ASSESSING SAVANNA SHRUB ROOTS DEPLOYMENT USING RADIOGENIC STRONTIUM ISOTOPES IN THE NORTH-EASTERN KALAHARI, NAMIBIA

A THESIS SUBMITTED IN FULFILMENT OF THE

REQUIREMENTS FOR THE DEGREE OF

MASTER OF ARTS (BY THESIS)

IN GEOGRAPHY

OF

THE UNIVERSITY OF NAMIBIA

BY

HELALIA N. IIPINGE

201410945

OCTOBER 2024

MAIN SUPERVISOR: Dr. J. NAKANYALA (UNIVERISTY OF NAMIBIA)

CO-SUPERVISOR: Prof. M. HIPONDOKA (UNIVERSITY OF NAMIBIA)

Abstract

The belowground interaction between trees and grasses in the savanna biome and mechanisms of moisture and nutrient uptake are poorly understood due to methodological challenges. To fill this gap, this study tested the robustness of the radiogenic strontium isotope method to study plant roots. The study was carried out at a landscape level, in Northeastern, Namibia. Plant leaves of four randomly selected shrubs and their 16 nearest neighborhood shrubs were collected within a 20 m x 20 m plot for isotopic analysis. This was done to assess the method's performance. The study characterised the belowground structure and roots deployment of 17 of the 20 shrubs, sampled for isotopic analysis. It also assessed soil isotope, soil physicochemical properties, and the distribution of root biomass and density. Soil isotopes and soil physicochemical properties were analysed in the laboratory from 50 samples taken from five soil cores (10 cm interval, down to 1 m depth). A total of 450 soil samples were taken from 45 soil cores, spaced 2.5 m in 5 transects to determine root density and biomass. Data analysis was done in R.4.2.1. Results showed a relatively poor soil nutrient at the study site, but a high concentration of nutrients was recorded in the first 50 cm soil profile, where 75% of the root biomass is invested. Comparable strontium ratios were recorded along the soil profile, which did not vary significantly with soil depth ($p = 0.44$). Results indicated that plants' isotopic ratios are neither attributed to rooting depths, nor to plant species. This study concluded that the isotope method lacks precision to establish shrubs' rooting depths for ecological studies. However, an overlap between sampled plants and soils' Sr ratios was recorded at the site, which makes the results precise for studies with interest in geographical variations of isotopes such as archaeology.

Keywords: Belowground biomass; Soil depth; Kruskal-Wallis test; Soil physicochemical properties; Plant canopy; Sr ratios; z-score

TABLE OF CONTENTS

List of Tables

List of Figures

List of Abbreviations and Acronyms

AGB	Above Ground Biomass
ASU	Arizona State University
BGB	Below Ground Biomass
Ca	Calcium
DRC	Democratic Republic of Congo
IDW	Inverse Distance Weighted
MC-ICPMS	Multicollector -Inductively Coupled Plasma Mass Spectrometer
ITCZ	Intertropical Convergence Zone
H_2O	Water
GPR	Ground Penetrating Radar
KB	Kalahari Basin
MAWLR	Ministry of Agriculture, Water and Land Reform
N	Nitrogen
Na	Sodium
NPP	Net Primary Productivity
OM	Organic matter
P	Phosphorus
ppm	parts per million

pH	Potential Hydrogen
Mg	Magnesium
K	Potassium
RSA	Root System Architecture
SD	Standard Deviation
SE	Standard Error
Sr	Strontium
USNSF	United States National Science Foundation

Acknowledgments

I would like to give special thanks to the Almighty God for his grace, strength, and protection He has given me throughout the completion of my study. The completion of this study could not have been possible without the support and advice offered by various institutions and people. Firstly, I would like to thank my amazing supervisors Prof Martin Hipondoka and Dr Jesaya Nakanyala for their immeasurable support, good advice, and constructive criticisms during my study. I would like to thank the United States National Science Foundation (USNSF) for funding my research project through the School of Human Evolution and Social Change at Arizona State University (ASU).

I would like to thank the Ministry of Agriculture, Water and Land Reform (MAWLR) for their laboratory analysis service rendered to me by analysing the soil's chemical properties. I would like to extend my appreciation to Mrs. Alma Frans at the University of Namibia (Ogongo Campus) for her assistance in the laboratory and arrangements of laboratory equipment. I would also like to thank the Multicollector -Inductively Coupled Plasma Mass Spectrometer (MC-ICPMS) laboratory of South Africa for carrying out the isotopic analysis on both plants and soil samples. I also thank Ms Anneli Nghikembua for proofreading and editing this thesis.

I am eternally grateful to the field assistants (Gcao Kashe and Coma Tshao), for spending their time with me in the field and to help with the excavation of shrubs despite the tedious, time-consuming, and laborious tasks involved in the process. They also assisted with identifying the local names of different shrubs. I would like to give thanks to my family members especially my husband Mr. Petrus Iipinge, who has not only been my pillar of strength but also patient enough when I spent most of my time

on this research project. Their support and words of encouragement relieved the stress during my postgraduate journey.

Dedication

I would like to dedicate this thesis to my grandmother, Gongaleni Jafet who has been encouraging me to focus on my education since my childhood until the completion of this study. Her prayers and blessings kept me going through my postgraduate journey. I would also like to dedicate this thesis to my mother Foibe Tangeni Shololo, for her understanding, encouragement, and emotional support during my stressful times. Thank you for believing in my capabilities.

Declarations

I, Helalia N Iipinge, hereby declare that this study is my work and is a true reflection of my research and that this work, or any part thereof has not been submitted for a degree at any other institution.

Name of Student: Helalia N Iipinge Signature:  Date: October 2024

CHAPTER 1: INTRODUCTION

1.1 Background of the study

The savanna is among the world's largest terrestrial biomes (Scholes & Archer, 1997). Widespread on the African continent, the savanna provides ecosystem services and sustains livelihoods for millions of people (Devine et al., 2017; Scholes & Archer, 1997). This unique biome is characterised by the occurrence of trees and shrubs interacting with grasses and herbs to create a biome that is neither grassland nor forest (Scholes & Archer, 1997). Those contrasting lifeforms arguably live in harmony, yet forage for the limited resources. The structure of the savanna is influenced by several factors such as rainfall and soil as well as fire and grazing, resulting in a tropical grassland with scattered trees and bushes (Werner, 2021). Despite the role of the savanna as a source of livelihood for pastoralists, our knowledge of its unique coexistence and the interactions between trees and grasses is still fragmented (Scholes & Archer, 1997).

The coexistence and the interaction between trees and grasses are gradually changing in many semi-arid savanna rangelands due to an ecological phenomenon called bush encroachment. Bush encroachment is defined as an undesired suppression of palatable grasses and herbs by encroaching shrubs, often unpalatable to livestock, resulting in an impenetrable thicket (Ward et al., 2013). Bush encroachment is considered a land degradation that affects the environment negatively, resulting in the loss of biodiversity and land productivity (Birch et al., 2017).

Several studies have investigated the savanna's unique coexistence and the occurrence of bush encroachment, of which earlier efforts focused on understanding the

mechanism under which tree-grass partition their roots for water and nutrient uptake (Walker et al., 1981; Walter & Mueller-Dombois, 1971). Subsequent studies provided various contrasting and diverging perspectives. However, the mechanisms underpinning the savanna's unique coexistence remain elusive to this day (Sankaran et al., 2004; Ward et al., 2013; Whitecross et al., 2017). Existing perspectives were largely influenced by the equilibrium and non-equilibrium ecological paradigms (Jeltsch et al., 2000; Sankaran et al., 2004; Scholes & Archer, 1997). The equilibrium paradigm considers the savanna as a stable biome, of which its unique coexistence is owed to the root niche partitioning between these two contrasting life forms, a natural regulating mechanism (Jeltsch, et al., 2000). This idea emerged following the work of Walter and Mueller-Dombois (1971) in the Kalahari of the then South West Africa, now Namibia, culminating in Walter's two-layer hypothesis. Walter's two-layer hypothesis argues that savanna is a result of vertical niche partitioning between trees and grasses whereby trees extend their roots into deeper soil layers to extract water from deep underground. Whereas, grasses with their shallow roots only forage for water on the topsoil layer (de Klerk, 2004; Ward et al., 2013).

However, this hypothesis has been disputed by several studies such as those of Nakanyala (2020), Ward et al. (2013) and Hipondoka et al. (2003). Nakanyala (2020) for example, argued that not all woody plants have tap roots to forage for water from deeper horizons. Additionally, some plants only have lateral roots limited to shallow soil depths and forage for water from the same subsurface layers as the grasses thus, leading to a direct niche competition between trees and grasses. Hipondoka et al. (2003) further stated that both trees and grasses deploy most of their root in the upper

soil layer, thus trees do not have a clear dominance over grasses at deeper soil layers during dry climates.

The non-equilibrium paradigm in contrast argues that savanna biomes are unstable and continuously change in space and time in response to various changes related to climate variability and anthropogenic drivers (Jeltsch et al., 2000). Thus, better explained by the state and transition model (Westoby et al., 1989). Transitions refer to the trajectories of change of ecosystems that are subjected either to natural events (e.g. drought or rainfall) or by the management (e.g. burning, destruction, or introduction of plant populations, stocking rates, and grazing) or by both natural and management activities (Westoby et al., 1989). The natural and management events often occur in the same ecosystem, i.e., savanna, however, the system does come to rest through a transition process (Westoby et al., 1989). Consequently, these competing views mean that the savanna's determinants are still inconclusive. This problem is further compounded by the fact that many savannas are significantly modified by anthropogenic activities, making it difficult to identify or quantify the key determinants of savanna structure in their natural settings (Scholes & Archer, 1997).

This lack of knowledge is particularly acute regarding their belowground biomass (BGB) and structure. Belowground biomass is an important component of terrestrial biomes' net primary production (NPP) and contributes significantly to carbon accumulation in forest ecosystems (Bijak et al., 2013). The belowground biomass is defined as the entire biomass of all live roots (Ravindranath & Ostwald, 2008). Unlike aboveground biomass (AGB), research on BGB is limited owing to the laborious efforts required for exposing the live roots.

This challenge has left a gap in knowledge on this essential component of many terrestrial biomes despite the significant role plant roots play in terms of acquiring soil moisture and soil nutrients (Laughlin et al., 2021). The plant roots determine the spatial distribution of water and nutrient uptake and could cause an increase or a decrease in resource availability (Smit & Rethman, 1998). Belowground structure could be quantified from two types of roots. Root segments larger than 2 mm in diameter are classified as coarse roots, whereas fine roots are those with less than 2 mm in diameter (Leuschner et al., 2007). Fine and coarse roots are regarded as the key contributors to the belowground net primary productivity and play critical roles in the biochemical cycling of forest and woodland ecosystems (Addo-Danso et al., 2015). Fine roots are homogenously distributed where water and nutrient distributions are not spatially patchy whereas coarse roots are generally more abundant close to stems (Macinnis-Ng et al., 2009). Access to the roots to take accurate and reliable empirical measurements is nevertheless a challenge.

Several direct methods such as excavation, soil coring, trenches, monolith, and ingrowth bags have been used to study plant roots (Chen et al., 2018; Hipondoka et al., 2003; Nakanyala, 2020; Rau et al., 2009; Smit & Rethman, 1997). However, these methods are considered destructive, labour-intensive, and time-consuming (Lui et al., 2016). Other indirect methods such as ground penetrating radar (GPR), minirhizotron, rhizotrons, scanners, and tracers have also been used to study plant roots dynamics (Kulmatiski et al., 2017; Kulmatiski & Beard, 2020; Raz-Yaseef et al., 2013; Taylor et al., 1990). This is because they are considered non-destructive and non-invasive. However, limitations are also evident with such methods. For example, the

minirhizotron technique is not suitable in environments with very low or very high temperatures because of the sensitivity of these devices to extreme temperatures (Gluszek, et al., 2013). Meanwhile, GPR is unable to detect fine roots owing to its coarse resolution (Maeght et al., 2013). While rhizotrons are expensive to construct and operate, they are at risk of disturbance from underground insects such as termites (Freschet et al., 2020; Neumann et al., 2009). On the other hand, tracers such as deuterium oxide could be affected by the hydrological redistribution process which results in water moving upwards or downwards along the soil profile (McCulley et al., 2004). Lastly, scanners have root length limits, thus being unable to determine full root systems (Zappala et al., 2013).

The challenges presented above call for alternative approaches. The radiogenic strontium (Sr) isotopes technique has the potential to understand the dynamics of water and nutrient uptake by savanna plants. This technique has been applied in archaeology as a tracer for geographic origins, migrations, trade, and exchange, as well as cultural change (Adams et al., 2019; Bentley et al., 2002; Frank et al., 2021; Slovak et al., 2018). Strontium could be picked up by plants through nutrient uptake and become part of the plant shoot (Gupta et al., 2018). McCulley et al. (2004) suggested that the radiogenic strontium isotope technique might be used as a proxy to determine the depth of water uptake by investigating the isotopic ratio of plant shoot about the isotopic ratio of the soil root zone. The purpose of this study was therefore to employ and gauge the ability of the radiogenic strontium isotope as a potential method for revealing savanna shrubs' root deployments in relation to nutrients and moisture uptake from distinct root zone depths. In order to calibrate the strontium isotope approach, the study assessed the distribution of root biomass and root density about soil depth and soil

nutrients in the Namibian savanna. The study area is inhabited by indigenous hunter-gatherer and has limited farming activities and human footprint, and thus a suitable area for this study. The area is part of the Kalahari from which Walter and Mueller-Dombois (1971) carried out their study that proposed Walter's two-layer hypothesis, a now debated model by numerous studies.

1.2 Statement of the problem

The belowground structure of terrestrial plants is an important component, yet it is a poorly explored zone. This is particularly so in the savanna where plants' belowground structure is claimed to be the main determinant of this biome. The major challenge to address this paradox is rooted in the methodological difficulties associated with exposing plant roots over a large area and achieving a large and representative sample size for empirical measurement. Both direct and indirect methods were put to the test in resolving this problem. However, limitations still exist. Limitations range from the destructiveness of the methods, labour intensive, time-consuming, and tedious processes associated with excavation, soil coring, monolith, and trenching methods, while indirect methods such as minirhizotrons, rhizotrons and GPR among other limitations, have difficulties in installations and setting up depending on the nature of the soil and unable to detect all the root types, i.e., fine roots (Maeght et al., 2013).

A lack of robust methods to study plants' belowground structure means that our current understanding of the mechanism of nutrients and moisture uptake by plant roots is flawed. These methodological challenges from both direct and indirect methods, associated with studying savanna plant roots could potentially be addressed by applying novel approaches to overcome existing challenges. To fill this gap, this study

tested the ability of the strontium isotopes method as a tracer for root depth and moisture uptake by analysing the isotopic ratio of plant leaves and the isotopic ratios of soil depths where plant roots are deployed. This technique has been used as a tracer in similar studies such as Coble et al. (2015) and Pozwa et al. (2009) to determine the source of nutrients in different environments. The use of strontium isotope is important because it has the potential to reveal depth uptake when plant leaves show Sr ratios to soil Sr ratios of their corresponding rooting depths. This helps to better understand the depths where trees and grasses acquire nutrients and moisture from the soil.

1.3 Aims and objectives of the study

This study aimed to assess the effectiveness of radiogenic strontium isotopes as a proxy for resolving distinct depths at which savanna shrubs obtain soil nutrients and moisture about their root structure. The objectives of this study were thus:

a) To characterise the belowground structure of the studied plants and assess their root deployments in the soil profile from 0 to 100 cm depth, where soil resource uptake is most likely to take place.
b) To assess the vertical distribution of soil physicochemical properties at the study area at the depth of 0 to 100 cm.
c) To determine radiogenic strontium isotope ratios of savanna shrub leaves at a landscape level in northeastern Namibia.
d) To determine radiogenic strontium isotope ratios of the soil profile from the under-canopy and outside canopy of the studied savanna shrubs.
e) To compare the isotopic ratios obtained from the soil profile with the shrub leaves isotopic ratios and assess the overall performance of the radiogenic strontium isotopes method in studying plant root deployment.

1.4 Significance of the study

The significance of this study rests in knowledge acquired on the belowground structure of savanna plants and their competitive interactions which was done by assessing the root deployment of savanna shrubs using the strontium isotopes approach. Knowledge of plant roots is important in the context of bush encroachment, a phenomenon whose occurrence in the savanna is still not well understood in terms of the mechanisms that regulate the coexistence between trees and grasses, owing to a lack of robust methods to study plant roots. In addition, this study supported a parallel study in the study area, focusing on archaeology by assessing the radiogenic isotopes of plant shoots at a district level, but only sampling a single plant at each site. Results from this study, therefore, help to understand the variation of plants' and soil's Sr ratios in proximity.

1.6 Delimitation of the study

The study was carried out in the Kalahari Basin, Tsumkwe district to complement a parallel study in the area. The study only sampled shrubs and not grasses since grasses in the Kalahari are already known to deploy their roots in the soil surface layers within 30 cm depth and overlap with shrub roots after 60 cm depth. (Hipondoka et al., 2003). The study was designed to only sample up to 1 m soil depth because it was reported that more roots are more abundant in the upper soil layer (Hipondoka et al., 2003; Hipondoka & Versfeld, 2006). Therefore, soil resource uptake is most likely to take place at this depth zone. Lastly, sampling into deeper depths would result in lots of soil samples which could cause excessive costs, particularly for laboratory analysis.

1.7 Limitations of the study

The study could not determine the rooting depths of all the sampled shrubs, particularly those of three *Grewia flava,* due to their complex fibrous root system. Additionally, the study could not expose the whole root structure of plants with roots beyond 1 m depth because the study was designed to only sample up to 1 m depth. The study was limited to a smaller size due to budget constraints.

1.8 Thesis outline

This thesis is arranged in six different chapters. The thesis first outlined the abstract which provides a summary of all chapters covered.

Chapter 1 offers a general idea of the savanna biome, emphasising on the importance of understanding the belowground structure of the savanna plants. It focuses on the problem associated with plant roots knowledge, mainly the methodological challenges. Different methods that have been used to study plant roots are listed in this chapter. The chapter also proposed a new method as a possible solution to the problem stated. Additionally, the main aim and research objectives are covered in this chapter.

Chapter 2 analyses the literature on the understanding of the savanna structure, focusing on different theories and models on the determinants of the savanna structure. This chapter details various methods used by researchers to study plant roots, ranging from direct to indirect methods. Challenges associated with each method are described in this chapter. The chapter also gives a detailed background of the strontium isotope method as a better method for studying savanna plant roots.

Chapter 3 details the procedures and equipment used by the study to successfully obtain the desired results. These procedures include data collection, laboratory analysis, and data analysis. The geographic location, vegetation, climate, and soil type of the study area have been also described in this chapter.

Chapter 4 presents the detailed findings obtained from the study with the study's objectives. This chapter demonstrates how root biomass, root density, and soil physicochemical properties vary with soil depths. The level of root biomass and soil nutrients at the study site are also presented in this chapter. The variation of plant leaves, and soil depths' Sr ratios are presented. This chapter also shows how plants' Sr ratios correspond with soils' Sr ratios of their rooting depths. Graphical presentations, tables, and texts have been used in this section to help explain the results better.

Chapter 5 interprets the main results presented in Chapter 4. The interpretation is based on the variation of root biomass, root density, and soil physicochemical properties of the savanna. The interpretation also focuses on the comparison between plant leaves and soil depths isotopic ratios as well as the ability of the radiogenic strontium isotope approach to determine the shrub roots deployment. This chapter explains how the research objectives were met, and the limitations and knowledge gaps that arise from the findings.

Chapter 6 explains in a nutshell the findings and discussions of the study. This chapter concludes the dynamic of the savanna belowground structure, particularly the distribution of root biomass, root density, and soil nutrients as well as the rooting

patterns of the sampled savanna shrubs. This chapter has concluded on the potency of the radiogenic strontium isotope approach in the study of plant roots. Recommendations arising from the findings of the study are also presented in this chapter.

CHAPTER 2. LITERATURE REVIEW

2.1 The savanna's unique coexistence

The savanna, one of the world's largest terrestrial biomes (Scholes & Archer, 1997), is characterised by a coexistence of two competitors, woody plants including trees and shrubs, and herbaceous plants including herbs and grasses (Scholes & Archer, 1997). This biome is considered unique because of its coexistence of contrasting life forms, not found in any other terrestrial biome. This coexistence also defies the competitive exclusion principle which states that two species competing for the same limited resource cannot coexist, but one species would outcompete the other and become dominant (Kneitel, 2013).

The savanna biome is widespread across tropical and temperate regions of the world (Rutherford et al., 2006). In tropical regions, this biome covers approximately 1.6 million ha (Scholes & Archer, 1997), which is equivalent to an eighth of the global land surfaces. Savanna covers about half of Africa and Australia, about 45% of South America, about 10% of India, and 10% of Southeast Asia (Scholes & Archer, 1997) (Figure 1).

The wide distribution of savanna means that this biome occurs over a broad range of climatic conditions ranging from sparse grassland with scattered trees, and average annual precipitation of less than 100 mm per annum to tall moist woodland savanna, with annual precipitation of more than 1500 mm per annum (Jeltsch et al., 2000). Climate has a deterministic effect on vegetation types, where the distributions of biomes on Earth such as savannas are expected to gradually change with climatic change (Staal et al., 2016). According to Scholes and Archer (1997), the savanna has

been broadly subdivided based on stature, canopy, cover and the arrangements of woody elements. For example, “Savanna grassland” consists of scattered trees or shrubs, and “shrub savanna” contains a vast number of trees and shrubs.

Figure 1. The distribution of savanna worldwide is indicated in yellow (source: Weaver, 2014).

2.2. Determinants of the savanna

The real determinants of the savanna are still uncertain, although several abiotic and biotic factors have been proposed (Axelsson, 2018; Sankaran, 2004; Scholes & Archer, 1997; Walter & Mueller-Dombois, 1971). Abiotic factors that have been attributed to the origin of savannas are largely soil moisture and soil nutrients. For instance, low and infrequent rainfall in savannas results in insufficient variability in soil moisture during the growing season, which could interact with trees and grasses differently (Sankaran et al., 2004). Meanwhile, nutrient availability influences plants’ growth rate, resulting in niche partition between trees and grasses (Konare´ et al., 2019). Fire is

also considered a determinant of the savannas (Axelsson, 2018). On the other hand, biotic factors such as grazing, and herbivores are considered important determinants of the savannas. Some scholars such as Carlsson (2005) considered savanna as an unnatural biome whose existence owes its origin to various anthropogenic disturbances such as grazing and the role of mega browsers.

2.2.1 Fire

Fire plays an important role in the savanna (Koffi et al., 2022). Savanna is the most frequently burned ecosystem on earth (Axelsson, 2018). According to Sheunyange et al. (2004), anthropogenic fires in Africa are prehistoric forms of environmental disturbance that shape the savanna vegetation more than any human-induced disturbance. Fire strongly impacts vegetation structure in savanna by suppressing the maturation of saplings into adult trees, hence, affecting tree density and cover (Staver et al., 2017). According to Baudena et al. (2015), grass constitutes much of the fuel load, therefore, highly flammable and fire spreads faster in open ecosystems. At the same time, grasses benefit from fire as they recover faster than trees. This results in forest areas turning into savanna-like, whereas grasslands turn into shrublands or woodlands with little grass biomass (Scholes & Archer, 1997). Sheunyange et al. (2004) suggested that vegetation patterns in savanna change according to fire frequency. For example, shorter fire return intervals reduce shrub cover and shrub species, while promoting herbaceous species (Sheunyange et al., 2004). In some areas, elders practice traditional burning to renew pasture quality when wood and grass litter accumulate as fuel load at landscape levels, however, bush expansion is experienced at a regional scale (Sheunyange et al., 2004).

2.2.2 Grazing

The savanna contains a large and rapidly growing proportion of the world's human population and most of its rangelands and livestock (Scholes & Archer, 1997). Savanna is a source of livelihood for most pastoralists of and the livestock survive through grazing (Scholes & Archer, 1997). However, the diversity of herbaceous and woody species decreases under high livestock pressure (Katjiua & Ward, 2012). Scholes and Archer (1997) argue that heavy grazing in savannas changes the patterns of herbaceous and vegetation in savanna. Higher grass biomass could affect tree biomass by fuelling fires while grazing reduces the fuel load and, hence, fire frequency (Scholes & Archer, 1997). According to de Klerk (2004), an increase in grazing causes an increase in woody vegetation in such a way that too much grazing reduces grasses and consequently a reduction in fire intensity which then gives a chance to the growth of woody vegetation.

2.2.3 Browsing

Herbivores, especially elephants, have also been proposed by some scholars to have contributed to the dynamics of savanna (Fullman & Bunting, 2014; Jeltsch et al., 2000; van der Waal, 2010; Valeix et al., 2010). Valeix et al. (2010) described African elephants as "ecosystem engineers" because they regularly uproot, and break trees and shrubs as well as browse. These have an impact on woody vegetation causing a reduction in the number of trees and woody vegetation cover (Jeltsch et al., 2000). When elephants cause direct mortality of trees, it leads to the vulnerability of trees to fire, hence, promoting grass production where trees are removed and, thus, changing the vegetation structure and nutrient cycling in the savanna (Fullman & Bunting, 2014).

Large herbivores potentially influence the structure of savanna vegetation indirectly via control over soil nutrient availability, which changes the establishment success of tree seedlings (van der Waal, 2010). This happens when they consume substantial amounts of the above-ground biomass, while depositing large amounts of soil nutrients through dung and urine, thus, providing a shortcut to nutrients released from plant litter through decomposition, by rapidly processing vegetation and depositing nutrients in plant-available forms (van der Waal, 2010). Because herbivores have recently been moved into conservation areas, an act done to prevent wildlife poaching and population reduction means that there is a reduction on the effect of browsing in the savanna vegetation structure.

