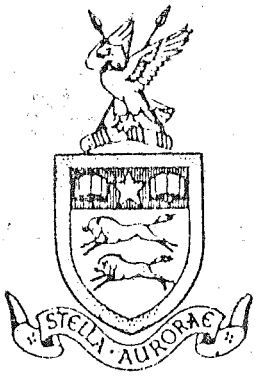

THE SEASONAL PATTERNS IN PLANT QUALITY IN VARIOUS ECOLOGICAL ZONES IN NATAL

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SEPTEMBER 1990

THE SEASONAL PATTERNS IN PLANT QUALITY IN VARIOUS ECOLOGICAL ZONES IN NATAL

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DECLARATION

The work reported in this thesis is the result of the authors original work, unless specifically acknowledged, or stated to the contrary, in the text.

P J K ZACHARIAS (BScAgric) September 1990

ABSTRACT

The objectives of this study required that the following investigations be undertaken:

1) to determine if plant quality can be altered by modifying growing conditions;
2) to quantify the seasonal trends in plant quality from different sites;
3) to relate differences between sites to environmental variables;
4) to develop an objective classification of SWEETNESS; and
5) to plan future research.

The majority of the commercial and subsistence livestock in southern Africa rely almost entirely on veld (rangeland) for their supply of nutrients. These rangelands are traditionally and conventionally managed according to their classification as 'SWEETVELD' and 'SOURVELD'. An intermediate group 'MIXED-VELD' is also recognised. The subjective classification is based on the quality (nutritive value) of the rangeland when it is mature (winter). Both extremes of 'sweet' and 'sour' rangeland contain many of the same species and this thesis considers the relationship between the *soil (chemical and physical)* and the *physical environment* and *plant quality* of a single indicator species *Themeda triandra* (red grass).

A glass house experiment was used to determine the effect of manipulating the soil environment on the quality of *T. triandra*. There were no significant differences ($P>0.05$) between any of the six treatments (combinations of *eutrophic, ameliorated* and *dystrophic* soil together with '*sweet*' and '*sour*' *T. triandra* plants). When compared as a group the SWEET group were significantly ($P\leq0.01$) higher in cellulase dry matter disappearance (CDMD) than the SOUR group. However the difference was only 1.5% CDMD units and is believed to be biologically

unimportant. It was concluded that the quality of *T. triandra* can not be altered by manipulating its growing conditions.

In a field investigation the seasonal pattern and the relationship between environmental variables and plant CDMD, N, P, K, Mg, Ca, S and Zn are described for Natal, South Africa. Most models have significant ($P \leq 0.01$) R^2 values but very few show any strong relationship between soil chemical status and forage quality. ALTITUDE appears most in the models as a related variable. The models would have little predictive value outside Natal and do not contribute or describe adequately the factors determining seasonal patterns in plant quality at different locations.

A multivariate approach is used to provide an objective index of 'SWEETNESS' (based on seasonal variations in plant quality at 31 sites over 23 months), and this is related to environmental variables. This analysis also showed that the soil environment was only weakly related to plant quality. The results are confusing given the wide variations in both plant quality and soil chemistry in the data presented.

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ACKNOWLEDGEMENTS

The work that has resulted in this thesis required a number of field trips and laboratory analyses to be undertaken. The sampling strategy that was used required over 350 000 individual tillers and an additional 9 000 tufts to be harvested, milled or separated. This task would have been impossible without the help of John Clayton (Senior Field Technician, Department of Grassland Science). His cheerfulness during wet, cold, hot and dry field trips ('*Pyotts eating meetings*') made the job enjoyable (even if he beat me to the bakkie ahead of the rhino).

The research programme would not have been undertaken without the co-operation of the Department of Forestry (now disbanded), The Natal Parks Board and the two private land owners, Rob Stock and Andre Nel (Mgudu Farmers). The hospitality we received from Colin and Terry Everson (Cathedral Peak), Jo Venter (previously Natal Parks Board, Hluhulwe), Rob and Andre (Mgudu Farmers) and Rich Davies (previously Natal Parks Board, Mkuze), made each field trip something to look forward to. Arrangements for working at Highmoor were made by Anthony Priday (formerly District Forest Officer, Drakensburg) and at Kranzkloof by Peter le Roux (Natal Parks Board). Both the department of Forestry and Natal Parks Board willingly issued collectors permits allowing me to sample in the areas under their control.

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Staff and students of the Grassland Science Department have contributed in a number of ways. In particular the Post-Graduate

class, spanning six years, have made useful suggestions on the analysis and treatment of these data. Professor Tainton has guided and encouraged that group. As far as my own project was concerned he ensured that I had the facilities and finance to complete the work. His comments on the earlier draft were instructive and have contributed to the current form of the report. As my academic supervisor he encouraged me to develop my own ideas without interference. When I needed comment he was always willing to assist, despite extremely heavy commitments to other post-grads.

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The final preparation of this thesis was carried out whilst I was on sabbatical leave as an Internal Research Fellow at Döhne Research Station, Stutterheim. Mr Frans du Toit (Deputy Director Research) assisted in securing the infrastructural support I needed to finish the task. The staff of the Pastures Section, in particular Jock Danckwerts, Greg Stuart-Hill, Peter O'Reagain and Glen Barnes provided valuable inputs regarding the approach and interpretation of the analyses. Carolyn Barnes (Biometrical Service, Döhne) assisted by improving my computing skills and understanding of some of the analyses. The preparation of the illustrations was made easier by the use of many printing tricks. These were willingly passed onto me by Brian Denyer, Dawn Vockerodt, Naomi Starbuck and Elize Vosloo of Media Services, Döhne.

The completion of this thesis has been protracted for a number of reasons. Whilst the details are not important there were many occasions when I lacked motivation to continue. Without the interest, support and encouragement of my entire family I might

have been tempted to discontinue. In particular, my wife Michèle has been subjected to the brunt of my inevitable periods of despair. The entire preparation of this document was done by her, mostly late at night after her 'family duties' were completed. Her support and willingness to be involved allowed me to make a number of changes to the style and format. I believe a better document has resulted.

Finally my parents have always encouraged and supported my academic pursuits. Without their confidence in my abilities I would not have entered University. Unfortunately my Father did not live to see the completion of this work. His advice and interest have always been of great value to me. He taught me the meaning of doing things to the best of ones ability. ***This thesis is dedicated to his memory.***

It is unlikely that this work is without error in logic and production. Although I have acknowledged the advise and assistance of a number of colleagues, any inaccuracy in the work, as reported, is entirely my responsibility.

P J K ZACHARIAS

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CHAPTER 1

OBJECTIVES AND LITERATURE REVIEW

INTRODUCTION

The South African livestock industry relies heavily on the veld for its supply of forage. Indeed the majority of livestock in this country uses the indigenous vegetation as its sole source of nutrients (Tainton *et al* 1989). It has long been recognised that not all of this vegetation has the same potential to support animals nor do all areas have the same grazing season.

Before the commencement of government sponsored pasture (grassland) research in South Africa, Pole-Evans (1920) suggested that a careful inter-disciplinary enquiry was required to explain why "... *stock fatten more rapidly in the Vaal kameel Dorn* (sic) *Veld in winter than in any other part of South Africa...*". In the same address he discussed the terms 'sweetveld' and 'sourveld' in relation to their geographic and altitudinal separation.

Development of grazing management systems in southern Africa has often been on the basis of the separation of 'sweet' and 'sour' veld, and for many years a system of trek farming was used. This system of land use required an individual grazier to own (control) farms in both the 'sweet' and 'sour' veld. The 'sour' veld is used for the summer grazing, when it is productive and palatable and the 'sweet' veld for the winter. It is highly likely that the other tribes in Africa used this method for their animals prior to settlement. The nomadic peoples of Africa, such as the San, would have followed the game species under a similar system as they moved to more palatable pastures between seasons. The early travellers in southern Africa noted in their diaries that the pastoralists they encountered used the rangeland in this

way (eg Alberti 1807).

Despite the fact that the use of veld in southern Africa has been based on this dichotomy very few detailed studies have been undertaken concerning the circumstances giving rise to this phenomenon, which is not unique and occurs elsewhere in the world (Schofield 1944; Heady 1975; Wilson 1982).

The purpose of this chapter is to define the terms '*sweet*' and '*sour*' veld, discuss previous studies and state the objectives of the study reported in this thesis.

DEFINITIONS

The earliest attempt at a definition of the terms sweet and sour veld was provided by Alberti (1807) who described the phenomenon as follows:

> "*In the Colony one distinguishes between sweet, sour and broken cattle land. The first two kinds, as their names already indicate, consist entirely either of sour or sweet plants, whereas the last contains a happy mixture of both. Sour veld has the draw-back, particularly in the case of cattle, that if they stay there throughout the year, they get so stiff that finally they cannot move at all and then die of hunger. Sweet pasturage causes a wasting disease (consumption) which also leads to death, unless such grazing grounds are exchanged at the appropriate time for sour ones, and such change may be effected regularly. Broken [gravelly loamy soil] pasturage, however, is suitable for cattle throughout the year and in general is best.*"

A more formal definition of the terms 'sweetveld' and 'sourveld' was first only published much later (Scott 1947) but the definitions are subjective and are at best descriptive. In terms of their common usage the following definitions apply:

1) **SOURVELD** - veld which provides palatable material only during the growing season, i.e. is unpalatable when mature;
2) **SWEETVELD** - veld which provides palatable material even when mature; and
3) **MIXED VELD** - veld which is intermediate between the above.

Since Scott's definition of the terms very little work has been conducted on the factors affecting seasonal variation of veld quality. However the extent and nature of these variations have been researched and reviewed periodically since the early work on correcting the nutritional deficiencies of South African veld (Staples and Taylor 1929; du Toit *et al* 1940a; Donnelly 1949; Tainton 1981b; Owen-Smith 1982). Tainton (1981b) summarised some of the characteristics of the various classes as follows:

1a) Sweetveld in summer rainfall areas;
   (a) occurs at low elevation,
   (b) rainfall scanty and erratic, and
   (c) spring period crucial for grazing due to lack of grazeable material.

1b) Sweetveld in all-year rainfall areas;
   (a) occurs at wide range of elevations,
   (b) growth periodic and erratic due to all-year rainfall, and
   (c) grazing most reliable in spring and autumn.

1c) Sweetveld in the winter rainfall areas;
   (a) occurs over a wide range of elevations,
   (b) rainfall variable, and
   (c) dry summer period crucial for grazing.

2) Sourveld;
   (a) occurs in areas of high altitude and relatively (to sweetveld) low temperatures,
   (b) rainfall high and reliable therefore growth more rapid and regular, and
   (c) grazing usually good in spring and early summer.

3) Mixed veld;
   (a) is intermediated between the extremes and may be described as mixed-sweet or mixed-sour depending on

which characteristics predominate.

Such definitions and reviews have allowed some veld farmers to manage their properties according to the principles derived from past studies. At present veld is, however, classified subjectively, by farmers and researchers alike, on its ability to support animals over a period of the year; i.e.

SOURVELD *provides grazing for 6 months or less,*
SWEETVELD *provides grazing for 12 months, and*
MIXED VELD *provides grazing for 6 - 11 months.*

JUSTIFICATION FOR THE STUDY

As mentioned earlier the terms 'sweet' and 'sour' are an integral part of our veld management literature (see Tainton 1981a) yet little attempt has been made to quantify these terms. There has been a call to place these terms on a more scientific footing (Mentis and Huntley 1982) and to improve the objectivity of their application. Whether this can be done remains to be seen. What is of importance though, is to improve the method of classifying veld in terms of its seasonal animal production potential. This need arises out of the growth in the use of computer based management models and expert systems. An environmentally based classification will obviate the need for empirical studies or subjective assessments.

AIM AND OBJECTIVES

The aim of this study is to develop an understanding of the factors which determine the degree of 'sweetness' or 'sourness' of veld and in particular to attempt to ascertain the extent to which soil and climate are implicated. Also an attempt will be made to investigate the degree to which plant quality is genetically controlled, or whether it changes with a change in growing conditions. From this aim one may formulate a number of

specific objectives.

Initially the following specific objectives will be addressed:

1) to establish what factors might play a role in seasonal variation in the digestibility of veld plants;
2) to provide a more objective definition of the terms 'sweetveld' and 'sourveld' than already exists;
3) to determine an objective and quantitative method of measuring (indexing) the degree of 'sweetness' or 'sourness' of *Themeda triandra* (used here as an indicator species);
4) to establish which factors, if any, may be manipulated in order to modify the digestibility of the indicator species *T. triandra*; and
5) to identify areas for further investigation and design experiments accordingly.

PREVIOUS STUDIES

The call by Pole-Evans (1920) for an inter-disciplinary study took over a decade to be implemented. The first study initiated was reported by Louw (1938) who considered the effect, on quality, of defoliation by clipping certain species. A related project was developed to determine the phosphorus deficient areas of South Africa (du Toit *et al* 1940a). This work however only elucidated the possible deficiencies in the veld with respect to animal feed requirements as the procedures adopted for sample collection did not allow the factors determining the seasonal variation in veld quality to be identified. Several studies were conducted by other workers on aspects of the seasonal variation of veld quality and the quality of veld plants and supplementation (Louw 1938, 1944; Louw and van der Wath 1943; Louw *et al* 1948). Most of these studies led to the common practice of feeding supplements to animals grazing veld in winter. Despite the need for supplements being established nearly 50 years ago (du Toit *et al* 1940b), there has apparently

been no attempt to study simultaneously potentially influential factors responsible for poor quality.

The subjective nature of the definitions of sweet and sour veld has not allowed objective classification of grazing land and this has hampered both farming and research in the area of veld management, and in particular it has precluded the ready description of forage flow in different veld communities. Outside Africa much work on the seasonal variation of plant quality has been undertaken (Rauzi *et al* 1969; Laycock and Price 1970; Kalmbacher 1983; Urness *et al* 1983; White 1983; Dickinson 1984). Unfortunately most of this work has been conducted on planted or improved species in temperate environments and is therefore likely to be a simplification of the situation under veld conditions in the tropics. A recent review of environmental and nutritional factors affecting herbage quality was presented in Australia (Wilson 1982). Wilson (1982) makes the point that although there is much information on the differences between species with respect to their digestibility, very few controlled experiments have been conducted to determine environmental effects on herbage characteristics in relation to animal intake. In his review he lists the following as being correlated with dry matter digestibility:

(1) climatic variables such as temperature, daylength irradiance, etc.;
(2) geographic location (latitude); and
(3) soil moisture (particularly moisture stress).

When considering the South African situation and the location of the various veld types (Tainton 1981b) it is obvious that these characters play a role here as well. Generally we may say that the high elevation humid areas are sour whilst the arid low elevation vegetation is sweet. Of course geographic location is more of a descriptive variable and should not be interpreted as playing a direct role in determining forage quality. In other words, on most continents the particular set of environmental variables giving rise to a particular forage quality shows some pattern in their geographic distribution. It is unlikely that

this relationship is consistent on a global or regional basis.

From the foregoing discussion one may get the impression that sweetveld and sourveld are two clearly distinguishable entities. The choice of the words ***sweet*** and ***sour*** may give the impression that the situation is analogous to ***honey/lemon*** with respect, say, to taste. If this analogy is accepted the differences between the two veld types would resolve into differences in species composition or plant types as was proposed by Alberti (1807). The analogy of honey/lemon is, of course, not correct as a study of Acocks's (1975) species list for each veld type will show. We see that a number of species of plants occur in both the sweet and sour veld when we compare Acocks's (1975) lists with the map provided by Tainton (1981b). For grass species in particular the overlap of species occurrence for sweet and sour veld is greatest amongst those species considered useful to the grazier eg *Themeda triandra, Heteropogon contortus, Setaria* spp., *Digitaria eriantha*, and others. From the research point of view then this is fortunate because it allows differences between species to be excluded and a single species to be researched.

Unfortunately the overlap of species does not cover all forms of each veld type. One area in particular is the Karoo. Because animals can be kept in a productive state for 12 months of the year in the Karoo, it is by definition a sweet veld. In this situation however it appears that the animals are relying on different components in each season ie. grasses in summer and bushes in winter (Botha 1981; Roberts 1981). In terms of investigating causal relationships between plant quality and the environment, such situations should be excluded because there is little basis for comparison with other areas where species types do not necessary play a role. The Karoo may therefore be treated as a separate situation and is essentially excluded from this investigation.

CONCLUDING COMMENTS

It is clear from the above discussion that the current definitions of sweet and sour veld are mainly descriptive of season of use rather than of plant quality *per sê*. Currently very little is known of the seasonal patterns in plant quality or the relationship between environment and the capacity of the veld to provide nutrients to sustain livestock. In particular the formulation of nutrient supplements is based on a gross level of resolution and requires improvement.

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CHAPTER 2

TECHNIQUES

INTRODUCTION

Although there have been very few detailed studies on the seasonal variation in plant quality in South Africa (Louw 1938; du Toit *et al* 1940a) the concept of sweet and sour veld has often been interpreted in terms of species composition (plant type). It is now generally accepted that considerable variation exists in the quality of different plant species and this difference is most marked between grass and non-grass plants (particularly legumes), as well as within the grasses (tropical versus temperate species). Theron (1966) has indicated that there is also a large difference in the quality of different species at any one location, with this difference increasing with distance between sites. Although Theron did not measure plant quality directly, he did attempt to record the relative palatability of a number of species. His results were not conclusive, due to inadequacies in the procedures used, but those differences that were recorded were explained in terms of the differences in the physical characteristics of the species studied as well as their differing rates of maturity. In the study by du Toit *et al* (1940a) the differences in the seasonal patterns of plant quality were clearly demonstrated. Unfortunately the types of forage samples collected varied a great deal in composition so that inter-plant differences negated any attempt to explain the factors affecting the seasonal change in plant quality.

As far as this project is concerned, therefore, it is important to avoid any confounding effects of comparisons across different species and it was decided to choose *Themeda triandra* as the indicator species. The choice of this species was based, firstly, on it being relatively well known to Grassland Scientists and Farmers alike, and, secondly because it is widely

distributed throughout southern Africa (Chippendall 1955; Acocks 1975; Chippendall and Crook 1976), as well as Asia (Bor 1960) and Australia (Tothill and Hacker 1983).

Having decided to limit this study to a single species it was then necessary to determine suitable techniques for harvesting the plants and analysing the soil and plant material. The purpose of this chapter is to provide some justification for the techniques used as well as to detail the procedures used for the study.

SAMPLING

PLANTS

Plant components

Sampling grass plant material for an investigation of this nature presents problems associated with the presence of a range in the ages of different components of the plant. In addition to this, the plants at different locations have differing ratios of the various plant components and it is therefore important, in order to provide meaningful data, to collect material of the same age. N M Tainton (personal communication 1983) suggests that grass leaves will be physiologically the same age at specific stages of growth; eg at the time of ligule exposure. There do not appear to be any data to support this notion, however in Langer's (1979) review of grass growth he suggest that rates of leaf production and death are approximately equal, indicating a constant age structure for each morphological unit. On the assumption that this is correct, the plants were sampled by removing the same morphological unit from plant tillers at each harvest; ie ***all the material above the second to uppermost exposed ligule*** (TWO-LEAVES-AND-A-BUD (Figure 2.1)). This method was used for both the field and the glasshouse trial and only vegetative tillers were included in the samples. For the field investigation plants

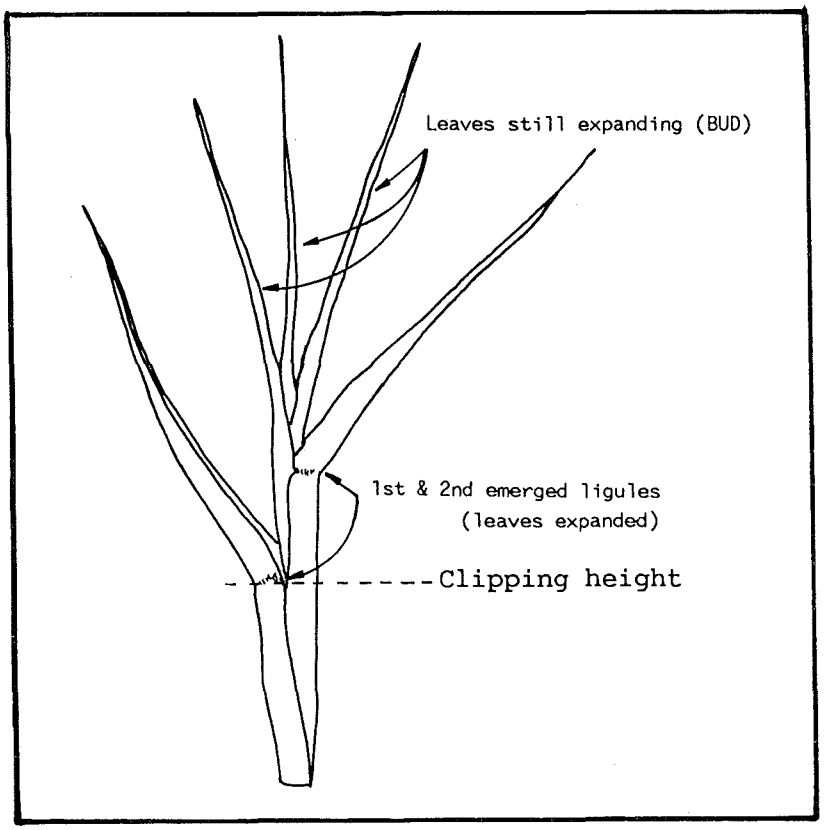

Figure 2.1 Diagrammatic representation of the harvesting unit TWO-LEAVES-AND-A-BUD.

for harvesting were randomly located within each site with a fresh sample being drawn on each occasion. In addition to the TWO-LEAVES-AND-A-BUD unit being collected on site, a number of additional plants were harvested by clipping at 5cm from the ground, half of which were milled as WHOLE plants, and the remainder retained for later separation (SEPARATES). Thus the material was classed into four components:

1) TWO-LEAVES-AND-A-BUD - described above;
2) WHOLE PLANTS - complete areal portions;
3) GREEN LEAF - all the green leaf blade from a number of plants; and
4) DEAD LEAF - all the dead (light brown or red colour) leaf blade from the same plants as the green leaf sample.

The need to separate these plant components was tested (Appendix 1) and showed clearly that the seasonal pattern for each is likely to be different. All plant material was dried in a forced draft oven at 65°C for 48h, milled to 1mm and then stored in glass jars at room temperature in the dark.

Site variability

At any one site in the veld *T. triandra* plants growing side by side are expected to have the same digestibility. However, the variation in the digestibility of plants located at subsites (a few meters apart) may be such that reliable estimates of the *in vitro* digestibility of the whole site may be costly to obtain because a large number of samples may be needed from each site. Furthermore, a large variance of the estimate of the mean digestibility of those estimates may make the detection of differences between the digestibility of plants at different sites (at separate geographic locations) impossible at any reasonable level of precision (say $P \leq 0.05$ to 0.10).

In order to be certain of obtaining a reliable estimate of the mean value of the digestibility of plants at different geographic locations throughout the year, a sampling investigation was carried out (Appendix 1).

From the analysis of those data it can be concluded that there are no significant differences ($P > 0.05$) between plants at a sub-site or between sub-sites at one location. This result enables a composite sample to be harvested. However the number of plants required to make up that sample must be calculated on the basis of the variability within and between sites (G P Y Clark and H M Dicks personal communications 1984).

The number of plants required to form a composite sample in order to detect differences between sites of 5% ($P \leq 0.01$) has been shown to be 17 (Appendix 1). On the basis of this calculation 20 plants were harvested at each location to form the composite

sample for the TWO-LEAVES-AND-A-BUD sampling units as well as for those plants collected as WHOLE and SEPARATES.

SOILS

Soils were collected at each location and returned to the laboratory for analysis. As the purpose of collecting the soil was to provide parameters from which to characterise each site, no test of the sampling variability of soil characteristics at each site was carried out. Apart from this, as each site covers only a small area (ca 0.1ha), chemical and physical characteristics are not likely to vary to any great extent (A Cass and M V Fey, personal communications 1983).

Soils were collected at two depths (200mm and 400mm) using an open bladed auger (75mm diameter). At each site 20 sampling points were randomly located and the samples from each auger hole combined. The samples were then sieved to 2mm and air-dried before storage in large airtight glass jars.

Inspection of the soil chemical variables measured will show that measurements in units of mg/kg and meq/100g were used. It is obvious that these will be highly correlated as one is simply some constant multiple of the other. Both were included here because they do not necessarily indicate the same activity of the element to the plant. In the case of the divalent ions, eg Ca and Mg, the two measures are not the same as for monovalent ions. In a mathematical sense the conversion from one to the other is simply a transformation in the same way as any other arithmetic transformation. As one of the distinguishing features between sweet and sour veld areas is expected to be their base status, it was felt that ion activity (meq/100g) may prove to be a more meaningful measure of chemical status for use in regression analysis than absolute measures of the elements in mg/kg.

PHYSIOGRAPHY

Physiographic information was collected at each site on the first visit, with the following information being collected:

1) ***aspect*** - this was measured, using a prismatic compass, as the bearing of a line perpendicular to the plane of the slope. Level sites were recorded as having zero aspect;
2) ***altitude*** - this was recorded from 1:50 000 Surveyor General Topocadastral Sheets which allowed recordings to the nearest 20m. It was originally intended to use an altimeter but this proved too sensitive as the recordings were affected by barometric pressure; and
3) ***geographic location*** - each site was identified trigonometrically and plotted on a 1:50 000 Surveyor General Topocadastral Sheet. The latitude and longitude at each location were recorded as six figure grid references which were then converted to degrees. These figures were rounded to four decimal places so that each site would be distinguished to the nearest second, ie. ca 50m on the ground.

CLIMATE

The climatic data for each site were derived from existing records. The source of the information used is the Agro-hydrological Research Unit of the Department of Agricultural Engineering, University of Natal. Mean monthly values for rainfall and maximum and minimum temperature were supplied by this unit (now the Computing Centre for Water Research) (M C Dent, personal communication 1984).

TIME

Each site was visited at ca 73 day intervals to give five harvests per year. In view of the distances covered as well as the dispersion of the sites through Natal, it was not possible

to harvest all sites within a week. Some of the harvest periods extended over three weeks, but this was unavoidable. In order to account for this difference in the day of harvest, the time on each occasion was identified by day and week. The first day of July was recorded as day 1 and July 1 to 7 as week 1 etc. This method of accounting for the date would enable time to be used as a variable in a more manageable form.

LOCATION OF SITES

Although it is desirable to have sites located at random through the study area this was not possible for a number of reasons. Firstly, the location of sampling sites was governed by accessibility as well as the wishes of the co-operating agency (Directorate of Forestry, Natal Parks Board and Private land owners). Secondly, the purpose of this study was primarily exploratory and consequently as wide a range of as many variables as possible was considered of prime importance. The third, and most important constraint, was the abundance of *T. triandra* at each location and to this end sites were selected judgementally to cover as wide a range of variables as possible, with a total of 32 sites being located (Figure 2.2; Appendix 1; Table A1.1)

ANALYSIS

This project has required that a considerable volume of laboratory analysis be undertaken. As there is no routine analytical laboratory facility available in Natal, the majority of the analyses were carried out by the author in laboratories of the Faculty of Agriculture at the University of Natal. The numbers of samples to be analyzed (duplicates of 1 600 plant and 64 soil samples) necessitated some modifications to existing techniques as well as the development of the cellulase method of determining digestibility. This method had not previously been used in South Africa.

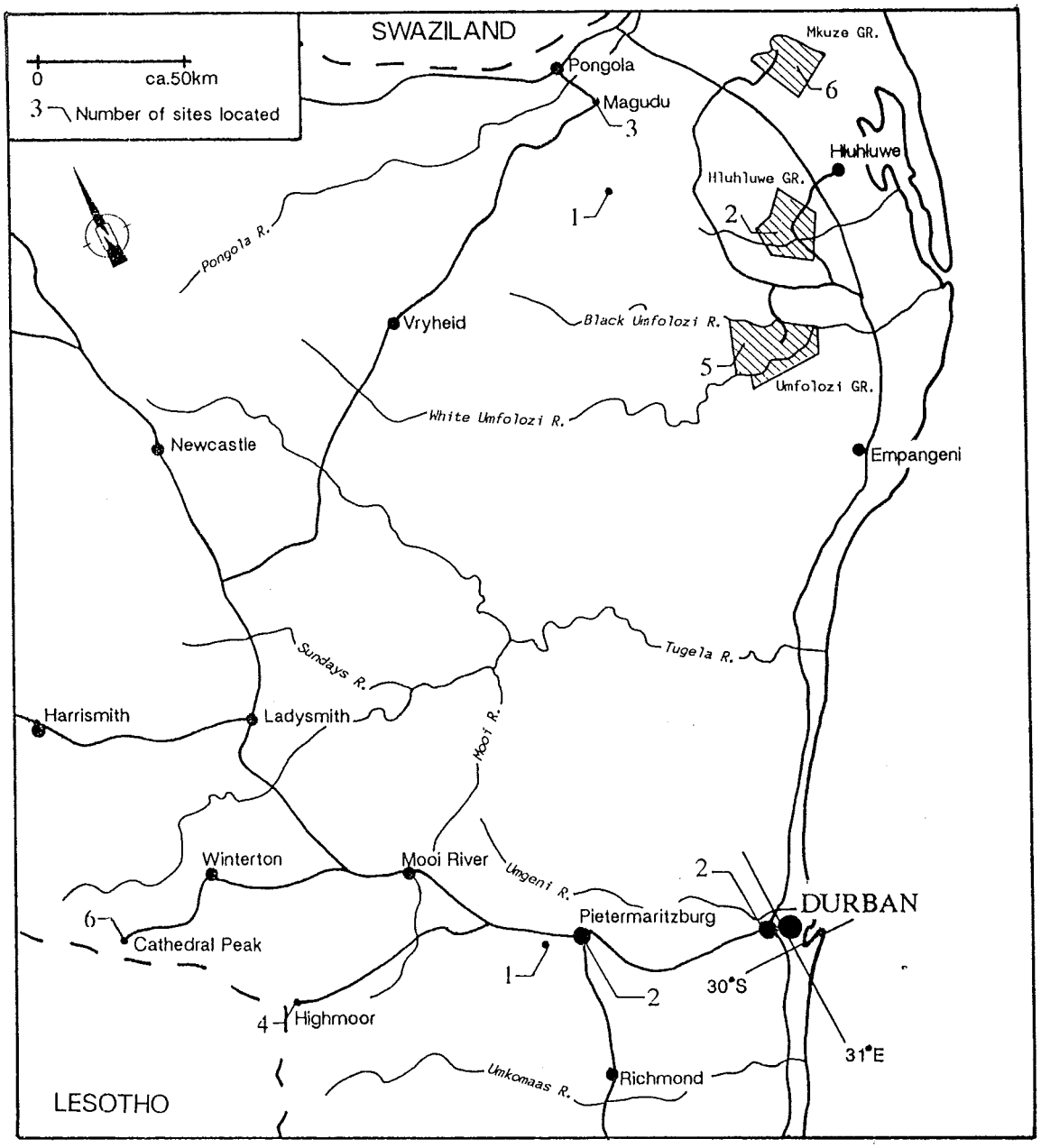

Figure 2.2 The distribution of sites and the number at each location throughout Natal.

