BASELINE OF SELECTED ESSENTIAL NUTRIENT ELEMENTS OF AN INDIGENEOUS FRUIT TREE (*MIMUSOPS ZEYHERI*) UNDER NATURAL CONDITIONS

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DECLARATION

I hereby declare that the work herein submitted as a dissertation for the degree Masters of Agricultural Management is the result of my own investigation and that it has neither wholly nor partially been presented as a dissertation for the degree in this University or elsewhere. Work by other authors which served as sources of information has duly been acknowledged by reference to the authors.

SIGNED BY STUDENT: DATE:

…… ……

MS. C. R. LEDWABA

DEDICATION

This work is dedicated to my mother Lydia Lenkwe Ledwaba; whose love, support and patience gave me the strength to trot on.

ACKNOWLEDGEMENTS

The author likes to thank God for His mercy upon her life. Giving respect and appreciation to her family that stood by her throughout the years. Many thanks go out to Professor Mashela for his supervision, encouragement and support. Professor Ayisi's support helped a lot in compiling of this thesis, especially with statistical analysis of the results. Lastly, the author also gives her appreciation to the National Research Foundation and the Department of Water Affairs and Forestry for financial support.

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ABSTRACT

The mineral nutrition of indigenous crop species is not well documented like other known crop species, thus making it difficult for one to know how to plant and maintain the crops. Mmupudu (M*imusops zeyheri*), which, happens to be a wild crop, is one of the indigenous trees of interest to the Discipline of Plant Production, University of Limpopo. The current study gives baseline information that will be important in various environmental physiology studies of this plant. Physiological studies will be necessary to assess the importance of "limiting" mineral nutrients in the accumulation of certain mineral nutrients in Mmupudu in relation to its productivity.

The experiment was arranged as a 2 x 3 factorial in RCBD, with the first and second factors being time of sampling and location, respectively. The three locations where data were collected were Chuenespoort, Bochum and Sekgosese. In each location, the experiment was replicated 10 times. Data were analyzed using ANOVA and means were separated using the least significant difference test.

The two-factor interaction was nonsignificant ($P \geq 0.10$) for both pH and electrical conductivity. Soil pH was not affected by time in all three locations suggesting that abscised flowers and fruitlets have no effect on pH. Leaf K experienced an increase of 65% at Chuenespoort and a decrease of soil K after fruiting by 44%. Leaf and soil P decreased after fruiting in all locations as was the case with Cu. Chuenespoort and Sekgosese experienced a decrease in leaf Mn after fruiting while soil Mn decreased in all

locations after fruiting. Sekgosese experienced an increase of 25% soil Mg after fruiting, whereas leaf Mg decreased in all locations.

CHAPTER 1
GENERAL INTRODUCTION

1.1 Introduction

The ability of plants to interfere successfully for nutrients, CO_2, light, water and temperature, determines their sustainable growth and abundance. In addition to the listed environmental factors, both internal and external, those that affect plant growths are complicated (Lochhart, 1965; Salisbury, 1975). Except for carefully managed orchards, most indigenous trees grow on marginal soils under intense interference with other plant species.

Under natural conditions, the abscission of leaves from deciduous fruit trees during autumn returns some of the absorbed nutrient elements to the soil (Salisbury, 1975). Also, in fruit trees, large amounts of flowers and fruits are abscised, thus returning most of the essential mineral nutrients into the soil. The abscission effect may result into the cyclic sustainability of nutrient elements under natural systems, with more or less equilibrium in soil and plant nutrient element contents (Lochhart, 1965). Thus, quantifying soil and plant nutrient elements may provide baseline information on the nutrient element requirements in undisturbed indigenous fruit trees.

Various indigenous plants are being evaluated as alternative crops in Limpopo Province, Republic of South Africa (Mashela and Mollel, 2001). Indigenous fruit trees were ranked

using edible qualities in fresh and/or beverage form. *Mimusops zeyheri* (Mmupudu = Sepedi, Moepel = Afrikaans, Transvaal red milkwood = English) was top-ranked for its edible fresh fruit qualities (Mashela and Mollel, 2001). Farmers in all six regions indicated that there was a great need for the domestication and commercialization of *M. zeyheri* trees. Eighty-three percent of farmers indicated that an average of 1.3 ha could be made available in their fields for cultivation of *M. zeyheri*.

Generally, *M. zeyheri* trees grow on wooded, rocky hillsides or river basins (Venter and Venter, 1996). The trees have a non-aggressive lateral root system, which is adapted to rocky conditions and marginal soils (Van Wyk, 1974). *Mimusops zeyheri* also have attributes that are associated with drought tolerance (Venter and Venter, 1996). In most drought tolerant plant species, more growth is partitioned towards root growth (Krieg, 1983). Thus, *M. zeyheri* is a slow grower.

Maila (2005) evaluated protocols for *in-vitro* propagation of *M. zeyheri*. In this study, clonal rootstocks were produced, which eliminate any genetic variation that is associated with seedling rootstocks. Maputla (2002) investigated genetic diversity of *M. zeyheri* in Limpopo Province. Cluster analysis using unweighted pair-group method with arithmetic means separated individual *M. zeyheri* bans into distinct clusters with average genetic similarity estimates ranging from 47% to 89%. The analysis showed that 91% genetic variance existed among populations of *M. zeyheri* in Limpopo Province, whereas within population variation was 9%. This was the first report on genetic variability and the partitioning of genetic diversity within and among populations of *M. zeyheri*. The data

provided important baseline information for future species identification and classification in an envisaged breeding program.

Sandy soil reduced productivity of seedlings, whereas loam and clay soils had no effect on various parameters (Ndhukula, 2006). *Mimusops zeyheri* seedlings did not respond to various $C\ell^-$ and Na^+ containing salt types, with the exception of sodium carbonate (Nchabeleng, 2004).

The physiological action of genes determines plant morphology, response to environmental conditions and yielding ability. However, the environment also plays a major role in determining the morphology of a plant.

1.2 Problem Statement

The nutrient element requirements of indigenous trees are not known. The researcher proposed to use *M. zeyheri* trees, in three growing regions of Limpopo Province to determine the limiting nutrient elements in this indigenous plant.

1.3 Motivation

Mimusops zeyheri is a popular indigenous fruit tree in Limpopo, South Africa, especially in rural areas and is known to have high vitamin C (Venter and Venter, 1996). Baselining the nutrient element requirements of this tree will facilitate its domestication to serve as a source of vitamin C, more especially in rural communities.

1.4 Aim and objectives

1.4.1 Aim

To baseline the selected essential nutrient elements of *M. zeyheri* under natural conditions

1.4.2 Specific objectives

a. To determine selected essential nutrient elements in leaves of *M. zeyheri* trees

b. To determine selected nutrient elements in the drip areas of *M. zeyheri* trees

c. To determine soil pH and EC in the drip areas of *M. zeyheri* trees

d. To determine soil factors that influence nutrient elements in *M. zeyheri* leaves

1.5 Hypothesis

a. Selected essential nutrient elements in the leaves of *M. zeyheri* trees do not differ.

b. Selected essential nutrient elements in the soil of the drip area of *M. zeyheri* trees do not differ.

c. Soil pH and EC in the drip areas of *M. zeyheri* trees do not differ

d. Soil factors have no influence on essential nutrient elements in *M. zeyheri*

CHAPTER 2
LITERATURE REVIEW

2.1 Introduction

In most environmental physiology studies, researchers are interested in the response of a particular experimental organism under specified artificial conditions. For instance, the response may be in relation to pest attack, salt stress or any other controlled factors (Mashela, 2007). The aim of environmental physiology studies is to derive data that can be analyzed statistically, with little or no interest in what the results might imply about plants under natural environments.