2.2.4 Soil moisture and soil nutrients

Water is a limiting factor in semi-arid savannas, on the tree-grass coexistence (Walter & Mueller-Dombois, 1971). Although trees and grasses are two different life forms with different shoot and root structures, they both depend on and compete for the same limited water resource within the savanna system (Whitecross et al., 2017). Soil moisture is important for plant growth because it is a reactant in photosynthesis, acts as a solvent for plant nutrients, and is a medium for the moderation of temperature in plants and soils (Ampofo, 2006). The main source of water in savanna is rainfall (Beudena et al., 2015). However, changes in rainfall patterns due to global climate change have affected the vertical distribution of soil moisture in the soil profiles and consequently affected the performance and relative abundance of shallow and deep-rooted plant species (Holdo & Nippert, 2015). Kgabi et al. (2016) further argued that

changes in average rainfall influence groundwater recharge rates, eventually affecting water supply.

Spatial competition for soil moisture is a basic component of the vegetation dynamics in savannas which leads to a stable coexistence between trees and grasses (Rodrígues-Iturbe & D'Odorico, 1999). In addition, water stress has multiple and global effects on tree-grass interaction as it influences the rate of photosynthesis, stomatal closure, and plant transpiration rate, which constitute a major constraint on primary production (Rodrígues-Iturbe & D'Odorico, 1999). Furthermore, tree abundance strongly influences savanna ecosystem dynamics, whereby maximum tree abundance in tropical savanna is found to be negatively correlated with rainfall intensity (Xu et al., 2018). This differentiation in the water use strategy creates a temporal niche-separation in water use between trees and grasses.

Savanna landscapes are heterogeneous systems and resources such as soil nutrients vary widely in space and time (van der Waal, 2010). The types of nutrients available influence how they are being portioned between co-existing trees and grasses (van der Waal, 2010). Moreover, nutritional necessities change depending on plant species as well as plant growth stage (Alaoui et al., 2022). According to Viani et al. (2011), soil nutrients availability is the key factor determining the distribution of species and composition of many tropical forests and woodlands. Plants vary in their absolute and relative requirements of nitrogen (N) and phosphorus (P), and changes in the relative availability of these nutrients could change the vegetation structure and composition, potentially affecting higher tropic levels (van der Waal, 2010). For example, trees are relatively more limited by N than P whereas grasses are more P than N limited (van de

Waal, 2010). Therefore, the availability of both N and P ensures a resource partition between trees and grasses. The variation in soil resource availability plays a fundamental role in structuring plant communities (Hold & Mack, 2014). For instance, Konare´ et al. (2019) suggested that the partitioning of two minerals of nitrogen namely: ammonium (NH_4) and nitrate (NO_3), allows the coexistence of trees and grasses in which they have contrasting preferences regardless of the nitrification rate. For example, when the nitrification rate is high, grasses prefer NH_4 and trees prefer NO_3, whereas at a low nitrification rate, grasses prefer NO_3 while trees prefer HN_4, hence, a root niche partitioning between tree-grass (Konare´ et al., 2019). Lastly, Holdo and Mack (2014) argued that changes in nutrient availability patterns lead to changes in forage quality, which influence large herbivory use and vegetation impact patterns, thus, indirectly influencing vegetation structure.

2.3. Different theories on the nature, structure, and dynamics of the savanna biome

Broadly, the explanation of the coexistence of tree-grass falls into two categories: a) competitive based models and b) demographic based models. A competitive based model argues that the coexistence of tree-grass is made possible because of a niche difference whereby trees and grasses avoid competition by using resources that are obtained at different places or obtained at different times (Scholes & Archer 1997). These models are considered to lead to a stable coexistence and, therefore, known as equilibrium (Scholes & Archers, 1997). In competitive based models, water and nutrients are considered to be the main determinant while fire and grazing are “modifiers”.

A popular competitive based model is a root niche separation model also known as Walters' two-layer model. This model states that grasses with their shallow roots are superior competitors for water in the upper soil layer, whereas trees with deep tap roots have exclusive access to water in the subsoil layer (Walter & Mueller-Dombois, 1971). Another competitive based model is a phenological niche separation model. According to this model, savanna trees are able to store water and nutrients and achieve a full leaf expansion on the onset of rain while peak leaf area for grasses is achieved at a later stage (Sankaran, 2004). Trees, therefore, have an exclusive access to resources early and late in the growing season and for grasses to prevail, they would have to be superior competitors for resources during periods of overlap with trees (Scholes & Archer, 1997).

In the past, the competitive model has been interpreted in the context of equilibrium paradigm. Equilibrium predicts a stable ecosystem determined by environmental factors such as rainfall and soil type (Gillson, 2004). This theory argues that terrestrial ecosystems are stable at an end of growth maturity level called climax, which represents a permanent stage of vegetation succession, whereby the ratio between trees and grasses is regulated by natural mechanism such as niche partitioning (Nakanyala, et al., 2017; Vetter, 2005).

In contrast, demographic based models state that the persistence of tree-grass in the savanna is a result of climatic variability and anthropogenic disturbances such as fire and grazing. According to these models, there are no 'primary determinants' of savanna and disturbances such as fire and grazing are not just 'modifiers' but they are also 'maintainers' of the savanna state (Sankaran et al., 2004). For example, in arid

regions, trees are assumed to be limited by drought at the seedling stage and by fire at the sapling stage. In mesic regions, trees are capable of dominating the system, but frequent high fire intensity limits tree seedlings from growing into adulthood (Sankaran, 2004). Additionally, Jeltsch et al. (2000) proposed the concept of 'ecological buffering mechanisms' which refers to buffering mechanisms such as fire, browsers, and microsites, that prevent the savanna system from crossing the boundaries to other types of systems which are either pure grassland or pure tropical forest. Fire, for example, is known to prevent the transition of savanna to woodland and microsites prevent the transition of savanna to grassland (Jeltch et al., 2000).

The demographic models have been predicted in the context of non-equilibrium. Non-equilibrium paradigm focuses on the processes that generate spatial and temporal heterogeneity, including the interactions between organism disturbances and environmental stochasticity. In this theory, savanna changes rather than stasis and disturbances by fire and herbivores are considered to be parts of the savanna function (Gillson, 2004).

However, limitations are also evident on these models. For example, the root niche separation theory under competitive model ignores the period of root overlap between older tree seedlings and grasses, thus the model is largely limited to adult trees. Nonetheless some studies also indicated and overlap of adult trees and grasses in the soil profile (Hipondoka et al., 2003). Additionally, this theory only applies to arid and semi-arid savannas where rainfall is limited, hence seasonal drought, unlike in mesic savannas where soil moisture is sufficient.

Demographic models on the other hand, although they consider disturbances based on the life history of tree-grass, the resource competition between trees and grasses is not explicitly incorporated (Jeltch et al., 2002). According to Sankaran et al. (2004) models where competition is not evoked, the coexistence results either from a stress tolerance colonization trade-off or is achieved broadly by default.

Although different theories and models have been proposed on the existence of the savanna, the origin, nature, and dynamics of savanna is still not clearly understood. Jeltch et al. (2000) stated that tree-grass interactions in savanna cannot be predicted by a simple model, but many interacting factors operating at various spatial and temporal scales are involved in creating and maintaining the savanna structure.

2.4. Trees-grass root niche partitioning, architecture, nutrient, and moisture uptake

The idea that rainfall and effective soil moisture is the main determinant of the savannas was first proposed by Walter & Mueller-Dombois (1971), who after visiting the savannas of the then South West Africa, now Namibia, hypothesised that the coexistence between trees and grasses is made possible by root niche partitioning. It is argued that the root niche partitioning occurs when trees develop tap roots to extract deep underground water, whereas grasses with their shallow roots extract water from the top layer, resulting in a vertical niche partitioning (Walter & Mueller-Dombois, 1971; Ward et al., 2013).

The Walter's two-layer hypothesis states that the ratio of trees to grasses is a function of vertical distribution of water in the soil profile because of their spatial differences

in their rooting depths (Sankaran et al., 2004). Accordingly, grasses become great competitors for water in the upper soil layer, while trees have exclusive access to water in the deeper soil depths by extending their roots systems into greater soil depths (Sankaran et al., 2004). This model further argues that when overgrazing occurs, the grass layer is reduced, resulting in the penetration of water further in the subsoil. This in turn favours trees and shrubs to become dominant, eventually leading to bush encroachment (de Klerk, 2004). Despite its popularity, this hypothesis has been disputed in several studies such as those of Holdo and Nippert (2015); Kulmatiski et al. (2020) and Nakanyala (2020), arguing that savanna plants develop contrasting root system architecture (RSA), not consistent with the root niche partitioning hypothesis.

Root system architecture is defined as the spatial configuration of the root system which is the explicit geometric deployment of root axes (Lynch, 1995). Savanna shrubs have different RSA, ranging from lateral root system, tap root system, dual root system and fibrous root system (Nakanyala, 2020). Therefore, not all shrubs forage for water and nutrients from deep soil layers. Those with lateral roots would absorb water from shallow soil, deploying their roots at the same depth as grasses, contrary to the root niche partitioning hypothesis. Kulmatiski et al. (2020), argue that there is a hydrological niche partitioning between trees and grasses that allow the coexistence, regulated by vertical rooting distribution and stochastic climate conditions. For example, trees have exclusive access to subsoil water because their roots are deeper than those of grasses whereas grasses with their shallow roots would only use water from the subsurface layer (Kulmatiski et al., 2020).

It has also been argued that rooting depth does not necessarily influence the uptake rate at which a plant forage or competes with neighbouring individuals, but their root size and shape determine the amount of water and nutrient uptake (Nippert & Holdo, 2015). For example, fine roots versus coarse roots (Nippert & Holdo, 2015). The cumulative transport capacity for fine roots and coarse roots differs, although their root biomass is similar at a given depth (Nippert & Holdo, 2015). Fine roots are described to be more active on water and nutrients acquisition and have a faster respiration rate than coarse roots (Satomura & Fukuzawa, 2007). Additionally, fine roots fulfil nutritional, metabolic, and symbiotic function and contribute to carbon stocks and terrestrial productivity (Jaloviar et al., 2009; Pang et al., 2022). Whereas, coarse roots act indirectly by providing connections between shoots and fine roots (Milikin & Bledsoe, 1999).

The root system architecture is one of the most important plant traits, as roots not only anchor plants' body to soil but also determine the availability of nutrients to plants (Teramoto et al., 2019). Root system architecture describes the structure and spatial arrangement of the root system within the soil and, therefore, crucial to nutrient and water uptake system (Freschet et al., 2020; Lynch, 1995; Rogers & Benfey, 2015). The RSA is an indicator of plants' ability to tolerate abiotic stressors and a driver for productivity enhancement (Lynch, 1995). Root spatial arrangement determines where the roots are in their growing environment, i.e., the soil. Generally, the partition and spatial distribution of the root system affect its water and nutrients uptake ability (Becker & Castillo, 1990). For instance, as the width, depth and branching of the root system increases, plant water stress decreases (Becker & Castillo, 1990). Many soil resources are unevenly distributed, or are subjected to localised depletion, so that the

spatial deployment of the root system would in large measure determine the ability of a plant to exploit those resources, hence, the importance of root system architecture (Lynch, 1995). The root structure refers to the diversity of individual roots and root segments within the root system and root system topology (Freschet et al., 2020).

Root system architecture is categorised in terms of their morphological organisation, ontogenesis, topology as well as root distribution (Doussan et al., 2003; Kong et al., 2014; Lynch, 1995). Root morphology is referred to as the surface features of a single root axis of an organ, including root hairs, root diameter and root cap (Lynch, 1995). Root topology refers to how individual root axes are connected to each other through branching (Lynch, 1995). Whereas root distribution refers to the presence of roots in a positional gradient of grid (Lynch, 1995). Root systems architecture have been further classified based on their developmental sequence defined by three fundamental groups which are: primary, adventitious, and lateral roots (Doussan et al., 2003). Primary roots are those formed in the embryo and grow directly downwards as the tap root and initiates lateral roots, referred to as the tap root system (Castañeda et al., 2019). The taproot system consists of a tap root and coarse lateral roots, and becomes the most prominent root, and then many smaller branch roots grow from this tap root (Yang et al., 2015). Nodal (or adventitious) roots originate from the shoot system, at locations such as stem nodes with different temporal patterns and are mostly abundant which give rise to a fibrous root system (Doussan et al., 2003). The fibrous root system forms with numerous fine roots about similar size developing from radical, in which the radical is short-lived (Hodge et al., 2009). The fibrous system has higher root number than taproot system (Yang et al., 2015). These roots tend to concentrate on the topsoil layer and a significant number of lateral roots might grow downward from

these roots to provide an effective absorption system (Mickovski, 2002). Lateral roots on the other hand develop when branching from the primary root at the right angle and differentiate from parent roots' younger tissues at a certain distance from the root apex. The branching process allows older lateral roots to be near the base of tap root and younger lateral roots are concentrated towards the root tip (Doussan et al., 2003). There could be several orders of lateral root branching as a result of much-branched condition characterised by a fibrous root system (Kerk & Sussex, 2001). Some plant species could have both primary and secondary root systems, and both remain important, often referred to as a dual root system (Nakanyala, 2020).

The root system architecture classification also reflects the differences between the two distinct vesicular plants which are monocotyledon and dicotyledon (Hodge et al., 2009). For dicotyledon plants, root system emerged from primary roots with branching secondary (lateral) roots that exhibit radical plants' growth (Hodge et al., 2009). These roots become woody, providing a strong anchor for plants and trees, which also help in adaptation for searching water (Hodge et al., 2009). Depending on the extension of lateral roots in relation to the primary axis, the morphology of the roots system would vary between tap rooted or fibrous rooted system (Doussan et al., 2003). In monocotyledons, the primary roots are generally short-lived and the plant's main root system develops from roots that arise from the shoot either adventitious or from nodal regions (Kerk & Sussex, 2001). They are characterised by a large number of root axes originating from the stem and they are generally not strongly gravitropic but quite sensitive to water and temperature tropism (Doussan et al., 2003). Lateral roots in monocotyledons species are initiated outside the phloem whereas, in the dicotyledons species they are initiated in the pericycle near protoxylem (Kerk & Sussex, 2001).

The analysis on the uptake of water and soil nutrients demonstrated that plant roots absorb soil water through the process of osmosis (Araya, 2007). Osmosis is the movement of water molecules from areas of high water potential (in the soil) to areas of low water potential (plant xylem) (Araya, 2007). In addition, the root uptake and nutrients availability depend largely on soil moisture because without water, plants cannot acquire nutrients regardless of the source (Reynolds et al., 2012). Nutrients are mineralised, transported to the root, and are taken up only in the presence of water (Scholers & Archer, 1997). According to van der Waal (2010), water availability contributes to the increase of nitrogen deposition in the soil.

Accordingly, plant roots take up soil nutrients through three different mechanisms such as root interception, mass flow, and diffusion (Alaoui et al., 2022). Mass flow is described as the convective passage of nutrients dissolved in plants when the plant takes up water for transpiration (Alaoui et al., 2022; Matimati et al., 2014). According to Matimati et al. (2014), high transpiration of water fluxes may be important in the acquisition of mobile nutrients or in zones with sparsely distributed roots. During this process, nutrient ions that are soluble in soil solution move with soil water as the water flows across the root system (Matimati et al., 2014). Mass flow is reported to transfer large amounts of calcium and magnesium, with little amount of potassium (Alaoui et al., 2022). Diffusion on the other hand takes place where nutrients ions move by active transportation from areas of high concentration to areas of lower concentration (Oyewole et al., 2014). When mineral concentration in the plant root system reduces, it creates a difference in concentration, therefore, nutrients naturally move to the region of lower concentration to reach equilibrium (Alaoui et al., 2022). The soil-root transfer

mechanism for phosphorus and potassium (K) is likely to occur during the diffusion process (Alaoui et al., 2022).

Root interception takes place during root growth when roots intercept with nutrients ions in the soil colloids and take up minerals from the root surface (Alaoui et al., 2022). This process requires a good structure for interception as soil compaction could limit root growth, hence, interception with nutrients in the soil. For example, sandy soils are considered favourable for root growth because of their sufficient pores and aeration (Zhou et al., 2009). Root interception is reported to only move a small amount of calcium (Ca) and magnesium (Mg) (Alaoui et al., 2022).

Despite the plant roots' mechanisms on water and soil nutrients, the direct mechanistic evidence for niche partitioning and understanding of the below-ground structure of these root architecture types is still lacking. This lack of knowledge is particularly because of the difficulties associated with studying the hidden half of the savanna plants (O'Donnell et al., 2015). No robust method has proven to effectively reveal the belowground life of savanna plants yet.

2.5 Distribution of the belowground biomass

Root distribution includes the horizontal and vertical extent of soil from which plant roots interact with the soil matrix to acquire soil resources (Freschet et al., 2020). Vertical root distribution refers to the distribution of roots in units of biomass or length over sequential soil layers ranging from the soil surface to deeper soil layers (Freschet et al., 2020). Efforts to understand the vertical distribution of root biomass include studies carried out in ecosystems such as the Mediterranean, woodland, tropical dry

forest, tropical moist forest, and semiarid steppe (Macinnis-Ng et al., 2009; Martínez et al., 1998; Noguchi et al., 2014; Raherison & Grouzis, 2005; Zhang et al., 2019). These studies show that belowground biomass allocation is vertically correlated with soil resources, i.e., water and soil nutrients. For example, more root biomass is allocated at the depth where water and nutrients are abundant (Cairns et al., 1997; Freschet et al., 2020).

Savanna landscapes are heterogeneous systems and resources such as soil nutrients vary widely in space and time (van der Waal., 2010). Furthermore, the quantities of root biomass and rooting depth in soils differ with the type of plants, types of soil, root system architecture, and plant canopy (Ansley et al., 2014; Hook et al., 1994; Macinnis-Ng et al., 2009). For example, root mass in forest ecosystems ranges from 2 to 5 kg m^{-2}, while in croplands, deserts, and grasslands is < 1.5 kg m^{-2} (Gregory, 2006). Tundra and tropical dry forest ecosystems almost have similar root biomass distribution whereby most of the root biomass in these environments is distributed in the first 30 cm of the soil profiles. For example, 80% of root biomass in Tundra and 60% of root biomass in tropical dry forests are distributed in the first 30 cm of soil depth (Gregory, 2006; Raherison & Grouzis, 2005). In contrast, Tropical grassland and the Mediterranean have superficial roots with more roots distributed below the soil surface with 44% of root biomass at 10 cm depth in tropical grassland and 50% of root biomass in the upper 12 cm in the Mediterranean (Gregory, 2006; Martínez et al., 1998) (Table 1). On the other hand, Noguchi et al. (2014) only studied fine roots in tropical moist forests and found that more than 74% and 93% of fine root biomass were distributed in the first 20 and 40 cm of the soil layers, respectively.

Table 1. Variation of root biomass at different soil depths in different ecosystems (source: Gregory, 2006; Martínez et al. 1998).

Type of ecosystem	Depths with root biomass (m)	Amount of root biomass (%)
Tundra	0.3	80
Tropical grassland	0.1	44
Tropical dry forest	0.3	60
Mediterranean	0.12	50

In terms of soil texture, more BGB is allocated to loamy sandy soil which is most likely to be on the upper layer as compared to clay soil which is most likely to be concentrated in the deep soil horizon (Macinnis-Ng et al., 2009). This is because loamy sandy soil would allow plant roots to have readily access to soil water during moist periods, unlike clay soil which is heavy-textured and thus, reduces the rate of deep percolation of water (Zhou et al., 2019). Therefore, it takes more time for water to reach the deeper soil layer compared to the upper soil layer (Macinnis-Ng et al., 2009).

2.6. Plants root research: A methodological challenge

Knowledge of the belowground interaction of plants in the savanna is very important but still limited due to the challenges associated with exposing plant roots for empirical measurements. Several direct and indirect methods have been used to expose plant roots. However, no robust method exists to study plants' root structure.

2.6.1 Direct methods used in plant roots studies

Various direct methods that have been used to study plant roots include excavation, coring, trenching, monolith, and ingrowth bags. The excavation method is the oldest method used in ecological root research which involves exposing the complete root system of a plant whereby surrounding soil is carefully being excavated manually (Böhm, 1979). The method is considered destructive, laborious, and time consuming (Hipondoka et al., 2003; Nakanyala 2020). Therefore, exposing several plants and covering a representative study area is nearly impossible.

The coring method is done manually by pushing or hammering sampling equipment into the soil using various devices from simple, sharpened steel augers to advanced cryogenic devices for sampling wetland soil (Maeht et al., 2013). The soil coring approach poses challenges in studying plant roots at greater depths, especially in rocky areas (Maeght et al., 2013; Rau et al., 2009). The method requires a large number of samples which could result in excessive costs (Maeght et al., 2013). Soil compaction from the sampling process might result in overestimation or root mass density (Freschet et al., 2020). Lastly, it is very challenging to sample on very thick roots (Freschet et al., 2020).

The trenching method, on the other hand, is done by digging a pit along the plant to determine the vertical rooting pattern and involves quadrat mapping and root counting (Schuurman & Goedewaagen, 1971). It is, thus, destructive and laborious. It is also difficult to establish a deep trench due to soil type and the influence of moisture levels. For example, the method cannot be applied in areas where clay soil is dominant

because clay swells to an extent that the nails used for trench-making disappear when the trench is sprayed with water (Kücke et al., 1995).

The monolith method involves taking soil cores and separating the soil from roots by washing, where trenches are first made before the sampling procedure (Böhm, 1979). The method is applicable for investigation where the aim is to make a quantitative determination of the root (Böhm,1979). However, the washing procedure is laborious and nearly impossible to wash all samples immediately after excavation (Smit & Rethman, 1997). Removing all organic matter particles from large samples is difficult and the washing prosses could result in root loss (Freschet et al., 2020).

Ingrowth bags method works by filling the root free soil in a mesh bag, bury it into the roots zone and allow root growth for a specified duration (Chen et al., 2018). The bag is later pulled out to determine root length inside the core (Chen et al., 2018). Although the method allows direct calculations of root productions, it has disadvantages of greatly modifying the root growth environment (Makkonen & Helmisaari, 1999). According to Steingrobe et al. (2000), the method is unable to obtain same soil conditions inside the bag as outside, which can result in a different root growth pattern between ingrowth soil and bulk soil. Nevertheless, this method is very labour intensive (Chen et al., 2018; Steingrobe et al., 2000).

2.6.2 Indirect methods used on plant roots studies

Indirect methods such as ground penetrating radar (GPR), minirhizotron, tracers, rhizotrons, and scanners have been employed to study the savanna's belowground structure. The GPR uses electromagnetic radiation to locate objects or interfaces buried

beneath the soil surface. The physical principle of GPR detection is based on the dielectric contrast between the buried target and the background material (Ferrara et al., 2014). It contains radio-wave emitter and a receiver antenna that picks up electromagnetic signals (Raz-Yaseef et al., 2013). This method has been used in studying the belowground structure of plants on the basis that it could provide non-invasive, areal, and repeatable underground measurements (Liu, et al., 2016). However, this method has complications that may result due to soil texture, water content, and root depth which largely affect the system performance (Liu et al, 2016). In addition, GPR is unable to detect individual fine roots (Liu et al., 2016).

The minirhizotron technique is a non-destructive approach, based on the application of transparent tubes, located in plant's root zone (Gluszek et al., 2013). Minirhizotron tubes are placed in the soil and are widely used for measuring fine roots (Maeght et al., 2013). Among other scholars, it has been used by Kulmatiski et al. (2017) to examine the effect of soil type and precipitation on fine-root abundance in savanna. However, the rooting depth was not well represented as the technique is more likely to capture the growth of smaller, faster growing roots compared to larger roots (Kulmatiski et al., 2017). The limitation associated with this approach is linked to the equipment and installation cost, potential changes in soil hydrology and physics and weather conditions (Dannoura et al., 2008; Mohamed et al., 2017). For example, when soil freezes due to too low temperature, some roots become invisible to the tube, thus, capturing, and image analysis become difficult. In addition, it is difficult to measure the whole root system as the measurable area of a single tube is small (Dannoura et al., 2008). Lastly, underground insects such as ants could destroy the tube (Gluszek et al., 2013), which limits the success of data collection.

Rhizotrons is a transparent wall technique that allows a researcher to observe plant roots and their rhizospheres and rhizoplanes on a serial basis while the roots are growing in the soil (Taylor et al., 1990). The method involves the use of root windows adapted to the well depth (Maeght et al., 2013), or covered underground walkaways with clear windows on one or both sides through which roots are observed at regular intervals (Maeght et al., 2013; Taylor et al., 1990). The method is laborious to install and results in disturbances of the study area (Neumann et al., 2009). Termites and other wood-eating insects might temper with root windows and lessen the accuracy (Freschet et al., 2020). The window used for observation is static and represents a limited two-dimensional area which does not provide details on the full extension of root systems (Neumann et al., 2009). Nevertheless, the method is expensive to construct and operate (Taylor et al., 1990).

Another indirect approach that has been used to study savanna plant roots involves the use of tracers. Tracers can be injected at different depths for later recovery in the biomass and the amount of tracers in the plant biomass is related to the uptake from each plant. In their study, Kulmatiski et al. (2020), used isotope depth-specific tracers namely H_2O to study trees and grasses root distribution. However, results obtained by tracers could be highly manipulated by hydraulic uplift or redistribution that occurs underground (McCulley et al., 2004). This water movement makes it difficult to conclude that the results obtained at a specific depth pertain to the corresponding depth or might have been carried along by moving water.