Where the techniques have been developed or modified the procedures are detailed below. Where no modifications to standard techniques have been made these are referenced accordingly. Each analysis, unless stated to the contrary, was carried out in duplicate for all samples. For each analysis of plant material each batch contained a standard in order to detect

possible batch errors and to test the reliability of the technique.

PLANT

In terms of the current definition of sweet and sour veld (Scott 1947) the distinction is made by the ability of the veld to carry animals in winter. It would seem logical therefore, that any investigation that attempts to provide information about the relationships between the components that affect the yearly pattern of plant quality should have some direct relation to animal nutrition. The systems currently employed by Animal Scientists for forage analysis are Total Digestible Nutrients (TDN) and the Metabolisable Energy/Net Energy System (ME/NE) (Stielau 1985; van Ryssen 1985). As far as the analysis of grass is concerned these two systems have a number of shortcomings relating to:

1. the inability of the TDN system to distinguish between feeds that have different potentials to promote production (Meissner 1985);
2. the inaccurate subdivision of feed into fractions for TDN calculations which bear little relation to forage quality (van Ryssen 1985);
3. the use of factors, in calculating TDN values, that include non-feed substances;
4. the approximation of efficiency of use of energy in the ME system being dependent on the concentration of energy in the ration; and
5. the cost of determining energy values for feeds because a sophisticated feeding trial is required (Stielau 1985).

A further problem relating to the determination of forage quality is the quantification of palatability to, or acceptance by, the animal (Ivins 1955; Heady 1964). Both these terms are relative, but are said to determine intake (Mentis 1981). If intake is a function of forage quality, then it is safe to consider intake as an absolute index of quality and measurements relating to

intake will be more appropriate in order to index plant quality. The relationship between intake and digestibility has been researched in Australia and recently reviewed (Minson 1982) and found to be generally linear for both tropical and temperate grass species. Methods of estimating or indexing the digestibility of a forage are more reliable than those for intake. As a result of this, it can be assumed that reliable indices of digestibility should provide a means of ranking forage in relation to animal preference. One obvious drawback of this assumption, based on digestibility, is that factors, such as alkaloids or hairiness, that may deter animals from consuming a plant are not accounted for. It is hoped that these differences will be relatively unimportant in this study because only a single species will be used.

Apart from this, the digestibility of the feed (or a suitable index of digestibility) is one factor that relates directly to the portion of feed that is useful to the animal (Bransby 1981). Measures of digestibility are therefore appropriate in this study. In addition to digestibility some measure of the elemental status of plant tissue will provide information on the potential to sustain animals. In particular the concentration of total nitrogen provides an indication of the protein available to the animal (Cullinson 1979). Other elements important in animal nutrition are Calcium, Phosphorous, Potassium and Magnesium and measures of these assist in determining the nutritional adequacy of plant material. The data generated here can, with time, be compared with animal requirements that are described by various feeding tables (Anon 1968, 1970).

Cellulase dry matter disappearance

The 'standard' method of determining dry matter digestibility in South Africa has been the *in vitro* technique of Tilley and Terry (1963). Although this technique has been used successfully it is often criticised because of the variability within and between runs.

This variability occurs mainly from the variation in the composition of rumen micro-flora which alters the digestive characteristics of rumen liquor. In an attempt to control this animals are kept on a standard diet, mainly of lucerne. For the purposes of this study such a diet is not satisfactory because tropical grass species and legumes are likely to have different micro-floral populations used by the animal for digestion (C Dennison, personal communication 1984). This could be overcome by holding all analyses until the end of the investigation but this is not practical because the volume of rumen liquor that could be extracted on one occasion would be insufficient for the number of samples generated here.

In order to overcome the problems discussed above an investigation to compare the *in vitro* rumen liquor method with the cellulase dry matter disappearance (CDMD) method was carried out. The full detail of those tests are reported elsewhere (Zacharias 1986), but a summary of the laboratory procedure for this investigation is provided together with the major findings of the comparison between CDMD and rumen liquor (Appendix 1).

Elemental analysis

As one of the objectives of this project was to determine factors affecting the seasonal decline in plant quality, some method of indexing plant quality was required. Unfortunately there are no satisfactory methods of analyzing grasses to provide an index which relates plant quality directly to animal nutrition (Anon 1985a). The techniques that have been developed (eg van Soest 1982, Dennison and Phillips 1983) are expensive and time consuming and so are not suitable for an exploratory investigation of this nature. The main reason for this is the large number of samples to be generated here. The ME/NE system (van Ryssen 1985) could be used, however most grasses, especially tropical grasses, yield much the same value for gross energy determinations. As a result of this it was decided to determine the elemental status of the samples using wet chemistry methods.

Nitrogen

Nitrogen is determined in forage analysis as this is said to give an indication of the protein content. Traditionally the percentage of nitrogen has been multiplied by a factor of 6.25 to give percent crude protein (CP) (McDonald *et al* 1981). Measures of CP are usually determined from the Kjeldahl technique which converts the protein and most other N sources to ammonium sulphate. The assumptions made in calculating CP using the conversion factors are:

1) that all N in the forage is protein and
2) that protein contains 16% N.

As neither of these assumptions hold, there seems little point in changing the scale of a data set if there is no sound biological reason for doing so (McDonald *et al* 1981). In any event this arithmetical adjustment of %N to give CP, means that published feeding standards actually reflect animal requirements for N and not protein. For the purposes of this study then, all data referring to N will be expressed as %N on a dry matter basis.

A modified micro-Kjeldahl technique was used to determine plant nitrogen. The major modification is that a titration was not carried out but N concentration was determined using a specific ion electrode (Anon undated). The procedure that was used for the plant samples is provided in Appendix 1.

Other elements

The remaining elements (Ca,Mg,K,P,S,Zn) measured in the plant material were extracted from a single sample by ashing and acid digestion (Appendix 1). All the glassware used for these analyses was acid washed to prevent contamination (especially of phosphorous) from detergents and municipal water.

SOIL

Soil samples were analyzed with a view to characterising each site in terms of possible factors affecting plant growth and quality. As mentioned earlier, samples were collected from the A(0-200mm) and B(>200-400mm) horizons at each site. These samples were analyzed for both chemical and physical characters.

Chemical

The chemical analysis of the soils was carried out by the Soil Testing laboratories at Cedara. As this laboratory has recently changed the methods used for soil testing, (Shona Wood, personal communication 1984) a summary of the procedures used for analyzing the samples collected for this investigation is provided (Appendix 1). The choice of elements to be analyzed in the soil was based on macro-nutrient requirements of plants (eg phosphorous). In addition to this those factors which distinguish one soil's chemistry from another and which may reflect the overall environmental condition were considered to be useful separators of sweet and sour veld. Included in this group of parameters are exchangeable bases (calcium, magnesium, potassium and sodium) pH and extractable aluminium as these are used to distinguish eutrophic from dystrophic soils (Macvicar *et al* 1977). The measures of effective cation exchange capacity (ECEC) and acid saturation were calculated from the soil test data. Organic carbon was also determined as this may have a major influence on the availability of soil nutrients for uptake by the plant (M V Fey, personal communication 1983).

Physical

Analysis of the physical parameters of the soil provides information on the inherent ability of a soil to support plant growth. The use of soil texture is becoming increasingly important in the determination of fertilizer recommendations for crop growth because of the influence of particle size on soil

chemistry (Johnston *et al* 1986). The ability of a particular soil to hold plant nutrients or to buffer against extraction of the nutrients by plants or leaching is governed largely by the colloidal fraction (A Cass, personal communication 1983).

Apart from the considerations of soil chemistry, a primary determinant of plant available moisture in a soil profile is the textural class (Johnston *et al* 1986). As the amount of rainfall is a major factor distinguishing sweet from sour veld areas it was considered important to determine parameters that affect the *soil/moisture/plant interface*.

For the glass-house trial (see Chapter 3) the field moisture capacity (FMC) of the soil was required to determine the watering regime required for each treatment.

The techniques used to determine the sand, silt, clay and FMC values for both the field and glass house studies were modified in order to accommodate the relatively large numbers of samples. The details of these minor modifications and their possible effects are described in Appendix 1.

CONCLUDING REMARKS

The variables that have been recorded in this study cover climate, soil chemistry, the physical status of the soil and plant quality. Although the individual units in each of these four groups do not form an exhaustive list, as many as 70 variables will be used to characterise each site. It may be argued that this is far too many variables to be handled in any meaningful way. It must be borne in mind though that this is essentially an exploratory investigation and it is therefore better to have 'too much' rather than 'too little' information. In addition to this, a major expense in this study in the cost of visiting each site and it is the opinion of the author that as much information as is practicable should therefore be collected during each visit to characterise each site. In this

way at least the most obvious parameters should be included in the initial investigation.

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CHAPTER 3

THE EFFECT OF SOIL ENVIRONMENT ON DIGESTIBILITY

INTRODUCTION

In their description of the grassland Biome Project, Mentis and Huntley (1982) identified as a key question in the definition and description of the grassland biome, a need to determine the relationship between grassland types and soil nutrients. They feel that this should be done in terms of so called sweet and sour veld.

One of the first factors that needs to be established then is whether or not the quality of plant material can be altered by altering the nutrient status of the soil that it is growing in. Indeed, Soil Scientists have argued in the past that applications of lime are all that is required to improve the quality of sour veld (M V Fey and N M Tainton, personal communications 1983). Some studies have been carried out in South Africa to investigate the effects of the addition of nutrients to the soil on herbage (qv Booysen 1981). Unfortunately few have considered plant quality and the results are not clear. No studies have been carried out in order to determine how an entirely different soil regime would affect the quality of a plant.

A glass-house experiment was designed in order to address the question of the effect that soil amelioration and soil type has on plant quality.

OBJECTIVE

The objective of this glasshouse trial was to address the question: *Can the quality (digestibility) of veld plants be modified by altering the soil environment in which they are growing?*

PROCEDURE

EXPERIMENTAL DESIGN

A four-by-four latin square (split-plot) design was laid out in a glass-house (modified environment) at the Faculty of Agriculture, University of Natal. The whole plots were *times of harvest* (four harvests 73 days apart (T_1 to T_4)), and the sub-plots the six treatments from combinations of *three soil types* and *two plant types*. The 73 day time interval was chosen to give five equal time intervals per year. The latin square (split-plot) design has been selected to overcome the problem of shading of the harvested plants by adjacent unharvested plants. In addition the design does not require the treatments to be rotated to balance light effects, as would be the case with a randomised blocks design.

The null hypothesis (H_o) under test was; **the *in vitro* digestibility (sweetness/sourness) of *Themeda triandra* is not affected by altering the soil conditions in which it is growing.** Soil nutrient status was chosen according to the following definitions (Macvicar et al 1977):

1. **dystrophic** - soil that is markedly leached such that the sum of the exchangeable Ca, Mg, K and Na is less than 5meq/100g soil.
2. **eutrophic** - soil that has little or no leaching such that the exchangeable Ca, Mg, K and Na is greater than 15meq/100g soil.

The third soil was produced by ameliorating the dystrophic soil with the addition of Ca and Mg to raise the base status to that of the eutrophic soil. The two plant types were the *T. triandra* plants collected from the same sites as those where the soils were obtained.

The six treatments tested were combinations of:

1. dystrophic soil (D);
2. eutrophic soil (E);

3. dystrophic soil ameliorated (A);
4. sour veld plants (SR); and
5. sweet veld plants (SW).

From each of the above combinations the following treatment codes were developed:

SWE = sweetveld plants with eutrophic soil;
SWD = sweetveld plants with dystrophic soil;
SWA = sweetveld plants with ameliorated soil;
SRE = sourveld plants with eutrophic soil;
SRD = sourveld plants with dystrophic soil; and
SRA = sourveld plants with ameliorated soil.

All treatments were harvested as TWO-LEAVES-AND-A-BUD (see Chapter 2) and the REMAINING material. Inflorescences were selectively removed and were **not** included in the samples as not all plants flowered simultaneously.

LAYOUT OF THE EXPERIMENT

Soil was collected for the experiment at the following locations;

a) Dystrophic soil (Cedara)
   Map: South Africa 1:50 000 topocadastral sheet
   2930CA MERRIVALE
   LATITUDE - 29° 33' 48"
   LONGITUDE - 30° 14' 39"

b) Eutrophic soil (Ukulinga)
   Map: South Africa 1:50 000 topocadastral sheet
   2930CD PIETERMARITZBURG
   LATITUDE - 29° 39' 48"
   LONGITUDE - 30° 24' 15".

Once collected the soil was passed through a 5mm screen and then air dried. After drying the soil, 9.0kg was placed in each of ninety six 10dm^3 plastic buckets. Each group of buckets was colour-coded for ready recognition and all buckets were adjusted to equal mass with the addition of glass beads.

Soil physical and chemical analysis

Physical

The field moisture capacity (FMC) was determined using the cylinder method (see Appendix 1) in order to calculate the amount of water to add to each bucket to raise the soil to field capacity (Table 3.1). As moisture level was not a treatment variable, maintaining the soil at near field capacity would reduce the likelihood of moisture stress affecting the growth of the plants.

Table 3.1 Field moisture capacity (FMC) determination for eutrophic and dystrophic soils.

	[1]EUTROPHIC	[1]DYSTROPHIC
Beaker+wet soil (g) (A)	561.00	509.10
Beaker+dry soil (g) (B)	454.90	399.80
Water (g) (A - B = C)	106.10	109.30
Beaker (g) (D)	144.84	153.65
Soil (g) (B - D = E)	305.06	246.15
Field capacity water content (Kg_{water}/kg_{soil}) (C/E)		
MEAN	0.3478	0.4440
± s (kg_{water}/kg_{soil})	± 0.0151	± 0.0092

[1] each value is the mean of 10 samples

Chemical

Soil analysis was carried out by the Department of Soil Science and Agrometeorology at the University of Natal so that differences in Ca and Mg levels, between the two soils, could be determined (Table 3.2).

Table 3.2 Chemical analysis of eutrophic and dystrophic soils showing the difference in Ca and Mg levels.

SOIL PARAMETER	EUTROPHIC	DYSTROPHIC
AL (meq/100g)	0.12	2.10
Ca (meq/100g)	10.00	1.34
Mg (meq/100g)	1.10	5.50
Amount Ca required (meq/100g)[1]		8.46
Amount Mg required(meq/100g)		4.40

[1] this represents the amount required to ameliorate the dystrophic soil to the levels found in the eutrophic soil

Soil adjustments

Once soil analysis was completed a portion of the dystrophic soil was ameliorated by adding Ca and Mg as $Ca(OH)_2$ and MgO. Sufficient $Ca(OH)_2$ and MgO were added to bring the levels of the two elements in the dystrophic soil to those of the eutrophic soil (Table 3.2). The amounts to be added (Table 3.3) were adjusted to account for impurities in the reagents and mixed in a cement mixer in 20kg batches. This ameliorated soil was also weighed into 9kg units in the same manner as the eutrophic and dystrophic soils.

Table 3.3 Amelioration calculation for adjusting levels of Ca and Mg in dystrophic soil to levels in eutrophic soil.

	Ca	Mg
meq/100g required (A)	8.460	4.400
Molecular mass of element (B)	40.080	24.305
Valance of element (C)	2	2
$\Rightarrow \mu g_{element}/g_{soil} = A \times \frac{B}{C} \times \frac{1000}{100}$ (D)	1695.384	534.710
$\Rightarrow g_{element}/kg_{soil} = D \times \frac{1}{1000}$ (E)	1.695	0.535
Molecular mass of compound (F)	74.084	40.299
$\Rightarrow g_{compound}/kg_{soil} = \frac{E \times F}{B}$	3.133	0.886

Planting

Whole *T. triandra* plants were removed from the same locations as the soil and the soil washed from their roots. The plants were then planted in the buckets according to treatment (see above). The buckets were watered to field capacity (Table 3.1) and placed in the shade.

As this first establishment was only partially successful (20 plants survived) subsequent replanting was required. This was done in 700g units (air dry soil) to enable larger numbers than required to be established. The 700g pots were treated in the same way as the original plants with the exception that they were placed in a mist chamber for 21 days to reduce stress. Although this proved more successful a third replanting was necessary before sufficient plants were established.

In order to transplant the replacement material, 700g of air dry soil was removed from the original buckets and the entire 'transplant' placed in the bucket. This procedure was adopted to prevent damage to the root system. The buckets were then watered as usual.

Layout and randomisation

Once the required numbers of plants were established in buckets the experiment was laid out according to the design (see above and Figures 3.1 and 3.2). As there were effectively three establishment dates these were randomised over the whole experiment to reduce the effect of any possible error that this may cause (ie treated as a random element).

Each plot was surrounded by a white masonite screen to reduce the effect of insolation on soil temperature. This method is considered preferable to using white buckets without a surround as air movement is also restricted (M J Savage, personal communication 1983), reducing the likelihood of temperature

fluctuation.

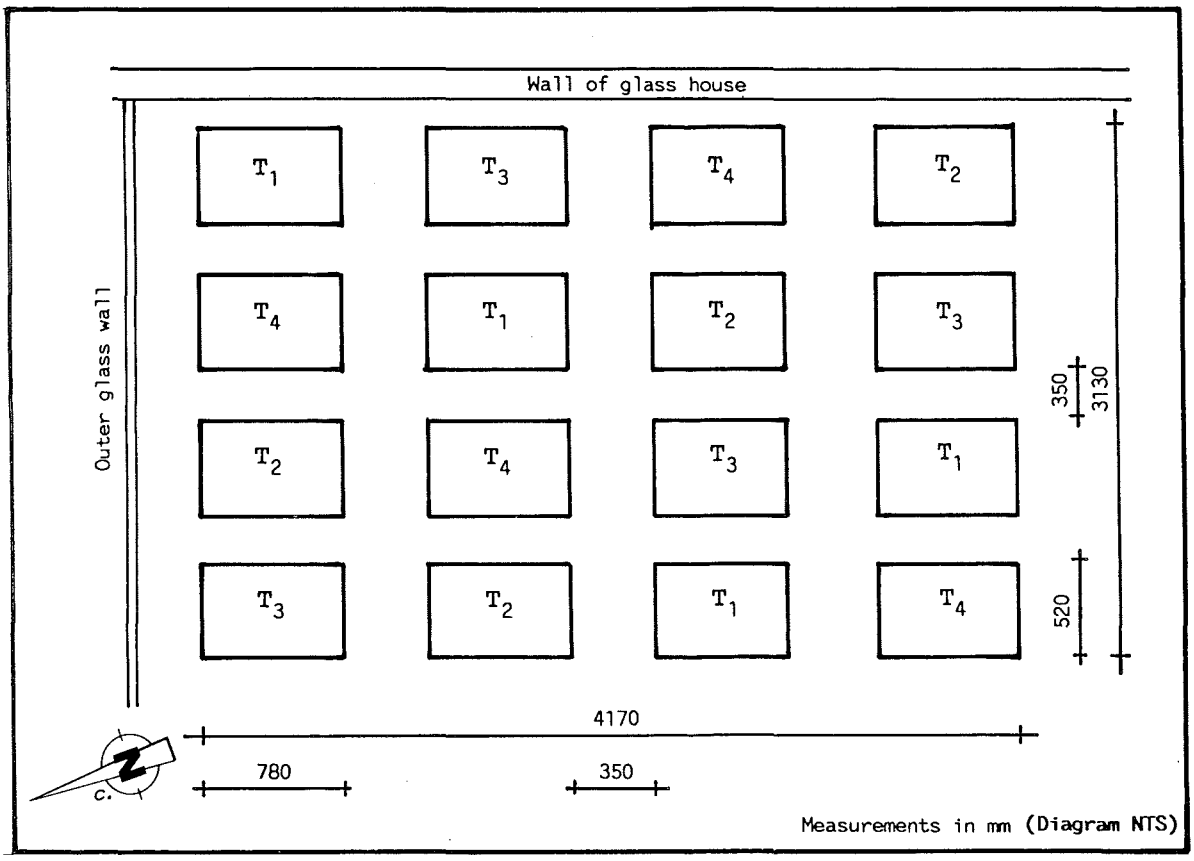

Figure 3.1 Layout of whole plots (T_1....T_4 refers to the time cutting intervals 73 days apart).

Watering

The pots were watered to the nearest 10g of field capacity every second day using an electronic balance with a check weighing facility that was fitted with a water flow controller. This devise was built for the experiment (C W Zacharias, personal communication 1983) and enabled the operator to ensure that each bucket was watered to the correct weight. A counter weight was used to avoid having to reset the apparatus for the two different soil types. The ameliorated soil was watered to the same level as that of the dystrophic soil.

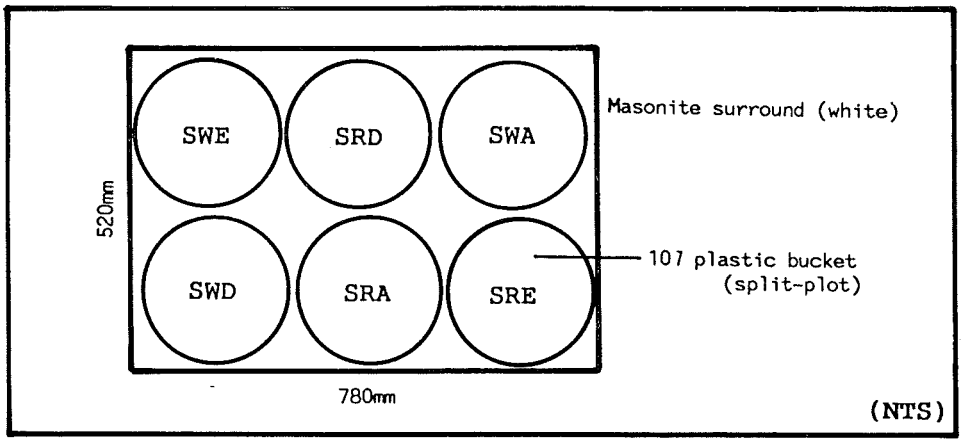

Figure 3.2 Layout of split-plots with one randomization of the six treatments (treatment codes are combinations of sweet (SW) and sour (SR) plants by eutrophic (E),dystrophic (D) and ameliorated (E) soils).

RESULTS AND DISCUSSION

SOIL AMELIORATION

The chemistry of bases in the soil is somewhat difficult to predict and it is therefore necessary to determine the success of the amelioration of the dystrophic soil. This is best done after a number of wet/dry cycles (M V Fey, personal communication 1984). The soils were sampled after ca 12 months using a core sampler and tested in the laboratories at Cedara (see Chapter 2) (Table 3.4).

The amelioration was only partially successful, with the adjustment bringing the base status to only about half the level of the eutrophic soil. The amelioration did not however increase the pH of the soil but reduced the Al concentration (3.30 to 0.06meq/100g) which had the effect of decreasing the acid saturation to a level very close to that of the eutrophic soil (0.40 vs 0.22) relative to its previous value of 44.82%.

Table 3.4 A comparison of the soil chemical status of three soils after ca 12 months of wet/dry cycles (each value is the mean of four replications analysed as individual samples).

	SOIL TYPE		
ELEMENT	DYSTROPHIC	AMELIORATED	EUTROPHIC
P mg/kg	5.60	4.25	2.50
K meq/100g	0.15	0.14	0.19
Ca meq/100g	1.95	8.41	13.16
Mg meq/100g	1.71	4.85	7.88
Na meq/100g	0.16	0.16	0.27
Al meq/100g	3.30	0.06	0.05
pH	4.75	4.75	4.85
Acid saturation (%)	44.82	0.40	0.22
ECEC meq/100g	7.38	13.03	21.68

SEASONAL TREND IN DIGESTIBILITY

Introduction

One of the major differences between sweet and sour veld, in terms of the current definition (Scott 1947), is the ability of sweetveld to carry stock throughout the year. By implication then, some seasonal trend in plant quality is expected and this should be demonstrable in plants from the two situations. For this glass-house investigation the seasonal trend in plant quality could be determined from a number of serially harvested samples.

Results and discussion

After the initial establishment of the trial, which was completed by May 1984, the plants were allowed to grow undisturbed until 20 August 1984. On this date, all 96 plants were clipped and the TWO-LEAVES-AND-A-BUD as well as the REMAINDER components dried

and milled. This initial harvest served as a pre-treatment so that all plants would have been defoliated at the same time at the start of the cycle of TIME treatments.

After this initial pre-treatment harvest the appropriate treatments, T_1 to T_4 (see Figure 3.1), were harvested on 1 November, 13 January, 27 March and 8 June respectively (ie 73 days apart). All plants were then harvested on the 20 August so that the cycle could be repeated, as well as to enable all the treatments to be compared using material that was of the same chronological age (73 days) by taking the appropriate subset of data.

Pre-treatment harvest

As some of the plants had not developed sufficient leaf the yield of milled material was very low and the treatments could not be analyzed using the individual replications. It was then decided to combine the four replications within each TIME treatment (sub-plots) so that sufficient material was available for analysis. The four separate TIME treatments (whole plots) were used as replications. A preliminary analysis of variance (randomised blocks) was carried out on these data.

The analysis of these data showed, for TWO-LEAVES-AND-A-BUD, a range from 38.73 to 42.40% CDMD. However, there were no significant differences between treatment groups ($P>0.05$)(Table 3.5). Although this result is based on only a few degrees of freedom (5) it does nonetheless suggest that the differences in plant quality between sweet and sour veld are less marked in spring (see Chapter 4). This result is in keeping with the descriptions of sweet and sourveld with respect to their relative quality.

The analysis of variance of the data from the REMAINDER component showed significant differences ($P\leq0.05$) between treatments. On closer inspection using least significant differences (LSD), only

Table 3.5 Analysis of variance table for TWO-LEAVES-AND-A-BUD harvested on 20 August 1984 (each treatment replication (4) consists of pooled material from four sub-plot plants).

SOURCE	DF	SS	MS	F
Blocks	3	24.1959	8.065	
Treatments	5	28.8502	5.770	1.1207 $NS_{(5;15)}$
Error	15	77.2160	5.147	
Total	23	130.2621		

$SE_{single\ treatment}$ = 2.27 CV% = 5.57

a single treatment was lower than the rest (Table 3.6). This treatment (SRA) did not behave as expected if soil amelioration is able to alter plant quality. However, as this was only a preliminary analysis on an incomplete data set, no further conclusions can be drawn.

Table 3.6 Comparison of mean cellulase dry matter disappearance (CDMD) values for the REMAINDER harvested on 20 August 1984 (treatment means with the same underscore are not significantly different P>0.05).

TREATMENT	[1]SRA	SRE	SRD	SWD	SWE	SWA
MEAN[2]	30.34	37.46	37.91	38.37	39.78	40.66

$SE_{single\ treatment}$ = 4.02 CV% = 10.74

1 combinations of sweet (SW) and sour (SR) plants and eutrophic (E), dystrophic (D) and ameliorated (A) soils

2 each value is a mean of 12 values

Material with different rest periods

According to the design of the experiment (see above) material from the TIME treatments was harvested from plants with periods of rest increasing from 73 days (T_1) to 292 days (T_4) (Figure 3.3 and 3.4).

Although there was a clear decline in plant quality over all treatments the difference between sweet and sour plants is relatively small. In general the sweet plants have a slightly higher CDMD value than do the sour plants, as would be expected. On the basis that the individual soils are not significantly different within plant types (see later), the data were pooled and the average value for each harvest used to plot the seasonal decline in CDMD (Figure 3.5). From these pooled data a far clearer picture emerges, with the CDMD for both treatments falling rapidly from November to January. The sour plants had a larger decline in CDMD than did the sweet plants and this decline continued throughout the year. It appears from these

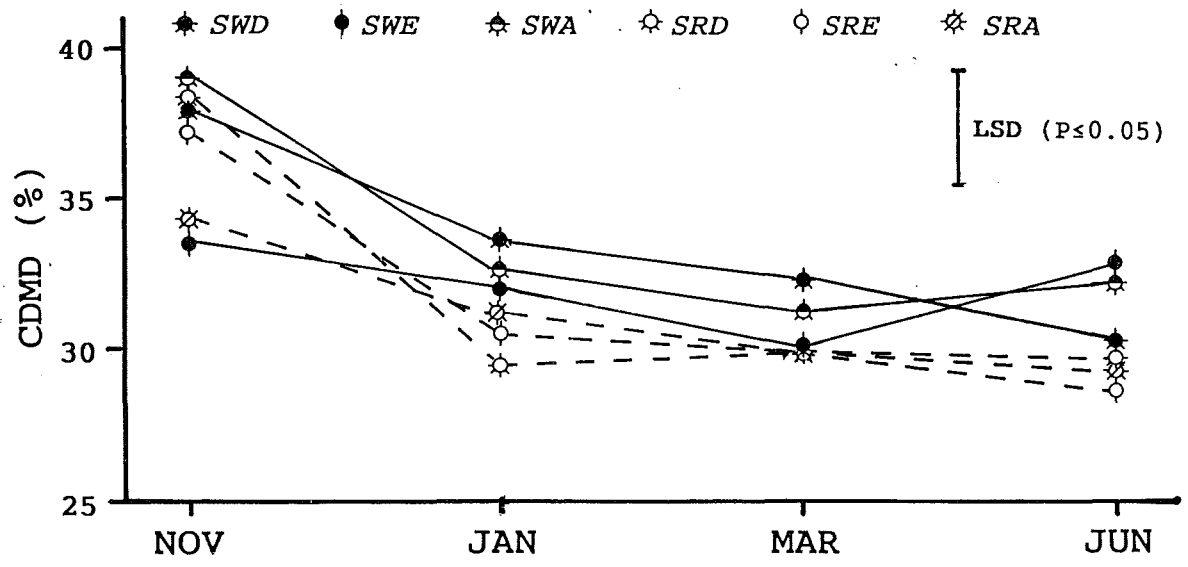

Figure 3.3 Seasonal trend in cellulase dry matter disapperance (CDMD) for TWO-LEAVES-AND-A-BUD harvested through year after increasing periods of rest from each of six treatments.