Environmental physiology is mainly concerned with plants in natural or cultivated ecosystems. In environmental physiology, it is important to establish a proper balance between natural and artificial ecosystems. In most studies of *M. zeyheri,* data has mainly been collected under artificial environments (Maila, 2005; Maputla, 2002; Nchabeleng, 2004; Ndhukula, 2006). It is natural to find oneself becoming much more involved in either artificial or natural systems. This should be controlled because unbiased answers certainly emerge when a suitable balance between artificial and natural ecosystems is not established.

The guiding concept in field research is to measure and then apply deductive logic to the analysed data (Salisbury, 1975). Many environmental variables may be measured, and then statistical methods are applied to identify factors which have a bearing in plant

growth, whether positive or negative. Controlled experiments may, on the other hand, hold particular variables constant, except one variable of interest (Petersen, 1994). The latter may be allowed to vary and then held responsible for the observed plant responses. The focus of this study was to baseline the environmental physiology of *M. zeyheri* under natural conditions, using the nutrient element content in leaves and in soil collected under the tree canopy.

2.2 Plant environmental physiology

Environmental physiology is a broad field. However, in this study, the scope of the review was limited to ten basic principles of environmental physiology. Most of the basic principles as reviewed, hereafter, will be helpful in discussing the data derived in subsequent trials of the study.

2.2.1 Saturation point

The most fundamental principle of plant response to the environment is the saturation point which was borrowed from chemistry. Various biotypes respond to various environmental factors according to a constant characteristic pattern (Seinhorst, 1975). When the factor reaches a threshold above which it begins to have an effect, the response increases until the system becomes saturated by the factor (Mohr, 1972; Seinhorst 1975). Then, as the intensity of the factor continues to increase, the response remains either constant or begins to decrease if the factor at its high level becomes toxic or inhibitory or competitive (Levitt, 1972; Mohr, 1972; Seinhorst, 1975). Saturation at inhibiting or competing levels is generally documented for temperature, photosynthesis, mineral

nutrition, enzyme action, ion transport across membranes, salt stress, pathogens and plant responses to hormones (Highkin and Lang, 1966; Mohr, 1972; Salisbury, 1975; Vernberg, 1975; Seinhorst, 1975).

In systems that are amenable to saturation points, it is possible to speak of three cardinal points: minimum, optimum and maximum. In classical studies Went (1957) used plant responses to hormones to demonstrate that biotypes use the factor being provided until their capacity to use it is saturated. Seinhorst (1975), using the 1933 Nicholus' model demonstrated that after a saturation point, where initial number of nematodes (Pi) equaled final number of nematodes (Pf), there was a decline in reproductive potential of nematodes.

The law of minimum was derived and understood from the saturation curves (Salisbury, 1975). This law states that: "The growth of a plant is dependent upon the amount of a growth factor that is presented to it in minimum quantities".

Salisbury (1975) illustrated the concept of a "limiting factor" with a simple experiment in mineral nutrition, involving two levels of P which were given to plants over a wide range of N concentrations. In the study, the threshold for N was kept low, and below that level plants did not grow. When N was gradually increased, plants responded the same at both P levels until P became limiting at the N saturation level. Higher P levels resulted in higher N saturation levels.

The practical implications of the law of minimum continue to be important in crop production systems. In crop production, the challenge is to identify the limiting nutrient element and then improve it. For instance, if plants are limited in their growth by the deficiency of N, more N fertilizer needs to be applied in order to increase yield. However, when enough N has been applied, another element, for example P, might become limiting and then needed to be applied. In environmental physiology, a plant's distribution might be limited at its boundary by some single environmental factor presented to it in the "limiting" amount. However, because plants require highly disparate quantities of different nutrient elements and environmental factors, the "limiting factor" is always an insignificant issue due to the smothering effect.

2.2.2 Interaction of factors

In nature, factors do not operate in a simplistic way as the law of minimum suggests. Under controlled conditions such as those of Salisbury (1975), everything might work as the law predicted. In nature, the curves may not be identical in the ascending parts where only one factor is supposed to be limiting.

Salisbury (1975) demonstrated that for fresh fruit weight, curves in the "limiting" N levels were not exactly superimposed to those of P. Salisbury and Ross (1978) provided several explanations for the failure of the law of minimum in Salisbury's (1975) example. Generally, the extent to which the law of minimum might function in multiple-factor conditions depends upon the extent to which the factors under consideration are able to interact within the plant. The inability of a factor to interact may depend on a number of

factors. For instance, restrictions upon diffusion of CO_2 through stomata or movement of ions in the apoplast may alter the chemical rate for various reactions in Krebs and Calvin cycles (Campbell, 1990).

The law of minimum guides assessments in environmental physiology, and it serves as a good starting point in this complex field. One can start one's studies of nutrient element accumulation or plant distribution by identifying the "limiting factors". However, one has to progress beyond these initial steps in order to have a clue of conditions through the use of controlled experiments.

Powerful mathematical tools have been developed to enhance the understanding of factor interactions in controlled experiments (Petersen, 1994). Generally, where an experiment cannot be carefully designed in advance, analysis of variance is inappropriate. These tools indicate whether or not factors interact to produce a response. When the two factors, each influencing separate responses do not interact, the factors may be additive in their effects or they may be multiplicative (Mohr, 1972). When two factors have additive effects, they act upon different casual sequences that lead to the response. For example, when a given compound is made in two different compartments in the cell, one factor may influence one compartment and another factor the other compartment (Mohr, 1972). Multiplicative responses occur when two or more factors act on different steps in the same causal sequence (Mohr, 1972). Analysis of variance tells us whether responses are additive (i.e. when there are interactions) or when they are multiplicative (i.e. when there is no interaction). One of the useful tools, which is independent of the design, a stepwise

regression analysis, may serve as a valuable tool in field observations where limiting factors are being identified.

2.2.3 Noninductive environmental responses

Noninductive environmental responses, when an environmental factor changes, the plant response changes directly and almost immediately (Salisbury, 1975). For instance, the rate of photosynthesis changes within seconds as light intensity changes. Transpiration is a comparable example, however, this is somewhat complicated by the stomatal responses, which may be a little less immediate and noninductive (Lang, 1961). Most enzyme-controlled reactions respond directly to temperature and in some cases, to the presence of specific molecules in their environment (Lehninger, 1979).

2.2.4 Triggered induction responses

Certain plant responses to the environmental change may proceed even if the environment returns to its initial state. This is referred to as triggered response, or on-off response or inductive response (Salisbury and Ross, 1978). Usually, there is a delay between the inductive action and the response itself. Frequently, there is also amplification. Amplification in triggered responses occurs when the energy supplied to the plant by the environmental change is well below the energy required to bring about the response. Thus, the energy for such a response is provided by the plant's metabolism. An example of a triggered response is seed germination that is triggered by light (Highkin and Lang, 1966). In a given seed, when the absorbed light reaches a point above the threshold level for response, germination begins and will continue even if the triggering

light is removed. Examples of triggered responses are rare, and most responses that were previously classified as triggered responses are actually modulated responses (Salisbury, 1975).