Lastly, the scanning method includes the visualisation of plant roots growth in the soil (Rogers et al., 2016). It can be done using photos, and the digital output of root image is then stored in the computer for image analysis (Dannoura et al., 2008). Example of scanning is an X-ray computed tomography which enables three-dimensional reconstruction of soil cores to estimate a wide range of soil features including root system architecture (Teramoto et al., 2020). This method has a challenge on the limited resources available for data analysis (Rogers et al., 2016). Other limitations include long scanning and reconstruction times, small scanning areas, and laborious processes involved in root segmentation (Teramoto et al., 2020). The accuracy of scanning could be affected by a lower scan resolution which is unable to capture fine lateral roots, thus, the full root system cannot be determined (Daly et al., 2017). Results could also be affected by the sample preparation process. For instance, the root length limit on the scanner bed would force large samples and root bunches to be cut into small segments, which is considered a major error in root length estimation (Zappalla et al., 2013).

2.7 Prospects of radiogenic strontium isotopes as a tracer for moisture uptake in the savannas

The search for sound methods to study the depth of moisture uptake by savanna plants has gone as far as the use of isotopes as tracers for studying the depth at which plants source moisture. Isotopes are elements in nature that are made up of atoms with the same number of protons and electrons, but different numbers of neutrons (Michell & Kendall, 2008).

Two categories of isotopes exist, stable isotopes and radiogenic isotopes (Lewis et al., 2017). Stable isotopes are those that occur naturally and do not undergo radioactive decay (Schwarcz et al., 2010). Radiogenic isotopes are produced by a radioactive decay process of elements such as rubidium (^{87}Rb) which slowly decays with a half-life of almost 50 billion years (Bayon et al., 2021). The radiogenic isotopes such as lead (Pb), strontium (Sr), and neodymium (Nd) are mostly used as tracers in the environment because they are heavy isotopes, unlike light isotopes such as hydrogen (H), oxygen (O), and sulfur (S) (Vallero, 2014). Heavy isotopes are relatively unaffected by changes in temperature and pressure during transport and accumulation, variations in the rates of chemical reactions, and the coexistence of different chemical species available in the environment (Vallero, 2014).

Strontium has four naturally occurring isotopes which are ^{88}Sr (82.53%), ^{87}Sr (7.04%), ^{86}Sr (9.87%), ^{84}Sr (0.56%) (Slovak et al., 2018). Three of these naturally occurring isotopes are stable whereas one (^{87}Sr) is radiogenic, and is, therefore, variable, as it is partially formed by the radioactive decay of the naturally occurring ^{87}Rb, which has a half-life of nearly 50 billion years (Frei, 2012). However, the Sr isotopic tracer system relies on the use of two naturally occurring isotopes (^{86}Sr and ^{87}Sr), because their isotopic ratios are nearly related to their natural abundance (between 7% ^{87}Sr and 10% ^{86}Sr) (Frei, 2012). Other radioactive strontium isotopes are such as ^{85}Sr, ^{89}Sr, and ^{90}Sr, with ^{90}Sr being the major radioisotope as well as the long-lived isotope that has been applied mostly in the soil studies (Burger et al., 2019). The natural isotopes are commonly occurring alkaline earth metals, usually found in nature in the form of minerals like celestite and strontianite (Gupta et al., 2018). It is contained in all plant and animal organisms in an amount of 10^{-2} to 10^{-3}% of dry mass (Dubchak, 2018).

Strontium isotopes could also be added into the soil during soil formation processes through the mechanism of different weathering processes (Aguzzoni et al., 2019). In addition, human activities such as milling and processing of strontium compounds, burning of coal, land application of phosphate fertilizer, as well as the usage of phosphate devices contribute to the addition of strontium in the atmosphere (Höllriegl, 2019). On the other hand, radioactive strontium and calcium are released into the environment due to atmospheric nuclear weapons tests and subsequently fall out onto the soil (Dubchak., 2018).

Strontium itself is not a plant's essential nutrient, but it is common where Ca is present because they have similar chemical properties (Reynolds et al., 2012). Strontium and calcium are homologues, both are alkaline earth metals, and behave similarly in the environment (Gupta et al., 2018). Therefore, strontium is highly plant available and becomes part of plants' shoots (Gupta et al., 2018). The absorption of Sr in soils is mainly caused by ion exchange (Dubchak, 2018). Plant species that take up large amounts of calcium absorb more Sr from the soil, therefore, Sr and Ca concentrations for different species grown in the same soil are positively and linearly related (Veresoglou et al., 1996). According to Höllriegl (2019), strontium is a natural constitute of food because once absorbed into a plant via the roots, it is distributed to other parts of the plant such as leaves or fruits.

Strontium and calcium can either come from deep soil through the slow chemical weathering of local substrates or from shallow soil depths by the alterations of silt-sized, carbonate-rich dust (Reynolds et al., 2012). The rate of Sr migration under

experimental conditions increases with an increase of exchangeable calcium content (Dubchak, 2018). The migration is influenced by soil properties, plants, and calcium, e.g., Sr migrates faster downward into the soil especially in sandy and organic soils compared to other radionuclides (Gupta et al., 2018).

Radiogenic strontium isotopes have mostly been applied in provenance studies such as tracing the origins, mobility patterns, trade, and exchange networks among ancient people, fauna, and artefacts (Frei, 2012; Slovak et al., 2018). Strontium isotopes have also been used in archaeology to study migration and movement patterns of people. For example, Bentley et al. (2002) used Sr isotopes to study the prehistoric migration of central Europe by measuring Sr isotopes from human skeletons, human teeth, and bones, which provides a geochemical signature of the place of residence. Moreover, Sr isotopes have also been used in the field of geology to understand the chronology of rock formation (Chang et al., 2022; Zhou et al., 2013). Whereas Bayon et al. (2020), investigated the radiogenic Sr composition of modern river sediments from various geological, tectonic, and climatic settings worldwide.

Moreover, radiogenic Sr isotopes have been applied in soil science, weathering, and hydrological studies, because of the large availability in isotopic composition and interest in tracing sources and cycling of analogy element calcium, (Blum & Erel, 2003; Nakano et al., 1993). According to Adams et al. (2019), each region has a specific $^{87}Sr/^{86}Sr$ ratio based on the surrounding geology and water sources, for example, the geology in the soil has a specific Sr signature, thus plants inherit isotopic ratios of their habitat. Strontium isotope was used by Aguzzoni et al. (2019) to study the variability of Intra and intertree Sr ratios in different apple orchards and recorded a moderate homogeneity of Sr ratio among subsamples of the same tree parts, high

homogeneity among different tree parts and low Sr ratio homogeneity among different trees. Additionally, Poszwa et al. (2009) studied the supply of mineral nutrients (atmosphere and soil mineral weathering) of different plant communities in tropical inselberg using Sr isotopes. Likewise, Coble et al. (2015) used Sr isotope ratios $^{87}Sr/^{86}Sr$, as a tracer along a semiarid substrate age gradient within San Francisco volcanic field, and measured Sr values in aeolian dust, soils, and vegetation to determine whether the contribution of atmospheric sources of rock-derived nutrients to soil and vegetation pools varied with substrate age. Critical understanding of the belowground interaction, soil resources acquisition and roots deployment in the savanna requires the use of vigorous approaches. It is, therefore, for this reason that this study assessed the potential of radiogenic Sr isotope as an effort to develop a robust method and help to expand our knowledge of the belowground structure of savanna plants.

2.8 Conclusion

The savanna is the largest and unique biome in Africa with such a unique coexistence of tree-grass that live together and forage for the same resource. The unique coexistence is influenced by several biotic factors such as soil moisture and nutrients availability and abiotic factors such as fire, grazing and browsing, of which soil moisture is considered the main determinant factor. Several studies have taken interest in the belowground structure of savanna plants to understand the dynamics under which trees and grasses forage for moisture without displacing each other. Different theories have been developed by different scholars and among others is the initial hypothesis known as the Walter's two-layer model which states the coexistence of tree-grass in savanna is regulated by a root niche partitioning. However, this model

has been challenged by several researchers, some arguing that RSA, root distribution, root size and shape contribute to nutrients and soil moisture uptake as opposed to rooting depth. Some studies explained the dynamics of savanna structure in the context of competitive models, which state that savanna is an equilibrium biome that is regulated by natural events while other studies used demographic models which refer to savanna as a non-equilibrium biome whose structure is constantly modified by anthropogenic disturbances.

Despite all these contrasting theories, our knowledge of savanna coexistence is still fragmented. For example, it is still not known whether there is a root niche differentiation or a root niche competition between trees and grasses. The subject on shrubs' root system architecture and factors that influences rooting patterns as well moisture and nutrients uptake is still poorly understood. This lack of understanding is due to several methodological challenges, particularly in accessing the plant root systems. Numerous methods ranging from direct to indirect methods have been used by several studies to study plant roots. However, access to plant roots for empirical measurements have been a challenge due to limitations associated with such methods. Although recent studies have moved away from the use of direct methods due to challenges such as their destructiveness and time consuming, indirect methods used up to date also have shortcomings associated with equipment costs and installation, and unfavourable to certain weather and soil conditions, among others. It is, therefore, for this reason that this study tested the ability of radiogenic strontium isotopes approach as a robust method to help understand the savanna's belowground structure. This was done because plant roots take along strontium during nutrients uptake. Therefore, the isotopic ratios in plant shoots are expected to be the same with the

isotopic ratio of soil depths at which plant roots are deployed, hence, determining their root deployments.

CHAPTER 3: MATERIALS AND METHODS

3.1 Description of the study area

This study was conducted in the savanna shrubland of the Kalahari Basin (KB), near Tsumkwe. The Kalahari Basin is a mega structural basin in Southern Africa, extending across parts of several countries including Angola, Namibia, Botswana, South Africa, Zambia, and Democratic Republic of Congo (DRC) (Thomas & Shaw, 1991). The study was done at a site located some 30 km north of Tsumkwe, along the D3315 road (Figure 2). The area is dominated by relict longitudinal sand dunes, fixed by vegetation (Goudie & Viles, 2015). This study was done at a landscape level, to complement a parallel study with a focus on constructing bioavailable strontium isoscapes at a district level for provenience studies of archaeological and ethnographic ostrich eggshell beads. The study site is located 10 km from the nearest settlements occupied by the Ju/'hoansi community, who are part of Khoisan population. They are involved in small-scale livestock farming and small business enterprises including selling craft works, and wildlife conservation (through Nyae Nyae conservancy).

3.1.1 Vegetation

The vegetation around Tsumkwe is classified as savanna known as savanna shrubland because it is dominated by savanna shrubs (Thomas & Shaw, 1991). The surrounding area is of typical savanna structure, characterised by shrubs, and herbaceous plants, with few trees. Vegetation density at the site is moderate, with dispersed shrubs, averaging 0.2 shrubs per m^2 (83 shrubs in a 20 m x 20 m plot), while their heights range from 0.8 m to 1.9 m. The most dominant shrub species are *Terminalia sericea, Grewia flava, Acacia mellifera and Dichrostachys cinerea.*

3.1.2 Climate

The study area is characterised by a summer rainfall of approximately 450 mm/year (Hüttich et al., 2009). There is a distinct seasonality of dry winters and wet summers due to the influence of Intertropical Convergence Zone (ITC) associated with warm and moist air masses in the summer months (Hüttich et al., 2009). Mean daily temperature ranges from 21 ºC to 22 ºC of which maximum and minimum temperatures are higher during the wet season (Atlas of Namibia Team, 2022).

3.1.3 Soils

The Kalahari Basin is dominated by sandy soils also known as the Kalahari sands, covering up to 2.5 million km^2, making it one of the largest sand seas in the world (Thomas & Shaw, 1991). The Kalahari sand soils where this study was conducted consist of more than 95% fine sand-sized, aeolian-deposited sediment, structureless and relatively infertile, lacking in N, P, and organic matter (Dougill & Thomas, 2004; Mendelsohn & Obeid, 2002). The sand sheet is generally deep to very deep, well drained and leached, with low agricultural productivity (Strohbach & Kutuahuripa, 2014). The soils have low organic matter which vary from 0.20% in the upper soil layer to 0.08% in the subsoil, on average (Ringrose et al, 1998). Dominant minerals are iron and oxide which give the soil a reddish colour and poor in nutrients (Nakanyala, 2020). The Kalahari sands are relatively uniform on the surface; however, a heterogeneity prevails on the subsurface as sands are interbedded with layers of calcretes and silcretes of different thickness (Ringrose et al., 1998). The Kalahari sands further consist of small pans and dry valleys which resulted from duricrusts formation and ground water weathering (Hauwanga, 2018). The pan soils comprise of either sand

pan when they are shallow, rocky soil when calcrete layer crops out or sandy clays mainly rich in sodium (Na) potassium, and magnesium ions (Werger, 1978).

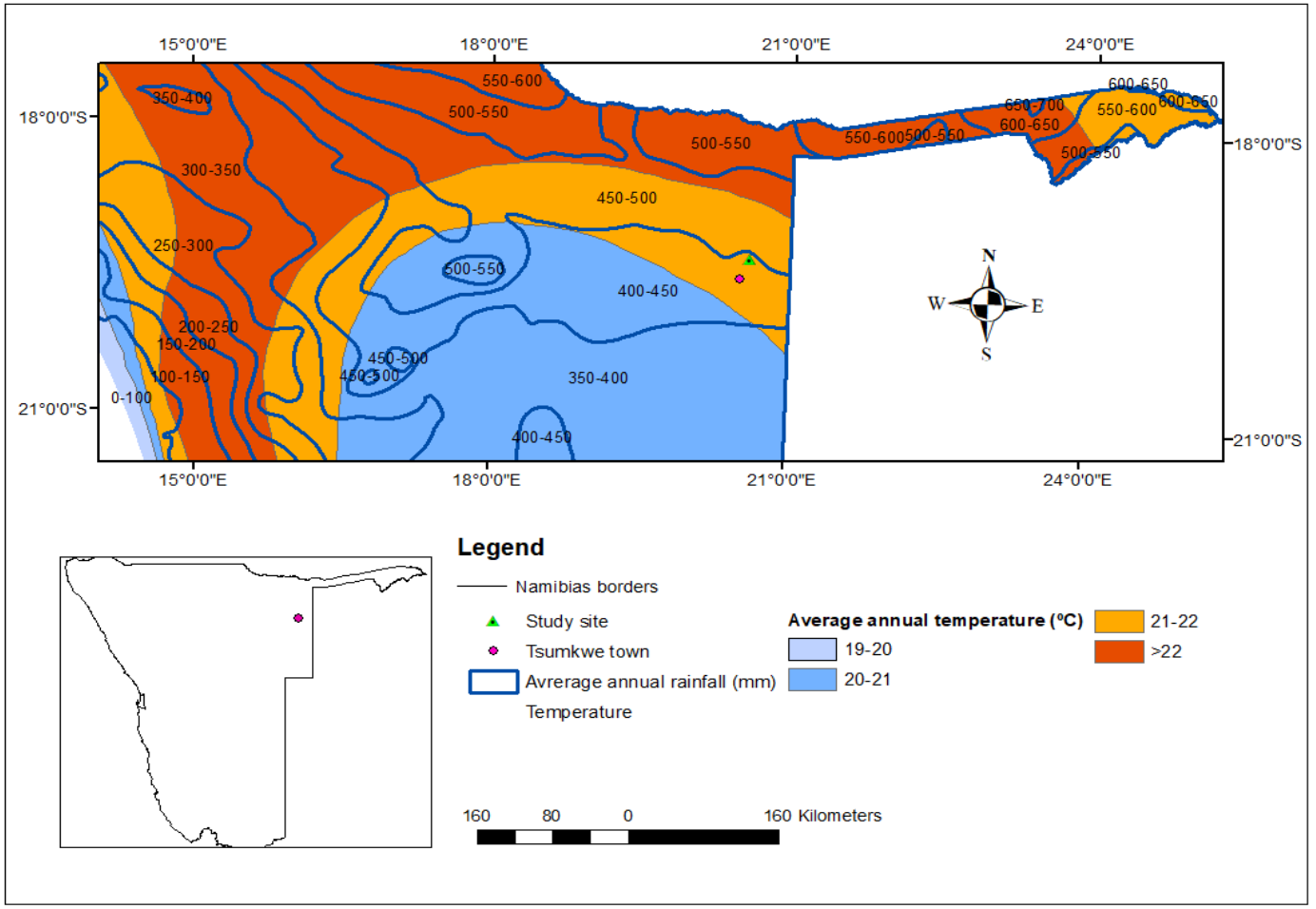


Figure 2. A study area map showing the Namibian average annual rainfall gradient (mm) and Namibian average annual temperature (ºC) (source: Atlas of Namibia Team, 2022).

3.2 Data Collection

Data collection started with a reconnaissance survey around the study area to identify suitable sites with natural vegetation. Sites to be considered for the study should at least be more than 200 m from any anthropogenic disturbance such as villages/settlements and roads. Three sites were subsequently identified as suitable for sampling and one site was randomly selected. The selected site was located in a transition zone. Transition zone is a geographic area of overlap, which displays a set of different plants species, or a different trend of vegetation with both broad-leave and narrow-leave plants.

3.2.1 Data collection for root biomass

Data collection started with demarcating a plot of 20 m x 20 m using a measuring tape and market using ropes and poles. Within that plot, five transects were laid with an interval of 5 m. Thereafter soil samples were collected using a soil auger (7 cm diameter) along each transect, at an interval of 2.5 m distance. Soil coring from 45 sampling points was done at an interval of 10 cm along the soil profile up to 1 m depth. A total of 450 soil samples were collected from the site. Each soil sample was sealed in a ziplock bag and air dried for 3 weeks at a room temperature before laboratory analysis commenced.

3.2.2 Data collection for strontium isotopes, soil physicochemical properties, and the excavation process

The 20 m x 20 m plot was divided into four quadrants and one shrub was randomly selected in each quadrant. For each selected shrub, the four nearest shrubs were selected using a nearest-neighbouring method. Species names for each selected shrub

were identified and recorded. About 100 g of fresh leaves were collected from all the selected shrubs and enclosed in sample paper bags.

The above ground structures of selected shrubs such as shrubs height, shrub canopy, and distance to the nearest neighbour were measured. In order to ascertain plant roots architecture and development in the soil profile, the shrub roots were exposed using the excavation method (Böhm, 1979). A spade and trowel were used to excavate both lateral and taproots and had their rooting system exposed up to a depth of 1 m. Root features such as root lengths, root depths, and root diameters were measured using a measuring tape and a vernier calliper. The types of root system architecture for each excavated shrub such as lateral root system, taproot system and dual root system were also recorded. *Grewia flava* was one of the dominant shrubs at the site and due to their complex, fibrous rooting structure which posed difficulties in exposing them, only 2 of the 5 selected *Grewia flava* shrubs were excavated.

The distribution of strontium isotopes and soil physicochemical properties in the soil profile was assessed from soil samples collected from five cores. Four of these cores were sited within plant canopies of *Grewia flava*, *Dichrostachys cinerea*, *Senegalia mellifera* and, *Terminalia sericea*. These same shrubs are among the shrubs whose leaves were sampled for isotopic analysis. One core was taken beyond plant canopies. Samples were once again collected using a soil auger (7 cm diameter) at a 10 cm interval up to a depth of 1 m. The sampling spots were randomly selected. Both plant leaves and soil samples were sent and analysed for Sr isotopes at the Multicollector - Inductively Coupled Plasma Mass Spectrometer (MC-ICPMS) laboratory, South Africa.

3.3 Laboratory procedures for root biomass and soil physicochemical properties

Air-dried soil samples were weighed and sieved using a 5 mm and 2 mm sieve to retain coarse and fine roots respectively. Non-root organic matters were separated manually from both sets. Both fine and coarse roots were then weighed, and oven dried overnight at 105 ºC temperature and weighed again, for biomass determination. Soil physicochemical properties such as: phosphorus, potassium, calcium, magnesium, nitrogen, sodium, potential hydrogen (pH), organic matter (OM), electrical conductivity (EC), and soil texture were analysed in the soil laboratory at the Ministry of Agriculture, Water and Land Reform, Windhoek.

3.4 Data analysis

Data analysis for root biomass, root density, soil physicochemical properties, and Sr ratios was done in RStudio version R 4.2.1. Firstly, a Shapiro-Wilk test was run to test the data for normal distribution, which confirmed that the data were not normally distributed. A Kruskal-Wallis test was conducted to assess the effect of depth and canopy status on dependent variables such as root biomass, root density, and soil physicochemical properties. A Kruskal-Wallis test was also conducted to assess the variation of Sr ratios across the soil profile and the variation of Sr ratios at different canopy statuses. A post hoc pairwise comparison for the Kruskal-Wallis test was done using the Dunn's test. Results were interpreted at a significance level of 0.05 alpha. Root density was determined by calculating the weight of dry roots in mg per volume of soil (cm^3).

The average root biomass (g) of each sampling point was interpolated by the Inverse Distance Weighted (IDW) interpolation technique, to assess the spatial variation of

root biomass across the study site. ArcMap 10.3 was used to perform the IDW analyses and to produce root biomass distribution maps. Interpolation is the method of estimating the value of properties at unsampled sites within the area covered by existing point data (Achilleos, 2008). The interpolation method that was used in this study is based on the concept that points close to each other are similar compared to those that are far away (Sentianto & Triandini, 2015). Three different root biomass maps presenting a) fine root biomass, b) coarse root biomass and c) total root biomass with five classes ranging from very low to very high were generated. The threshold values for each class were generated through statistical method: maximum - minimum/ number of classes.

In order to establish the relationship between plants' Sr ratios and those of soil depths, the plants' Sr ratios were compared with the cumulative average of soil depths' Sr ratios at which their roots were deployed as well as with the soil mean Sr ratios of their maximum rooting depths. Shrubs whose canopies were sampled for soil Sr analysis where also compared with the soil's Sr ratios of their respective cores. A z-score test was performed to measure the variation of plants' and soil depths' Sr ratios from the mean.

3.5 Research Ethics

The researcher obtained an ethical clearance certificate from the Decentralized Ethics Committee (DEC) at the University of Namibia before proceeding with the research. The temperature and rainfall data were freely available and were downloaded from Atlas of Namibia Team website. After shrub excavation and soil coring, the land was levelled back to its original level.

CHAPTER 4: RESULTS

4.1 The belowground structure and plants root deployments (as part of objective a)

4.1.1 Roots biomass and root density along the soil profile

There was a significant variation in total root biomass ($H(9) = 189.66$, $p = 0.00$) and total root density ($H(9) = 138.59$, $p = 0.00$) with depth along the soil profile. Total root biomass was highest at the depth of 10-20 cm and lowest at the depth of 90-100 cm (Figure 3a). Post hoc pairwise comparison (Table 2) and descriptive statistics for fine and coarse root biomass (Table 4) revealed that total root biomass varied across different soil depth intervals, with the upper depth at 0-10 cm significantly different from that of 20-30 cm and that of 30-40 cm depth. Furthermore, significant differences were noted between the upper and middle depths of 10-20 cm and 40-50 cm; 20-30 cm and 50-60 cm; 40-50 cm and 60-70 cm, as well as the lower depths such as 70-80 cm and 80-90 cm; 80-90 cm and 90-100 cm. In contrast, there was no significant difference between some successive soil depths, starting from the depth of 10-20 cm and 20-30 cm and up to the depth of 70-80 cm and 80-90 cm (Table 2).

Table 2. Dunn test pairwise comparison results for total root biomass. The number of stars shows a significant level. Note: ***p < 0.001, **p < 0.01, * p < 0.05. Values with no alpha indicate that there was no significant difference between the depths.

Depth (cm)	0-10	10-20	20-30	30-40	40-50	50-60	60-70	70-80	80-90	90-100
0-10										
10-20	1.34									
20-30	2.49**	1.15								
30-40	3.13***	1.79*	0.64							

40-50	3.72***	2.38**	1.22	0.58					
50-60	5.30***	3.96***	2.80**	2.17*	1.58				
60-70	6.46***	5.12***	3.97***	3.33***	2.75**	1.16			
70-80	6.66***	5.32***	4.17***	3.53***	2.95***	1.36	0.20		
80-90	6.84***	5.50***	4.35***	3.71***	3.12***	1.54	0.38	0.18	
90-100	8.71***	7.35***	6.19***	5.54***	4.95***	3.35***	2.18**	1.97*	1.79*

Root density followed the same pattern with a significant variation with soil depths (Figure 3b). A Post hoc pairwise comparison revealed that the depth class of 0-10 cm was significantly different from the depth of 20-30 cm. Other depth classes which were significantly different from each other are such as depth 10-20 cm and 30-40 cm; 40-50 cm and 60-70 cm, 70-80 cm and 80-90 cm; 80-90 cm and 90-100 cm. Whereas other numerous depths did not yield any significant difference, ranging from the depth of 10-20 cm and 20-30 cm, 30-40 cm and 40-50 cm, 40-50 cm and 50-60 cm, 50-60 cm and 60-70 cm, 70-80 cm, and 80-90 cm (Table 3).

Table 3. Dunn test pairwise comparison results for total root density. Note: ***p < 0.001, **p < 0.01, * p < 0.05. Values with no alpha indicate that there was no significant difference between the depths.

Depth (cm)	0-10	10-20	20-30	30-40	40-50	50-60	60-70	70-80	80-90	90-100
0-10										
10-20	1.43									
20-30	2.51**	1.08								
30-40	3.16***	1.73*	0.65							
40-50	3.76***	2.33**	1.25	0.61						
50-60	5.36***	3.94***	2.85***	2.21*	1.60*					

60-70	6.60***	5.18***	4.09***	3.44***	2.84**	1.24			
70-80	6.66***	5.24***	4.16***	3.51***	2.91**	1.30	0.06		
80-90	6.92***	5.49***	4.41***	3.76***	3.16**	1.56	0.32	0.26	
90-100	8.69***	7.25***	6.16***	5.51***	4.89***	3.27***	2.02*	1.96*	1.70*

Both fine root biomass and root density were highest at the depth class of 0-10 cm and lowest at depth of 90-100 cm. Coarse root biomass and density were highest at 10-20 cm depth and lowest at 90-100 cm (Table 4).