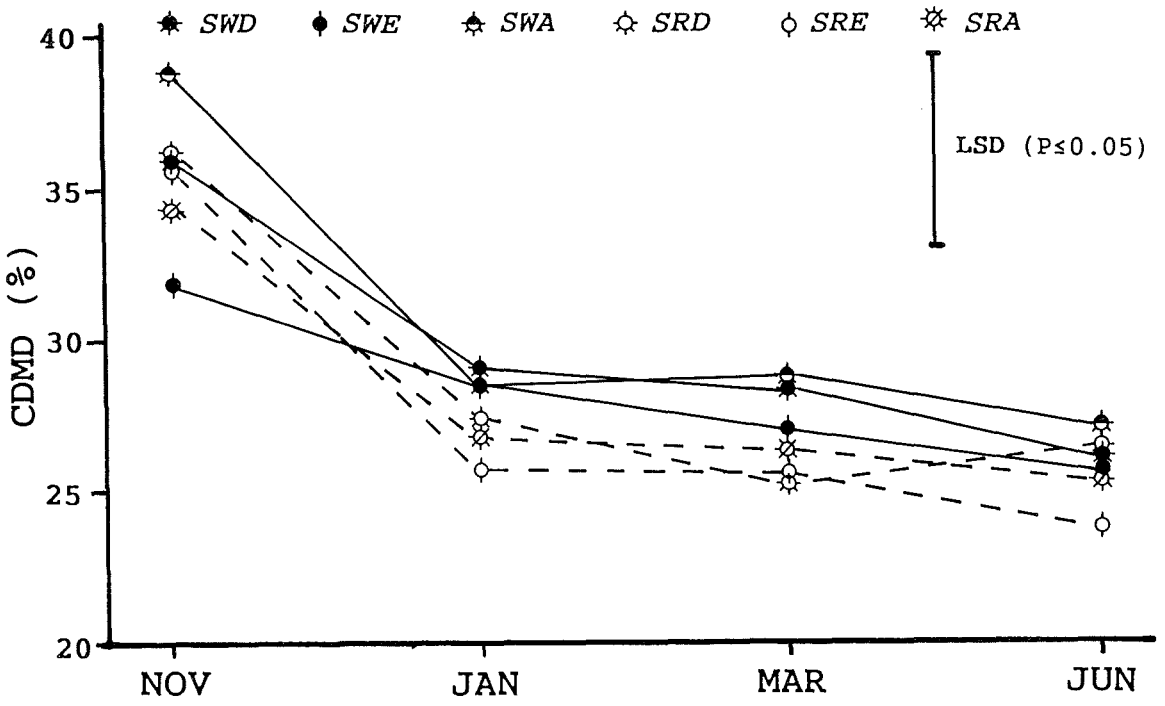

Figure 3.4 Seasonal trend in plant quality cellulase dry matter disappearnce (CDMD) for REMAINDER harvested throughout the year after increasing period of rest.

data that the sweet plants begin to increase in quality (CDMD) before the sour plants at the start of the next spring.

Material of the same age

At each harvest the plants that had previously been harvested were also clipped. This additional harvest was carried out so that material of the same age could be compared throughout the year.

A trend similar to that determined for the material of different ages was also found from January to June (Figure 3.6) although the differences between the two plant types were smaller.

The effects of soils on digestibility

In addition to the study of the seasonal trend in CDMD these data were also subject to analysis of variance. In the original

year. From these data one could ascertain whether the use of the TWO-LEAVES-AND-A-BUD component provides similar results to those from re-growth of the same age. The results of the analysis of variance are provided in Table 3.7.

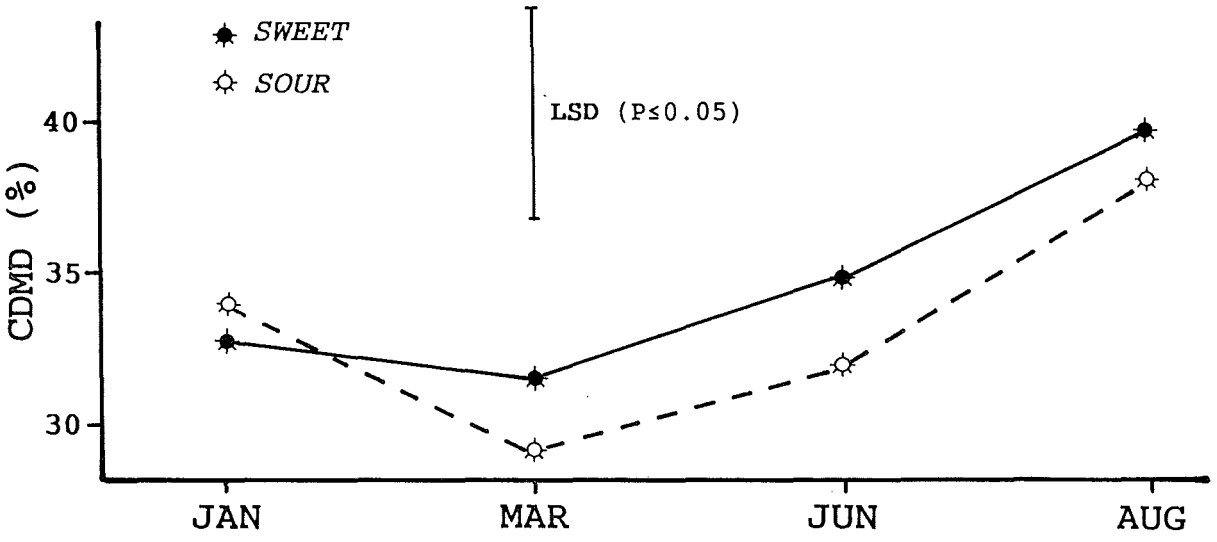

Figure 3.6 Seasonal trend in cellulase dry matter disappearance (CDMD) for the TWO-LEAVES-AND-A-BUD component that is 73days old from sweet and sour plants).

As has been demonstrated earlier (Figures 3.3 to 3.5) there were significant differences in the digestibility of plants harvested at different times of the year. There was a significant decline in quality from the spring to the following autumn for both plant types with the sour plants having a lower value than the sweet veld plants (Table 3.8). Although this confirms what we might expect from the sourveld plants, investigation of the full analysis summarised in Table 3.7 showed the difference in quality to be 1.5% CDMD units. The biological validity of a significant (P≤0.01) difference of 1.5% in the digestibility of forages is questionable. There is some doubt whether animal performance would be affected by such small differences in forage quality. It could be concluded therefore that although these differences may be statistically different, biologically they are not likely to be meaningful.

Table 3.7 Summary of test of various components by analysis of variance of cellulase dry matter disappearance (CDMD) data from the glasshouse trial.

	Design[1]		73 day	
	B	R	B	R
Time	***	**	*	**
Plants	***	**	**	**
Soils	NS	NS	NS	NS
Time x Plants	*	NS	*	NS
Time x Soils	NS	NS	NS	NS
Plant x Soils	NS	NS	NS	NS
Time x Plants x Soils	**	*	NS	NS

[1] Design = material ranging from 73 to 266 days old.
73 day = material all 73 days old but harvested at different times.
B = TWO-LEAVES-AND-A-BUD; R = Remainder
*** = P≤0.001 ** = P≤0.01 * = P≤0.05 NS = P>0.05

The original purpose of this glass house trial was to determine the effect of soil chemistry on plant quality measured by CDMD. The data from this experiment showed no significant differences (P>0.05) between soils (Table 3.7). Furthermore, none of the soil interactions were significant (P>0.05), suggesting that TIME of year and plant type have a greater influence on the quality of plant material than soil type.

CONCLUSION

The results obtained from this glasshouse investigation appear to be in conflict with field observations (see later) with the exception of the seasonal trends in plant quality. It would be expected that the plants growing in their normal soil environment (eg sweet plants on the eutrophic soil) should behave similarly to those in the field (see later) where plants on dystrophic soil (eg Highmoor) had considerably lower digestibility than plants on eutrophic soils (eg Umfolozi). In addition to this plant nutrient status has been shown to be directly affected by the

Table 3.8 Mean cellulase dry matter disappearance (CDMD) values (%) of sweet and sour *Themeda triandra* plants harvested at different times of the year.

	[1]Design		[1]73 Day	
	[2]B	R	B	R
Sweet	33.19	29.61	34.63	34.14
Sour	31.52	28.28	33.13	32.15

[1]Design = material increasing in age from 73 to 292 days
73 day = material 73 days old but harvested at different times
[2]B = TWO-LEAVES-AND-A-BUD R = REMAINDER

nutrient levels in the soil (Laycock and Price 1970; Mengel and Kirkby 1978).

The outcome of this experiment may be due to radical changes that have been made to the plant-soil-environment system during the establishment of the trial. Tisdale *et al* (1985) list (1) soil texture, (2) moisture content and (3) temperature as the most important factors which modify the delivery of ions to the roots. Of all the features of the environment that are modified when taking plants and soil from the field to the glasshouse, these are most likely to be radically altered. During this experiment the soil texture has been modified by sieving, the moisture regime by keeping the soil at field capacity year round and the temperature controlled (especially the diurnal fluctuations) by air conditioning. The interaction of these three modifications is likely to have had the effect of improving the availability of nutrients to the plants in all treatments. The result of this was that the plants had an adequate environment to produce quality material from all six treatments. Coupled with this, is the possibility that all the treatments may have experienced a rapid depletion of nutrients in the pots. Essentially, therefore, all treatments may have enjoyed a similar soil environment. This is one of the many problems concerned with the

validity of pot trials whose results are to be extrapolated to the field. Such a problem is difficult to overcome because adding nutrients to maintain the treatments would have deviated from the purpose of this study.

In terms of the objective of this experiment it appears that manipulation of the soil environment may alter CDMD. However the small adjustments that were made to digestibility (CDMD) are not likely to have a significant effect on animal performance.

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CHAPTER 4

PROPOSAL FOR THE FIELD STUDY: factors affecting the quality of *Themeda triandra*

INTRODUCTION

The broad purpose of this field study was to quantify the seasonal variation in plant quality and to determine, if possible, the relationship between these patterns and the environment that the plants are growing in.

The specific purpose of this Chapter is to present the proposal for that investigation. The justification for the project and a description of the techniques will not be presented as these have already been covered in Chapters 1 and 2.

NULL HYPOTHESIS

From the available literature and observations (Chapter 1) it is expected that the quality of veld in the sweetveld areas, as measured by digestibility, will decline less markedly than that in the sourveld areas. This then forms the basis of the conceptual model on which this study is based (Figure 4.1). In addition to these two a third group is recognised as intermediate. This group is referred to as mixed veld.

PROCEDURE

It was intended that this field study be conducted with a view to determining the relationships between four major groups of variables. These are grouped for convenience as this may facilitate an inter-disciplinary approach. The groupings are:

1) plant,
2) soil,
3) physiographic, and
4) climatological.

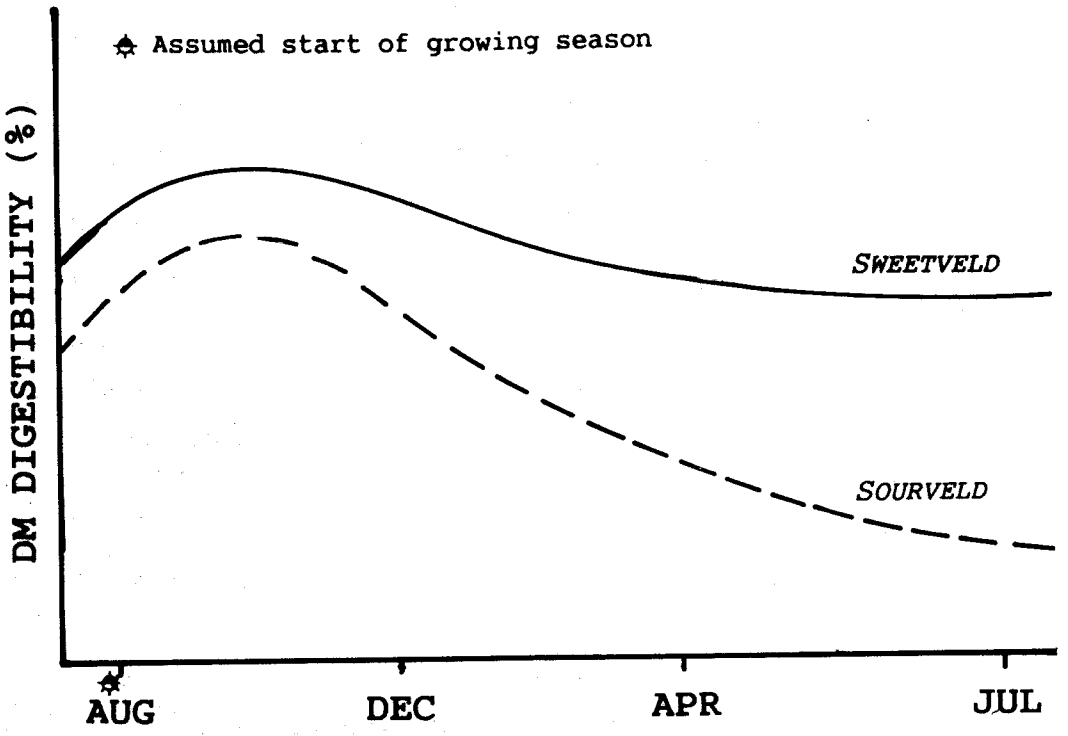

Figure 4.1 Expected relationship between dry matter digestibility and season for **sweetveld** and **sourveld** areas of South Africa.

PLANT

Included in this group are the following:

1) digestibility of organic matter *in vitro* (Zacharias 1986) and
2) chemical status.

The above analyses should provide an indication of the quality (usefulness) of the plant material to the ruminant.

SOIL

The soil investigation can be further sub-divided into:

1) physical aspects - (a) particle size distribution, and
   (b) organic carbon.
2) chemical aspects - (a) pH,
   (b) exchangeable acidity,

(c) nutrient status (P levels), and
(d) base status (extractable bases).

PHYSIOGRAPHIC

Quantification of the environment is a complex task and because of this only those factors which are known to influence vegetation type are included. The following are to be considered for this investigation:

1) aspect (compass bearing),
2) altitude, and
3) geographic location (6 figure grid reference).

CLIMATOLOGICAL

Those factors influencing plant growth have been chosen with only rainfall and temperature data being available.

1) Rainfall - (a) season
   (b) mean annual rainfall and range
2) Temperature - (a) maximum
   (b) minimum.

Information regarding these parameters will be recorded at about 30 sites in Natal to provide as many combinations and variations of them as possible. The indicator plant to be used will be *Themeda triandra* because of its wide distribution in South Africa (Chippendall 1955; Acocks 1975; Chippendall and Crook 1976).

ANALYSIS

The data will be analyzed using multivariate statistical procedures and methods appropriate to the data. There are a number of alternative techniques and these will be applied as the data, or portions of the data, becomes available (Chapter 5).

CONCLUSION

The data from the plant quality determinations will be used to

test the validity of this theoretical relationship (Figure 4.1) as well as providing input for the multivariate analysis.

This theoretical relationship, it is hoped, will form the basis of more objective method of indexing the degree of sweetness and sourness of veld. By determining the nature and the degree of the decline in plant digestibility with time, an index of digestibility of *Themeda triandra* can be used to rate the veld in terms of its sweetness. The digestibility value together with elemental analyses can be used to determine the value of *Themeda triandra* as herbivore food at different times of the year and so provide a biologically based definition of sweetness or sourness which will replace the current subjective method.

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CHAPTER 5

INVESTIGATING THE FIELD DATA

INTRODUCTION

The data that were collected for this investigation have covered most of the factors of soil and plant chemistry which are used to determine plant and animal nutrition (Cullinson 1979). An attempt has been made to cover as much variation of these factors as is available in Natal. This has left a large data set from which no outstanding features are immediately apparent.

The purpose of this Chapter is to examine these data using a variety of techniques in order to determine which factors bear further investigation. Once this is done, a set of hypotheses can be erected and tested more formally.

TREATMENT OF MISSING DATA

In any field-sampling exercise it is inevitable that some samples will not be available on every occasion. The field excursions for this study were no exception and as a result of floods, fire and of insufficient material on some sites, 10% of the plant quality data are missing. In an univariate situation this is usually not a problem as there are procedures for accounting for the missing values (Rayner 1967). Most statistical software has algorithms which calculate appropriate missing values for least squares or associated analyses (eg Anon 1985b; 1988). In the case of this exploratory investigation it is intended to use multivariate methods. Unfortunately multivariate pattern seeking procedures do not have methods for handling missing data and they ignore (reject) variable sets that contain missing values. As far as the plant quality data are concerned this resulted in over 60% of the data being excluded from the analyses. This is obviously unacceptably high given that only 10% of these data are missing. The problem now is how to 'make up' these missing data?

A number of options are available in this case:

1) Accept that 60% of the available information will be excluded from the analyses at the investigatory level. This is clearly not a logical option because the majority of the available information would not be used. This would be inefficient.
2) Guess the missing values on the basis that we are more interested in trends than absolute values. It is my opinion that, from a biological point of view, a qualified guess is better than no value at all. Unfortunately such an option is not possible because it will be criticised on the basis of not being objective and therefore biased.
3) Use the available information to calculate arithmetic means between the previous and following points. Such an option resolves to ignoring the variability of the associated variables in the set and is a form of total smoothing. This option is at least objective and on that basis would be acceptable.
4) Use some interpolation procedure to calculate the missing values. This alternative is similar to 3) above, but this option takes into account all the values of the set and not just those on either side of the missing value. Such a procedure therefore uses all the available information and is considered the most desirable option.

The course of action in option 4) was taken and the algorithm used to estimate the missing data is based on Reinsch (1967) and is supplied by SAS (Anon 1985b). Once calculated the imputed data were included in the data set and will be used in all cases where multivariate methods are employed.

THE CHARACTER OF THE DATA

In terms of the objectives of this project (Chapter 1) a major approach has been one of explaining possible relationships between plant and soil chemical status. Because little is known of the factors affecting seasonal changes in plant quality many

of the variables measured here might be cross-correlated and non-normal. This needs to be tested to ensure that the mathematical procedures used are not invalidated.

CORRELATION

As one of the intentions of this study is to determine a relationship between plant quality and environment it is important to ensure that the independent variables used in any model are not themselves correlated. Further, if a useful index of sweetness can be developed those variables which are easy or inexpensive to measure should be used in preference to expensive ones. Another advantage of this investigation of the data is that the numbers of variables can be decreased so that only the most important variables are used. This reduces the amount of unnecessary computation.

Site characteristics

There are a number of methods for determining the correlation between a number of variables. However, because little is known at this stage of the distributions of the data Spearmans Rank Correlation Coefficient (r_s) was used as no assumptions need to be made regarding the underlying distribution of the data (Siegel 1956; Neave and Worthington 1988). The algorithm used for this test of these data was that included by SAS (Anon 1985b). Variables were considered non-correlated if the Spearman Rank Correlation Coefficient had a value greater than that specified by Siegel (1956) as being significant (ie $P \leq 0.05$). In their discussion on correlation Neave and Worthington (1988) warn that such an approach does not necessarily efficiently remove variables that are biologically correlated. In particular, high correlation coefficients of what ever type (eg Pearsons (r), Kendalls (τ) or Spearman (r_s)), do not necessarily imply a causal relationship. The problem of choice here is a mathematical one rather than a biological one. Unfortunately there is no objective method of choosing appropriate values for significant

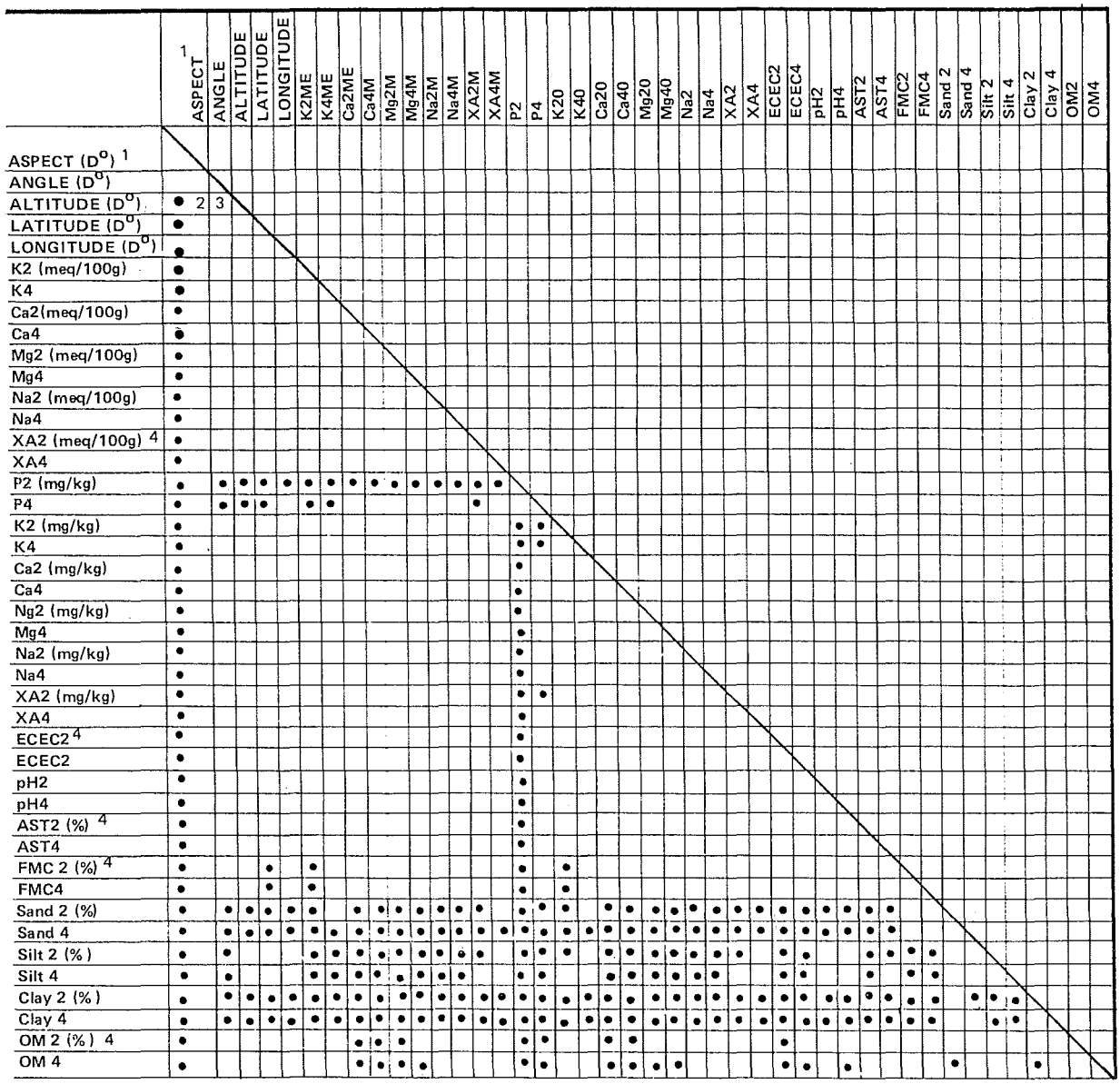
or meaningful correlations. The problem is increased in multivariate cases where a large number of variables from a number of sites increases the probability of calculating a significant correlation value even though it might be of the order of 0.38 (on a scale of 0 to 1). A correlation coefficient of this size would not usually be considered biologically meaningful but could be statistically highly significant. In such cases a subjective decision is needed and for both the environmental and the plant data r_s values ≥ 0.65 were taken to be biologically meaningful (significant). Variables with r_s values with a probability of >0.05 were taken as non-significant according to convention (Rayner 1967). Variables with a coefficient greater then these cut offs were taken as being not associated in the population from which they were drawn (Table 5.1). It must be stressed here that tests of multiple association or non-linear association are not necessarily detected using this method. The method is however more efficient in detecting gross association, which is likely to affect regression procedures used later, than Pearsons r as this assumes a specific linear model from normal data.

Soil horizons

In the initial planning of this experiment both the A(0-200mm) and B(200-400mm) horizons (Macvicar *et al* 1977) were considered as potentially influencing plant quality. It is generally accepted that the majority of the rooting zone of grasses is within the top 150mm of soil but in the sour veld areas in Natal high levels of Al in the sub-soil (just below this zone) may affect plant growth and consequently seasonal changes in plant quality (N M Tainton, M V Fey, personal communications 1983). The soil analyses data (Chapter 2) were compared to see if the 200 and 400mm samples were correlated. Spearmans Rank Correlation Coefficients (see above) showed that, with the exception of P, the chemical and physical status of soils in the two horizons is highly associated ($P \leq 0.01$). These relationships are best illustrated using scatter plots (Figure 5.1) which

confirm that most of these associations are linear.

Table 5.1 Cross correlation matrix showing those variables which are not significantly correlated based on Spearman rank correlation coefficients (r_s).



1 in all cases 2 or 20 is the 0-200mm and 4 or 40 is the 0-400mm soil horizons

2 • = correlation not significant (P>0.05)

3 BLANK = $r_s \geq 0.65$

4 ECEC = effective cation exchange capacity; XA = exchangeable acidity; AST = acid saturation; OM = organic matter; FMC = field moisture capacity

As the chemical and physical status of the A and B horizons is associated only one data group needs to be used. Since the A

horizon is easier to sample, and represents the major rooting zone of grasses, the B (200-400mm) horizon data will be excluded from further analyses. Although the values for P are not correlated (Table 5.1) P values for the 200-400mm horizon will also be excluded as these are correlated with pH, Ca and Mg in the 0 to 200mm horizon.

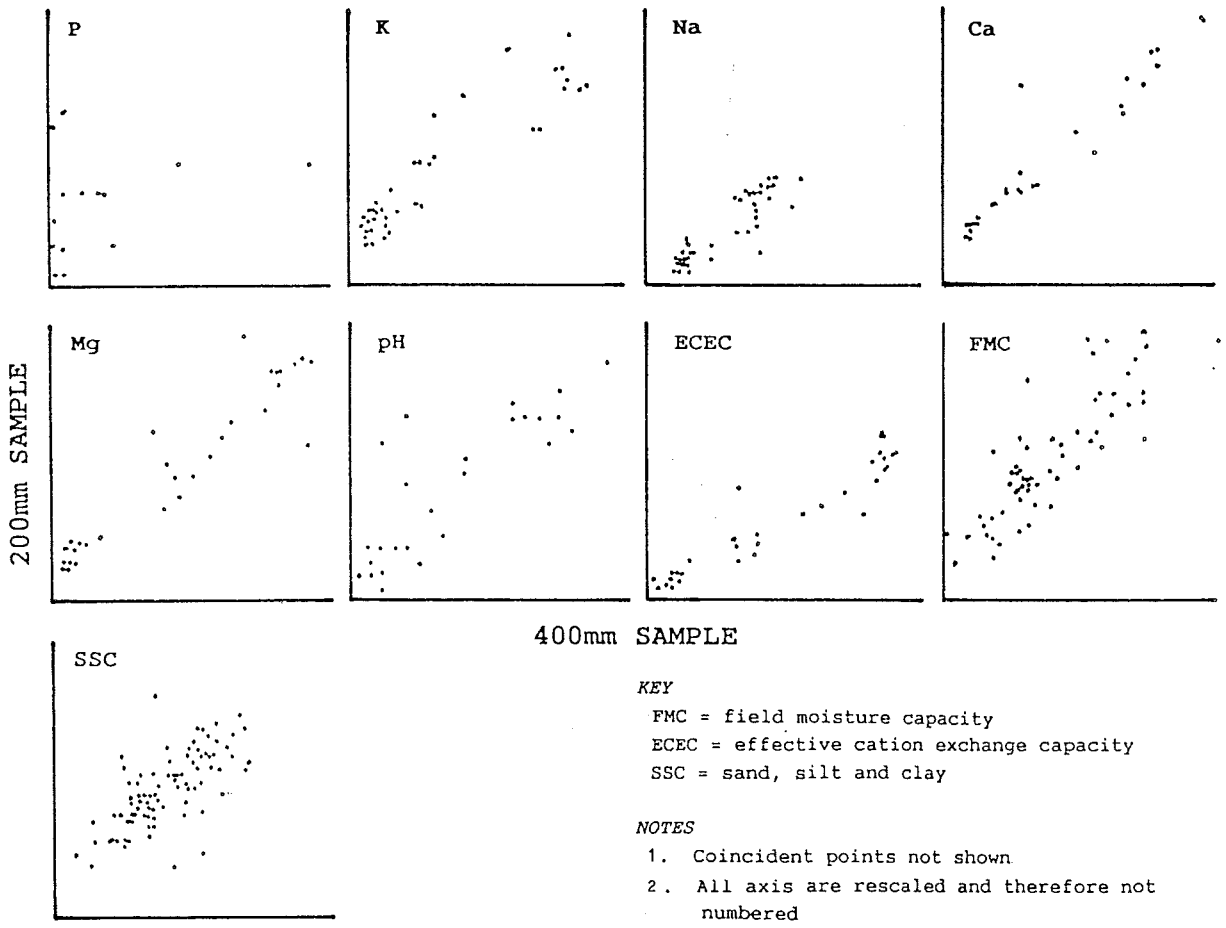


Figure 5.1 Scatter diagrams showing the association between A (0-200mm) and B (0-400mm) soil horizon chemical and physical characters at 32 locations in Natal.

Plant quality

In analysis of the plant material a total of nine parameters were measured viz; digestibility (CDMD), N, P, K, Ca, Mg, Na, Zn and S. As was the case with the soil parameters, the use of

regression techniques requires that cross correlated variables are removed. Spearman Rank Correlation Coefficients (r_s) were also calculated for these data (Table 5.2). Inspection of the cross-correlation matrix showed that digestibility (measured by CDMD, see Chapter 2) is highly correlated with N, P and K. Examination of the data (Table 5.2) will suggest that N would be the best choice to represent this group because it has higher r_s values than any of the others. It was nonetheless decided to use CDMD as the indicator variable of the group because of its relative ease and low cost of determination, which is in keeping with the objective of finding an inexpensive index of sweetness.

Table 5.2 Spearman Rank Correlations (r_s) between each of the plant variables used to index quality (data based on 217 observations from 32 sites in Natal).

	Spearman Rank Correlations							
	CDMD	N	P	K	Mg	Ca	S	Zn
CDMD	1.000[1]	.702	.670	.749	.359	.297	.391	.283
	1.000[2]	.000	.000	.000	.000	.000	.000	.000
N	.702	1.000	.870	.809	.503	.315	.364	.425
	.000	1.000	.000	.000	.000	.000	.000	.000
P	.670	.870	1.000	.771	.472	.255	.214	.442
	.000	.000	1.000	.000	.000	.000	.001	.000
K	.749	.809	.771	1.000	.450	.182	.235	.286
	.000	.000	.000	1.000	.000	.007	.000	.000
Mg	.359	.503	.472	.450	1.000	.401	.374	.317
	.000	.000	.000	.000	1.000	.000	.000	.000
Ca	.297	.315	.255	.182	.401	1.000	.439	.172
	.000	.000	.000	.007	.000	1.000	.000	.011
S	.391	.364	.214	.235	.374	.439	1.000	.117
	.000	.000	.001	.000	.000	.000	1.000	.084
Zn	.283	.425	.442	.286	.317	.172	.117	1.000
	.000	.000	.000	.000	.000	.011	.084	1.000

[1]Coefficient (r_s); [2]Significance level (P≤ value given)

The remaining plant quality variables ie Ca, Mg, S and Zn will be used together with CDMD in further analyses.