2.2.5 Modulated responses

Modulated responses are also referred to as quantitative responses (Campbell, 1990). A modulated response is clearly not an on or off. However, the level of response is determined by the level of the environmental factor. The existing amplification is proportional to the environmental input. Also, like in triggered responses, there is a delay in the response of modulated responses. Phototropism is a good example of a modulated response. The degree of bending of a stem is a function of the number of quanta of light absorbed by the coleoptiles, but the light does not supply the energy for bending (Went, 1957). Another simple example of modulated response is the accumulation of carbohydrates in roots when the root/shoot ratio is reduced through root pruning (Mashela, 2007). Thus, the energy for this response is provided by the plant's metabolism.

In modulated responses, when the response to the environmental stimulus is lengthy, the response is referred to as inductive response (Went, 1957). Flowering, a classical example of inductive response, occurs long after the plant has been exposed to vernalization. Numerous plant responses are time-related. For instance, in flowering, dormancy, tuber formulation and other photoperiodic responses, the time of light

interruption during the dark period is very important (Salisbury, 1975). Similarly, time of light application in circadian rhythms is also an important controlling factor.

2.2.6 Homeostasis and feedback

Homeostasis is the principle whereby body mechanisms strive to maintain a constant inner environment through self-regulation (Campbell, 1990). In animals, body temperature, blood chemistry and other many parameters tend to meet the homeostasis principle. Although highly documented in animals, the homeostasis principles also apply to plants. Mechanisms such as stomatal regulation of transpiration and maintenance of various hormone levels are amenable to the homeostasis principle (Campbell, 1990).

Generally, homeostasis is achieved through feedback mechanisms (Campbell, 1990). It has been established that feedback is the primary manner through which homeostasis is achieved. For instance, CO_2 levels control stomatal apertures, whereas products of reactions affect reaction rates by acting allosterically on the enzymes that catalyze their production (Fitter and Hay, 1987).

2.2.7 Conditioning effects

In conditioning effects, there are no delay responses. The change is gradual and occurs in response to continuous application of the environmental factor. Examples of conditioning effects include the development of frost tolerance and drought tolerance in plants (Salisbury and Ross, 1978). In pea seeds, the germination temperature determines the

nature and extent of subsequent seedling growth (Highkin and Lang, 1966). Although this is not certain, it may also be an example of a conditioning effect.

2.2.8 Carryover effects

Genetically pure inbred peas grew poorly when day/night temperatures were held equal and constant (Highkin and Lang, 1966). When peas were grown up to the fifth generation under equal and constant day/night temperatures, each generation grew more poorly than the previous one. When the situation was reversed by raising germinated seeds from stunted plants under optimum conditions, Highkin and Lang (1966) noted that it required at least three generations of harvesting and replanting to reach the optimum level of growth. The two authors proposed that developing embryo was in some way conditioned by the environment so that the effect was carried over through a number of succeeding generations. However, this carryover of environmental effects from one generation to the next is contrary to known and established concepts of genetics (Campbell, 1990).

2.2.9 Role of genes

Both environment and genetics are important in the expression of phenotypes in plants (Campbell, 1990). Adoptive effects of the environment on plant morphology and physiology are common. Plants having similar genetic, but exhibiting differences due to different natural environments are referred to as ecophenes (Salisbury and Ross, 1978). The environment can and does produce different ecophenes from any uniform genetic stock. The effects of temperature, light, nutrient elements and other environmental factors

on growth and development of plants are extensively documented in Levitt‘s (1972) reviewes of the topic.

Genetic differences may occur in representatives taken from the different areas of species’ distribution. Similar populations which contain different genetic make-ups are called ecotypes (Campbell, 1990). Various ecotypes exist and it is now apparent that different environments exert different selection pressures, which result in different genetic compositions that are directly correlated with location.

Selection pressures also work for the physiological responses to the environment. For instance, photoperiodic ecotypes have been demonstrated in several species. The carryover effects could be a significant confounding effect in studying plants of the same species collected from different locations. Observed differences in the first three generations could be due to carryover effects rather than genetics. This possibility should not be ignored when comparing *M. zeyheri* seedlings where seeds were collected from different geographic locations.

2.2.10 Allelochemicals

An allelochemical is a chemical produced by one plant that influences another plant of a different species (Whittaker, 1975). The production by plants of allelochemicals that are harmful to other plants referred to as allelopathy is well documented (Inderjit and Keating, 1999).

Fungi produce antibiotics that are effective against bacteria. Also, higher plants produce allelochemicals that suppress growth of other plant species. The *Juglans* species produce a quinone called juglone, which inhibit the germination and root growth of other plants (Inderjit and Keating, 1999). Generally, when allelochemicals induce sickness or death to animals, they are termed poisons.

Some plants have allelopathic potential by releasing allelochemicals to their surroundings that have either deleterious or beneficial effects on other plants in the vicinity. Mashela (2005) demonstrated that *Casuarin cunninghamiana*, which is a widely used windbreak tree, released substances that are allelopathic to citrus trees. Autotoxicity, allelopathic effects among plants of the same species, has been documented in a number of plant species and is believed to be involved in peach short life and citrus replant problems (Inderjit and Keating, 1999). In cucumber, it was established that plant growth was inhibited by its own root extract and root exudates, but improved by removal of these substances from the rhizosphere (Yu, 1999). Allelopathic interference is due to the stifling of various physiological processes, which include ion uptake and water uptake (Inderjit and Keating, 1999).

CHAPTER 3
BASELINE OF NUTRIENT ELEMENTS IN *MIMUSOPS ZEYHERI* LEAVES

3.1 Introduction

The law of minimum, modulated responses and interactions were singled-out as principles of environmental physiology that could apply to nutrient element accumulation in plants. In all three responses, time plays a major role. In this study, the nutrient content of M*imusops zeyheri* in both leaves and within soil canopy was baselined using time-location factors.

3.2 Material and Methods

3.2.1 Location

The experiment was initiated on 11 October 2004 at three climatologically different districts of Limpopo Province. Leaf sampling was done during fruiting at Bochum (23° 16’ 59" S; 29° 8' 5" E) on 11 October 2004, Chuenespoort (24 ° 21' 4" S; 29° 48' 4" E) on 12 October 2004 and Sekgosese (23° 37' 8" S; 30° 4' 7" E) on 13 October 2004 and after fruiting on 24 March 2005, 25 March 2005 and 26 March 2005, respectively.

3.2.2 Experimental design

The experiment was arranged as a 2 x 3 factorial in a randomized complete block design (RCBD), with the first and second factors being time of sampling and location, respectively. The trees were blocked for slope and marked for future sampling.

3.2.3 Data collection

Ten representatives at Chuenespoort, Bochum and Sekgosese were chosen and the profile pits prepared underneath the tree crowns in order to establish soil form. A square pit was dug to 1 m deep with the centre of the square being 2 m from the crown of the tree. The pit was left for 10 minutes to allow the sides to dry. When the sides were dry, the soil was cut using a spade and thrown out separately and carefully examined for structure. Three soil samples were collected from 25, 50, and 75 cm depth respectively from each horizon. Samples were transported to the laboratory for analysis of electrical conductivity (EC) and pH.