Table 4. Measured vertical fine root biomass (g ± SE) and fine root density (mg/cm^3 ± SE) as well as coarse root biomass (g ± SE) and coarse root density (mg/cm^3 ± SE), along the soil profile.

Depth (cm)	Fine root biomass (g)	Fine root density (mg/cm^3)	Coarse root biomass (g)	Coarse root density (mg/cm^3)
0-10	0.67 ± 0.08	1.75 ± 0.22	0.35 ± 0.11	1.41 ± 0.41
10-20	0.35 ± 0.05	0.92 ± 0.12	0.73 ±0.30	1.91 ± 0.77
20-30	0.23 ± 0.05	0.60 ± 0.14	0.24 ± 0.07	0.63 ± 0.18
30-40	0.18 ± 0.03	0.44 ± 0.06	0.25 ±0.08	0.64 ± 0.20
40-50	0.19 ± 0.05	0.49 ± 0.12	0.53 ±0.23	1.27 ± 0.60
50-60	0.10 ± 0.02	0.27 ± 0.06	0.25 ± 0.10	0.66 ± 0.23
60-70	0.06 ± 0.01	0.16 ± 0.03	0.30 ± 0.16	0.79 ± 0.41
70-80	0.06 ± 0.02	0.15 ± 0.05	0.18 ± 0.08	0.47 ± 0.19
80-90	0.06 ± 0.02	0.14 ± 0.04	0.20 ± 0.08	0.53 ± 0.22
90-100	0.03 ± 0.01	0.09 ± 0.02	0.01 ± 0.01	0.04 ±0.02

The proportion for root biomass (Figure 3c), illustrates that overall, the depth of 10-20 cm has the highest proportion of total root biomass (21.69%) while the lowest total root biomass (0.98%) was recorded at the depth of 90-100 cm. Although the percentage of root biomass decreases with increasing soil depths, the depth of 40-50 cm comes in the 3rd position with more percentage total root biomass (14.38%), higher than those of depths 20-30 cm and 30-40 cm (9.46% and 8.59%, respectively). The cumulative percentage on the variation of the root biomass along the soil profile showed that 52% of the total root biomass were found in the first 30 cm of the soil profile and 75% within 50 cm soil depth (Figure 3d).

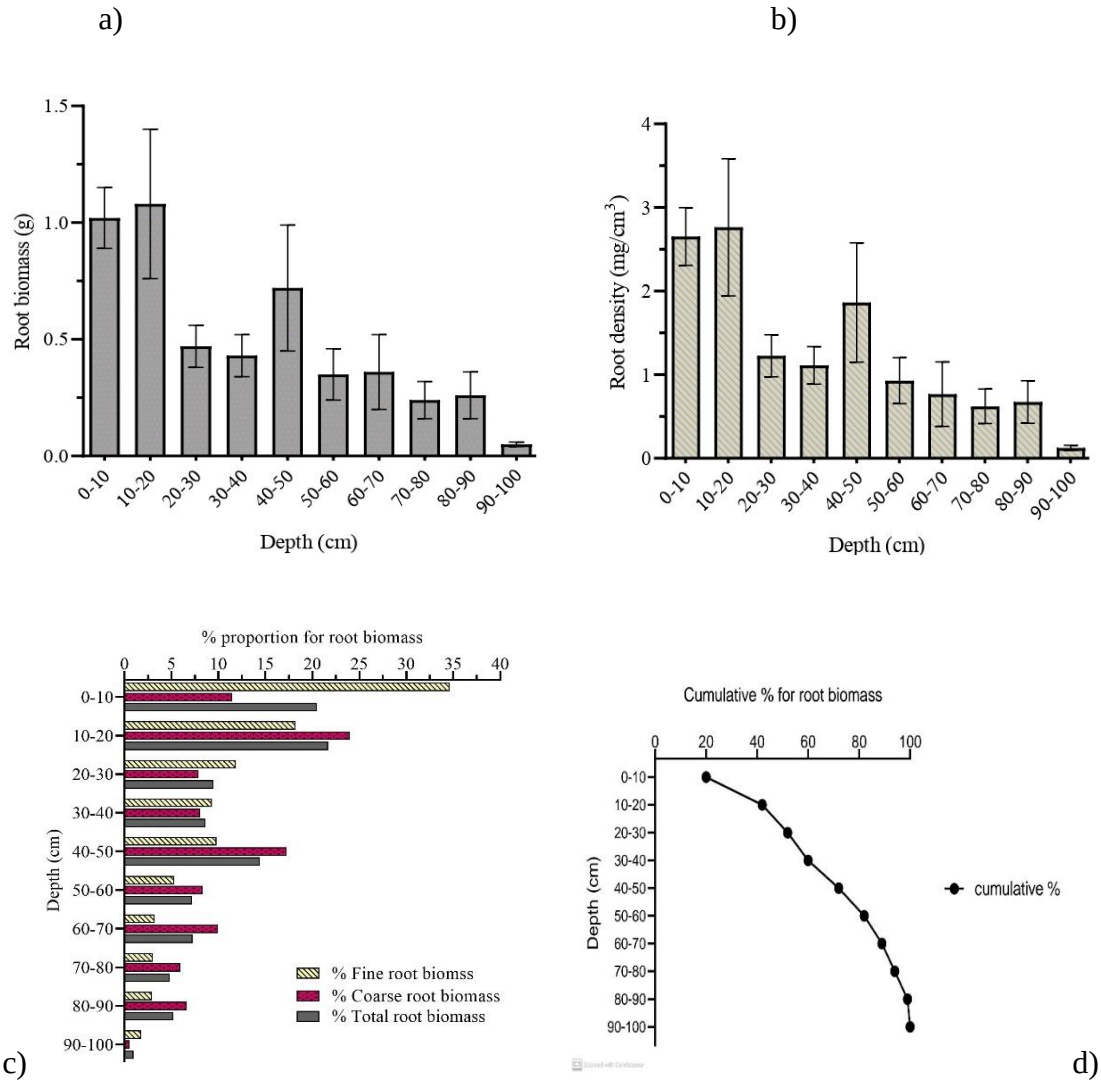


Figure 3. Measured vertical total root biomass (g ± SE) (a); and measured vertical total root density (mg/cm^3 ± SE) (b), along the soil profile; percentage proportion of root biomass at different soil depths (c); and percentage biomass cumulative curve along the soil profile (d).

4.1.2 Variation in total root biomass and total root density within and beyond plant canopy

There was a significant variation in total root density (H (1) = 4.30, p = 0.04) and total root biomass (H (1) = 5.37, p = 0.02) between the inside plant canopy and the outside plant canopy. Root biomass was higher under plant canopy (0.63 ± 0.10 g) as compared

to outside canopy (0.40 ± 0.05 g). Similarly, root density was higher under canopy (0.77 ± 0.21 mg/cm^3) as compared to outside canopy (0.52 ± 0.10 mg/cm^3). Overall, samples taken under the canopy covered about 53.35% of the total root biomass. Results indicated a decrease in total root biomass and total root density with increasing soil depth within and outside plant canopy (Figure 4a & b), respectively.

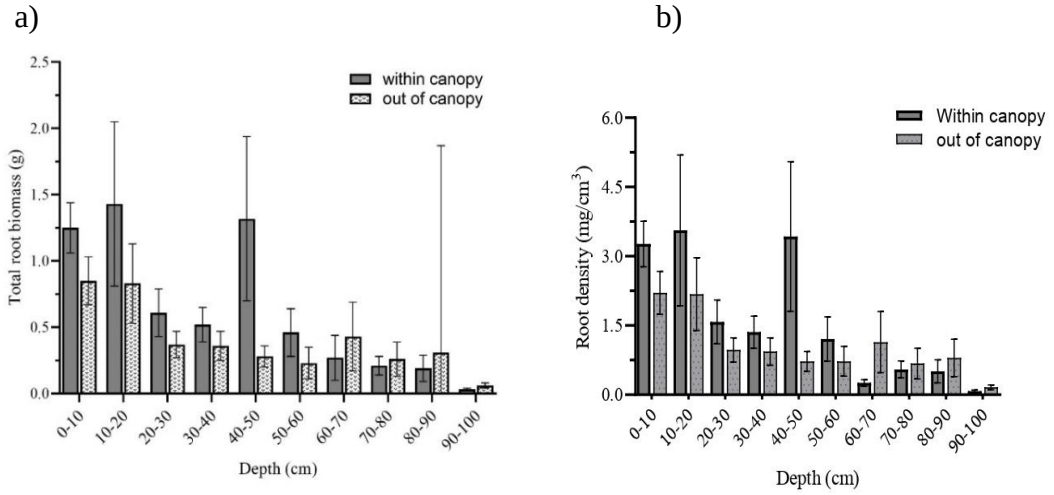


Figure 4. Measured vertical total root biomass within plant canopy and outside plant canopy (g ± SE) along the soil profile (a) and measured vertical total root density within plant canopy and outside plant canopy (mg/cm^3 ± SE) along the soil profile (b).

4.1.3 Spatial distribution of root biomass (IDW) and vegetation density across the study site

Firstly, the distribution of root biomass was found to relate with the number and distribution of plants across the plot. Overall, results from interpolation showed that there was a low spatial distribution of total root biomass across the study site (Figure 5c). The "low" class covered the highest proportion of the study site. This class was widely spread across the plot but with low distribution towards the south-western area and covering 64% of the area. The second dominant class is the "very Low" which

covers 20.7% of the study site. This class was widely distributed at the south-western part of the plot but could be seen with isolated patches across the entire plot. The "moderate" class is in the third position, and it covered about 9.9% of the study area. This class is widely distributed across the area with dispersed patches. The "high" class and "very high" class have the least distribution and cover 4% and 1.4%, respectively (Figure 5c).

The distribution of fine root biomass followed a similar pattern with total root biomass distribution, whereby the low class dominated the site with 60.6%, and it was widely distributed across the plot except in the south-western section which is dominated by a very low class. The moderate class came near second with 20.8%. The very low class is also seen with dispersed patches across the plot and became the third dominant class covering 15% of the area and mostly dominated the northwestern part of the study site. The high and very high class have a low distribution with 2.7% and 0.9%, respectively in small, dispersed patches across the plot. However, these classes were more distributed at the centre, northwestern part and southeastern of the plot (Figure 5a).

On the contrary, the spatial distribution of coarse root biomass was very low at the study site with the 'very low' class covering more than half of the area (55.9%). This class was widely distributed across the entire plot except some parts of north-western and north-east. The low class came second with 34.5% of the area covered. The low class was mostly distributed at the western and north-eastern parts of the area with dispersed patches on the south-eastern areas. The moderate class came third and it covered (6.7%) of the area with small patches. There was a low distribution of high and very high class with 2.4%, 0.5%, respectively (Figure 5b). Overall, the result from

interpolation indicates that there is a low spatial distribution of root biomass at the study area.

The vegetation distribution indicated that quadrant 1 (southwest) and quadrant 3 (northeast) were found to have comparable and lower vegetation density. For example, quadrant 3 had 18 shrubs and they were distributed linearly in a north south direction. In quadrant 1, there were about 19 shrubs, and they were mostly dominated at the centre of the quadrant. Whereas quadrant 4, in the northwest of the plot, had about 22 shrubs that were randomly distributed across the quadrant. Lastly, in Quadrant 2 (southeast of the plot), the shrubs were randomly distributed across the quadrant. This quadrant was also found with a high number of shrubs (28).

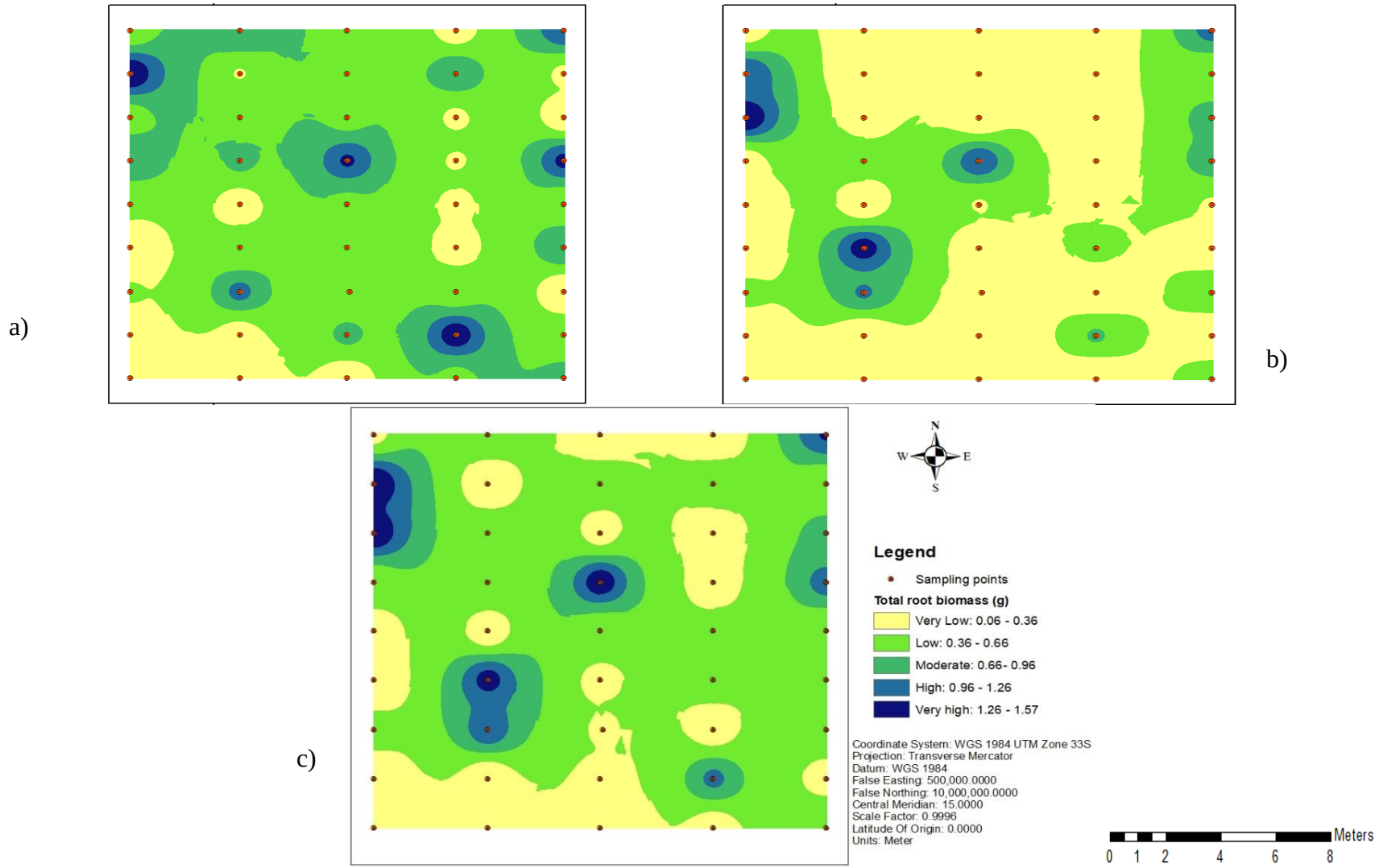


Figure 5. Spatial distribution of root biomass across the study site, where a) is fine root biomass; b) is coarse root biomass and c) is total root biomass.

4.2 Vertical distribution of soil physicochemical properties at the study area (as part of objective b)

Soil was largely made up of sand (89.5%), while the proportion of clay and silt was 6.2% and 4.3%, respectively. Although the soil was largely dominated by sand, the sand was more abundant on the topsoil layer and reduced slightly with soil depth, i.e., 90.5% of sand at 0-10 cm and 88.9% at 90-100 cm. Silt and clay, on the other hand were lower at the upper depth and increase slightly with increasing soil depth. For example, there was 3.4% of silt and 5.5% of clay at 0-10 cm and increased to 4.1% and 7.0% at 90-100 cm, respectively.

Results showed that among all the nutrients, there was only a significant variation in P ($H(9) = 16.9$, $p = 0.05$) and OM ($H(9) = 26.7$, $p = 0.00$), with different soil depths. Both P, OM and OC were highly concentrated on the topsoil layer and reduced significantly with increasing soil depth (Table 5). Other elements such as K ($H(9) = 4.0$, $p = 0.91$) and N ($H(9) = 8.65$, $p = 0.47$ did not yield any significant difference across soil depths.

Table 5. The soil physicochemical properties at different soil depths (cm) across the study site.

Depth (cm)	P (ppm)	K (ppm)	Ca (ppm)	Mg (ppm)	Na (ppm)	pHw	EC (μS/cm)	OM (%)	N (%)
0-10	1.24 ± 0.13	66.00 ± 17.10	2472.20 ± 681.77	47.60 ± 6.65	84.00 ± 10.42	8.55 ± 0.13	78.6 ± 4.43	0.68 ± 0.13	0.18 ± 0.04
10-20	0.74 ± 0.10	58.20 ± 21.22	2953.6 ± 619.36	39.60 ± 7.26	80.80 ± 40.65	8.55 ± 0.11	68.02 ± 6.03	0.53 ± 0.04	0.13 ± 0.02
20-30	0.70 ± 0.19	54 ± 15.18	2741.40 ± 567.66	37.60 ± 3.91	87.40 ± 19.48	8.63 ± 0.02	65.24 ± 6.75	0.45 ± 0.04	0.11 ± 0.02
30-40	0.72 ± 0.19	39.80 ± 12.66	2356.00 ± 573.73	30.40 ± 3.83	87.40 ± 19.48	8.73 ± 0.09	59.94 ± 6.29	0.41 ± 0.03	0.10 ± 0.02
40-50	0.44 ± 0.17	36.40 ± 13.79	2483.00 ± 501.84	27.80 ± 4.85	77.00 ± 38.45	8.79 ± 0.90	58.9 ± 8.92	0.38 ± 0.02	0.09 ± 0.01
50-60	0.54 ± 0.13	40.40 ± 13.60	3076.00 ± 583.42	24.60 ± 4.23	98.60 ± 33.80	8.73 ± 0.12	66.8 ± 5.53	0.39 ± 0.05	0.10 ± 0.01
60-70	0.54 ± 0.13	46.60 ± 11.66	2891.00 ± 614.69	25.40 ± 4.96	111.20 ± 34.05	8.74 ± 0.09	68.62 ± 3.49	0.35 ± 0.02	0.09 ± 0.01
70-80	0.63 ± 0.33	47.00 ± 12.02	3295.20 ± 687.95	24.20 ± 3.35	91.20 ± 5.46	8.77 ± 0.08	68.9 ± 2.41	0.33 ± 0.02	0.09 ± 0.01
80-90	0.62 ± 0.22	46.80 ± 9.92	3394.00 ± 604.67	26.40 ± 3.80	109.20 ± 12.24	8.78 ± 0.06	70.46 ± 3.22	0.33 ± 0.02	0.09 ± 0.01
90-100	0.36 ± 0.10	51.00 ± 11.30	3406.80 ± 847.48	27.00 ± 3.58	102.20 ± 27.16	8.78 ± 0.13	67.88 ± 7.50	0.35 ± 0.02	0.09 ± 0.01

A Post hoc pairwise comparison for P and OM is presented in Table 6, 7, and 8, respectively. Results for P pairwise comparison revealed that there was a significant difference between the upper and lower soil depths of 0-10 cm and 30-40 cm; 10-20 cm and 40-50 cm; 0-10 cm and 50-60 cm; and up to 0-10 cm and 90-100 cm depth. Moreover, significant difference was noted between the middle soil depths such as depth 40-50 cm and 70-80 cm. On the contrary, there was no significant difference between some upper soil depths such as 0-10 cm and 10-20 cm; 10-20 cm and 30-40 cm, some middle soil depths such as 40-50 cm and 50-60 cm; 40-50 cm and 60-70 cm; as well as deeper soil depths such as 70-80 cm and 80-90 cm; 80-90 cm and 90-100 cm (Table 6).

Table 6. Dunn test pairwise comparison results for P. Note: ***p < 0.001, **p < 0.01, p < 0.05. Values with no alpha indicate that there was no significant difference between the soil depths.

Depth (cm)	0-10	10-20	20-30	30-40	40-50	50-60	60-70	70-80	80-90	90-100
0-10										
10-20	1.23									
20-30	1.63	0.40								
30-40	2.03*	0.79	0.39							
40-50	3.55***	2.32*	1.92*	1.52						
50-60	2.63**	1.39	0.99	0.60	-0.93					
60-70	2.26*	1.02	0.62	0.23	-1.29	-1.37				
70-80	1.67*	0.44	0.03	-0.36	-0.88*	-0.96	-0.59			
80-90	1.97*	0.74	0.34	-0.05	-1.58	-0.65	-0.28	0.31		
90-100	2.87**	1.63	1.23	0.83	-0.69	0.24	0.61	1.19	0.89	

A pairwise comparison for OM revealed that there was a significant difference between the depths of 0-10 cm and 30-40 cm; 0-10 cm and 40-50 cm; 0-10 cm and 50-60 cm; 10-20 cm and 60-70 cm; 20-30 cm and 70-80 cm; 10-20 cm and 90-100 cm. Whereas, other depth classes did not yield a significant difference, ranging from the upper depths such as 0-10 cm and 10 cm; 10-20 cm and 20-30 cm, middle class soil depths such as 40-50 cm and 50-60 cm; 50-60 cm and 60-70 cm to lower depths classes such as 70-80 cm and 80-90 cm; 80-90 cm and 90-100 cm depths (Table 7).

Table 7. Dunn test pairwise comparison results for OM. Note: ***p < 0.001, **p < 0.01, * p < 0.05. Values with no alpha indicate that there was no significant difference between the soil depths.

Depth (cm)	0-10	10-20	20-30	30-40	40-50	50-60	60-70	70-80	80-90	90-100
0-10										
10-20	0.19									
20-30	0.96	0.77								
30-40	1.65*	1.47	0.70							
40-50	2.17*	1.99*	1.22	0.52						
50-60	2.00*	1.82*	1.04	0.35	-0.17					
60-70	2.97**	2.78**	2.01*	1.32	0.79	0.97				
70-80	3.28***	3.09***	2.33**	1.63	1.11	1.28	0.32			
80-90	3.19***	3.01**	2.24*	1.54	1.02	1.19	0.23	-0.09		
90-100	2.93**	72.75**	1.98*	1.28	0.76	0.93	-0.03	-0.35	-0.26	

Although Na does not significantly vary with soil depths (H (9) = 8.66, p = 0.47), results showed that it was relatively lower in the upper soil layers and more in the deeper layers. Nitrogen on the other hand was more at the upper depths compared to deeper depths (Table 5). Results recorded for soil physicochemical properties at the

site, including macro nutrients such as P and N showed a low amount of soil nutrients with the mean of 0.81 ± 0.17 ppm and 0.11 ± 0.01% respectively. Results only recorded an adequate amount of Ca among all nutrients at the depths of 50-60 cm, 70-80 cm, 80-90 cm, and 90-100 cm with 3076 ppm, 3295.2 ppm, 3394 ppm and 3406.8 ppm, respectively. The highest Ca was recorded at the depth of 90-100 cm and the lowest at the depth of 30-40 cm. Results also showed that the soil is alkaline with pH value of 8.17 ± 0.03 on average and that the pH was relatively constant throughout the soil profile. Lastly, results also recorded a very high saline soil at the study site with an average of 67.33 ± 1.73 μS/cm. The variation of soil salinity was also quite constant along the soil profile (Table 5).

4.3 Radiogenic strontium isotopes of plant leaves and the soil profile (as part of objective c and d)

4.3.1 Shrubs species, root's structure, and rooting depths

A total of 9 different species from 20 shrub samples across the study site were selected for sampling (Table 9). The excavated shrubs were recorded to have different rooting systems, namely: fibrous roots, taproots, and dual roots system. Some shrubs of the same species had similar rooting systems whereas others had different rooting systems. For example, all three *Terminalia sericea* that were excavated had a fibrous root system, while all three sampled *Dichrostachys cinerea* had a dual root system. However, species like *Ximenia caffra* had a completely different rooting system i.e., one had a fibrous root system, one had a taproot system and the other had a dual root system (Table 9). Overall, the fibrous root system dominated at the plot, and it was recorded from at least 9 shrubs of 6 different species. The taproot system was found to

be the least common with only one shrub (*Ximenia caffra*) recorded to have a taproot system and the remaining had a dual root system (Table 9).

These dynamics of rooting systems also applies to rooting depths. Some shrubs of the same species had nearly the same maximum rooting depths and some had different maximum rooting depths although they carry similar rooting systems. For example, one of the most dominant species at the site (*Terminalia sericea)* had maximum rooting depths that were closer from all the three shrubs, ranging between 30 and 53 cm. Nevertheless, there was a high distinction on the maximum rooting depths between some shrubs of the same species. These species were *Ximenia americana* whereby one shrub with fibrous roots was having a maximum rooting depth of 55 cm (1.4 m height), whereas those with dual root and taproot system had a maximum rooting depth beyond 1 m (1.9 m height). Additionally, one shrub of *Ximenia caffra* with a fibrous root system had a maximum rooting depth of 55 cm, one with a taproot system had a maximum rooting depth beyond 1 m and the other with a dual root system had a maximum rooting depths beyond 1 m as well. Lastly, three shrubs of *Dichrostachys cinerea* with a dual root system had different maximum rooting depths ranging from 0.65 m to beyond 1 m.

Consequently, these results indicated that the type of root system also influences the shrubs' root deployments, whereby those shrubs with lateral or fibrous roots deployed their roots between 10 cm and 55 cm and those with dual roots and taproots deployed their roots from 65 cm and beyond 1 m depth, respectively. Among all the excavated shrubs, two shrub species *Acacia hereroensis* and *Rhus tenuinervis* deployed their roots on a shallow soil layer which is 10 cm and 20 cm, respectively (Table 9). Results

from data collection also indicated that most shrubs with fibrous root system start to grow from a shallower depth i.e., 0.05 m, while some shrubs with dual root system had lateral roots that start to grow from deeper depths. For example, *Senegalia mellifera* was found to have some lateral roots that started to grow from as deep as 0.70 m that originated from the taproot as the shrub was recorded to have a dual root system (Table 9).