NORMALITY AND RANGE

The reliability of any statistical procedure depends on the validity of the assumptions that are made when applying the test to the data (Siegel 1956; Rayner 1967; Sokal and Rohlf 1981). As this is an exploratory investigation a range of analytical methods may be followed and it is therefore important to understand the underlying distribution of each of the variables measured here.

The data were subjected to the Shapiro-Wilk test for normality using analysis of variance (Table 5.3) (Shapiro and Wilk 1965), with the algorithm provided by SAS (Anon 1985b). By this method the W statistic is calculated and compared with an expected tabular value for a normal population. Values for the W statistic which are greater than the tabular value indicate data from a normal population.

It can be seen that only three variables have normal distributions (Table 5.3), indicating that non-parametric statistics should be favoured for this investigation (Siegel 1956).

This test for normality also provides the coefficient of variations (CV) for these data (Table 5.3). Most of the chemical and physical data have high CV's, indicating that there are wide ranges in these data. This is considered to be a desired outcome because these characters are needed to separate the sites. The high CV's shown here indicate that the soil environments of each of the sites are heterogeneous in relation to each other. The sites were selected subjectively in an attempt to cover a wide range of soil condition. In terms of that objective, the sampling strategy was therefore successful.

Table 5.3 Summary of mathematical characteristics of field data used to characterise 32 sampling sites in Natal.

VARIABLE	UNITS	MEAN	CV(%)	W	P[1]
ASPECT	D°	150.2	80.6	0.90	**
ANGLE	D°	8.8	85.3	0.90	**
ALTITUDE	m	807.4	96.2	0.76	**
LATITUDE	D°	28.5	2.7	0.88	**
LONGITUDE	D°	30.9	3.8	0.81	**
K	meq/100g	0.5	74.8	0.84	**
Ca	meq/100g	6.7	89.5	0.86	**
Mg	meq/100g	4.1	75.1	0.88	**
Na	meq/100g	0.2	92.3	0.77	**
EXCH'ABLE ACIDITY	meq/100g	0.7	135.9	0.74	**
P	mg/kg	2.0	136.7	0.72	**
K	mg/kg	213.7	74.5	0.84	**
Ca	mg/kg	1351.5	89.5	0.86	**
Mg	mg/kg	496.7	75.1	0.88	**
Na	mg/kg	35.4	92.3	0.77	**
ECEC	meq/100g	131.0	81.1	0.86	**
pH		4.7	10.7	0.89	**
FMC	g_{water}/g_{soil}	0.52	28.8	0.95	NS
SAND	%	43.7	39.1	0.97	NS
SILT	%	15.0	63.5	0.90	**
CLAY	%	41.7	36.8	0.98	NS
ORGANIC MATTER	%	4.5	42.0	0.92	*

[1] ** = P≤0.01 ie data are NOT normal

PLANT COMPONENTS

One of the objectives of designing this investigation was to quantify the variations in plant quality that are a characteristic of sweet and sour veld. As discussed earlier (Chapter 2), it was felt that separation of plant parts was essential to reliably detect these trends. One of the shortfalls of earlier studies (eg du Toit et al 1940a) was the combination of species and different plant components into a single sample. As an attempt is being made here to determine a causal relationship, care has been taken to exclude potentially

confounding factors such as inter-species differences. The same can be said of plant parts and so separation was carried out in this study.

It seems logical then that the data from the plant components should be investigated to determine which of these shows the greatest variation, as was the case for the site characteristics (see earlier). This procedure should not be seen as a deliberate attempt to 'select' the 'best' data. The objective here is to reliably quantify a relationship which is generally known to exist. In order to do this a range of sites, covering each ecological zone, were selected, and the data for each of the four plant parts compared (Figure 5.2).

Inspection of this figure shows that the TWO-LEAVES-AND-A-BUD data have the greatest variation both across and seasonally within sites. The data presented here are taken from the four sampling dates for which complete data are available. It can also be seen from the figures that the DEAD and WHOLE components have lower values (as expected) and are relatively uniform. It makes little sense to use a component that shows little variations when one is specifically attempting to explain known variation. For this reason the data for the DEAD and WHOLE components were excluded from further analysis. A review of the summary presented in Table 3.7 (Chapter 3) supports this, where the TIME by PLANT interaction of the WHOLE samples was not significant ($P>0.05$) whilst the TWO-LEAVES-AND-A-BUD samples were.

On the other hand, distinction between the TWO-LEAVES-AND-A-BUD component and the GREEN leaf blade is not that clear. There appears to be a clearer pattern amongst the former data (Figure 5.2a vs 5.2b) both within and between sites. Correlation analysis of these two sources of data confirms that the quality of the two sources is closely related with the Spearmans r_s being significant ($P\leq0.01$).

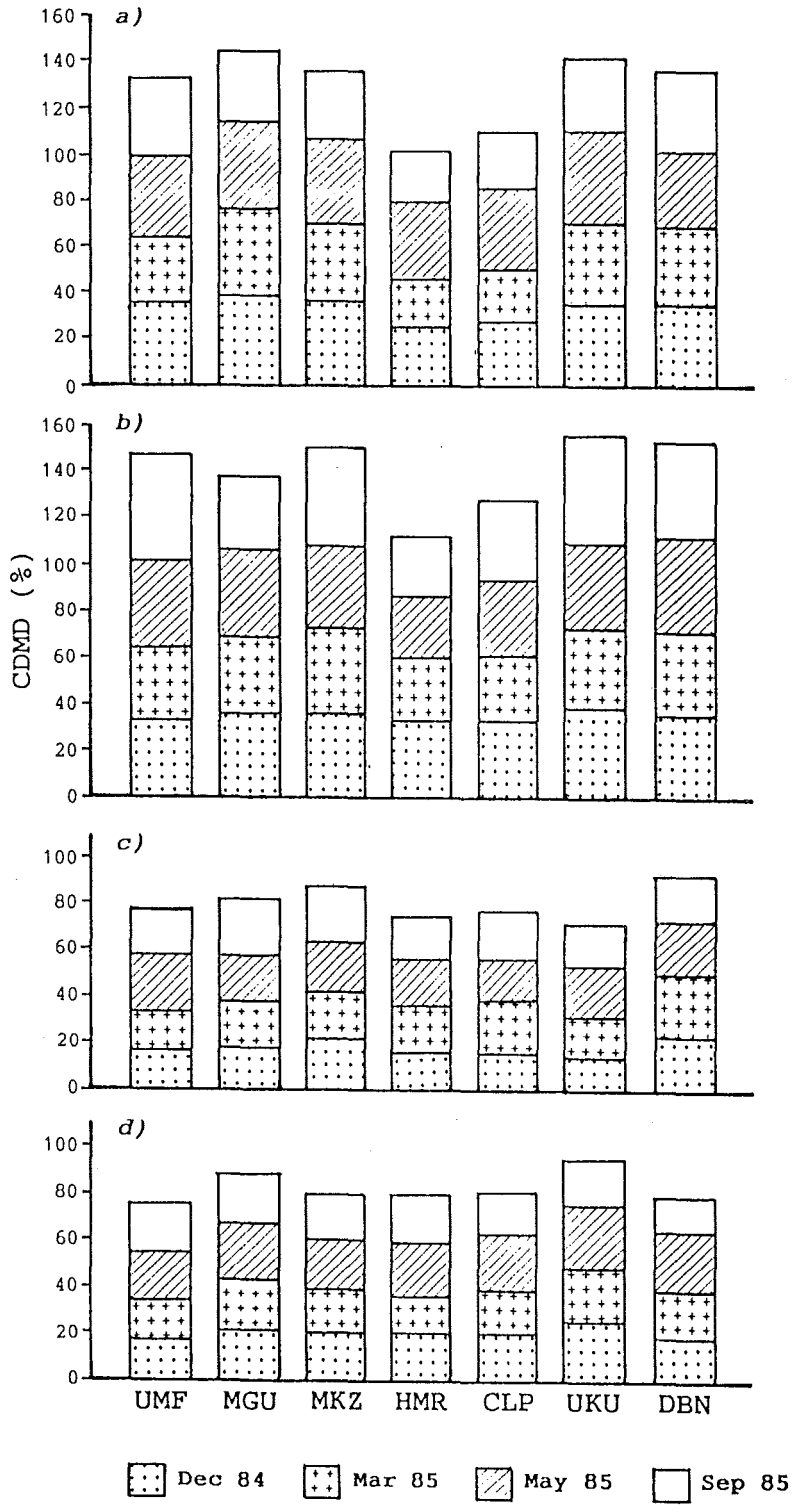


Figure 5.2 Comparison of the cellulase dry matter disappearance values (CDMD(%)) of **a)** TWO-LEAVES-AND-A-BUD, **b)** GREEN LEAF, **c)** DEAD LEAF and **d)** WHOLE PLANTS, for a range of sites in Natal (UMF = Umfolozi; MGU = Mugudu; MKZ = Mkuze; HMR = Highmoor; CLP = Cathedral Peak; UKU = Ukulinga; DBN = Durban (Kranzkloof).

On the basis of the above preliminary investigation of the plant quality data it was decided to use only the TWO-LEAVES-AND-A-BUD component for further analysis, as this shows the greatest variability with respect to the objectives of this study.

GROUPING OF SITES

As mentioned earlier (Chapters 1, 2, and 4) this study has been based on the separation of sites according to environmental characters and sites were subjectively chosen to cover a range of plant quality. At each of these sites a broad range of sites descriptors were chosen to characterise the locations, in terms of their differences, in order to sample a range of plant qualities and environmental characteristics.

The data that have been collected here have met that requirement reasonably well for the site characteristics (Table 5.3) and the plant quality data. Because of the large number of data collected, sites cannot be easily compared and so a method is needed to group the sites on an objective basis. The purpose of this grouping is; (1) to reduce the number of cases to deal with by combining sites that are similar with respect to both environmental and plant data; and (2) to test the validity of the original groupings.

CLUSTER ANALYSIS

There are a number of methods of grouping multivariate data based on some form of reduction in dimensions (Manly 1986; Jongman *et al* 1987). A useful procedure for grouping sites is cluster analysis, as one may use of number of methods for determining groups. The objective in all cases is to group a number of samples based on similarities or differences in a number of variables. The data collected in this study lend themselves to such an approach.

In subjecting these data to cluster analysis an attempt was made

to achieve the following:

1) to determine if the subjective grouping used to choose the sites could be duplicated in an objective way; and
2) to see if these data could be grouped into three groups (sweet, sour and mixed) according to the conceptual model (Chapter 4).

In order to do this the climatic data were excluded from the analyses. The reason for this was to prevent the repetition of information in the climatic data from over influencing the results (Jolliffe *et al* 1989). As each location has a limited number of weather stations (usually only 1), the climatic data are duplicated for all the sites at each location. As the cluster analysis procedure uses Euclidean distance as a measure of dissimilarity, all sites would have the same distance measure for each climatic variable. This situation would certainly result in the grouping of those sites being determined to a large extent by these variables. It seems certain that such a situation would distort any biological interpretation of the clusters as they would merely reflect the low within cluster variance for each of the four climatic variables used.

SOIL DATA

The sites were clustered on the basis of the soil data from the 0-200mm horizon (see CORRELATION, **Soil horizons** above) together with the physiographic data. These data were standardised and Euclidean distances calculated as recommended by Manly (1986) and then clustered using the furtherest neighbour criterion (Milligan 1980; Anon 1988). An initial attempt was made to determine eight clusters to reflect the sampling locations of this study (Table 5.4).

From these analyses it can be seen that there are distinct groups of sites. The separation within each location is not distinct except that the Highmoor and Cathedral Peak sites are separated from all the other groups. Included with these is one site from Kranzkloof (Durban). This particular site is located in a

remnant of 'Ngongoni veld (Acocks 1975, veld type 5)) while the other one is located in the valley bottom at Kranzkloof (DBN in Group 8). In terms of soil characters therefore it appears that it may be closer to the dystrophic humid soils of the Drakensberg. The two sites from Ukulinga are placed in separate clusters. A possible explanation is that they have different parent material, with one on shale and the other on dolerite. The clustering of the various sites from Zululand (Hluhluwe, Umfolozi, Mugudu and Mkuze) are grouped together but sites from each location are mixed across groups.

Table 5.4 Grouping of 31 sampling locations based on furtherest neighbour (Euclidean distance) from 24 environmental variables.

	GROUP							
	1	2	3	4	5	6	7	8
	[1] [2]							
SITE POSS	UMF(1) HLU(1) MGU(2)	UMF(3) HLU(1) MKZ(5) MGU(2)	MKZ(1)	HMR(1)	HMR(3) CLP(6) DBN(1)	UKU(1)	(UKU(1)	PMB(1) DBN(1)

[1] UMF = Umfolozi; MGU = Mugudu; MKZ = Mkuze; HLU = Hluhluwe; HMR = Highmoor; CLP = Cathedral Peak; UKU = Ukulinga; DBN = Kranzkloof; PMB = Worlds View, Pietermaritzburg; POSS = location

[2] number of sites from each location

On the basis of this analysis it appears that the eight groups can be combined into four broader groups. Those in Group 1 to 3; 4 and 5; 6 and 7; and then Group 8 alone (Table 5.4). In terms of the subjective classification these would represent the sweet, sour, sweet-mixed and sour-mixed respectively.

If the separation into sweet-mixed and sour-mixed is ignored we may consider the overall data to form three major groups corresponding to sweet, sour and mixed. In order to test this the cluster analysis was re-run with sufficient iterations to form three groups (Table 5.5).

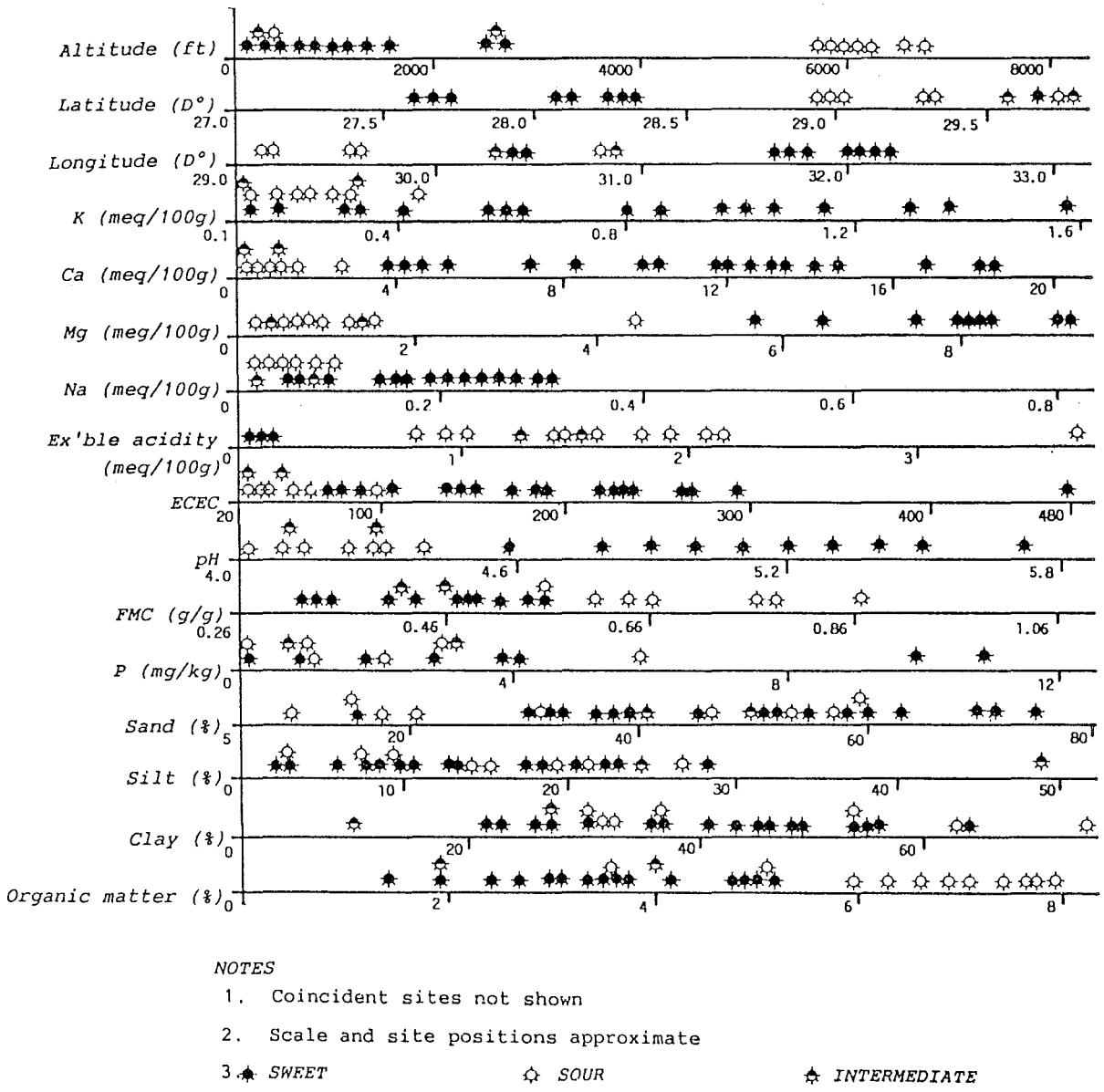
Table 5.5 Clusters from the analysis of environmental data for 31 sites in Natal using the furtherest neighbour criterion.

GROUP	LOCATION	NO OF SITES
1	Umfolozi, Hluhluwe, Mugudu Mkuze	16
2	Highmoor, Cathedral Peak	10
3	Ukulinga, Kranzkloof, Pietermaritzburg	5

Unfortunately the algorithms available do not provide a direct indication of which variables were responsible for the separation of sites into groups. This can be determined by plotting the positions of each site (by group) along a relative scale for each variable. If sites are plotted as a scatter diagram between axes representing each of two variables' scales, the degree to which each variable pair accounts for clustering can be seen. This method however does not allow comparison of the effects of all variables simultaneously. In order to facilitate this comparison a composite plot of all variables was developed (Figure 5.3).

The sites are not readily distinguished on the basis of a number of these variables (Table 5.3). On the other hand some are clearly separating the majority of sites from each group. Those that do clearly separate the groups include ALTITUDE, LATITUDE, LONGITUDE, Ca, Mg, EXCHANGEABLE ACIDITY, pH and FMC. Organic matter could also be used as the groups separated reasonably well with the exception of two sour sites and the intermediate group. The intermediate group do not show a clear pattern as far as any of these variables are concerned and share values with both sweet and sour sites across all variables.

Those variables that do show separation of the sites will be used to test the relationship between plant quality and environment (Chapter 7).



NOTES

1. Coincident sites not shown
2. Scale and site positions approximate
3. *SWEET* *SOUR* *INTERMEDIATE*

Figure 5.3 Grouping of 32 sites using furtherest neighbour (Euclidean distance) on environmental measures (position of each site is relative to all others).

PLANT QUALITY

The plant quality data were subjected to cluster analysis in the same way as the soil data (Table 5.6). In this case the separation of sites was more clear, with less overlap between groups from each location.

Table 5.6 Clustering of 31 sites in Natal based on plant quality data using the furtherest neighbour criterion (data are from seven sampling dates from July 1984 to March 1986).

	GROUP						
	1	2	3	4	5	6	7	8
	[1] [2]							
SITE	UMF(1)	MGU(2)	UMF(3)	MKZ(2)	MKZ(1)	HMR(3)	HMR(1)	UKU(1)
POSS			HLU(2)			CLP(6)		
			MGU(2)					
			MKZ(3)					
			PMB(1)					
			DBN(2)					

[1] UMF = Umfolozi; MGU = Mugudu; MKZ = Mkuze; Hluhluwe; HMR = Highmoor; Clp = Cathedral Peak; UKU = Ukulinga; DBN = Kranzkloof; PMB = Worlds View, Pietermaritzburg; POSS = location

[2] number of sites from each location

The only exception to this are the sites from Kranzkloof and Pietermaritzburg which are clustered here with sites from Zululand and not the Drakensberg, as they were in the case of the environmental data. This is then indicative of their intermediate nature. This was clarified with a further cluster analysis where these three sites were again grouped with the Zululand sites when only three clusters were formed. The three groups in this case were: Zululand and Kranzkloof with Pietermaritzburg; Drakensberg; and Ukulinga.

It must be noted that the use of cluster analysis on the plant data at this point only provides confirmation that the site characteristics and the patterns in plant quality are related in some way. It may be argued that this is inevitable because sites were chosen according to an *a priori* grouping of **sweet, sour** and **mixed.** The cluster analysis therefore provides only a corroboration of that apparently contrived selection. This is not the case, however, because the selection of sites was based on the requirement for a wide range of environmental parameters. No deliberate attempt was made to select sites in terms of plant quality profiles. Indeed this was not possible as no data were

available on which to base such a selection.

The results of the cluster analysis of the plant data is presented here because it shows very clearly that the character of the environment reflects the patterns in plant quality. Whether any driving (causal) variables can be detected remains to be tested with sufficient care to isolate cause from chance correlation. If the clusters extracted on the basis of the plant data were not similar to those from the clustering of the environmental data, there would be little point in pursuing these data further. Clearly a lack of similarity between the two analyses would suggest that none of the environmental variables measured here influenced, or could be used to determine, plant quality.

CONCLUDING REMARKS

On the basis of the foregoing investigation a number of characteristics of the original data become evident. One outstanding feature is the degree to which the variables are cross-correlated. This will allow the number of variables for further analysis to be reduced and hopefully improve the possibility of detecting any relationships that may exist between plant quality and environment.

From these investigations a sub-set of variables from the environmental and plant quality data can be used to test relationships between variables from the following groups:

1) **Environmental**

   The following site characteristics will be used for further analysis:

   a) P,
   b) Field moisture capacity,
   c) Exchangeable acidity,
   d) Ca,
   e) Mg,
   f) pH,

g) Altitude, and
h) Aspect.

Geographic location (latitude and longitude) were shown to possibly influence plant quality. However they are not included in this list because as far as these data are concerned, they reflect the geographic separation only. This feature is reflected by altitude, as shown in Table 5.1. In addition to these selected variables the climatic variables will also be added. These include the long term MEAN RAINFALL (mm), MEDIAN RAINFALL (mm), MAXIMUM TEMPERATURE (C°) and MINIMUM TEMPERATURE (C°), for each month of sampling. The median rainfall has been included here because it is likely to reflect the true amount of rainfall more closely than the mean as it is less affected by excessively high or low values as is the case for the mean.

2) **Plant quality**

The data from the plant components are best investigated using those from TWO-LEAVES-AND-A-BUD as these show the greatest variation across sites and seasons. As far as measure of plant quality are concerned the following will be used:

a) CDMD,
b) Ca,
c) Mg
d) S, and
e) Zn.

Others may be added in cases where multiple regression is not used or where auto-correlation is not important in the interpretation of the analyses.

The variables selected from both the environmental and plant data sets have been extracted from a very large amount of information. It is hoped that the complexity of the original data has not been over simplified during this process. There is a danger in missing the biological reality because of an obsession with statistical nicety. The author is well aware of this danger and does not consider the above lists to be finite. Depending on the outcome of further analyses of these data this list may be

expanded or contracted. This is likely to be on the basis of biological interest rather that mathematical rigor. In doing so I hope to maintain a reasonable level of usefulness of the results.

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CHAPTER 6

TRENDS IN PLANT QUALITY

INTRODUCTION

The measurement of plant quality that has been used in this study has relied on laboratory analyses. Quality here refers to the *usefulness of the forage to the animal*. This is difficult to measure, and analysis of elemental status and digestibility are used to index quality (see Chapter 2; PLANT). Although never specifically included in the definitions of sweet or sour veld (Scott 1947), the concept embodies palatability and maturity. It is generally accepted amongst Grassland Scientists in southern Africa that the differences between sweet and sour veld are mainly a function of palatability and acceptability of the mature forage: especially in winter. There is little doubt in my mind that this is correct. However this would be difficult to test in any rigorous way. The reason for this is the measurement of **palatability** and **acceptability**. There are many variations on the theme but Ivins (1955) provides a relatively succinct definition viz; the sum of the factors which operate to determine the attractiveness of food to animals. Included here are the factors determining the selection of food items by animals (Heady 1964; Theron 1966) or acceptability. The definitions are confusing and Bransby (1981) suggests that **acceptability** is a relative term (depending on circumstances) and **palatability** is absolute determined by physical factors of the plant itself. In any event the intake of forage by the grazing animal is influenced by both and therefore in their biological context both terms have a relative nature and are not readily measurable. Because of this the aspect of maturity has not been specifically included in this study and indices of quality are used (see Chapter 2). The objective of the study relates specifically to the relationship between quality and the environment (Chapter 1). Maturity incorporates the relationship between the animal and the plant

via its effect on palatability. As such, then, a study of maturity is beyond the scope of this preliminary investigation.

The conceptual model proposed in Chapter 4 (Figure 4.1) summarises the classical approach to separating veld with respect to plant quality, as indexed by digestibility. The trends presented for sweet and sour veld are a graphical representation of the possible extremes in quality. Quality is used here because it has a clearer definition, being *the sum of the factors influencing the absorption of nutrients by the animal* (Ulyatt 1973). Nutritive value is the true 'value' of the forage to the animal and this cannot be measured in the laboratory as it requires the use of metabolic crates. The measures of quality used in this study relate to the nutritive status of material and provides an index of the usefulness of the material to the animal. This then is a function of the chemical composition and digestibility of the forage that is measured in the laboratory. The current definitions of sweet and sour veld (Scott 1947) imply that quality of sweetveld does not vary over the year. The conceptual model suggests that the sweetveld does vary but not to the same extent as sour veld.

The purpose of this chapter is to present the data for each of the plant quality components and examine their seasonal trends within each group.

VALIDITY OF THE CLUSTERS

The presentation of results and the analyses that follow are based on the clusters of SWEET, SOUR and MIXED identified in Chapter 5. As mentioned there, the clustering of the plant data allowed the reduction in the number of cases (sites) as well as a confirmation of the groups determined on the basis of the environmental data.

It may be argued that the clustering has removed a great deal of information from the data by forcing the formation of homogeneous

units from a heterogenous data set. To some extent this is true, in so far as the separate geographic sub-regions have been lumped together (eg Cathedral Peak with Highmoor and Mugudu with Umfolozi). No attempt has been made to summarise the information so all of the original data describing the sites is still to be used to relate the environment to plant quality (see Chapter 7). In essence the clustering procedure used here amounts to a form of *post hoc* stratification as described by Rayner (1967). However, instead of using variance heterogeneity as a basis for separation, Euclidean distance has been employed. The later approach is favoured because it is non-parametric while the former is likely to be invalid as the sites were not chosen randomly *sensu stricto*.

A difficulty that some readers may have is the justification of what seems, initially at least, to be a circular argument ie:

> *the sites occurred in preconceived sweet and sour areas and the clusters have been interpreted on the basis of that preconception.*

This is not the case, however, because the groups were clustered on the basis of environmental data and only corroborated using the plant data. These two data sets are essentially independent and so the clustering procedure is considered valid.

The only exceptions to this are those sites grouped as MIXED (5 of the 31 sites). These were not separated independently by each of the two data sets. Consequently their grouping is subjective because they in fact represent those sites which did not occur in the same clusters in the analysis of both the plant and environmental data. The easiest way of solving the problem would be to generate only two clusters and deal with the extremes irrespective of the outcome. This was not done for two specific reasons:

(1) the raw data presented earlier (Chapter 5; see Figure 5.2) suggest there is a range in the plant quality data; and

(2) to do so would require that I ignore the biological intergrade.

The fact that there are some cases where the two classifications do not overlap verifies that the range that the sampling strategy was designed to capture is reflected in those data.

SEASONAL AFFECTS

The sampling of plant material for this study extended over a period of 21 months (July 1984 to March 1986). This was done to enable the investigation of any seasonal trends in the parameters of plant quality measured here. Unfortunately the sampling dates were not evenly spaced as this was not practical. However an attempt was made to sample at approximately three month intervals. The original intention was to sample at the same times over two successive seasons. Due to the unseasonal and excessive rainfall resulting in several floods over the initial period of this research, a five month gap occurred between the first and the second harvests. In the event seven sampling trips were undertaken (Table 6.1) (see also Chapter 5; **TREATMENT OF MISSING DATA**). Each field trip lasted approximately two weeks and this was determined by the time required to complete each harvest. As a result of the unforseen circumstances only two sampling dates coincided across years (Table 6.1) so the data base on which to detect inter seasonal differences is shallow.

Table 6.1 Sampling dates on which plant material was harvested.

DATE	JULY 1984	DEC 1984	MAR 1985	MAY 1985	SEP 1985	DEC 1985	MAR 1986
TIME (months)	1	6	9	11	15	18	21

It is felt, by the author, that in spite of this such a comparison is necessary as the development of a model describing seasonal trend in plant quality is dependent on a reasonably

consistent trend between years. If no between season differences are detectable this would justify grouping the data and rearranging them into a single chronological sequence of twelve months.

The two sampling dates that do coincide were subjected to analysis using Spearman Rank Correlations (Table 6.2). The data for N, P and K were excluded from this analysis because these are correlated within periods (see Chapter 5, CORRELATION **Plant quality**). The diagonal of this matrix (Table 6.2) is the main component of interest here as it reflects the rank correlations between each of the quality variables with themselves one year later. With the exception of CDMD, S and Zn for the March data there is little evidence to suggest that the data are consistent across the seasons. The r_s values for Mg and Zn in December are significant but they are only weakly correlated. On the basis of this analysis it was decided not to lump the data from the two seasons but to treat them separately. The data presented here suggests that autumn (March) quality may be more consistent then that of the summer (December).

The balance of the r_s values suggest that the trends between seasons are not consistent. This will be investigated later (see **SEASONAL TRENDS**)

The lack of consistency between years is unfortunate as it precludes the development of a model describing the season trends. If the data from the two seasons were highly correlated it would allow the development of the model on the first seasons data and the second to test the model. This will be discussed more completely in Chapter 7.

SEASONAL TRENDS

Before an investigation of factors determining seasonal trends can be undertaken it is necessary to determine if any seasonal trends do exist. In terms of the general application of the

STATISTICAL CONSIDERATIONS

One of the problems in designing experiments in a field situation is that replication (*sensu stricto*) is often not practicable. Simply one cannot duplicate ones treatments (in this case environmental conditions). This then often precludes the use of powerful statistical procedures such as analysis of variance to compare sites. Even at one location two plots are not strictly replicates because they do not have the same 'treatment'; eg aspect and altitude may be different.

It may be argued that this is not the case as the interpretation of replication depends on the objectives of the study (G P Y Clarke, personal communication 1983). In this case the site is the treatment and this can not be replicated as its description includes the unique combination of soil chemistry and physiography which are relatively site specific. The analysis of variance approach would be valid if the intention here was to determine if the locations were significantly different with respect to their measures of plant quality. It is clear, based on the known characteristics of sweet and sour veld, that they are different. What is of interest here is why they are different. The gradient analysis approach is likely to be more useful. For this reason the results are presented in graphical form with a mean for each group (SOUR, SWEET OR MIXED), together with twice the standard error (SE) of the mean. Straight lines join successive points.