Five matured and healthy leaves from fruit-bearing branches were sampled from each of the four cardinal positions of the tree. Soil samples were collected from the cardinal points at 0.5 m from the trunk in the drip area using a 2-cm-diameter auger. The 10 cores per tree were mixed in a plastic bag and labeled. In the laboratory leaf samples were oven-dried for 72 hours at 57^{o} C using air-forced ovens, and ground in a Wiley mill to pass through a 1 mm pore sieve. Soil samples were shade dried and passed through a 2 mm pore sieve. Leaf and soil samples were analysed for EC, pH and nutrient elements using the standard laboratory techniques.

a. Soil EC and pH

Fifteen grams soil sample was incubated in 75 mℓ distilled water and shaken for 1 hour at 175 cycles per minute (cpm). The solution was filtered using Whatman no. 42 into 100

mℓ beakers and EC of the filtrates was measured with EC meter (model WTW CF 318) using Longenecker and Lyerlys 1964 method. Five grams soil sample was incubated in 25 mℓ of distilled water plus 75 mℓ KCℓ for 50 minutes while being stirred and incubated for 10 minutes prior to measuring pH using a pH meter (model 420 A).

b. Soil nutrient elements

Copper, Fe, Mn and Zn in the soil samples were quantified by suspending 10 g of soil samples in 20 mℓ DTPA solution in the extracting bottles and shaken for 2 hours at 180 cycles per minute (Beyers *et.al*, 1971). The solution was filtered using Whatman no. 42 and filtrates were used to quantify Cu, Fe, Mn and Zn concentrations using Perkin Elmer atomic absorption spectrophotometer 3110 (Page *et al*., 1982).

The macronutrients were quantified using the Schollenberger and Simon 1945 method, which is briefly outlined. Five grams soil sample were suspended in ammonium acetate and shaken for 30 minutes and filtered using Whatman no. 42. Ten-milliliter filtrates were pipetted into 100 mℓ volumetric flasks, and de-ionized water added to the mark. Potassium, Mg, and Na were quantified with Perkin Elmer atomic absorption spectrophotometer 3110.

c. Leaf tissue nutrient elements

Ground leaf at 2 g per plant were ashed in a furnace at 550^{o} C for 24 hours and 10 mℓ solution of 1:1 (v/v) de-ionized water and hydrochloric acid was added to the ash and dried in a steam bath (Page *et al*., 1982). The resultant mixture was then transferred into

100 mℓ volumetric flasks, filled up to the mark with de-ionized water and filtered with Whatmann no. 42. Copper, Fe, Mn, and Zn were quantified using Perkin Elmer atomic absorption spectrophotometer 3110. The 2 mℓ filtrate was pipetted into 100 mℓ volumetric flasks, and de-ionized water added up to the mark. Potassium, Mg and Na were quantified using Perkin Elmer atomic absorption spectrophotometer 3110. Leaf P was quantified as described for soil P. To determine soil P, 10 mℓ of nitric acid was added in 5 mℓ of the filtrate solution in 100 mℓ beakers and dried in a steam bath. Ten millilitres nitric acid, ammonium molybdate and ammonium vanadate were each added in the filtrate and the whole solution was transferred into 100 mℓ volumetric flasks and water added up to the mark (Milton, 1992). The soil P was quantified using a Spectronic 301 spectrophotometer.

3.2.4 Data analysis

The data were analyzed using analysis of variance and evaluated for time x location interactions (Petersen, 1994). When interactions were significant ($P \leq 0.05$), the data were presented using a two-way table, along with the standard error of the means. However, when interactions were not significant ($P > 0.05$), mean comparison was attained using least significant difference test.

3.3 Results

(a) Soil pH and EC

Location had significant effects on soil pH and soil EC, while time has a significant effect on EC only (Table 3.1). Location contributed 27% and 16% to the total treatment

variation (TTV) in soil pH and EC, respectively. Time had no effect on the TTV in soil pH, but contributed 33% to the TTV in EC. Location x time interactions were not significant ($P > 0.05$) for both pH and EC. Soil pH was significantly high in Bochum compared to Sekgosese and Chuenespoort (Table 3.2). The EC values were significantly high in Chuenespoort, compared to Bochum and Sekgosese ($P \leq 0.05$).

Table 3.1 Variation of soil pH and electrical conductivity from the drip areas of *Mimusops zeyheri* in the three regions of the Limpopo Province.

		pH-H_2O			EC (dS/m)		
Source	Df	SS	%		SS	%	
Replication (R)	9	2.89	11.0	ns	0.01	4.7	ns
Location (L)	2	7.48	27.0	***	0.04	16	**
Time (T)	1	0.24	0.9	ns	0.09	33	***
L × T	2	0.10	0.4	ns	0.01	1.3	ns
R × L × T	45	16.51	61.0	ns	0.11	44	ns
Total	59	27.22			0.26		

ns Not significant at $P \geq 0.10$; * significant at $P \leq 0.10$; ** significant at $P \leq 0.05$; *** significant at $P \leq 0.01$.

The soil pH during fruiting and after fruiting did not differ, whereas soil EC after fruiting was higher than that during fruiting by 78% i.e. [(0.16/0.09-1) x 100 = 77.7%](Table 3.3). Location alone had a significant effect on leaf and soil K (Table 3.4). Location contributed 23% and 32% to the TTV in the content of K in leaves and soils, respectively. Time had no effect to the TTV of K in leaves or soils. Location x time interaction was

highly significant for leaf K and contributed 11% to the TTV, whereas the interaction for soil K only was slightly significant ($P \leq 0.10$).

Table 3.2 Soil pH and electrical conductivity from drip areas of *Mimusops zeyheri* in three regions of Limpopo Province.

Region	pH-H_2O	EC (dS/m)
Chuenespoort	6.85 b	0.17 a
Bochum	7.28 a	0.12 b
Sekgosese	6.42 c	0.10 b
LSD (0.05)	0.39	0.03

Column means (n = 10) with the same letter are not different at $P \leq 0.05$ according to the least significant difference test. Ns = not significant.

Table 3.3 Soil pH and electrical conductivity from drip areas of *Mimusops zeyheri* in two seasons across three regions.

Season	pH-H_2O	EC (dS/m)
During fruiting	6.79 a	0.09 b
After fruiting	6.91 a	0.16 a
LSD (0.05)	0.32	0.02

Column means (n = 10) with the same letter are not different at $P \leq 0.05$ according to the least significant difference test. Ns = not significant.

Leaf K was the highest after fruiting in Chuenespoort, but did not differ from that of during fruiting in Bochum and Sekgosese (Table 3.5). On the other hand, soil K had no significant difference during and after fruiting in all regions.

Location had a significant effect on soil P (Table 3.6). Both location and time had a significant effect ($P \leq 0.10$) on leaf P, contributing 4% and 11% to the TTV, respectively. Location x time interaction contributed 11% to the TTV of P leaves.

Leaf P at Bochum increased after fruiting, whereas at Sekgosese and Chuenespoort it remained the same (Table 3.7). Sekgosese had the lowest soil P after fruiting which decreased by 43%, whereas those in Bochum and Chuenespoort remained unchanged.

Table 3.4 Variation of potassium in leaves of *Mimusops zeyheri* and soil from drip areas in three regions of Limpopo Province over two seasons.

		Leaf K (%)			Soil K (ppm)		
Source	Df	SS	%		SS	%	
Replication (R)	9	1.07	18	*	129498	15	ns
Location (L)	2	1.34	23	***	272444	32	***
Time (T)	1	0.03	0.5	ns	4267.27	0.5	ns
L × T	2	0.66	11	***	57465.7	7	*
R × L × T	45	2.70	47		387735	46	
Total	59	5.79			851408		

ns Not significant at $P \geq 0.10$; * significant at $P \leq 0.10$; ** significant at $P \leq 0.05$; *** significant at $P \leq 0.01$.

Table3. 5 Leaf and soil potassium from drip areas of *Mimusops zeyheri* in three regions of Limpopo Province during two seasons.