Lateral roots length of sampled shrubs ranged from 0.8 m to 5 m with the average of 1.97 ± 1.10 m (Table 9). The shorter lateral roots (0.8 m) were recorded from a *Dichrostachys cinerea* with a dual root system while the longest (5 m) lateral roots were recorded from a *Terminalia sericea* with a fibrous root system. Results also showed that the type of root system influenced the lateral roots length whereby shrubs with fibrous root system had longer lateral roots (2.43 m on average) while shrubs with dual root system had shorter lateral roots (1.46 m on average).

Table 8. Shrubs' species, root systems, and rooting depths of the sampled shrubs.

Shrubs' specie names	Type of root system	Average lateral root length (m)	Maximum rooting depth (m)	Depth at which roots start to grow (m)	Average maximum rooting depth (m)
Acacia hereroensis	fibrous root system	1.1	0.10	0 - 0.10	0.10
Commiphora angolensis	dual root system	1.98	> 1	0.20 - 0.30	> 1
Dichrostachys cinerea	dual root system	0.8	0.65	0.20 - 0.60	
Dichrostachys cinerea	dual root system	1.2	> 1	0.20 - 0.30	> 0.83
Dichrostachys cinerea	dual root system	1.8	> 1	0.05	
Grewia flava	fibrous root system	2.3	0.55	0.05 - 0.10	
Grewia flava	fibrous root system	4.1	0.50	0 - 0.20	
Grewia flava	not excavated	not excavated	not excavated	not excavated	52.5 cm Note: not all *Grewia flava* were excavated due to their complex root system.
Grewia flava	not excavated	not excavated	not excavated	not excavated	
Grewia flava	not excavated	not excavated	not excavated	not excavated	
Rhus tenuinervis	fibrous root system	> 2	0.20	0-10	0.20
Senegalia mellifera	dual root system	1.78	> 1	0.05 - 0.70	> 1
Terminalia sericea	fibrous root system	2.3	0.40	0.05 - 0.10	
Terminalia sericea	fibrous root system	2.37	0.30	0.03 - 0.10	0.41
Terminalia sericea	fibrous root system	5	0.53	0.03 - 0.10	
Ximenia americana	fibrous root system	1.9	0.55	0- 0.24	> 0.08
Ximenia americana	dual root system	1.42	> 1	0.03 - 0.30	

Ximenia caffra	tap root system	N/A	> 1	0	> 0.85
Ximenia caffra	fibrous root system	0.83	0.55	0	
Ximenia caffra	dual root system	1.27	> 1	0.05 - 0.10	

Results on the above-ground data showed that shrub canopies were comparable to the shrub heights whereby the average for both canopy diameter and shrub height was recorded to be 1.4 ± 0.59 m and 1.4 ± 0.47 m, respectively. However, a shrub (*Dichrostachys cinerea)* that recorded the highest height (2.5 m) was found to have the shortest canopy diameter (0.5 m). Shrub heights range from 0.13 m to 2.5 m whereas canopy diameter ranges from 0.5 m to 2.35 m.

4.3.2 Variation of Sr ratios among shrub species and different soil depths

All but two shrubs' Sr ratios varied within the range of 0.721509 and 0.721958. For example, one shrub (*Ximenia caffra*) with maximum rooting depth beyond 1 m recorded the lowest Sr ratio of 0.721222 (Figure 6a). Additionally, there was one of the three shrubs (*Grewia flava*) that was not excavated and recorded the highest Sr ratio (0.722600). The overall average Sr ratio for plants was 0.721725 ± 0.000268. The Sr ratios of some shrubs were very close to the overall average. For example, one shrub of the *Dichrostachys cinerea* in Figure 6(b) had a z-score value of -0.023945, and a *Senegalia mellifera* with a z-score value of 0.0829401 among others. Meanwhile, the z-score of 3.265378 for the *Grewia flava* with the highest Sr ratio, falls outside the range of the mean. Any z-score value above 3 or below -3 was considered an outlier, and, therefore, out of the range. Consequently, 95% of shrubs were within the range of the shrubs' Sr ratios and one plant (5%) was out of this range.

Moreover, results indicated that 13 shrubs had Sr ratios below the mean while 7 had Sr ratios that were above the mean (Figure 6b). Results also showed that the first 3 shrubs with the highest Sr ratio (0.722600; 0.721958; and 0.721933) were of the same species (*Grewia flava*). Although the Sr ratio was not species-specific, the result

revealed that there were shrubs of the same species with Sr ratios that were either below the mean or above the mean. For instance, all three shrubs of *Terminalia sericea* as well as the three shrubs of *Ximenia caffra* had Sr ratios that were below the mean, while both shrubs of *Ximenia americana* had Sr ratios above the mean (Figure 6b).

Although most shrubs recorded different Sr ratios, two shrubs of the same species (*Ximenia caffra*) were found to have the same Sr ratio of 0.721679. At the same time, these two shrubs had different maximum rooting depths of 55 cm and beyond 1 m, as well as different rooting systems (fibrous and taproot) (Table 9 and Figure 6a). There were also shrubs with comparable Sr ratios but of different species and different maximum rooting depths. These shrubs included *Commiphora angolensis* (0.721559), *Terminalia sericea* (0.721560), and *Dichrostachys cinerea* (0.721563), which had maximum rooting depths of > 1 m, 53 cm and > 1 m, respectively. Additionally, there were two shrubs of different species with Sr ratios closer to each other and their maximum rooting depths were also closer. These shrubs were *Terminalia sericea* (0.721596) and *Grewia flava* (0.721597) with maximum rooting depths of 30 cm and 55 cm, respectively, (Figure 6a). Results also showed that among all the sampled shrubs, there were shrubs of different species (*Grewia flava, Ximenia caffra, and Ximenia americana*) with similar maximum rooting depths (55 cm) and similar rooting systems as they were all fibrous root systems, but their Sr ratios were not comparable (0.721597; 0.721679 and 0.721893, respectively) (Table 9 and Figure 6a).

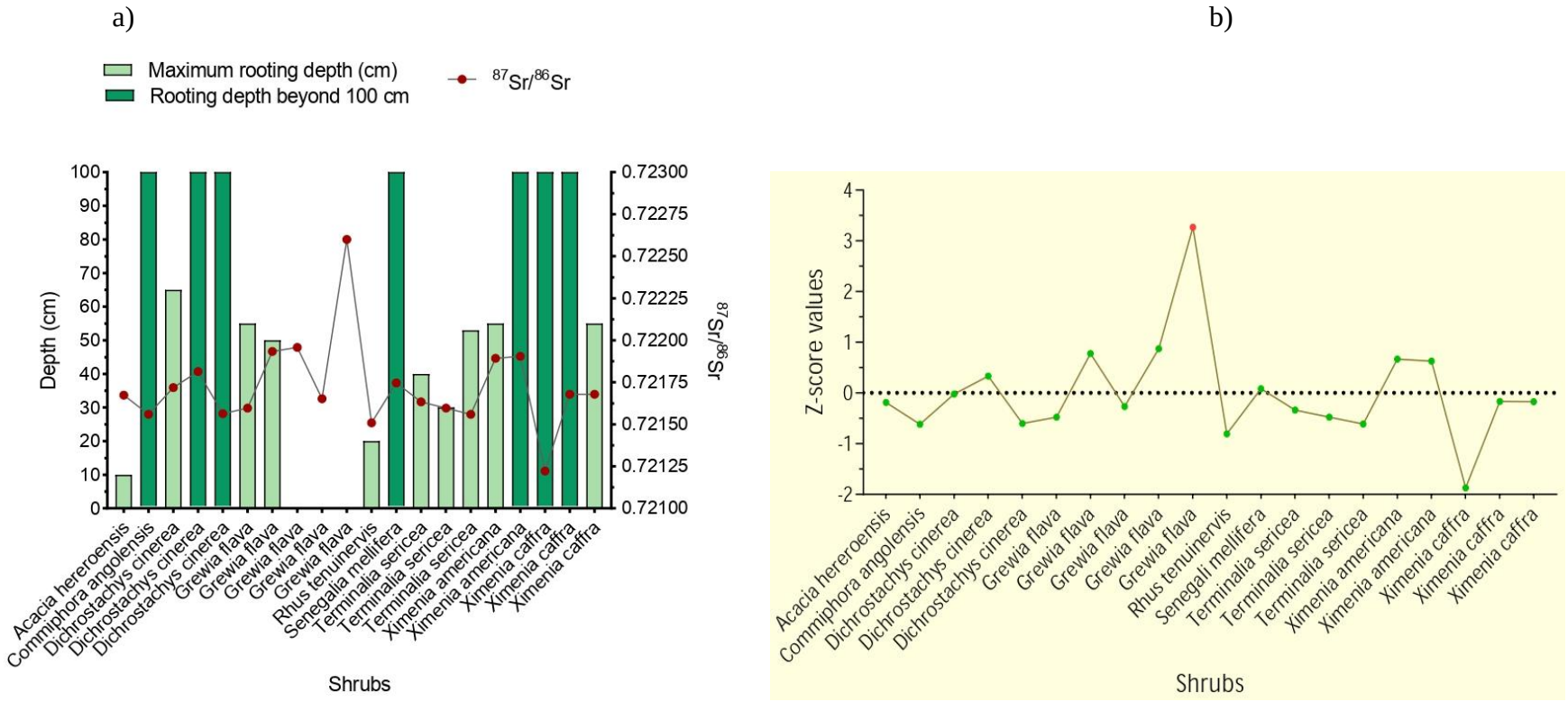


Figure 6. a) The variation of Sr ratios among different shrub species and their maximum rooting depths. Note: The rooting depths of the three *Grewia flava* are not known as they were not excavated. And b) Z-score values of shrub species where 0 represents the mean, positive (+) values are above the mean and negative (-) values are below the mean. The distance from the trendline indicates how far the Sr ratio of individual shrubs is from the mean. The closer the Z-score values are to the trendline, the closer the Sr ratios are to the mean and vice versa. Value marked with red colour was considered an outlier.

In terms of the Sr ratio variation with soil depths, the Sr values at the site did not show much distinct variation along the soil profile. Therefore, Sr was not significantly influenced by soil depth (H (9) = 9, p = 0.44). However, there was a slight variation of Sr ratios at different soil depths with the lowest $^{87}Sr/^{86}Sr$ ratio being 0.721422 at 70-80 cm depth and the highest being 0.722334 at 90-100 cm, (Figure 7a). The overall average soil Sr ratio at all depths was recorded to be 0.721636 ± 0.000272. The z-score results for soil depths showed that the Sr ratios from all the soil depths were found within the soil Sr ratio range. However, depth of 90-100 cm had a Sr ratio far from the mean with a z-score of 2.563258 while depths 50-60 cm and 80-90 cm had Sr ratios very close to the mean with a z-score of 0.036608 and -0.033195, respectively. Results also show that 6 soil depths (10-20 cm, 30-40 cm, 40-50 cm, 60-70 cm, 70-80 cm, and 80-90 cm) had Sr ratios below the mean and the remaining 4 soil depths had Sr ratios above the mean (Figure 7b).

The overall average of Sr ratios for all the samples collected for isotopic analysis (both plant and soil samples) was 0.721661 ± 0.000478. In terms of the z-score for the entire site, results indicated that all but one sample had Sr ratios within the site range. This outlier, with a z-score value of 4.972664 was recorded from a soil sample that was sampled at a depth of 90-100 cm, within the plant canopy. One of the soil samples from the depth of 20-30 cm was recorded with the lowest Sr ratio at the site, with a Sr ratio of 0.720904 and a z-score value of -1.583811. Nevertheless, results showed that most samples, both soil and plant samples had Sr ratios closer to the mean, (Figure 7c). The plant sample with the Sr ratio closest to the mean was a *Grewia flava* (one of the shrubs that were not excavated), with a Sr ratio of 0.721652 and a z-score value of -0.01835. Netherless, the results indicated that 69 (98.6 %) of all the samples were

found within the Sr ratio mean and one (1.4%) was out of the Sr ratio mean of the study site (Figure 7c).

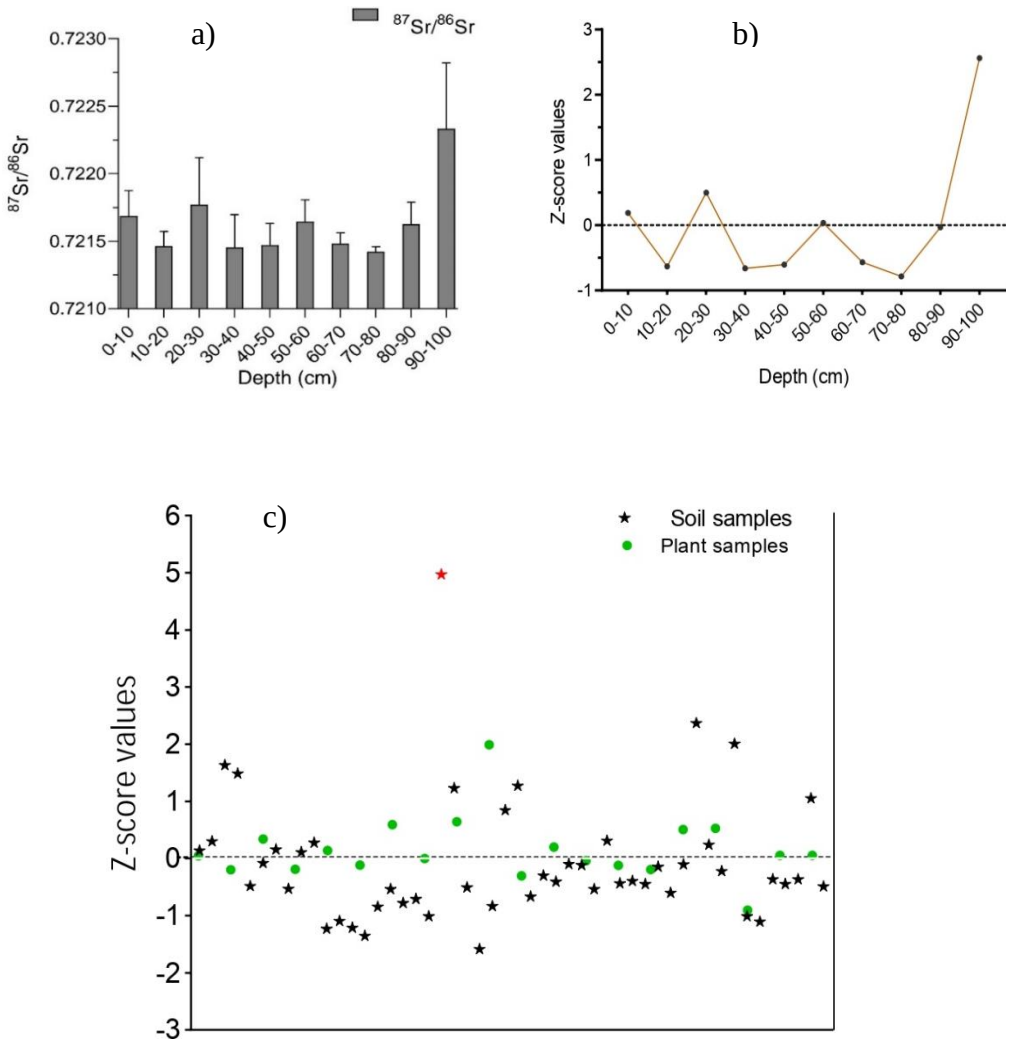


Figure 7. a) Average Sr ratio ($^{87}Sr/^{86}Sr \pm SD$) at different soil depths, (b) Z-score values of soil samples at different soil depths, and (c) Z-score values of all plants and soil samples at the study site. Where 0 represents the mean, positive (+) values are above the mean and negative (-) values are below the mean. Distance from the trendline indicates how far the Sr ratio of the individual sample is from the mean. The closer the z-score values are to the trendline, the closer the Sr ratios are to the mean and vice versa. Value marked with red colour was considered an outlier.

4.3.3 Variation of soil Sr ratios inside and outside the plant canopy

There was a significant variation in Sr ratio (H (1) = 3.8629, p = 0.05) between the soil samples taken from outside canopy and inside canopy. The Sr ratios from the outside canopy were slightly higher (0.721805 ± 0.000109) as compared to those under canopy which recorded a lower Sr ratio (0.721583 ± 0.000107) on average. The results also revealed that samples taken outside the canopy had higher a Sr ratio in the first 40 cm soil depth whereas samples taken within the canopy had a higher Sr ratio at a greater depth, as illustrated in Figure 8. The highest Sr ratio within the canopy was recorded at the depth of 90-100 cm while the lowest Sr ratio was recorded at a depth of 30-40 cm. The highest Sr ratio outside the canopy was recorded at the depth of 20-30 cm and the lowest was recorded at a depth of 70-80 cm.

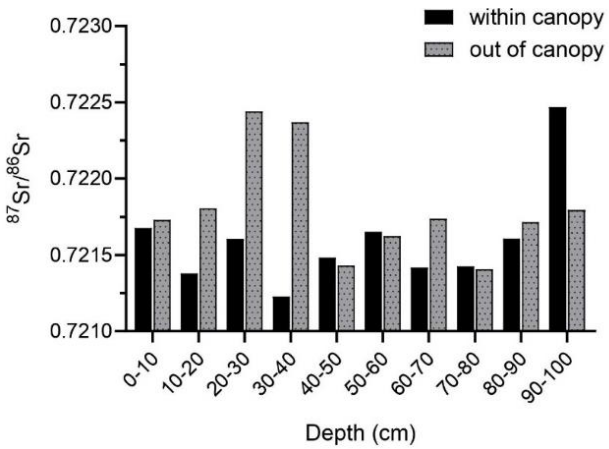


Figure 8. Variation of soil Sr ratios from inside and outside plant canopy, at different soil depths.

4.4 Comparison between plants' Sr ratio and soil depths Sr ratio (as part of objective e)

The shrubs' Sr ratios were compared with the cumulative average of soil depths at which individual plant roots start to grow until their maximum rooting depths. For

example, the Sr ratio for a *Dichrostachys cinerea* (0.721718) whose roots started to grow from the depth of 20 cm and had a maximum rooting depth of 65 cm was compared with the soil Sr average of depth 20 - 70 cm (0.721565 ± 0.000139) (Table 10 and Figure 9a).

Results from the first comparison showed that 13 shrubs had Sr ratios found within the same range of the average soil Sr ratios at which their roots were deployed. One of the good correspondences was that of *Acacia hereroensis* with a Sr ratio of 0.721674 and whose roots were deployed between 0-10 cm and an average Sr ratio of 0.721687 ± 0.000188 at such depths (Table 10 and Figure 9a). Other shrubs with Sr ratios closer to the average Sr ratios of their rooting zones include *Grewia flava* (0.721597) and a *Terminalia sericea* (0.721560), with roots deployed between 0-60 cm, whose Sr ratios were closer to the cumulative average of their rooting zone (0.721582 ± 0.000137). Some shrubs had Sr ratios that were found within the standard deviation however, they were distant from the mean. For example, *Ximenia americana* with roots deployed between 0-60 cm had a Sr ratio (0.721893) that was far from the mean (0.721582 ± 0.000137) of such depths.

The five shrubs whose maximum rooting depths go beyond 1 m and whose roots started to grow from 0 cm, were compared with the average soil Sr of 0-100 cm that was sampled for the study. Although their maximum rooting depths could not be determined, results showed that 4 of these shrubs had their Sr ratios corresponding with the average soil Sr ratio of the 0-100 cm depth (Figure 9a). Two of these shrubs (*Ximenia caffra and Senegalia mellifera)* had Sr ratios (0.721679 and 0.721747) closer to the average Sr ratio of 0-100 cm (0.721636 ± 0.000272) and the remaining two

(*Ximenia americana* and *Dichrostachys cinerea*) had Sr ratios (0.721904 and 0.721814, respectively) that were far from the Sr ratios mean of such depths.

Moreover, shrubs' Sr ratios were also compared with the soil mean Sr ratios per 10 cm interval at their correspondence maximum rooting depths (Figure 9b). It should also be noted that the soil Sr ratios presented were not obtained at every single depth but at an interval of 10 cm. Therefore, Sr ratios for the soil depths were recorded as an average at those 10 cm intervals. For example, depths 50-60 cm had a Sr ratio of 0.721646 on average but the Sr ratio of individual soil depths between such ranges is not expected to be exact with that of the mean.

Results from this comparison indicated that 10 shrubs had Sr ratios that were found within the standard deviation of soil mean at their respective maximum rooting depths. Four of these shrubs had Sr ratios closer to the soil mean Sr ratios of their respective maximum rooting depths. For example, *Acacia hereroensis* with a maximum rooting of 10 cm had a Sr ratio (0.721679) very close to that soil mean for depth 0-10 cm (0.721687 ± 0.000421). The same correspondence with *Rhus tenuinervis* with a maximum rooting depth of 20 cm had Sr ratios (0.721509) closer to that of soil mean Sr ratio for depth 10-20 cm (0.721464 ± 0.00243). Additionally, *Ximenia caffra* with a maximum rooting depth of 55 cm had a Sr ratio (0.721679) closer to the soil means' a Sr ratio of depth 50-60 (0.721646 ± 0.000357). Moreover, *Grewia flava* with a maximum rooting depth of 55 cm had a Sr ratio (0.721597) closer to the soil mean Sr ratio for depths 50-60 cm. The remaining 6 shrubs had Sr ratios found within the standard deviation of soil mean Sr ratios of their respective maximum rooting depths however, their Sr ratios were far from such means (Figure 9b).

The results also revealed that there were three shrubs with Sr ratios that did not correspond neither with the soil cumulative average of their rooting zones, nor with the soil mean of their maximum rooting depths at 10 cm intervals. These shrubs were *Grewia flava* with a maximum rooting depth of 50 cm and Sr ratio of 0.721933, *Dichrostachys cinerea* with a maximum rooting depth of 65 cm and Sr ratio of 0.721718, as well as *Ximenia caffra* with a maximum rooting depth beyond 1 m and Sr ratio of 0.721222. However, for *Dichrostachys cinerea,* its Sr ratio corresponds with the overall average of soil Sr ratio (0.721636 ± 0.000272), unlike for *Grewia flava* and *Ximenia caffra*. Nevertheless, *Ximenia caffra* was also recorded to have the lowest Sr ratio among all the shrubs sampled (Table 10 and Figure 6).

Although plants recorded a slightly higher Sr ratio on average (0.721725 ± 0.000268) than the average Sr ratios for soils (0.721636 ± 0.000272). Overall, results showed that there was an overlap between the shrubs' Sr ratios (n = 20) and the soils' Sr ratios (n = 50) in terms of their standard deviations, (Figure 9c).

Table 9. The Sr ratios of different shrub species and average soil's Sr ratios ($^{87}Sr/^{86}Sr$ ± SD) at different rooting zones

Depth (cm)	Specie Names	Plant Sr	Soil Sr ± SD
0-10	*Acacia hereroensis*	0.721674	0.721687 ± 0.000188
0-20	*Rhus tenuinervis*	0.721509	0.721575 ± 0.000158
0-30	*Terminalia sericea*	0.721596	0.721641 ± 0.000159
0-40	*Terminalia sericea*	0.721634	0.721575 ± 0.000158
0-50	*Grewia flava*	0.721933	0.721570 ± 0.000149
0-60	*Grewia flava*	0.721597	0.721582 ± 0.000137
	Terminalia sericea	0.721560	
	Ximenia caffra	0.721679	
	Ximenia americana	0.721893	
0-100	*Dichrostachys cinerea*	0.721814	0.721636 ± 0.000272
	Senegalia mellifera	0.721747	
	Ximenia americana	0.721904	
	Ximenia caffra	0.721679	
	Ximenia caffra	0.721222	
20-70	*Dichrostachys cinerea*	0.721718	0.721565 ± 0.000139
20-100	*Dichrostachys cinerea*	0.721563	0.721651 ± 0.000301
	Commiphora angolensis	0.721559	

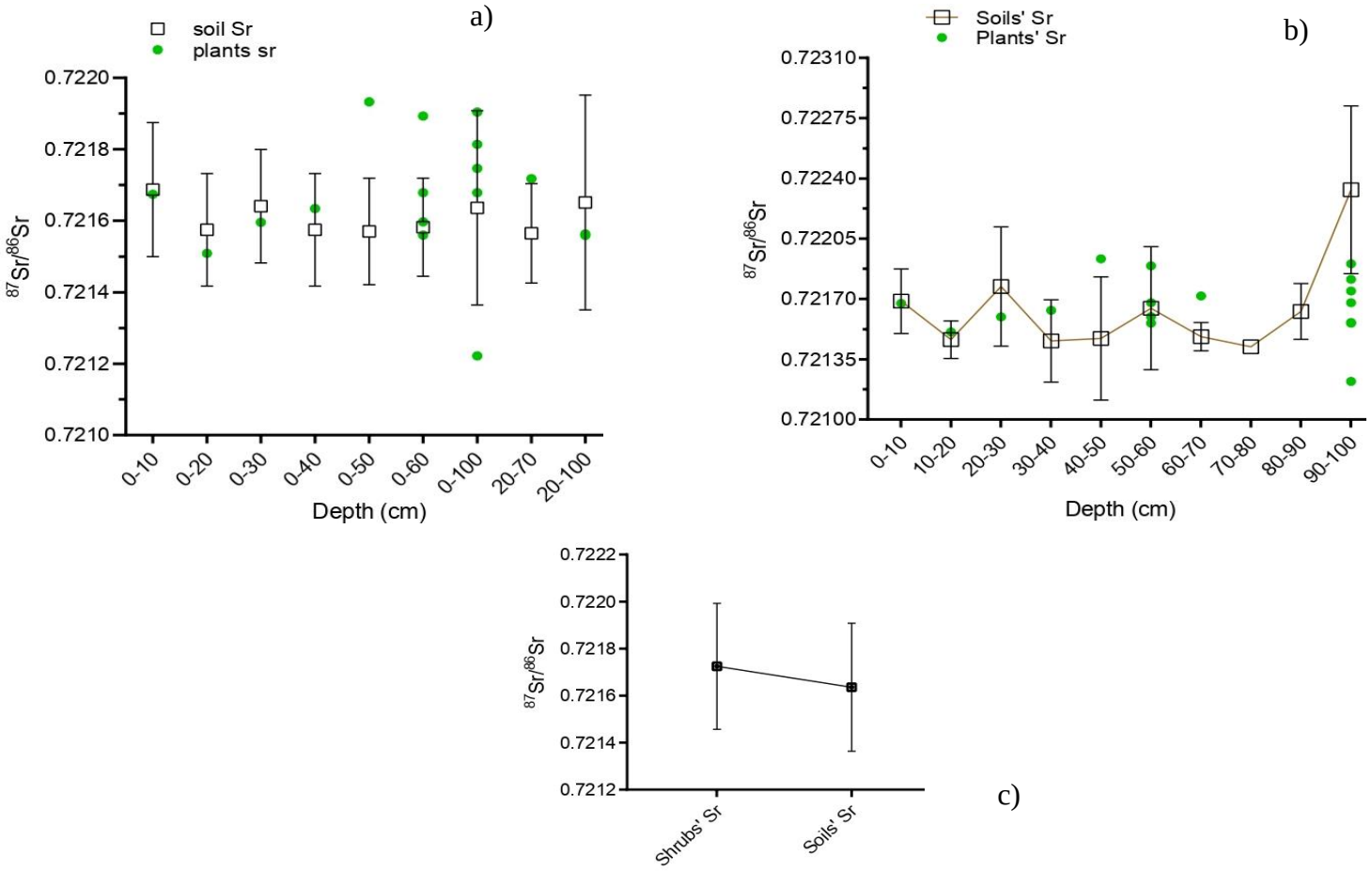


Figure 9. A comparison of shrubs' Sr ratios with the cumulative average of soil Sr ratios ($^{87}Sr/^{86}Sr \pm SD$) of their rooting zones (a). A comparison of shrubs' Sr ratios with the average of soil Sr ratios ($^{87}Sr/^{86}Sr \pm SD$) at a 10 cm depth interval of their corresponding maximum rooting depths (b), while (c) is an overlap between all sampled shrubs' and soil depths' average Sr ratios ($^{87}Sr/^{86}Sr \pm SD$) at the study site.