This approach is similar to that of Hunt (1982) and allows convenient graphical representation of the data. Unfortunately the few data points per year does not allow a regression approach and 95% confidence intervals to be used reliably. The approach of Hunt (1982) relies on a large number of closely spaced points to fit a trend to the data (on a time scale) but these are not available here. A further point is that the apparent differences between seasons also makes the development of a reliable equation from these data unlikely. The use of two times the variance (SE

The data for CDMD (Figure 6.1) shows distinct differences in the pattern throughout the year. The SWEET and the MIXED group vary from about 32 to 42 percent whilst the SOUR group ranges from about 21 to 38 percent. Comparison of the data values for the two December harvests shows that the three groups had similar values in the first year but not in the second. The SOUR group having a much lower CDMD value, then the other two groups, during the second year. Closer inspection of the error bands (for SOUR) shows they do not over lap between December 1984 and 1985 and so the values for CDMD are likely to be significantly different. The error bands from the December harvests of the SWEET and MIXED groups are overlapping between years but are both fairly broad. This suggests that they may not be significantly different but both groups show wide differences between harvest times within years. The mean value for the SWEET group was about 37% in the first year and 39% the second and these are not likely to be biologically different. For the MIXED group the difference is 8% CDMD with fairly wide ranges at each date (ie 6 and 10 units in the first and second years respectively).

The same pattern is not reflected for the March data as the error bands for the SWEET and MIXED at these times are just overlapping (Figure 6.1), suggesting they may not be significantly different. In the summer of the second year the values for CDMD for both groups is marginally higher the in the first year. In the case of the SWEET group the inter-seasonal differences is nearly 10%. The data for the SWEET group generally have narrower ranges than either the SOUR or the MIXED group so these differences are likely to be significant.

Despite the inconsistency between years in these data it is clear that there are seasonal patterns in plant quality and so they support the conceptual model. The differences between years is disappointing because they invalidate any attempts to fit a model to these data which will allow one to predict plant quality at any given time of the year. However the SOUR group were consistently lower in value than the SWEET group, with the MIXED

changing its position in the order of the three groups, but was much closer to the sweet then the sour group.

NITROGEN

The pattern in the nitrogen data show similar trends to those discussed for CDMD (Figure 6.2). In the case of the data for the

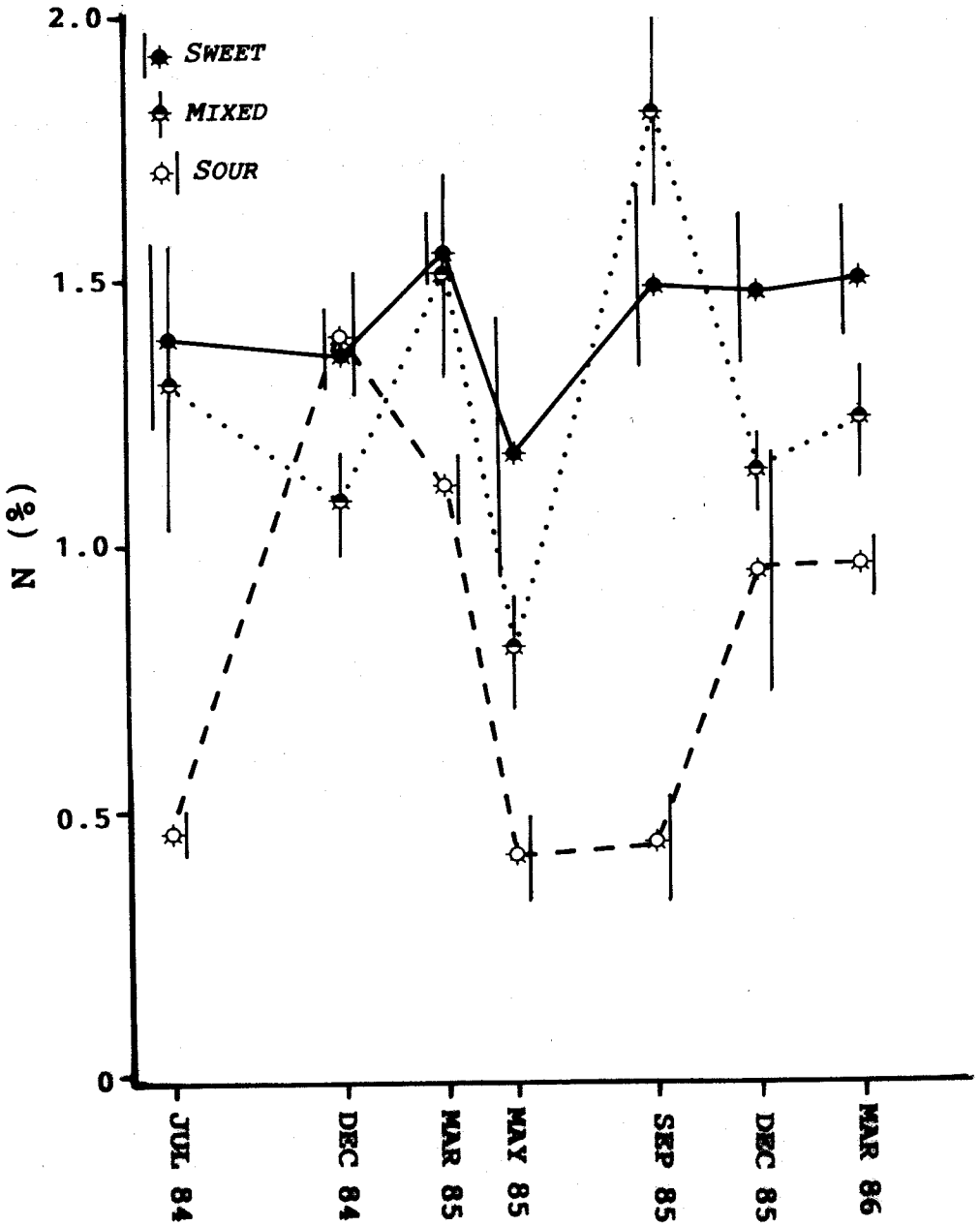

Figure 6.2 Seasonal trends in Nitrogen (%) for the three groups of sites in Natal (vertical bars represent ± 2SE).

SWEET and MIXED groups the inter-seasonal differences are not as marked as they were for CDMD (Figures 6.1 and 6.2). On the other hand the SOUR group showed narrow ranges within dates but the inter-seasonal difference is again evident. With the exception of the December 1984 data the SWEET group is consistently higher in N value then the SOUR group, as expected. In the second season the means of the two groups are likely to be significantly different whereas in the first season they were not (yielding approximately the same mean value of 1.4%N) in December 1984.

Again all three groups showed the highest values in early spring and summer, with a decline in autumn and winter. The SOUR group declined to under 0.5%N, with SWEET group retaining N levels above 1.1% over the study period. Also in a similar fashion to the CDMD data, the SWEET and MIXED groups apparently increase in quality earlier in the season then the SOUR group. The trend for the MIXED group is more consistent for these data, with this group being more clearly intermediate between the SWEET and SOUR groups. In addition to this, these data are less variable then those for CDMD. This suggests that N status may be a better measure of plant quality then CDMD for the objectives of this study.

PHOSPHORUS

The data for P (Figure 6.3) are almost identical to those of N and the description of the data will be the same and so will not be repeated. The only difference between the two measures are the nature of the separation of the groups. In the case of the data for P the groups separate out more clearly, with lower error bands at each sampling date. This was not the case for the nitrogen data. Again there was a difference between seasons which was more marked for December than for March.

In the SWEET group values declined only slightly in autumn (March/May), with values being relatively constant throughout the year. In the case of the MIXED group the data are inconsistent

and vary from point to point with no consistent trend. This group showed the same range (varying from 0.04%P to 0.15%P) as the SOUR group which range from 0.01%P at the lowest level (July 1984) to 0.1% in December 1984.

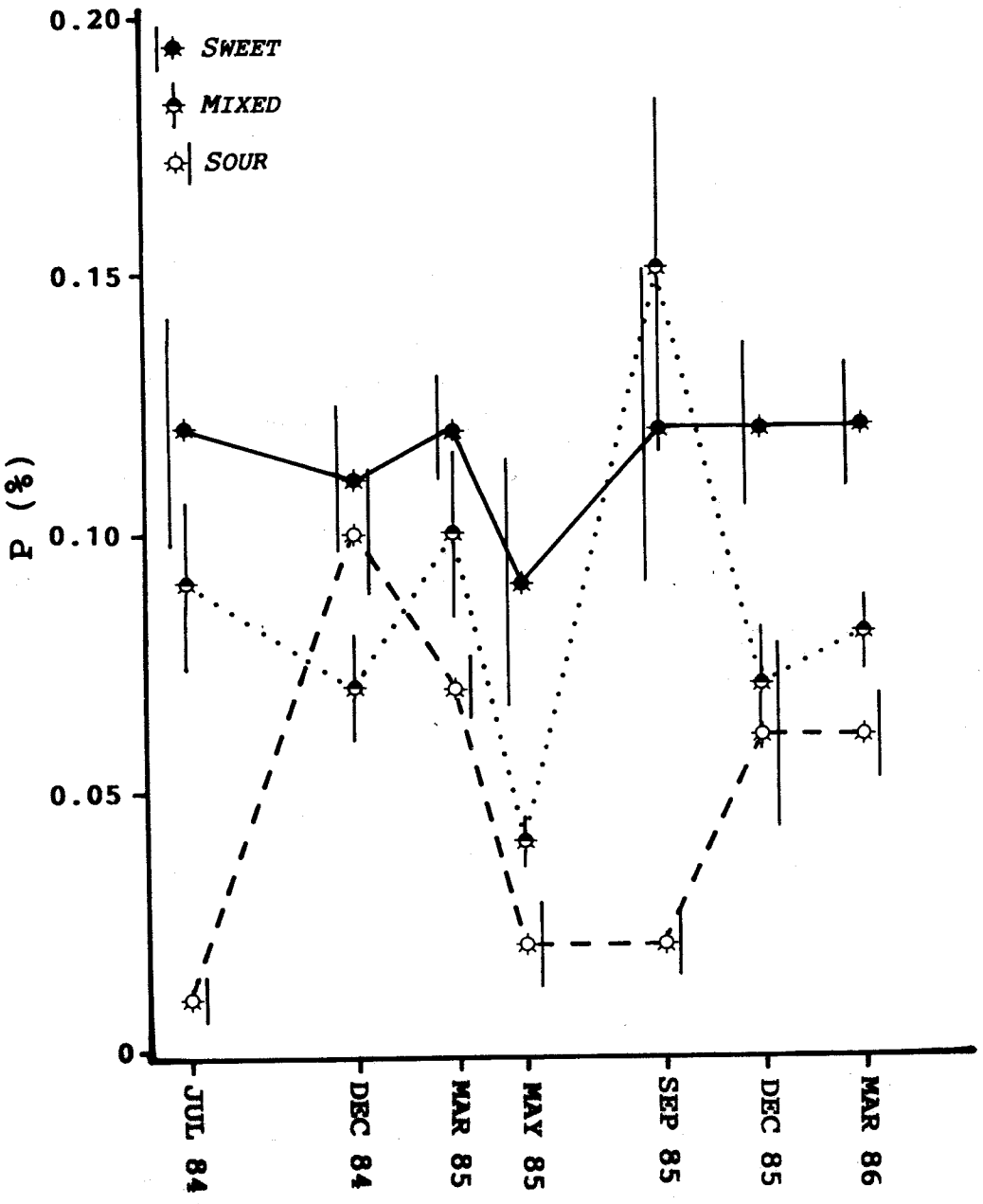

Figure 6.3 Seasonal trends in Phosphorus (%) for the three groups of sites in Natal (vertical bars represent ± 2SE).

POTASSIUM

The data for K (Figure 6.4) do not show any consistent trends amongst the groups and therefore do not support the conceptual model. The SWEET group generally increased in K levels over the sampling period. In the first period the levels for this group increased from December to March whereas they declined over this period in the second year. Both the SOUR and the MIXED groups

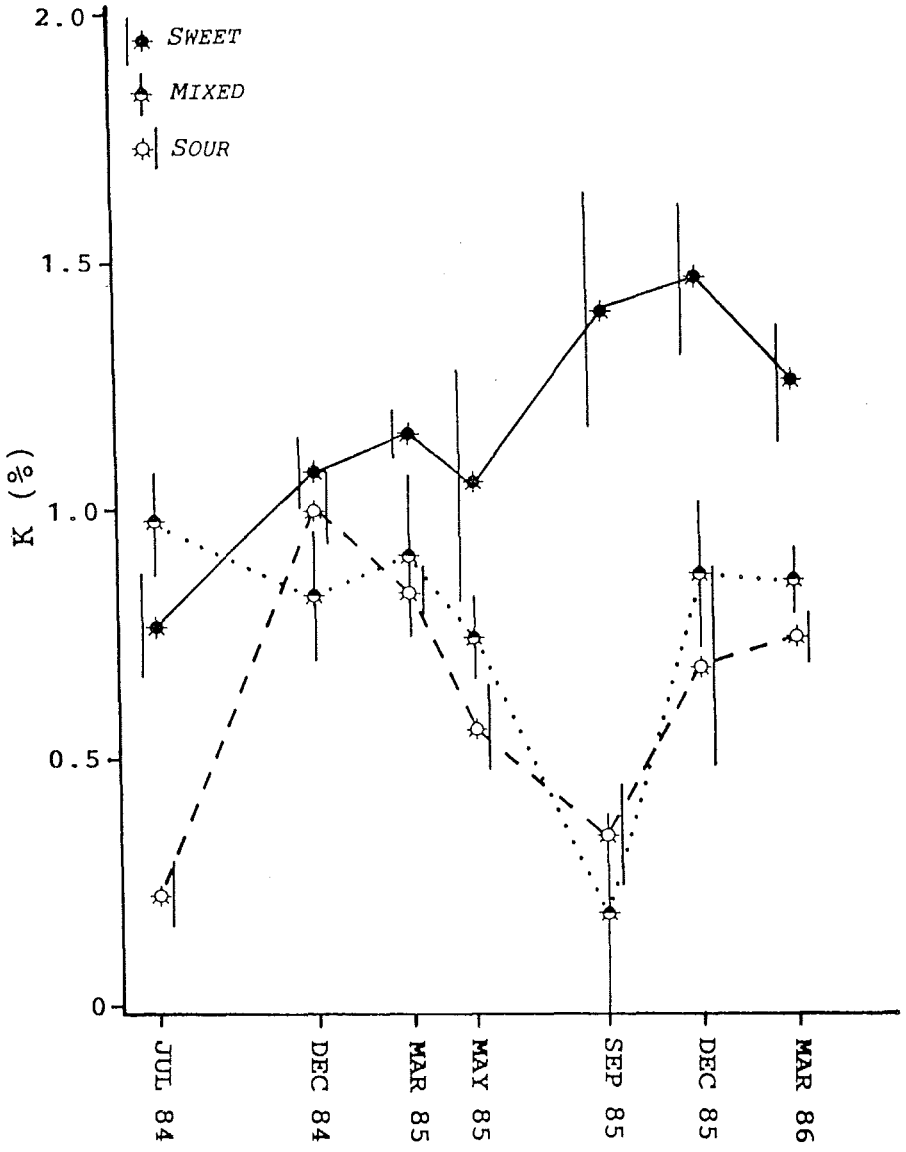

Figure 6.4 Seasonal trends in Potassium (%) for the three groups of sites in Natal (vertical bars represent ± 2SE).

decreased into the 1985 winter (March to May) whilst the SWEET group increased for the same period. As a result of this little can be concluded from these data other than that the K values for the SWEET group were consistently higher then the SOUR group.

MAGNESIUM

For magnesium the trends in the data conform relatively closely to those expected as far as the SWEET and SOUR groups are concerned (Figure 6.5). An interesting point there is that the

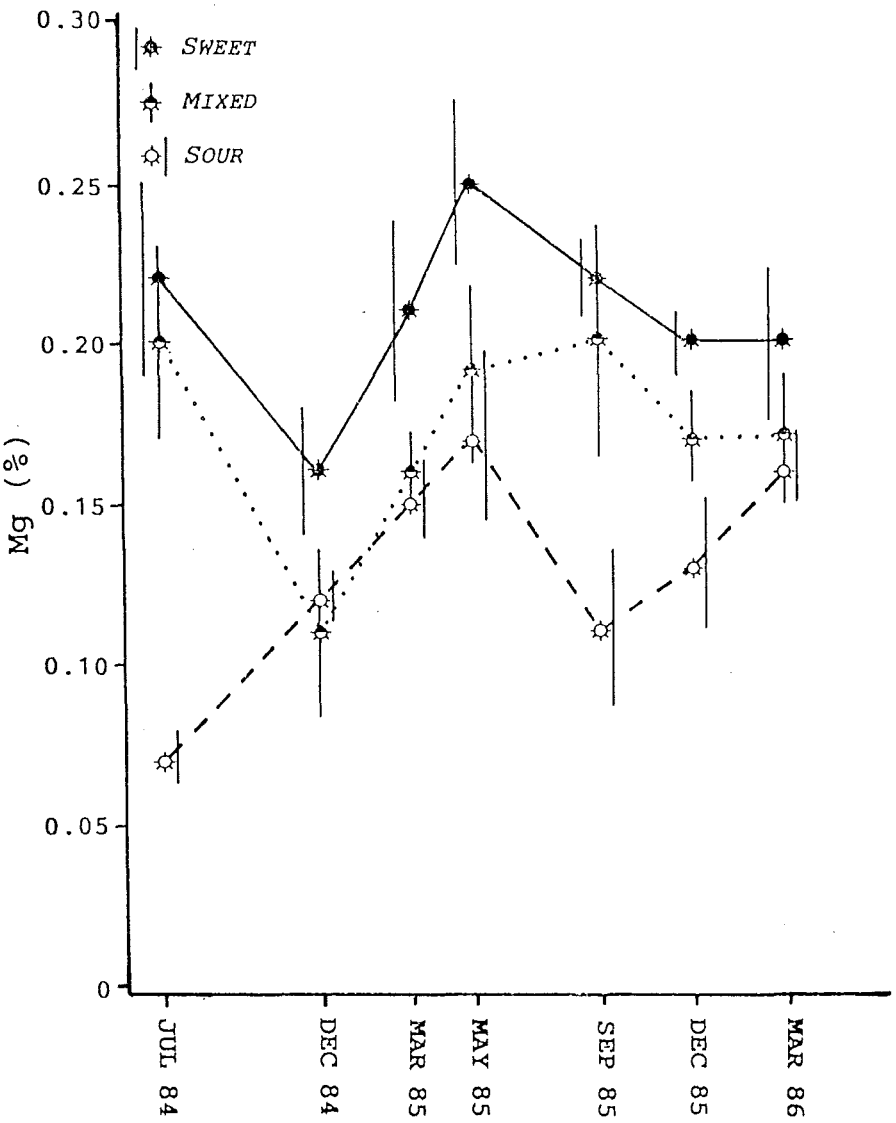

Figure 6.5 Seasonal trends in Magnesium (%) for the three groups of sites in Natal (vertical bars represent ± 2SE).

Mg levels appear to increase into autumn (May) and they decline through winter to spring. The increase Mg levels takes place later in the year than for the other elements considered so far. For the SWEET group the May 1985 levels are highest (0.25%Mg) with July 1984 highest for the MIXED group (0.20%Mg) whilst the SOUR group has the lowest level 0.07% in July 1984.

CALCIUM

The Ca levels recorded in the material showed a variation over a fairly narrow range in all three groups (Figure 6.6). For most

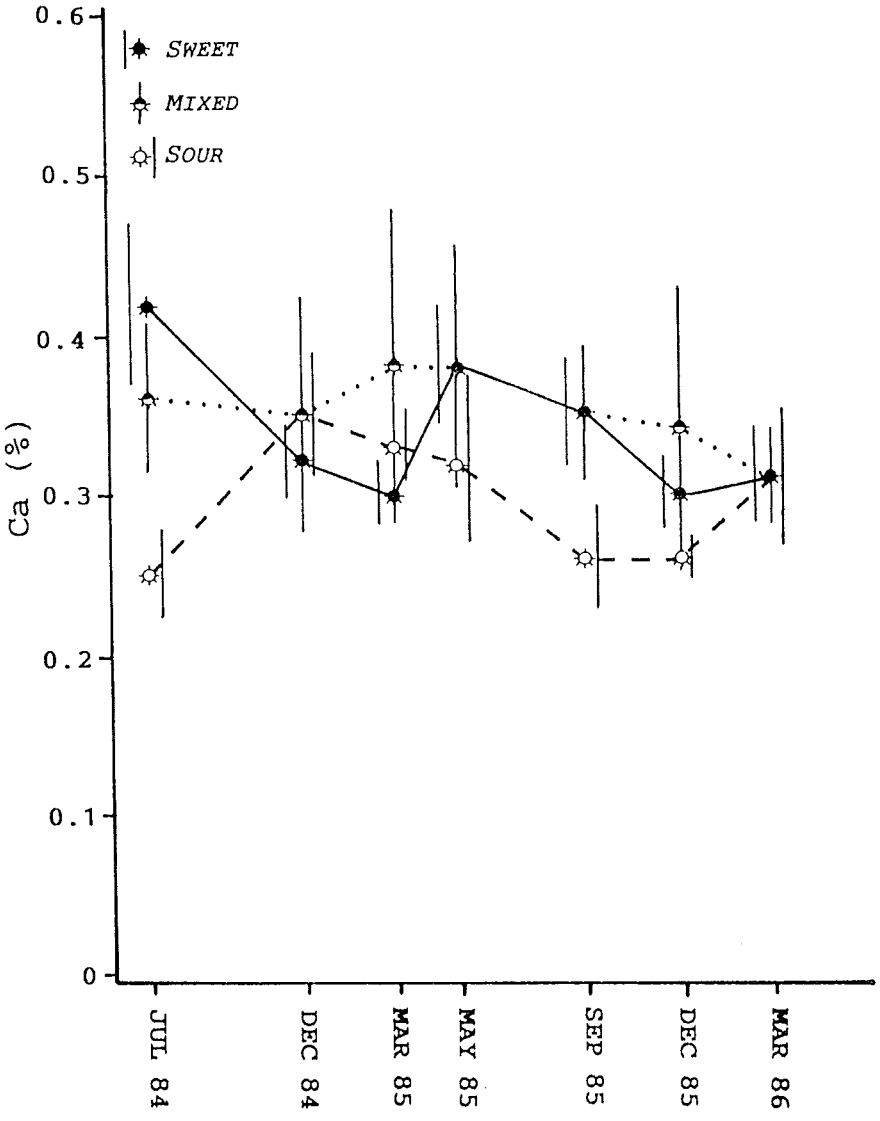

Figure 6.6 Seasonal trends in Calcium (%) for the three groups of sites in Natal (vertical bars represent ± 2SE).

of the sampling periods there appears to be little differences between the three groups. The trends, if any, for each group are not clear with the SWEET group having lower levels then the SOUR group during the first summer but not the second. The MIXED group shows the smallest variation, ranging from 0.38% to 0.32% from May 1985 to March 1986.

SULPHUR

The pattern in the S data is generally similar to that of Calcium. The MIXED group is consistently higher then either the SWEET or the SOUR sites (Figure 6.7). It must be noted though that these data are very variable and the differences reflected are not likely to be significant. The SOUR group has the least variable data and has a relatively consistent seasonal trend. However S levels for this group are lowest in winter and spring

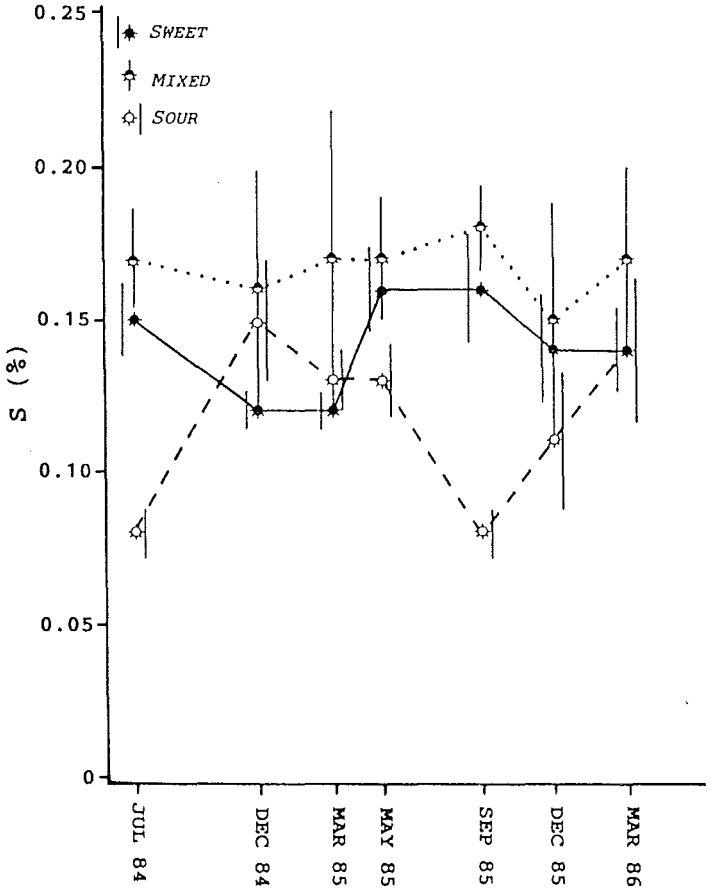

Figure 6.7 Seasonal trends in Sulphur (%) for the three groups of sites in Natal (vertical bars represent ± 2SE).

and highest in mid-summer (December) whilst in the MIXED and SWEET groups have low values in mid summer and relatively high values in spring and winter. Relative to the other elements discussed so far the SWEET group are occupying the position usually taken by the MIXED group.

ZINC

Another element that yielded variable and inconsistent data was Zn (Figure 6.8). The levels of Zn for the SWEET and SOUR group shows some weak trends with the SWEET group marginally higher then the SOUR group. This trend is at best weak as inspection of the error ranges will show. It is unlikely that the levels are significantly different across sampling times for those data, and the differences between groups is also only marginally significant.

As was the case for P, the Zn levels for the MIXED group showed no consistent behaviour. The total range of these data is very small for all groups, with the means varying from 25mg/kg to 13mg/kg. It is likely that the methods of measurement are too insensitive to detect what appear to be relatively minor variations in Zn between sites.

CONCLUSIONS

The data presented in this chapter do not provide a clear and consistent pattern for changes in plant quality. There is however a pattern for most elements within years but this is not clearly shown because the sampling took place across three years. This was compounded by the fact that only two sampling occasions (December and March) coincided across years. There appears to be a significant difference in levels of certain elements of quality of one year to the next and this does not allow the development of a simple model at this stage. This can only be more fully addressed if more frequent harvest dates are used over a longer period. The use of means from clustered sites is also

a limitation but it was felt that the presentation of 31 separate sites would have been far too confusing to interpret clearly.

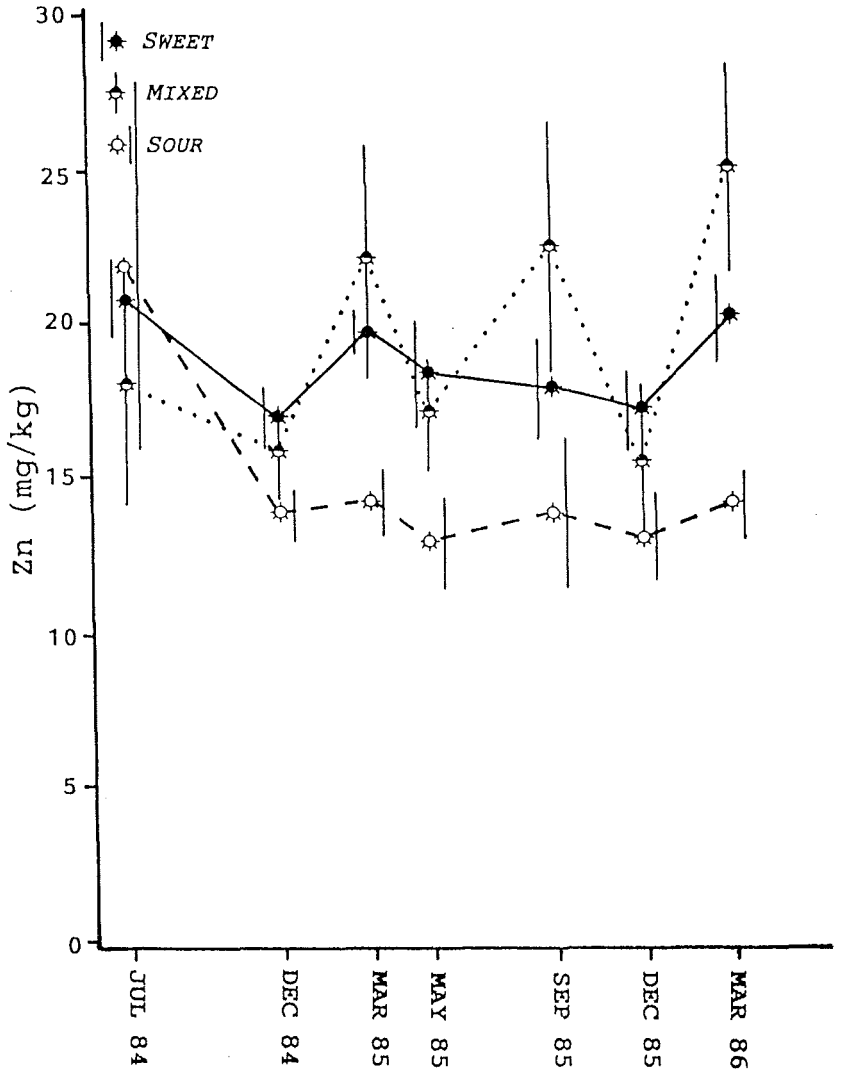

Figure 6.8 Seasonal trends in Zinc (mg/kg) for the three groups of sites in Natal (vertical bars represent ± 2SE).

In essence there are clear differences between the groups and the inconsistent behaviour of the MIXED group explains its poor performance during the cluster analysis. The trends of this group are not distinct but generally it lies between SWEET and SOUR. From this it can be concluded that the conceptual model is basically sound and the differences between the various groups can be quantified. The data suggest that the winter period is the period of greatest difference and so any attempt to detect

a relationship between environment and plant quality should concentrate on those periods where differences have been shown. In this way any variation within groups is unlikely to confuse the interpretation of results because the values are separated by relatively large margins. One point in reviewing the trends here (Figures 6.1 to 6.8) is that the increase or decrease in each quality factor does not take place at the same time. Also the time of increase or decrease is not consistent across groups. This is clearly a function of the sampling interval and it suggests that the switching mechanism may be triggered by some environmental variable at each site. This could only be detected with an intensive long term study.

