WATER USE EFFICIENCY AND CARBON SEQUESTRATION POTENTIAL OF INDIGENOUS CROPS

Report to the
Water Research Commission

by

R Zengeni, V Chaplot, H Shimelis, I Mathew, N Mbava, S Ntonta, N Mbambo, K Shayamurira

School of Agricultural, Earth and Environmental Sciences
University of KwaZulu-Natal

WRC Report No. 2721/1/21
ISBN 978-0-6392-0258-7

July 2021

Obtainable from

Water Research Commission
Private Bag X03
Gezina, 0031
PRETORIA

EXECUTIVE SUMMARY

Water is fundamental to agricultural development and enhancing food security. However, climate change is a serious threat to provision of adequate water for agricultural, industrial and municipal use. South Africa and sub-Sahara Africa, in general, are challenged by water scarcity and should use the limited available water more efficiently. Collaborative thinking is central to mitigate climate change and improve water use efficiency in agriculture by adopting strategies that enhance long-term storage of carbon in the soil, and cultivating crops and varieties that utilize water efficiently. Most smallholder farmers in South Africa cultivate crops in regions that have limited water availability and marginal soil fertility to achieve meaningful agricultural productivity. This project sought to explore ways of using crops that are readily available to local farmers to simultaneously cope with unfavorable water supply while improving soil fertility and mitigating against climate change. It sought to enhance crop production by selecting local crop varieties that have high capacity for soil carbon sequestration and efficient utilization of limited water supply. Increased carbon sequestration ultimately improves the water and nutrient holding capacities of the soil. The specific objectives of the project were:

i. To evaluate water-use efficiency and soil C sequestration potential of sorghum varieties in comparison with maize and wheat varieties;
ii. To assess the effect of crop residue incorporation on soil biological properties (microbial biomass, enzyme activity, C pools, mineralizable N and P);
iii. To build capacity of post-graduate students and smallholder farming communities on water-use efficiency and soil C dynamics;
iv. To build a multidisciplinary team of soil scientists, crop breeders and hydrologists/climatologists;
v. To identify the contribution of crop production to global carbon credits.

The major findings of the project were that biomass production, grain yield, water use efficiency and carbon sequestration potential varied significantly among maize, wheat, sorghum and pearl millet varieties. Differences in carbon sequestration were direct results of variation in biomass productivity and chemical composition of the biomass since carbon sequestration is achieved primarily via two processes, i.e. decomposition of plant matter and rhizodeposits. In general, crops differed significantly in their water use efficiency for grain yield production. Similar to other reports, C4 plants such as maize and sorghum exhibited significantly higher WUE than C3 types (e.g. wheat and barley). Water-use efficiency was affected by climatic and soil factors. Higher WUE values were observed for warm and wet climates than cooler environments, while clay soils induced lower WUE values in crops compared to loam soils. Maize exhibited generally high WUE in all soil textures. Specifically, maize grain yield was 8.2 t ha^{-1} followed by sorghum (6.5 t ha^{-1}), millet (6.3 t ha^{-1}) and wheat (3.1 t ha^{-1}). The high biomass production potential of maize, sorghum and millet is supported by their C4 photosynthesis compared to wheat, which is a C3 crop. The average root to shoot ratio was highest for sorghum with 0.55, followed by maize (0.18) and wheat (0.12). Drought stress had the highest impact on grain yield production on maize (52%) and wheat (43%) compared to sorghum (26%) and millet (25%). Amongst cereals, maize (3.78 kg m^{-3}) and sorghum (2.52 kg m^{-3}) were more water-use efficient than wheat (1.02 kg m^{-3}), barley (1.21 kg m^{-3}) and millet (0.47 kg m^{-3}). Overall, maize was the most water use efficient crop under well-watered conditions (9.90 kg m^{-3}), but sorghum was the most efficient under dry conditions (5.99 kg m^{-3}). Similarly, grain yield production followed a similar trend with maize attaining 8.2 t ha^{-1} followed by sorghum (6.5 t ha^{-1}), millet (6.3 t ha^{-1}) and wheat (3.1 t ha^{-1}).

The review of global data showed that C transfer was highest in maize, which yielded the greatest soil C sequestration potential (1.0 Mg C ha^{-1} yr^{-1} or 19% total assimilation) followed by sorghum (1.0 Mg C ha^{-1}, 17%) and wheat (0.8 Mg C ha^{-1} yr^{-1}, 23%). These rates were comparable to potential C sequestration of most land management practices showing that selecting suitable crops was equally important. Only 7% of the assimilated C was stabilized in the soil but wheat transferred 23% of its total C allocation to the soil followed by rice with 20%, and maize and ryegrass at 19%. Higher C transfer to soils occurred under clayey soils and warmer climates provided that exudation is high enough to offset respiration C losses. These results could be used in selecting crop species and ideotypes for high soil C sequestration, while the variability within species could be useful in breeding efforts to enhance the genetic capacity of existing crops to sequester C.

The study highlighted C allocation within plants parts in maize, sorghum and wheat, which can be used as a basis for identifying crops and genotypes with high C sequestration potential. Sorghum showed the highest potential for C sequestration based on high root biomass productivity. Among sorghum genotypes, AS15 exhibited the highest potential for rooting biomass and C stocks. For maize, Nel and Obanapta would be superior in shoot C sequestration under well-watered and drought stressed conditions respectively. BW140 is the most suitable genotype of wheat to promote C sequestration in water limited environments. The ability of the genotypes to sequester atmospheric carbon into the soil varied highly with differences of up to 87% under drought conditions, and up to 117% under well-watered conditions. Secondly, C transfer to the soil was higher under well-watered than water-stressed conditions. Thirdly, high allocation of carbon to shoots reduced potential carbon sequestration in the soil. Thus, incorporation of crop residues from different sorghum and wheat genotypes had significant effects on CO_2 emission, N and P mineralization. Sorghum residues generally evolved lower CO_2 (in both roots and shoots) compared to wheat residues, showing greater potential for sorghum to sequester carbon. The decomposition of residues is largely regulated by microbial activity which is influenced by the residue quality. In general, the residue quality is negatively related to its C:N ratio, lignin: N and lignin content. Low C: N ratio causes rapid mineralization of organic N, whereas high C: N in residues results in immobilization of mineral N as it is locked up in microbial biomass. Genotypes exhibiting lower C: N ratio, low lignin: N ratio and low lignin content (BW162 and BW140 shoots for wheat; and 05 POTCH roots and shoots for sorghum) showed rapid mineralization thereby evolving higher CO_2-C and mineralizing more N. Those with high C:N ratio, lignin: N ratio and lignin content (LM70 and BW152 roots for wheat and AS8 roots and KZ5246 root and shoots for sorghum residues) showed slow decomposition, thus releasing low CO_2-C and low mineral N. This would be beneficial for carbon sequestration and longer-term availability of nutrients. Phosphorus mineralization is mainly influenced by the initial P concentration of residues. Residues with higher initial P content (BW152, LM70 and BW162 shoots for wheat residues, and AS8 roots for sorghum residues) released higher concentrations of extractable P. Overall, the residues that evolved the lowest CO_2-C, namely LM70 wheat roots, as well as AS8 and KZ5246 sorghum roots and KZ5246 shoots, are recommended to farmers for carbon sequestration and improvement of soil organic carbon (SOC). On the other hand, shoots of wheat genotypes BW162, BW140 and BW152 as well as sorghum roots and shoots from 05-POTCH-138 could be a good source of N recommended to farmers. Lastly shoots from wheat genotypes BW162, BW152 and LM75 as well as sorghum roots from AS8 could be ready sources of mineral P into the soil that can potentially serve as organic P fertilizer.

Wide intra-specific variation in biomass production and its allocation to roots and shoots, and biochemical composition was found among the different genotypes of wheat, maize and sorghum, showing that there was potential for selecting superior varieties for improving water

use efficiency, drought tolerance and carbon sequestration in different crops. The results from correlation analyses showed that it was possible to simultaneously select for high grain yield and root biomass production to satisfy both food production and C sequestration needs but with consideration for water availability.

The project designed protocols for investigating water use efficiency, carbon sequestration and drought tolerance using wheat as a model crop. Previously, these three phenomena were not investigated simultaneously despite their interlinkage. The methods described in this study can be adapted for any crop that can be grown in the field and greenhouse over a relatively short period of time such as 5 months, which is the average growth and reproduction cycle for most annual field crops. The following methodological considerations are recommended by this project:

i. There is a lack of information in South Africa, particularly on carbon sequestration and the available information does not meet international standards to a larger extent; thus, more research should be done on this. Available literature is mostly found as technical reports that have not been published in peer reviewed journals and is largely inaccessible to a wider audience.

ii. When conducting WUE trials, the water balance must be considered and measured as accurately as possible. Irrigating with known quantities of water is necessary to calculate WUE independently for each variety and by considering the duration from sowing to senescence. Mechanisms for continuous evaluation of the required amount of water must be developed and implemented because plant available water in the soil and water requirements change with crop growth stage. There is also need to reduce constraints such as weeds, pest and diseases, nutrient imbalances, soil type variability and soil acidity that directly reduce plant growth or indirectly compete for water availability.

iii. The quantification of water applied and transpired is a highly contentious issue in crop-water relations and is dependent on the type of probes used for measuring water in the soil prior to, during and after experimentation. Highly specialized probes have been developed that are ideal for pot and field trials, and researchers must carefully select the most suitable for their circumstances. Probes must be inserted to reach and account for water in the deeper horizons at least 60 cm deeper for most annual field crops. There are instances where the minimum depth can be 1 m, e.g. in deep soils where there are no impediments for root growth. It is also important to use probes that ensure that soil moisture availability can be measured at incremental depths.

iv. In the greenhouse pot trials, the pot size must be large enough (between 5 and 10 litres) to simulate field conditions and allow reasonable root growth. However, this also brings the challenge of plant density per unit area. Sparsely planted crops tend to compensate by developing large canopies that can be detrimental to reproductive growth and increase evapotranspiration. Prolific vegetative growth beyond the optimal architecture reduces water use efficiency and increases competition for resources between roots and shoots. Small root biomass reduces carbon sequestration capacity and water exploration acquisition in general but may not be very critical in pot trials.

v. Increase in soil organic matter content in the soil will induce a greater water retention capacity of the soil. Thus, the water balance in the soil will be incremental with improvement in soil fertility status. Ultimately, the water balance must be calculated each year to determine critical points for drought stress induction, wilting points and optimal water application during experimentation.

vi. The project also learnt that it is important to include locally varieties of crops to improve adoption rates among stakeholders. For a project dealing with crop varieties, it is imperative to include local adapted varieties. However, the challenge of low productivity among local varieties remains pertinent especially where the farmer is concerned with high yield as a priority over any other attribute. Developing or identifying varieties that outperform local varieties in grain yield, water use efficiency and carbon sequestration will be important for adoption and lobbying governments for carbon credits. The accrual of benefits such as reduced water and fertilizer application to farmers and reduced carbon emission for the environment are appealing to governments.

vii. The development of crop varieties that are going to be adopted by farmers needs to look at all the characteristics of the crops that enhance C sequestration capacity, drought tolerance and high water use efficiency while at the same time fulfilling the purpose of food production.

viii. Carbon sequestration using carbon isotope tracing methods is still largely confined to the greenhouse using a small number of genotypes grown for a relatively short period of time due to huge costs. We improvised the method for greenhouse using a subset of 10 genotypes of wheat and could not extend the experiment to other crops because of the cost. There are methods that have been developed for field experiments using isotopes but these are currently out of reach for many projects in sub-Sahara Africa; due to technical and financial constraints that prevent their implementation in developing countries.

ix. The main limitation of the study was that the C fluxes were estimated from planting to harvest, while multiple cycles or across year experiments should possibly provide a more accurate estimation of soil C sequestration by crops.

The study also identified challenges and limitations that could be encountered in implementing the recommendations proposed. Recommendations for wheat and sorghum production under smallholder farmers in South Africa are limited despite their huge potential for carbon sequestration and water use efficiency. Promoting wheat as a candidate for C sequestration among smallholder farmers in South Africa will be challenged by acute water shortages in communal farming systems where there is a lack of provision for irrigation infrastructure. On the other hand, sorghum is less preferred as a food crop among smallholder farmers in South Africa, which will complicate its promotion for carbon sequestration and water use efficiency because the farmers may find it less useful for food production. Maize is widely grown among communal farmers, but is particularly sensitive to water limitations resulting in reduced C sequestration potential under marginal conditions. For accurate quantification of carbon deposits into the soil, the isotope tracing method is widely used but the method is still largely confined to the greenhouse. There are methods that have been developed for field experiments using isotopes but these are currently out of reach for many projects in sub-Sahara Africa due to technical and financial constraints.

The conclusions derived from this study suggest that more still needs to be understood on the main factors controlling water use efficiency and carbon sequestration in crops. There are many interactions between crop water use efficiency and carbon sequestration in relation to agronomic practices such as fertilizer application, irrigation scheduling, tillage system, planting dates, and plant architecture. Such interactions could not be investigated in the framework of the present study but present important grey areas for further research. Such an improved understanding of the links between plant characteristics, water use efficiency and soil C storage may open opportunities for plant breeding and improved agronomic practices with climate change mitigation in mind. Research on stover management strategies that address farmer needs while enhancing soil C is also needed. There is need to do a cost benefit analyses for C sequestration between accumulating high biomass compared to having more favorable biochemistry such as high C: N ratio, high lignin and high cellulose contents to increase recalcitrance of plant residues. The biochemistry of crop residues and carbon deposits have huge implications for microbial populations in the soil that affect the stability and mineralization of the deposited carbon. It will be imperative to conduct assessment on changes in microbial population in the soil following incorporation of different crop residues and determine carbon sequestration potential in that regard. The main limitation of the study was that the carbon fluxes in the soil were estimated from planting to harvest in a single season. Carbon sequestration in the soil is a long-term process and may be difficult to accurately quantify over short periods. Conducting multiple year experiments should possibly provide a more accurate estimation of soil C sequestration by crops.

Lastly, our research was done as on-station experimentation, where plant growth conditions are ideal and controlled. More research needs to be done on-farm to see the performance of the different crop varieties under real field conditions that the farmers are experiencing.

ACKNOWLEDGEMENTS

The project thanks the following reference group members:

Prof Sylvester Mpandeli	Water Research Commission
Dr Luxon Nhamo	Water Research Commission
Dr S Hlophe-Ginindza	Water Research Commission
Dr G Backeberg	Water Research Commission
Prof Trevor Hill	University of KwaZulu-Natal
Dr Willem Jansen van Rensburg	Agricultural Research Council
Dr JJ Botha	Agricultural Research Council
Prof W van Averbeke	Tshwane University of Technology
Mr JA Denison	
Dr DJ Beukes	

The following project team members were instrumental to the research:
Dr Rebecca Zengeni
Prof Vincent Chaplot
Prof Hussein Shimelis
Dr Macdex Mutema
Dr Isack Mathew
Miss Nozibusiso Mbava
Mr Sipho Ntonta
Mr Nhlakanipho Mbambo
Mr Kwame Shayamurira

We would also like to acknowledge the following for their contributions to the project:
Prof Pardon Muchaonyerwa
Dr Alistair Clulow, UKZN
Mr Ian Doidge, UKZN
The staff at Ukulinga farm and Agriculture campus of the University of KwaZulu-Natal for technical assistance.

The following partner institutions supported the project immensely:
University of KwaZulu-Natal, Pietermaritzburg Campus
African Centre for Crop Improvement (ACCI)
Institute for Research and Development-France (IRD)
National Research Foundation of South Africa (NRF)

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

LIST OF ABBREVIATIONS

ACCI	African Centre for Crop Improvement
ARC	Agricultural Research Council of South Africa
C	Carbon
CO_2	Carbon dioxide
Cseq	Carbon sequestration
GY	Grain yield
NH_4^+-N	Ammonium- nitrogen
NO_3^-N	Nitrate -nitrogen
P	Phosphorous
PB	Plant biomass
Pc	Plant carbon
RB	Root biomass
Rc	Root carbon
Rcs	Root carbon stocks
SB	Shoot biomass
Sc	Shoot carbon
Scs	Shoot carbon stocks
SOC	Soil organic carbon
UKZN	University of KwaZulu-Natal
WRC	Water Research Commission
WUE	Water use efficiency
^{13}C	Carbon 13 isotope

Chapter 1 INTRODUCTION

1.1. Background and motivation of the project

Climate change, aridity and poor soil fertility are major constraints threatening crop production in the world and South Africa, in particular. Thus, it was envisaged that identifying crops and varieties with high water use efficiency, and developing strategies that would enhance deposition of carbon into soils would prevent further degradation of soil quality, and help in mitigating carbon emission into the atmosphere. This will have cascading positive impact on the environment by improving water usage and reducing the impact of drought stress on crop production. The initial stages of the project focused on varietal selection of the different crops to identify varieties that are carbon and water efficient. Subsequently, the impact of the residues of each crop on soil carbon dynamics was elucidated to determine the residues with the highest potential for carbon sequestration and minimum carbon dioxide emission over a stipulated period. Crop residues are readily available and can directly contribute to soil organic matter build up and nutrient cycling. Wheat, maize and sorghum are amongst the most commonly grown crops in South Africa. These produce large amounts of biomass with high potential to contribute to carbon sequestration and building up of soil organic matter reserves. However, their contribution to soil carbon emission mitigation, increasing soil organic carbon stocks and preserving soil moisture have not been well investigated in the South African context.

1.2. Aim and objectives

The aim of this study was to evaluate water use efficiency and carbon sequestration potential of sorghum, maize and wheat. Simultaneous assessment of WUE, crop productivity and carbon cycling for different cultivars of these three crops were carried out to fulfil these objectives. Specifically, the study evaluated biomass production, grain yield, C sequestration potential and water use efficiency of maize, sorghum and wheat varieties over a four-year period. The study sites selected for this research were guided by availability of ancillary infrastructure such as adequate security, access roads and presence of weather stations, and these included the Ukulinga University of KwaZulu-Natal Research Farm and Cedara experimental stations both in Pietermaritzburg. The specific objectives of the project were:

- **i.** To evaluate water use efficiency and soil C sequestration potential of sorghum varieties in comparison with maize and wheat varieties.
- **ii.** To assess the effect of crop residue incorporation on soil biological properties (microbial biomass, enzyme activity, C pools, mineralisable N and P).

iii. To build capacity of post-graduate students and smallholder farming communities on water-use efficiency and soil C dynamics.

iv. To estimate the potential contribution of crop production to global carbon credits.

Chapter 2 WATER USE EFFICIENCY OF DIFFERENT CROPS (A META-ANALYSIS)

Abstract

Water is a limiting natural resource for agricultural production. While it is well accepted that crop types differ in their water use efficiency (WUE), there is no consensus on the main factors affecting WUE of main field crops, which was the motivation of this meta-analysis. The effects of rainfall pattern, soil type and climatic regime on crop WUE were evaluated using data from 514 experiments around the world published in ISI journal papers. The results confirmed that crop type had a significant effect ($p < 0.05$) on WUE with cereals producing on average 2.37 kg of dry grain per cubic meter (m^{-3}) of water followed by oilseeds (0.69 kg m^{-3}), fibre crops (0.45 kg m^{-3}) and legumes (0.42 kg m^{-3}). Amongst cereals, maize (3.78 kg m^{-3}) and sorghum (2.52 kg m^{-3}) were more water-use efficient than wheat (1.02 kg m^{-3}), barley (1.21 kg m^{-3}) and millet (0.47 kg m^{-3}). Overall, maize was the most water use efficient crop under well-watered conditions (9.90 kg m^{-3}), but sorghum was the most efficient under dry conditions (5.99 kg m^{-3}). WUE of crops increased from desert to tropical through sub-tropical climate. Moreover, WUE tended to correlate positively with soil organic carbon content ($r=0.86$) and negatively with clay content ($r=-0.78$) and soil bulk density ($r=-0.85$). These results provide information that is important for making decisions on crop selection in a context of increased climate variability and for crop variety development with enhanced WUE. However, there is need for more research to gain understanding on the mechanisms responsible for the observed trends and causes of the unexplained variability.

Keywords: Climate variability, Crop water use efficiency, Crop management, Photosynthetic process, Soil water availability

2.1. Introduction

Water scarcity is a major global environmental problem of the 21st century (Srinivasan et al., 2012). Globally, agriculture accounts for 80-90% of all freshwater used by humans, and most of that water is used for crop production (Morison et al., 2008). While irrigation development has increased crop productivity in arid and semi-arid areas for decades, water scarcity and escalating costs of setting and managing the infrastructure hamper further expansion of irrigation in developing countries. Rising demand for water by other sectors such as domestic use, mining, industries, and recently severe pressures from climate uncertainties, exacerbates water shortages for further irrigation development. Therefore, there is need for new paradigms for agriculture to at least keep pace with rising demand for food, while using the lowest possible amounts of water.

Different disciplines define water use efficiency (WUE) differently. Originally, crop physiologists defined WUE as the amount of carbon assimilated and crop yield per unit of transpiration (Viets, 1962) and later it was referred to as amount of biomass or marketable yield per unit of evapotranspiration. Irrigation scientists view WUE as a ratio of total irrigation water transpired to water diverted from the source (Israelsen, 1932), while crop scientists define it as the ratio of total biomass or grain yield to water supplied (Sharma et al., 2015). The present paper adopted WUE as the ratio of grain yield achieved to the amount of water made available to the crop (i.e. stored in the soil plus rainfall and irrigation water). It is expressed as grain yield per unit of land area (Y, kg m^{-2}) divided by the amount of water consumed by the crop per unit land area (ET, m^3 m^{-2}), usually reported as mm of water needed to produce that yield (Blum, 2005).

The need for crops to be drought tolerant and have higher WUE has been the focus of breeding efforts for many years (Sivamani et al., 2000; Tilman et al., 2002; Condon et al., 2004; Blum, 2005; Ruggiero et al., 2017) to alleviate water scarcity and food insecurity. Improved soil fertility and crop management practices have also been developed to enhance better yields with less water (Evans and Sadler, 2008; Busari and Salako, 2013; Busari et al., 2015; Wang et al., 2018). However, water scarcity remains an issue and there is still need for better understanding of the underlying controls of crop WUE.

It is well accepted that crops differ in WUE abilities. Zwart and Bastianssen (2004), in a global meta-analysis, found WUE to increase from 0.6-1.7 kg m^{-3} for wheat, 0.6-1.6 kg m^{-3} for rice to 1.1-2.7 kg m^{-3} for maize. Several other studies pointed that cereals tend to have higher WUE than oilseed crops (Norton and Waschsmann, 2006; Sadras and Mcdonald, 2012). Water supply also showed significant effect on crop WUE, with several studies reporting a general increase of WUE with decreasing water supplied (e.g. Erdem et al., 2001; Rusere et al., 2012; Mabhaudhi et al., 2013; Chibarabada et al., 2015). For example, Erdem et al. (2001) reported higher sunflower WUE under water stressed than under well-watered conditions in their study at Tekirdag, Turkey. In South Africa, Chibarabada et al. (2015) reported the highest Bambara groundnut WUE under severely water stressed conditions (WUE of 0.006 g mm^{-1} at 25% Field Capacity, FC) and lowest WUE under non-stressed conditions (0.003 g mm^{-1} at 75% FC).

Rusere et al. (2012) also reported increasing WUE with decreasing amount of water applied in irrigation-based winter silage maize trials in Zimbabwe. Type of irrigation system used was also shown to have significant effect on WUE with drip systems yielding higher cotton WUE (0.77 kg m^{-3}) than the furrow systems (0.49 kg m^{-3}) (Ibragimov et al., 2007). In addition, other factors such as soil type, climate, crop and land management practices also exhibit significant effects on crop WUE. Ismail and Ozawa (2007) reported from 45 to 64% higher WUE in crops grown on clayey than sandy soils. In a different study, Dou et al. (2016) also reported (25%) higher WUE for rice grown on clayey than sandy loam soils; and higher WUE for wheat, oats, potato and maize under warm-dry than warm-wet climate in northern China. Fan et al. (2005) reported higher WUE in fertilized (0.95 kg ha^{-1} mm^{-1} and 0.96 kg ha^{-1} mm^{-1}) than unfertilized crops (0.32 kg ha^{-1} mm^{-1} and 0.47 kg ha^{-1} mm^{-1}) for wheat and maize respectively in Gansu, China. Moreover, Wang et al. (2018) reported an increase of water use efficiency of winter wheat by 5.1%, 13.8% and 29.3% respectively at an application rate of 60, 120 and 180 kg N ha^{-1} respectively. Zhang et al. (2017) pointed to a combined effect of water and nitrogen applications and showed that a decreasing water supply with nitrogen fertilizer application before cotton flowering and increasing the amounts at later growth stages thereby increasing WUE. Sinclair (2018) gives some explanation on how water shortage may improve WUE. This author showed that drought occurring early in the crop growth cycle partially closes stomata which results in the conservation of soil water and subsequent improved crop yield per unit of water.

Conservation tillage also had the ability to enhance WUE in comparison with conventional tillage in Henan Province, China. In addition, mulching of crop residues was reported to reduce evapotranspiration (Xie et al., 2005; Zheng et al., 2018) with, for instance, Xie et al. (2005) reporting 16% higher WUE in mulched than un-mulched wheat (0.875 and 1.02 kg m^{-3}, respectively). Several studies have reported disparities in crop WUE under different soil types, climates and water regimes, and there is no consensus yet on the impact of these factors. However, the existence of studies across the world provides an opportunity for a comprehensive analysis of the main controls of crop WUE. Therefore, the objectives of the current study were to compile and analyze available studies on the WUE of main crops in order to assess and quantify variations due to soil and environmental conditions. In pursuit of these objectives, the study compiled data from 514 experiments around the world.

2.2. Materials and methods

2.2.1. Database preparation

Online sources of published ISI journal papers sources such as Google Scholar, Science Direct, Springerlink, Scopus, Web of Science and Researchgate were searched for literature on WUE using key terms such as “crop water use”, “crop water use efficiency”, “water use efficiency” and “water use efficiency for different crops”. Only papers reporting WUE for named field crops and environmental conditions, e.g. site, climate, soil type, whether the trial was in an open or controlled environment, amount of water applied and/or rainfall received were considered. In addition, data on plant population, fertilizer application rate, yield, and aboveground biomass were also captured. The final database, summarised in Table 2.1, consisted of 684 WUE data points from 60 peer-reviewed ISI journal papers. The main data contributors were USA (n=138), Turkey (n=130), China (n=100), Egypt (n=41) and Syria (n40). Southern Africa had South Africa (n=16), Malawi (n=10) and Zimbabwe (n=6) as the main contributors.