Variable	Season	Region		
		Chuenespoort	Bochum	Sekgosese
Leaf K (%)	During fruiting	0.62 b	0.65 a	0.92 a
	After fruiting	1.02 a	0.47 a	0.85 a
	LSD (0.05)	0.23	ns	ns
Soil K (ppm)	During fruiting	237.70 a	285.40 a	332.60 a
	After fruiting	133.30 b	312.20 a	359.60 a
	LSD (0.05)	117.95	ns	ns

Column means (n = 10) with the same letter are not different at $P \leq 0.05$ according to the least significant difference test. Ns = not significant.

Table 3.6 Variation of phosphorus in leaves of *Mimusops zeyheri* and soil from drip areas in three regions of Limpopo Province over two seasons.

Source	Df	Leaf P (%)			Soil P (ppm)		
		SS	%		SS	%	
Replication (R)	9	0.004	7	ns	1135.29	12	ns
Location (L)	2	0.006	11	*	4615.23	49	***
Time (T)	1	0.002	4	*	14.1130	0.2	ns
L × T	2	0.006	11	*	148.252	1.6	ns
R × L × T	45	0.037	67		3488.45	37	
Total	59	0.56			9401.34		

ns Not significant at $P \geq 0.10$; * significant at $P \leq 0.10$; ** significant at $P \leq 0.05$; *** significant at $P \leq 0.01$.

Table 3. 7 Soil phosphorus from drip areas of *Mimusops zeyheri* in three regions of Limpopo Province in two seasons.

Variable	Season	Region		
		Chuenespoort	Bochum	Sekgosese
Leaf P (%)	During fruiting	0.13 a	0.08 b	0.09 a
	After fruiting	0.09 a	0.80 a	0.10 a
	LSD (0.05)	ns	0.3	ns
Soil P (ppm)	During fruiting	24.66 a	2.57 a	3.88 a
	After fruiting	19.85 a	2.27 a	2.23 b
	LSD (0.05)	ns	ns	1.57

Column means (n = 10) with the same letter are not different at P≤ 0.05 according to the least significant difference test. Ns = not significant.

Location had a significant effect on leaf and soil Mg and contributed 23% and 8 % to the TTV, respectively (Table 3.8). Time had no significant effect on both leaf Mg and soil Mg. Location x time interaction contributed 12% to the TTV of leaf Mg.

Chuenespoort, Bochum and Sekgosese experienced a decrease in leaf Mg after fruiting by 29%, 55% and 90%, respectively whereby Sekgosese experienced the highest decrease (Table 3.9).

Location had a significant effect on leaf Mn (Table 3.10). Time had no significant effect on both leaf Mn and soil Mn. Loaction x time interactions had a significant effect on both leaf and soil Mn contributing 14% and 44% to the TTV, respectively.

Leaf Mn content decreased after fruiting at Chuenespoort whereas at Bochum it increased (Table 3.11). Chuenespoort and Bochum experienced a decrease in soil Mn after fruiting whereas it increased at Sekgosese.

Location had a significant effect on Leaf Zn (Table 3.12). Location and time had no significant effect on soil Zn. Location x time interaction was highly significant on leaf Zn and contributed 35% to the TTV.

Leaf Zn was the highest at Chuenespoort after fruiting (Table 3.13). Bochum experienced a decrease in leaf and an increase in soil Zn after fruiting. Soil Zn was not affected by season at Chuenespoort and Sekgosese.

Table 3.8 Variation of magnesium in leaves of *Mimusops zeyheri* and soil from the drip areas in three regions of Limpopo Province over two seasons.

		Leaf Mg (%)			Soil Mg (ppm)		
Source	Df	SS	%		SS	%	
Replication (R)	9	0.02	4	ns	334 227.0	20	ns
Location (L)	2	0.11	23	***	129 180.0	8	*
Time (T)	1	0.02	3	ns	21 736.1	1	ns
L ×T	2	0.06	12	**	83 654.4	5	ns
R × L × T	45	0.28	58		1 099 822.0	66	
Total	59						

ns Not significant at $P \geq 0.10$; * significant at $P \leq 0.10$; ** significant at $P \leq 0.05$; *** significant at $P \leq 0.01$.

Table 3.9 Leaf magnesium from drip areas of *Mimusops zeyheri* in three regions of Limpopo Province in two seasons.

Variable	Season	Region		
		Chuenespoort	Bochum	Sekgosese
Leaf Mg (%)	During fruiting	0.41 a	0.93 a	0.30 a
	After fruiting	0.29 b	0.42 a	0.03 a
	LSD (0.05)	0.11	ns	ns
Soil Mg (ppm)	During fruiting	621.40 a	495.70 a	632.30 a
	After fruiting	611.50 a	531.70 a	474.00 b
	LSD (0.05)	ns	ns	154.80

Column means (n = 10) with the same letter are not different at $P \leq 0.05$ according to the least significant difference test. Ns = not significant.

Table 3.10 Variation of manganese in leaves of *Mimusops zeyheri* and soil from the drip areas in three regions of Limpopo Province over two seasons.

Source		Laef Mn (ppm)		Soil Mn (ppm)	
	Df	SS	%	SS	%
Replication (R)	9	22633.7	2 ns	37.26	3 ns
Location (L)	2	436688	47 ***	70.54	6 *
Time (T)	1	493.07	0 ns	17.40	2 ns
L × T	2	129844	14 ***	472.98	44 ***
R × L × T	45	344205	37	481.98	45
Total	59	933864		1080.15	

ns Not significant at $P \geq 0.10$; * significant at $P \leq 0.10$; ** significant at $P \leq 0.05$; *** significant at $P \leq 0.01$.

Table 3.11 Leaf and soil manganese from drip areas of *Mimusops zeyheri* in three regions of Limpopo Province in two seasons.

Variable	Season	Region		
		Chuenespoort	Bochum	Sekgosese
Leaf Mn (ppm)	During fruiting	207.60 a	227.60 b	56.60 a
	After fruiting	76.80 b	319.80 a	75.04 a
	LSD (0.05)	73.79	84.36	ns
Soil Mn (ppm)	During fruiting	10.9 a	6.04 a	4.48 b
	After fruiting	2.74 b	3.89 b	10.75 a
	LSD (0.05)	5.19	1.58	1.71

Column means (n = 10) with the same letter are not different at $P \leq 0.05$ according to the least significant difference test. Ns = not significant.

Table 3.12 Variation of zinc in leaves of *Mimusops zeyheri* and soil from drip areas in three regions of Limpopo Province over two seasons.

Source	Df	Leaf Zn (ppm)			Soil (ppm)		
		SS	%		SS	%	
Replication (R)	9	699.350	4	ns	355.04	15	ns
Location (L)	2	2698.030	17	***	98.63	24	ns
Time (T)	1	1000.420	6	**	43.12	2	ns
L × T	2	5498.030	35	***	71.86	3	ns
R × L × T	45	5910.350	38		1767.01	76	
Total	59	15776.200			2335.66		

ns Not significant at $P \geq 0.10$; * significant at $P \leq 0.10$; ** significant at $P \leq 0.05$; *** significant at $P \leq 0.01$.

Table 3.13 Leaf zinc from drip areas of *Mimusops zeyheri* in three regions of Limpopo Province over two seasons.