When considering these data in relation to the environmental variables there is a distinct difference between the SWEET and SOUR groups. This is supported by the results of the cluster analysis. The next Chapter will attempt to determine which of these characters are related to the trends of plant quality across all sites.

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CHAPTER 7

THE RELATIONSHIP BETWEEN ENVIRONMENT AND PLANT QUALITY

INTRODUCTION

The trends over the season that were described (see Chapter 6) for each of the plant quality components showed clearly that quality varies at different locations throughout the year. In Chapter 5, sites were clustered according to both environmental and plant characteristics. The analyses of the data so far have identified a great deal of cross-correlation amongst these variables and this has resulted in a reduction in the number of variables to deal with. One aspect that has not been addressed so far is the component of time. In order to approach this study on a broad scale the description of the physical environment was considered as static. Only the climatic data used here incorporate time as they are the long term records for each sampling date. The soil data were determined from a single set of samples collected in July 1984, the assumption being made that their values would not vary with season (see Chapter 2). One of the reasons this date was used to collect these samples was because mid-winter was perceived to be the time when greatest differences between sweet and sour veld would be apparent. As little is known of the seasonal trends in soil chemistry there was, in any event, little option. For practical and financial reasons it was not possible to determine the seasonal variation of soil chemistry during this study.

The author is not aware of a suitable technique that will incorporate an analysis of the time dimension adequately for these data. Time series analyses could be used but this usually requires an even and relatively short time interval. The data describing each of the sites have therefore not been collected in a suitable manner to apply this technique.

In order to keep the amount of analysis within reasonable limits it was decided to analyse only one time period. As the data for July shows the greatest differences between groups (Chapter 6), this period was chosen for the investigation of factors determining plant quality.

The purpose of this chapter is to present the results of regression analyses between plant quality variables and the environment.

REGRESSION ANALYSIS

The most convenient method of describing the relationship between variables is to use regression analysis. A single dependent parameter can be easily regressed with a number of independent variables using stepwise multiple regression. Each of the plant quality variables was subjected to step wise regression. The independent variables used included those identified in Chapter 5, together with the climatic variables.

Prior to the analysis all the independent variables were subjected to Spearmans rank correlation to detect the presence of correlations between the environmental variables and climate. This was necessary at this stage as the climatic variables had not been included in any of the previous tests for correlation (due to them being repeated at each location). The climatic variables used were;

1) MEAN RAINFALL (mm),
2) MEDIAN RAINFALL (mm),
3) MAXIMUM TEMPERATURE (C°), and
4) MINIMUM TEMPERATURE (C°).

When grouped with the other environmental variables, Spearmans rank correlation showed both temperature measures to be highly correlated with other environmental variables, in particular ALTITUDE. As ALTITUDE is one of the test variables identified in Chapter 5, models which included temperature with altitude were rerun.

DIGESTIBILITY

Cellulase dry matter disappearance (CDMD) was found to be significantly correlated with ALTITUDE, SOIL CALCIUM, FIELD MOISTURE CAPACITY and both MEAN and MEDIAN rainfall. The linear model developed took the following form:

$$CDMD = 46.0592 - 0.00174ALTI + 0.278Ca - 10.154FMC + 0.475JMDR - 0.696JMNR$$

where

CDMD = cellulase dry matter disappearance (%),
ALTI = altitude (ft),
Ca = Ca in 0-200mm horizon (meq/100g),
FMC = Field moisture capacity (kg_{water}/kg_{soil}),
JMDR = Median rainfall in July (mm), and
JMNR = Mean rainfall in July (mm).

The model was highly significant ($P \leq 0.001$) and accounted for a large proportion of the variance ($R^2_{adjusted} = 0.87$). Analysis of variance of the model yielded an $SE_{estimate}$ of 2.458%.

NITROGEN

Nitrogen was found to be negatively correlated with the level of P in the A horizon and positively with the maximum temperature in July. The model fitted to the data for all sites had a $R^2_{adjusted}$ of 0.83 with an $SE_{estimate}$ of 0.211%N. All coefficients were significant ($P \leq 0.01$). The equation was:

$$N = 0.142JMAX - 0.041P - 1.717$$

where

N = Nitrogen content (%)
JMAX = Mean Maximum temperature for July (C°) and
P = P content of 0-200mm horizon (mg/kg)

Although this linear model tested to be highly significant a plot of the predicted versus actual value suggested a minor deviation

from linearity for high levels of N (Figure 7.1).

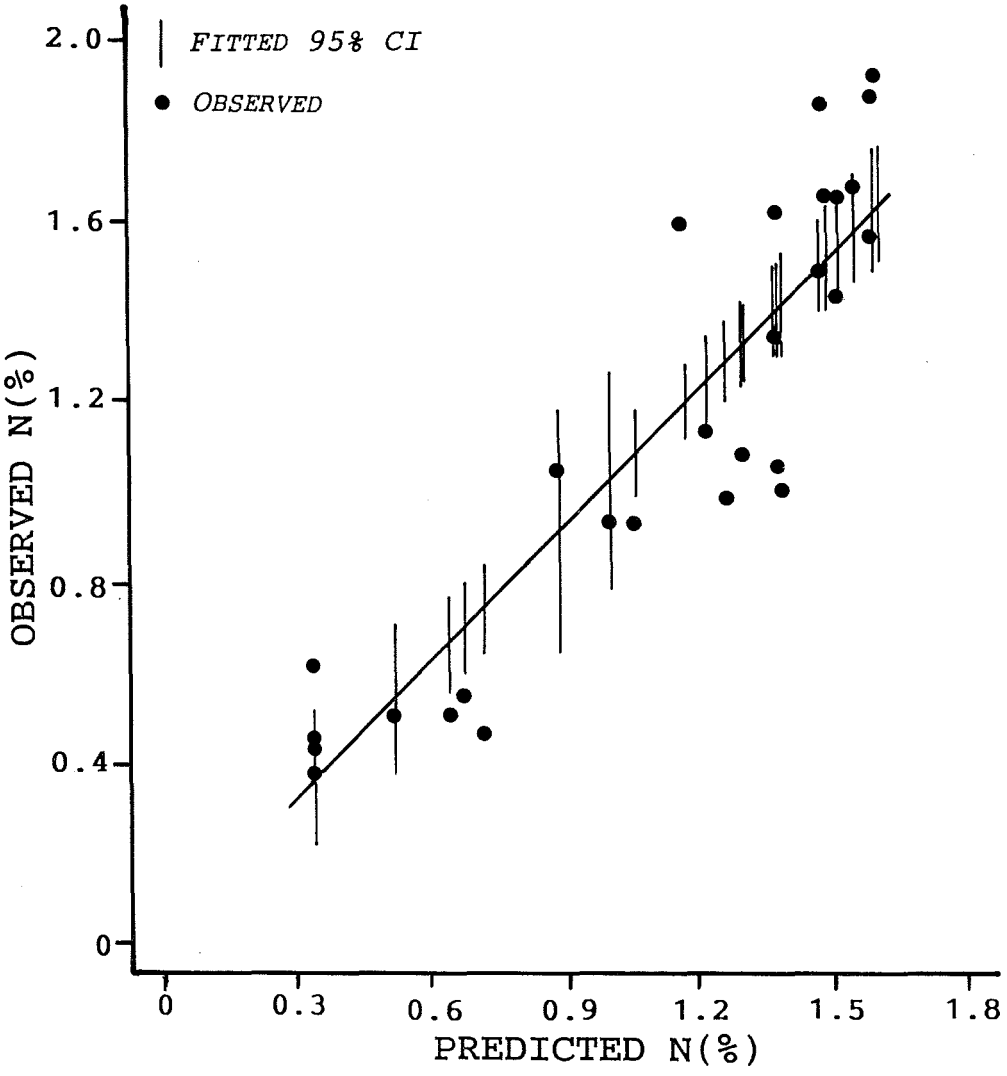

Figure 7.1 Plot of predicted versus observed values for model estimating Nitrogen from maximum July temperature and soil P.

PHOSPHORUS

The level of P in the TWO-LEAVES-AND-A-BUD material was shown to be correlated with a number of environmental variables. The stepwise multiple regression procedure included the following in the model:

$$P = 0.184 - 1.5\times10^{-5}ALTI + 0.011Ca + 3.57\times10^{-4}ECEC + 4\times10^{-3}JMDR - 7.1\times10^{-3}JMNR$$

where

P = P in plant tissue (%),
ECEC = Effective cation exchange capacity, and
other symbols as for CDMD as above.

The model has similar variables to those for CDMD (see above) and confirms the correlation between these two quality components identified earlier (see Chapter 5). The model presented for P has an $R^2_{adjusted}$ of 0.82 and an $SE_{estimate}$ = 0.024%P.
The plot of predicted versus actual data showed the model to fit the linear relationship closely.

POTASSIUM

The level of K in plant tissue was also found to be a function of ALTITUDE and RAINFALL. The model and its coefficients were all highly significant ($P \leq 0.001$) and yielded an $R^2_{adjusted}$ = 0.87. The equation describing K levels in plant material is:

$$K = 1.398 - 1.29 \times 10^{-4} ALTI - 0.035 JMNR + 0.022 JMDR$$

where

K = K in plant material (%), and
other symbols are defined above.

The $SE_{estimate}$ was calculated as 0.120%K which is relatively high given that the data ranged from 0.1 to 1.2%K. Despite this the observed and predicted data followed a linear 1:1 fit very closely.

MAGNESIUM

The Mg level in the plant samples collected in July was found to be significantly ($P \leq 0.001$) related only to MAXIMUM TEMPERATURE but the $R^2_{adjusted}$ was only 0.65. The model determined by stepwise regression was:

$$Mg = 0.019 JMAX - 0.226$$

where

Mg = Mg level in plant material (%), and
other symbols as before.

The plot of observed and predicted was disappointing with a high degree of deviation from the 1:1 line (Figure 7.2). The result of this figure suggests that the model is not likely to be very meaningful despite being statistically significant.

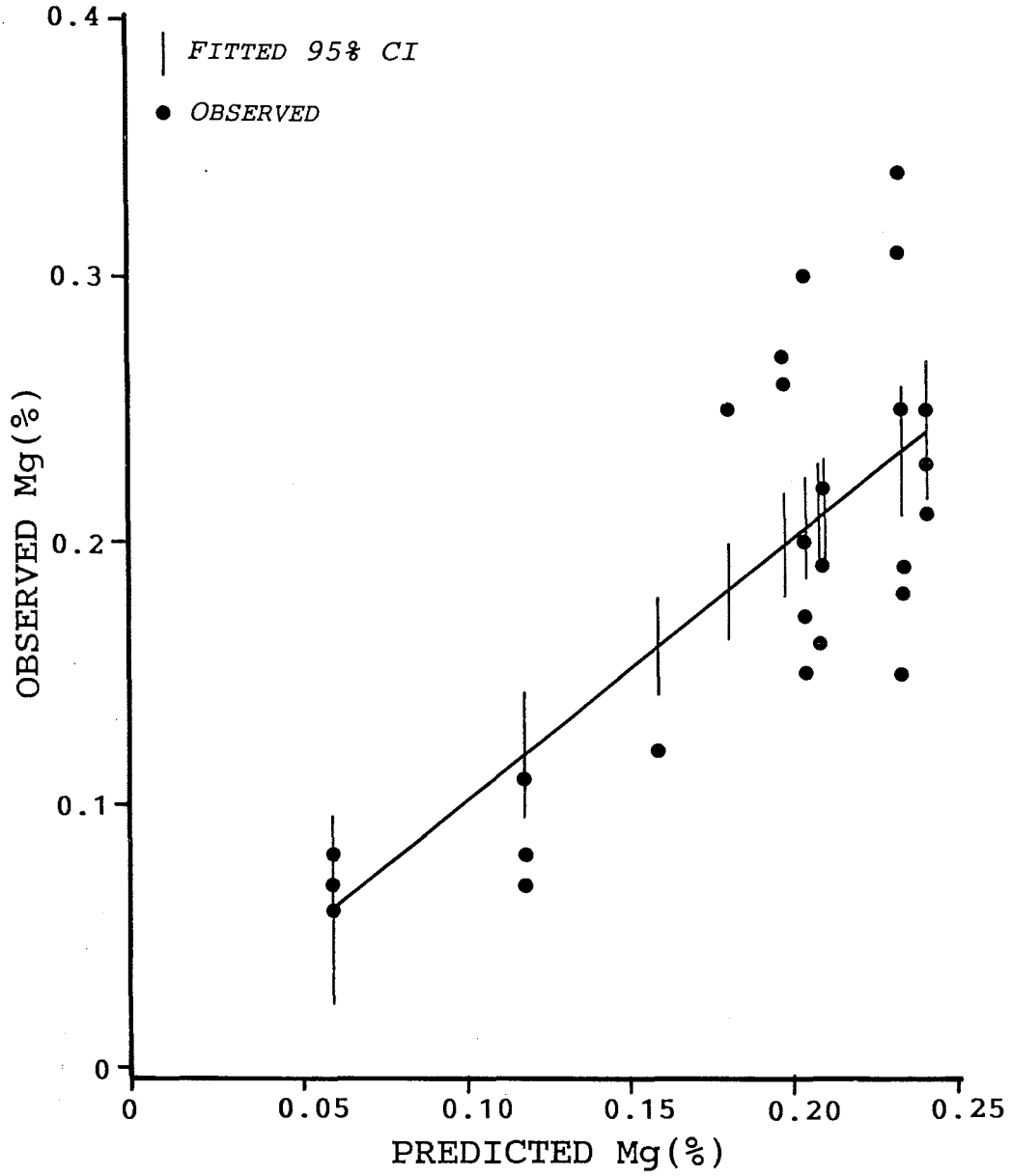

Figure 7.2 Plot of predicted versus observed values for model predicting Magnesium from maximum temperature in July.

CALCIUM

The level of Ca in the plant material was shown to be related to ORGANIC MATTER and MAXIMUM TEMPERATURE in the following form:

$$Ca = 0.0329ORG + 0.0385JMAX - 0.575$$

where

Ca = amount of Ca in plant material (%),
ORG = percent organic matter in the A horizon, and
other symbols are described above.

This was one of the poorer models with an adjusted R^2 of 0.63. Although significant ($P \leq 0.01$), the observed and predicted values tended to deviate at upper range of the data.

SULPHUR

The development of a model relating the level of S in plant material to the environmental variables included ALTITUDE, Mg in the soil, pH and EXCHANGEABLE ACIDITY together with MEAN RAINFALL. All coefficients were significant with the function as follows:

$$S = 0.707 - 1.9x10^{-5}ALTI + 7.3x10^{-3}Mg - 0.022XA - 0.105pH - 2.17JMNR$$

where

S = level in S in plant tissue (%),
XA = exchangeable acidity (meq/100g),
pH = pH of soil in 0-200mm horizon, and
other symbols are described above.

The model has a $R^2_{adjusted} = 0.77$ with the observed and predicted values very closely related.

ZINC

The poorest model that was produced here was that for Zn. It was the only element where ASPECT was included in the model. The equation generated was:

$$Zn = 10.986 + 0.022ASP + 8.95x10^{-4}ALTI - 0.99Ca + 2.61Mg$$

where

Zn = level of Zn in the material (%),
ASP = aspect measured in degrees (D°), and
other symbols as before.

The analysis of variance of this model showed it to be highly significant ($P \leq 0.01$) but with an $R^2_{adjusted}$ of only 0.33. This suggested that only 33% of the variance in the Zn data is accounted for by this model. It is unlikely that such a model will be of any practical value.

The inclusion of aspect in the model is interesting since this is the first time aspect has featured in any of the analyses so far. Aspectal differences are a well known phenomenon and it was expected that these would have been detected in a number of the analyses.

CONCLUSIONS

The models that have been presented in this section have shown some interesting features. A large number of them have included ALTITUDE and this suggested that the overall structure of the environment in Natal is related to altitude. Consideration of the data set shows that the location of sites has been unavoidably graded from SOUR to SWEET by descending elevation. A possible interpretation of these models is that ALTITUDE is therefore a 'driving variable' with respect to plant quality. I do not believe this to be the case because all these models would have very poor predictive capacities if applied in other regions of South Africa eg *Stormberg plateau sweetveld* and *Pondoland sourveld* (Acocks 1975; veld types 59 and 3 respectively). This interpretation then suggests that these models are little more than a description of the underlying structure of the field data. The lack of soil chemical variables in these equations supports this argument. Several authors have shown correlations between soil chemistry and plant quality (Meredith 1947; Anon 1965; Laycock and Price 1970; Wilson 1982; Coughenor *et al* 1985) yet such relationships are not clear in

these data.

An obvious counter to the argument above is to remove ALTITUDE from the analysis. This was attempted and resulted in a failure to produce any significant coefficients in the models. For this reason the models presented for each element should be treated with caution as they are likely to be of greater mathematical than biological value.

The purpose of carrying out these analyses was to find some common variable set that determines plant quality. It was hoped that this would lead to an objective classification of different types of veld with respect to veld quality. The inconsistency and range of variables included across all the equations above has not achieved that purpose. It appears that the analysis techniques used are limited in their usefulness in explaining what is clearly a complex interaction. It is unlikely that a workable index of sweetness, based on the environment, will be developed because a major limitation is the need to handle each variable separately using this regression approach. An alternative approach would be to use multivariate methods in an attempt to simultaneously consider the inter-relationships within the measures of plant quality. In this way all the measures of quality can be included in any index that may be developed.

MULTIVARIATE METHODS

INTRODUCTION

In the development of this programme a range of environmental and plant quality variables were collected. The analyses that have been carried out previously (Chapter 6) have shown that most parameters of plant quality vary within the groups determined by cluster analysis. The net result of that exercise however was to reduce the data into three groups which have a relatively large within-group range. This was demonstrated by the standard error ranges presented in Figures 6.1 to 6.8. Although the

grouping of sites was very successful and corroborated the conceptual model, it resulted in a compartmentalization of the intergrade. Initially such an approach was useful in determining the general nature of the system. However, to continue analysis on the basis of the three groups would eventually lead to a relatively inflexible model. It is likely that separate indexes of SWEETNESS would be developed for such groups. This would mean that some predetermined classification would be needed before the index could be applied. Such a system would be little different from the current subjective classification.

A multivariate approach would allow each site to be considered separately but together with all sites in the same classification. In this way the intergrade between the two extremes of SWEET and SOUR could be retained. An attempt at such a procedure will be presented in this section.

ANALYTICAL APPROACH

The use of multivariate methods is well established in biology (Morrison 1967; Tatsuoka 1971; Manly 1986). The main purpose of these is to reduce the dimensionality of data and to detect patterns. Most of the techniques are used to describe a complex system in terms of a few major variables. These are usually defined as those accounting for the greatest proportion of the total variation. They also allow for the development of hypotheses for testing in some more rigorous way, but their most useful application is the convenient description of complex data. Use of techniques such as principal components analysis will allow each multivariate case (site) to be described in terms of a scalar value. This site description represents the placement of that site in multidimensional space. Such placement is relative to all other sites. Indirect gradient analysis, which is widely used in vegetation studies, relies on this feature (Gauch 1982).

It is proposed therefore that the successful development of an

index of SWEETNESS needs to follow a gradient approach. The assumption that is made is that there is an intergrade between the two extremes. The following procedure is to be adopted:

1) use the plant data for July as a multivariate data set and subject these to Principal component analysis (PCA);
2) extract the main components from the analysis and rank each site according to its score on the component axes (eigen values);
3) provided the major measures of quality (eg N, P, K and CDMD) are extracted, this ranking will be used as an INDEX of quality; and
4) if the above are successful the INDEX developed can be regressed with environmental variables.

Should this approach work the INDEX can then be regressed with each of the plant quality variables. This will allow the levels of each of these to be determined once an INDEX has been determined from the model developed in 4) above.

The final stage of this proposed approach may be criticized on the basis that the INDEX and plant quality variables are highly auto-correlated, so models with high R^2 values are expected. The author is aware of this but, since no hypotheses are being tested, auto-correlation is of little consequence. The regressions are only being used in a descriptive manner and are essentially providing a method of converting from one measure to another. For this reason the model with the best fit should be used in all cases. This methodology is similar to that described by Stuart-Hill *et al* (1986) for the development of an ecological index of veld condition in the Valley bushveld.

DEVELOPMENT OF A SWEETNESS INDEX

The data for all plant quality parameters in July was subjected to Principal Component Analysis (PCA) using the sites as rows and the variables as columns. This resulted in the calculation of eigen values for each of eight axes, one for each variable (Table

7.1). These data show that the first two components (AXIS 1 plus 2) account for about 81% of the variation. This is relatively high and suggested that the PCA provides a very good summary of the original data (Williams 1976). Furthermore the rapid drop in the magnitude of the eigen value shows that high proportions of the total variance is accounted for by only a few axes. This allows the axis site scores (rankings) to be used as site descriptors (see later).

Table 7.1 Eigen values and variable weights for first two Principal Component Axes from plant quality derived from 32 sites in Natal during July 1984.

COMPONENT NUMBER	EIGEN VALUE	QUALITY VARIABLE	AXIS 1 WEIGHTS	AXIS 2 WEIGHTS
1	68.89	CDMD	0.395	-0.035
2	12.44	N	0.407	0.042
3	7.91	P	0.388	0.058
4	5.71	K	0.392	-0.081
5	2.18	Mg	0.371	0.062
6	1.29	Ca	0.327	0.096
7	0.94	S	0.355	0.030
8	0.63	Zn	-0.060	0.986

The weights of the plant quality variables shows that the first axis represents a combination of all the measures except Zn. This element is accounted for by AXIS 2 (Table 7.1). As each of the remaining axes accounts for less that 10% of the variance these have been excluded from further analysis. The relative positions of the plant quality variables are shown in Figure 7.3. It should be noted however that the position of Zn on COMPONENT 2 is exaggerated by scaling, given that the entire axis accounts for only 12% of the variance whereas COMPONENT 1 accounts for 69% (Table 7.1). Unfortunately using an equal scaling would compress the positions of the variables clustered on COMPONENT 1 so this has not been done. The relative position of Zn when axes are proportionally scaled is indicated by an asterisk (*Zn).

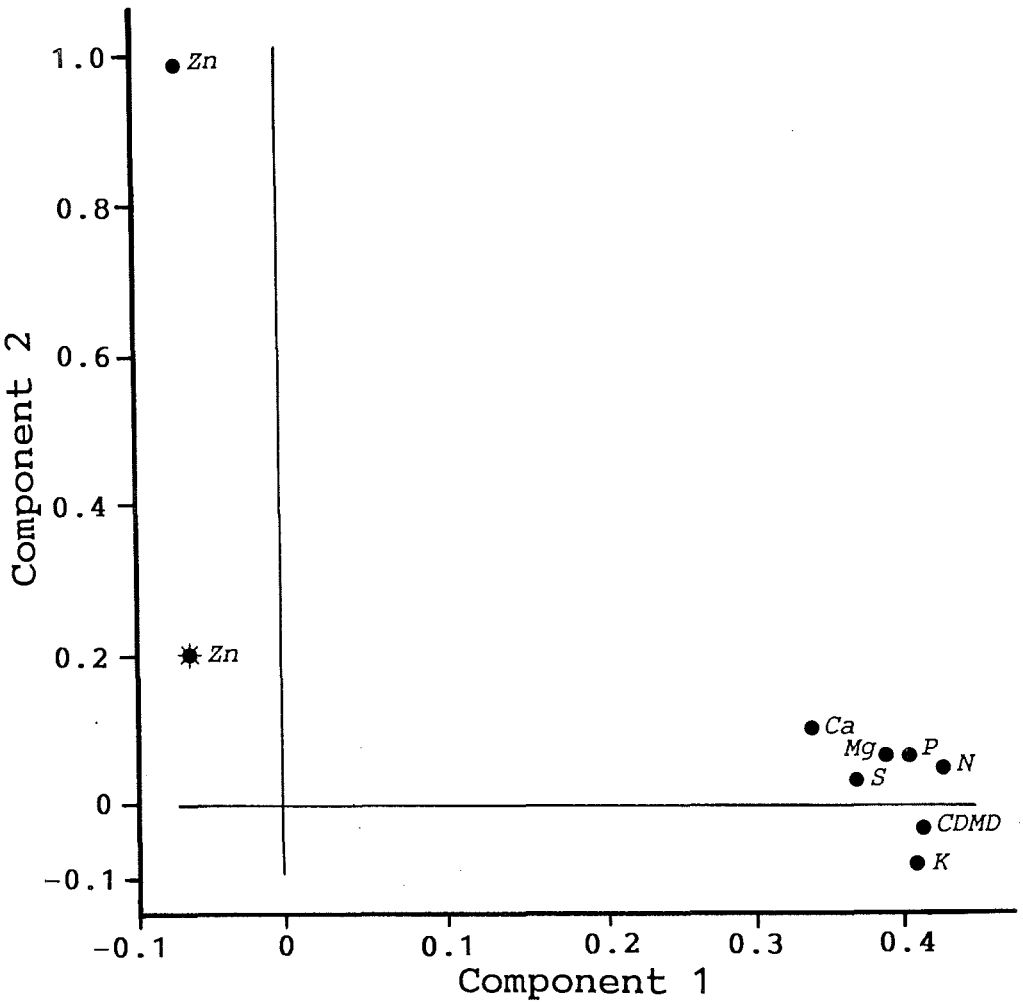

Figure 7.3 Positions of each plant quality parameter in relation to the first two Principal components for *Themeda triandra* collected in July (✸Zn represents the relative position of Zn if both components are scaled according to their eigen values).

On the basis of the grouping of these parameters of quality, COMPONENT 1 is considered an AXIS of quality. Unfortunately no measures of low quality (eg high fibre) were recorded in this study so those are not incorporated here. It is hypothesised therefore that COMPONENT 1 is positively linked to plant quality. On this basis COMPONENT 1 would represent the gradient of SWEETNESS. This however cannot be tested as this stage.

The next stage in the development requires the ranking of each

site along COMPONENT 1. The component scores for each site range from -3.779 to 3.288 (Table 7.2) with a reasonable spread of sites along the axis. The sites are grouped at one end of COMPONENT 2 with the exception of sites 26 and 27 (Table 7.2).

Table 7.2 Principal Component scores for 31 sites in Natal derived from the analysis of plant quality data for *Themeda triandra* in July 1984.

SITE NO	AXIS 1	AXIS 2	AXIS1+6 (INDEX)	SORTED INDEX	SORT NO	CLUSTER GROUP [1]	LOCATION [2]
2	-0.678	0.632	5.32	9.29	8	*	MGU
3	-0.550	-0.164	5.45	9.16	9	*	MGU
4	0.309	-0.032	6.31	9.07	13	*	MKZ
5	0.066	-0.335	6.07	9.00	16	*	MKZ
6	1.203	0.184	7.20	8.73	10	*	MGU
7	1.511	0.497	7.51	8.68	14	*	MKZ
8	3.288	0.773	9.29	8.55	12	*	MKZ
9	3.162	0.379	9.16	8.20	31	+	DBN
10	2.726	0.555	8.73	8.03	29	+	UKU
11	-0.330	-0.210	5.67	7.90	30	+	PMB
12	2.552	-0.311	8.55	7.52	15	*	MKZ
13	3.067	0.953	9.07	7.51	7	*	HLU
14	2.677	0.001	8.68	7.49	17	*	MKZ
15	1.519	-0.262	7.52	7.20	6	*	HLU
16	2.997	0.239	9.00	7.13	32	+	DBN
17	1.492	-0.770	7.49	6.59	28	+	UKU
18	-2.926	-0.938	3.07	6.31	4	*	UMF
19	-2.809	-0.455	3.19	6.07	5	*	UMF
20	-2.948	-1.168	3.05	5.67	11	*	MGU
21	-3.194	-0.037	2.81	5.45	3	*	UMF
22	-3.606	-0.950	2.39	5.32	2	*	UMF
23	-3.411	-0.987	2.59	3.19	19	#	HMR
24	-3.611	-0.348	2.39	3.10	27	#	CLP
25	-3.779	-0.490	2.22	3.07	18	#	HMR
26	-3.686	3.885	2.31	3.05	20	#	HMR
27	-2.901	1.832	3.10	2.81	21	#	HMR
28	0.592	0.472	6.59	2.59	23	#	CLP
29	2.031	-1.450	8.03	2.39	22	#	CLP
30	1.902	-0.422	7.90	2.39	24	#	CLP
31	2.203	-0.371	8.20	2.31	26	#	CLP
32	1.131	-0.702	7.13	2.22	25	#	CLP

[1] * = SWEET; + = MIXED; # = SOUR

[2] UMF = Umfolozi; MGU = Mugudu; MKZ = Mkuze; HLU = Hluhluwe; HMR = Highmoor; CLP = Cathedral Peak; UKU = Ukulinga; DBN = Kranzkloof; PMB = Worlds View, Pietermaritzburg

The position of sites along COMPONENT 1 reflects and range within the SWEET and SOUR groups identified in Chapter 5 (Table 7.2; Figure 7.4). This COMPONENT is considered to provide the basis for the **SWEETNESS INDEX (SW_I)** which is calculated as:

SW_I = COMPONENT SCORE + 6.

The addition of 6 to each of the scores has been done for two reasons:

1) the original site rankings have both negative and positive values and these are inconvenient to work with so the addition converts all ranking to positive numbers; and
2) the amount added (+6) has resulted in a range from 2.22 to 9.29 and this will allow for extensions of the rankings as more data are made available.

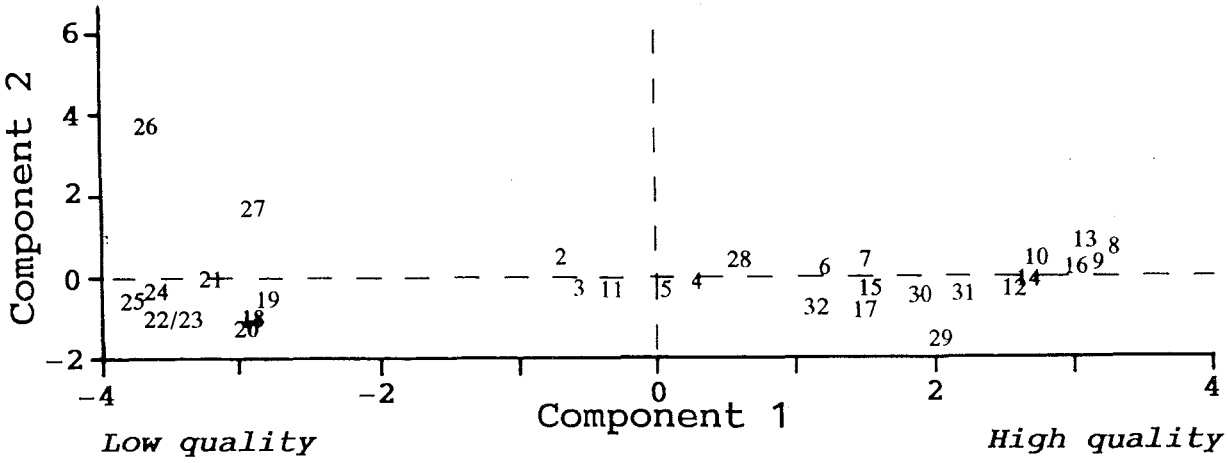

Figure 7.4 Position of 31 sites in Natal according to the nutritional status of *Themeda triandra* in July 1984 (COMPONENT axes have been rescaled to reflect their proportional contribution to the variance of these data).