Table 2.1. Database compiled using data from ISI journal papers showing references and averages of selected environmental values

Crop Name	Country	n	MAP (mm y^{-1})	MAT (°C y^{-1})	TW (mm)	AWC %	BD (g cm^{-3})	Clay %	Sand %	Silt %	WUE (kg m^{-3})	Bio (t ha^{-1})	References
Wheat	Argentina	18	577	18	63						3.43	378	Abbate et al., 2004
Cowpea	Egypt	3	0	22	360	6	1.5				0.62		Aboamera, 2010
Maize	Egypt	6	0	21	935	25	1.31				1.2		Abuarab et al., 2013
Barley, Faba bean	Saudi-Arabia	16	100	16	479	8	1.58	3	81	16	0.72		Al-Neem 2008
Bambara	South Africa	12	845	19	110	34					0.005	1	Chibarabada et al., 2015
Sorghum	USA	8	198	22	808						3.05		Conley et al., 2001
Sorghum	Spain	9	456	14	137	8		17	79	7	5.29	2856	Curt et al., 1995
Cotton, Maize	Turkey	20	657	18	374	21	1.46	12	45	26	1.32	1380	Dağdelen et al., 2006
Cotton	Turkey	4	657	18	548	23	1.46	17	55	28	0.85	968	Dağdelen et al., 2009
Sunflower	Turkey	15	575	14	387	15	1.56				0.74		Erdem et al., 2001
Elephant grass, Energycane, Giantreed	USA	6	1228	27	1159						3.23	3270	Erickson et al., 2012
Maize													
Wheat	China	6	580	14	766	16	1.35				2,36		Gao et al., 2008
Maize	USA	12	397	20	592	13					1.29	1626	Howell et al., 1995
Sorghum	Egypt	12	0	22	7300						5.99		Hussein and Alva, 2014
Cotton	Syria	8	120	17	600	19	1.16				0.66		Hussein et al., 2011
Cotton	Uzbekistan	9	228	26	541						0.67		Ibragimov et al., 2007
Maize	Iran	6	279	14	0	8	1.42				7.13		Kanani et al., 2016
Wheat	China	45	542	9	477		1.2	31	4	66	1.09	1036	Kang et al., 2001
Linseed, Mustard, Safflower	India	9	1472	27	171	15	1.58	27	47	26	0.19	495	Kar et.al., 2007
Maize	Turkey	17	679	14	1391	15	1.4	33	24	44	3.45	2893	Kuscu and Demir, 2013
Maize	Turkey	12	126	21	669	15					1.71		Kuscu et al., 2013
Alfalfa	Turkey	12	409	6	474	8	1.42	26	37	38	1.13	502	Kuslu et al., 2010
Sunflower	Egypt	20	0	22	1315						0.93		Mahmoud et al., 2016
Sorghum	Italy	3	876	15	0	12		44	30	26	5.5	2694	Mastrorilli et al., 1999
Sorghum	South Africa	4	710	17	950						3.39	1318	Mengistu et al., 2016
Maize	Ethiopia	15	831	21	188	17	1.17				10.67	2115	Meskelu et al., 2014
Cotton	Pakistan	2	252	27	154						0.6		Muhammad et al.,2016
Millet	Tunisia	8	207	27	296						0.7	811	Nagaz et al., 2009
Cotton	Turkey	8	1109	18	1447	25	1.43	69	15	15	0.74		Onder et al., 2009
Chickpea	Syria	12	330	28	816						0.51	415	Oweis et al., 2003
Maize	Italy	3	650	15	340	18	1.25	40	6	53	1.93	1652	Di Paolo and Rinaldi 2008
Maize	USA	16	508	10	570	26					1.45	190	Payero, et al., 2008
Maize	USA	16	508	10	439	26					1.5	36	Payero et al., 2009

Crop Name	Country	n	MAP (mm y^{-1})	MAT (°C y^{-1})	TW (mm)	AWC %	BD (g cm^{-3})	Clay %	Sand %	Silt %	WUE (kg m^{-3})	Bio (t ha^{-1})	References
Sunflower	Pakistan	2	178	24	933						0.37		Qureshi et.al., 2015
Maize	Zimbabwe	6	850	22	388						11.81	4205	Rusere et al., 2012
Millet	Niger	8	252	29	0		1.65	4	91	5	0.24		Sivakumar and Salaam, 1999
Cotton	USA	42	142	25	501						0.22		Snowden et al., 2013
Maize, Sorghum, Sunflower	USA	14	419	14	568						1.18		Stone et al., 1996
Wheat	China	4	614	14	439		1.38	15	2	83	1.18		Su et al., 2007
Wheat	Turkey	22	318	12	365	14	1.59				1.18	1611	Tari,2016
Maize	Malawi	10	1142	24	0			23	74	4	7.6		Teravest et al., 2015
Wheat, Maize	China	12	540	7	345		1.3				0.09		Fan et al., 2005
Sorghum	USA	24	177	13	235						1.49		Tolk and Howell, 2003
Bean, Green gram	Uzbekistan	24	261	16	275	14					1.71	312	Webber et al., 2006
Wheat	Syria	20	252	27	430	16					2.29	796	Zhang et al., 1998
Wheat	China	24	129	7	433	23	1.37				0.95		Xie et al., 2005

mean annual precipitation and temperature, MAP and MAT respectively; number of data points (n), total amount of water onto the plot, TW; available water capacity of soil, AWC%; topsoil bulk density, BD; topsoil clay, sand and silt content, Clay, Sand and Silt, respectively) and response variables (water use efficiency, WUE; and above ground biomass, Bio).

2.2.2. Definitions of WUE and environmental factors

In the current study, WUE was defined as the amount of crop yield produced per unit volume of water used (kg m^{-3}), regardless of differences in growing seasons for different crop types and varieties. Biomass is the amount of aboveground dry matter. All WUE data came from peer reviewed papers. Table 2.2 presents the definitions of selected soil and environmental conditions. The soil parameters used were topsoil clay, sand and silt content, bulk density, field capacity moisture, permanent wilting point and available water capacity. The other environmental factors related to site (LONG: longitude, LAT: latitude and Z: altitude), and climate (MAP: mean annual precipitation, and MAT: mean annual air temperature). When climatic characteristics were not available in the papers, surrogate data from nearby prominent features (e.g. town) were used from Wikipedia. Environmental conditions varied widely and categories were generated for analyses (Table 2.3). Climates are defined here in terms of MAP and MAT following Mathew et al. (2017), and do not necessarily comply with the Köppen (1936) system. Tropical represents hot (MAT>20°C $year^{-1}$) and wet (MAP>1000 mm $year^{-1}$) climate; subtropical depicts warm (MAT: 10-30°C $year^{-1}$) and arid to humid (MAP: 100-1110 mm $year^{-1}$); temperate represents cool (MAT<10°C $year^{-1}$) and arid to moist (MAP: 120-1000 mm $year^{-1}$); while desert corresponds to warm (MAT>15°C $year^{-1}$) and dry (MAP: 0-100 mm $year^{-1}$) zone. Soil texture (clay, sand and silt content) and amount of water supplied (precipitation plus irrigation) were categorised into low, medium and high class (Table 2.2). Crops were categorised by type and growing season (Table 2.3).

Table 2.2. Definitions of environmental factors, water use efficiency, yield and biomass used in the analysis

Environmental factors	Symbols	Units	Definitions
Mean annual precipitation	MAP	mm $year^{-1}$	Long-term (at least 30 year) mean precipitation per year for the study location from the papers
Mean annual air temperature	MAT	°C $year^{-1}$	Long-term (at least 30 year) mean temperature per year for the study location from the papers
Longitude	LONG	°	Longitude of the midpoint of study site as given in papers
Latitude	LAT	°	Latitude of the midpoint of study site as given in papers
Altitude	Z	m.a.s.l	Average elevation above sea level of the study site as given in the papers
Rooting depth	Zd	m	Depth within the soil profile where most of the crop roots were found
Soil bulk density	BD	g cm^{-3}	Bulk density of the top soil layer as given in papers
Total Water	TW	mm	Total amount of water received by the crop during the full crop cycle (i.e. precipitation + irrigation)
Available water capacity	AWC	%	Amount of soil water available to crops (i.e. the calculated as the difference between field capacity and permanent wilting point)
Clay content	Clay	%	Average clay content (or fine textured soil particles) of the top soils in the plot
Silt content	Silt	%	Average silt content (or medium textured soil particles) of the top soils in the plot
Sand content	Sand	%	Average sand content (or coarse textured soil particles) of the top soils in the plot
Water use efficiency	WUE	kg m^{-3}	Amount of yield per unit volume of the amount of water received in the plot
Biomass	Bio	t ha^{-1}	Total biomass above the ground when the crop had matured

Table 2.3. List of classes describing the environmental conditions and crops used in the analysis

Environmental factors	Remarks	Class range	Name
Climate (MAP, mm year^{-1}; MAT, °C year^{-1})	Hot and wet	MAT>20 MAP>1000	Tropical
	Warm and arid-humid	MAT: 10-20 MAP:<100-1110	Subtropical
	Cool and arid-moist	MAT<10 MAP: 120-1000	Temperate
Clay (%)	Average clay content of the top soil horizon	0-20 20-35	Low Medium
Sand (%)	Average sand content of the top soil horizon	0-25 25-50	Low Medium
Silt (%)	Average silt content of the top soil horizon	0-20 20-40	Low Medium
Total water (TW, mm)	Amount of water received by a plot (sums of precipitation and irrigation water applied)	0-500 500-1000	Low Medium
Crop type	Field grains crops	wheat, maize, sorghum, millet, barley	Grain
	Field legume crops	Beans, peas	Legume
	Field oil seed crops	linseeds, mustard, safflower, sunflower, nuts	Oilseed
	Field fibre crops	cotton	Fibre
	Field fodder crops	alfalfa, energycane, giantreed, elephant grass	Grass
Growing season	Field crops grown under the different seasons	wheat, barley	Winter
		Maize, sorghum, pearl millet	Summer

Some of the factor classes were adapted from Mutema et al. (2015)

2.2.3. Data analyses

Descriptive statistics (minimum, maximum, median, mean, SEM: standard error of mean, quartile 1 and quartile 3 representing 25th and 75th percentiles, respectively, skewness (Skew), kurtosis (Kurt) and coefficient of variation (CV%) were calculated for all study variables. Mean WUE values were computed for different crops types, crop growing seasons and environmental factor classes. Significant differences between factor class values were tested at $p<0.05$ using nonparametric test (Statistica 10.0) because skewness and kurtosis statistics suggested the datasets were not normally distributed. In addition, one to one Spearman rank correlations and principal component analysis (PCA) for multiple correlations were performed. PCAs convert non-linear factors and variables into linear combinations called principal components (Jambu, 1991).

2.3. Results

2.3.1. General statistics of environmental variables and WUE

The type of environment and climatic conditions prevailing in a particular area varied widely, with MAP (mean 420±13 mm y^{-1}) ranging from 0 mm y^{-1} in Egypt where sorghum production was under sole irrigation (Hussein and Alva, 2014) to 1 472 mm y^{-1} in India (Kar et al., 2007) (Table 2.4). The lowest irrigation was 12 mm applied to wheat in Turkey (Tari, 2016), while the highest amount was 3381 mm to sorghum in Egypt (Hussein and Alva, 2014). The average supplementary irrigation was 569±55 mm. MAT (mean=17.3±0.3°C y^{-1}) showed less variation than MAP (CV of 75 and 38% for MAP and MAT, respectively) and ranged from 6.20°C y^{-1} in Turkey (Kuslu et al., 2010) to 29.0°C y^{-1} in Niger (Sivakumar and Salaam, 1999). Relative humidity (RH, 61.2±0.5%) showed even lower variation (CV=11%) ranging from 48.4% in Syria (Hussein et al., 2010) to 81.5% in Argentina (Abbate et al., 2004). Soil bulk density (BD, 1.39±0.01 g cm^{-3}) ranged from 1.16 g cm^{-3} in Damascus, Syria (Hussein et al., 2010) to 1.65 g cm^{-3} in Niger (Sivakumar and Salaam, 1999). Sand content (37.2±2.4%) exhibited the highest variation amongst the textural properties (CV=82%) with a minimum value of 2.3% found in China (Su et al., 2006) and a maximum of 91% in Niger (Sivakumar and Salaam, 1999). Soil clay content (mean=24.6±1.1%; CV=60%) varied from 3.24% in Saudi Arabia (Al-Neem, 2008) to 68.9% in Turkey (Onder et al., 2009). The lowest soil organic carbon content (SOCc) was 0.2% for Niger (Sivakumar and Salaam, 1999) and the highest was 1.4% in Malawi (Teravest et al., 2015), with a mean of 0.66±0.1%.

The mean WUE was 2.03±0.11 kg m^{-3} with values ranging from 0.03 kg m^{-3} for Bambara groundnuts in South Africa (Chibarabada et al., 2015) to 14.8 kg m^{-3} for maize in Ethiopia (Meskelu et al., 2014) (Table 2.4). Aboveground biomass (mean=1191±63 g m^{-2}) varied from 6.30 g m^{-2} for wheat in Argentina (Abbate et al., 2004) to 5950 g m^{-2} for maize in Zimbabwe (Rusere et al., 2012). Plant population (107±32 plants m^{-2}) varied widely (CV 199%), with maize in China being planted at 3.70 plants m^{-2} (Gao et al., 2008), while alfalfa in Turkey was densest at 1000 plant m^{-2} (Kuscu et al., 2012). Fertilizer usage also varied with N application rate (145±6 kg ha^{-1}) ranging from 36 kg ha^{-1} in Egypt (Hussein and Alva, 2014) to 500 kg ha^{-1} for barley in Saudi Arabia (Al-Neem, 2008).

Table 2.4. Descriptive statistics of environmental factors and crop variables included in the global database

Variables	n	Mean	Median	Min	Max	Qt 1	Qt 3	SEM	Skew	Kurt	CV%
WUE	548	2.03	1.16	0.03	14.8	0.66	2.09	0.11	2.55	6.92	124
Biomass	301	1191	867	6.30	5950	395	1671	62.5	1.50	2.46	91
MAP	548	420	370	0.00	1472	142	575	13.4	1.17	1.72	75
MAT	548	17.3	17.5	6.20	29.0	11.9	21.9	0.28	0.04	-1.14	38
Z	548	686	608	4.00	1835	93.0	1146	22.7	0.27	-1.11	77
RH	174	61.2	59.5	48.4	81.5	56.8	64.4	0.50	0.62	0.33	11
BD	263	1.39	1.40	1.16	1.65	1.20	1.53	0.01	-0.04	-1.23	11
Clay	168	24.6	27.2	3.24	68.9	16.8	30.9	1.14	0.86	1.87	60
Sand	168	37.2	34.8	2.30	91.0	3.50	69.2	2.35	0.37	-1.29	82
Silt	168	36.3	28.0	3.40	82.5	15.6	65.6	1.78	0.24	-1.27	64
Soil pH	128	7.66	8.06	4.22	8.75	7.70	8.30	0.11	-1.78	1.98	16
SOM	158	1.70	1.55	0.34	5.50	0.53	2.15	0.10	1.53	2.49	72
SOCc	33	0.66	0.34	0.20	1.40	0.34	1.03	0.08	0.46	-1.30	67
SONc	120	0.17	0.11	0.00	1.25	0.10	0.11	0.03	3.16	8.43	181
Soil P	142	313	15.0	2.90	950	13.4	950	36.6	0.79	-1.39	139
Soil K	85	286	176	75.0	544	165	544	19.5	0.46	-1.51	63
Population	370	107	10.26	3.70	1000	7.14	42.1	11.1	2.80	8.04	199
Plot size	444	189	33.6	1.00	6400	21.0	225	29.6	9.04	87.7	330
Precipitation	425	252	240	0.00	1169	73.0	330	10.7	1.68	3.98	87
Irrigation	447	569	306	12	3381	150	518	55.1	5.28	28.3	205
Total water	548	660	445	0.00	7960	295	645	45.3	5.48	32.2	161
Water used	109	460	432	3.60	1304	272	593	28.5	0.95	1.06	65
ETc	268	530	472	37.9	1886	363	652	14.7	1.43	3.99	45
AWC%	306	16.7	16.0	6.32	26.0	12.5	22.5	0.32	0.17	-0.85	34
FC%	282	29.9	32.0	12.7	51.3	24.0	35.0	0.52	0.22	-0.05	29
WP%	264	13.4	11.2	6.00	26.3	9.00	17.5	0.34	0.71	-0.63	41
Zd	168	1.33	1.20	0.50	2.00	0.90	1.83	0.04	-0.06	-1.51	40
LAI	53	13.7	3.70	0.80	103	2.70	4.78	3.55	2.37	4.40	189
N rate	302	145	113	36	500	87.1	174	5.88	1.71	2.90	70
P rate	270	81.2	60.0	8.40	400	31.00	100	4.66	2.46	7.04	94
K rate	86	109	75.0	20.0	840	60.0	100	17.9	4.07	15.8	152

Min: minimum, Max: maximum, Qt1 and Qt3: quartile 1and quartile 3, respectively, SEM: standard error of mean, Skew: skewness, Kurt: kurtosis, CV%: coefficient of variation, MAP=mean annual precipitation (mm yr^{-1}); MAT=mean annual air (^{o}C); Z=altitude (masl); RH%=relative humidity (%); BD=top soil bulk density (g cm^{-3}); Clay, Sand and Silt for top soil clay, sand and silt content (%); Population=plant population (plants m^{-2}); Irrigation=amount of irrigation water applied (mm); Total water= Sum of precipitation and irrigation (mm),Water used= amount of water used by crop (mm); ETc=Crop evapotranspiration for the entire crop cycle (mm); AWC%=available water capacity of soils (%); FC=field capacity (%),WP=permanent wilting point (%); Zd=rooting depth of crops (m); LAI=leaf area index; N, P and K rate=application rate of N, P and K (kg ha^{-1}); Biomass=above ground biomass (g m^{-2}); WUE=water use efficiency (kg m^{-3})

2.3.2. Impact of crop type on WUE

Grain crops had the highest WUE amongst the cultivated crops studied (Figure 2.1). Their median WUE (1.48 kg m^{-3}) was not significantly different from non-cultivated grasses (1.24 kg m^{-3}). WUE decreased from grain to legume (0.80 kg m^{-3}), oilseed (0.61 kg m^{-3}) and to fibre crops (0.33 kg m^{-3}), with all differences significant except between oilseed and fibre crops. Natural grass (median=3.60 kg m^{-3}) had 1.5 and 2-fold greater WUE than sorghum and maize, respectively (Figure 2.2). Maize and sorghum WUE were significantly higher than barley (1.17 kg m^{-3}) and wheat (1.15 kg m^{-3}). Pearl millet had the lowest WUE (0.51 kg m^{-3}) amongst the grain crops.

Beans (1.15 kg m^{-3}) and alfalfa (1.07 kg m^{-3}) had similar WUE to wheat. However, both had significantly higher WUE than sunflower (0.77 kg m^{-3}). In turn, sunflower had significantly higher WUE than peas (0.52 kg m^{-3}), cotton (0.33 kg m^{-3}), linseeds (0.25 kg m^{-3}), safflower (0.21 kg m^{-3}) and mustard (0.17 kg m^{-3}). Overall, Bambaranuts had the lowest WUE of 0.005 kg m^{-3} (Chibarabada et al., 2015).

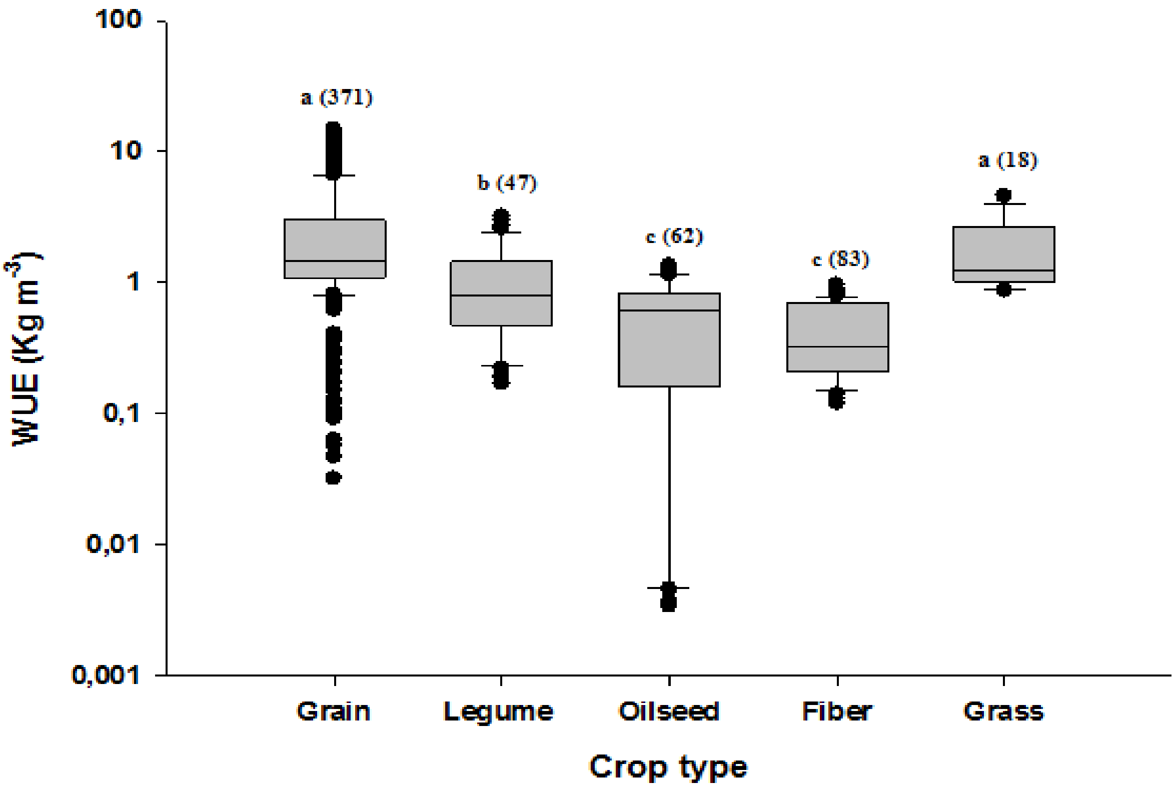


Figure 3.1. Box plots comparing WUE (kg m^{-3}) variations among field crop types/categories used in the analysis.

Box plots accompanied by similar letters were not significantly different at <0.05. Numbers between brackets are the sample sizes. Y-axis is in logarithmic scale.

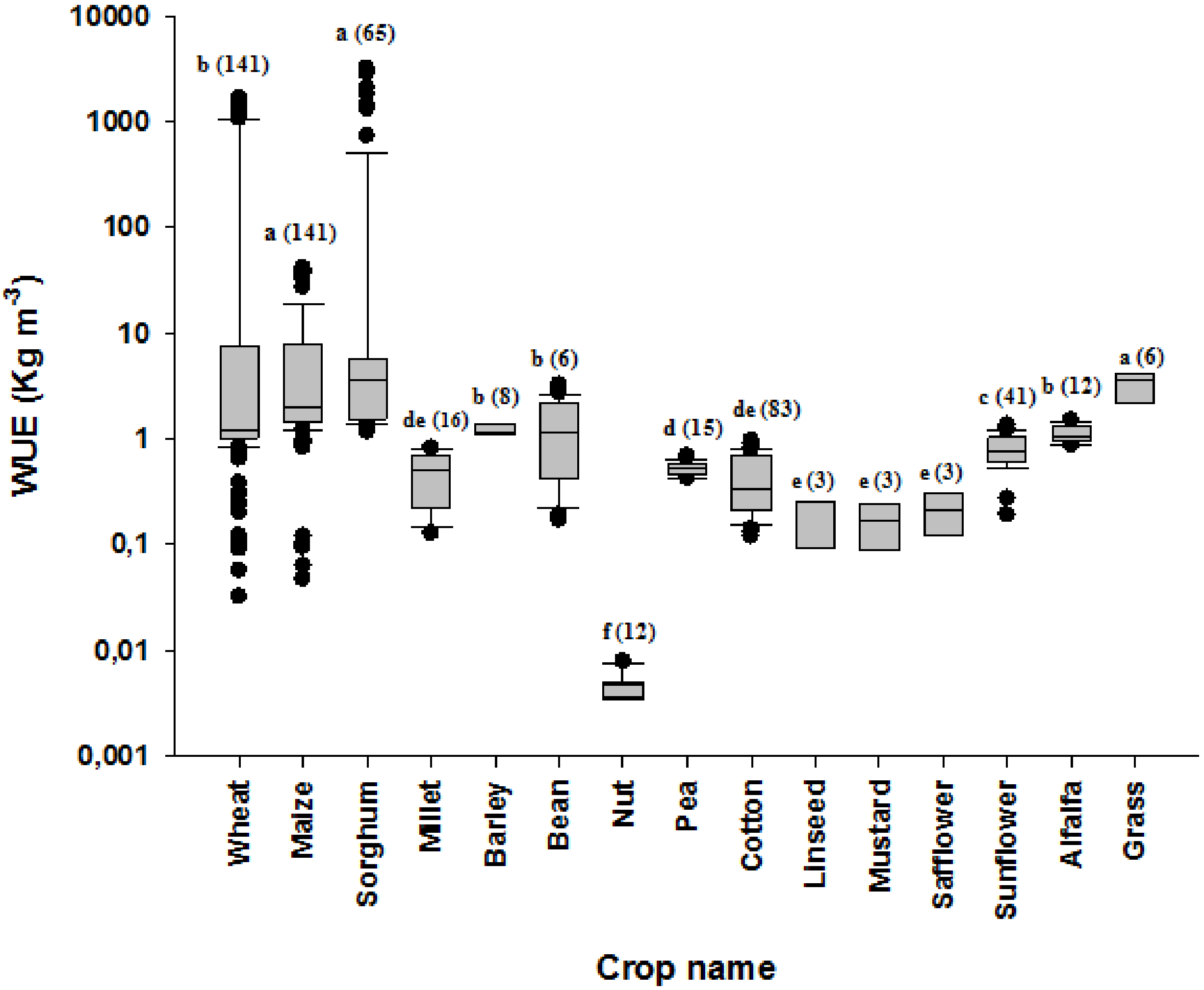


Figure 3.2. Box plots comparing WUE (kg m^{-3}) variations among the different field crops used in the analysis.

Box plots accompanied by similar letters were not significantly different at <0.05. Numbers between brackets are the sample sizes. Y-axis is in logarithmic scale.

2.3.3. Impact of environmental conditions on WUE

2.3.3.1. Climate

Growing season showed significant effect on WUE with summer crops having higher WUE (1.72 kg m^{-3}) than winter crops (1.15 kg m^{-3}) suggesting that growing crops off-season might decrease WUE by over 30% (Fig 2.3). Figure 2.4 confirms that WUE increase with MAP from desert (median 1.08 kg m^{-3}) to subtropical (1.34 kg m^{-3}) and to tropical climate (1.83 kg m^{-3}). The increase from desert to subtropical climate was 24%, while that from subtropical to tropical climate was 37%.

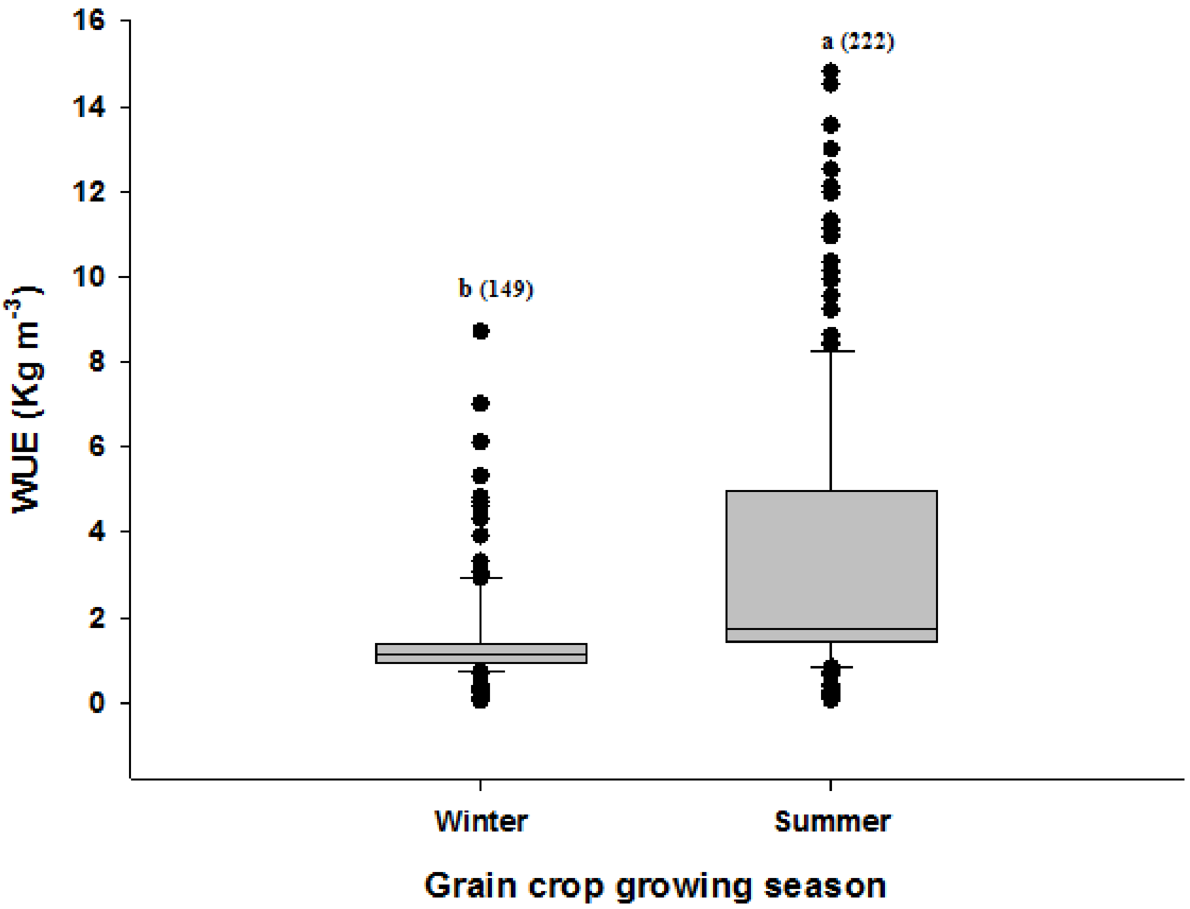


Figure 3.3. Box plots comparing WUE (kg m^{-3}) variations for field grain crops grown in winter and summer used in the analysis.