Variable	Season	Region		
		Chuenespoort	Bochum	Sekgosese
Leaf Zn (%)	During fruiting	18.40 b	47.50 a	16.00 a
	After fruiting	30.80 a	13.80 b	12.80 a
	LSD (0.05)	6.19	14.93	ns
Soil Zn (ppm)	During fruiting	0.20 a	0.08 a	0.54 a
	After fruiting	0.21 a	0.37 b	0.33 a
	LSD (0.05)	ns	0.12	ns

Column means (n = 10) with the same letter are not different at $P \leq 0.05$ according to the least significant difference test. Ns = not significant.

Location and time had no significant effect on leaf Cu (Table 3.14), but they both had a significant effect on soil Cu. Location x time interaction had a significant effect on soil Cu which contributed 20% to the TTV. Leaf Cu was not affected by season in all locations (Table 3.15). Soil Cu decreased after fruiting at Bochum and Sekgosese, but increased at Chuenespoort by 19%.

Table 3.14 Variation of copper in leaves of *Mimusops zeyheri* and soil from drip areas in three regions of Limpopo Province over two seasons.

Source	Df	Leaf Cu (ppm) SS	%		Soil Cu (ppm) SS	%	
Replication (R)	9	6.67	1.65	ns	0.10	13.80	ns
Location (L)	2	0.23	0.58	ns	0.14	20.01	***
Time (T)	1	0.27	0.66	ns	0.06	8.89	**
L × T	2	5.63	13.97	*	0.11	15.22	**
R × L × T	45	27.53	68.26		0.30	42.59	
Total	59	40.33			0.71		

ns Not significant at $P \geq 0.10$; * significant at $P \leq 0.10$; ** significant at $P \leq 0.05$; *** significant at $P \leq 0.01$.

Table 3.15 Leaf and soil copper from drip areas of *Mimusops zeyheri* in three regions of Limpopo Province over two seasons.

Variable	Season	Region Chuenespoort	Bochum	Sekgosese
Leaf Cu (ppm)	During fruiting	2.70 a	3.40 a	2.60 a
	After fruiting	3.00 a	2.60 a	2.90 a
	LSD (0.05)	ns	ns	ns
Soil Cu (ppm)	During fruiting	0.32 b	0.29 a	0.37 a
	After fruiting	0.38 a	0.17 b	0.25 b
	LSD (0.05)	0.09	0.7	0.08

Column means (n =10) with the same letter are not different at $P \leq 0.05$ according to the least significant difference test. Ns = not significant.

3.4 Discussion

Bochum has low rainfall, thus, explains the high pH values in the district, whereas Sekgosese with high rainfall had low pH values. The soil in Chuenespoort, with moderately low rainfall, was lying between that of Bochum and Sekgosese. The soil pH values were within the normal crop pH values of 6 and 7. It appears that location differences were mainly due to rainfall level as opposed to soil morphology.

In all three districts soil pH was not affected by time, suggesting that the large number of flowers and fruitlets that abscised in the drip area had no effect on pH. Trace mineral availability provides an excellent example of pH interaction with nutrient availability. Soil pH has been identified as a particularly critical parameter in the regulation of micronutrient availability. At high pH values, P, Cu, Zn and Mn become less available and at low pH levels P and Mg become unavailable and Mn concentrate to toxic levels (Relf, 1997).

Electrical conductivity also plays a significant role in nutrient availability. High EC values are associated with water stress and they also temper with nutrient element uptake, fruit growth and quality in various plant species (Ehnret and Ho, 1986). Electrical conductivity was high after fruiting, suggesting that abscised materials, which occur in large numbers during fruiting, release compounds after decomposition that have an influence on soil EC. In various organometric studies, application of small amounts of plant materials into soil consistently increased soil EC (Mashela, 2007).

Leaf K was affected by both location and season. Clear patterns in changes of leaf K in terms of location or time did not exist. However, when comparing leaf K of trees in the semi-arid hot Bochum district and humid-hot Sekgosese district there was a clear increasing pattern of leaf K during both seasons. Due to the possible high level of microbial activity in Sekgosese, K in plant materials was probably rapidly released into the soil and thus, may account for high levels of K in leaves.

Generally, nutrient uptake is faster in warmer soils than in cold soils. Since most nutrients are taken up via soil solution, soil water is needed to dissolve them. In humid climates where moisture effectiveness is high, soluble materials are easily leached and in arid climates where moisture effectiveness is low, salinity occurs (White, 1997). Nutrients may be present but may first require conversion to available forms that the plant is capable of taking and utilizing. Conversion to available form is affected by soil pH, microorganisms and chemical reaction.

In all three regions, horizon A soil was the same but horizon B differed. These suggest that M. *zeyheri* can do well in different soil types (Appendix A). The soil pH of the horizons also did not differ even though at Bochum the pH was slightly higher.

CHAPTER 4
DRIP AREA FACTORS THAT INFLUENCE COMPOSITION OF NUTRIENT ELEMENTS IN *MIMUSOPS ZEYHERI* LEAVES

4.1 Introduction

Saturation curves with three cardinal points (minimum, optimum and maximum) are suitable models for studying accumulation of nutrient elements in plants. On the minimum side of the curve, the accumulation response is proportional to the concentration of the factor in the soil. During the minimum effect, the organism uses the factor being considered until its capacity for using the factor becomes optimum. After optimal concentration, the accumulation response curve descends, resulting into inhibitory effects on the plant (Salisbury and Ross, 1978).

Under natural conditions, plants absorb nutrients from the soil, leaves are abscised in drip areas, decompose and release nutrients into the rhizosphere. This results into a sustainable soil-plant nutrient cycle. Stepwise regression can be used to determine the position of the response on the saturation curves. Because on the minimum side of the curve the response increases as the concentration of the element increases, the coefficients of the elements are positive ($P \leq 0.05$). On the other hand, at optimum level, the coefficients of the factors are not contributing towards the response ($P > 0.1$) because the capacity of the plant to use the elements being considered is saturated. When the intensity of the concentration of the elements being considered is saturated, the coefficients of the elements that are contributing towards the response (in accumulation of the nutrients) become negative, thus, resulting into the reduction of the response.

Under natural conditions, nutrient elements of plants are in equilibrium. In this study, stepwise regression was used to baseline the accumulation of selected nutrient elements in the leaves of *M. zeyheri* using the concentrations of these nutrients in the soil, along with pH and electrical conductivity.

4.2 Materials and Methods

4.2.1 Location

The experiment was initiated on 11 October 2004 at three climatologically different districts of Limpopo Province. Leaf sampling was done during fruiting at Bochum on (23° 16’59" S; 29° 8’5" E) on 11 October 2004, Chuenespoort (24 ° 21’4" S; 29° 48’4" E) on 12 October 2004 and Sekgosese (23° 37 8" S; 30° 4'7" E) on 13 October 2004 and after fruiting on 24 March 2005, 25 March 2005, and 26 March 2005, respectively.

4.2.2 Experimental design

The experiment was arranged as a 2 x 3 factorial in a randomized complete block design (RCBD), with the first and second factors being time of sampling and location, respectively. The trees were blocked for slope and marked for future sampling.

4.2.3 Data collection

Ten representatives at Chuenespoort, Bochum and Sekgosese were chosen and the profile pits prepared underneath the tree crowns in order to establish soil form. A square pit was dug to 1 m deep with the centre of the square being 2 m from the crown of the tree. The pit was left for 10 minutes to allow for the drying up of sides. When the sides

were dry, the soil was cut using a spade and thrown out separately and carefully examined for structure. Three soil samples were collected from 25, 50, and 75 cm depths, respectively from each horizon. Samples were transported to the laboratory for analysis of electrical conductivity (EC) and pH.