Furthermore, leaf-soil Sr ratios comparison was done between the four shrubs (*Grewia flava, Dichrostachys cinerea, Senegalia melifera,* and *Terminalia sericea*) whose canopies were sampled for soil Sr with different soil depths of their respective soil cores. Results from this comparison revealed that all four shrubs had Sr ratios that were within the soil average Sr ratios of their respective cores, in terms of their standard deviations (Figure 10b). Two shrubs had Sr ratios closer to the Sr ratios of some specific soil depths of their correspondence cores. These shrubs were *Senegalia melifera* and *Terminalia sericea* of which, *Senegalia melifera* had Sr ratio (0.721747) that closely corresponds with that of depth 20-30 cm (0.721811) of the core under its canopy. *Terminalia sericea* on the other hand had a Sr ratio (0.721596) that nearly corresponds with those of depth 10-20 cm (0.721554) of its corresponding core (Figure 12a). Results also showed that all four shrubs had Sr ratios that were in close range, ranging from 0.721597, 0.721718, 0.721747, and 0.721596, respectively. In contrast, the Sr ratios of the soil profile from the four cores showed a higher variation with the highest being recorded at the depth of 90-100 cm (0.724037) from the *Grewia flava's* core, while the lowest was recorded at the depth of 20-30 cm (0.720904) from the *Dichrostachys cinerea's* core (Figure 10a).

Lastly, the Sr ratios of these shrubs were also compared with the average soil Sr ratios of their correspondence cores, as presented in Figure 10b. Results from this comparison indicated that although none of the shrub's Sr ratio corresponds with any specific soil depth's Sr ratio, they all fell within their respective soil average Sr ratios in terms of their standard deviations. A *Terminalia sericea* had a Sr ratio (0.721596) very close to the average soil Sr ratio (0.721627 ± 0.000455) of its core. The same with *Senegalia mellifera*, whose Sr ratio (0.721747) was somewhat closer to the

average of Sr ratios of its core (0.721656 ± 0.000420). On the other hand, *Grewia flava* (0.721597) and *Dichrostachys cinerea* (0.721718) recorded Sr ratios that were quite far from the average soil Sr ratio of their cores (0.721479 ± 0.000907; 0.721611 ± 0.000448, respectively). However, their Sr ratios were still within the same range in terms of their standard deviation (Figure 10b).

a) b)

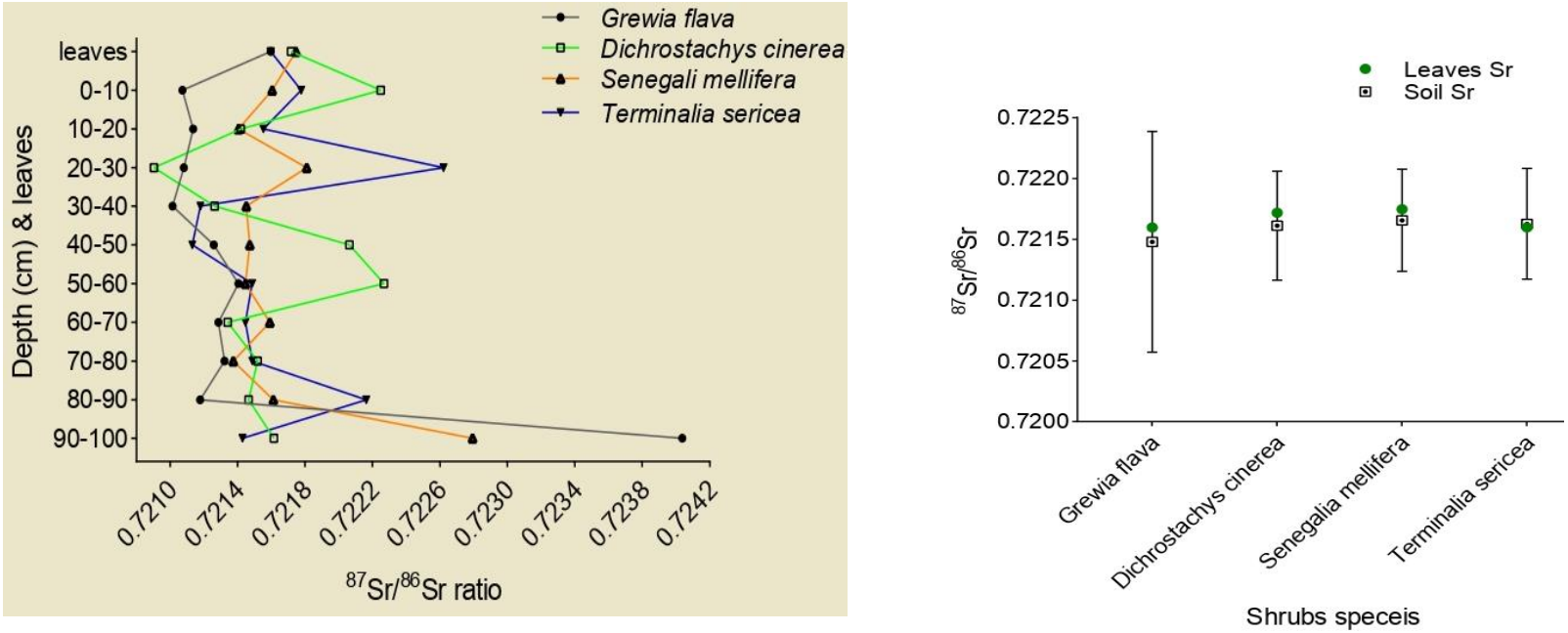


Figure 10: a) Leaf $^{87}Sr/^{86}Sr$ ratios for shrubs whose canopies were sampled for soil Sr analysis, plotted together with the $^{87}Sr/^{86}Sr$ ratios of different soil depths from their respective soil cores. Each line connects values of the same sampling point. While b) is the Leaf $^{87}Sr/^{86}Sr$ ratios of the four shrubs compared with the average Sr ratios ($^{87}Sr/^{86}Sr \pm SD$) of all soil depths of their respective cores.

CHAPTER 5: DISCUSSIONS

The main aim of this study was to assess radiogenic strontium isotopes as a potential method for studying the root deployment of the savanna plants. To set the stage, the study contextualised the belowground structure of savanna plants about their root deployments as well as to determine the distribution of physicochemical properties of the study site. The study revealed that 75% of the root biomass and most of the soil nutrients such as OM, P, N, and K are concentrated in the upper 50 cm of the soil layer. Among these, P, N, and K are classified as macronutrients that are highly cycled by plants (Jobbagy & Jackson, 2001; Paganeli & Batalha, 2022). This signifies a strong correlation in the vertical distribution between the plants' essential nutrients, root biomass, and root density in the soil. The study reported a significant effect of soil depths on vertical root distribution, where root biomass and root density decrease as soil depth increases. This decline of root biomass and root density with increasing soil depth has been associated with the inflow of nutrients returned to the soil via litter fall, canopy leachates, stem flow tropism for organic matter and increasing aluminium (AL) toxicity with increasing soil depth (Cains et al., 1997; Cavelier 1992; Jobbagy & Jackson, 2001).

Similarly, the study revealed that there is a strong correlation between the spatial (horizontal) distribution of root biomass and the distribution of plants, as well as the vegetation density. In other words, there was more root biomass within the plant canopies and less root biomass outside plant canopies. More root biomass within plant canopy is once again attributed to more organic matter inside plant canopies due to litterfall, leading to a fertile soil (Cavelier, 1992; Hipondoka et al., 2003; Hook et al., 1994; Zhou et al., 2009).

Results also indicated that the upper soil layer (0-50 cm) where most of the root biomass, root density, and soil nutrients are concentrated is dominated by a fibrous roots system, extended horizontally up to 5 m. In contrast, dual and tap roots systems have deeper rooting depths. Although most of the roots are concentrated in the upper soil layer, this study reported that the extent of rooting depths and lateral root length are also influenced by the shrub's root systems. For example, those with dual and taproots were found to be deep-rooted with shorter lateral root lengths as they extended their roots vertically into deeper soil depths, unlike fibrous root systems with shallower and longer lateral roots that run parallel with the soil layers. The difference in rooting system and root deployment is also true for shrubs of the same species. For example, the three sampled shrubs of *Ximenia caffra* were found with distinct rooting system and rooting depths: one shrub had fibrous roots with a maximum rooting depth of 0.83 m, another had a taproot that went beyond 1 m; and the remaining had a dual root system whose roots go beyond 1 m.

Those results are comparable with Nakanyala (2020) whose study obtained similar results from a similar environmental setting and asserted that plants develop their root systems plastically due to prevailing environmental conditions. Soil moisture, soil nutrients, and plant hormones are some of these conditions (Cains et al., 1997; Morris et al., 2017; Waidmann et al., 2020; Zhang et al., 2019). For example, phosphate, an organic form of P is highly immobile and is reported to have high a concentration in the topsoil (Morris et al., 2017). Its high concentration in the topsoil increases the foraging for P acquisition and consequently the RSA by increasing the root biomass, root lengths, and shallower root angle (Morris et al., 2017). The distribution of the hormone auxin is reported to vary from plant to plant and mediates responses such as

gravitropism which determines the direction of roots downward (Waidmann, et al., 2020). On the other hand, the higher precipitation in the upper soil layer results in high moisture concentration at such soil layer, and consequently increases topsoil foraging (Zhang et al., 2019). Therefore, these findings signify that the belowground structure of shrubs, specifically the rooting depths is not related to species but rather an individual strategy.

The assessment of the savanna shrubs' root deployments and the depths at which they forage for soil resources using the radiogenic strontium isotope method included determining the Sr ratios of plant leaves and those of soil from different soil depths. Findings relating to this objective signify that the study site has the same level of strontium that is being taken up by the plant roots from the soil profile, with a strong correlation between plants and soils' Sr ratios. For example, results revealed that 95% of sampled shrubs had Sr ratios that were within the overall mean of all the shrubs' Sr ratios. This study also reported that most of these shrubs' Sr ratios are closer to the overall mean of the shrubs' Sr ratios. Additionally, the average Sr ratio of all soil depths was found within the soil mean's Sr ratios. Furthermore, results also revealed that there is a slight variation in Sr ratios at different soil depths, whose variation was not significantly influenced by soil depth (p = 0.44). Consequently, results obtained from the z-score revealed that nearly all the samples (98%) collected for isotopic analysis recorded Sr ratios that are within the site mean. Lastly, the homogenous Sr ratios at the site was proved by the overlap between the average soil depths' Sr ratios and the average shrubs' Sr ratios in terms of their standard deviations.

Adams et al. (2019) and Aguzzoni et al. (2019) reported a strong association between plants and soils' Sr ratios from the same location. Adams et al. (2019) further stated that this correspondence is to confirm that the Sr ratios represent a chemical fingerprint of the growing area. Aguzzoni et al. (2019) suggested that the plant root systems either spread mainly in the same soil layer hence plants absorb Sr from the same nutrient pool, or the Sr isotope ratio in the soil is very homogeneous along the soil profile. This study supports the idea of Aguzzoni et al. (2019) on the homogeneity of soil depths' Sr ratios. However, the study could not conclude whether all shrubs deploy their roots in the same soil layer, thus foraging for soil moisture and nutrients from the same layer, since sampling was abandoned at 1 m depth. Additionally, Chizzali et al. (2021) stated that the landscape of the area also influences the variation of Sr in the soil.

Although results reported a homogeneity from both plant's and soil's Sr ratios, an outlier was recorded from a *Grewia flava*, whose Sr ratio was found outside the average Sr ratios of all plants (a z-score value >3). This shrub was one of the three shrubs whose roots were not exposed for empirical measurements due to their complex root structures; thus, its rooting depths are unknown. It should be noted that besides the higher Sr ratios in this shrub, its z-score value was still found within the average Sr ratio of the entire site.

While plants' isotopic ratios are not associated with plant species, since this study reported different Sr ratios from shrubs of the same shrubs' species, results revealed that the first three shrubs with the highest Sr ratios are of the *Grewia flava* species. Therefore, this study suggests that this could be due to their complex rooting patterns which consist of numerous and longer fibrous roots that are more concentrated in the

middle and upper soil layers and extended in opposite directions. Root morphology and the release of Sr in the atmosphere have been associated with high Sr ratios in some plants (Burger & Lichtscheidl, 2019; Morte & Varma, 2014). For example, the placement of roots determines the maximum amount of resources that could be absorbed, whereby shallow roots take up much strontium where it is concentrated in the upper soil depths (Burger & Lichtscheidl, 2019; Morte & Varma, 2014). Additionally, the release of Sr in the atmosphere which is later retained as a mixture of dry and wet deposition on the plant surface and absorbed by plants leaves (Burger & Lichtscheidl, 2019), could also be a reason for high Sr ratios in these shrubs.

Additionally, although the shrub that recorded the lowest Sr ratio had a rooting depth beyond 1 m, the highest Sr ratio recorded, which was also considered an outlier at the entire site (z-score value of 4.972664) was also obtained from a deeper soil depth (90-100 cm). Nonetheless, Adams et al. (2019) stated that factors such as anthropogenic, meteorological, and geological processes could lead to a divergence of plant and soil Sr ratios from the same location. These findings indicate that the accumulation of Sr ratios in plants and soil is neither a species based, nor it is attributed to their rooting depths, or soil depths.

Furthermore, the soil samples used for isotopic analysis were sampled from inside plant canopies and from outside plant canopies. The results revealed that canopy significantly affects strontium isotopic ratios ($p = 0.05$), whereby higher Sr ratios were recorded from the outside canopy compared to the inside canopy, on average. The study reported that soil samples taken inside plant canopy have higher Sr ratios at greater soil depths and lower at the upper soil depths. Whereas Sr ratios of soil sampled

from the outside canopy are higher at the upper soil depths and lower at the greater soil depths.

The study's last objective was to compare the obtained plant isotopic ratios with different soil depths' isotopic ratios, to determine their correspondence and assess the overall performance of the strontium isotope method. Results from this objective indicated that most shrubs (13) correspond with the soil Sr ratios of their rooting zones' cumulative average and half of the studied shrubs (10) correspond with the soils' Sr ratios recorded at their maximum rooting depths. Results also revealed that three of the sampled shrubs did not correspond with any of the comparisons. Although most shrubs are found within the range of their correspondence soil mean in terms of their standard deviations, some shrubs recorded Sr ratios closer to the mean and others had Sr ratios far from their correspondence mean. The study suggests that the correspondence of plant leaves, and soil depths Sr ratios could be influenced by the extent of individual plants' rooting depths. For example, plants with upper rooting depths would have a better correspondence than those with deeper roots because their roots are not exposed to multiple soil depths, therefore their uptake is only limited to a specific soil depth zone. In this context, *Acacia hereroensis* (with a maximum rooting depth of 10 cm) was reported to have a good correspondence with the Sr ratio of its rooting zone (0-10 cm). The study suggests that this is because this shrub's roots were only exposed to Sr ratios that are available on topsoil. Hence, it does not absorb soil resources from multiple soil depths. The study also suggests that those shrubs with Sr ratios far from the mean could be a result of root uptake from several soil depths, especially those with roots deployed beyond 1 m depth, which the study could not determine the extent of all their maximum rooting depths.

Although not all shrub's Sr ratios matched with soil Sr ratios of their correspondence soil depths, findings revealed that a total of fourteen shrubs from the two comparisons had Sr ratios that corresponded with soil depths' Sr ratios at which their roots were deployed. Results also revealed that between the two comparison approaches that were used, the cumulative average of Sr ratio was a better comparison because the majority of shrubs (13) corresponded better with the soil depths Sr ratios, at which their roots were found. Unlike the maximum rooting depths comparison which only shows ten shrubs that correspond with the average soil Sr ratios at their maximum rooting depth. The study suggests that this was the case because most plants carry Sr ratios of all soil depths at which their roots are deployed, and not only from their maximum rooting depths. Most shrubs with rooting depths beyond 1 m were reported to have Sr ratios far from the overall soil mean. The study also suggests that this is because their roots uptake goes beyond the sampled depth and thus soil Sr ratios of their maximum depths are unknown. However, their Sr ratios were still within the range of the overall mean because their roots started to grow from the topsoil, thus, it is certain that they absorb Sr from all the sampled soil depths.

Those shrubs whose canopy was sampled for soil Sr analysis also show a similar pattern when compared with the soil Sr ratios of their corresponding soil cores. For instance, two shrubs showed a close correspondence with the soil's Sr ratio of some specific soil depths of their cores, compared to the remaining two. One of these shrubs was a *Senegalia mellifera* which was found to have an Sr ratio that nearly corresponds with soil depth of 20-30 cm. The other was a *Terminalia sericea* which had a Sr ratio closer to that of soil depth 10-20 cm from their respective soil cores. The study suggests that these shrubs are likely to forage most of the soil resources at such above-

mentioned soil depths. As for a *Terminalia sericea,* the excavation method revealed that its roots were deployed between 0.5 to 30 cm soil depths. Therefore, it is evident that it indeed forages for soil resources at the upper soil depths thus, had a good correspondence with soil Sr ratio of 10-20 cm depth. However, for *Senegalia melifera,* although it recorded a Sr ratio comparable to that of 20-30 cm, it was found to have a maximum rooting depth that goes beyond 1 m, with roots starting to grow from a depth of 0.5 to 70 cm. Although this shrub may obtain soil resources at different soil depths, especially because it is a dual root system, the study suggests that it may prioritise foraging for soil resources at the upper depths, inclusive of the depth of 20-30 cm, as most of its roots are concentrated at the upper soil layer. According to Schenk and Jackson (2002), shallow roots are generally favoured over deep roots because the energy cost for resource uptake and maintenance is lower for shallow roots. Additionally, shallow soil layers have high nutrient concentrations and sufficient oxygen compared to greater soil depths (Schenk & Jackson, 2002). Overall results indicated that all these four shrubs had Sr ratios that were found within the average soil Sr ratios of their corresponding cores. Two of these shrubs had Sr ratios closer to the mean and the remaining two shrubs had Sr ratios quite distant from the mean, the same correspondences that were also found from the rest of all the sampled shrubs as discussed earlier.

Although most shrubs' Sr ratios were found within the average soil Sr ratios at which their roots were deployed, the study concluded that the radiogenic strontium isotope method could not determine plant root deployment at the level required for ecological studies. The excavation method was used to validate the new method and this study

revealed that strontium isotope was unable to pick up the complexity of savanna shrub roots deployment in the soil profile because of the following four main reasons:

i) Some shrubs with different rooting depths carried the same Sr ratios while those shrubs with the same rooting depths had distinct Sr ratios.

ii) Some shrubs had Sr ratios that nearly corresponded with some soil depths' Sr ratios at which their roots were not deployed, while others had Sr ratios far from the average soil depths' Sr ratios of their rooting zones.

iii) The study also revealed that three shrubs had Sr ratios that did not correspond with the soils' Sr ratios of their corresponding soil depths, in all the two comparisons that were used.

iv) Comparable isotopic ratios along the soil profile were reported in the study area. For example, the study revealed that there was a slight variation between Sr ratios of sampled soil depths with the lowest difference being 0.000001 Sr ratio between some soil depths.

The study suggests that this homogeneity was the possible cause of the challenges on plant leaves and soil depths' Sr ratios correspondence, presented above. Additionally, results also revealed that the variation of strontium isotopic ratio in the soil, at the study site is not significantly influenced by soil depths (p = 0.44). For example, the study recorded lower Sr ratios at both lower and upper soil depths, i.e., the depth of 30-40 cm and 70-80 cm, and higher Sr ratios were recorded at both lower and upper soil depths, i.e., depth 20-30 cm and 90-100 cm. Therefore, shrubs with deeper roots could carry Sr ratios that correspond with soil Sr ratios in the upper soil depths and those with shallower roots may as well carry Sr ratios similar to the deeper soil's Sr ratios, as reported in this study.

Although the Sr ratios obtained are not precise enough to reveal the complexity of the savanna plant rooting depths, their homogeneity makes them accurate in representing the site geographically. For instance, this study recorded the smallest difference of 0.000001 between soil depths Sr ratios, while in some cases, some shrubs recorded similar Sr ratios. Despite the lowest difference obtained, the fact that 98.6% of the samples at the site were found within the average of the site's Sr ratio implies that their Sr ratios have the potential to distinguish the site from other geographic locations. Therefore, the radiogenic strontium isotope method remains relevant in studies with an interest in geographical variations of isotopes such as in archaeology, geology, and geomorphology. In this regard, the plants' average Sr ratio (0.721725 ± 0.000268) from this study differs (with at least 0.0073) from those of Zipkin et al. (2021) who recorded from plant materials with higher Sr ratios of up to 0.7290, in the east of Tsumkwe and lower Sr ratios (0.7144) further north of Tsumkwe. The difference in Sr ratios (0.0073) obtained between the study site and those of Zipkin et al. (2021) makes it adequate to differentiate the area from other locations. For example, several studies for provenance in archaeology such as those of Frank et al. (2021) and Frei (2012), recorded a much lower difference (0.0005) between their sites that are > 200 km apart.

CHAPTER 6: CONCLUSIONS AND RECOMMENDATIONS

Knowledge of the belowground biomass and belowground structure of the savanna provides insights into how plant roots extract soil resources and the mechanism under which they forage for such resources. This study shows that the belowground biomass of the savannas, which includes root biomass and root density decreases with increasing soil depths. More root biomass and root density from this study were found in the first 30 cm of the soil profile and fewer are present at the deeper soil depths. These results have been related to the high distribution of P, N, OM, and OC in the topsoil layer and lower distribution at the greater soil depths. Therefore, the study concluded that the vertical distribution of root biomass and root density in the soil strongly correlated with soil nutrient availability along the soil profile. Plant canopies were also found to influence the spatial distribution of root biomass whereby more roots were found inside plant canopies compared to the outside canopy, due to more organic matter inside plant canopies resulting from litterfall. The concentration of soil nutrients at the study site is generally poor including macronutrients such as P and N. The spatial distribution of root biomass has been associated with plant distribution and vegetation density at the study site.

Despite the higher concentration of root biomass, root density and soil nutrients on the upper soil depths, this study reported a dynamic in the rooting systems and rooting depths of the savanna shrubs which varies among different and same shrub species. Different root system architectures reported from the studied shrubs were fibrous root system, taproot system and dual root system. The fibrous root system was reported to be the most dominant, while taproot was the least dominant at the site. The study concluded that the types of shrubs' root systems and rooting depths are not species-

specific but different shrubs of the same species could have different root systems as well as different rooting depths. The study also concluded that the type of root system has an influence on the shrubs' rooting depths whereby those shrubs with fibrous roots are likely to deploy their roots at the upper soil depths while those with dual and taproot systems have deeper rooting depths.

Results from the strontium isotope analysis revealed that the variation of Sr ratio in the soil was not influenced by soil depths, hence this study recorded a homogenous isotopic signature from both plant leaves and soil depths. In some cases, some shrubs were found to have similar and some very close Sr ratios although there is a high distinction in their rooting depths. On the contrary, those shrubs with similar rooting depths recorded different Sr ratios. Since soil Sr ratio is not highly different at various sampled soil depths, it would thus be challenging to determine different rooting depths of shrubs with Sr ratios that are similar or in close range. Therefore, the study concluded that the variation of Sr ratios in plant leaves is not attributed to plants' rooting depths. Hence with these findings, the strontium isotope method lacks the precision required for determining the roots deployment of savanna shrubs. Nevertheless, the study also suggests that the root system of plants might have an influence on the amount of Sr ratio absorbed by a plant. For example, the *Grewia flava* has a complex fibrous root system which consists of numerous and longer roots and recorded the highest Sr ratios among other shrubs.