Box plots accompanied by similar letters were not significantly different at $p<0.05$. Numbers between brackets are the sample sizes.

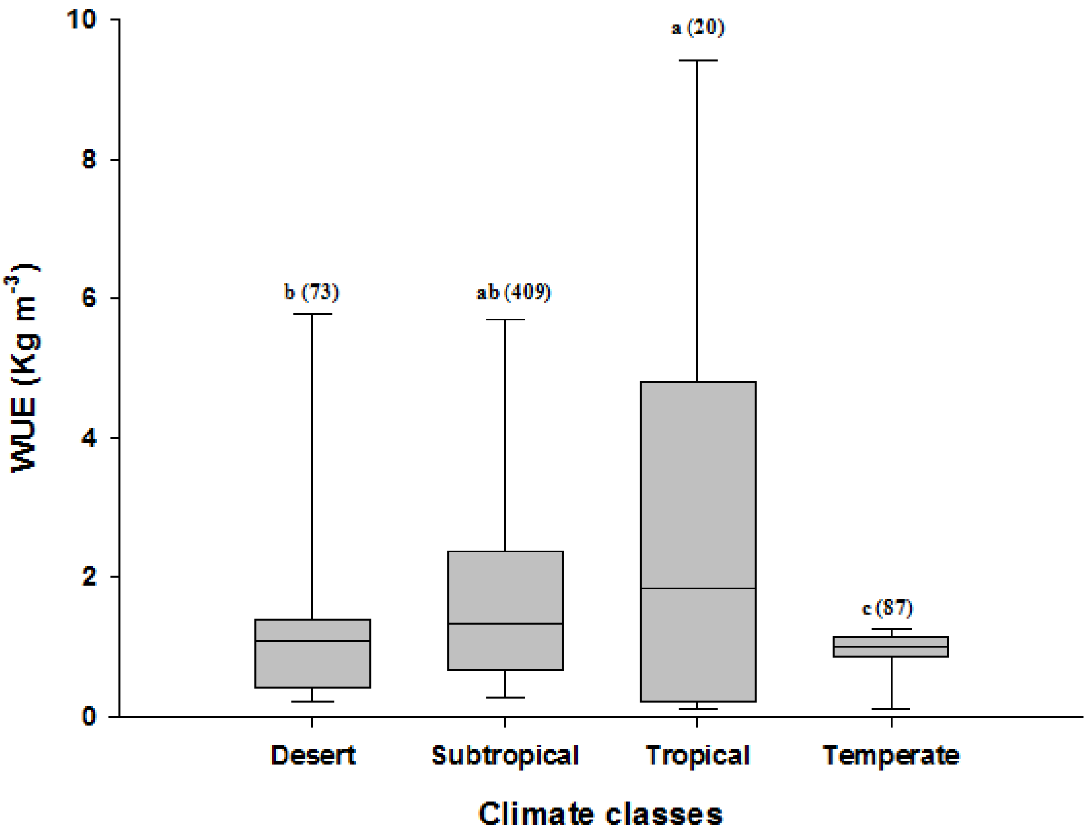


Figure 3.4. Box plots comparing WUE (kg m^{-3}) variations for field crops under different climatic regions.

Box plots accompanied by similar letters were not significantly different at $p<0.05$. Numbers between brackets are the sample sizes.

Sorghum had the highest WUE (5.60 kg m^{-3}) followed by maize (1.21 kg m^{-3}) and barley (1.20 kg m^{-3}) under desert conditions (Fig 2.5A). However, maize WUE (3.92 kg m^{-3}) was not significantly different from sorghum (3.78 kg m^{-3}) under subtropical conditions (Fig 2.5B). Surprisingly, wheat and beans were also not significantly different under the subtropical conditions, but peas (0.51 kg m^{-3}), pearl millet (0.47 kg m^{-3}) and cotton (0.45 kg m^{-3}) showed significantly lower WUE. Maize had significantly higher WUE (8.37 kg m^{-3}) than grass (3.37 kg m^{-3}) in the tropical climate (Fig 2.5C). However, grass WUE was still higher than mustard (0.71 kg m^{-3}), safflower (0.21 kg m^{-3}) and linseed (0.20 kg m^{-3}). Temperate climate had fewer crop types than other climates (Fig 2.5D), and grass had highest WUE (3.23 kg m^{-3}) followed by wheat (0.97 kg m^{-3}) and lowest was maize (0.9 kg m^{-3}).

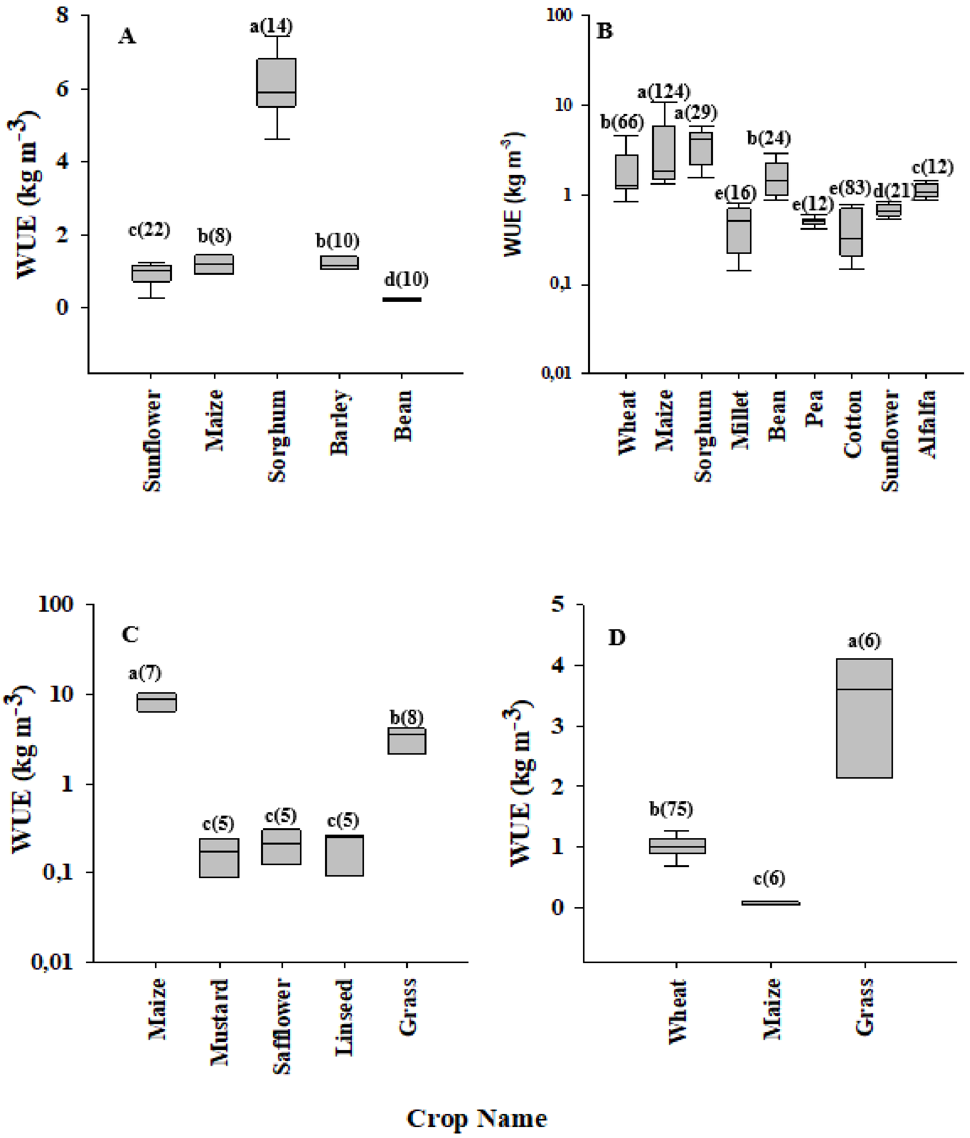


Figure 3.5. Box plots comparing WUE (kg m^{-3}) variations amongst field crops grown in different environments (A) Desert, (B) Subtropical (C) Tropical and (D) Temperate.

Box plots accompanied by similar letters were not significantly different at $p<0.05$. Numbers between brackets are the sample sizes.

2.3.3.2. Water received

While amount of precipitation had significant effect on WUE (r_s=0.16) and biomass (r_s=0.13), supplementary irrigation showed significant effect on biomass only (r_s=0.27) (Table 2.**5**). Subsequently, total water (precipitation plus irrigation) received showed significant effect on biomass only (r_s=0.28). WUE decreased significantly from a lowly watered regime (median=1.17

kg m^{-3}) to a moderately watered regime (1.08 kg m^{-3}) (Figure 2.6). Unexpectedly, a fully watered regime had the highest WUE (1.60 kg m^{-3}); however, it was not significantly different from the regime receiving lower amount of water. The fully watered regime resulted in 37 and 48% higher WUE than lower and moderately watered regimes, respectively. However, the lowly watered regime had highest variability in WUE (CV=133%) followed by fully watered (CV=79%), and least moderately watered regime (CV=72%).

Table 3.1. Spearman rank correlation coefficients between selected controlling factors and crop variables

	Biomass	WUE
MAP	0.21*	0.19*
MAT	0.05	-0.09*
Altitude	0.11*	0.01
RH%	-0.16	0.23*
Soil bulk density	-0.08	-0.25*
Clay content	0.28*	0.20*
Sand content	-0.09	-0.11
Silt content	-0.07	-0.01
Soil pH	0.48*	-0.04
OM	-0.03	0.17*
SOCc	nd	0.77*
SONc	-0.24*	0.06
Soil P	0.18	0.56*
Soil K	0.55*	0.41*
Plant population	-0.08	-0.16*
Plot size	-0.51*	0.04
Precipitation	0.13*	0.16*
Irrigation water applied	0.27*	0.04
Total water	0.28*	-0.01
Water used	0.71*	-0.06
AWC%	-0.47*	-0.16*
FC%	-0.17*	0.07
WP%	0.20*	0.03
Root depth	0.40*	-0.25*
LAI	0.51*	0.14
ETc	0.13	0.09
N application rate	0.06	0.00
P application rate	0.28*	0.03
K application rate	-0.03	-0.14

* significant at $p < 0.05$, ** significant at $p < 0.01$., nd not determined due to inadequate data. MAP=mean annual precipitation; MAT=mean annual air temperature; RH%=relative humidity of the atmosphere; OM=soil organic matter content; SOCc=soil organic carbon content; SONc=soil organic nitrogen content; Soil P=soil phosphorus content; Soil K=soil potassium content; Total water=some of rainfall and irrigation water applied to a crop; AWC%=available water capacity of the soil; FC%=field capacity; WP%=permanent wilting point; LAI=leaf area index; ETc=evapotranspiration of the crop

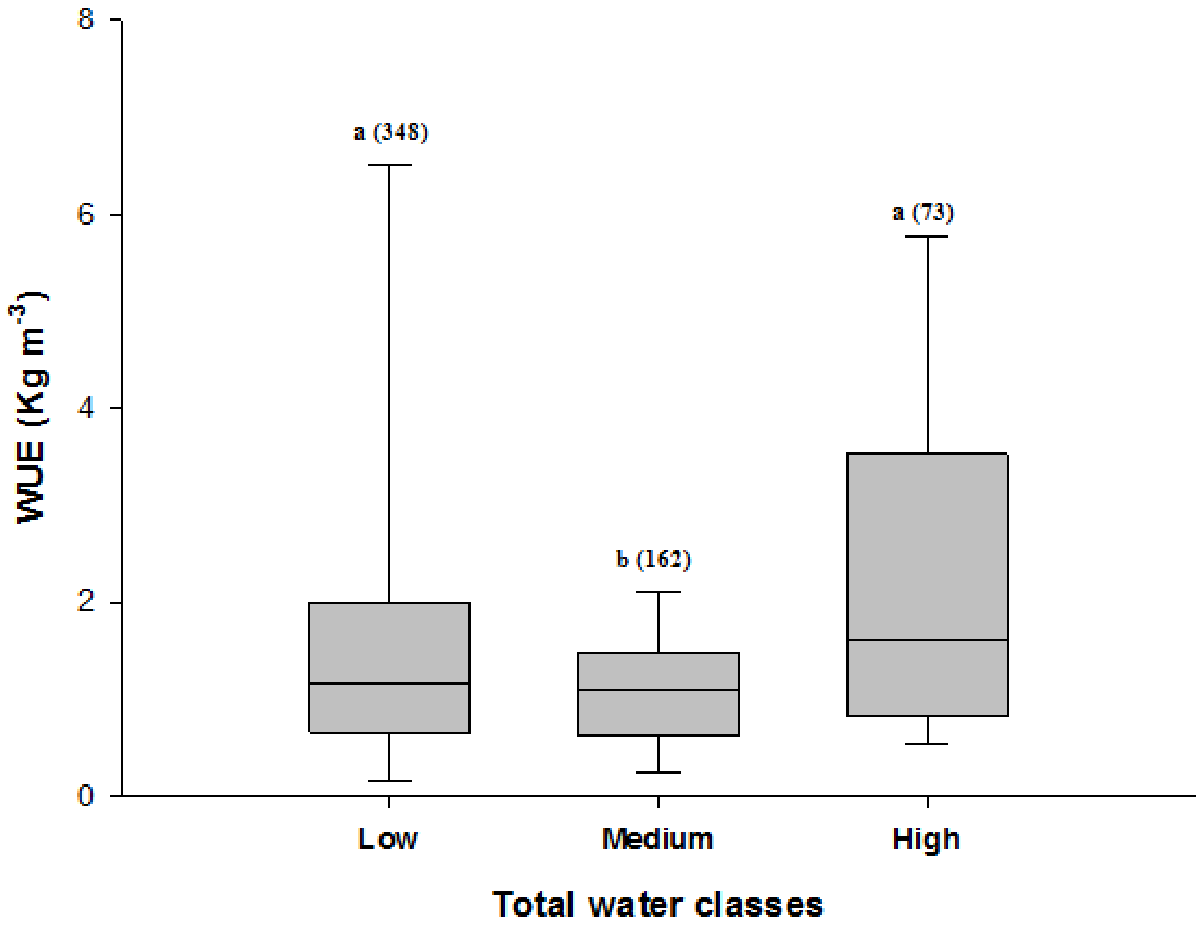


Figure 3.6. Box plots comparing WUE (kg m^{-3}) variations for different field crops under three different total water classes.

Box plots accompanied by similar letters were not significantly different at $p<0.05$. Numbers between brackets are the sample sizes.

2.3.3.3. Soil texture

Clay content was the only textural property that exhibited significant effect on both WUE (r=0.20) and biomass (r=0.28) (Table 2.5). The other soil physical properties exhibited moderate effects with, for example, bulk density showing negative effect on WUE only (r=-0.25). Box plots showed significant effects of soil texture on WUE (Figure 2.7). Thus, average WUE of field crops in general was highest under silt loam (1.43 kg m^{-3}) and clay loam soil (1.27 kg m^{-3}). However, no significant difference was observed between loam and clay soil.

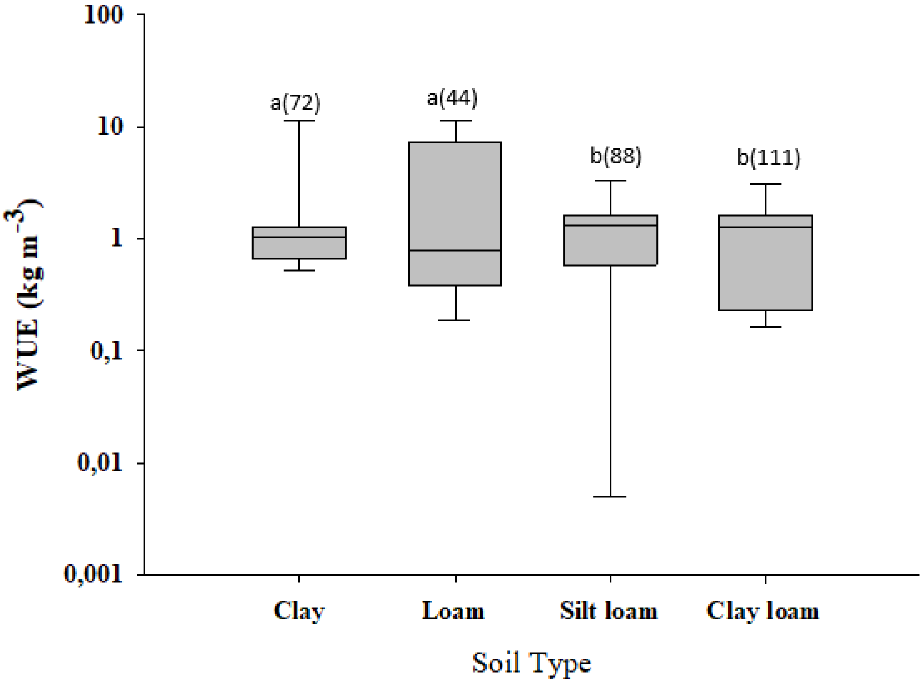


Figure 3.7. Box plots comparing field crops WUE (kg m^{-3}) variations under different soil types.

Box plots accompanied by similar letters were not significantly different at $p<0.05$. Numbers between brackets are the sample sizes. Y-axis in logarithmic scale

Figure 2.8 indicates that maize had the most significant WUE on clay (10.67 kg m^{-3}, Fig 2.8A), loam (9.47 kg m^{-3}, Fig 2.8B) and clay loam soils (2.31 kg m^{-3}, Fig 2.8D). While safflower (0.21 kg m^{-3}) linseed (0.20 kg m^{-3}) and mustard (0.18 kg m^{-3}) had much lower WUE than cotton on loam soil. Wheat on the other hand had the second most significant effect on WUE in clay

(1.18 kg m^{-3}) and loam soil (1.18 kg m^{-3}); but had the most significant WUE on silt loam soil (3.04 kg m^{-3}, Fig 2.8C). Sorghum (1.48 kg m^{-3}) and maize (1.43 kg m^{-3}) were not significantly different on silt loam soil. While cotton had the least WUE (0.28 kg m^{-3}) on clay loam soil (Fig 2.8D).

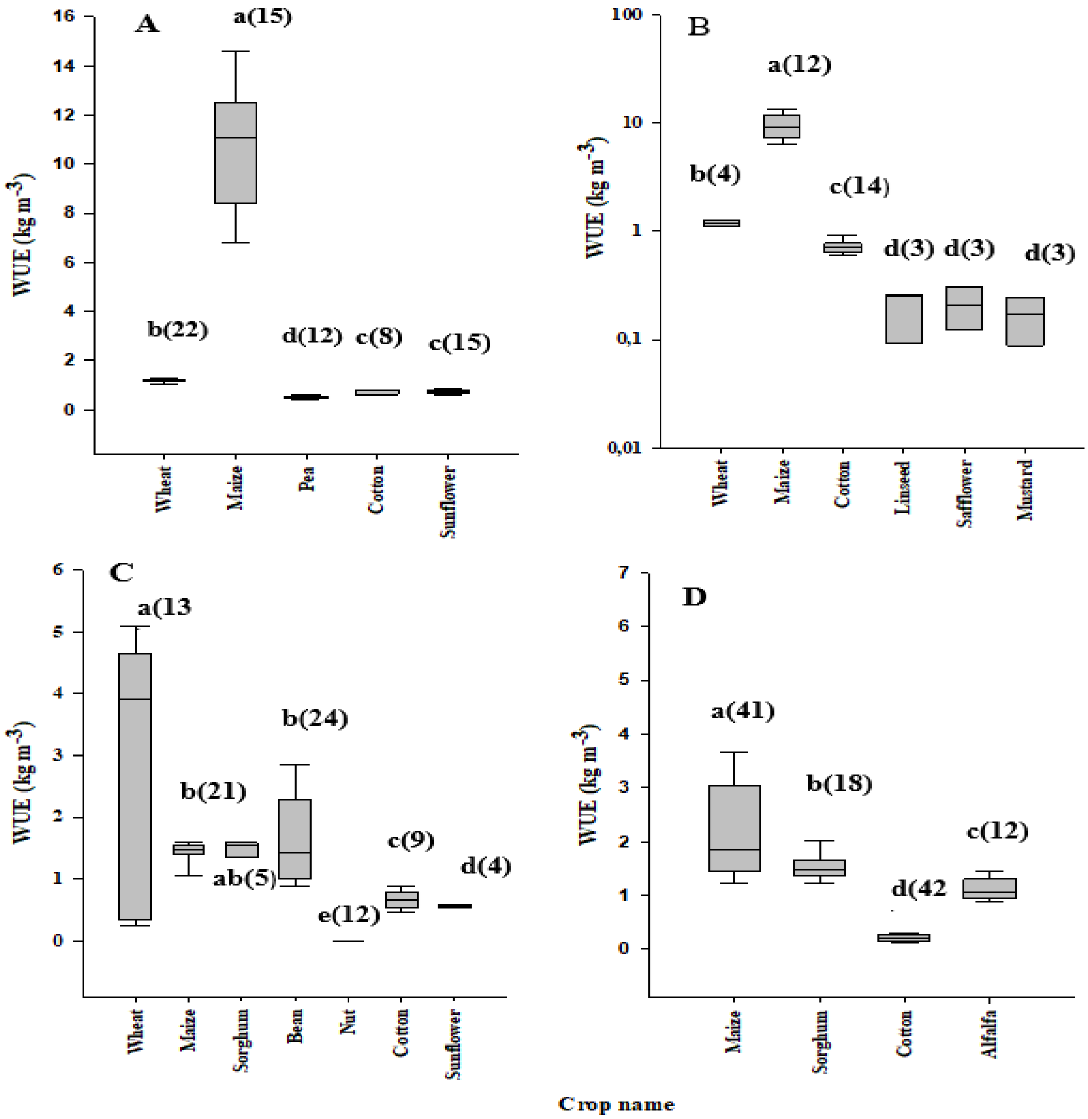


Figure 3.8. Box plots comparing WUE (kg m^{-3}) variations for field crops under different soil types (A) Clay soil, (B) Loam soil, (C) Silt loam soil and, (D) Clay loam soil.

Box plots accompanied by similar letters were not significantly different at $p<0.05$. Numbers between brackets are the sample sizes

2.3.4. Correlations between environmental factors and WUE

Five principal components accounted for 50.44% of the total variation. The first two principal components with PC1 and PC2 accounting for 26.54 and 23.9%, respectively (Fig 2.9). Although many factors were studied and had influence on crop WUE, only a few factors with positive correlation with WUE were included in the PCA (Table 2.5). PC1 was closely associated with total water and MAT on the positive coordinates (Fig 2.9). On the other hand, PC2 was closely associated with clay and MAP with both factors exhibiting positive coordinates. This PC represented the axis of increasing soil moisture. Therefore, the results of this PCA implied that WUE increased with soil moisture and temperature. Clay and SOC contents were also important promoters of higher WUE. These associations are in general agreement with the Spearman rank correlation results in Table 2.5.

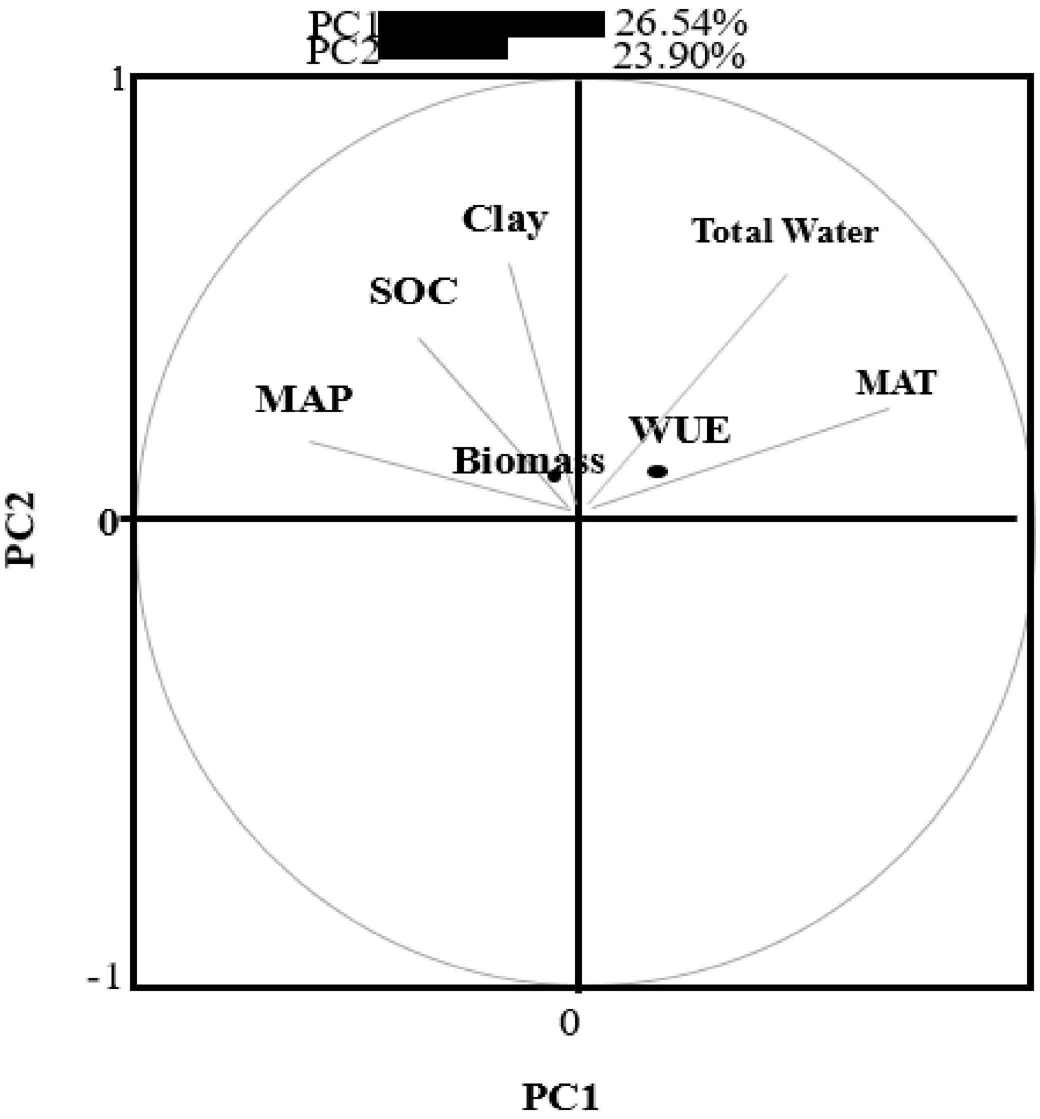


Figure 3.9. Principal component analysis of selected environmental factors and the water use efficiency parameters (Biomass and WUE).

Environmental factors were used as variables for analysis while the water use efficiency parameters were the supplementary variables

2.4. Discussion

2.4.1. Impact of crop type on WUE

Water use efficiency (WUE) varied with crop type, with our results indicating that maize and sorghum tended to have higher WUE than other members of the grass family such as wheat. This may be explained by differences in the genetic make-up of the crops. These two groups have different photosynthetic pathways linked to the way they evolved genetically leading to plants being classified as either C4 or C3 types. C4 plants (which include sugarcane, maize, sorghum and millet) originated from subtropical areas, while C3 plants (e.g. wheat, barley and rice) populated a far broader range of climates (Yamori et al., 2014). Studies have generally shown that C4 plants tend to be more water use efficient than C3 species under both natural and managed ecosystems (Sage and Manson, 1998; Zwart and Bastiaanssen, 2004). The higher WUE in C4 plants is attributed to increased CO_2 concentration mechanisms in their special bundle sheath cells, which allow higher photosynthetic rates while reducing stomatal conductance. This reduced stomatal conductance reduces transpiration, resulting in water savings (Sage and Manson, 1998). However, whether C4 are more water use efficient than C3 plants or do not appear to be controlled by the growing environment. Hence, C4 plants tend to outperform C3 plants in photosynthetic prowess under hot and dry conditions, while C3 plants perform better under sufficient water and sunlight conditions.