Five matured leaves from fruit-bearing branches were sampled from each of the four cardinal positions of the tree. Soil samples were collected from the cardinal points at 0.5 m from the trunk in the drip area using a 2 cm diameter auger. The ten cores per tree were mixed in a plastic bag and labeled. In the laboratory leaf samples were oven-dried for 72 hours at 57^{o} C using air-forced ovens, and ground in a Wiley mill to pass through a 1 mm pore sieve. Leaves were analyzed for nutrient elements as described previously in Chapter 3

4.2.4 Data analysis

The during and after fruiting data were analyzed using stepwise regression. In both cases, leaf nutrient elements were regressed against soil Ca, Cu, K, Fe, Mn, Na, Zn, P, pH-(H_2O) and EC. Unless stated otherwise, treatment effects were significant at the probability level of 5 %.

4.3 Results

Accumulation of six essential nutrient elements, Zn, P, Mn, Mg, K and Cu were influenced by the physico-chemical properties of the soil.

4.3.1 Leaf zinc

During fruiting, the soil pH and all other soil nutrients except Cu, Fe, K and P were optimal for the accumulation of Zn because their individual coefficients were not significant ($P \geq 0.05$). Soil Cu, Fe, K and P as shown by their negative coefficients, were at saturation level for the accumulation of Zn in leaves of *M. zeyheri* (Table 4.1) The model explained 45% of the total treatment variation (TTV) in the accumulation of Zn in leaves of *M. zeyheri*.

After fruiting, all variables except Cu and K, were eliminated suggesting that they were optimal for the accumulation of Zn in leaves, thus, it had a positive coefficient. On the other hand, soil K had reached a saturation level and was no longer essential for the accumulation of Zn in leaves. The model explained 66% of the total treatment variation (TTV) in the accumulation of Zn in leaves of *M. zeyheri.*

4.3.2 Leaf phosphorus

During fruiting, soil Mg, P, and pH positively contributed to the accumulation of leaf P in *M. zeyheri* leaves (Table 4.2), whereas after harvest only soil Fe and soil K had positive effects in the accumulation of leaf K. However, in either case, the TTV due to the segregated soil elements was low.

Table 4.1 Leaf zinc (ppm) content of *Mimusops zeyheri* determined using concentrations of selected soil nutrient elements, soil electrical conductivity and soil pH.

Time	Variable	Coefficient	Std error	t-value	P ≤	R^2
During fruiting	Constant	85.6039	16.0269	5.34	0.01	0.45
	Cu	– 67.7771	30.7931	– 2.20	0.05	
	Fe	– 0.4012	0.14731	– 2.72	0.05	
	K	– 0.0534	0.02585	– 2.06	0.05	
	P	– 1.0185	0.26641	– 3.82	0.01	
After fruiting	Constant	16.2956	4.9746	3.28	0.01	0.66
	Cu	67.4906	11.9185	5.66	0.01	
	K	– 0.0446	0.0104	– 4.27	0.01	

During fruiting: Leaf Zn = 85.6039 – 67.777 Cu – 0.401 Fe – 0.053 K –1.018 P, R^2= 0.45
After fruiting: Leaf Zn = 16.2956 + 67.4906 Cu – 0.044 K, R^2 = 0.66

4.3.3 Leaf manganese

During fruiting, Ca, K, Mn and Na contributed positively to the accumulation of leaf Mn, whereas Cu, Fe and Mg had a negative impact (Table 4.3). The four elements contributed 75 % to the TTV in Mn content. However, after fruiting both Cu and Fe still had a negative impact on the accumulation of Mn by *M. zeyheri* leaves.

Table 4.2 Leaf phosphorus (%) content of *M. zeyheri* determined using concentrations of selected soil nutrient elements, soil electrical conductivity and soil pH.

Time	Variable	Coefficient	Std error	t-value	P ≤	R^2
During fruiting	Constant	– 0.0696	0.0658	– 1.06	0.10	0.3
	Mg	0.0810	0.0344	2.36	0.05	
	P	0.0014	0.0472	2.85	0.01	
	PH	0.0181	0.00854	2.12	0.05	
After fruiting	Constant	0.0979	0.0118	8.27	0.01	0.2
	Fe	0.0872	0.0254	2.31	0.05	
	K	0.0071	0.0003	– 2.16	0.05	

During fruiting: leaf P = – 0.070 + 0.081 Mg + 0.001 P + 0.181 pH, R^2 = 0.30
After fruiting: Leaf P = 0.098 + 0.087 Fe – 0.007 K, R^2 = 0.22

4.3.4 Leaf magnesium

During fruiting K and Mn contributed 62% to the TTV in Mg content of *M. zeyheri* leaves (Table 4.4). Magnesium and Fe had a negative impact on the accumulation of leaf Mg.

Table 4.3 Leaf manganese (ppm) content of *M. zeyheri* determined using concentrations of selected soil nutrient elements, soil electrical conductivity and soil pH.

Time	Variable	Coefficient	Std error	t-value	P ≤	R^2
During fruiting	Constant	308.382	73.6604	4.19	0.01	0.75
	Ca	0.0281	0.0096	2.93	0.01	
	Cu	– 749.829	214.167	– 3.50	0.01	
	Fe	– 2.398	0.7711	– 3.11	0.01	
	K	0.3372	0.1311	2.57	0.01	
	Mg	– 0.4600	0.1174	– 3.92	0.01	
	Mn	16.3123	3.7629	4.34	0.01	
	Na	13.8386	4.0522	3.42	0.01	
	EC	– 1529.56	354.517	– 4.31	0.01	
After fruiting	Constant	649.146	55.6008	11.68	0.01	0.77
	Cu	– 1032.48	122.746	– 8.41	0.01	
	Fe	– 4.6632	0.8240	– 5.66	0.01	
	Na	– 4.5274	1.2689	– 3.58	0.01	

During fruiting: Leaf Mn = 308.382 + 0.0281 Ca + 13.839 Na, $R^2 = 0.75$
After fruiting: Leaf Mn = 649.146 – 1032.48 Cu – 4.663 Fe – 4.527 Na, $R^2 = 0.77$

4.3.5 Leaf potassium

During fruiting, Mg and Na contributed 49 % to the TTV of K accumulation in leaves of *M. zeyheri* (Table 4.5). After fruiting, Cu, Fe and P contributed 62 % to the TTV of K accumulation in the leaves of *M. zeyheri*.

Table 4.4 Leaf magnesium (%) content of *M. zeyheri* determined using concentrations of selected soil nutrient element, soil electrical conductivity and soil pH.

Time	Variable	Coefficient	Std error	t-value	P ≤	R^2
During fruiting	Constant	0.4801	0.0551	8.71	0.01	0.62
	Fe	– 0.0020	0.0065	– 3.06	0.01	
	K	0.0024	0.0011	2.12	0.05	
	Mg	– 0.0018	0.0007	– 2.43	0.05	
	Mn	0.0087	0.0029	2.79	0.01	
	EC	– 0.9028	0.2722	– 3.32	0.01	
After fruiting	Constant	0.3929	0.0354	11.11	0.01	0.56
	Fe	– 0.06588	0.1135	– 5.80	0.01	
	P	0.7120	0.1997	3.56	0.01	

During fruiting: Leaf Mg = 0.480 – 0.002 Fe + 0.002 K – 0.002 Mg + – 0.009 Mn 0.903 EC, R^2 =0.62
After fruiting: Leaf Mg = 0.39 – 0.07 Fe + 0.71 P, R^2 = 0.56

4.3.6 Leaf copper

During fruiting, the accumulation of Cu in leaves was restricted by Mg and Na, both with negative coefficients (Table 4.6). After fruiting P had a positive impact on the accumulation of Cu in the leaves of *M. zeyheri*, whereas Mg had a negative impact. The model contributed 27% to the TTV accumulation of Cu.