Although the radiogenic strontium method is not the best approach to unfold the challenges to study root deployment as presented, the results obtained are accurate enough to represent the study site. This isotopic method is therefore sufficient for the

provenance studies. Evidence was derived from the overlap of the sampled plants' and soil depths' Sr ratios which signifies a homogenous Sr ratio at the study site, thereby representing a fingerprint of their growing area. Additionally, most of both plant and soil samples (98.6%) were found with Sr ratios within the average Sr ratios of the entire study site. Lastly, the differences between the Sr ratios of the study site and those of other studies from different locations in the surroundings make the study's Sr results accurate enough to represent its immediate location.

Since the strontium isotope method could not reveal in detail the belowground structure of savanna plants, the study recommends that future studies should engage in finding a better and innovative approach to unlock the complexity of savanna plant roots, hence understanding the savanna's tree-grass coexistence. The study would also like to recommend that future studies consider a larger sample size that would include more shrub species.

REFERENCES

Achilleos, G. (2008). Errors within the Inverse Distance Weighted (IDW) interpolation procedure. *Geocarto Internation, 23*(6), 429-449. https://doi.org/10.1080/10106040801966704

Adams, S. Grün, R., McGahan, D., Zhao, J., Feng, Y., Nguyen, A., Willmes, M., Quaresimin, M., Lobsey, B., Collard, M., & Westaway, M. C. (2019). A strontium isoscape of north-east Australia for human provenance and repatriation. *Geoarchaeology, 2019*(34), 231-251.

Addo-Danso, S. D., Predcott, C. E., & Smith, A. R. (2015). Methods for estimating root biomass and production in forest and woodland ecosystem carbon studies: A review. *Forest Ecology and Management, 359*(206), 332-351. https://doi.org/10.1016/j.foreco.2015.08.015

Ampofo, E. A. (2006). Soil moisture dynamics in coastal savanna soils in the tropics under different soil management practices. *Hydrological Sciences Journal, 51*(6), 1194-1202. https:s//doi.org/10.1623/hysj.51.6.1194

Aguzzoni, A., Bassi, M., Robatscher, P., Scandellari, F., Tirler, W., & Tagliavini, M. (2019). Intra- and Intertree Variability of the ^{87} Sr/ ^{86} Sr Ratio in Apple Orchards and Its Correlation with the Soil ^{87} Sr/ ^{86} Sr Ratio. *Journal of Agricultural and Food Chemistry*, *67*(20), 5728–5735. https://doi.org/10.1021/acs.jafc.9b01082

Ansley, R. J., Boutton, T. W., & Jacoby, P. W. (2014). Root biomass and distribution patterns in a semi-arid Mesquite savanna: Responses to long-term rainfall manipulation. *Rangeland Ecology & Management, 67*(2), 206-218. https://doi.org/10.2111/REM-D-13-00119.1

Alaoui, I., el ghadraoui, O., Serbouti, S., ahmed, H., Mansouri, I., Kamari, F. E., Taroq, A., Ousaaid, D., Squalli1, W., & Farah, A. (2022). The Mechanisms of Absorption and Nutrients Transport in Plants: A Review. *Tropical Journal of Natural Product Research*, *6*(1), 8–14. https://doi.org/10.26538/tjnpr/v6i1.2

Araya, Y. N. (2007). Ecology of Water Relations in Plants. In John Wiley & Sons, Ltd (Ed.), *Encyclopaedia of life sciences* (1st ed.). Wiley. https://doi.org/10.1002/9780470015902.a0003201

Atlas of Namibia Team. (2022). Atlas of Namibia. *Its land, water and life.* https://atlasofnamibia.online/chapter-3

Axelsson, R. C. (2018). *A study of African savanna vegetation structure, patterning, and change.* (Doctoral dissertation, South Dakota State University). https://openprairie.sdstate.edu/etd/2639

Baudena, M., Dekker, S. C., van Bodegom, P. M., Cuesta, B., Higgins, S. I., Lehsten, V., Reick, C. H., Rietkerk, M., Scheiter, S., Yin, Z., Zavala, M. A., & Brovkin, V. (2015). Forests, savannas, and grasslands: bridging the knowledge gap between ecology and dynamic global vegetation models. *Biogeoscience, 12*(6), 1833-1848. https://doi.org/105194/bg-12-1833-2015

Bayon, G., Freslon, N., Germain, Y., Bindeman, I., Trinquier, A., & Barrat, J.-A. (2021). A global survey of radiogenic strontium isotopes in river sediments. *Chemical Geology*, *559*(43), Article e119958. https://doi.org/10.1016/j.chemgeo.2020.119958

Becker, P., & Castillo, A. (1990). Root architecture of shrubs and saplings in the understory of a tropical moist forest in lowland Panama. *Biotropica*, *22*(3), 242-249. https://www.jstor.org/stable/2388534

Bentley, R. A., Price, T. D., Lning J., Gronenborn, D., Wahl, J., & Fullagar, P. (2002). Prehistoric Migration in Europe: strontium isotope analysis of early neolithic skeletons. *Current Anthropology, 43*(5), 799-804. https://www.jstor.org/stable/10.1086/34437

Bijak, S., Zasada, M., Bronisz, A., Bronisz, K., Czajkowski, M., Ludwisiak, L. Tomusiak, R., & Wojtan, R. (2013). Estimating coarse roots biomass in young silver birch stands on post-agricultural lands in central Poland. *Silva Fennica, 47*(2), Article id963. http://www.silvafennica.fi/article/963

Birch, C., Middleton, A., Harper-Simmonds, L., & Lindeque, P. (2017). *Economics of Land Degradation Initiative: Benefits of bush control in Namibia*. Report for the Economics of Land Degradation Initiative. www.eld-initiave.org

Blum, J.D., & Erel, Y. (2003). Radiogenic Isotopes in weathering and hydrology. *Treatise on Geochemistry, 5*, 365-392. https://doi.org/10.1016/B0-08-043751-6/05082-9

Böhm, W. (1979). Methods of studying root systems. *Ecological studies, 33*,20-38. https://doi.org/10.1007/978-3-642-67282-8-4

Burger, A., & Lichtscheidl, I. (2019). Strontium in the environment: Review about reactions of plants towards stable and radioactive strontium isotopes. *Science of The Total Environment, 653*, 1458–1512. https://doi.org/10.1016/j.scitotenv.2018.10.312

Cairns, M. A., Brown, S., Helmer, E., Baumgardner, G. A (1997). Root biomass allocation in the world's upland forests. *Oecologia, 111*(1), 1-11. https://www.jstor.org/stable.4221653

Carlsson, M. (2005). *Vegetation succession in savanna determined by interaction of grazing, browsing, and fire; a comparison between hypotheses.* [Master thesis,

Institutionen för Livsvetenskaper.] Semantic Scholar. https://api.semanticscholar.org/CorpusID:111382904

Castañeda V., Peña, M., Azcárate, L., Aranjuelo, I., & Gonzalez, E. M. (2019). Functional analysis of the taproot and fibrous roots of *Medicago truncatula:* Sucrose and proline catabolism primary response to water deficit. *Agriculture Water Management*, *216*, 473-483. https://doi.org/10.1016/j.agwat.2018.07.018

Cavelier, J. (1992). Fine-root biomass and soil properties in a semideciduous and a lower montane rain forest in Panama. *Plant and soil*, *142*(2), 187-201. https://www.jstor.otg/stable/42938029

Chang, C., Beckford, H. O., & Ji, H. (2022). Indication of Sr isotopes on weathering process of carbonate rocks in karst area of southwest China. *Sustainability*, *14*(8), Article e4822. https://doi.org/10.3390/su14084822

Chen, W., Koide, R. T., & Eissenstat, D. M. (2018). Root morphology and mycorrhizal type strongly influence root production in nutrient hot spots of mixed forests. *Journal of Ecology*, *106*(1), 148–156. https://doi.org/10.1111/1365-2745.12800

Chizzali, S., Aguzzoni, A., Pignotti, E., Zelger, J., Voto, G., Zignale, P., Tagliavini, M., Tirler, W., & Robatscher, P. (2021). Sr isotope ratio in vegetable crops and apple trees depends on that of the soil environment while is unaffected by the genotype. *Italus Hortus*, *28*(3), 49-58. https://doi.org/10.26353/j.itahort/2021.3.4958

Coble, A. A., Hart, C. S., Ketterer, E.M., Newman, S. G., & Kowler, L. A. (2015). Strontium source and depth of uptake shifts with substrate age in semiarid

ecosystems. *Journal of Geophysical Research: Biogeosciences, 120*(6), 1069-1077. https://doi.org/10.1002/2015JG002992

Daly, K. R., Tracy, S.R., Crout, N. M. J., Mairhofer, S., Pridmore, T. P., Mooney, S. J., & Roose, T. (2017). Quantification of root water uptake in soil using X-ray computed tomography and image-based modelling. *Plant, Cell & Environment, 41*(1), 121-133.

Dannoura, M., Kominami, Y., Oguma, H., & Kanazawa, Y. (2008). The development of an optical scanner method for observation of plant root dynamics. *Plant Root, 2*, 14–18. https://doi.org/10.3117/plantroot.2.14

de Klerk, J. N. (2004). *Bush encroachment in Namibia: Report on phase 1 of the Bush Encroachment Research, Monitoring, and Management Project*. Ministry of Environment and Tourism, Windhoek.

Devine, A. P., McDonald, R. A., Quaife, T., & Maclean, I. M. D. (2017). Determinants of woody encroachment and cover in African savannas. *Oecologia, 183*, 939-951. doi.10.1007/s00442-01-3807-6

Doudill, A. J., Heathwaite, A. L., & Thomas, D. S. G. (1998). Soil water movement and nutrient cycling in semi-arid rangeland: vegetation change and system resilience. *Hydrological Processes, 12*(3), 443-495.

Dougill, A. J., & Thomas, A. D. (2004). Kalahari sand soils: Spatial heterogeneity, biological soil crusts and land degradation. *Land Degradation and Development, 15*(3), 233-242. https://doi.org/10.1002/ldr.611

Doussan, C., Pagès, L., & Pierret, A. (2003). Soil exploration and resource acquisition by plant roots: an architectural and modelling point of view. *Agronomie, 23*(5–6), 419–431. https://doi.org/10.1051/agro:2003027

Dubchak, S. (2018). Distribution of strontium in soil: Interception, weathering, speciation, and translocation to plants. In D. Gupta & C, Walther (Eds.), *Behaviour of strontium in plants and the environment* (pp. 33-43). Springer International Publishing. https://doi.org/10.1007/978-3-319-66574-0

Ennos, R. a., & Fitter, H. A. (1997). Comparative functional Morphology of the Anchorage Systems of Annual Dicots. *British Ecological Society, 6(*1), 71-78. https://www.jstor.org/stable/2389773

Ferrara, C., Barone, P. M., Salvati, L., & Pettinelli, E. (2014). Ground penetrating radar as remote sensing technique to investigate the root system architecture. *Applied ecology and environmental research, 12*(3), 695-702.

Frank, A. B., Frei, R., Moutafi, I., Voutsaki, S., Orgeolet, R., Kristiansen, K., & Frei, K. M. (2021). The geographic distribution of bioavailable strontium isotopes in Greece – A base for provenance studies in archaeology. *Science of The Total Environment, 791*, Article e148156. https://doi.org/10.1016/j.scitotenv.2021.148156

Frei, K. M. (2012). Exploring the potential of the strontium isotope tracing system in Denmark. *Danish Journal of Archaeology*, *1*(2), 113–122. https://doi.org/10.1080/21662282.2012.760889

Freschet, G. T., Pagès, L. L., Iversen, C., Comas, L. H., Rewald, B., Roumet, C., Klimešová, J., Zadworny, M., Poorter, H., Postma, J. A., Adams, T. S., Bagniewska-Zadworna, A., Bengough, A. G., Blancaflor, E. B., Brunner, I., Cornelissen, J. H. C., Garnier, E., Gessler, A., Hobbie, S. E., … Batterman, S. A. (2020). *A starting guide to root ecology: Strengthening ecological concepts and standardizing root classification, sampling, processing and trait measurements*. https://hal.science/hal-02918834

Fullman, T. J., & Bunting, E. L. (2014). Analyzing vegetation change in an elephant-impacted landscape using the moving standard deviation index. *Land, 3*(1), 74-104. https://doi.org/10.3390/land3010074

Gillson, L. (2004). Testing non-equilibrium theories in savannas: 1400 years of vegetation change in Tsavo Nation Park, Kenya. *Elsevier*, *1*(4), 281-298.

Gluszek, S., Paszt, L. S., Sumorok, B., Derkowska, E., & Kozera, R. (2013). Application of the minirhizotron technique to studying the roots of fruit plants. *Advances in Science and Technology*, *7*(18), 45–53. https://doi.org/10.5604/2080404075.1049605

Goudie, A. S., Vile, H. A. (2015). *Landscapes and Landforms of Namibia*. Springer Netherlands. https://doi.org10.1007/978-94-017-8020-9 21

Gregory, P. J. (2006). Roots, rhizosphere and soil: the route to better understanding of soil science? *European Journal of Soil Science*, *57*(1), 2-12. https://doi.org/ 10.1111/j.1365-2389.2005.00778.x

Gupta, D. K., & Walther, C., & Chatterjee, S. (2018). Distribution in the ecosystem: transfer in plants via root system. In D. Gupta & C, Walther (Eds.), *Behaviour of strontium in plants and the environment* (pp. 1-18). Springer International Publishing. https://doi.org/10.1007/978-3-319-66574-0

Hauwanga, W. N. (2018). *Vegetation-environment relations along an aridity gradient on kalahari sands in central Namibia*. [Master's thesis, Namibia University of Science and Technology]. Ounongo Repository. http://ir.nust.na:8080/jspui/handle/10628/604

Hipondoka, M. H. T., Aranibar, J. N., Chirara, C., Lihavha, M., & Macko, S. A. (2003). Vertical distribution of grass and tree roots in arid ecosystems of Southern

Africa: Niche differentiation or competition? *Journal Arid of Environments*, *54*(2), 319–411.

Hipondoka, M., & Versfeld, W. (2006). Root system of Terminalia sericea shrubs across rainfall gradient in a semi-arid environment of Etosha National Park, Namibia. *Ecological Indicators*, *6*(3), 516–524. https://doi.org/10.1016/j.ecolind.2005.07.004

Hodge, A., Berta, G., Doussan, C., Merchan, F., & Crespi, M. (2009). Plant root growth, architecture and function. *Plant and soil*, *321(*1-2), 153-187.

Höllriegl, V., & Müchen, H. Z. (2011). Strontium in the Environment and possible human health. In J. O. Nriagu (Ed.), *Encylopedia of Environmental Health, Elsevier,* (pp. 268-275). University of Michigan, United States. https://doi.org/10.1016/B978-0-12-409548-9.11195

Holdo, R. M., & Mack, M. (2014). Functional attributes of savanna soils: contrasting effects of tree canopies and herbivores on bulk density, nutrients and moisture dynamics. *Journal of ecology*, *102*(5), 1171-1182. https://doi.org/10.1111/1365-2745.12290

Holdo, R. M., & Nippert, J. B. (2015). Transpiration dynamics support resource partitioning in African savanna trees and grasses. *Ecology*, *96*(6), 1466–1472. https://doi.org/10.1890/14-1986.1

Hook,P. B., Lauenroth, W. K., & Burke, I. C. (1994). Spatial patterns of roots in a semiarid grassland: abundance of canopy openings and regeneration gaps. *British Ecological Society, 82*(3), 485-494.

Hüttich, C., Gessner, U., Herold, M., Strohbach, B. J., Schmidt, M., Keil, M., & Dech, S. (2009). On the Suitability of MODIS Time Series Metricts to Map Vegetation Types in Dry Savanna Ecosystems: A Case Study in the Kalahari

of NE Namibia. *Remote Sensing*, *1*(4), 620–643. https://doi.org/10.3390/rs1040620

Jaloviar, P., Bakošová, L., Kucbel, S, Vencurik, J. (2009). Quantity and distribution of fine root biomass in the intermediate stage of beech virgin forest Badínsky prales. J*ournal of Forest Science, 55*(11), 502-510.

Jeltsch, F., Weber, G. E., & Grimm, V. (2000). Ecological buffering mechanisms in savannas: A unifying theory of long-term tree-grass coexistence. *Plant Ecology, 150*(1-2), 161-171.

Jobbagy, E. G., & Jackson, R, B. (2001). The distribution of soil nutrients with depth: Global patterns and the imprint of plants. *Biogeochemistry*. *53*, 51-77. https://doi.org/10.1023/A:1010760720215

Kgabi, N., Uugwanga, M., & Ithindi J. (2016). Atmospheric conditions and precipitation in arid environments: A case of Namibia. *International Journal of Environmental Protection, 6*(1), 148-159.

Katjiua, M., & Ward, D. (2012). Soil and vegetation changes under livestock production in the northern Kalahari, Namibia. *DINTERIA*, *32*, 52-69.

Kerk, N. M., & Sussex, I. M. (2001). Roots and Root Systems. In John Wiley & Sons, Ltd (Ed.), *ELS* (1st ed.). Wiley. https://doi.org/10.1038/npg.els.0002058

Kneitel, J. (2013). Gause's Competitive Exclusion Principle. In S. A. Elias (ed.), *Reference Module in Earth Systems and Environmental Sciences* (pp. 20-24). Elsevier. doi:10.1016/B978-012-409548-9.00816-2

Koffi, K. F., N'Dri, A. B., Srikanthasamy, T., Lata, J-C., Konaté, S. Konan, M., & Barot, S. (2022). The demographic response of grass species to fire treatments in a Guinean savanna. *Fire, 5*(6), Article 193. https://doi.org/10.3390/fire5060193

Konare´, S., Boudsocq, S., Gignoux, J., Lata, J., Raynaud, X., & Barot, S. (2019). Effects of mineral nitrogen partitioning on tree-grass coexistence in west African savannas. *Ecosystems*, *22*(7), 1676-1690. https://doi.org/10.1007/s10021-019-00365-x

Kong, X., Zhang, M., Smet, D. I., & Ding, Z. (2014). Designer crops: optimal root system architecture for nutrients acquisition. *Trends in Biotechnology*, *32*(12), 597-598. https://doi.org/10.1016/j.tibtech.2014.09.008

Kulmatiski, A., Beard, K. H., Holdrege, M. C., & February, E. C. (2020). Small differences in root distributions allow resource niche partitioning. *Ecology and Evolution*, *10*(18), 9776–9787. https://doi.org/10.1002/ece3.6612

Kulmatiski, A., Sprouse S, R. C., & Bead, K. H. (2017). Soil type more than precipitation determines fine-root abundance in savannas of Kruger National Park, South Africa. *Plant and Soil*, *417*(1-2), 523-533. https://doi.org/10.1007/s11104-017-3277-y

Kücke, M., Schmid, H., & Spiess (1995). A comparison of four methods for measuring roots of field crops in three contrasting soils. *Plant and soil 172*(1), 63-71. https://www.jstor.org/stable/42947463

Laughlin, D. C., Mommer, L., & Alexandra, W. (2021). Root traits explain plant species distributions along climatic gradients yet challenge the nature of ecological trade-offs. *Nature Ecology & Evolution*, *5*(8), 1123- 1134. https://doi.org/10.1038/s41559-021-01471-7

Leuschner, C., Moser, G., Bertsch, C., Röderstein, M., & Hertel, D. (2007). Large altitudinal increase in tree root/shoot ratio in tropical mountain forests of Ecuador. *Basic and Applied Ecology*, *8*(3), 219-230.

Lewis, J., Pike, A. W. G., Coath, C. D., & Evershed, R. P. (2017). Strontium concentration, radiogenic (^{87} Sr/ ^{86} Sr) and stable (δ ^{88} Sr) strontium isotope systematics in a controlled feeding study. *STAR: Science & Technology of Archaeological Research*, *3*(1), 45–57. https://doi.org/10.1080/20548923.2017.1303124

Liu, X., Dong, X., & Leskovar, D. I. (2016). Ground penetrating radar for underground sensing in agriculture. *International Agrophysics*, *30*(4), 533–543. https://doi.org/10.1515/intag-2016-0010

Lynch, J. (1995). Root architecture and plant productivity. *Plant Physiol, 109*(1), 7-13. https://doi.org/10.1104/pp.109.1.7

Macinnis-Ng, C.M.O., Fuentes, S., O'Grady, A. P., Palmer, A. R., Taylor, D., Whitley, R.J., Yunusa, I., Zeppel, J. B., & Eamus, D. (2009). Root biomass distribution and soil properties of an open woodland on a duplex soil. *Plant and soil, 327*(1), 377-388. https://doi.org/10.1007/s11104-009-0061-7

Maeght, J.-L., Rewald, B., & Pierret, A. (2013). How to study deep roots- and why it matters. *Frontiers in Pant Science*, *4*, Article e299. https://doi.org/10.3389/fpls.2013.00299

Makkonen, K., & Helmisaari, H.-S. (1999). Assessing fine-root biomass and production in a Scots pine stand – comparison of soil core and root ingrowth core methods. *Plant and Soil, 210*(1), 43-50. http://www.jstor.org/stable/42949606

Martínez, F., Merino, O, Martin, A, Martin, D. G, Merino, J. (1998). Belowground structure and production in a Mediterranean sand dune shrub community. *Plant and soil, 201*(2), 209-216.

Matimati, I., Verboom, G. A., & Cramer, M. D. (2014). Nitrogen regulation of transpiration controls mass-flow acquisition of nutrients. *Journal of Experimental Botany*, *65*(1), 159-168. https://doi.org/10.1093/jxb/ert367

McCulley, R. L., Jobbagy, E. G., & Pockman, W. T. (2004). Nutrient uptake as a contributing explanation for deep rooting in arid and semi-arid ecosystems. *Oecologia*, *141*(4), 620–628. https://doi.org/10.1007/s00442-004-1687-z

Mendelsohn, J., & Obeid, S. E. (2002). *The Communal Lands in Eastern Namibia*. Research and Information Services of Namibia. Windhoek, Namibia. http://theeis.com/elibrary/sites/default/files/downloads/literature/Communal%20Lands%20in%20Eastern%20Namibia.pdf

Mickovski, B. S. (2002) *Anchorage mechanics of different types of roots system. Ecology, environment.* [Doctoral dessertation, University of Manchester]. HAL open science. https://theses.hal.science/tel-00003454

Millikin, C. S., & Bledsoe, C. S. (1999). Biomass and distribution of fine and coarse roots from blue oak *(Quercus douglasii)* trees in the northern Sierra Nevada foothills of California. *Plant and Soil*, *214*(1/2), 27-38. https://www.jstor.org/stable/42950535

Mohamed, A., Monnier, Y., Mao, Z., Lobet, G., Maeght, J.-L., Ramel, M., & Stokes, A. (2017). An evaluation of inexpensive methods for root image acquisition when using rhizotrons. *Plant Methods*, *13*(1), article 11. https://doi.org/10.1186/s13007-017-0160-z

Morte, A., & Varma, A. (Eds.). (2014). *Root Engineering: Basic and Applied Concepts*. Springer Berlin Heidelberg. https://doi.org/10.1007/978-3-642-54276-3

Morris E. C., Griffiths, M., Golebiowska, A., Mairhofer, S., Burr-Hersey, J., Goh, T., von Wangenheim, D., Atkinson, B., Sturrock, C. J., Lynch, J. P., Vissenberg, K., Ritz, K., Wells, D. M., Mooney, S. J., & Bennett, M. J. (2017). Shaping 3D root system architecture. *Current Biology, 27*(11), R919-R930. https://doi.org/10.1016/j.cub.2017.06.043

Nakano, T., Tanaka, T., Tsujimura, M., & Matsutani, J. (1993). *Strontium isotopes in soil-plant-atmosphere continuum (SPAC).*

Nakanyala, J. (2020). *Rooting strategies of savanna shrubs in the Kalahari basin: Implications for the coexistence of woody and herbaceous plants and shrub encroachment in the African savannas.* [Doctoral dissertation, University of Namibia]. UNAM Repository. https://repository.unam.edu.na

Nakanyala, J., Kosmas, S., & Hipondoka, M. (2017). The Savannas: An integrated synthesis of three major competing paradigms. *International Science and Technology Journal of Namibia, 10*(1), 108-121.

Neumann, G., George, T. S., & Plassard, C. (2009). Strategies and methods for studying the rhizosphere-The plant science toolbox. *Plant and Soil*, *32*(1), 431–456. https://doi.org/DOI 10.1007/s11104-009-9953-9

Nippert, J. B., & Holdo, R. M. (2015). Challenging the maximum rooting depth paradigm in grasslands and savannas. *Functional Ecology, 29*(6), 739-745.