The resulting index is now in the range of 0 to 10 with zero being SOUR and 10 being SWEET (Table 7.2). Once this adjustment was made the sites were sorted in descending order and their locations identified (Table 7.2). From this arrangement it can be seen that sites from each geographic location are not all grouped together. In particular the MIXED GROUP (see Chapter 5)

have separated into two sub-groups. All of the SOUR sites are concentrated at the bottom of the scale, with Umfolozi closest to that group.

This index (SW_I) provides a first approximation at an objective classification of sweetness.

FACTORS RELATED TO THE SWEETNESS INDEX

The SWEETNESS INDEX (SW_I) developed in the previous section has provided an objective method of rating the quality of *Themeda triandra* using a scalar. What needs to be developed is a method of determining that scalar value for a particular site given some site descriptors. The sites in this study have been described using environmental variables. These need to be related to SW_I to test whether an objective classification of SWEETNESS is possible.

Stepwise multiple regression was once again used to develop a model after the independent variables were subjected to analysis using Spearmans Rank correlations and correlated variables (r_s >0.65) removed. The following uncorrelated variables were used to develop the model:

a) ASPECT (0°),
b) Ca (meq/100g),
c) Mg (meq/100g),
d) EXCHANGEABLE ACIDITY (meq/100g),
e) P (mg/kg),
f) pH, and
g) FIELD MOISTURE CAPACITY (g_{water}/g_{soil}).

The regression procedure produced the following model:

$$SW_I = 11.14 - 1.02x10^{-3}ALTI - 0.18JMNR + 0.08JMDR$$

where

SW_I = SWEETNESS INDEX,
ALTI = ALTITUDE (ft),
JMNR = long term mean rainfall for July, and

JMDR = long term median rainfall for July.

The model has a $R^2_{adjusted}$ of 0.86 with an $SE_{estimate}$ = 0.94. All coefficients were highly significant ($P \leq 0.001$), as was the total model when subjected to analysis of variance.

The equation developed here suggests that climate plays the over-riding role in determining the SW_I as none of the soil or other environmental variables were shown to be important. It is of interest to note that Ca and Mg levels were the last two variables to be removed from the model. Forcing these variables into the model reduced the $R^2_{adjusted}$ to ≤ 0.65 and both coefficients were non significant ($P > 0.05$).

CORRELATION BETWEEN SWEETNESS INDEX AND PLANT QUALITY

The final stage in the development of an objective index of SWEETNESS (SW_I) requires the *calibration* of plant quality with the index. The term calibration is used here specifically to indicate that the determination of plant quality from the index is nothing more than a conversion from one measure to another. The approach adopted uses regression analysis with the objective of finding the model with the best fit. The reader is reminded that the obvious auto-correlation in this methodology is of no consequence because the regression equations provide only a means of converting from SW_I to the plant quality parameter of interest. A simple regression approach was adopted for this purpose.

Each of the plant quality parameters was separately regressed with the SWEETNESS INDEX (SW_I) with the objective of finding a model that accounted for the greatest variance (Table 7.3). As expected most of the variables are highly correlated with SW_I because of the autocorrelation. The conversion models for Ca and S are disappointingly low but are a feature of the high variation of these elements across the sites (Figure 7.5a and b). The model for Zn is extremely poor and this is expected as it had a

Table 7.3 Models used to convert a predicted SWEETNESS INDEX (SW_I) to each of eight plant quality parameters for Natal in July.

QUALITY VARIABLE	MODEL	$SE_{estimate}$	$R^2_{adjusted}$
[1]CDMD(%)	$19.88 + 2.46SW_I$	2.61	0.86
N(%)	$0.19SW_I - 0.087$	0.13	0.94
P(%)	$2.63x10^{-3}\ SW_I^{1.841}$	0.25	0.93
K(%)	$5.81x10^{-2}\ SW_I^{1.30}$	0.25	0.88
Mg(%)	$2.85x10^{-2}\ SW_I^{0.99}$	0.20	0.87
Ca(%)	$e^{(0.098SW_I - 1.65)}$	0.18	0.65
S(%)	$0.058 + 0.013SW_I$	0.02	0.68
Zn(mg/kg)	$22.56 - 0.31SW_I$	5.90	0.18

[1]CDMD = Cellulase dry matter disappearance

very low ranking on COMPONENT 1 from which SW_I was developed. At this stage therefore this model does not provide a means of determining Zn levels in plants from given SWEETNESS INDEX.

CONCLUSION

The methodology presented in this chapter followed two approaches: a) the conventional use of regression techniques and b) the use of pattern seeking multivariate methods. Both of these analyses have produced 'significant' results but neither has shown the soil environment to be important in determining plant quality. This result is somewhat surprising as the practice of fertilizer application is based on the assumption that both production and the quality of the pasture will be improved. One would have expected that the range in soil chemistry and physical values together with apparently corresponding ranges in plant quality parameters would have

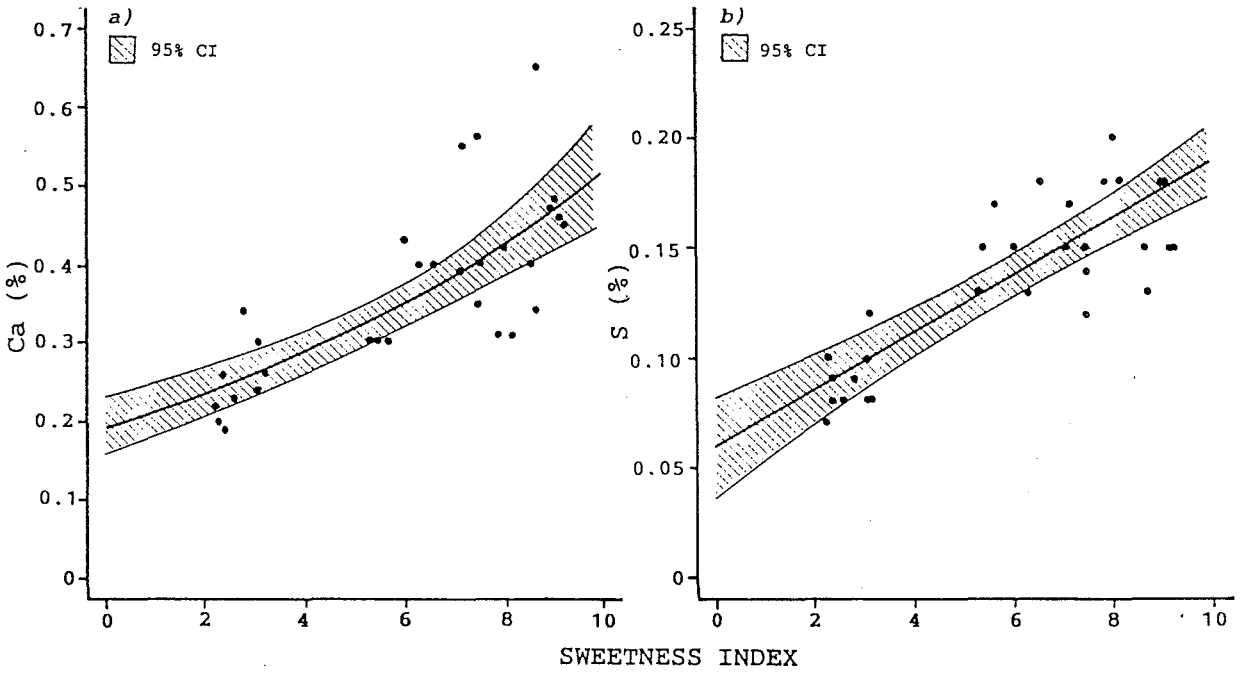

Figure 7.5 Calibration curve used to determine **a)** Ca(%) and **b)** S(%) in *Themeda triandra* in July given a SWEETNESS INDEX.

enabled the detection of causal relationships. This was not so and possible reasons for this apparent failure will be discussed in Chapter 8.

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CHAPTER 8

DISCUSSION AND CONCLUSION

INTRODUCTION

The objectives that were set at the start of this project covered a vast field of study (Chapter 1). The formulation of the objectives was based on a number of assumptions relating to the use of the terms sweet and sour veld. In essence the objectives required the following investigations to be undertaken:

1) to determine if plant quality can be altered by modifying growing conditions;
2) to quantify the seasonal trends in plant quality from different sites;
3) to relate differences between sites to environmental variables;
4) to develop an objective classification of SWEETNESS; and
5) to plan future research.

These tasks were ambitious given the constraints of man power and finance as well as the relatively short sampling period. In spite of this, a relatively large body of information has been collected which has allowed the attainment of some of these objectives to be attempted. The overall research topic covered here is complex, and the potential for inter disciplinary study is enormous. The project that has been described here has made a start to what needs further integrated long term research.

If this initial programme is to be of any benefit in the long term a review of its success is necessary at this stage. The purpose of this chapter is to determine the extent to which the project has achieved its objectives. This needs to be done in terms of the objectives and interpreted in terms of inadequacies of the design.

THE RELATIONSHIP BETWEEN GROWING CONDITIONS AND PLANT QUALITY

The intensive study that was conducted under controlled conditions (Chapter 3) was designed to establish if plant quality could be altered by manipulating growing conditions.

From the point of view of plant growth this experiment was conducted under ideal conditions. The soils were kept at field capacity with watering by mass at a frequency of less then every second day. Temperature was modified with air conditioning to maintain a range of between ca 12°C and ca 25°C, which is ideal for tropical grasses (Langer 1979). Under such circumstances plants are expected to produce high quality material similar to pastures under irrigation (Laycock and Price 1970). The general treatment of all plants was carried out to ensure that factors, such as growing environment, were controlled in order to investigate the effects of soil conditions alone. The use of a cross-over experiment (ie crossing soil and plant types) was considered the best approach.

There is a great deal of speculation amongst 'field scientists', who separate themselves from 'physiologists', regarding the validity of glass house and pot experiments (personal observation). The main objections are that such experiments separate the plant from the complex interactions of a myriad of environmental variables. The author agrees with this sentiment in general. However there was little option in this case. A specific objective of this sub-project was to remove the plant from its own growing medium (especially the soil) and this necessitated the removal of all soil from the plants' roots. In a cross-over experiment in the field this could only be achieved by the radical manipulation of soil that was used for this experiment (Chapter 3). It was far more economical to conduct the trial on a small scale in the glass house and it is unlikely that a different result would have been attained from a field experiment. What is likely is that the results would have been more confused as the environmental effect at each location would

probably not be accounted for in such a short duration. The use of seed and establishment of each plant type in a field cross-over experiment is a possible alternative. This approach was not taken here because the question being asked related specifically to *altering* plant quality by modifying existing soil chemistry. This required that quality of the plants *in situ* be known so that the extent of any changes could be recorded. Using seed would not have allowed the separation of genotypic from phenotypic effects on, or changes to, plant quality (B J Cilliers, personal communication 1983). The genetic complex of *Themeda triandra* is well known (Goosens and Theron 1934; Liebenberg 1966) and separation of these affects would require a complex study in genetics. Although this is an important aspect of the greater project it was beyond the scope of this initial study.

The results of this experiment showed a seasonal trend in plant quality. The quality of the two plant types (sweet and sour) was significantly different ($P \leq 0.01$). However this was only 1.5% CDMD units and it was not considered to be biologically meaningful. The effect of soil, or its interaction with plants, was found to be non-significant in all analyses. This is perhaps not surprising as the amelioration of the soils did not alter the pH despite a large change in acid saturation (44.0% to 0.22%). If an increase in pH had taken place it is expected that nutritional availability to the plant would have been improved and so too plant quality (Jeffrey 1987). It must be noted that the initial differences in pH were small between the eutrophic and dystrophic soil despite a fairly large difference in effective cation exchange capacity (21.68 meq/100g vs 7.38 meq/100g). It is proposed here that the results of this experiment have assisted in describing the seasonal behaviour of plant quality. The effect of soil nutrient status has not been clearly demonstrated so the results are not conclusive. A possible reason for the lack of result here is that the initial soils that were selected were not sufficiently different in terms of the availability of nutrients to plants (see Table 3.4).

It is suggested therefore that the experiment be repeated with careful selection of soils to ensure a wider range in soil nutrients status. A limitation to doing this however would be the cost of transporting the soil and plants to suitable glass house facilities.

A further improvement would be to increase the duration of the experiment as there is a possibility that the soil chemical differences had not manifested themselves due to the short time span of this experiment (1 year). Instituting this improvement would need a larger amount of soil to prevent the plants depleting the nutrients in the soil (see Chapter 3 CONCLUSION).

CONCLUSION

In terms of the objective which this experiment was designed to address, it appears that the soil environment cannot be manipulated to alter the quality of *Themeda triandra,* at least in the short term. The effects that have been shown here are likely to have little effect on animal performance.

SEASONAL TRENDS IN PLANT QUALITY

The conceptual model describing seasonal trends in plant quality (Figure 4.1) has been quantified successfully for most of the measures of plant quality (Chapter 6). Those variables for which the trends are not clear (eg K, Mg, Ca, S and Zn) either do not fluctuate much during the year or alternatively have a high variation within each group of SWEET and SOUR. The overall variation between years prevented the establishment of an equation describing the seasonal trends for the groups. The development of such an equation would have assisted in the objective classification of veld as well as relating plant quality (SWEETNESS) to animal production potential.

The variation in the data that is described above is derived from two sources:

1) the inherent variation in the data, and
2) the pooled variation as a result of grouping sites.

The first of these is a function of the analytical procedures adopted as well as the true values in the plant. All necessary steps and checks were taken during analyses to ensure that the data were as reliable as possible (Chapter 2; Appendix 1). It is believed, therefore, that little can be done about this source of variation. The second source of variation has led to a degree of uncertainty regarding the seasonal trends in plant quality which have been presented (Figures 6.1 to 6.8). The uncertainty is due entirely to the overlapping error bands for many of the variables (Chapter 6). The problems could be overcome by presenting the data for each location separately but this would make ready comparison between groups (ranging from SWEET to SOUR) very difficult. An attempt was made here to present the complex intergrade on the basis of only three groups, SWEET, SOUR and MIXED. Removing the MIXED group improves the visual picture but I believe that its inclusion reflects the variation that is present in the field. Their removal would probably simplify the picture to the extent where any models developed would be of academic rather than practical value.

The data that were collected here provide a foundation on which a number of modelling developments could be based. Essentially these would relate to animal production potential with respect to feed nutrient requirements. As the objectives of this study relate specifically to the plant/soil interaction, such developments are beyond the scope of this report. However including the animal production aspects of the SWEETNESS question is a logical development from this investigation.

CONCLUSION

The quantification of seasonal trends in plant quality has been successfully achieved. The data collected for this purpose have corroborated the conceptual model and will in the future provide a useful basis on which to introduce aspects relating to animal

production.

FACTORS RELATED TO PLANT QUALITY

One surprising outcome from this research programme is the relatively poor degree to which plant nutrient status and soil chemistry have been linked. The use of cluster analysis on both the environmental and the plant data showed very clearly that plant and soil parameters are related in some way. With the exception of the so called MIXED sites the two analyses provided identical clusters. It must be remembered that the plant analysis contained all the data from each of the seven sampling periods so to an extent the time dimension (or seasonal trend) was included in that analysis. The point here is that the separation on the basis of plant material was not only for July (the period of maximum difference between groups) as has been the case with the other analyses.

I expected that there should be some detectable underlying relationship between these independently determined groups. This was not strongly demonstrated here. Some of the equations describing the relationship between plant nutrient status and environment were shown to be highly significant from a statistical point of view. However most had very large SE's of the estimate, suggesting that there was a large variation in the independent variables (Daniel and Wood 1971). An investigation of scatter plots shows this clearly and a typical example is that for the SWEETNESS INDEX (SW_I) and field moisture capacity (FMC) (Figure 8.1). Another feature reflected in these data is the discontinuity between the points at the low end of the SW_I and those of the higher group. The two clusters therefore provide a mathematical means of fitting a straight line between the two groups. Because there are a relatively large number of degrees of freedom from these data (usually >25 depending on the model), highly significant F ratios are generated.

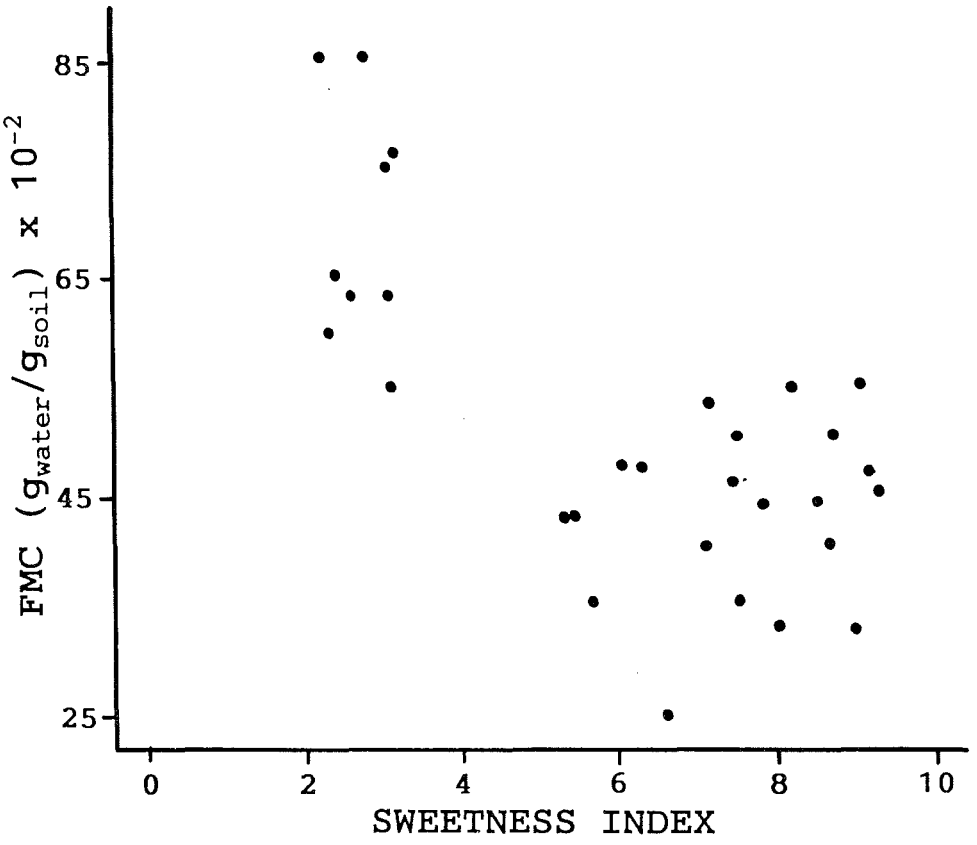

Figure 8.1 Scatter diagram between plant quality and field moisture capacity (SWEETNESS INDEX being the summary of N, P, K, Ca, Mg, S, Zn, and cellulase dry matter disappearance (CDMD) using Principal components analysis).

It will be remembered that ALTITUDE featured in most regression models from both the individual nutrients and SW_I. The grouping of sites is most strongly reflected in these data (Figure 8.2). The significant regressions were developed on the basis of a straight line joining the centroids of two essentially separate clouds of points. Again this has more mathematical value than biological, but explains why ALTITUDE features so prominently in the regression analysis. Unfortunately there is also little to support the validity of any of the models developed here.

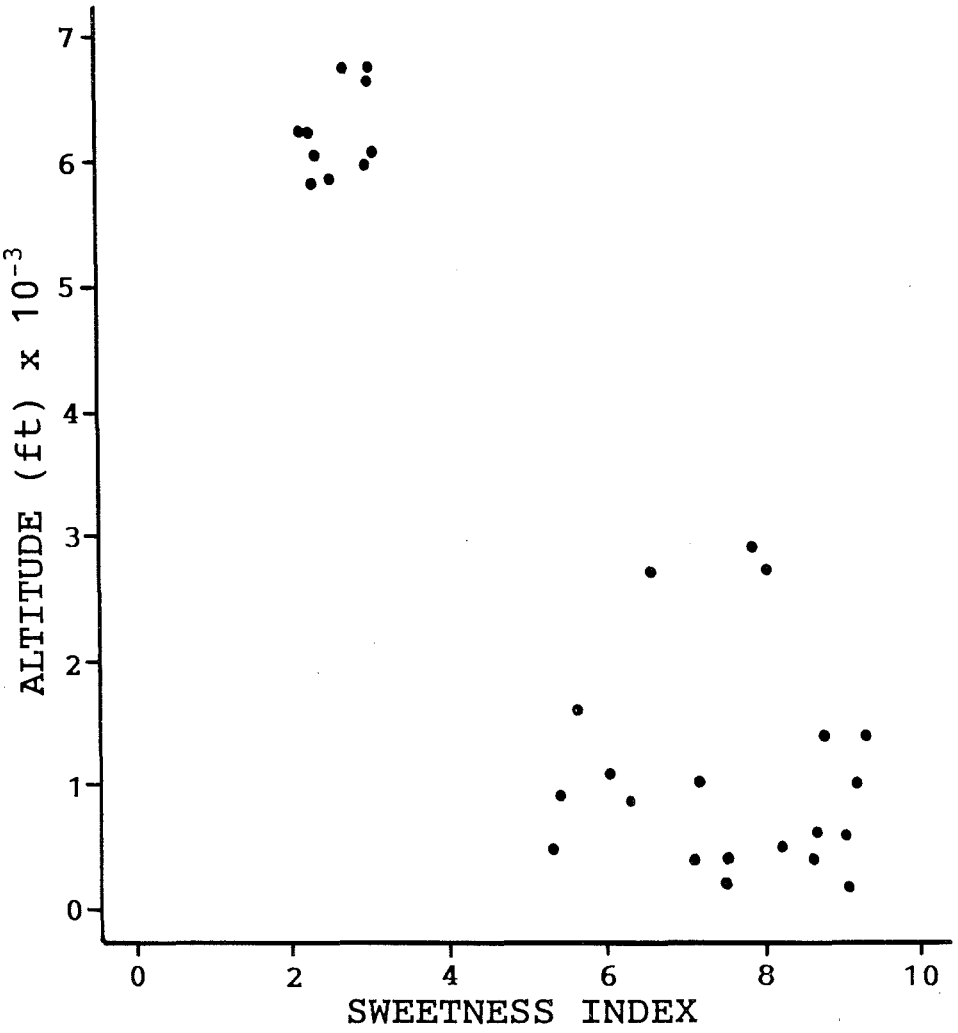

Figure 8.2 Scattergram of SWEETNESS INDEX (SW_I) and ALTITUDE showing the discontinuity in the data set for this variable.

CONCLUSION

The data that have been used in an attempt to determine the relationship between plant quality and the environment is deficient. A major problem is the discontinuity in the data with respect to environmental variables. The results of stepwise regression analysis was overly influenced by ALTITUDE, which is highly discontinuous in these data. The models that have been developed are apparently only significant as a result of a large number of degrees of freedom. Most of them have $SE_{estimates}$ in excess of 10% mean of the dependent variable and are therefore

of little biological use (Daniel and Wood 1971). The only option available therefore is to reject the models as invalid.

At this stage it appears that the objective of determining factors affecting plant quality is unattainable.

AN ALTERNATIVE INTERPRETATION

The apparent lack of association between plant quality and environment that is reflected in these data has been confirmed in a later study by Kirkman (1988). This study was designed as an extension of this programme and many of the sites used by Kirkman (1988) were initially located during this study. The data collected by Kirkman were therefore an extension of the time series of these data. However the location of sites in Kirkman's study extended over a wider area and he also concluded that there were no obvious environmental factors determining plant quality in July.

It appears therefore that the objective classification of sweet and sour veld will need to be plant based. Essentially then the subjective classification currently in use will have to be used as no definite improvements have been made at this stage. What has been achieved is an objective summary of a range of plant quality characters. A possible alternative approach would be the use of norms.

One of the problems that has been encountered in these data is the high variability in the soil data. A review of the scatter diagrams reveals that the variability is largely associated with the values for the sweetveld sites, or at least those sites at the high end of the sweetness index (SW_1). This then suggests that there is some critical value above or below which only sour veld sites will occur. This concept will be explained using the scatter diagrams of K and Ca with the SWEETNESS INDEX (SW_I) (Figure 8.3). These data presented in Figure 8.3 show very clearly the lack of values in the top left hand section of each

diagram and matches the pattern for other elements eg Mg, Na, pH and ECEC. If this pattern is consistent then the upper limits for certain classes can be developed with the stippled lines on each diagram representing the boundaries. The placement of the boundary must be developed in some objective way (eg cluster analyses) and formerly tested.

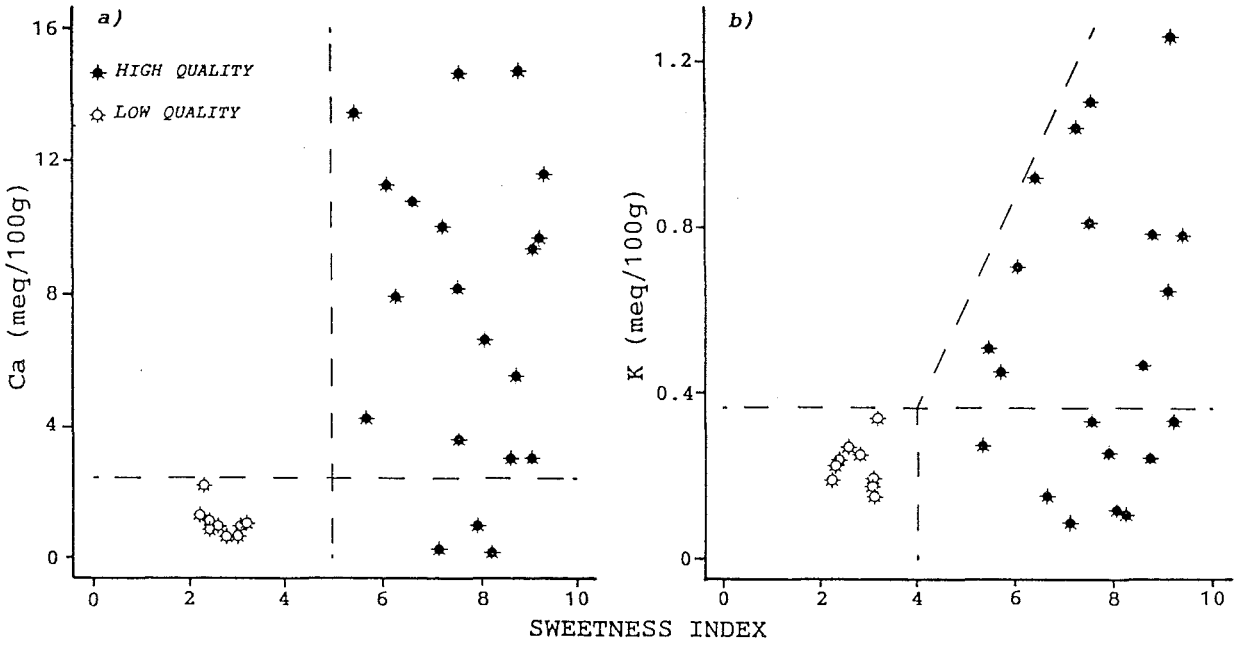

Figure 8.3 Scatter diagrams for **a)** Ca and **b)** K which illustrate the low variation in LOW QUALITY sites and high variation in HIGH QUALITY sites (data are from 31 sites in Natal; see text for details).

The method proposed here needs further development. It is entirely empirical and will rely on a large body of data from both plant and soils in order to develop reliable norms. Once developed these norms can be related to potential animal production via feeding tables. If it can be shown that the norms are consistent then they may provide a mechanism for a more detailed classification of SWEETNESS on the basis of soil chemistry.

CONCLUSION

The research that has been reported in this document is based on

a single species (*Themeda triandra*). The analysis of the data has not provided a clear picture of the functioning of the process of seasonal decline in plant quality. An objective index of quality based on plant parameters has been successfully developed. However this is not apparently correlated with any environmental variables.

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APPENDIX 1

DESCRIPTION OF TECHNIQUES

INTRODUCTION

In any investigation where a number of analyses are to be carried out the researcher is faced with a choice of analytical techniques. There do not seem to be any universal guidelines for the choice of suitable techniques. In addition to this there are very few routine laboratory facilities available which can accommodate the numbers of samples which it was anticipated would be generated during this investigation. This necessitated the development of sufficient analytical skill by the author in order to carry out the analysis of plant and soil samples.

A further complicating factor is that once a technique has been decided upon, there are a number of variations in the laboratory procedures detailed in the literature. Some procedures that are recommended however, were not suitable for this study because they are either too expensive, when applied to large numbers of samples, or required large quantities of material (particularly for the glass house trial).

In order to overcome the problems relating to the relatively large numbers of samples some of the methods were modified for this study and those modifications are detailed here. In addition to considerations of modification it is also important that the reader is made aware of the procedures used so that comparisons with studies of a similar nature can be made on the basis of the procedures used and not only on the outputs from the study.

This appendix is included to provide the details of the field and laboratory procedures used in this study. They are presented in point form for the sake of brevity.

SITE LOCATIONS

The location of the 32 sites for this study were determined using Surveyor General 1:50 000 topocadastral sheets. The position of each site was determined using triangulation to give a six figure grid reference (Table A1.1) This should enable the relocation of the sites which were not permanently marked. Permanent markers were not used because the collaborating agencies considered this undesireable. In any event no long term monitoring of any of the sites was planned at this preliminary stage.

PLANT COMPONENTS

In previous studies (eg du Toit *et al* 1940a,b) the herbaceous sward was harvested as a single unit (usually whole plants). When dealing with a multispecies situation plant quality data from each sample are confounded with inter and intra-species aspects. As far as inter-species relationships are concerned these have been excluded from this study by choosing only a single species. The intra-species factors of concern here are the different qualities of each component of the plant.

In order to ascertain which component would be used for this study the quality (CDMD) of WHOLE plants and TWO-LEAVES-AND-BUD was compared for July and November (Figure A1.1)

From these data it can be seen that data from the WHOLE plant samples do not clearly show differences between the two harvest times over the range of sites spanning both sweetveld and sourveld. However the TWO-LEAVES-AND-A-BUD samples do show differences, especially in the typically 'sourveld' sites. As these data are from sites collected in the Drakensberg (sour) and Zululand (sweet) it is expected that there should be detectable differences. One possible reason for there being no differences between sites in the WHOLE plant data is the large difference in leaf:stem ratio of plants from the two areas (Table A1.2).