The growing conditions for the studies used in the current analysis appear to have favoured C4 plants more than the C3 plants. Though not significantly different, sorghum and maize had higher WUE than most crops, which agree with results from other studies. Katerji and Mastorilli (2014) for example, found higher WUE in sorghum than other major grain crops including maize, wheat and barley in a Mediterranean environment. The impact of growing environment may also explain the low WUE for pearl millet, a C4 plant type, which was even lower than wheat WUE. Pearl millet is one of the least popular grain crops worldwide but is highly adapted to hot and dry conditions, where research efforts are generally limited. Hence, research related activities around this crop tend to occur in environments less favourable for its optimum performance.

The current results also showed that grain crops had the highest WUE, which was largely anticipated because the group was dominated by C4 crops (data points totalled 222 for maize, sorghum and millet *vs*. 149 for wheat and barley). The WUE tended to decrease from grain crops, to legumes, oilseeds and to fibre crops, which was also expected because other studies elsewhere showed a similar trend. Angadi et al. (2008), whilst evaluating WUE of different crops, found that wheat (grain crop) had the highest WUE followed by pulses (legumes) with the oilseeds having the lowest WUE. Legumes are C3 plants and would be expected to have lower WUE than grain crops dominated by C4 plants. It is also generally known that yield per unit area tends to decrease as crops are improved for such properties as high protein; oil and fibre production, which might explain the general decrease because WUE was on the basis of yield. Thus, the very low WUE for nuts would not be surprising because they are generally improved for either protein or oil content. Another possible explanation for the low nut WUE could be errors when normalising young crop WUE data to full maturity basis. The WUE data for nuts mostly came from young plants in the early stages of the growth cycles. The lack of significant difference between oilseeds and fibre crops may be attributed to higher variability of WUE under the oilseeds.

The result that summer grain crops had significantly higher WUE than winter grain crops was reflective of the performance of C4 vs. C3 plants. The summer crop group consisted of maize, sorghum and millet which are C4, while winter crops comprised wheat and barley which are C3. Summer conditions might also be more conducive for crop productivity especially in the southern hemisphere where summers are characterised by relatively higher temperatures, soil moisture and more sunshine hours. Southern hemisphere winters are generally cooler and drier with less sunshine hours. However, high variability of WUE for both groups might be indicative of the existence of other critical controls of WUE. Thus, the impacts of selected environmental controls were also elucidated.

2.4.2. Impact of environmental conditions on WUE

2.4.2.1. Climate

Climate is very crucial for growth, as it largely determines whether crops would be successful or not in a particular environment. Climatic factors (MAP and MAT) showed significant effects on WUE with a tendency for WUE to increase from desert-like environments to subtropical and tropical zones. There was no significant difference between WUE in tropical and subtropical environments. This is mainly due to the greater number of crops favoured by these environments compared to desert and temperate environment. Maize and sorghum exhibited higher WUE in desert, subtropical and tropical environments. This was due to the fact that both crops are C4. Thus, they have higher drought tolerance and WUE compared to other crops (Chipanshi et al., 2003). The increase of WUE from desert to subtropical and tropical environments can be explained in terms of high temperature and soil moisture stresses, which decrease with change of environmental conditions. High temperature stress on crops is likely to be highest in the deserts due to high temperatures and low humidity (Sherwood and Huber, 2010; Tardieu, 2013). These drive soil moisture losses through increased evaporation. The combined effect of these two factors depresses crop WUE (Shah and Paulsen, 2003; Boutraa et al., 2010; Tardieu, 2013) due to subdued yields. Heat and soil moisture stress generally decrease with the transition towards the tropics due to a general increase of precipitation which raises humidity. Though subtropical and tropical regions may be characterised by high average air temperatures, higher precipitation incidences tend to increase soil water availability and humidity which results in a number of crops being favoured by these environments. This school of thought is supported by the PCA results that showed positive and negative association of WUE with MAT and LAT (latitude), respectively. It is also clear in the same figure that WUE tended to increase with precipitation (MAP), soil moisture (AWC), soil clay and carbon content. The temperate climate produced field crops that had the least variation in WUE (CV 42%), than the other climates (119, 120 and 114% for desert, subtropical and tropical climate, respectively). This was also confirmed by the negative association between WUE and latitude and by the fewer crops found under this environment. Despite relatively high soil moisture levels due to low air temperatures in the temperate regions, most crop growth cycles are quite long due to low metabolic rates, (Hendrickson et al., 2004) yet yields seldom outperform warmer regions by significant margins. Since WUE used in the current study was based on yield, it therefore means that it is bound to be much lower in temperate than warmer regions where plant growth is faster. Of course, the foregoing assumes good management practices for optimal production in all regions. It is also appreciated that WUE only increase up to peak levels dependent on crop type and variety (Tang et al., 2014), provided heat and soil moisture stress are kept low, and management practice enhances attainment of maximum potential. It was also interesting to note that field crops produced in the desert climate characterised by generally dry soils and high air temperatures still had significantly higher WUE than crops produced in the temperate climate.

2.4.2.2. Amount of water supplied

The results suggested that, regardless, of crop type and environmental conditions such as air temperature and soil type, crop WUE tends to be significantly higher in both low and high watering regimes than moderate watering regime. This result was surprising because low watering was expected to result in lower WUE than moderate watering because moisture stress was expected to be higher. However, deliberate moisture stress in crop production can be beneficial depending on its timing during the growth cycle. For example, moisture stress in the early growth stages of cereals is known to enhance deep rooting (Çakir, 2004; Chemura et al., 2014), essential for anchorage and reaching out to deeper horizons for moisture and nutrients. The high variation of WUE in the low water category suggests that some crops indeed benefit from moisture stress. However, the most ideal practice remains the supply of adequate water complemented by good management practices for optimal growth and yield levels. Our results are consistent with findings by other studies. For instance, Chibarabada et al. (2015), when evaluating the effect of water regime on groundnut WUE, reported higher WUE under stressed than well-watered conditions. However, Boutraa et al. (2015), examining the effect of moisture stress on wheat WUE in Saudi Arabia found higher WUE in well-watered than moisture stressed wheat. Therefore, the impact of watering regime on WUE, indeed, also depends on other factors including crop type (Chibarabada et al., 2015).

2.4.2.3. Soil texture

Texture is one of the most important soil properties which affects WUE and crop production (Mojid et al., 2012). Soil clay content had some impact on crop WUE indicating that high clay soils tends to depress crop yields. In clayey soil, maize had highest WUE than all crops, mainly because it does well in most soil types (Bennetzen and Hake, 2008). High clay content soils promote waterlogging, which has a negative effect on crop productivity due to poor soil respiration (Greenway et al., 2006; Morales-Olmedo et al., 2015). Moreover, high soil water levels in high clay content soils do not always result in high water availability (Reichert et al., 2009). Poorly drained soils may also promote proliferation of fungal diseases. In addition, high clay content presents other challenges to crop production such as crusting and restrictions on root development, which are detrimental to crop yield. Most crops exhibited high WUE under loam, silt clay loam and clay loam soil compared to clay soil, even though silt loam and clay loam soil had lower WUE in general. WUE of wheat was higher in both silt loam and loam soil compared to clayey soil, as loam textured soils are said to be highly suitable for growing wheat and other crops due to good aeration, drainage and water holding capacity (Russell, 2002). Katerji et al. (2009) also found out that WUE of potato, maize, sunflower and sugar beet was significantly higher in a loam soil compared to a clay soil.

2.5. Conclusion

The study investigated the impacts of soil and environmental conditions on water use efficiency (WUE) of main field crops. The first main conclusion using data from 514 sites across the globe was that crops differed significantly in their abilities to efficiently use water for grain yield, with C4 plants such as maize exhibiting significantly higher WUE than C3 types (e.g. wheat). The second conclusion was climate had significant impact on water use efficiency with a tendency for warm and wet climates to yield higher crop WUE than cooler environments. The third conclusion was that soil texture significantly impacted crop WUE with lowest values occurring in clayey soils and highest values under loamy conditions. However, maize performed very well in all soil textures. There is still a certain level of unexplained variability in crop WUE that could potentially be attributed to factors not considered in the present study, which are seldom reported in literature on WUE such as seasonal variations of climate, land management and agronomic practices such as fertilization application, tillage and crop residue mulching. The knowledge generated in the framework of the present study is expected to provide useful insights for policy and decision making in the context of climate uncertainties. The results also suggest that more still needs to be understood about the main controls of field crop water use efficiency. For instance, there seems to exist many interactions between crop water and nutrient availability. Such interactions couldn't be investigated in the framework of the present study but present an important grey area for further research.

Chapter 3 WUE OF SORGHUM AND PEARL MILLET COMPARED AGAINST MAIZE AND WHEAT VARIETIES

Abstract

The need to increase water productivity in agricultural systems has intensified in recent years due to diminishing water resources and recurrent droughts. Food production is critically connected to water availability but the world is now facing intense conflicts on water use among agriculture, industry and human needs. One strategy to improve water productivity in agriculture and minimize the conflict is to identify crops and varieties with high water use efficiency biomass and grain yield production. Indigenous crops such as sorghum and pearl millet have been promoted as water use efficient crops compared to commodity crops such as maize and wheat but there are a few studies that comprehensively compared these four crops simultaneously. A meta-analysis was conducted using data from different countries to compare their biomass and grain yield production and water use efficiency under optimal and drought stressed conditions. The study included 700 observations on biomass and grain yield production and water use efficiency from studies across the world. The data was subjected to statistical analyses in Genstat and R softwares and the impact of drought stress on these parameters was calculated using MetaEasy software. The means for biomass production for maize, sorghum, millet and wheat were 24.0, 18.4, 14.6 and 8.1 t ha^{-1}, respectively, under optimal conditions. Similarly, grain yield production followed a similar trend with maize attaining 8.2 t ha^{-1} followed by sorghum (6.5 t ha^{-1}), millet (6.3 t ha^{-1}) and wheat (3.1 t ha^{-1}). The high biomass production potential of maize, sorghum and millet is supported by their C4 photosynthesis compared to wheat, which is a C3 crop. Drought stress had the highest impact on grain yield production on maize (52%) and wheat (43%) compared to sorghum (26%) and millet (25%). The meta-analysis showed that the effect size for drought on yield and biomass production potential were highest for maize (-0.6) followed by wheat (-0.55), showing that these commodity crops were highly sensitive to drought stress compared to sorghum and millet. Again, sorghum and millet were more efficient in water use, attaining more than 4.0 kg of grain m^{-3} of water used. In general, drought stress induced higher water use efficiency in crops although it significantly reduced biomass and grain yield production potential as signified by a negative association between water use efficiency and biomass traits. In conclusion, sorghum and millet are recommended for water-constrained ecologies to improve water use efficiency and food production.

Keywords: biomass production, drought stress, water productivity, water availability

3.1. Introduction

South Africa is a semi-arid country and is projected to experience below 1000 m^3 per capita freshwater availability by the year 2025 (Otieno and Ochieng 2004). Currently, 65% of the country is arid and receives less than 450 mm/year compared to the global average of 860 mm/year (Donnenfeld et al., 2018). The demand for water is expected to increase due to agricultural, industrial and population growth. This has led to proposals to increase extraction of underground water to supplement agriculture use, which consumes about 60% of all water in South Africa (WRC, 2002). However, water is a finite resource and should be replenished for sustainable agriculture. Projections show that water levels in 60% of dams are critically low and more than 66% of the rivers have been exploited with adverse consequences on ecological stability (IPCC 2014). The critical shortage of water and its potential catastrophic impact on food production and life demands that we develop strategies that minimize water losses in agriculture. Most agricultural systems around the world experience shortage of water and inefficient water use utilization (Mokgope et al., 2001). Water use efficiency in agriculture systems is estimated to be less than 30% (Eliasson et al., 2003), which is a huge loss in finite water resources that must be conserved to prevent a crisis in the near future.

Given the scarcity of water in South Africa and the world in general, it is important to maximize yield and biomass production in crops. The use of supplement irrigation in agriculture is common to mitigate water shortages. However, the applied water either by irrigation or rainfall should be used efficiently to produce food and biomass for feed or carbon retention in soil. Current projections show that improving efficiency in water use to maximize production holds much more promise to address food and water shortages than expansion of irrigation capacity because the water resources are already strained. Irrigation scheduling has been used to improve water use efficiency in agriculture but this is influenced by plant, soil and climatic factors (Katerji and Mastrorilli 2014), which must be included in crop models that seek to understand and improve water use in agriculture. Thus, it would be important to evaluate different crops and determine their water use efficiency for recommendation under different water scenarios. Water use efficiency is normally quantified in terms of primary productivity as biomass or yield for a unit of water applied. Generally, a linear relationship between productivity and water use efficiency has been reported although the relationship has been found to be more complex than that. For instance, optimized application of water was reported to achieve higher water use efficiency for equal amount of water compared to blanket application of water (de Lima et al., 2015; Du et al., 2010; Tambussi et al., 2007). This has led to the development of deficit irrigation and other strategies that seek to optimize water application at critical moments in the crop growth stages.

Most commercial commodity crops such as maize and wheat are increasingly cultivated under irrigation to increase production and meet food requirements. The diminishing water resources would not be able to cope and support such agriculture systems especially among smallholder farmers who have no provision for water abstraction (Chauke and Anim 2013; Shah et al., 2002). One strategy to improve water productivity on farms is to select crops or varieties with enhanced water use efficiency for biomass or grain yield. In sub-Sahara Africa, agro-ecosystems are dominated by cereals crops and it will be prudent to evaluate their relative water use efficiency. Traditional crops such as sorghum and millet have been reported to be more drought tolerant compared to maize or wheat. There are very few studies that have compared these under similar conditions, for instance wheat is predominantly a winter crop. Environmental conditions such as wind speed, temperature and relative humidity prevailing in winter are far more different in summer, and these have a strong bearing on water use in crops (Durand et al., 2020; Linderson et al., 2007). Thus, it would be difficult to establish a fair comparison of these. The existence of a number of studies across the world gives an opportunity to conduct a comprehensive meta-analysis to establish the impact of water availability on water use efficiency, biomass and yield production in different crops and obtain a global view of the dynamics of water use efficiency. Evaluating water use efficiency in traditional crops will improve information and extension services to promote their production and improve food and water security under communal farming systems. Thus, the study analysed a large dataset of water use efficiency in maize, millet, sorghum and wheat to draw comparisons on water use efficiency, biomass and grain yield production and the impact of drought on these parameters. The purpose of the analysis is to deduce quantitative information for advisory and extension provision to promote cultivation of traditional and indigenous crops in South Africa as a mitigation strategy for water scarcity.

3.2. Materials and methods

3.2.1. Data collection

The study collated data from peer reviewed publications obtained from academic databases. The papers were obtained by using search words and phrases that included “biomass production”, “grain yield production”, “water use efficiency”, “water productivity”, “sorghum”, “maize”, “wheat” “pearl millet” and “drought stress”, among others. The papers were screened and selected based on quantitative reporting of total biomass, grain yield and amount of water used per unit grain yield produced in maize, sorghum, millet or wheat. Priority was given to papers that compared water productivity for biomass and grain yield production under contrasting water scenarios of optimal and drought stressed conditions. Drought was defined as any level of water availability that was inadequate to support full potential for biomass and grain yield production in either of the four crops, and this definition was arbitrary for the purpose of this study only. Based on this criterion, 46 papers (Table 3.1) were selected and data was extracted accordingly and used to create a database with about 566, 617 and 707 observations for biomass, grain yield and water use efficiency, respectively. The database was subsequently subjected to statistical analysis as presented below.

3.2.2. Data analyses

3.2.2.1. Variation in biomass yield production and water use efficiency

The data variability was assessed by deriving summary statistics in Excel. The minimum, maximum and mean values for each parameter were deduced. In addition, the standard error, standard deviation, kurtosis and skewness were calculated. The difference between means for parameters measured under drought and optimal conditions were compared for significance using the t test in Genstat 18th edition (Payne et al., 2017). Further, the variability in the dataset was depicted by boxplots constructed in SigmaPlot 11.0 (Systat Software 2008). For each parameter, the boxplots presented the 5 and 95th percentile interval, 25, 50 and 75th quartiles and mean.

3.2.2.2. Trait-crop associations

The relationship among the parameters and their correlations with each crop were determined by the principal component analysis in R software (R Core Team 2020) using the “FactorMiner”

(Husson et al., 2016) and “factoextra” (Kassambara et al., 2017) packages. The relationships were depicted in principal component analysis biplot.

3.2.2.3. Determination of effect sizes

The effects of drought stress on biomass, yield production and water use efficiency were calculated by comparing the datasets and deducing the mean effect size using MetaEasy software v1.0.2 (Kontopantelis and Reeves 2009). The MetaEasy software assists in standardising the effect size and increases statistical precision and reduces bias in data by factoring in sample sizes (Kontopantelis and Reeves 2009). The Cochrane Collaboration (Stokes et al., 2015) was used in the meta-analysis of the data. The effect sizes were estimated within 95% confidence interval based on the Dersimonian-Laird (DL) random effects model that accounts for heterogeneous studies (DerSimonian and Laird 1986). An effect size whose cumulative value did not cross the zero line of the biplot was considered significant.

Table 3.1. List of studies used in meta-analysis showing the number of observations for each parameter

No.	Author	Country	Crop	Biomass	Grain yield	WUE
1	Abdel-Motagally (2010)	Egypt	Sorghum	18	18	18
2	Conley et al. (2001)	USA	Sorghum	8	8	8
3	Curt et al. (1995)	Spain	Sorghum	9	9	9
4	Dağdelen et al. (2006)	Turkey	Maize	10	10	10
5	Djaman and Irmak (2012)	USA	Maize		10	10
6	Du et al. (2010)	China	Maize		17	17
7	El-Hendawy et al. (2008)	Egypt	Maize		8	8
8	Farre and Faci (2006)	Spain	Maize	6	6	6
			Sorghum	6	6	6
9	Farre and Faci (2009)	China	Maize	18	18	18
10	French and Schultz (1984)	Australia	Wheat	11	11	11
11	Garba and Renard (1991)	Niger	Millet	14	12	15
12	Hao et al. (2014)	USA	Sorghum	24		24
13	Howell et al. (2014)	USA	Maize	12	12	12
14	Ibrahim et al. (1995)	USA	Millet	12	11	12
15	Ibrahim et al. (2015)	Niger	Millet	16	16	16
16	Kang et al. (2001)	China	Wheat	38	45	45
17	Kang et al. (2000)	China	Maize	18	18	18
			Wheat	45	45	45
18	Kiziloglu et al. (2008)	Turkey	Maize	12	12	12
19	Kresovic et al. (2016)	Serbia	Maize	16	16	16
20	Kuscu and Demir (2013)	Turkey	Maize	25	29	29
21	Mastrorilli et al. (1995)	Italy	Sorghum	5		5
22	Mesfine et al. (2005)	Ethiopia	Sorghum	3	3	3
23	Mishra et al. (2001)	India	Maize		14	14
24	Moroke et al. (2010)	USA	Sorghum		6	6
25	Nagaz et al. (2009)	Tunisia	Millet	8	8	8
26	Paolo et al. (2007)	Italy	Maize	3	3	3
27	Payero et al. (2008)	USA	Maize	31	31	32
28	Peng et al. (1991)	USA	Sorghum	5		5

No.	Author	Country	Crop	Biomass	Grain yield	WUE
29	Rostamza et al. (2011)	Iran	Millet	4		4
30	Rusere et al. (2012)	Zimbabwe	Maize	6	6	6
31	Sawargaonkar et al. (2013)	India	Sorghum	10	10	10
32	Seghatoleslami et al. (2008)	Iran	Millet		10	10
33	Siddique et al. (1990)	Australia	Wheat	11	11	11
34	Singh and Singh (1995)	India	Maize	4		4
			Millet	4		4
			Sorghum	4	4	4
35	Sivakumar and Salaam (1999)	Niger	Millet	1	7	8
36	Stewart et al. (1983)	USA	Sorghum	2	18	18
37	Stone et al. (1995)	USA	Maize	3	5	5
			Sorghum	4	5	5
38	Tari (2016)	Turkey	Wheat	18	22	22
39	Tinglu et al. (2005)	China	Maize		12	12
40	Tolk and Howell (2003)	USA	Sorghum	17	22	24
41	Xie et al. (2005)	China	Wheat	23		24
42	Zegada-Lizarazu et al. (2012)	Japan	Millet	12		12
43	Zhang et al. (2004)	China	Maize	4	4	4
			Wheat	15	15	15
44	Zhang et al. (2008)	China	Wheat	36	36	36
45	Zhang et al. (2014)	China	Maize		8	8
46	Zhang et al. (1998)	Syria	Wheat	15	20	20
	Total observations			566	617	707

3.3. Results

3.3.1. Variation in biomass and grain yield production and water use efficiency

Biomass production in maize ranged from 2 to 39.38 t ha^{-1} with a mean of 24.02 t ha^{-1} under optimal conditions (Table 3.2). In contrast, severe stress caused significant loss in biomass production where below 1 t ha^{-1} of maize biomass was recorded. The mean maize biomass production under drought stress was 10.24 t ha^{-1}, which was more than 50% lower than the average biomass production under optimal conditions. In comparison, the highest attainable biomass for maize under drought stress was 17.85 t ha^{-1}. The differences in mean biomass production between drought stressed and optimal conditions were significant ($t= -8.89$, $p<0.007$). Similarly, the impact of drought was significant on biomass production in pearl millet ($t= -2.82$, $p<0.001$). The mean biomass production in millet was 14.56 t ha^{-1} under optimal conditions, which was 43% higher than 8.31 t ha^{-1} attained under stress conditions. The maximum attainable biomass for millet under drought stressed conditions was 12.31 t ha^{-1}. Sorghum attained a maximum of 33.21 t ha^{-1} with means of 18.39 and 11.91 t ha^{-1} under optimal and drought stressed conditions, respectively, which were significantly different ($t=-5.58$, $p<0.001$). A minimum of less than 1 t ha^{-1} of biomass was achieved for sorghum under severe stress. For wheat, biomass production ranged between 0.18 and 13.86 t ha^{-1}. The mean wheat biomass production under optimal conditions was 8.12 t ha^{-1}, which was 52% higher and significantly different ($t=-9.23$, $p<0.001$) from 3.89 t ha^{-1} attained under stressed conditions. In comparison, the maximum attainable biomass in wheat under drought stress conditions was 17% of that achievable in maize.

Grain yield production followed an almost similar trend as biomass production (Table 3.2). Under optimal conditions, maize, millet, sorghum and wheat attained average yields of 8.19, 6.29, 6.45 and 3.11 t ha^{-1}, respectively. The average yield for each crop was also significantly reduced by drought stress. Maize and wheat however incurred high average losses, recording more than 52% losses in yield potential compared to 25% reduction realised in millet or sorghum due to drought stress. Severe drought stress caused minimum losses of below 0.6 t ha^{-1} while the average yield was significantly reduced in all crops. Maximum attainable yields in maize, sorghum and wheat were 77, 88 and 30% of the maximum recorded for millet. Water use efficiency for grain production showed significant variation among the crops (Table 3.2.). There was a general trend showing that water use efficiency increased under drought stress although maize showed a significant reduction while millet exhibited no significant differences. The mean water use efficiency of maize was 2.12 kg m^{-3} under optimal conditions compared to 1.87 kg m^{-3} under drought stressed conditions. The mean water use efficiency for grain production for millet was 2.18 and 4.66 kg m^{-3} under optimal and drought stress conditions, respectively. In comparison, sorghum attained high water use efficiency for grain yield under drought stress where 4.06 kg of grain were produced per m^3 litre of water applied. The water-use efficiency of sorghum under drought stress improved by 43% compared to 2.31 kg m^{-3} under optimal conditions. Drought stressed wheat had the second highest water use efficiency for grain production after sorghum. Wheat achieved water use efficiency for grain production of 3.01 kg m^{-3}, which was 60% higher than 1.18 kg m^{-3} attainable under optimal conditions. However, the maximum attainable water use efficiency for grain production of 9.65 kg m^{-3} was recorded for millet.

Table 3.2. Summary statistics showing biomass, yield and WUE attainable in different crops under different water scenarios

Biomass (t ha^{-1})								
	Maize		**Millet**		**Sorghum**		**Wheat**	
Statistic	***Optimal***	***Stressed***	***Optimal***	***Stressed***	***Optimal***	***Stressed***	***Optimal***	***Stressed***
Obs	84	130	119	86	64	43	28	12
Min	2.00	0.00	0.72	0.00	0.00	0.19	0.20	0.18
Max	39.38	17.85	23.63	12.31	33.21	10.21	13.86	3.50
Median	23.57	8.10	12.29	8.90	7.70	4.68	5.83	1.66
Mean	24.02	10.24	14.56	8.31	18.39	11.91	8.12	3.89
se	1.63	0.87	0.96	1.28	1.28	0.96	0.60	0.42
st dv	14.90	9.93	10.44	11.86	10.24	6.28	3.17	1.44
Variance	221.92	98.65	108.97	140.71	104.79	39.40	10.05	2.08
Kurtosis	1.27	2.66	3.79	2.29	1.11	7.05	0.07	-2.04
Skew	0.73	1.53	1.79	1.60	1.37	2.53	0.35	0.06
t statistic	-8.89		-2.82		-5.58		-9.23	
df	123.66		50.41		74.37		151.75	
p value	0.001		0.007		0.001		0.001	
Grain yield (t ha^{-1})								
	Maize		**Millet**		**Sorghum**		**Wheat**	
Statistic	***Optimal***	***Stressed***	***Optimal***	***Stressed***	***Optimal***	***Stressed***	***Optimal***	***Stressed***
Observations	124	130	119	87	55	39	25	21
Minimum	0.71	0.02	1.16	0.28	0.81	0.60	0.87	0.30
Maximum	15.39	12.59	19.80	13.33	17.43	11.75	5.97	2.90
Median	9.73	2.57	5.59	3.89	3.96	2.21	2.14	1.08
Mean	8.19	3.89	6.29	4.74	6.45	4.80	3.11	1.78
se	0.36	0.27	0.30	0.35	0.39	0.39	0.11	0.14
st dv	4.00	3.13	3.32	3.23	2.92	2.42	0.55	0.64
Variance	16.03	9.81	11.01	10.42	8.55	5.84	0.31	0.41
Kurtosis	-1.30	-0.14	-0.73	4.50	0.34	6.07	0.42	1.66
Skew	-0.40	0.89	0.53	1.66	0.97	2.37	-0.49	1.10
t statistic	-10.99		-2.19		-5.10		-8.61	
df	215.55		46.60		64.08		115.33	
p value	0.001		0.034		0.001		0.001	
Water use efficiency (kg m^{-3})								

	Maize		**Millet**		**Sorghum**		**Wheat**	
Statistic	***Optimal***	***Stressed***	***Optimal***	***Stressed***	***Optimal***	***Stressed***	***Optimal***	***Stressed***
Obs	125	130	119	99	89	56	45	44
Min	0.05	0.05	0.13	0.09	0.33	0.39	0.81	1.14
Max	5.93	3.90	8.45	9.65	6.50	5.79	3.11	3.93
Median	1.59	2.20	1.70	1.23	1.46	4.35	1.09	3.06
Mean	2.12	1.87	2.18	4.66	2.31	4.06	1.18	3.01
se	0.08	0.08	0.14	0.27	0.16	0.15	0.07	0.10
st dv	0.92	0.88	1.50	2.68	1.55	1.13	0.44	0.66
Variance	0.84	0.78	2.24	7.19	2.39	1.28	0.19	0.43
Kurtosis	2.96	-0.05	1.82	0.66	0.27	2.29	10.89	1.09
Skew	0.94	-0.58	1.25	1.43	1.13	-1.33	3.15	-1.00
t statistic	2.17		142.70		7.85		15.42	
df	253.00		1.61		139.70		74.83	
p value	0.031		0.110		0.001		0.001	

df=degrees of freedom, se=standard error, st dv=standard deviation

3.3.2. Variation among parameters of different crops

The differences in performance were further elucidated with boxplots to explore the variability in data among the crops and different water availability. Figure 3.1A shows that maize and sorghum exhibited significantly higher mean biomass production and wider variability compared to millet or wheat under optimal conditions. Their mean biomass was about 25 t ha^{-1}. Millet had the next highest mean biomass production capability, attaining about 19 t ha^{-1}. While wheat had the least biomass production capacity of slightly more than 10 t ha^{-1} under optimal conditions. The trend under drought stress was slightly different (Fig 3.1B). Sorghum and millet had significantly higher biomass production of about 11 t ha^{-1} compared to 8 t ha^{-1} attained by maize. Again, wheat had the lowest biomass production of about 6 t ha^{-1} under drought stressed conditions.