Table 4.5 Leaf potassium (%) content of *M. zeyheri* determined using concentrations of selected soil nutrient elements, soil electrical conductivity and soil pH.

Time	Variable	Coefficient	Std error	t-value	P ≤	R^2
During fruiting	Constant	– 0.0505	0.1649	– 0.31	1.00	0.49
	Mg	0.0007	0.0002	2.99	0.01	
	Na	0.0204	0.0088	2.31	0.05	
After fruiting	Constant	– 0.0260	0.1440	– 0.18	0.00	0.62
	Cu	1.2305	0.4489	2.74	0.01	
	Fe	0.0142	0.0029	4.82	0.01	
	P	0.0169	0.0050	3.36	0.01	

During fruiting: leaf K= – 0.051+ 0.001 Mg + 0.020 Na, R^2 = 0.49
After fruiting: Leaf K= – 0.026 + 1.231 Cu + 0.014 Fe + 0.017 P, R^2 = 0.62

4.3.7 Leaf calcium

During fruiting, all factors measured were optimal for the accumulation of Cu in leaves of *M. zeyheri*, thus they were eliminated by stepwise regression (data not shown). After fruiting, Ca, Cu, and Fe were super optimal, thus, they had a negative impact on the accumulation of Ca in the leaves of *M. zeyheri*. The model contributed 62% to the TTV of Ca content in leaves.

Table 4.6 Leaf copper (ppm) content of *M. zeyheri* determined using concentrations of selected soil nutrient elements, soil electrical conductivity and soil pH.

Time	Variable	Coefficient	Std error	t-value	P ≤	R^2
During fruiting	Constant	5.1363	0.4962	10.35	0.01	0.45
	Mg	– 0.0015	0.0007	– 2.24	0.05	
	Na	– 0.0707	0.0266	– 2.66	0.05	
After fruiting	Constant	4.1810	0.6989	5.98	0.01	0.27
	P	0.0495	0.0159	3.12	0.01	
	Mg	– 0.0032	0.0014	– 2.36	0.05	

During fruiting: leaf Cu = 5.136 – 0.002 Mg – 0.071 Na, R^2 = 0.45
After fruiting: Leaf Cu = 4.181 + 0.050 P – 0.003 Mg, R^2 = 0.27

Table 4.7 Leaf calcium (%) content of *M. zeyheri* determined using concentrations of selected soil nutrient elements, soil electrical conductivity and soil pH.

Time	Variable	Coefficient	Std error	t-value	P ≤	R^2
After fruiting	Constant	1.70086	0.11571	14.7	0.01	0.62
	Ca	– 0.00006	0.00002	– 2.68	0.01	
	Cu	– 1.47259	0.29510	– 4.99	0.01	
	Fe	– 0.00554	0.00205	–2.70	0.01	

After fruiting: Leaf Ca = 1.7009 – 0.001 Ca – 1.473 – 0.006 Cu, Fe, R^2 = 0.62

4.4 Discussion

According to the law of the minimum, "The growth of a plant is dependant upon the amount of foodstuff that is presented to it in minimum quantities" (Salisbury and Ross, 1978). Applied to mineral nutrition, the ascending part of the saturation curves represents those elements that are in relatively small amounts, whereas the descending parts are those that are in toxic amounts (Salisbury and Ross, 1978). The uniqueness of stepwise regression, eliminates those factors that the organism is saturated with (i.e. no longer has capacity to use them).

In this study, it was demonstrated that various soil factors play a role in the status of mineral nutrition of *M. zeyheri* during fruiting and after fruiting. The study of interactions of mineral in plants is complicated (Bonn *et al*., 1985). In this study, for one mineral element, a particular element may be limiting for the accumulation of a given element in leaves during fruiting, whereas after fruiting the same element becomes either limiting, optimal or toxic (inhibitory) to the accumulation of the element. For instance, for the accumulation of Cu in the leaves of *M. zeyheri*, Mg was inhibitory during fruiting and after fruiting, whereas it had limited influence on the accumulation of K during fruiting.

The challenge in crop production is to discover the limiting factors and to rectify the situation through fertilization. In another study (Ndhukula, 2006), it was demonstrated that *M. zeyheri* seedlings do not respond to fertilization under greenhouse conditions, which further complicates the fertilization studies of *M. zeyheri.*

The current study gives baseline information, which may be important in various environmental physiology studies of this plant. Physiological studies will be necessary to assess the importance of “limiting” mineral nutrients in the accumulation of certain mineral nutrients in *M. zeyheri* in relation to its productivity.

CHAPTER 5
SUMMARY AND CONCLUSIONS

Mimusops zeyheri managed to perform well in three regions with different climatic conditions and soil types. This shows that the plant can be produced in areas with more or less the same climatic conditions with the sampled areas, as long as the nutrient status of the plant can be taken into consideration. As an indigenous fruit tree, *M. zeyheri* has attributes to serve as a vitamin C source in rural areas of Limpopo Province.

In all the three regions soil pH was not affected by time of sampling (during and after fruiting), meaning that the abscission of flowers and fruitlets did not have an effect on soil pH. At high pH levels, P, Cu, Zn and Mn became less available and at low pH levels P and Mg become unavailable and Mn concentrate was at toxic levels.

Electrical conductivity (EC) also plays a significant role in nutrient availability. The high EC after fruiting suggested that the abscised materials, which occur mostly during fruiting (flowers and fruitlets) may have released compounds after decomposition, thus influencing its (EC) value.

The challenge in crop production is to discover the limiting factors so that we can be able to rectify them through fertilization. The results from this experiment can serve as a guideline on the nutrients requirement of *M. zeyheri.* Further studies can be conducted on nutrient uptake of the plant in a long term experiment to have definite findings about it.

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LIST OF APPENDICES

Appendix A. Soil forms from the three regions of Limpopo Province.

DESCRIPTION	REGION		
	CHUENESPOORT	BOCHUM	SEKGOSESE
Horizon A	Orthic A	Orthic A	Orthic A
Horizon B	Luthocutanic B	Red appedal B	Yellow brown appedal B
Soil form	Glenrosa	Hutton	Clovelly

Soil pH in horizon A and B within the region did not differ. Also, Chuenespoort and Sekgosese did not differ, where values in Bochum were slightly higher (Appendix B). Electrical conductivity values in all regions averaged 0.03 for A horizon and 0.05 for horizon B.

Appendix B. Soil pH and electrical conductivity of the different horizons in three regions of Limpopo Province.

Region	Horizon	pH-H_20	pH-KCl	EC (dS/m)
Chuenespoort	A	6.82	6.50	0.03
	B	6.84	5.68	0.01
Bochum	A	7.23	6.89	0.03
	B	7.50	6.99	0.06
Sekgosese	A	6.78	5.05	0.05
	B	6.80	4.22	0.08

In all three sampling sites, Horizon A comprised Orthic A. However, Horizon B's differed, and comprised Luthocutanic B, Red Apeddal B and Yellow brown appedal B in

Chuenespoort, Bochum and Sekgosese respectively (Appendix A). Soil forms in the three regions consisted of Glenrosa, Hutton and Clovelly, respectively.