.V., KHAITHO, R.J. & WILLIAMS, B.A., 1997. Relationships between preference, rumen degradability, gas production and chemical composition of browsers. *Agrofestry Sysytems*. 39: 129-144.

OAKS, A.J., 1968. *Leucaena leucocephala*-description, culture, utilization, *Adv. Front Pl. Sci.* 20: 1-114.

OBA, M. & ALLAN, M.S, 1999. Effects of brown midrib 3 mutation in corn silage on dry matter intake and productivity of high yielding dairy cows. J. Dairy Sci. 82 (in press). ØRSKOV, E.R. & McDONALD.I, 1979. The estimation of protein degradation in the rumen from the incubation measurement weighted according to rate of passage. *J. Agric. Sci.*, Cambridge, 92: 499-503.

PARSONS, A.J., JOHNSON, I.R. & HARVEY, A., 1988. Use of model to optimize the interaction between frequency and severity of intermittent defoliation and to provide a fundamental comparison of the continuous and intermittent defoliation of grass. *Grassl. Forage Sci.* 43: 49-60.

PARSONS, A.J. & PENNING, P.D., 1988. The effect of the duration of regrowth on photosynthesis, leaf death and the average rate of growth in a rotationally grazed sward. *Grass Forage Sci.* 43: 15-27.

PHILLIPS, W.A., RAO, S.C., FITCH, J.Q. & MAYAIX, H.S., 2002. Digestibility and dry matter intake containing alfalfa and kenaf. *J. Anim. Sci.*, 80: 2989-2995.

PLUCKNETT, D.C., 1970. Productivity of tropical pastures in Hawaii, *Proc.* 11th Int. Grassld Congr., Surfers Paradise, 1970, A38-A49.

POSSINGHAM, J.V., GROOT OBBINK, J. & JONES, R.K., 1971. Tropical legumes and vesicular arbuscular mycorrhiza. *J. Aust. Inst. Agric. Sci.*. 37: No.2, 160-161.

PRICHARD, D.A., STOCKS, D.C., O'SULLIVAN, B.M., MARTIN, P.R., HURWOOD, I.S. & O'ROURKE, P.K., 1988. The effect of Polyethylene glycol (PEG) on wool growth and live weight of sheep consuming a Mulga diet. *Proc. Aust. Soc. Anim. Prod.* 17: 290-293.

PROVENZA, F.D., BURRIT, E.A., CLAUSEN, T.P., BRYANT, J.P., REICHARDT, P.B. & DISTEL, R.A., 1990. Conditional flavor aversion: A mechanism for goats to avoid condensed tannins in blackbush. *American Nature*. 136: 810-828.

REED, J.D., SOLLER, H. & WOODWARD, A., 1990. Fodder tree and straw diets for sheep: intake, growth, digestibility and the effect Phenolics on nitrogen utilization. *Anim. Feed Sci. Technol.* 30: 39-50.

REED, J.D., 1995. Nutritional toxicology of tannins and related Polyphenol in forage legumes. Pharmacology/Toxicology Symposium on toxic legumes. *J. Anim. Sci.* 73: 1516-1528.

REED, J.D., KRUEGER, C., RODRIGUEZ, G. & HANSON, J., 2000. Secondary plant compounds and forage evaluation. In: Forage Evaluation in Ruminant Nutrition. Eds. Givens, D.I., Owen, E., Oxford, R.F.E. & Omed, H.M., CAB International, pp. 433-448.

REID, R.L., JUNG, G.A. & THAYNE, W.V., 1988. Relationship between nutritive quality and fibre components of cool season and warm season forages: Retrospective study. *J. Anim. Sci.* 66:1275-1291.

REIS, P.J., TUNKS, D.A. & HEGARTY, M.P., 1975. Fate of mimosine administered orally to sheep and its effectiveness as a defleecing agent. *Aust. J. Bio. Sci.* 28: 495-501.

RELLING, E.A., VAN NIEKERK, W.A., COERTZE, R.J. & RETHMAN, N.F.G., 2001. An evaluation of *Panicum maximum* cv. Gatton: 2. The influence of stage and maturity on diet selection, intake and rumen fermentation in sheep. *S. Afr. J. Anim. Sci.* 31: 85-91.

RITTNER, U. & REED, J.D., 1992. Phenolics and *in vitro* degradability of protein and fibre in West African browse. *J. Sci. Food Agric*. 21: 28.

ROBERTSON, J.B. & VAN SOEST, P.J., 1981. The detergent system of analysis and its application to human foods. In: The Analysis of Dietary Fibre in Food. Eds. James, W.P.T. & Theander, O., Marcel Dekker, New York. pp. 123-158.

ROBERTSON, B.M., 1988. The nutritive value of five browse legumes fed as supplements to goats offered a basal rise straw diet. Master of Agricultural Studies Thesis. The University of Queensland.

ROBINSON, P.J., 1985. Trees and Fodder crops. Institute of Terrestrial Ecology, Huntington, U.K.

ROBBINS, C.T., 1993. Wildlife feeding and nutrition. 2nd ed. Academic Press, New York.