Grain yield production among the four crops followed a similar trend to biomass production (Fig 3.2). Under optimal conditions, sorghum and maize had similar average grain yield, which was significantly higher than that attained in millet and wheat. The average grain yield in maize and sorghum was 8 t ha^{-1} compared to about 6 t ha^{-1} attained by millet and wheat under optimal conditions (Fig 3.2A). The average yield in sorghum and millet was higher than maize and wheat under drought stressed conditions. In comparison sorghum and millet attained about 4.5 t ha^{-1} compared to less than 3.5 t ha^{-1} in maize or wheat (Fig 3.2B).

The water use efficiency for grain yield (under optimal conditions) in sorghum was significantly higher and exhibited wider variation compared to any other crop (Fig 3.3). Sorghum produced about 3.00 g of grain per m^{-2} per litre of water applied under optimal conditions (Fig 3.3A). In comparison, maize and millet exhibited similar water use efficiency of about 2.00 kg m^{-3} under optimal conditions while wheat had significantly the least water use efficiency for 1.5 g kg m^{-3}. However, water use efficiency for millet significantly improved to 4.5 kg m^{-3} under drought stressed conditions, which was significantly higher than 3.6 kg m^{-3} for sorghum and 2.0 kg m^{-3} for maize or wheat (Fig 3.3B). Millet also exhibited the widest variability in water use efficiency under drought stress conditions.

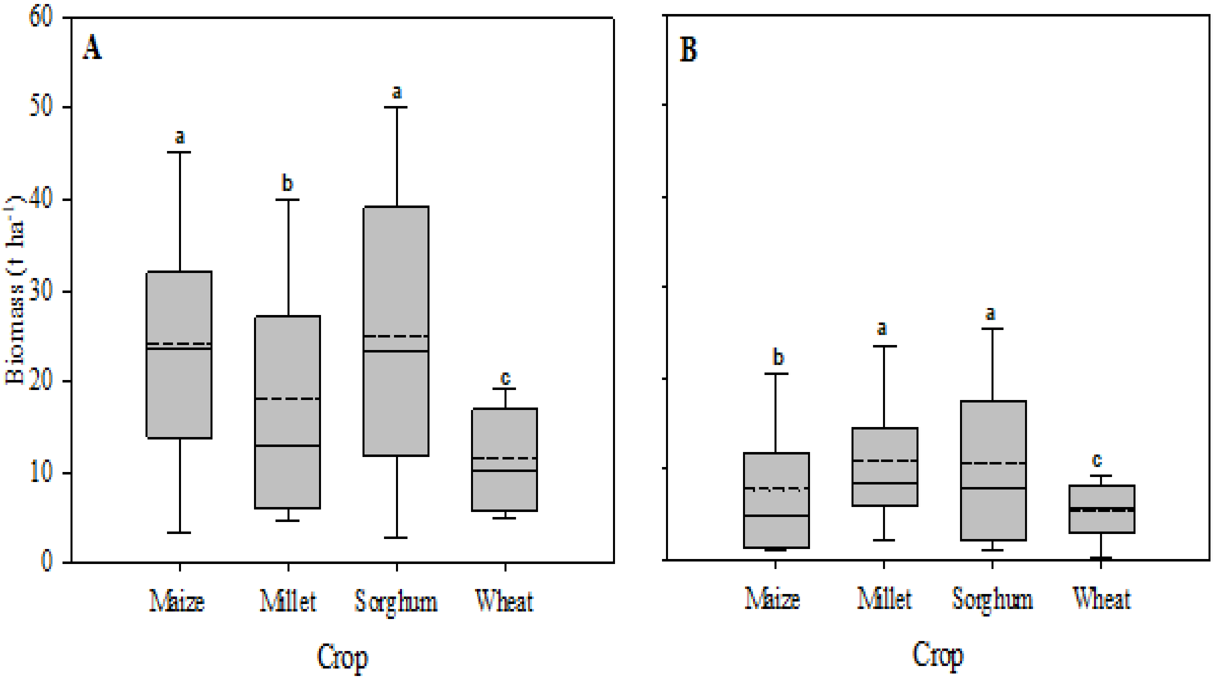


Figure 3.1. Biomass production among four crops grown under A) optimal and B) drought stress conditions.

Whiskers show 5 and 95th percentiles, dashed line is the mean the horizontal lines represent 25, 50 and 75th percentiles. Different letters above whiskers show significant differences at 0.05 using t statistic.

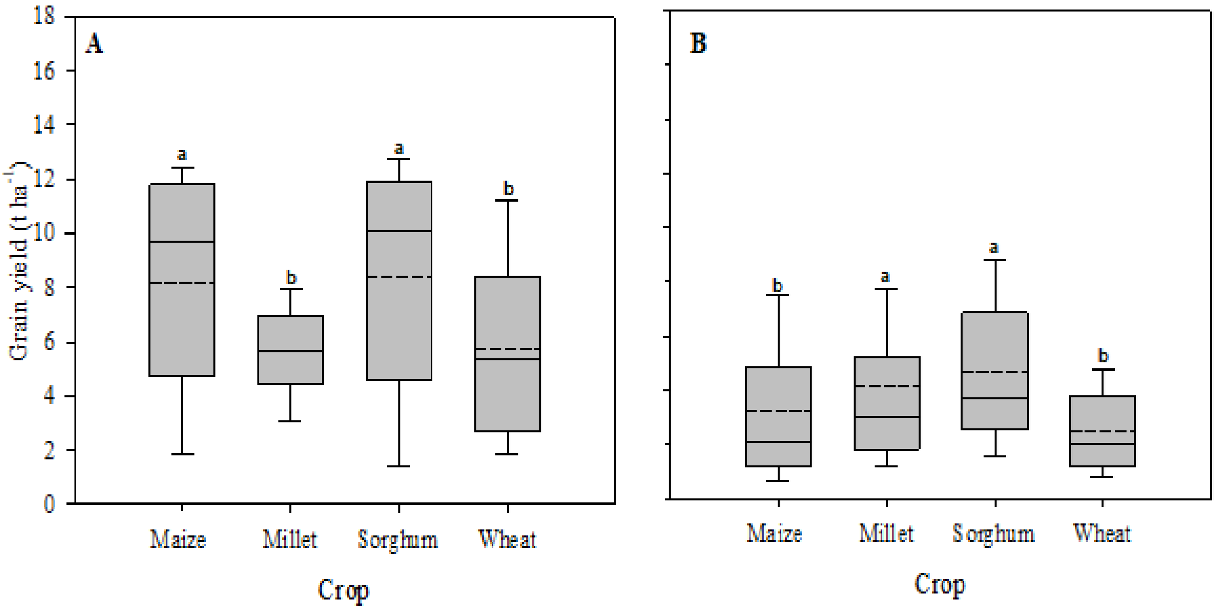


Figure 4.1. Grain yield production among four crops grown under A) optimal and B) drought stress conditions.

Whiskers show 5 and 95th percentiles, dashed line is the mean the horizontal lines represent 25, 50 and 75th percentiles. Different letters above whiskers show significant differences at 0.05 using t statistic.

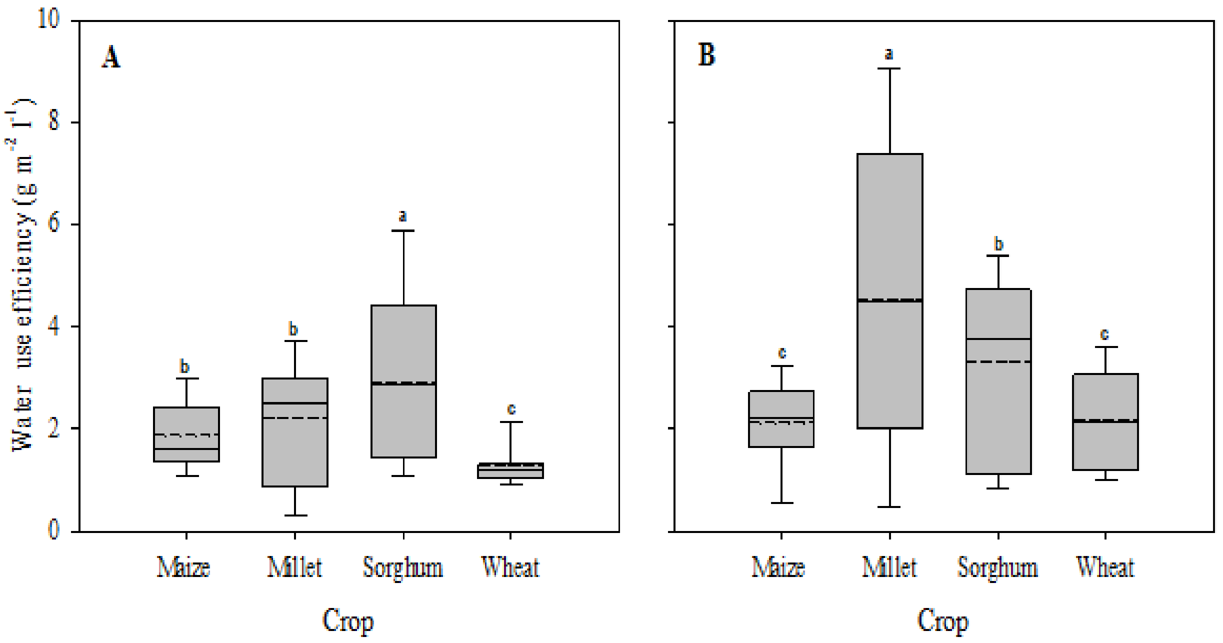


Figure 4.2. Water use efficiency among four crops grown under A) optimal and B) drought stress conditions.

Whiskers show 5 and 95th percentiles, dashed line is the mean the horizontal lines represent 25, 50 and 75th percentiles. Different letters above whiskers show significant differences at 0.05 using t statistic.

3.3.3. Crop-trait relationships among the four crops and measured parameters

The relationship among the parameters and crops was highlighted by a principal component analysis bi-plot using the first and second principal components. The first and second principal components explained 50.8 and 34.2% of the variation in biomass and grain yield production and water use efficiency among the four crops (Fig 3.4). There was a strong and positive correlation between biomass and grain yield production. Maize and sorghum were plotted along the vector for biomass and grain yield concomitant with the high biomass production capability. Maize also exhibited wide variation for biomass and grain yield production. On the other hand, wheat exhibited lower biomass and grain yield potential in comparison to the other crops. Water use efficiency showed weak correlations with grain yield and biomass production. In general, millet and sorghum were highly correlated with the vector for water use efficiency although some few maize crops exhibited exceptional water use efficiency. Again, wheat was mostly plotted in the negative x-axis showing its low potential for water use efficiency.

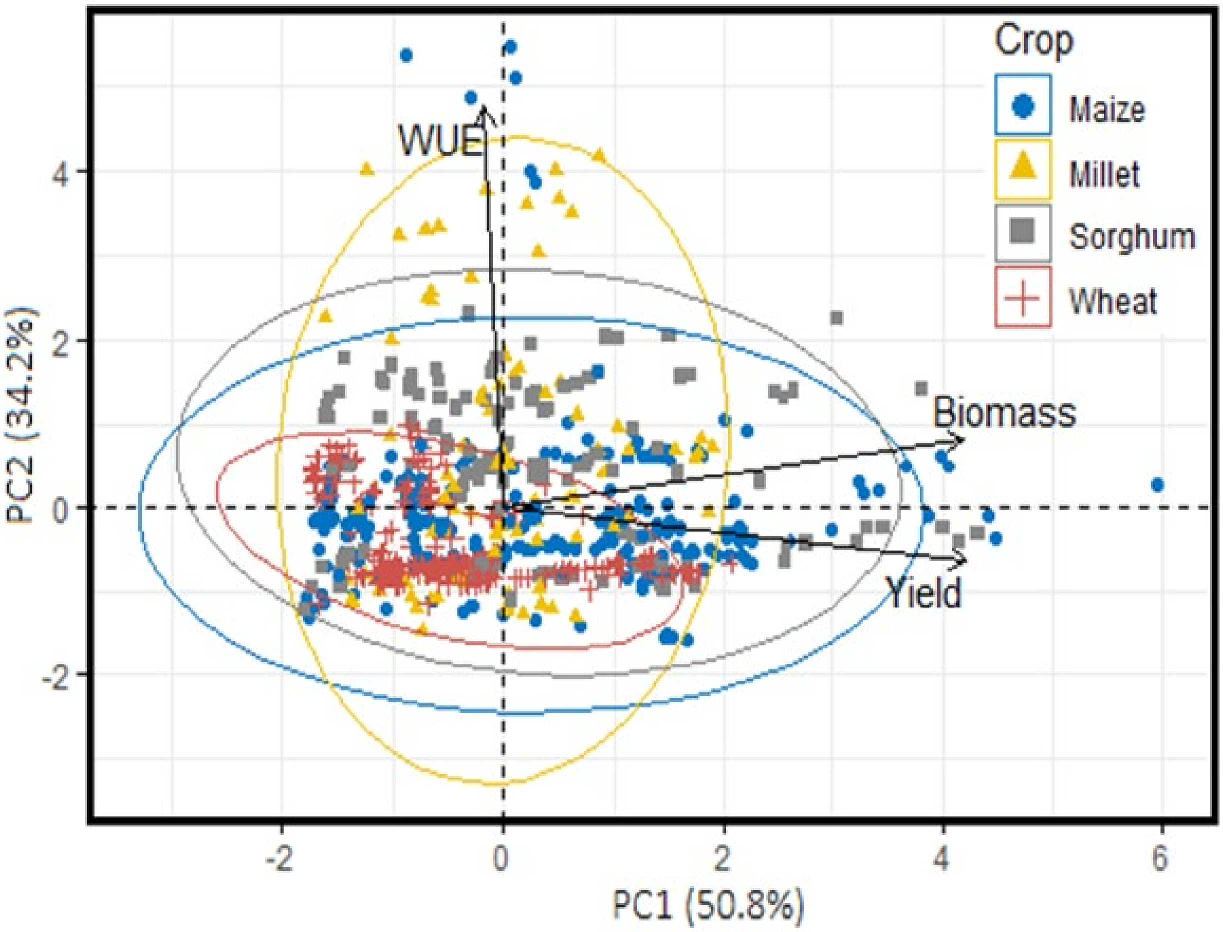


Figure 4.3. Principal component biplot of crop-trait relationships among four crops evaluated under drought and optimal conditions

3.3.4. Meta-analysis of effects of drought stress on parameters

Drought stress had a significant impact on the parameters (Fig 3.5). The highest impact of drought on biomass and yield production was observed on maize followed by wheat. The effect size of drought on biomass and grain yield in maize were -0.56 and -0.60 compared to -0.50 and -0.53, respectively for the same traits in wheat. Sorghum suffered the least effects of drought on biomass production while millet incurred the least reduction in grain yield production. On the other hand, drought generally stimulated an improvement in water use efficiency in the crops except maize. For maize there was a reduction in water use efficiency while millet exhibited an increase, although the effects of drought on the water use efficiency of these two crops was not significant. The effects of drought were significant on the water use efficiency of sorghum and wheat, with the highest mean value for effects of drought stress on water use efficiency observed on sorghum.

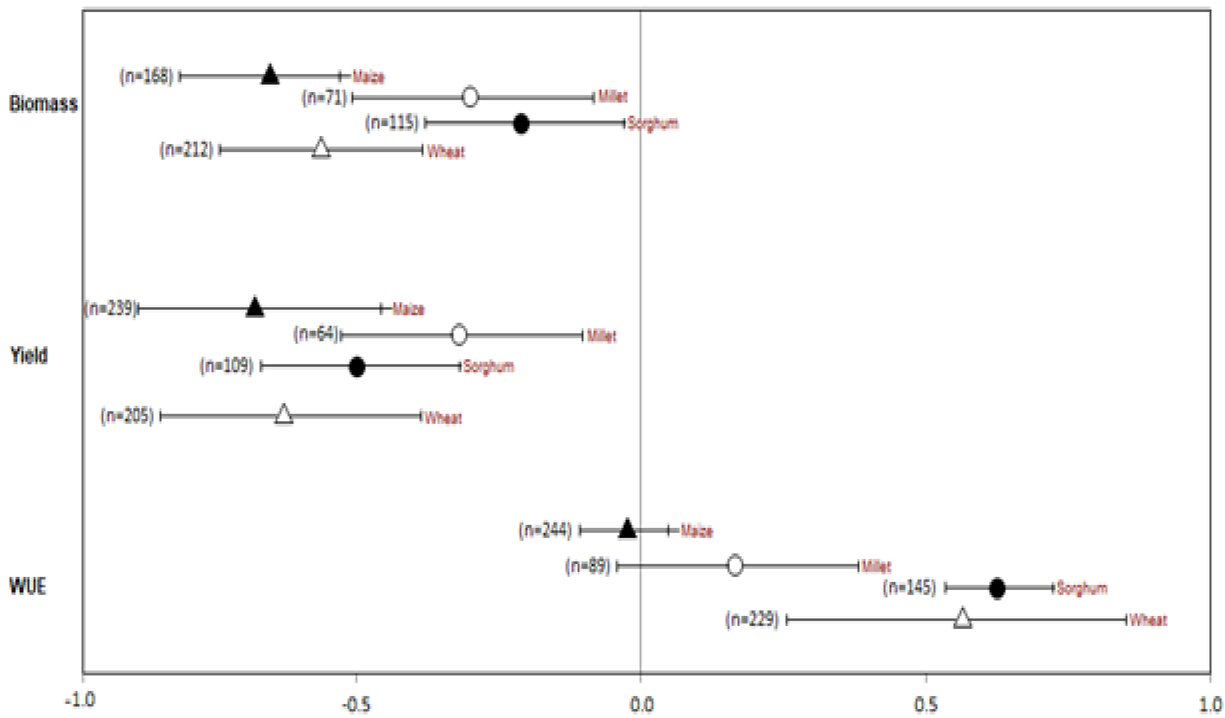


Figure 4.4. The impact of drought stress on biomass and yield production and water use efficiency (WUE) among four crops.

Error bars are effect size means±95% bootstrap CIs. Where the CIs do not overlap the vertical zero line, the effect size for that parameter is significant

3.4. Discussion

3.4.1. Biomass and grain yield production

The trend in biomass and grain yield production was similar for all crops showing that these two parameters are positively correlated. This positive link between above ground biomass traits has been identified in many studies (Peng et al., 1991; Ghassemi-Golezani and Tajbakhsh 2012; Habyarimana et al., 2019; Shamuyarira et al., 2019; Yang et al., 2019; Chen et al., 2020). Maize attained the highest mean biomass and grain yield production showing its high potential to accumulate biomass under optimal conditions. The mean biomass production by maize of between 10.24 and 24.02 t ha^{-1} found in this study was similar to a range between 7.00 and 20.00 t ha^{-1} reported previously for maize (Langholtz et al., 2016; Wilhelm et al., 2011). The variation depends on soil amendments such as fertilizer application, climatic conditions and nature of germplasm used in different studies. Tropical germplasm widely adapted to sub-Sahara Africa is known to be taller and accumulates higher vegetative biomass than temperate maize. For instance, Infante et al. (2018) found that tropical maize accumulated up to 40% higher non-grain biomass than temperate maize. There are also differences among open pollinated varieties, hybrids or inbred lines that contributed to the observed variation. In general, hybrids accumulate large amounts of grain and biomass yield compared to inbred lines or open pollinated varieties. The average maize yield found in this study was not significantly different from 9.2 t ha^{-1} reported by Muchow (1989). The maximum attainable biomass for millet and sorghum were 23.63 and 33.21 t ha^{-1}, respectively, showing that they also have high potential in biomass production for residue retention and as feedstock for biofuel. Like maize, millet and sorghum are C4 species characterised by high efficiency even under water limiting conditions. The average biomass production of 18.4 t ha^{-1} for sorghum was lower than 35.0 t ha^{-1} reported by Bhattarai et al. (2020). In comparison, the average biomass of 14.6 t ha^{-1} for millet under optimal conditions was similar to 12.2 t ha^{-1} reported by Tine et al. (2016). There are different types of sorghum for instance, biomass sorghum is bred specifically for producing large quantities of biomass for silage or biofuel production (Rooney 2014). The average grain yield for sorghum found in this was about 6.5 t ha^{-1} (under optimal conditions), which was comparable to 5.6 t ha^{-1} reported by Muchow (1989) but the average yield of 6.3 t ha^{-1} for millet was relatively higher than 2.9 t ha^{-1} reported by Muchow (1989). In comparison, wheat is a C3 species that is easily affected by oxidation of substrates and water shortages. Concomitantly, wheat had the lowest biomass production potential with a mean biomass potential of 8.1 t ha^{-1} and grain yield potential of 3.1 t ha^{-1}, which were comparable to 8.2 and 2.5 t ha^{-1} for biomass and grain yield, respectively, reported by Mathew et al. (2019). The variation in potential biomass production among the crops was attributed to differences in genetic constitution and environmental conditions. The C4 species are generally superior in biomass production compared to C3 species. The possession of a C4 photosynthetic pathway renders an efficient accumulation of biomass in maize, sorghum and millet compared to C3 plants in field and controlled environments (Katerji and Mastrorilli, 2014). On the other hand, maize is known to be superior in solar radiation interception and maximizing resource utilization under optimal conditions compared to sorghum and millet (Amanullah and Stewart, 2013). However, other researchers have reported higher biomass production in sorghum compared to maize or millet (Hallam et al., 2001). This variation exhibited by maize and sorghum is also affected by whether the varieties grown are bred for grain or biomass production. Biomass varieties used for silage or biofuels have higher potential for non-grain biomass production.

3.4.2. Impact of drought stress on biomass and grain yield production

Both biomass and grain yield production were significant reduced by drought stress. Drought stress causes variation within and among the crops as they have different responses to the effects of drought. In general, drought impacts on biomass and grain yield through its negative effects on leaf water potential, stomatal opening and cell turgidity (Riboldi et al., 2016). Sorghum incurred the least reduction in biomass production of 25% due to drought stress, which was lower than 42% reported by Perrier et al. (2017). The biomass production capacity of maize and millet were reduced by 50 and 43%, respectively, under water stressed conditions in comparison to optimal conditions, which was comparable to reports by Mi et al. (2018) who observed that drought reduced biomass production in maize by up to 46.6%. The reduction in biomass and grain yield of wheat were slightly higher than 35% reported by Mathew et al. (2019). The impact of drought on a crop is influenced by its genetics in addition to environmental conditions. The high impact on maize has been reported previously showing that maize is more sensitive to drought compared to sorghum or millet (Rooney, 2014). The relative reduction in biomass (52%) and grain yield (43%) production in wheat under drought stressed conditions could be due to effect of environmental factors such as humidity and temperature. Despite its low prolificacy in biomass or grain yield production, wheat is usually grown in winter when temperatures are low and humidity relatively higher than hot and drought summer seasons for sorghum, millet and maize production. This could cushion it against extreme adverse effects of drought stress combined with other environmental factors. High temperature and low humidity exacerbate the impact of drought stress on crops, leading to higher reduction in biomass and grain yield production or complete crop failure (Killi, 2017). The impact of environmental conditions on crop performance in biomass and grain yield production is affected by their pre-existing acclimation and photosynthetic pathway (Berry and Björkman 1980). Thus, variation in biomass accumulation among the different crops point to their different acclimation, genetic constitution and potential, which govern their efficiency in utilizing water resources under the different water availability scenarios. Reports have shown that crops such as sorghum, millet or maize vary in biomass and grain yield accumulation depending on agronomic practices, varieties and water availability (Zegada-Lizarazu et al., 2012; Oikawa et al., 2014); and it would be prudent to evaluate their water dynamics for informed decision making in agriculture systems.

3.4.3. Variation in water use efficiency among the crops

The highest water use efficiency under optimal conditions was found in sorghum and millet followed by maize, which corroborated reports by Bhattarai et al. (2020). In this study, the average water use efficiency of sorghum, millet and maize under optimal conditions were almost similar at about 2 kg m^{-3}, which was underpinned by their highly efficient C4 photosynthetic pathway compared to wheat, which is a C3 species. The reported water use efficiency estimates for sorghum were comparable to 4.3 kg m^{-3} reported by Roy et al. (2019). Sorghum is known to be more water use efficient than most cereals including maize, wheat or millet (Mbava et al., 2020; Rooney, 2014). There was a wide variation and a general increase in water use efficiency under drought stress conditions. The variability is also attributable to inter- and intraspecific differences alluded to biomass production. For instance, differences in agronomic performance among hybrids, varieties and un-improved traditional varieties in maize, wheat, sorghum and millet have been highlighted previously (Mbava et al., 2020; Ficiciyan et al., 2018; Lamptey et al., 2014). On the other hand, Fang et al. (2014) asserted that modern improved varieties have better water use efficiency compared to old and un-improved landraces even under water stressed conditions. It is therefore imperative to identify crops and varieties that would maximize productivity in water constrained environments such as agro-ecological zones of sub-Sahara Africa. The average estimates for water use efficiency calculated for crops in this study were similar to those reported by Mbava et al. (2020) and Zhao et al. (2018) for cereals but slightly higher than those reported by Jabereldar et al. (2017) for sorghum. In general, drought stress increases water use efficiency in crops although maize showed a significant decrease in water use efficiency due to drought. The decrease in water use efficiency of maize is not unique to this study as Kiziloglu et al. (2009) also reported that maize exhibited higher water use efficiency under irrigated treatment compared to stress treatment. The increase in water use efficiency under drought stress is facilitated by closure of stomatal opening that prevents transpirational water loss. The closure of stomata decreases water loss but has a negative impact on gaseous exchange and leads to photorespiration especially in C3 species such as wheat (Killi et al., 2017). In comparison, C4 plants such as sorghum and millet have the capacity to avoid the detrimental effects of accumulated carbon dioxide during stomatal closure, which enable them to maintain relatively higher photosynthetic activity even under limiting conditions compared to wheat (Sage and Monson, 1998). In the case of maize, its water use efficiency under drought stress is relative lower than sorghum and millet due to differences in rooting ability. Bhattaraj et al. (2019) elaborated that sorghum and millet were more drought tolerant than maize owing to their extensive root system that were efficient in water acquisition. For instance, rooting depths of up to 1.6 m have been reported in sorghum and millet compared to 0.7 m in maize and wheat (Faye et al., 2019; Porter 2010). Sorghum has higher root density and better partitioning of biomass to roots under drought stress (Zegada-Lizarazu et al., 2012).

3.4.4. Multivariate associations

The large proportion of variation explained by the principal component biplot shows that the two components of the PCA could adequately represent the variation. The high variation could also be due to the fact that only three parameters were used to discriminate the genotypes. However, the variation shows that the crops were largely different in these parameters, which was expected given that they belong to different species and they were also varietal differences. Variation in agronomic performance among crops is attributed primarily to genetic differences and the impact of environment on the performance of the different genotypes (Oikawa et al., 2014). The correlation between biomass and grain yield production shows that crops or varieties with strong above ground sinks were likely to produce more grain yield. This is important for food security especially in grain cereals such as maize and wheat, which are strategic commodity crops for South Africa. However, the partitioning of excessively high amount of above ground biomass can be detrimental during periods of constrained moisture availability (Haque et al., 2016; Kashiwagi et al., 2006; Manschadi et al., 2006). As previously alluded, sorghum is able to maintain drought tolerance due to its ability to partition relatively larger proportion of biomass below ground compared to maize or millet. On the other hand, the negative or weak correlation between water use efficiency and the other parameters shows that although sorghum and millet have ability to conserve water during stress periods their maximum attainable biomass is still lower than that of maize. Yield and biomass production are genetically governed and each species has a limit. Under optimal conditions, maize has been reported to have higher genetic potential to produce biomass but at a larger water cost. In comparison, sorghum and millet are efficient and they would be more productive under drought stress but may not have an advantage over maize in well managed systems. Wheat would likely attain less biomass or grain compared to the C4 crops under any water availability scenario.