Noguchi, H., Suwa, R., Souza, C. A. S. D., Silva, R. P. D., Santos, J. D., Higuchi, N., Kajimoto, T., & Ishizuka, M. (2014). Examination of Vertical Distribution of Fine Root Biomass in a Tropical Moist Forest of the Central Amazon, Brazil. *Japan Agricultural Research Quarterly: JARQ*, *48*(2), 231–235. https://doi.org/10.6090/jarq.48.231

O'Donnell, F. C., Caylor, K. K., Bhattachan, A., Dintwe, K., D'Odorico, P., & Okin, G. S. (2015). A quantitative description of the interspecies diversity of belowground structure in savanna woody plants. *Ecosphere, 6*(9), 1-15. https://doi.org/10.1890/ES14-00310.1

Oyewole, O. A., Inselsbacher, E., & Näsholm, T. (2014). Direct estimation of mass flow and diffusion of nitrogen compounds in solution and soil. *New Phytologist*, *201*(3), 1056–1064. https://doi.org/10.1111/nph.12553

Paganeli, B., & Batalha, M. A. (2022). Effects of nitrogen and phosphorus availability on the early growth of two congeneric pairs of savanna and forest species. *Brazilian Journal of Biology*, *82*(4), Article e235573. https://doi.org/10.1590/1519-6984.235573

Pang, Y., Tian, J., Yang, H., Zhang, K., & Wang, D. (2022). Response of fine roots at different soil depths to different thinning intensities in a secondary forest in the Qinling mountains, China. *Biology*, *11*(3), Article 351. https://doi.org/10.3390/biology11030351

Pearce, C. R., Parkinson, I. J., Gaillardet, J., Charlier, B. L. A., Mokadem, F., & Burton, K. W. (2015). Reassessing the stable ($\delta^{88/86}$Sr) and radiogenic (^{87}Sr/^{86}Sr) strontium isotopic composition of marine inputs. *Geochimica et Cosmochimica Acta*, *157*, 125-146. https://doi.org/10.1016/j.gca.2015.02.029

Poszwa, A., Ferry, B.,Pollie, B., Grimaldi, C., Charles-Dominique, P., Loubet, M., & Dambrine, E. (2009). Variations of plant and soil 87Sr/86Sr along the slope of a tropical inselberg. *Annals of Forest Science, 66*(5), 1-13. doi:10.1051/forest/2009036

Raherison, S. M., & Grouzis, M. (2005). Plant Biomass, Nutrient Concentration and Nutrient Storage in a Tropical Dry Forest in the South–west of Madagascar. *Plant Ecology*, *180*(1), 33–45. https://doi.org/10.1007/s11258-005-8063-x

Ravindranath, N. H., & Ostwald, M. (2008). Methods for Below-Ground Biomass. In M. Beniston (Ed.), *Carbon Inventory Methods Handbook for Greenhouse Gas Inventory, Carbon Mitigation and Roundwood Production Projects* (pp.149–156). Springer Netherlands. https://doi.org/10.1007/978-1-4020-6547-7_11

Rau, B. M., Johnson, D. W., Chambers, J. C., Blank, R. R., & Lucchesi, A. (2009). Estimating roots biomass and distribution after fire in a great basin woodland using cores and pits. *Western North American Naturalist, 69*(4), 459–468.

Raz-Yaseef, N., Koteen, L., & Baldocchi, D. D. (2013). Coarse root distribution of a semi-arid oak savanna estimated with ground penetrating radar. *Journal of Geophysical Research: Biogeosciences*, *118*(1), 135–147. https://doi.org/10.1029/2012JG002160

Reynolds, A. C., Quade, J., & Betancourt, J. L. (2012). Strontium isotopes and nutrient sourcing in a semi-arid woodland. *Geoderma*, *189*, 574–584. https://doi.org/10.1016/j.geoderma.2012.06.029

Rodríguez-Iturbe, I., & D'Odorico, P. (1999). Tree-grass coexistence in savannas: The role of spatial dynamics and climate fluctuations. *Geophysical Research Letter, 26*(2), 247-250.

Rogers, E. D., Monaenkova, D., Mijar, M., Nori, A., Goldman, D. I., & Benfey, P. N. (2016). X-Ray Computed Tomography Reveals the Response of Root System Architecture to Soil Texture. *Plant Physiology*, *171*(3), 2028–2040. https://doi.org/10.1104/pp.16.00397

Rogers, E. D., & Benfey, P. N. (2015). Regulation of plant root system architecture: implications for crop advancement. *Current Opinion in Biotechnology, 32*, 93-98. https://doi.org/10.1016/j.copbio.2014.11.015

Ringrose, S., Matheson, W., & Vanderspost, C. (1998). Analysis of soil organic carbon and vegetation cover trends along the Botswana Kalahari Transect. *Journal of Arid Environments*, 38, 379-396.

Rutherford, M.C., Mucina, L., Lötter, M.C., Bredenkamp, G. J., Smit, J. H. L., Scott-shaw, R., Hoare, D. B., Goodman, P. S., Bezuidenhout, H., Scott, L., Ellis, F., Powrie, L. W., Siebert, F., Mostert, T. H., Henning, B. J., Venter, C. E., Camp, K. G. T., Siebert, S. J., Matthews, W. S., … Hurter, J.H. (2006). Savanna Biome. *Trelitzia, 19*, 439-539.

Sankaran, M., Ratnam, J., & Hanan, N. P. (2004). Tree–grass coexistence in savannas revisited – insights from an examination of assumptions and mechanisms invoked in existing models. *Ecology Letters*, *7*(6), 480–490. https://doi.org/10.1111/j.1461-0248.2004.00596.x

Satomura, T., Fukuzawa, K., & Horikoshi, T. (2007). Considerations in the study of tree fine-root turnover with minirhizotrons. *Plant Root*, *1*(1), 34-35. https://doi.org/10.3117/plantroot.1.34

Schenk, H. J., & Jackson, R. B. (2002). Rooting depths, lateral root spreads and below-ground/above-ground allometries of plants in water-limited ecosystems. *Journal of Ecology*, *90*(3), 480–494. https://doi.org/10.1046/j.1365-2745.2002.00682.x

Scholes, R. J., Archer, S. R. (1997). Tree-grass interactions in Savannas. *Annual Reviews of Ecological Systematics*, *28*, 517-544. https://www.researchgate.net/publication/30509618

Schuurman, J. J., & Goedewaagen, M. A. (1971). *Methods for the examination of root systems and roots*. Centre for Agricultural Publishing and Documentation, Wageningen.

Schwarcz, H. P., White, C. D., & Longstaffe, F. J. (2010). Stable and radiogenic isotopes in biological archaeology: some applications. In J.B. West, G. J. Bowen, T. E. Dawson, & K.P. Tu, (Eds.), *Isoscapes: Understanding movement, pattern, and process on Earth through isotope mapping* (pp. 335-356). Springer Dordrecht Heidelberg London New York. https://doi.org/10.1007/978-90-481-3354-3

Setianto, A., & Triandini, T. (2015). Comparison of kriging and inverse distance weighted (IDW) interpolation methods in lineament extraction and analysis. *Journal of Applied Geology*, *5*(1), 21–29. https://doi.org/10.22146/jag.7204

Sheuyange, A., Oba, G., & Weladji, R. B (2004). Effects of Anthropogenic Fire History on Savanna Vegetation in Northeastern Namibia. *Journal of Environmental Management*, *75*(3), 189-198. http://dx.doi.org/10.1016/j.jenvman.2004.11.004

Slovak, N. M., Paytan, A., Rick, J. W., & Chien, C. (2018). Establishing radiogenic strontium isotope signatures for Chavin de Huàntar Peru. *Journal of Archaeological Science: Reports*, *19*, 411-419. https://doi.org/10.1016/j.jasrep.2018.03.014

Smit, G. N., & Rethman, N. F. G. (1997). Root biomass, depth distribution and relations with leaf biomass of *Colophospermum mopane*, *64*(1), 38–43.

Staal, A., Dekker, S. C., Xu, C., & van Nes, E, H. (2016). Bistability, spatial interaction, and the distribution of tropical forests and savannas. *Ecosystems*, *19*(6), 1080-1091. https://doi.org/10.1007/s10021-016-0011-1

Staver, A. C., Botha, J., & Hedin, L. (2017). Soils and fire jointly determine vegetation structure in an African Savanna. *New Phytologist, 216*(4), 1151-1160. https://doi.org/10.1111/nph.14738

Steingrobe, B., Schmid, H., & Claassen, N., (2000). The use of the ingrowth core method for measuring root production of arable crops – influence of soil conditions inside the ingrowth core on root growth. *Journal of Plant Nutrition and Soil Science, 163*(6), 617-622. https://doi.org/10.1002/1522-2624(200012)163:63.0.CO;2-0

Strohbach, B. J., & Kutuahuripa, J. T (2014). Vegetation of the eastern communal conservancies in Namibia: II. Environmental drivers. *Koedoe 56*(1), 1-12. http://dx.doi.org/10.4102/koedoe.v56i1.1117

Taylor, H. M., Upchurch, D. R., & McMichael, B. L. (1990). Applications and limitations of rhizotrons and minirhizotrons for root studies. *Plant and Soil, 129*(1), 29–35. https://www.jstor.org/stable/4293911

Teramoto, S., Kitomi, Y., Nishijima, R., Takayasu R.,Maruyama, N., & Uga Y. (2019). Backhoe-assisted monolith method for plant root phenotyping under upland conditions. *Breeding Science, 69*(3), 508-513. https://doi.org/10.1270/jsbbs.19019

Teramoto S., Takayasu, S., Kitomi, Y., Aral-Sanoh, Y., Tanabata, T., & Uga, Y. (2020). High-throughput three-dimensional visualization of root system architecture of rice using X-ray computed tomography. *Plant Methods,16,* 66. https://doi.org/10.1186/s13007-020-000612-6

Thomas, D. S. G., & Shaw, P. A. (1991). *The Kalahari environment. C*ambridge University Press.

http://theeis.com/elibrary/sites/default/files/downloads/literature/The_Kalahari_Environment.pdf

Valeix, M., Fritz, H., Sabatier, R., Murindagomo, F., Cumming, D., & Duncan, P. (2010) Elephant-induced structural changes in the vegetation and habitat selection by large herbivores in an African savanna. *Biological Conservation, 144*(2011), 902-912.

Vallero, D. (2014). *Fundamentals of Air Pollution.* (5th ed.). Academic Press. https://doi.org/10.1016/C2012-0-01172-6

van der Waal, C. (2010). *Nutrients in an African savanna: The consequences of supply heterogeneity for plants and animals.* [Doctoral thesis, Wageningen University]. https://core.ac.uk/download/pdf/29242724.pdf

Vetter, S. (2005). Rangelands at equilibrium and non-equilibrium: recent developments in the debate. *Journal of Arid Environments*, *62(2),* 321-341. https://doi.org/10.1016/j.jaridenv.2004.11.015

Veresoglou, D. S., Barbayiannis, N., Matsi, T., Anagnostopoulos, C., & Zalidis, G. C. (1996). Shoot Sr concetrations in relation to shoot Ca concetration and to soil properties. *Plant and soil, 178(*1), 95-100. http://www.jstor.org/stable/42946642

Viani, R. A. G., Rodrigues, R. R., Dawson, T, E., Oliveira, R. S. (2011). Savanna soil fertility limits growth but not survival of tropical forest tree seedlings. *Plant and Soil*, *349*(1-2), 341-353. https://doi.org/10.1007/s11104-011-0879-7

Waidmann, S., Sarkel, E., & Kleine-Vehn, J. (2020). Same, but different: growth responses of primary and lateral roots. *Journal of experimental botany*, *71*(8), 2397–2411. https://doi.org/10.1093/jxb/eraa027

Walker, B. H., Ludwig, D., Holling, C. S., & Peterman, R. M. (1981). Stability of semi-arid savanna grazing systems. *The Journal of Ecology*, *69*(2), 473-498.

Walter, H., & Mueller-Dombois, D. (1971). *Ecology of Tropical and Subtropical Vegetation*. Edinburgh, United Kingdom, Oliver & Boyd.

Ward, D., Wiegand, K., & Getzin, S. (2013). Walter's two-layer hypothesis revisited: Back to the roots! *Oecologia*, *172*(3), 617–630. https://doi.org/10.1007/s00442-012-2538-y

Weaver, M. (2014, May 25). *Searching the Savanna*. ASU-Ask a biologist. Retrieved April 2023. https://askabiologist.asu.edu/explore/savanna

Werger, M. J. A. (1978). Vegetation Structure in the Southern Kalahari. *Journal of Ecology*, *66*(3), 933-941. https://doi.org/10.2307/2259305

Werner, C. M., Kimuyu, D., Veblen, K. E., Sensenig, R. L., LaMalfa, E., Young, T. P. (2021). Synergistic effects of long-term herbivory and previous fire on fine-scale heterogeneity of prescribed grassland burns. *Ecology, 102*(4), Article e03270. https://doi.org/10.1002/ecy.3270

Westoby, M., Walker, B., & Noy-Meir, I. (1989). Opportunistic management for rangelands not at equilibrium. *Journal of Range Management*, *42*(4) 266–274.

Whitecross, M. A., Witkowski, E. T. F., & Archibald, S. (2017). Savanna tree-grass interactions: A phenological investigation of green-up in relation to water availability over three seasons. *South African Journal of Botany*, *108*(6), 29–40. https://doi.org/10.1016/j.sajb.2016.09.003

Xu, X., Medvigy, D., Rodriguez-Iturbe, I. (2015). Relationship between rainfall intensity and savanna tree abundance explained by water use strategies. *Proceedings of the National Academy of Sciences*, *112*(42), 12992-12996. https://doi.org/10.1073/pnas.1517382112

Yang, H., Zhang, Q., Dai, Y., Liu, Q., Tang, J., Bian, X., & Chen, X. (2015). Effects of arbuscular mycorrhizal fungi on plant growth depend on root system: A meta-analysis. *Plant and Soil*, *389*(1–2), 361–374. https://doi.org/10.1007/s11104-014-2370-8

Zappala, S., Helliwell, J, R., Tracy, S. R., Mairhofer, S., Sturrock, C. J., Pridmore, T., Bennett, M., & Mooney, S. J. (2013). Effects of X-ray dose on rhizosphere studies using X-ray computed tomography. *PLoS One*, *8*(6), Article e67250. https://doi.org/10.1371/journal.pone.0067250

Zhang, B., Cadotte, M.W., Chen, S., Tan, X., You, C., Ren T., Chen, M., Wang, S., Li, W., Chu, C., Jiang, L., Bai, Y., Huang, J., & Han, X. (2019). Plants alter their vertical root distribution rather than biomass allocation in response to changing precipitation. *Ecology, 100*(11), 1-10. https://www.jstor.org/stable/26825330

Zhou, J., Huang, Z., Zhou, M., Li, X., & Jin, Z. (2013). Constraints of C-O-S-Pb isotope compositions and Rb-Sr isotopic age on the origin of Tianqiao carbonate-hosted Pb-Zn deposit, SW China. *Ore Geology Reviews*, *53*, 77-92. https://doi.org/10.1016/j.oregeorev.2013.01.001

Zhou, Y., Watts, S., Boutton, T. W., & Archer, S. R. (2019). Root density distribution and biomass allocation of co-occurring woody plants on contrasting soils in a subtropical savanna parkland. *Plant and Soil*, *438*(1/2), 263-279. https://www.jstor.org,stable/48703780

Zipkin, A. M., Hipondoka, M., & Le Roux, W. (2021). *Radiogenic strontium isoscape construction in the Kalahari and Namib deserts for provenience studies of ostrich eggshell ornaments*. [paper presentation].9th Developing International Geoarchaeology (DIG) Conference, University of Algarve, Faro, Portugal.

APPENDICES

Appendix 1: Ethical clearance certificate from the University of Namibia

UNAM
UNIVERSITY OF NAMIBIA

ETHICAL CLEARANCE CERTIFICATE

Ethical Clearance Reference Number: SOS-0045 Date: 04 March 2022

This Ethical Clearance Certificate is issued by the University of Namibia Ethics Committee (REC) in accordance with the University of Namibia's Research Ethics Policy and Guidelines. Ethical approval is given in respect of undertakings contained in the Research Project outlined below. This Certificate is issued on the recommendations of the ethical evaluation done by the ethics committee.

Title of Project: ASSESSNG SAVANNA SHRUB ROOTS DEPLOYMENT USING RADIOGENIC STRONTIUM ISOTOPES IN THE NORTHEASTERN KALAHARI, NAMIBIA

Student: HELALIA AMBAMBI

Student Number: 201410945

Supervisor(s): DR. JESAYA NAKANYALA; DR. MARTIN HIPONDOKA

Centre for Research Services

Take note of the following:

1. Any significant changes in the conditions or undertakings outlined in the approved Proposal must be communicated to the ethics committee. An application to make amendments may be necessary.
2. Any breaches of ethical undertakings or practices that have an impact on ethical conduct of the research must be reported to the ethics committee
3. The Principal Researcher must report issues of ethical compliance to the ethics committee (through the Chairperson) at the end of the Project or as may be requested by the ethics committee
4. The ethics committee retains the right to:

i) Withdraw or amend this Ethical Clearance if any unethical practices (as outlined in the Research Ethics Policy) have been detected or suspected,

ii) Request for an ethical compliance report at any point during the course of the research.

The ethics committee wishes you the best in your research.

Dr. Zivayi Chiguvare (Chairperson Ethics Committee)

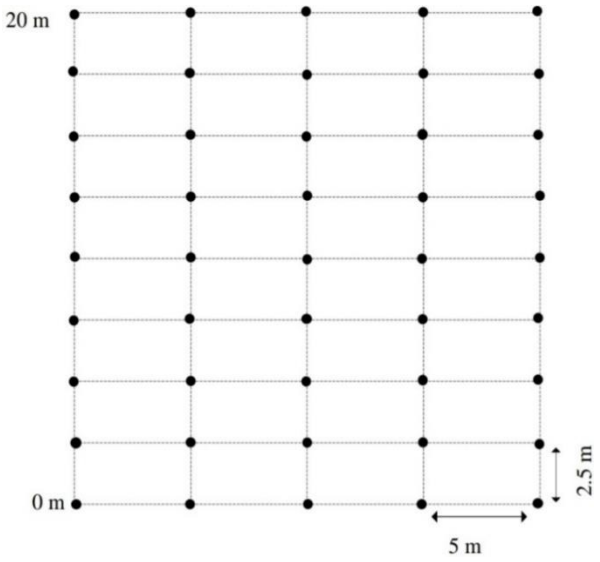
Prof. Davis Mumbengegwi (Head, Multidisciplinary Research)

Appendix 2: Field Data Collection Sheet

Date…… Name……

Shrub particulars: ID…… Species name …… Canopy diameter (m)…… Shrub height (m)……	Type root system:	Maximum root depths (cm)	Lateral roots length(cm)	Root diameter (cm)
Distance from nearest neighbours (m) 1st neighbour …… 2nd neighbour…… 3rd neighbour…… 4th neighbour……	Comments…… …….. …….. …….. ……..			

Appendix 3: A 20 x 20 m plot sketch with sampling points from which soil for root biomass was sampled.



- Sampling points

Appendix 4: Laboratory procedures for root biomass and soil moisture content

Equipment:

- Analytical balance with 0.01 g accuracy
- Drying oven

- Paper bags
- A stainless sieve (5 mm & 2 mm)
- Lab oven tray
- Small bucket

Step 1: Determine soil sampling weight

A. Arrange the soil samples per transect and depth.
B. Weigh each sample on a standard scale and record the weight.
C. Weigh a few empty plastics 3-4 empty plastic bags and determine the average weight of each sampling bag.

Step 2 (A): Sieving the soil sample

A. Place a 5mm sieve on top of a 2mm sieve and a container underneath to collect the soil when sieving.
B. Pour the entire soil sample from the plastic bag onto the 5mm sieve, break down aggregates if any and shake the sieve for soil grains to pass through the mesh.
C. Collect the coarse and fine root material retained by the two sieves, weigh them separately on analytical balance, and record their weights.
D. Repeat the process on the remaining samples.

Step 3. Determining root biomass

E. Label each sample paper bag, weigh them on an analytical balance separately, and record their weight correctly.
F. Place the air-dried roots in paper bags (fine and coarse roots separately), weigh them on analytical balance, and record their weight correctly.
G. Place the paper bags in an oven overnight at 105 ^{o}c.
H. Remove the paper bags from the oven and cool down at room temperature.
I. Weigh the oven-dried samples and record their weight.

Appendix 5: Description of the methods that were used by the Ministry of Agriculture, Water and Land Reform soil laboratory for soil physicochemical properties analysis.

SAMPLE PREPARATION	Soil samples are dried at a temperature not greater than 35 degrees C. The part of the sample retained on a 2 mm sieve, called the fine earth fraction, is used for analysis. The fraction > 2mm is referred to as stones and gravel.
AVAILABLE PHOSPHORUS	Ohlsen method: Extraction with sodium bicarbonate. Phosphate was measured spectrophotometrically using the phosphomolybdate blue method.
EXTRACTABLE CATIONS (AVAILABLE K, Mg, Ca)	Extraction with 1M ammonium acetate at pH 7. Measurement of calcium, magnesium, potassium, and sodium by inductively coupled plasma (ICP).
EXCHANGEABLE CATIONS & CATION EXCHANGE CAPACITY(CEC)	Extraction with 1M ammonium acetate at pH 7 if pH (H2O) < 6.8 & EC < 0.4 mS/cm. Extraction with 50:50 ammonium acetate (1M) and ethanol at pH 7 if pH (H2O) > 6.8 & EC > 0.4mS/cm. Calcium, magnesium, sodium, and potassium are measured by atomic absorption spectrophotometry.
TEXTURE and PARTICLE SIZE ANALYSIS (SAND, SILT and CLAY)	Dispersion of soil with sodium hexametaphosphate/sodium carbonate. Determination of silt and clay by pipette method. Sand fraction determined by sieving to retain 53 micron fraction. Textural Class using the USDA classification system.
ORGANIC CARBON (ORGANIC MATTER CONTENT)	Walkley-Black method (sulphuric acid-potassium dichromate oxidation). A factor is included in calculations to take account of incomplete oxidation. Organic matter content calculated as organic-C x 1.74
ORGANIC MATTER (by loss on ignition)	Organic matter is estimated by measuring the weight loss when dried samples are heated in a muffle furnace at 360 degrees C for 4 hours.

pH (KCl)	Measured in a 1:2.5 soil: IM potassium chloride ratio suspension on a mass-to-volume basis.
pH(water)	Measured in a 1:2.5 soil: water ratio suspension on a mass-to-volume basis.
ELECTRICAL CONDUCTIVITY (SOLUBLE SALT CONTENT)	Measurement in the supernatant of the 1:2.5 soil: water suspension before measurement of pH. Units of measurement are mS/cm (1 mS=1000 uS). High results indicating possible salinity hazard are repeated on the extract of a saturated soil paste.
TOTAL NITROGEN	The sample is then introduced to the furnace containing only pure oxygen, resulting in a rapid and complete combustion(oxidation). Nitrogen present is oxidized to NOx respectively. The NOx gases are passed through a reduction tube filled with copper to reduce the gases to N and onto a thermal conductivity cell (TC) utilized to detect the N2.
EXTRACTABLE ACIDITY	Extraction with 1M KCl and titration of extract to determine acidity.
CARBONATE (as Calcium Carbonate)	Reaction of soil with hydrochloric acid and estimation of acid consumed by titration with sodium hydroxide.
CARBONATE (estimation)	Treatment of dry soil with 10% hydrochloric acid and observation of effervescence.
AVAILABLE SULPHUR (as SULPHATE)	1:2 weight: volume extraction of soil with 0.01M calcium chloride. Sulphate-S was estimated by measuring turbidity at 600 nm following treatment with acidified barium chloride.
SULPHATE (estimation)	Soil: water extract from pH/EC measurement made 0.01M concerning calcium by addition of 1M calcium chloride. Filtered extract reacted with acid barium chloride and turbidity visually compared with standard solution of sulphate-S.
SALINITY ANALYSIS	

	Saturated soil: water paste prepared, and the extract recovered by vacuum filtration. Anions and cations are measured in the extract. Sodium adsorption ratio (SAR) is a diagnostic criterion for assessing salinity. It is equal to the concentration of sodium divided by the square root of one-half the combined calcium and magnesium in the extract. All concentrations measured in me/l
AVAILABLE MICRONUTRIENTS (Zinc, manganese, copper, and iron)	Extraction with 0.5M ammonium acetate: 0.5M acetic acid: 0.02M EDTA at pH 4.65 at a 1:5 extraction ratio. Fe, Mn, Cu and Zn were measured by Inductively Coupled Plasma (ICP). Available calcium, potassium, and magnesium can also be measured in the extract.

Note: 1 ppm (part per million) = 1 mg/kg = 1 ug/g

1% = 10 000 p

Appendix 6: (a) One of the complex rooting systems of *Grewia flava* that was exposed during excavation and (b) shallow roots deployment of *Acacia hereroensis*, (10 cm maximum rooting depth).

(a)

(b)

Appendix 7: The above-ground data of the sampled shrubs

Shrubs species	Shrubs' height (m)	Shrubs' canopy (m)
Acacia hereroensis	1.1	1.8
Commiphora angolensis	1.3	0.95
Dichrostachys cinerea	1.4	0.95
Dichrostachys cinerea	2.5	0.5
Dichrostachys cinerea	1.7	1.8
Grewia flava	1.1	1.2
Grewia flava	1.5	2.1
Grewia flava	1.3	2
Grewia flava	1.6	2
Grewia flava	0.8	1.2
Rhus tenuinervis	1.7	1.45
Terminalia sericea	1.1	0.95
Terminalia sericea	1.8	2.35
Terminalia sericea	1.77	1.5
Senegalia mellifera	1.63	2.8
Ximenia americana	1.9	0.85
Ximenia americana	1.4	1.35
Ximenia caffra	1.2	0.9
Ximenia caffra	0.13	0.6

Appendix 8. The percentage of vertical distribution of sand, clay, and silt soils of the studied soil depths

	Soil type		
Depth (cm)	Sand (%)	Clay (%)	Silt (%)
0-10	90.5	3.4	5.5
10-20	90.0	4.3	5.7
20-30	88.0	6.0	6.0
30-40	90.4	3.7	6.0
40-50	90.3	3.7	6.0
50-60	89.9	4.1	6.0
60-70	90.1	3.8	6.1
70-80	88.1	5.4	6.4
80-90	89.2	3.9	6.9
90-100	88.9	4.1	7.0