ROUX, C.Z. & MEISSNER, H.H., 1984. Growth and feed intake patterns. The derived theory. In: Herbivores Nutrition in the Subtropics and Tropics. Eds. F.M.C. Gilchrist & R.I. Mackie. The Science Press, Graighall, pp. 67.

RUIZ, T.M., BERNAL, E., STAPLES, C.R. SOLLENBERGER, L.E. & GALLER, R.N., 1995. Effect of dietery neutral detergent fiber concentration and forage source on performance of lactating cows. *J. Dairy Sci.* 78:305.

SAMUEL, M.L., 1989. Statistics for the life sciences. Collier MacMillan Publishers, London, UK.

SAS., 2001. Statistical Analysis System User's Guide: (Statistics Version 8) SAS Institute Inc, Cary, North Carolina, USA.

SCA, 1990. Feeding standards for Australian Livestock Ruminants. Standing Committee on Agriculture and CSIRO, Melbourne, pp. 266.

SHEHU, Y., ALHASSAN, W.S., PAL., U.R. & PHILLIPS, C.J.C., 2001. Yield and chemical composition response of *Lablab purpureus* to nitrogen, phosphorus and potassium fertilizers. *Trop. Grassl*, 35: 180-185.

SKERMAN, P.J., 1970. *Stylosanthes mucronata* Willd., an important natural perennial legume in Eastern Africa. *Proc. XI Int. Grassl. Congr.* 196-198.

SKERMAN, P.J., 1977. Tropical forage legumes. FAO Plant Production and Protection. Rome.

SMITH, F.W., VAN DEN BERG, P.J., GONZALEZ, A., ANDREW, C.S. & PLATERS, W.H.J., 1992. Foliar symptoms of nutrient disorders in the tropical shrub legume *Leucaena leucocephala*. Division of Tropical Crops and Pastures. Technical Paper No. 43, CSIRO, Australia.

TABE., L.M., HIGGINS, C.M., MACNABB, W.C. & HIGGINS, T.J.V., 1993. Genetic engineering for grain and nutritive value. *Genetica*, 90: 181-200.

TANNER, J.C., REED, J.D. & OWEN, E., 1990. The nutritive value of fruits (pods with seeds) from four *Acacia* spp. Compared with extracted noug (*Guizotia abyssinica*) meal as supplements to maize stover for Ethiopian Highland sheep. *Anim. Prod.* 51: 127-133.

THOMAS, C. & THOMAS, P.C., 1985. Factors affecting the nutritive value of grass silages. Chapter 13. In Recent Advances in Animal Nutrition. Haresign. Butterworths, London.

THOMPSON, F.N. & STUEDEMANN, J.A., 1993. Pathophysiology of fescue toxicosis. Agriculture Ecosystem and the Environment, 44: 263-281.

THORTON, R.F. & MINSON, D.J., 1973. The relationship between apparent retention time in the rumen, voluntary intake and apparent digestibility of legume grass diets in sheep. *Aust. J. Agric. Res.* 24, 889-898.

TILLEY, J.M.A. & TERRY, R.A., 1963 A stage technique for *in vitro* digestion of forage crops. *J. British Grassl. Soc.*, 18: 104-111.

TUDSRI, S., ISHII, Y., NUMAGUCHI, H. & PRASANPANICH, S., 2002. The effect of cutting interval on the growth of *Leucaena leucocephala* and three associated grasses in Thailand. *Trop. Grassl.*, 36: 90-96.

VAN RENSBURG, H.J., 1968. Palatability of plants in Zambia. Govt. Printer. Lusaka.

VAN SOEST, P.J., 1982. Nutritional ecology of the ruminant. Chapter 13. O and B Books, Inc. Corvallis, O.R.

VAN SOEST, P.J., 1987. Practical aspects of forage quality. Page 90 in Proc. Southwest Nutr. Conf. Temp, AZ.

VAN SOEST, P.J., 1994. Nutritional ecology of the ruminant, 2nd ed. Cornell University Press. Ithaca, N.Y, 9.

WEDEKIND, K.J. & BAKER, D.H., 1990. Effect of varying calcium and phosphorus levels on manganese utilization. *Poultry Sci.* 69: 977.

WEIS, W.P., 1993. Symposium: Prevailing concepts in energy utilization by ruminants: Predicting energy values of feeds. *J. Dairy Sci.* 76: 1802-1811.

WILDIN, J.H., 1985. Tree Leucaena-Permanent high quality pastures. Queensland Department of Primary Industries, Rockhampton, pp 8.

WILMAN, D. & ASIEDU, F.H.K., 1983. Growth, nutritive value and selection by sheep of sainfoin, red clover, lucerne and hybrid ryegrass. *J. Agric. Sci.* (Camb). 100:15.

WILSON, A.D., 1977. A review of browse in the nutrition of grazing animals. *J. Range Man.* 22: 23-28.

WILSON, J.R., 1983. Effect of water stress on *in vitro* dry matter digestibility and chemical composition of herbage of tropical pasture specie. *Aust. J. Agric. Res.* 34(4): 377-390.

WOODWARD, A. & REED., J.D., 1995. Intake and digestibility for sheep and goat consuming supplementary, *Acacia brevispica* and *Sesbania sesban*. *Anim. Feed Sci. Technol*.56:207-217.