3.4.5. Crop response to drought stress

The highest impact of drought on biomass and yield production was observed on maize followed by wheat, which could be attributed to their generally high sensitivity to moisture stress. Maize and wheat are known to be highly sensitive to moisture stress at all stages of growth (Daryanto et al., 2016). The impact of drought is exacerbated by other environmental factors such as heat stress that often occur simultaneously or in sequence with drought (Rezaei et al., 2018; Ferreira, 2016). On the other hand, the confounding effects of heat stress on wheat are less evident since most wheat is grown in cooler seasons when temperature rarely rise to cause significant heat stress. Sorghum and millet are widely reported to be tolerant to drought through their C4 photosynthetic pathways, deep and extensive roots systems and their reduced leaf area indices that minimize water loss compared to maize or wheat. Biomass and grain yield production are most affected by drought in agreement with several reports that suggest that the grain-filling period, responsible for grain production, is the most sensitive stage followed by the seedling stage, responsible for biomass accumulation (Rezaei et al., 2018; Ferreira 2016). Sorghum suffered the least effects of drought on biomass production while millet incurred the least reduction in grain yield production due to their relative ability to maintain viable assimilate partitioning even under water stress (Killi et al., 2017). On the other hand, drought generally stimulated an improvement in water use efficiency in the crops, a phenomenon attributable to stomatal closure for reducing transpirational losses while maintaining conductance for CO_2 to proceed with photosynthesis (Killi et al., 2017; Riboldi et al., 2016).

3.5. Conclusion

The study evaluated the relative differences among sorghum, maize, millet and wheat in biomass and grain yield production and water use efficiency, and their response to water limitations. It was found that, firstly, the crops differed significantly in all these parameters, which opens opportunities for selecting crops and varieties suitable for production under different production systems. The means for biomass production for maize, sorghum, millet and wheat were 24.0, 18.4, 14.6 and 8.1 t ha^{-1}, respectively, under optimal conditions. Similarly, grain yield production followed a similar trend with maize attaining 8.2 t ha^{-1} followed by sorghum (6.5 t ha^{-1}), millet (6.3 t ha^{-1}) and wheat (3.1 t ha^{-1}). The high biomass production potential of maize, sorghum and millet is supported by their C4 photosynthesis compared to wheat, which is a C3 crop. Secondly, drought significantly reduced biomass and yield production while water use efficiency significantly increased under drought stress. Drought stress had the highest impact on grain yield production on maize (52%) and wheat (43%) compared to sorghum (26%) and millet (25%). The meta-analysis showed that the effect size for drought on yield and biomass production potential were highest for maize (-0.6) followed by wheat (-0.55), showing that these commodity crops were highly sensitive to drought stress compared to sorghum and millet. The third most important conclusion drawn from this study was that indigenous crops, sorghum and millet, were more efficient in water use efficiency and should be promoted for production under low management smallholder farming systems. Again, sorghum and millet were more efficient in water use, attaining more than 4.0 kg of grain m^{-3} of water used. In conclusion, sorghum and millet are recommended for water-constrained ecologies to improve water use efficiency and food production.

Chapter 4 WATER USE EFFICIENCY OF WHEAT UNDER GLASSHOUSE CONDITIONS

Abstract

The biomass allocation pattern of plants to shoots and roots is key in the cycle of elements such as carbon, water and nutrients with, for instance, the greatest allocations to roots fostering the transfer of atmospheric carbon to soils through photosynthesis. Several studies have investigated the root to shoot ratio (R:S) biomass of existing crops but variation within a crop species constitutes an important information gap for selecting genotypes aiming for increasing soil carbon stocks for climate change mitigation and food security. The objectives of this study were to evaluate agronomic performance and quantify biomass production and allocation between roots and shoots, in response to different soil water levels to select promising genotypes for breeding. Field and greenhouse experiments were carried out using 100 wheat genotypes under drought stressed and non-stressed conditions. The experiments were set up using a 10*10 alpha lattice design with two replications under water stress and non-stress conditions. The following phenotypic traits were collected: number of days to heading (DTH), number of productive tillers per plant (NPT), plant height (PH), days to maturity (DTM), spike length (SL), kernels per spike (KPS), thousand kernel weight (TKW), root biomass (RB), shoot biomass (SB), root to shoot ratio (R:S) and grain yield (GY). There was significant ($p<0.05$) variation for grain yield and biomass production because of genotypic variation. The highest grain yield of 247.3 g m^{-2} was recorded in the genotype LM52 and the least was in genotype Sossognon with 30 g m^{-2}. Shoot biomass ranged from 830 g m^{-2} (genotype Arenza) to 437 g m^{-2} (LM57), whilst root biomass ranged between 603 g m^{-2} for Triticale and 140 g m^{-2} for LM15 across testing sites and water regimes. Triticale also recorded the highest R: S of 1.2, while the least was 0.30 for wheat genotype LM18. Overall, water stress reduced total biomass production by 35% and R: S by 14%. Genotypic variation existed for all measured traits useful for improving drought tolerance, while the calculated R: S values can improve accuracy in estimating C sequestration potential of wheat. Wheat genotypes LM26, LM47, BW140, LM70, LM48, BW152, LM75, BW162, LM71 and BW141 were selected for further development based on their high total biomass production, grain yield potential and genetic diversity under drought stress.

Keywords: agronomic traits, Carbon sequestration, grain yield, soil water, wheat

4.1. Introduction

Evaluating biomass allocation to plant roots, shoots and economic traits in plants can help to predict genetic gains achievable through breeding and to assess the impact of crop biomass on several ecosystem functions such as carbon, water and nutrient cycles, which affect crop production (Litton et al., 2007). Up to 80% of soil C comes from root activity and turnover (Yang et al., 2012); consequently, the allocation of C to roots has important consequences for transfer of C to soil since fostering atmospheric C allocation to soils mitigates against climate change (Minasny et al., 2017). Soil organic matter, which constitutes the bulk of soil C, retains essential nutrients, improves water holding capacity and provides energy for soil living organisms, all of which enhance soil ecosystem functioning and crop production.

Biomass allocation between roots and shoots, expressed as root to shoot ratio (R: S), is highly variable amongst plant species. R: S ratio above unity shows that production of root biomass exceeded that of above ground biomass. The R: S ratios reported for annual cereal crops such as wheat are comparably lower than for perennial grasses. Amanullah and Stewart (2013), for instance reported R: S ratios of 0.41 and 0.29 for sorghum and maize, respectively while Yang et al. (2010) reported a mean of 0.25 for maize and wheat. Bolinder et al., (2002b) reported R: S ratios of up to 7.00 for forage grasses compared to a range of 0.1 to 0.5 cited for many annual crops including cereals and legumes (Mathew et al., 2017). There are also variations in R: S within single species which can be attributed to genotypic differences.

Intra-specific variation in any trait results from genotypic differences among individuals in a given species. King et al. (2007) asserted that there is wide genotypic variation for biomass allocation. Fang et al. (2017) found a R: S ratio of 1.13 in a wheat landrace compared with 0.61 and 0.81 found in two modern cultivars. Similarly, Siddique et al. (1990) reported higher R: S ratios ranging between 0.74 and 1.18 for obsolete varieties than modern cultivars of wheat which ranged from 0.72 to 0.84. Most crop improvement programs in wheat have focused on channeling more biomass towards economic traits such as grain and above ground biomass for food, feed and biofuel production without improving root systems leading to low R: S in modern cultivars compared to landraces (Wasson et al., 2012). However, variations do occur even in genotypically identical individuals due to environmental factors.

Biomass allocation is also affected by genotype x environment interaction. It indicates environmental plasticity to soil properties, temperature and soil water availability (Sánchez et al., 2014, Pittelkow et al., 2015). Water availability is a key factor influencing biomass allocation as drought stress is known to reduce crop growth. This was illustrated by Perdomo et al. (2015) who reported biomass reductions of 60% in maize and rice, and 90% in wheat. However, drought stress tends to increase R: S ratio as reported by Tatar et al. (2016) who found 7% higher ratio in wheat produced under 25% soil water content than wheat grown at 75% soil water content. However, Vanaja et al. (2011) reported a contrasting trend with 60 and 7% reduction in R: S for maize and sunflower, respectively, as a result of 30% reduction in soil water content. Biomass allocation also vary with intensity and duration of drought stress (Farooq et al., 2009). Instantaneous and short-lived drought spells may not cause a significant shift in biomass allocation, while excessive drought stress beyond a threshold level causes plants to lose their biomass allocation regulatory ability completely (Xu et al., 2010). Conversely, Sharp and Davies (1985) indicated that soil water stress has greater negative impact on shoot than root growth which led to an increase in R: S.

Several, studies have investigated variations in R: S between plant species but little is known about intra-specific variations. This constitutes an important information gap when selecting crop varieties for specific objectives (e.g. grain or biomass production, soil C sequestration, drought tolerance). Moreover, in the context of global warming (Ashraf and Fooled, 2007), there is need to investigate intra-specific variation in response to drought stress. Therefore, the objective of this study was to characterize intra-specific variations in R: S ratios and agronomic performance of diverse wheat genotypes sourced from the International Maize and Wheat Improvement Centre (CIMMYT) and subjected to water stressed and non-stressed conditions. The results can be helpful to crop breeders in evaluating diversity in biomass allocation and agronomic performance, which is important for developing varieties with greater water use efficiency and drought tolerance for grain yield and C sequestration into soils.

4.2. Materials and methods

4.2.1. Plant material

One hundred genotypes of wheat were evaluated to develop a selection protocol for evaluating water use efficiency in the greenhouse. Subsequently, 10 genotypes of wheat were selected from the panel of 100 genotypes. The 100 genotypes included drought and heat tolerant wheat accessions, 2 commercial wheat varieties from France and triticale (×Triticosecale Wittmack) (Appendix 1). The drought and heat tolerant genotypes were obtained from the International Maize and Wheat Improvement Center (CIMMYT). The CIMMYT genotypes were used owing to their genetic variability for rooting abilities and breeding history for drought tolerance.

4.2.2. Growing conditions and trial management

The experiments were carried out at the Controlled Environment Facility of the University of KwaZulu-Natal Pietermaritzburg Campus. The initial panel of 100 wheat genotypes was planted in a 10 × 10 alpha lattice design with two replications. Ten seeds were sown in each pot and thinned to 8 plants per pot, 3 weeks after emergence. Ten pots were allocated per block and genotypes were randomly assigned to pots to minimize the experimental error associated with water discharge from drip irrigation. Fertilizer was applied through automated drip irrigation at a rate of 300 kg N ha^{-1} and 200 kg P_2O_5 ha^{-1}. The different water regimes were initiated 6 weeks after planting to ensure good establishment but also to ensure early exposure of all growth stages to drought. In the well-watered regime, the plants were watered to field capacity (FC) whenever average soil water content fell to 80% of FC, while in the water stress conditions volumetric soil water content was allowed to drop to 30% of FC before watering to FC. The soil water content was monitored by a soil moisture probe and weighing of the pots. The two watering treatments were maintained until maturity for each crop. The first greenhouse experiment (GH1) was carried out in summer (October 2016 to February 2017), while the second one (GH2) ran concurrently with an open field experiment in winter (May to September 2017).

4.2.3. Data collection

Agronomic traits were recorded during the growth period. At maturity, 8 plants were harvested from each pot. All the roots in the soil volume were collected per genotype per pot. The larger roots were manually separated from the soil and the finer roots were collected by wet sieving. The separated plant parts were oven dried at 60°C for 72 hours to measure the dry weight. Plant parts for each pot were separated at maturity into grain, shoot and root and oven dried at 60°C for 72 hrs to measure the dry weight. The weight was converted to gram per square meter (g m^{-2}) accordingly by factoring in plant population. Root: shoot (R:S) ratio and total plant biomass (PB) were computed after determining GY, RB, and SB.

4.2.4. Data analysis

Analysis of variance (ANOVA) was conducted using the lattice procedure using Genstat 18th edition (Payne et al., 2017). In addition, the means of genotypes and the different water regimes were separated by Fischer's unprotected least significant difference (LSD) at 0.05 significance level to quantify the effects of genotype, environment and water regime. The cumulative amount of irrigation water was calculated at the end of the growing period. WUE was calculated as the ratio of the amount of water used by the plant to amount of biomass accumulated yield or carbon in the soil at the end of experimentation.

$$\boldsymbol{WUE = \frac{DM}{TIW}}$$

where DM is plant dry mass and TIW is total amount of irrigation water.

4.3. Results

4.3.1. Variations in grain yield and plant biomass amongst wheat cultivars

The average grain yield (GY) amongst the initial set of 100 wheat genotypes was 1387 g m^{-2} with a standard error of ± 84 g m^{-2} (Table 4.1). GY varied by a 62 factor from 75 to 4696 g m^{-2} and showed a positively skewed distribution (Skew=1.53). Total plant biomass (PB) was much less variable than GY as values ranged from 1967 to 13529 g m^{-2}, a 6.8 times difference, which was significant at $P<0.001$. The 100 selected genotypes had an average root to shoot ratio for biomass of 0.12, meaning that they allocated an average 12% of their total biomass to their roots, with values from 3 to 38% (Table 4.1). The sub-set of 10 wheat genotypes showed much less variations in grain yield and plant total biomass and biomass allocation with for instance GY ranging from 621 to 4383 g m^{-2} (a 7.0-fold increase *vs* 62 times for the 100 genotypes), but similar variations in PB with values between 2475 and 13529 g m^{-2} (5.4 *vs* 6.8) and in R: S (Table 4.1).

4.3.2. Variation in carbon concentration and stocks

The average carbon content for the selected 10 genotypes was 34±0.9% in the shoots (SC_C) and 30±1.1% in the roots (RC_C). SC_C ranged from 30 to 37%, while RCc varied from 21 to 39%. However, the carbon stocks in the shoots, roots and in the total plant biomass showed greater variations. For instance, the total plant carbon stocks (PCs) exhibited a mean of 1174± 64 g m^{-2}

with values from 582 to 3022 g m^{-2} (Table 4.1). Most of the PC_S was contributed by the carbon stocks in the shoots (91%), while only 9% of the carbon stocks was contributed by the roots.

Table 4.1. Summary statistics for grain yield, selected morphological variables for the 100 and selected 10 wheat varieties grown in the greenhouse under different moisture regimes

	GY	SB	RB	PB	R:S	SC_C	RC_C	SC_S	RC_S	PC_S
	----------------------g m^2 ----------------------					----------%--------		--------------g m^2 ----------------		
100 genotypes										
Mean	1387	2498	305	4189	0,12					
Median	1309	2332	263	3930	0,11					
Minimum	75	1179	65	1976	0,03					
Maximum	4696	8658	1219	13529	0,38					
Q1	959	1827	189	3026	0,09					
Q3	1644	2908	365	4893	0,15					
CV%	47	37	57	37	41					
SEM	84	121	22	200	0,01					
Skewness	1,53	2,04	2,03	1,82	1,67					
Kurtosis	4,35	7,15	5,76	5,66	4,39					
10 selected genotypes										
Mean	1882	3169	356	5408	0,12	34	30	1067	106	1174
Median	1624	2929	326	5214	0,10	34	30	940	93	1061
Minimum	621	1598	152	2475	0,06	30	21	534	45	582
Maximum	4383	8658	1006	13529	0,26	37	39	2869	302	3022
Q1	1349	2317	249	3952	0,09	32	28	792	72	862
Q3	2500	3691	427	6140	0,13	35	33	1210	136	1384
CV%	45	44	47	42	35	6	14	44	49	42
SEM	109	182	22	290	0,01	0	1	61	7	64
Skewness	1,05	2,03	1,91	1,65	1,60	-0,29	-0,19	1,98	1,86	1,77
Kurtosis	1,03	6,08	5,89	4,19	3,40	-0,85	-0,01	5,62	5,17	4,71

Q1: 25th percentile, Q3: 76th percentile, CV: coefficient of variation, SEM: standard error of mean, GY: grain yield, SB: shoot biomass, RB:Root Biomass, PB: Total Plant Biomass, SC_C: shoot carbon content, RCc: Root carbon content, SCs: Shoot carbon stocks, RCs: Root carbon stocks and PCs: Plant carbon stocks

4.3.3. Agronomic performance under variable moisture regimes

The average grain yield for the selected 10 wheat genotypes increased from 326±28 g m^{-2} under drought stress (25% FC) to 414±27 g m^{-2} for 75% field capacity (75FC), which corresponded to a 27% increase (Table 4.2). At the same time the total plant biomass (PB) increased by 110% (from 2992±94 to 6289±247 g m^{-2}) (Table 4.2). This was concomitant to an increase in the R: S ratio (from 0.43 to 0.57). The greatest increase occurred for root C stocks (170%), followed by plant C stocks (118%).

Table 4.2. Summary statistics for grain yield, selected morphological variables among the selected 10 wheat varieties grown in the greenhouse under different moisture regimes

	GY	SB	RB	PB	R:S	SCC	RCC	SCS	RCS	PCS
	-----------------------g m^{2} -----------------------					----------%--------		--------------g m^{2} ----------------		
25% Field capacity										
Mean	326	1873	792	2992	0,43	33	31	625	239	865
Median	266	1826	762	2918	0,44	34	31	621	225	854
Minimum	58	1349	368	2156	0,15	25	21	409	103	592
Maximum	869	2415	1263	3983	0,70	37	39	805	379	1109
CV%	68	24	36	24	34	19	23	25	34	23
SEM	28	57	37	94	0,02	1	1	20	10	26
75% Field capacity										
Mean	414	3726	2149	6289	0,57	33	31	1243	647	1890
Median	397	3440	1823	5508	0,57	34	31	1186	605	1716
Minimum	136	2779	1290	4531	0,37	25	21	851	333	1324
Maximum	811	5192	3575	9305	0,78	37	39	1822	1059	2881
CV%	50	26	38	30	26	19	23	27	35	28
SEM	27	127	104	247	0,02	1	1	44	29	69

Q1: 25th percentile, Q3: 76th percentile, CV: coefficient of variation, SEM: standard error of mean, GY: grain yield, SB: shoot biomass, RB:Root Biomass, PB: Total Plant Biomass, SCC: shoot carbon content, RCc: Root carbon content, SCs: Shoot carbon stocks, RCs: Root carbon stocks and PCs: Plant carbon stocks

4.3.4. Water use efficiency for grain, biomass production and carbon storage

The average WUE for GY was 0.74 g m^{-2} l^{-1} in the glasshouse under 25FC, with values from 0.12 to 2.10 g m^{-2} l^{-1} (Table 4.3). In the glasshouse and under 75FC WUE for GY ranged from 0.12 to 0.65 g m^{-2} l^{-1} with an average of 0.33 and standard error of ±0.02. The WUE for total plant biomass (PB) was higher than that of shoots (WUE-SCS), roots (WUE-RB) and grain yield (WUE-GY), as it had an average of 6.58 g m^{-2} l^{-1} and 5.07 g m^{-2} l^{-1} in the glasshouse under 25% and 75% FC respectively. Also, the WUE of carbon was higher in the total plant (WUE-PSC) than in roots or shoots as it ranged from 1.16 to 2.68 g m^{-2} l^{-1} with an average of 1.89 g m^{-2} l^{-1} in the glasshouse under 25% FC (Table 4.3).

Table 4.3. Summary statistics for biomass and plant carbon WUE for the 10 selected wheat varieties grown in the field and glasshouse under different moisture regimes

	GY	SB	RB	PB	SC_S	RC_S	PC_S
				----------g m^2 l^{-1}----------			
25% Field capacity							
Mean	0.74	4.10	1.74	6.58	1.37	0.52	1.89
Median	0.55	4.16	1.71	6.49	1.38	0.53	1.93
Min	0.12	2.63	0.89	4.21	0.80	0.25	1.16
Max	2.10	5.84	3.06	9.63	1.95	0.77	2.68
CV	75	20	34	22	20	28	17
SEM	0.07	0.10	0.08	0.19	0.04	0.02	0.04
75% Field capacity							
Mean	0.33	3.00	1.74	5.07	1.00	0.52	1.52
Median	0.31	2.73	1.58	4.51	0.94	0.49	1.42
Minimum	0.12	2.28	0.94	3.56	0.64	0.30	1.10
Maximum	0.65	4.42	3.04	7.92	1.55	0.90	2.45
CV %	51	26	39	31	27	36	29
SEM	0.02	0.10	0.09	0.20	0.04	0.02	0.06

Q1: 25th percentile, Q3: 76th percentile, CV: coefficient of variation, SEM: standard error of mean, WUE-GY: Grain yield WUE, WUE-SB: Shoot Biomass WUE, WUE-RB:Root biomass WUE,WUE-PB: Plant biomass WUE, WUE SCS: Shoot carbon stocks WUE;WUE-RCS: Root carbon stock WUE and WUE-PCS: Total plant carbon stocks WUE

4.3.5. Varietal differences in water use efficiency for grain yield, biomass production and carbon storage

WUE for grain yield (GY) was much higher at 25FC than 75FC, with all the 25FC values above 0.75 g m^{-2} l^{-1} and 75FC values below 0.75 g m^{-2} l^{-1} (Fig 4.1). Under stressed conditions (25FC) the two best cultivars for WUE for GY were LM75 with 1.76 g m^{-2} l^{-1} followed by LM71 with 1.37 g m^{-2} l^{-1}. LM 70 and LM 48 had the lowest WUE for GY with values of 0.76 and

0.78 g m^{-2} l^{-1} respectively. This corresponded to a maximum 130% difference in WUE for GY. Under well-watered conditions (75FC), BW162 exhibited the highest WUE for GY amongst all the cultivars (0.66 g m^{-2} l^{-1}), whilst LM47 had the lowest (0.29 g m^{-2} l^{-1}). Irrespective of the water regime, LM75 was ranked first for GY WUE. The highest difference in WUE for the different plant parts occurred for roots biomass at 25FC with values between 0.66 g m^{-2} l^{-1} LM75 and 1.5 g m^{-2} l^{-1} for LM26, a 130% difference significant at $p<0.05$. In contrast, at 75FC the highest WUE for RB was 1.56 g m^{-2} l^{-1} obtained for BW140; while the lowest value 0.54 g m^{-2} l^{-1} (185% lower) was obtained for LM48 (Fig 5.1). Plant carbon stocks varied within the plant parts (Fig 4.2). Overall, the study cultivars only slightly differed in their ability to store carbon in their shoots, with for instance SCs values at 25FC ranging from 0.82 g C m^{-2} l^{-1} for LM71, to 1.13 g C m^{-2} l^{-1} for LM26 and BW141, i.e. a 38% difference. In contrast, WUE for RCs at 75FC varied between 0.19 g C m^{-2} l^{-1} for LM47, LM48 and LM26 to 0.44 g C m^{-2} l^{-1} (BW140), i.e. a 131% difference.

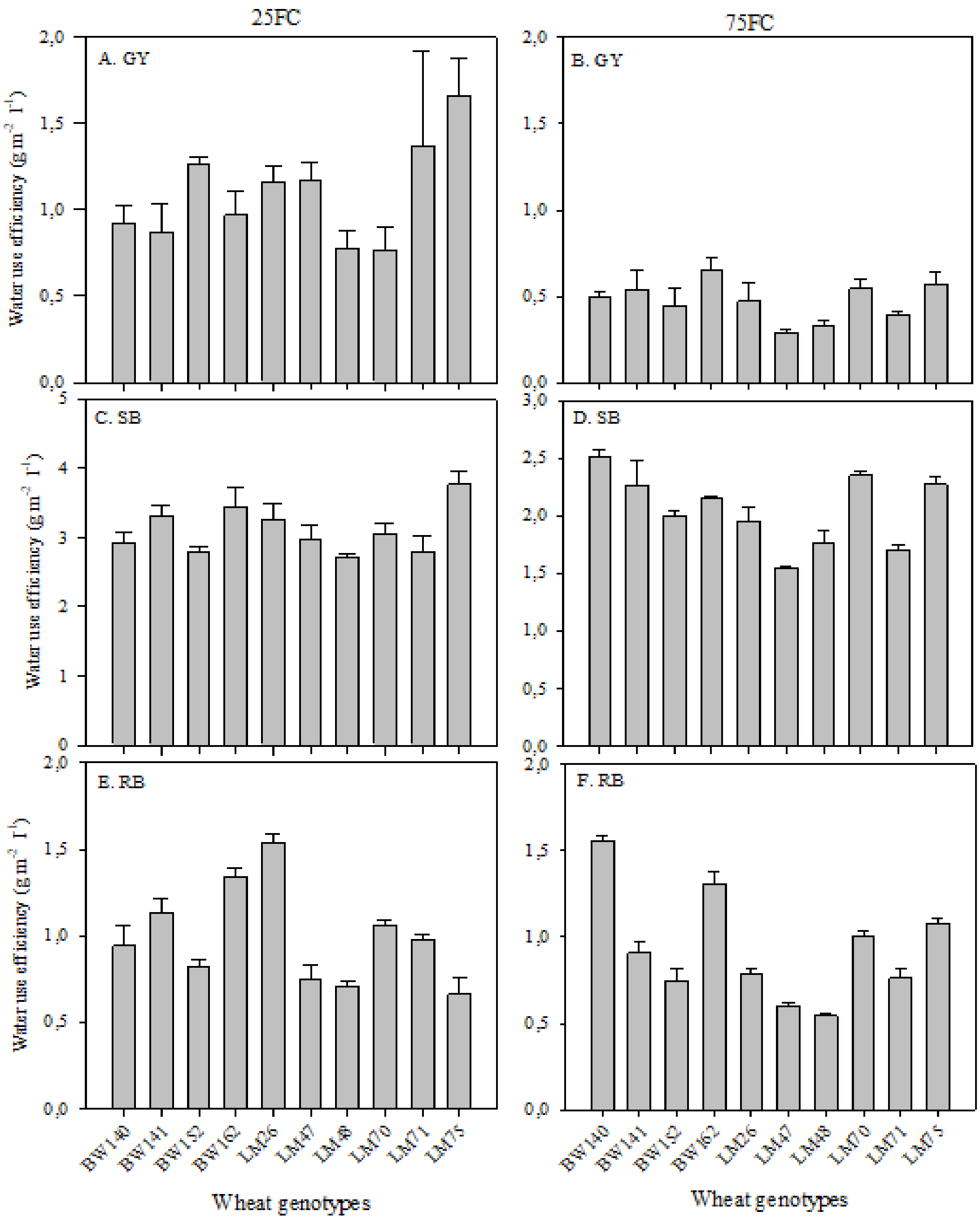


Figure 4.1. Water use efficiency for grain yield, shoot and root biomass of 10 selected wheat genotypes under two water regimes

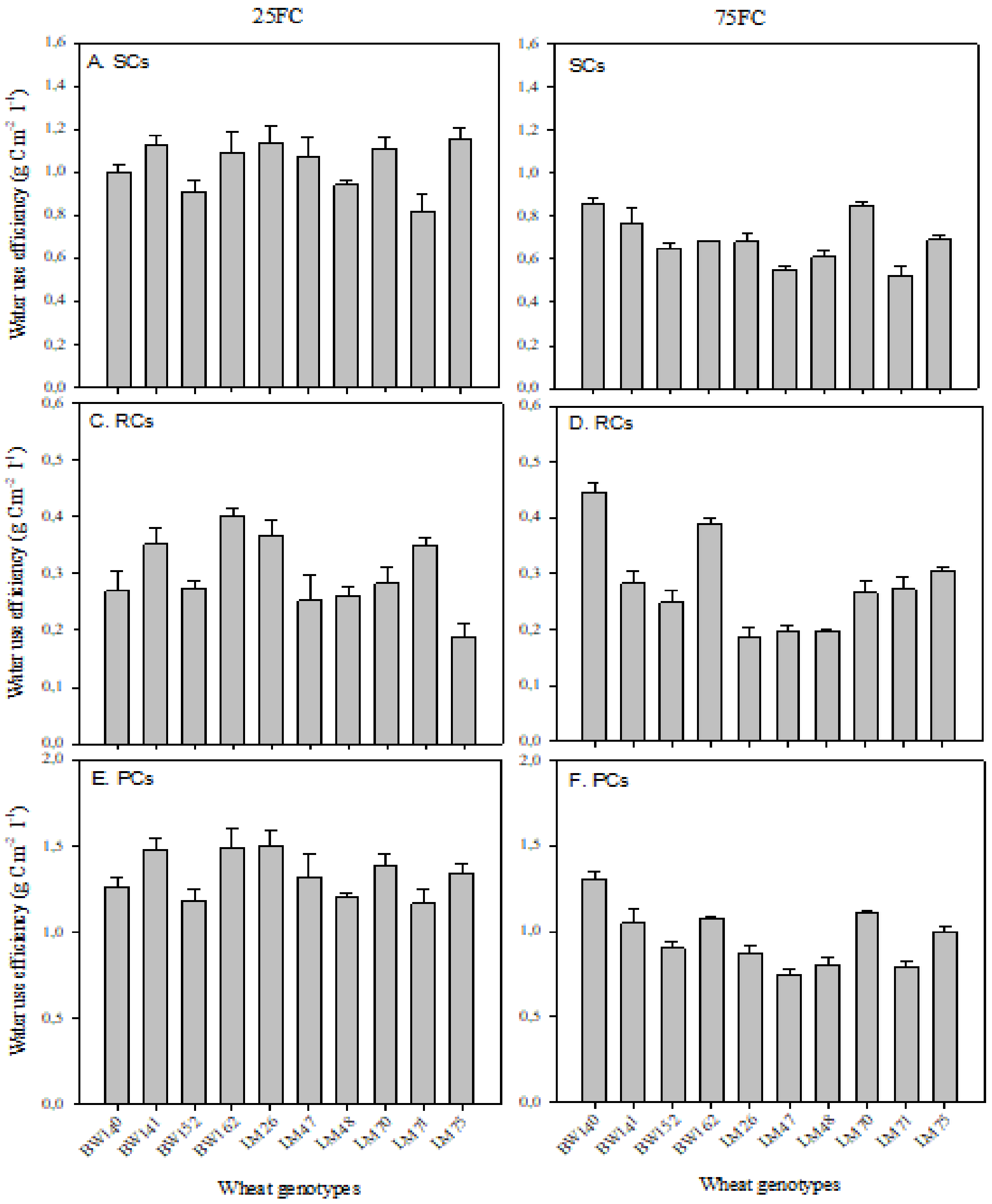


Figure 4.2. Water use efficiency for carbon stocks in different parts of wheat

4.4. Discussion

4.4.1. Genotypic variations in biomass and agronomic performance

The 100 genotypes accumulated different biomass, which they allocated in variable proportions between roots and shoots ($p<0.05$). This agreed with Akman et al. (2017), who reported significant genotypic variation among 47 wheat genotypes evaluated under field conditions in Turkey.