Table A1.1 Location of sampling sites in Natal.

SITE NO	MAP[1] SHEET	LOCATION	LATITUDE	LONGITUDE
1	2831BC	UMFOLOZI	28°16'22"	31°44'00"
2	2831BB	UMFOLOZI	28°14'20"	31°48'20"
3	2831BD	UMFOLOZI	28°19'00"	31°50'20"
4	2831BD	UMFOLOZI	28°19'10"	31°50'20"
5	2831BD	UMFOLOZI	28°19'20"	32°50'20"
6	2832AA	HLUHLUWE	28°06'10"	32°02'30"
7	2832AA	HLUHLUWE	28°05'06"	32°07'25"
8	2731DA	MGAMBO FARM	27°30'35"	31°40'10"
9	2731DA	MGAMBO FARM	27°30'35"	31°40'12"
10	2731DA	MGAMBO FARM	27°30'50"	31°40'00"
11	2731DA	RONDEBOSCH FARM	27°46'40"	31°39'10"
12	2732CA	MKUZE	27°38'40"	32°09'20"
13	2732CA	MKUZE	27°38'40"	32°09'30"
14	2732CA	MKUZE	27°38'30"	32°09'45"
15	2732CA	MKUZE	27°36'40"	32°13'15"
16	2732CA	MKUZE	27°39'50"	32°14'30"
17	2732CA	MKUZE	27°40'00"	32°15'00"
18	2929BC	HIGHMOOR	29°19'15"	29°37'25"
19	2929BC	HIGHMOOR	29°15'00"	29°38'40"
20	2929BC	HIGHMOOR	29°18'55"	29°38'50"
21	2929BC	HIGHMOOR	29°19'30"	29°37'10"
22	2829CC	CATHEDRAL PEAK	28°57'40"	29°14'20"
23	2829CC	CATHEDRAL PEAK	28°57'40"	29°14'05"
24	2829CC	CATHEDRAL PEAK	28°58'20"	29°15'05"
25	2829CC	CATHEDRAL PEAK	28°58'55"	29°14'30"
26	2829CC	CATHEDRAL PEAK	28°58'55"	29°14'31"
27	2829CD	CATHEDRAL PEAK	28°58'00"	29°16'00"
28	2930CB	UKULINGA	29°39'48"	30°24'08"
29	2930CB	UKULINGA	29°40'06"	30°24'15"
30	2930CB	WORLDS VIEW	29°34'49"	30°20'00"
31	2930DD	KRANZKLOOF	29°45'18"	30°50'06"
32	2930DD	KRANZKLOOF	29°45'52"	30°51'23"

[1] Surveyor General Topocadastral 1:50 000 map sheet.

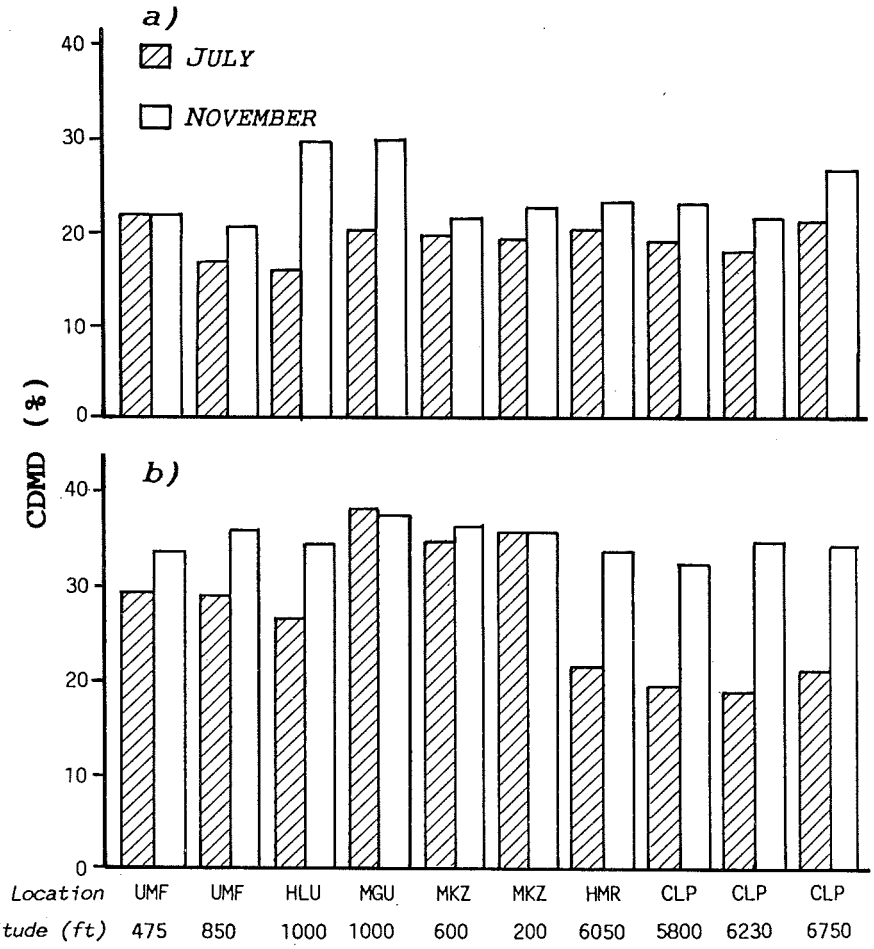

Figure A1.1 A comparison of the cellulase dry matter disapperance (CDMD) of **a)** WHOLE plants and **b)** TWO-LEAVES-AND-A-BUD for ten sites in July and November 1984. (UMF = Umfolozi; HLU = Hluhluwe; MGU = Mgudu; MKZ = Mkuze; HMR = Highmoor; CLP = Cathedral Peak).

Table A1.2 Leaf:stem ratios of *Themeda triandra* for various areas in Natal.

LOCATION	LEAF:STEM RATIO[1]	
	mean	SE
Mkuze	0.206	0.0036
Hlobane	0.546	0.0968
Mugudu	0.250	0.0581
Cathedral Peak	3.029	0.3946

[1] refers to leaf blade removed at the collar

The plants from Zululand are generally tall and stemmy and with much aerial tillering, while those in the Drakensberg are shorter and have high leaf:stem ratios. As one of the key questions of

this project concerns the development of a rapid technique of indexing veld in terms of its sweetness or sourness, it was decided to consider a component that is more easily harvested than TWO-LEAVES-AND-A-BUD, but that would also show differences between sites. For this reason the plants were harvested in the field and later separated in the laboratory. Each component was then analysed separately (Figure A1.2).

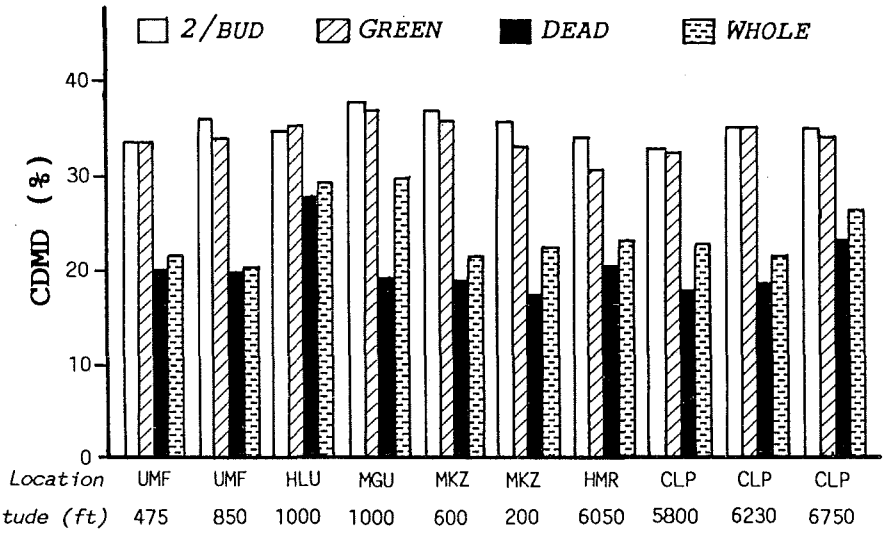

Figure A1.2 A comparison of the four components of *Themeda triandra* harvested in November 1984.

From these data it can be seen that GREEN LEAF material and the TWO-LEAVES-AND-A-BUD sample are closely correlated whereas the DEAD LEAF and WHOLE plant differ across sites. As animals are likely to select the quality components it is important to attempt to harvest each component separately so that their differences can be highlighted. In this way a clearer picture of what is available to the animals will be built up. In time such an approach may lead to the developement of more reliable forage flow models, based on the nutritional information presented here.

SITE VARIABILITY

One of the dangers of conducting a field study, which relies on laboratory analyses, is that the sampling strategy may not be

intense enough to capture the range of values of interest. As far as forage analysis is concerned the researcher should be sure that he has collected a *representative* sample. What constitutes a representative sample is a function of the variance of the parameter of interest.

In order to make a sampling strategy efficient, therefore, a preliminary harvesting technique study should be carried out. The objectives of this technique investigation are to:

1) ensure that the harvesting procedure is practical;
2) estimate the mean and variance of the parameter to be measured; and
3) calculate the number of samples required to detect differences of a particular magnitude at some chosen level of precision.

This section describes the techniques investigation carried out to determine the numbers samples required at each site for the field study.

PROCEDURE

A site for this investigation was located at Ukulinga Research Farm. Five subsites were randomly located and at each subsite seven *Themeda triandra* plants were identified. The plants were then individually harvested by clipping all the material above the second to uppermost exposed ligule on each tiller. The material was then dried by forced draft (65°C for 48h), milled to 1mm, weighed and stored. The *in vitro* dry matter disappearance was then determined (Tilley and Terry 1963; Jones and Hayward 1973) and the results subjected to analysis of variance.

RESULTS AND DISCUSSION

Dry matter yields of the milled material above the second to uppermost exposed ligule on all tillers of each tuft indicated

that harvesting individual plants would not be feasible as many plants yielded insufficient material for *in vitro* digestibility determinations (Table A1.3 & A1.4). Such small samples would not allow any further analysis to be carried out in order to obtain other measures of forage quality.

For the purposes of this harvesting investigation, all plants that provided at least one sample (ca 0.55g) for analysis were included.

Table A1.3 Dry matter yields (g) of milled material form *T. triandra* plants harvested individually.

	Subsites				
Plant	1	2	3	4	5
1	0.6777	2.9108	0.8290	2.0568	0.4916
2	1.0644	2.6460	1.5456	0.7443	1.7342
3	1.4085	1.8397	0.7754	4.1229	0.3453
4	0.8351	3.2852	0.5837	1.1402	0.4520
5	0.5325	1.2418	0.3107	2.7425	1.7137
6	1.5763	2.5966	1.3882	0.6069	2.3689
7	0.9257	1.0537	0.5710	1.6953	1.4087
MEAN (g)	1.0029	2.2248	0.8577	1.8727	1.2163
s	0.3782	0.8560	0.4507	1.2448	0.7905

Table A1.4 Numbers of plants yielding sufficient material for a single assay for *in vitro* digestibility analysis.

N° 0.55g samples	N° of plants
2	18
1	10
0	7
Total	35

This meant that for some plants only a single *in vitro* determination was carried out. However, they were included in the analysis (Table A1.5) so that a more reliable estimate of between plant error could be obtained.

Table A1.5 Summary of the analysis of variance for harvest technique to determine the variability between plants at any one location.

SOURCE	DF	SS	MS	$F_{(4,19)}$
BETWEEN SUBSITES	4	122.6295	30.6574	1.4450 NS
BETWEEN PLANTS WITHIN SUBSITES	25	530.4159	21.2166	2.3070 NS
BETWEEN DUPLICATE DETERMINATIONS WITHIN SAMPLES	19	174.7360	9.1966	
TOTAL	48	827.7814		
	MEAN =	66.4891		
SE OF SINGLE DETERMINATION	=	3.0326		
	CV% =	4.54		

This result enables a composite sample to be harvested but, the number of plants required to make up that sample needs to be calculated as follows (G P Y Clarke and H M Dicks personal communications 1984);

(let s^2 = estimate of variance)

$$s^2_{duplicates} = 9.1966$$

$$s^2_{whole\ plots} = (122.6295 + 530.4159)/(4 + 25)$$

$$= 22.5188$$

$$s^2_{total} = (22.5188 - 9.1966)/2$$

$$= 13.3222/2$$

$$= 6.6611$$

Now, $var(x_i)$ (ie of a composite sample) is given by;

$$var(x_i) = (s^2d + r.s^2_T)/r.n_i$$

where r = number of duplicate runs for *in vitro* digestibility.

To calculate LSD ($P \leq 0.01$) to estimate a 5% difference between sites;

$$LSD = \sqrt{[2.var(x_i)]}.t_{(0.01(19))}$$

$$d.m/100 = \sqrt{[2.var(x_i)]}.t_{(0.01(19))}$$

where d = % detectable difference required

m = mean per sample

therefore;

$$var(x_i) = [(d/100m)/t_{0.01}]^2/2$$
$$= (((5x66.489)/100)/2.861)^2)/2$$
$$= ((3.3245/2.861)^2/2$$
$$= 0.6751$$

Assuming duplicate analyses as standard laboratory practice the number of plants (n_i) required is calculated from (Rayner 1967);

$$n_i = (s^2_d + rs^2_t)/(r\ var(x_i))$$
$$= ((9.1966 + (2 \times 6.6611))/(2 \times 0.6751)$$
$$= 16.67$$

ie 17 plants.

CELLULASE DRY MATTER DISAPPEARANCE

INTRODUCTION

Currently in South Africa the two stage *in vitro* technique of Tilley and Terry (1963) is widely used for the determination of forage dry matter disappearance (DMD) in the laboratory. While this method has been shown to be reasonably accurate (Engels and van der Merwe 1967; McLeod and Minson 1969; Barnes 1973; Welch 1983), there are a number of problems associated with its application. These include 1) the maintenance of donor animals under controlled conditions, 2) the difficulties of collecting and handling rumen liquor, 3) the maintenance of anaerobic conditions during the collection of rumen liquor as well as during the analyses, 4) the difference between the type of donor

animal (usually sheep) and those to which the research applies and 5) the fact that forages under test are not usually the same as the maintenance diet of the donor animals, thus affecting the rumen micro-organism population. Although these problems are not insurmountable they provide a number areas for potential error.

Work conducted in Wales and Australia has led to the development of a modified two stage technique in which cellulase enzyme replaces rumen liquor in the digestion process (Jones and Hayward 1973,1975; Goto and Minson 1977). Using this method McLeod and Minson (1978) have studied the effect of incubation time, temperature, sample size, cellulase concentration, and grinding size on the accuracy of the technique for predicting in vivo digestibility (here *in vivo* refer to digestibility determinations of forages from feeding trials). They concluded that the technique was statistically superior to the *in vitro* rumen liquor procedure, by yielding a lower residual standard deviation (RSD) ($P \leq 0.01$) for the regression of *in vivo* upon *in vitro* values.

The cellulase technique has a number of advantages over the rumen liquor method, including the following: 1) no animals are required; 2) anaerobic conditions are not required; 3) analysis may be conducted when required without ensuring the correct pre-treatment to donor animals; and 4) large numbers of samples can be handled simultaneously as the volume of digesting agent (ie cellulase rather than rumen liquor) is not limiting.

Zacharias (1986) provides a more complete explanation of the comparison between rumen liquor and cellulase as digesting agents in the laboratory.

LABORATORY PROCEDURE

Throughout the investigation the following procedure has been adopted for cellulase dry matter disappearance (CDMD) determinations:

1. approximately 0.5g samples (dried in a forced draft oven at 65°C for 48 hours and milled to 1mm, are weighed directly into 120ml glass tubes in duplicate with a known *in vivo* standard included in each run;
2. approximately 2g samples are weighed into oven dry porcelain crucibles of known weight and their DM content determined gravimetrically;
3. an acid-pepsin solution (4.8g pepsin powder per litre of 0.125M HCl (sp.gr. 1.18) per litre) is made up fresh and 25ml is dispensed by an automatic pipette into each tube;
4. tubes are covered with parafilm, agitated gently and incubated (water bath) at 39°c for 48 hours;
5. tubes are agitated gently three times a day during incubation;
6. after 48 hours the tubes are removed from the water bath and 1.5ml of 1M sodium carbonate are added down the sides of each tube to adjust the pH to between 4.5 and 4.7;
7. cellulase-buffer solution (800 IU cellulase per litre of acetate buffer (4.10g anhydrous sodium acetate and 2.9ml glacial acetic acid per litre)) is made up when required and 50ml are added to each tube;
8. tubes are resealed and incubated at 39°C for a further 48 hours;
9. tubes are removed after incubation and the contents transferred quantitatively to preweighed, oven dry sintered glass crucibles (porosity 1) and filtered under vacuum;
10. samples are rinsed in the sintered glass crucibles with 10ml acetone and then oven dried overnight at 105°C; and
11. after drying, the crucibles are removed, desiccated to cool and weighed for CDMD determination by weight lost from the sample.

VARIABILITY

Every batch of samples that were analyzed included a standard of the same material each time. These standard samples were used to 1) ensure that no major problems had arisen during the

digestion (eg wrong chemicals) as well as 2) to determine whether adjustments for between-runs variations was required. The mean CDMD values of a standard sample included in 20 runs (4 replications) over an 18 month period were not significantly different ($P>0.05$) and yielded a coefficient of variation of 8.3% (MEAN = 17.45 ± 1.451%). The technique has therefore shown to be reliable for indexing the digestibility of grass samples for the purposes of this study. Furthermore these data show that no adjustment for runs was required (H M Dicks, personal communication 1985). As standard laboratory practice, duplicate analyses that differed by more than 2% were discarded. As the SE of the difference between two samples is 1.451% the LSD ($P\leq0.05$) between duplicates would be 2.90%. This result confirms that this practice was sound.

PLANT ELEMENTAL STATUS

NITROGEN

Procedure

The procedure was streamlined as far as possible in order to use the available equipment most efficiently. The following method was used:

1) weigh out ca 0.5g of sample into a 100ml Kjeldahl flask;
2) add ca 0.15g K_2SO_4 to raise the boiling point of the acid;
3) burette 0.5ml $HgSO_4$ (10g HgO in 90ml deionized water with 11ml conc. H_2SO_4);
4) pipette in 10ml conc. H_2SO_4 by automatic pipette;
5) boil in a fume cupboard until solutions are clear, swirl tubes occasionally;
6) allow to cool and slowly add ca 10ml deionized water,
7) filter and transfer solution to 250ml volumetric flask;
8) dispense 50ml with a measuring cylinder into a clean beaker;
9) determine the N content with a specific ion electrode (ammonium electrode); and

10) present results as %N in plant tissue.

Variability

The standard was analyzed using 12 replications (Table A1.6). The readings from the ammonium electrode were taken at intervals during the recording of samples to determine the consistency of the instrument.

Although this coefficient of variation (17.08%) is relatively high (Rayner 1967) there is no alternative method of determining plant nitrogen that can be used for such large numbers of samples. In any event this coefficient of variation merely reflects the inherent variability of the technique and provides no statistical justification for rejecting the procedure or these data (Sokal and Rohlf 1981).

Table A1.6 The values for N content (%N in plant tissue) of the standards used to determine the reliability of the instrument.

REPLICATE	1	2	3	4	5	6	7	8	9	10	11	12
N content%	0.07	0.09	0.08	0.08	0.09	0.09	0.12	0.09	0.09	0.08	0.12	0.08

MEAN = 0.09% s = 0.015% CV = 17.08%

OTHER ELEMENTS

Ashing procedure

Before extraction of the elements from the material the organic matter in the sample was removed by ashing. The following procedure was adopted:

1) ca 0.5g of milled samples was weighed directly into a wide-form porcelain crucible ca 20ml capacity;

2) samples were placed on an asbestos tray, their positions carefully noted, and then placed in a pre-warmed (ca 200°C) muffle furnace;
3) the furnace was then set to ca 500°C and the material ignited for 2 hours before being removed to a desiccator to cool;
4) once cool, samples were moistened with a few drops of deionised water before 10ml 4M HCl:HNO_3 was slowly added to each crucible;
5) samples were then digested for ca 20min on a sand bath; and
6) once digested, samples were cooled and transferred quantitatively, through Whatman N°41 (ashless) filter paper, using deionised water, into 250ml volumetric flasks.

Once all samples had been ashed, digested and transferred, separate aliquots of the extractant were drawn for the various analyses.

Calcium and magnesium

Calcium and magnesium were determined using an Atomic Absorption Spectrophotometer (AAS). A 5ml aliquot was pipetted into a dry plastic bottle and 1ml of strontium buffer solution (2 500mg/l strontium in a solution of 7.61g/l $SrCl_2.6H_20$) was added. Results from the AAS were recorded in mg/kg and these were converted to express the concentration as percent of plant tissue.

Potassium

Potassium was also determined using an AAS with 1ml of extractant being used. This was mixed with 5ml of caesium buffer solution (1 200mg Cs/l as 1.52g/l CsCl) and read as mg/kg K, with the results being expressed as %K in oven-dry material.

Phosphorus

The vanadamolybdate yellow method was used to determine P with

readings of absorbence from a colorimeter set at a wavelength of 440nm. The vanadamolybdate yellow was made up as follows:

1) 20g ammonium molybdate was dissolved in 400ml deionised water;
2) 1.0g ammonium metavandate was dissolved in 300ml boiling water, cooled and then 200ml conc. HNO_3 added;
3) once both solutions (1 and 2 above) were thoroughly cooled the molybdate was added, with continual stirring, to the vandate solution and then made up to 1 000ml with deionised water.

A 10ml aliquot of plant extractant was pipetted into a dry acid-washed glass bottle. A 2ml volume of vanadamolybdate was added and the colour allowed to develop for 15 minutes. Absorbence readings were taken and converted into mg/kg using a calibration curve. These mg/kg readings were then used to calculate P as percent in oven dry material.

Variability

For each determination of elements the standard used previously was analyzed a number of times (13) as a check (Table A1.7). The highest CV% was for the phosphorus determination and this is probably due to errors in reading the second decimal place on the colorimeter as well as from errors in taking reading from the calibration graph.

Table A1.7 The summary statistics for the standard used to check the variability of determinations for Ca, Mg, K and P in plant material by wet chemistry methods (all values in mg/kg).

	Ca	Mg	K	P
max. value	3.9	1.4	7.9	1.4
min. value	3.2	1.1	6.1	0.83
mean	3.39	1.21	6.86	1.02
std. dev.	0.18	0.10	0.53	0.15
CV%	5.31	8.26	7.73	14.71

SOIL ELEMENTAL STATUS

CHEMICAL

The determination of soil chemistry included P extractable cations (Ca, Mg, K, Na,), Aluminium content and pH.

Phosphorus

Soil P was extracted using 0.05N H_2SO_4 in a 1:10 soil to extractant mixture at a nominal temperature of 25°C. The method adopted is as follows:

1) weigh 5g air dry soil (sieved to 1mm) in a dry 200ml Erlenmeyer flask;
2) check that the flasks and extractant are ca 25°C;
3) add one scoop (ca 5ml) of activated carbon (Darco G60 charcoal washed in 1:1 H_2SO_4; add 5l acid per 1kg charcoal, rinse in deionised water until wash water gives a pH of 2.5 or higher) to remove soil derived colour;
4) add 50ml 0.05N H_2SO_4 and shake immediately (4 min at 175 cycles/min);
5) filter the extractant through two circles of Whatman N°41 filter paper into 4ml Elkay Auto Analyzer sample containers; and
6) determine P in mg/kg.

Extractable cations

The method employed at Cedara uses a neutral salt solution to replace the cations present on the soil exchange complex. Therefore the cation concentrations determined by this method are referred to as 'exchangeable + soluble'.

Extraction

This is carried out using a 1N ammonium acetate solution as

follows:

1) weigh 5.0g of 1mm, air dry soil into an 80ml extraction tube;
2) add 50ml of extracting reagent (1N ammonium acetate adjusted to pH7);
3) place tube on reciprocal shaker and shake for 30 minutes (180 oscillations/min);
4) filter the extractant through Whatman N°30 filter paper, discarding the first few drops, refilter if filtrate is not clear; and
5) prepare extractant for analysis using flame emission or atomic absorption where appropriate.

Analysis

The determination of K, Ca, Mg and Na concentration in the filtrate is carried out using flame emmission according to the procedures described by Isaac and Kerber (1971). Standards used to calibrate the Atomic Absorption instrument are 1 000mg/kg Set Standards.

Aluminium

The method used to determine the Al content of soils involves a titration and because of this, 'exchangeable acidity' is a more accurate description of the measure (M V Fey, personal communication 1983).

Procedure

The extraction procedure is similar to that used for the other elements and is described as follows:

1) weigh 10g air dry soil (≤1mm) into a plastic extraction tube;
2) add 50ml 1N KCl (74.56g KCl/l distilled water) and shake end-over-end for 4 minutes;
3) filter through Whatman N°41 filter paper;

4) add 10ml phenolphthalein solution (5g phenolphthalein dissolved in 500ml 96% ethyl alcohol and make up to 1 000ml with distilled water) to the 10ml sample extractant; and
5) titrate with 0.05N NaOH.

pH

The determination of pH in soil is based on the activity of H^+ ions in a soil suspension. The activity of H^+ ions is dependent on the matrix of the suspension and the pH requirements differ for the same soil depending on the matrix used. For this reason pH is determined and recorded in respect to the matrix, KCl in this case. The following procedure is used;

1) weigh 10g air dry soil (≤1mm) into a 100ml container;
2) add 50ml of 1N KCl (74.56g KCl/l), stir and allow to stand;
3) after 50 minutes stir again and allow to stand for a further 10 minutes; and
4) stir again and read pH using a digital pH meter (calibrate using Beckman pH4 buffer solution) taking a reading only after the meter has stabilised.

Organic carbon

The organic carbon content of the soil was analyzed using the Wakley-Black method as described by Allinson (1965).

PHYSICAL

Field moisture capacity

Cylinder method

The method described below is used to determine FMC when pot trials are to be watered to mass (A Cass, personal communication 1983). The method used for the glass house trial was as follows:

1) soil (sieved to 5mm) was placed in a 1 000ml measuring cylinder and compacted to the same degree as that in the

buckets by tapping the cylinder three times on the laboratory bench (this procedure was also followed for the buckets);

2) a glass tube was placed up the centre of the soil column to allow air to escape;
3) water was then added to the cylinder (ca 150ml);
4) the cylinder was covered with aluminium foil to prevent surface evaporation and allowed to equilibrate for 24 hours;
5) once there was no apparent movement of the water-front the moist soil was removed, weighed immediately and dried in an oven at 55°C for 48 hours and then reweighed; and
6) FMC was calculated gravimetrically.

The recordings on the basis of this method were used to calculate the amount of water to add to each bucket to bring the soil to field capacity.

Syringe method

Although the cylinder method is relatively quick, fairly large quantities of soil are required in order to carry out duplicate determinations. Apart from this, the laboratory equipment needed to conduct the determinations of FMC on the samples for the field investigation for this study was not available in sufficient quantities. This meant that some suitable small scale method had to be devised that would yield satisfactory results.

The syringe method was developed and tested as a procedure for indexing FMC.

Procedure

The soil samples were air dried and passed through a 2mm screen. Ten plastic syringes with the plunger removed, were marked, oven dried and then weighed. These were then filled to the 10ml mark and tapped five times on the bench to compact the soil. The soil was then saturated several times with water and allowed to drain

freely. Cotton wool plugs were then placed in the top of the syringes to reduce surface evaporation. The soil was then left to equilibrate overnight before being weighed. The syringes were then placed in an oven at 95°C for 24 hours to dry (greater temperatures would have melted the syringes). The syringes were then desiccated to cool and weighed.

Results and discussion

Once all the weights had been determined FMC (Table A1.8) was calculated as follows:

$$FMC = \frac{(\text{syringe + wet soil}) - (\text{syringe + dry soil})}{(\text{syringe + dry soil}) - (\text{syringe})}$$

These data yielded a CV% of 3.31 which indicates the technique is repeatable by virtue of its inherent low variability.

Having shown the technique to provide a repeatable index the number of replicate determinations required to detect differences between soils was calculated. The number of replications (n) is given by:

$n = s^2(t_{0.05})^2L/2$ (Anon 1983)

where s = standard error of the mean
= 0.01224

$t_{0.05}$ = tabular value for the t-test
= 2.228 (DF=10)

L = 5% of the mean (ie detectable difference required)
= 0.0185

n = number of replications required
= 2.27

From this calculation it was decided that three replications should be used for the determination of FMC, from sieved soil samples, by the syringe method.

Table A1.8 Data used to calculate the number of replications required to determine Field Moisture Capacity (FMC).

	MEASUREMENT					
Rep N°	Syringe (g) (A)	Syringe+ wet soil (g) (B)	Syringe+ dry soil (g) (C)	H_2O (g) (B-C=D)	Soil (g) (C-A=E)	FMC gH_2O/g_{soil} (D/E)
1	4.4477	19.5313	13.5512	5.9801	15.0836	0.3965
2	4.4542	19.3130	13.7279	5.851	14.8588	0.3759
3	4.4522	19.2567	13.8067	5.4500	14.8045	0.3681
4	4.4110	20.2806	14.5035	5.7771	15.8696	0.3640
5	4.3722	19.2043	13.5992	5.6051	14.8321	0.3779
6	4.3954	20.3153	14.4314	5.8839	15.9199	0.3696
7	4.3820	19.8066	14.1611	5.6455	15.4246	0.3660
8	4.4509	19.5335	13.9538	5.5797	15.0826	0.3699
9	4.4739	20.2556	14.6518	5.6038	15.7817	0.3551
10	4.4973	19.9690	14.4997	5.4693	15.4717	0.3535

MEAN FMC = 0.3696
S = 0.01224
CV% = 3.3%

Particle size analysis

The pipette method for determining particle size distribution (Day 1965) was used with some minor modifications. These modifications were made in order to overcome a shortage of equipment as a result of the relatively large numbers of samples to be analysed.

Firstly, no sodium acetate was added to dissolve any lime ($CaCO_3$) present. This was considered unnecessary as all the soils being analyzed had pH's under pH7 and so Ca levels would not have affected the analysis (J Hutson, personal communication 1985) Secondly, the soil was not treated with hydrogen peroxide to remove organic matter as this is time consuming and often results in a loss of samples and therefore in further delays.

These two modifications are in any event likely to have had the same effect on all samples (J Hutson, personal communication 1985), and as an index of relative differences is required for this study, a consistent procedure is more important than the detailed method leading to an absolute determinations of particle size.

Variability

No standards were run for the soil tests carried out by the author. However, any pair of duplicates that differed by more than 10 units were re-analysed.

REFERENCES

See main reference list on PAGE 127 together with the addresses for personal communications on PAGE 133 of the main document.