Differences in performance among genotypes reflects genetic diversity (Bhutta et al., 2006), which is a consequence of variable genetic background of the genotypes. Twenty-one genotypes were from a drought tolerant nursery while 75 originated from the heat tolerant nursery of CIMMYT resulting in differences in their agronomic performance. The heat tolerant lines such as BW152, LM47, BW141, BW162 had higher shoot biomass compared to the drought tolerant genotypes such as LM48, LM71 and LM75; showing that ability to tolerate heat stress conferred an advantage in above ground biomass production. The high total and shoot biomass production by Triticale compared to wheat concurred with reports that Triticale has higher potential for biomass production which was attributed to its higher solar radiation conversion efficiency of 3.2 g MJ^{-1} compared to 2.0 g MJ^{-1} exhibited by wheat under similar conditions (Estrada-Campuzano et al., 2012).

Root biomass production was significantly different among the 100 genotypes, agreeing with a report by Nevo and Chen (2010) who asserted that there is wide genetic variability in root biomass production in wheat that is yet to be exploited in breeding programs. The range of root biomass found in this study differed from the range of 97 to 1176 g m^{-2} reported by Waines et al. (2012) under greenhouse condition, but confirms the existence of genotypic variation. The differences in root biomass among the CIMMYT accessions, French varieties and Triticale are apparent due to their different adaptations. Triticale is well known to combine an aggressive rooting capability of one of its parents, rye (Gelalcha et al., 2007), while wheat genotypes adapted to temperate ecologies can have double the amount of root biomass of genotypes grown in warmer winters (Thorup-Kristensen et al., 2009) such as the heat and drought tolerant CIMMYT accessions. However, the French varieties and Triticale could not reach their full grain production potential due to a shorter growing season, which could explain their low grain yield in comparison with other genotypes.

Biomass allocation between roots and shoots expressed as R: S ratios also varied among the 100 genotypes, which encompasses a range of 1.00 to 1.36 reported in wheat varieties by Fang et al. (2017). The high R: S exhibited by the heat tolerant genotypes suggests their ability to maintain productivity even under combined drought and heat stress, which allowed them to allocate more biomass towards root development compared to the drought tolerant genotypes. Such genotypes with heat tolerance and high biomass accumulation are more relevant for sub-Saharan Africa where drought and heat stress often occur simultaneously.

4.4.2. The impact of water regime and environmental conditions on trait expression

Water regime had significant ($p<0.05$) impact on biomass (GY, PB, SB and RB), biomass allocation to roots and shoot (R: S) and other agronomic traits, as reported by other studies (e.g. Fang et al., 2017; Mwadzingeni et al., 2016). The high impact of drought stress on biomass production revealed that drought stress imposed earlier during pre-anthesis vegetative stages affects all biomass and growth parameters, whereas other studies focused on terminal drought imposed at anthesis or post-anthesis (e.g. Foulkes et al., 2007; Mwadzingeni et al., 2016). However, the changes in ranking of genotypes in biomass accumulation across water regimes disagreed with Foulkes et al. (2007) who asserted that genotypes with high performance under optimal soil water conditions still perform well under drought stress.

Biomass allocation to roots (R: S) was reduced by 14% due to drought stress, unlike previous studies which supported the optimal partitioning theory (Poorter and Nagel 2012; Eziz et al., 2017). The present findings agree with Vanaja et al. (2011) who also found that drought reduced R: S ratios by 7% in sunflower and 60% in maize. The reason for decreased R: S ratios can be explained by the fact that prolonged drought stress may have exceeded a threshold level causing the plant to completely lose its biomass allocation regulatory ability (Xu et al., 2010). Nonetheless, the higher impact of drought on biomass production (>30%) compared to R:S ratio (14%) agreed with a report by Lopez-Castaneda and Richards, (1994) who indicated that drought stress has more impact on dry matter production than its allocation to organs.

4.4.3. Associations between biomass and agronomic traits under contrasting water regimes

Drought stress affected the relationship among agronomic traits. Similarly, Mwadzingeni et al. (2016) found differences in trait relationship explained by PC under different water regimes. Under both drought stress and non-stress conditions, variations in yield were largely explained by changes in shoot biomass than any other parameter. This agreed with Dodig et al. (2012) and Sareen et al. (2014), who reported moderate to high correlations ($r > 0.3$) between grain yield and biomass production under stressed conditions. Reynolds et al. (2009) reckoned that the association between grain yield and shoot biomass is vital since future yield improvement in wheat will be accountable to increase in above ground biomass rather than shifts in biomass partitioning. This presents a conflict in efforts to increase soil C input via root biomass while attempting to maintain or increase yield potential simultaneously. Root biomass and biomass allocation between roots and shoots (R:

S) were more important under drought stressed conditions. This is vital because under stressed conditions root biomass is pivotal in accessing water resources (Poorter et al., 2012; Tatar 2016; Eziz et al., 2017). In concurrence, Atta et al. (2013) attributed 45% of variance in grain yield to root traits. Similarly, Fang et al. (2017) reported significant correlation of 0.24 between root biomass and grain yield under irrigation and even stronger correlation of 0.78 under lower soil water availability in a wheat rain-fed system. Wheat genotypes behaved more or less the same way under field conditions as they did in the glasshouse so there was no need to duplicate discussion of trends under field conditions.

4.4.4. Water use efficiency for grain yield, biomass production and carbon storage

The best cultivar for WUE for GY was LM75 followed by LM71 while LM 70 and LM 48 had the lowest WUE for GY, showing that WUE exhibited genotypic variation. Similarly, it has been reported that varietal differences in WUE exist in wheat and other crops such as rice and maize (Zwart and Bastianssen, 2004). Under well-watered conditions, WUE was relatively lower than drought stressed conditions. Genotype BW162 exhibited the highest WUE for GY whilst LM47 had the lowest under well-watered conditions, which shows that genotypic variation for WUE still exists but responds to environmental conditions. Several other reports have shown that water supply has a significant impact on WUE in crops with a general increase in WUE as water supply decreases (e.g. Erdem et al., 2001; Rusere et al., 2012; Mabhaudhi et al., 2013; Chibarabada et al., 2015). Sinclair (2018) explained that drought stimulates the closure of stomata leading to decrease in transpirational water loss and a subsequent improvement in yield or biomass productivity per unit of water. Irrespective of the water regime, LM75 was ranked first for GY WUE. This shows that its WUE capacity is generally stable, showing static stability, which means it is less affected by water supply. Such a genotype with static stability for WUE will be suitable for environments with highly unpredictable rainfall or water availability to ensure reasonable yield or biomass production. The highest difference in WUE for the different plant parts occurred for shoots biomass showing that water stress has higher impact on shoot biomass than root biomass. Vegetative growth is known to be more sensitive to water stress compared to root production but less sensitive compared to grain production. In contrast, at 75FC the highest WUE for RB was 1.56 g m^{-2} l^{-1} obtained for BW140; while the lowest value 0.54 g m^{-2} l^{-1} (185% lower) was obtained for LM48 (Figure 25).

4.5. Conclusion

There was significant variation for biomass and grain yield production among the genotypes showing that there were opportunities for selecting superior genotypes. Concomitant to the genotypic variation for biomass and yield production, the genotypes exhibited differences in water use efficiency. Water use efficiency for biomass and grain yield production decreased with higher water availability.

Chapter 5 SELECTION OF WHEAT GENOTYPES FOR IMPROVED WUE AND ATMOSPHERIC CARBON SEQUESTRATION

Abstract

Sequestration of atmospheric carbon (C) into plants and ultimately to soils is becoming a credible strategy to mitigate against climate change and alternatively to restore land productivity as soil carbon is key for soil fertility and water storage. While some studies have compared the ability of existing crops to store carbon and allocate it to the soil through roots, yet the variations between crop genotypes have received less attention. The objective of this study was to compare the atmospheric carbon sequestration of selected wheat genotypes under different scenarios of soil water availability. The experiments were set up under field and greenhouse conditions with 100 wheat genotypes grown at 25% field capacity (drought stressed) and 75% (non-stressed) and using an alpha lattice with 10 blocks and 10 genotypes per block, replicated twice. The variables considered were as follows: plant shoot biomass (PSB), root (PRB), grain yield (GY), shoot carbon stock (SCS), root (RCS), root to shoot ratios (R:S), and water use efficiency (WUE) for GY and biomass production, and carbon storage. Genotypic variation existed for all measured traits useful for improving drought tolerance, while the calculated R:S values can improve accuracy in estimating C sequestration potential of wheat. Wheat genotypes LM26, LM47, BW140, LM70, LM48, BW152, LM75, BW162, LM71 and BW141 were selected for further development based on their high total biomass production, grain yield potential and genetic diversity under drought stress. These were used for further plant C assessment

Keywords: agronomic traits, grain yield, genotype by environment interaction, root to shoot ratio, water stress

5.1. Introduction

Evidence point to agriculture as the main source of greenhouse gases (GHGs) and climate change. This started several thousand years ago by the human-induced conversion of natural ecosystems (e.g. forests and grasslands) into croplands. Following the conversion, most of the biomass C and the soil C pool, which consisting of plant and animal residues at various stages of decomposition, have been emitted to the atmosphere through mineralization. Because the soil C pool has been largely depleted and shows dynamic and direct exchanges with the atmosphere and the biosphere, there is a growing belief that C sequestration into soils could exert a significant role in mitigating greenhouse gases emissions and thus constitute a credible solution to mitigate climate change (Paustian et al., 2016). Because soils store about three times more carbon than the atmosphere, a relatively small increase in soil carbon stocks could be a *game changer* for climate change. Indeed, Minasny et al. (2017) recently estimated that considering annual greenhouse gas emissions from fossil carbon of 8.9 giga tonnes C (8.9 × 1015 g) and an actual soil C stocks to 2 m deep of 2400 giga tonnes, Soil C stocks would have to be increased globally by an average of 8.9/2400 or 0.4% per year. Such an increase is not only expected to mitigate against climate change but also to provide additional benefits as soil organic matter of which carbon is a main component exerts numerous positive effects on soil quality, soil fertility, soil water holding capacity and biodiversity, amongst others. Enhancing soil C stocks by an average 0.4% per year equates an average 0.6 tonnes of C per hectare per year for all terrestrial soils which vary widely in their capacity to store carbon from deep tropical clayey soils to deserts.

Several studies have reported annual C sequestration rates in soils of between 0.2 to 0.5 tonnes C per hectare or even higher after the adoption of best management practices. These practices include land rehabilitation, conservation agriculture with reduced tillage or tillage cessation, alone or in combination with intercropping or cover cropping with legumes, crop residue management and addition of organic material. Amongst the latest studies, Luca et al. (2018) showed that green trashing in Brazilian sugar cane soils increases carbon sequestration by as much as 10.8% in the most carbon depleted soils, which were sandy soils with low concentration in chemical elements such as calcium, magnesium and potassium. According to these authors the resulting soil carbon sequestration rate for Brazil where 8.8 million hectares are under green-trashing of sugarcane residues is 9.3 million tons C y^{-1}. However, several authors concluded that land management may have a much less impact on the cycle of carbon and is thus expected to play a limited role in the mitigation against human-induced global changes such as global warming and land degradation. Poulton et al. (2018), who investigated 114 treatment comparisons found out severe limitations to achieving the 0.4% target over large areas because of the inability of farmers to access the necessary resources (e.g. insufficient manure). Indeed, these authors showed that SOC increases from 1.8 to 4.3% can only be achieved by application of 35 tonnes of fresh material per hectare. It appears that several agriculture-based greenhouse gas mitigation activities have been proposed but either their effectiveness or adoption at large scale remains a challenge. More research needs to be performed on the capacity of agriculture and in particular on the plant-soil system to capture more of the exceeding atmospheric carbon and that will be further translated into effective greenhouse gas policies.

Plants are crucial in the global C cycle because the amount of CO_2 that passes through them reaches each year 10% of all of the atmospheric CO_2 (Raich and Potter, 1995). Therefore, to foster the ability of plants to store atmospheric C into their body and ultimately transfer it to the soil may constitute a credible alternative to mitigate against global changes. Because most of the soil C comes from the decomposition of plant C and especially from root activity and residue decomposition (Yang et al., 2012), consequently selection of "carbon superior" crops and crop cultivars should be promoted. The R:S ratios reported for annual cereal crops such as wheat are comparably lower than for perennial grasses. For example, Amanullah and Stewart (2013) reported R:S ratios of 0.41 and 0.29 for sorghum and maize, respectively while Yang et al. (2010) reported a mean of 0.25 for maize and wheat. Bolinder et al., (2002) reported R:S ratios of up to 7.00 for forage grasses compared to a range of 0.1 to 0.5 cited for many annual crops including cereals and legumes (Mathew et al., 2017). There are also variations in R:S within single species which can be attributed to genotypic differences. Intra-specific variation in any trait results from genotypic differences among individuals in a given species. King et al. (2007) asserted that there is wide genotypic variation for biomass allocation. Fang et al. (2017) found that a R:S ratio of 1.13 in a wheat landrace compared with 0.61 and 0.81 found in two modern cultivars. Similarly, Siddique et al. (1990) reported higher R:S ratios ranging between 0.74 and 1.18 for obsolete varieties than modern cultivars of wheat which ranged from 0.72 to 0.84. Most crop improvement programs in wheat have focused on channeling more biomass towards economic traits such as grain and above ground biomass for food, feed and biofuel production without improving root systems leading to low R:S in modern cultivars compared to landraces (Wasson et al., 2012). However, variations do occur even in genotypically identical individuals due to environmental factors.

Biomass allocation is also affected by genotype x environment interaction. Biomass allocation pattern indicate environmental plasticity to soil properties, temperature and soil water availability (Sánchez et al., 2014, Pittelkow et al., 2015). Water availability is a key factor influencing biomass allocation. Drought stress is known to reduce crop growth; for example, Perdomo et al. (2015) reported biomass reductions of 60% in maize and rice, and 90% in wheat. However, drought stress tends to increase R:S ratios as reported by Tatar et al. (2016) who found 7% higher ratio in wheat produced under 25% soil water content than wheat grown at 75% soil water content. However, Vanaja et al. (2011) reported a contrasting trend when they recorded 60 and 7% reduction in R:S for maize and sunflower, respectively, as a result of 30% reduction in soil water content. Biomass allocation also vary with intensity and duration of drought stress (Farooq et al., 2009). Instantaneous and short-lived drought spells may not cause a significant shift in biomass allocation, while excessive drought stress beyond a threshold level causes plants to lose their biomass allocation regulatory ability completely (Xu et al., 2010). Conversely, Sharp and Davies (1989) indicated that soil water stress has greater negative impact on shoot than root growth which led to a reduction in R:S. Several, studies have investigated variations in R:S between plant species but little is known about intra-specific variations. The present study aims at filling an important information gap. Therefore, the objectives of this study were to characterize intra-specific variations in R:S ratios and agronomic performance of diverse wheat genotypes sourced from the International Maize and Wheat Improvement Centre (CIMMYT) and subjected to water stressed and non-stressed conditions. The results can be helpful to crop breeders in evaluating diversity in biomass allocation and agronomic performance, which is important for developing varieties with greater water use efficiency and drought tolerance for grain yield and C sequestration into soils.

5.2. Materials and methods

5.2.1. Plant materials

One hundred genotypes, consisting of 97 drought and heat tolerant wheat accessions, 2 commercial wheat varieties from France and Triticale were evaluated (Supplementary Table S1). The drought and heat tolerant genotypes were obtained from CIMMYT. The CIMMYT genotypes were used owing to their genetic variability for rooting abilities and breeding history for drought tolerance. The French varieties and Triticale were used as comparative controls because they are known for their high rooting capacities. The French varieties, which are winter wheat genotypes, have twice their rooting capacity of wheat grown in warmer winters (Thorup-Kristensen et al., 2009), while Triticale has an aggressive root system inherited from rye (*Secale cereale*) (Gelalcha et al., 2007).

5.2.2. Growing conditions and trial management

5.2.2.1. Greenhouse experiments

The experiments were carried out at the Controlled Environment Facility of the University of KwaZulu-Natal Pietermaritzburg Campus. The greenhouse experiment was conducted using a 10×10 alpha lattice design with two replications. Ten seeds were sown in each pot and thinned to 8 plants per pot, 3 weeks after emergence. Ten pots were allocated per incomplete block and genotypes were randomly assigned to pots to minimize the experimental error associated with water discharge from the drip irrigation. Fertilizer was applied through automated drip irrigation at a rate of 300 kg N ha^{-1} and 200 kg P_2O_5 ha^{-1}. The different water regimes were initiated 6 weeks after planting to ensure good establishment but also to ensure early exposure of all growth stages to drought. In the well-watered regime, the plants were watered to field capacity (FC) whenever average soil water content fell to 80% of FC, while in the water stress conditions volumetric soil water content was allowed to drop to 30% of FC before watering to FC. The soil water content was monitored by a soil moisture probe and weighing of the pots. The two watering treatments were maintained until maturity (~120 days). The first greenhouse experiment (GH1) was carried out in summer (October 2016 to February 2017), while the second one (GH2) ran concurrently with an open field experiment in a winter season (May to September 2017).

5.2.2.2. Field experiment

The field experiment was carried out at the University of KwaZulu-Natal Ukulinga Research farm (LAT: 29.667^{o} LON: 30.406^{o} and ALT: 811 m) using a 10×10 alpha lattice design with two replications. Long-term average temperature and rainfall for Ukulinga are 18°C and 738 mm, respectively. The field was ploughed in May 2017 to a depth of 30 cm and custom-made plastic mulch was used to exclude rainwater. Three seeds were planted per station at 10 cm intra-row spacing and 30 cm between rows soon after ploughing. Each row consisted of 10 genotypes and was treated as an incomplete block. Basal fertilizer was applied at a rate of 120:30:30 kg ha^{-1} (N:P:K). Other agronomic practices were as per normal wheat production practice in South Africa (DAFF, 2010). Irrigation was applied through a drip irrigation system with the aim to maintain soil water content at FC in the well-watered regime. Under the drought stress treatment, irrigation was withheld 5 weeks after crop emergence until just before signs of permanent wilting were observed upon which irrigation was reinstated. This differs from the 80 and 30% FC soil water regimes maintained in the greenhouse because it is more difficult to determine field capacity and regulate soil water content appropriately under field conditions compared to a controlled greenhouse environment. During the field experiment, irrigation was withheld before anthesis to induce drought stress in a way that simulated *in situ* wheat production under field conditions.

5.2.3. Data collection

Plant parts for each plot and pot were separated at maturity into grain, shoot and root and oven dried at 60°C for 72 hrs to measure the dry weight. The weight was converted to gram per square meter (g m^{-2}) accordingly using the plant population of 128 and 134 plants per square meter for the greenhouse and field experiments, respectively. Root: shoot (R:S) ratio and total biomass (PB) were computed after determining GY, RB, and SB. 250 kernels of wheat for each genotype were weighed in grams and the weight was multiplied by 4 to obtain the thousand kernel weight (TKW).

5.2.4. Data analysis

Analysis of variance (ANOVA) was conducted using the lattice procedure using Genstat 18th edition (Payne et al., 2017). In addition, the means of genotypes and the different water regimes were separated by Fischer's unprotected least significant difference (LSD) at 0.05 significance level to quantify the effects of genotype, environment and water regime. A multivariate procedure for hierarchical clustering was performed based on phenotypic data combined across water regimes and sites to group the genotypes for their similarity. A dendrogram was derived from a Euclidean similarity matrix using the Unweighted Pair Group Method with Arithmetic mean algorithm (UPGMA). Genotypes with high grain yield and biomass production in each cluster were selected (to capture high performance and as much diversity as possible) for further development.

5.3. Results

5.3.1. Cluster analysis

The dendrogram resulting from the UPGMA revealed two major distinct clusters of the 100 genotypes based on their similarity in agronomic performance (Fig 5.1). The first cluster comprised of 97 genotypes that were further divided into subgroups A and B at 0.95 similarity. They were all from CIMMYT heat and drought tolerant genotypes except LM70, which was a local line. The sub-cluster A was further divided into 4 clusters, which was comprised of genotypes such as LM26, BW141, BW140, LM70 and LM48. Sub-cluster B was further divided into 2 clusters with genotypes such as BW152, LM47, LM75, BW162 and LM71. Cluster 2 comprised of three genotypes, Triticale, Sossognon and Arenza. After clustering, the genotypes were ranked based on the total biomass production under drought stressed condition. The best performing genotypes in each cluster were selected for further study.

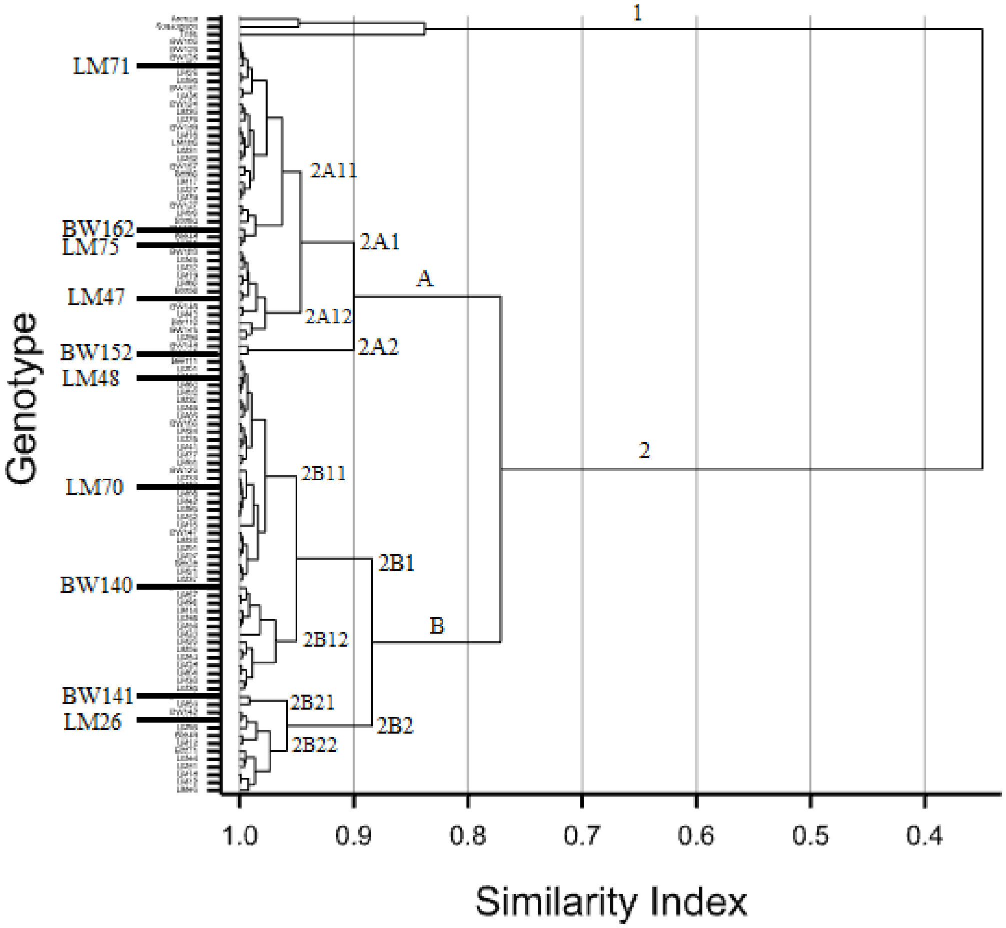


Figure 5.1. Dendrogram showing clusters according to phenotypic relatedness of 100 genotypes evaluated across greenhouse and field

5.3.2. Variation in biomass and grain yield production

The average grain yield (GY) amongst the initial set of 100 wheat genotypes was 1387 g m-^{2} with a standard error of ± 84 g m-^{2} (Table 5.1). GY varied by a 62 factor from 75 to 4696 g m-^{2} and showed a positively skewed distribution (Skew=1.53). Total plant biomass (PB) was much less variable than GY as values ranged from 1967 to 13528 g m-^{2}, a 6.8 times difference, which was significant at $P<0.001$. The 100 selected genotypes had an average root to shoot ratio for biomass of 0.12, meaning that they allocated an average 12% of their total biomass to their roots, with values from 3 to 38% (Table 5.1). The sub-set of 10 wheat genotypes showed much less variations in grain yield and plant total biomass and biomass allocation with for instance GY from 621 to 4383 g m-^{2} (a 7.0-fold increase *vs* 62 times for the 100 genotypes), but similar variations in PB with values between 2475 and 13529 g m^{2} (5.4 *vs* 6.8) and in R: S (Table 5.1).

Table 5.1. Summary statistics for grain yield, selected morphological variables, and plant carbon stocks for the 100 and selected 10 wheat varieties grown in the field under different water regimes.

	GY	SB	RB	PB	R:S	SC$_C$	RC$_C$	SC$_S$	RC$_S$	PC$_S$
	-----------------------g m^2 -----------------------					----------%--------		--------------g m^2 ----------------		
100 genotypes										
Mean	1387	2498	305	4189	0,12					
Median	1309	2332	263	3930	0,11					
Minimum	75	1179	65	1976	0,03					
Maximum	4696	8658	1219	13529	0,38					
Q1	959	1827	189	3026	0,09					
Q3	1644	2908	365	4893	0,15					
CV%	47	37	57	37	41					
SEM	84	121	22	200	0,01					
Skewness	1,53	2,04	2,03	1,82	1,67					
Kurtosis	4,35	7,15	5,76	5,66	4,39					
10 selected genotypes										
Mean	1882	3169	356	5408	0,12	34	30	1067	106	1174
Median	1624	2929	326	5214	0,10	34	30	940	93	1061
Minimum	621	1598	152	2475	0,06	30	21	534	45	582
Maximum	4383	8658	1006	13529	0,26	37	39	2869	302	3022
Q1	1349	2317	249	3952	0,09	32	28	792	72	862
Q3	2500	3691	427	6140	0,13	35	33	1210	136	1384
CV%	45	44	47	42	35	6	14	44	49	42
SEM	109	182	22	290	0,01	0	1	61	7	64
Skewness	1,05	2,03	1,91	1,65	1,60	-0,29	-0,19	1,98	1,86	1,77
Kurtosis	1,03	6,08	5,89	4,19	3,40	-0,85	-0,01	5,62	5,17	4,71

Q1: 25th percentile, Q3: 76th percentile, CV: coefficient of variation, SEM: standard error of mean, GY: grain yield, SB: shoot biomass, RB:Root Biomass, PB: Total Plant Biomass, SC$_C$: shoot carbon content, RCc: Root carbon content, SCs: Shoot carbon stocks, RCs: Root carbon stocks and PCs: Plant carbon stocks

5.3.3. Variations in the cultivar's ability to store carbon

The average carbon content for the selected 10 genotypes was 34±0.9% in the shoots (SC_C) and 30±1.1% in the roots (RC_C), which corresponded to a significant difference at $P<0.05$. SC_C ranged from 30 to 37%, while RCc varied from 21 to 39%, the later corresponding to a 90% increase. However, the carbon stocks in the shoots, roots and in the total plant biomass showed greater variations. For instance, the total plant carbon stocks (PCs) exhibited a mean of 1174± 64 g m^{-2} with values from 582 to 3022 g m^{-2} and corresponding to a 500% difference (Table 5.1). Most of the PCs was contributed by the carbon stocks in the shoots (91%), while only 9% of the carbon stocks was contributed by the roots.

5.3.4. Variations of crop agronomic traits with water regimes and environment

The average grain yield for the selected 10 wheat genotypes increased from 876±116 g m^{-2} under 25% field capacity (25FC) to 1074±130 g m^{-2} for 75FC, which corresponded to a 22% increase (Table 5.2). At the same time the total plant biomass (PB) increased by 74% (from 3598±165 to 6245±249 g m^{-2}) (Table 5.2). This was concomitant to an increase in the R:S ratio (from 0.3 to 0.39). The greatest increase occurred for root C stocks (140%), followed by plant C stocks (89%). In contrast, the C content in the plant tissues was not affected by the water regime (Table 5.2).

The environment (field *vs* glasshouse) had a significant impact on the study variables (Table 5.3). For instance, GY at 25FC increased 5.2 folds from glasshouse to the field (326 *vs* 1700 g m^{-2}). The increase was of 4.6-fold at 75FC. The C content in plant parts was not impacted by the environment. The shoot biomass overly increased from glasshouse to field at 25FC. In contrast, root biomass and root C stock decreased for both water regimes (Table 5.3).

Table 5.2. Summary statistics for grain yield, selected morphological variables and plant carbon of selected 10 wheat varieties grown in the field under different water regimes

	GY	SB	RB	PB	R:S	SC_C	RC_C	SC_S	RC_S	PC_S
	-----------------------	g m^2	-----------------------			----------%	--------	--------------	g m^2	----------------
25% Field capacity										
Mean	876	2127	596	3598	0,30	33	31	708	181	889
Median	549	2012	569	3289	0,30	34	31	684	179	858
Minimum	58	1349	152	2156	0,08	25	21	409	45	582
Maximum	4488	3775	1263	8100	0,70	37	39	1215	379	1409
Q1	242	1677	301	2707	0,13	32	28	578	97	752
Q3	1345	2344	837	3890	0,45	35	33	781	238	1005
CV%	102	27	53	35	59	8	14	27	52	21
SEM	116	74	41	165	0.02	0	1	24	12	24
Skewness	1,85	1,10	0,47	1,67	0,30	-1,26	-0,24	1,00	0,29	0,60
Kurtosis	4,29	0,54	-0,81	2,66	-1,26	2,10	-0,26	0,81	-0,91	0,26
75% Field capacity										
Mean	1074	3721	1450	6245	0,39	34	31	1248	436	1684
Median	643	3418	1462	5508	0,46	34	31	1183	472	1653
Minimum	136	1954	135	3555	0,06	25	21	668	48	715
Maximum	4383	8658	3575	13529	0,78	37	39	2869	1059	3022
Q1	344	2977	425	4967	0,12	32	28	994	133	1442
Q3	1542	4393	2142	7517	0,59	35	33	1392	633	2070
CV%	94	30	71	31	62	7	14	31	69	32
SEM	130	145	133	249	0,03	0	1	50	39	68
Skewness	1,49	1,81	0,48	1,35	-0,10	-1,09	-0,25	1,74	0,34	0,49
Kurtosis	1,85	6,31	-0,82	2,64	-1,57	1,97	-0,24	5,34	-1,02	0,02

Q1: 25th percentile, Q3: 76th percentile, CV: coefficient of variation, SEM: standard error of mean, GY: grain yield, SB: shoot biomass, RB:Root Biomass, PB: Total Plant Biomass, SC_C: shoot carbon content, RCc: Root carbon content, SCs: Shoot carbon stocks, RCs: Root carbon stocks and PCs: Plant carbon stocks

Table 5.3. Summary statistics for grain yield (GY), selected morphological variables and plant carbon stocks for selected 10 wheat varieties grown in the field and glasshouse under different water regimes

	GY	SB	RB	PB	R:S	SC_C	RC_C	SC_S	RC_S	PC_S
	-----------------------g m^2 -----------------------					----------%--------		--------------g m^2 ----------------		
					Glasshouse					
25% Field capacity										
Mean	326	1873	792	2992	0,43	33	31	625	239	865
Median	266	1826	762	2918	0,44	34	31	621	225	854
Min	58	1349	368	2156	0,15	25	21	409	103	592
Max	869	2415	1263	3983	0,70	37	39	805	379	1109
CV%	68	24	36	24	34	19	23	25	34	23
SEM	28	57	37	94	0,02	1	1	20	10	26
75% Field capacity										
Mean	414	3726	2149	6289	0,57	33	31	1243	647	1890
Median	397	3440	1823	5508	0,57	34	31	1186	605	1716
Min	136	2779	1290	4531	0,37	25	21	851	333	1324
Max	811	5192	3575	9305	0,78	37	39	1822	1059	2881
CV%	50	26	38	30	26	19	23	27	35	28
SEM	27	127	104	247	0,02	1	1	44	29	69
					Field					
25% Field capacity										
Mean	1700	2507	302	4508	0.12	33	31	832	95	927
Median	1401	2534	277	4042	0.12	34	31	827	80	968
Minimum	621	1598	152	2475	0.08	25	21	534	45	582
Maximum	4488	3775	642	8100	0.19	37	39	1215	214	1409
CV%	53	27	39	34	24	8	16	26	47	27
SEM	116	87	15	199	0.004	0.36	0.62	28.10	5.79	32.30
75% Field capacity										
Mean	2062	3714	403	6179	0.12	34	31	1254	121	1375
Median	1720	3154	382	5462	0.10	35	31	1052	112	1170
Minimum	1118	1954	135	3555	0.06	30	21	675	53	728
Maximum	4383	8658	1006	13529	0.26	37	39	2869	302	3022
CV%	44	42	44	39	43	6	15	42	43	39
SEM	117	202	23	313	0.01	0.26	0.61	67.57	6.69	69.93

Q1: 25th percentile, Q3: 76th percentile, CV: coefficient of variation, SEM: standard error of mean, GY: grain yield, SB: shoot biomass, RB:Root Biomass, PB: Total Plant Biomass, SC_C: shoot carbon content, RCc: Root carbon content, SCs: Shoot carbon stocks, RCs: Root carbon stocks and PCs: Plant carbon stocks

5.3.5. Water use efficiency for grain, biomass production and carbon storage

The average WUE for GY was 0.74 g m^{-2} l^{-1} in the glasshouse under 25FC, with values from 0.12 to 2.10 g m^{-2} l^{-1} (Table 5.4). In the glasshouse and under 75FC WUE for GY ranged from 0.12 to 0.65 g m^{-2} l^{-1} with an average of 0.33 with a standard error of ±0.02. The WUE for total plant biomass (PB) was higher than that of shoots (WUE-SCS), roots (WUE-RB) and grain yield (WUE-GY), as it had an average of 6.58 g m^{-2} l^{-1} and 5.07 g m^{-2} l^{-1} in the glasshouse under 25% and 75% FC respectively. Also, the WUE of carbon was higher in the total plant (WUE-PSC) than in roots or shoots as it ranged from 1.16 to 2.68 g m^{-2} l^{-1} with an average of 1.89 g m^{-2} l^{-1} in the glasshouse under 25% FC (Table 5.4). The conditions were conducive for WUE of GY in the field, since the average was 1.44 and 0.62 g m^{-2} l^{-1} under 25 and 75% FC respectively which was higher than WUE of GY in the glasshouse (Table 5.4). The WUE for total biomass in the field (WUE-PB) ranged from 2.19 to 7.24 g m^{-2} l^{-1} and from 1.09 g m^{-2} l^{-1} to 3.90 g m^{-2} for 25% FC and 75% FC respectively. The WUE for total plant carbon stocks (WUE-PCS) in the field had an average of 0. 78 g m^{-2} l^{-1} and 0.41 g m^{-2} l^{-1} under 25% and 75% FC respectively (Table 5.4).

Table 5.4. Summary statistics for biomass and plant carbon WUE for the 10 selected wheat varieties grown in the field and glasshouse under different water regimes

	GY	SB	RB	PB	SC_S	RC_S	PC_S
	---------------------------------------$g\ m^2\ l^{-1}$---------------------------------------						
				Glasshouse			
25% Field capacity							
Mean	0.74	4.10	1.74	6.58	1.37	0.52	1.89
Median	0.55	4.16	1.71	6.49	1.38	0.53	1.93
Min	0.12	2.63	0.89	4.21	0.80	0.25	1.16
Max	2.10	5.84	3.06	9.63	1.95	0.77	2.68
CV	75	20	34	22	20	28	17
SEM	0.07	0.10	0.08	0.19	0.04	0.02	0.04
75% Field capacity							
Mean	0.33	3.00	1.74	5.07	1.00	0.52	1.52
Median	0.31	2.73	1.58	4.51	0.94	0.49	1.42
Min	0.12	2.28	0.94	3.56	0.64	0.30	1.10
Max	0.65	4.42	3.04	7.92	1.55	0.90	2.45
CV	51	26	39	31	27	36	29
SEM	0.02	0.10	0.09	0.20	0.04	0.02	0.06
				Field			
25% Field capacity							
Mean	1.44	2.11	0.25	3.80	0.70	0.08	0.78
Median	1.25	2.00	0.23	3.37	0.69	0.07	0.78
Min	0.57	1.35	0.13	2.19	0.42	0.04	0.46
Max	4.01	3.49	0.55	7.24	1.12	0.18	1.21
CV%	55	28	40	36	27	48	28
SEM	0.10	0.08	0.01	0.18	0.02	0.00	0.03
75% Field capacity							
Mean	0.62	1.11	0.12	1.84	0.37	0.04	0.41
Median	0.53	0.94	0.11	1.69	0.32	0.03	0.35
Min	0.33	0.60	0.04	1.09	0.21	0.02	0.22
Max	1.26	2.50	0.30	3.90	0.83	0.09	0.87
CV%	43	40	44	37	40	43	37
SEM	0.03	0.06	0.01	0.09	0.02	0.00	0.02

CV: coefficient of variation, SEM: standard error of mean, GY: grain yield, SB: shoot biomass, RB:Root Biomass, PB: Total Plant Biomass, SC_C: shoot carbon content, RCc: Root carbon content, SCs: Shoot carbon stocks, RCs: Root carbon stocks and PCs: Plant carbon stocks

5.3.6. Impact of water regime on biomass and agronomic performance

The performance of the 10 genotypes under each soil water regime across the different environments were recorded. Mean shoot biomass was 2127 g m^{-2} under non-stressed condition (Table 5.2). Genotype BW152 exhibited the highest shoot biomass under stressed condition while

LM30 had the highest reduction (61%) in shoot biomass due to drought stress. On average, shoot biomass was reduced by 28% under drought stress relative to non-stressed condition.

On average, root biomass was 1450 g m^{-2} under non-stressed compared to 596 g m^{-2} attained under drought stress conditions (Table 5.2). LM71 produced the lowest root biomass of 218 g m^{-2} under non-stressed condition, while LM54 had the lowest under drought stress conditions. Overall, drought stress significantly reduced root biomass by 23% compared to non-stressed condition.

Biomass allocation between roots and shoot (R:S) was not significantly different between stressed and non-stressed conditions (Table 5.2). Under non-stressed condition, R:S ranged between 0.06 and 0.78 compared to 0.08 and 0.7 under drought stressed condition. Drought stress reduced mean R:S ratios across genotypes by 40% compared to non-stressed condition.

The mean grain yield under non-stress condition was 1074 g m^{-2} compared to 876 g m^{-2} under drought stress condition. The genotypes exhibited wide variation in grain accumulation under both soil water regimes. Under non-stressed condition, grain yield ranged from 136 to 4383 g m^{-2} compared to 58 to 4488 g m^{-2} recorded under drought stress condition. On average, drought stress reduced grain yield by 35% across the different soil water regimes.

5.3.7. The variation in the cultivar's ability to store carbon as influenced by water regimes

The plant carbon stocks (PCS) ranged from 582 to 1409 g m^{-2} under water stress, which was just a 2 times difference, however the carbon stocks in the roots (RCS) ranged from 45 to 379 g m^{-2} which was 8 times difference which is much greater variation than that of carbon stocks in the whole plant (Table 5.2). For 75% FC, the total plant biomass had an average of 6245 g m^{-2} with a standard error of ± 249 g m^{-2} (Table 5.2). PB varied by a 4 times factor from 3555 g m^{-2} to 13529 g m^{-2}. However, the total biomass (PB) was higher in 75% FC compared to 25% with their values being 6245 and 3598 g m^{-2} respectively indicating a 2 times fold increase from 25% to 75% FC (Table 5.2).

The field conditions were more conducive than glasshouse conditions for grain yield production since the average grain yield produced were 1700 and 326 g m^{-2} for field and glasshouse respectively under 25% FC (Table 5.3). GY ranged from 58 to 869 g m^{-2} with a standard error of ±28 under 25% FC in the glasshouse, whilst it ranged from 621 to 4488 g m^{-2} with a standard error of ±116 under 25% in the field. However, the fold increase was higher in the glasshouse than in the field (15 vs 7). Total plant biomass (PB) ranged from 4531 to 9305 g m^{-2} at 75% FC in the glasshouse, whilst it ranged from 3555 to13529 g m^{-2} in the field (Table 5.3). There was a 4-fold increase of plant carbon stocks (PCS) under 75% FC in the field ranging from 728 g m^{-2} to 3022 g m^{-2} while there was a 2-fold increase on plant carbon stocks (PCS) at 75% FC in the glasshouse with the range of 1324 to 2881 g m^{-2}.

5.3.8. Water use efficiency for grain yield, biomass production and carbon storage

The WUE for grain yield (GY) was much higher at 25FC than at 75FC, with all the 25FC values above 0.75 g m^{-2} l^{-1} and 75FC values below 0.75 g m^{-2} l^{-1} (Fig 5.2). Under stressed conditions (25FC) the two best cultivars for WUE for GY were LM75 with 1.76 g m^{-2} l^{-1} followed by LM71 with 1.37 g m^{-2} l^{-1}. LM 70 and LM 48 had the lowest WUE for GY with values of 0.76 and 0.78 g m^{-2} l^{-1} respectively. This corresponded to a maximum 130% difference in WUE for GY. Under well-watered conditions (75FC), BW162 exhibited the highest WUE for GY amongst all the cultivars (0.66 g m^{-2} l^{-1}), whilst LM47 had the lowest (0.29 g m^{-2} l^{-1}). WUE for total plant biomass (PB) at 25FC was, as for GY the highest for LM75 (6.1 g m^{-2} l^{-1}) and the lowest value of 4.2 g m^{-2} l^{-1} was obtained for LM48 which corresponded to a 45% difference. The highest difference in WUE for the different plant parts occurred for roots biomass with values between 0.66 g m^{-2} l^{-1} LM75 and 1.5 g m^{-2} l^{-1} for LM26, a 130% difference significant at $p<0.05$. In contrast, at 75FC the highest WUE for RB was with 1.56 g m^{-2} l^{-1} obtained for BW140. The lowest value (0.54 g m^{-2} l^{-1}) was 185% lower and was obtained for LM48 (Fig 5.2).

Plant carbon stocks varied within the plant parts (Fig 5.3). Overall, the study cultivars only slightly differed in their ability to store carbon in their shoots, with for instance SCs values at 25FC between 0.82 g C m^{-2} l^{-1} for LM71, and 1.13 g C m^{-2} l^{-1} for LM26 and BW141, i.e. a 38% difference. In contrast, WUE for RCs at 75FC varied between 0.19 g C m^{-2} l^{-1} for LM47-48 and LM26, and 0.44 g C m^{-2} l^{-1} (BW140), i.e. a 131% difference.

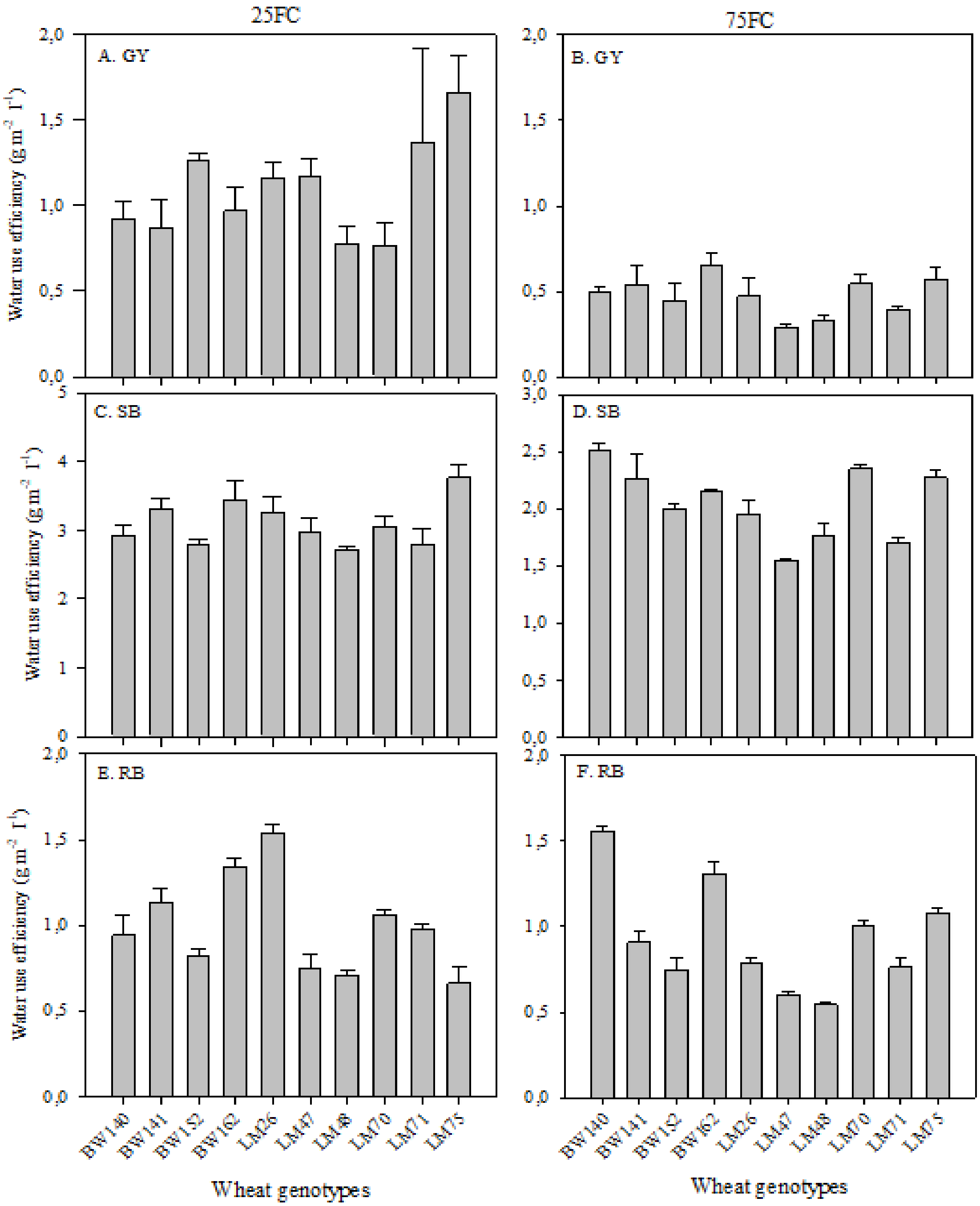


Figure 5.2. Water use efficiency for grain yield, shoot and root biomass of 10 selected wheat genotypes under two water regimes.

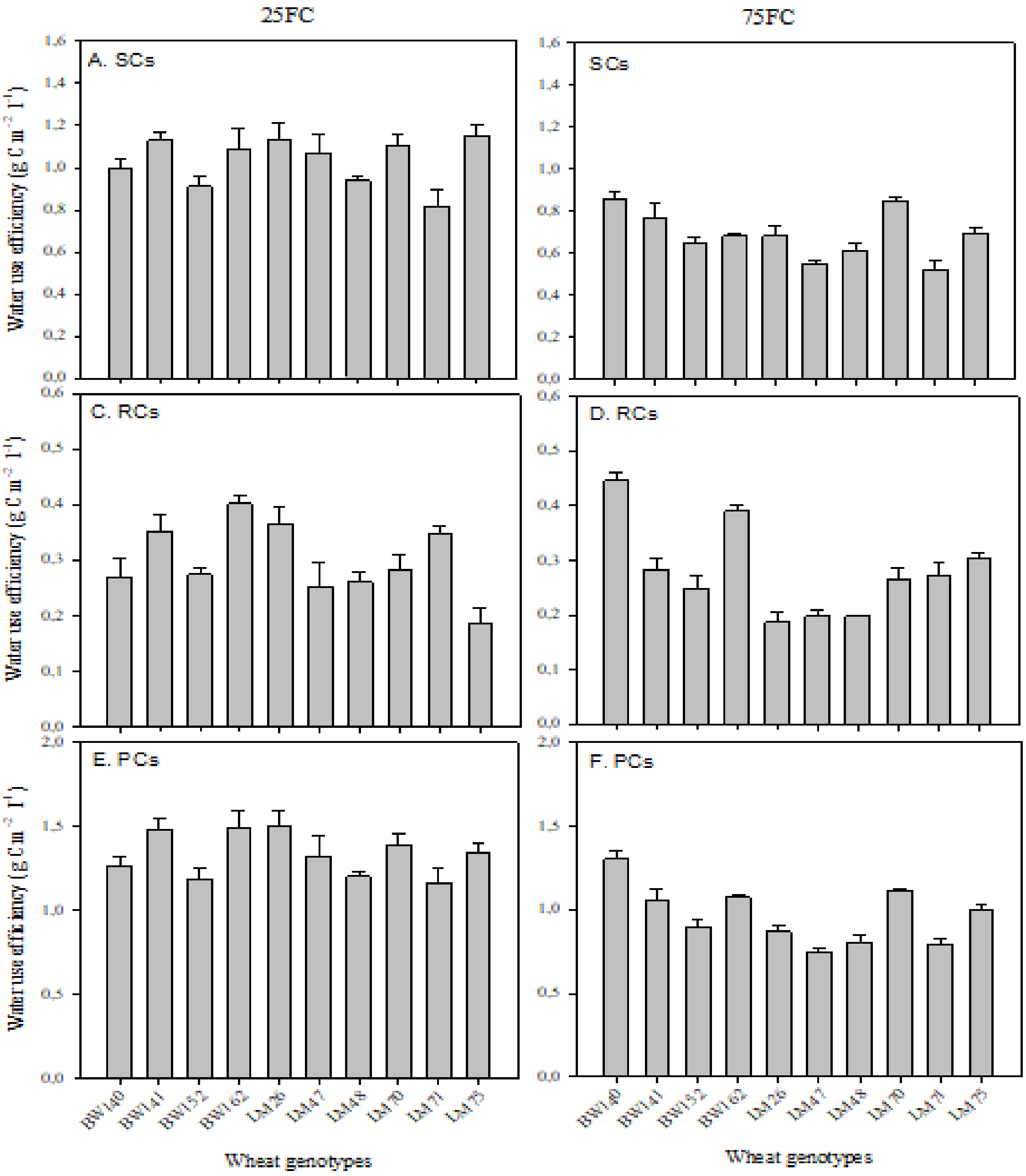


Figure 5.3. Water use efficiency for shoot, root and total plan carbon stocks of 10 selected wheat genotypes under two water regimes.

5.4. Discussion

5.4.1. Genotypic variations in biomass and agronomic performance

The 100 genotypes accumulated different biomass, which they allocated in variable proportions between roots and shoots ($p<0.05$). This agreed with Akman et al., (2017), who reported significant genotypic variation among 47 wheat genotypes evaluated under field conditions in Turkey. Differences in performance among genotypes reflects genetic diversity (Bhutta et al., 2006), which is a consequence of variable genetic background of the genotypes. Twenty-one genotypes were from the drought tolerant nursery while 75 originated from the heat tolerant nursery of CIMMYT resulting in differences in their agronomic performance. The heat tolerant lines such as BW152, LM47, BW141, BW162 had higher shoot biomass compared to the drought tolerant genotypes such as LM98, LM71 and LM75 showing their ability to tolerate heat stress conferred an advantage in above ground biomass production. The high total and shoot biomass production by Triticale compared to wheat concurred with reports that Triticale has higher potential for biomass production which was attributed to its higher solar radiation conversion efficiency of 3.2 g MJ^{-1} compared to 2.0 g Mj^{-1} exhibited by wheat under similar conditions (Estrada-Campuzano et al., 2012).

Root biomass production was significantly different among the 100 genotypes, agreeing with a report by Nevo and Chen (2010) who asserted that there is wide genetic variability in root biomass production in wheat that is yet to be exploited in breeding programs. The range of root biomass found in this study differed from the range of 97 to 1176 g m^{-2} reported by Waines et al., (2012) under greenhouse condition, but confirms the existence of genotypic variation. The differences in root biomass among the CIMMYT accessions, French varieties and Triticale are apparent due to their different adaptations. Triticale is well known to combine an aggressive rooting capability of one of its parents, rye (Gelalcha et al., 2007), while wheat genotypes adapted to temperate ecologies can have double the amount of root biomass of wheat genotypes grown in the warmer winters (Thorup-Kristensen et al., 2009) such as the heat and drought tolerant CIMMYT accessions. However, the French varieties and Triticale could not reach their full grain production potential due to a shorter growing season, which could explain their low grain yield in comparison to the other genotypes.

Biomass allocation between roots and shoots expressed as R:S ratios also varied significantly among the 100 genotypes, which encompasses a range of 1.00 to 1.36 reported in wheat varieties by Fang et al., (2017). The high R:S exhibited by the heat tolerant genotypes suggests their ability to maintain productivity even under combined drought and heat stress conditions that allowed them to allocate more biomass towards root development compared to the drought tolerant genotypes.

Such genotypes with heat tolerance and high biomass accumulation are more relevant for sub-Saharan Africa where drought and heat stresses often occur simultaneously.

Cluster analysis grouped the genotypes into distinct groups based on their phenotypic similarity (Fig 1). Phenotypic differences are important as they simultaneously reflect the influence of genes and environmental factors. The clustering of the genotypes into distinct clusters highlights variation in the pedigree of genotypes from the different nurseries (heat and drought tolerant) because clusters reflect relatedness in the genetic background (Sorkheh et al., 2007; Mofokeng et al., 2014). The wide genotypic variation in the germplasm opens opportunities for wheat improvement by selecting the best performing genotypes from different clusters to preserve genetic diversity that is critical for breeding (Nevo and Chen, 2010). For instance, LM26 has early maturity while BW152 has high biomass accumulation potential. The introgression of genes from one into the other genotype will potentially create a genotype which matures earlier but that can still maintain high biomass yield potential important for food production and C sequestration.

5.4.2. The impact of water regime and environmental conditions on trait expression

Water regime had significant ($p<0.05$) impact on biomass (GY, PB, SB and RB), biomass allocation to roots and shoot (R:S) and other agronomic traits, similar to other studies (e.g. Fang et al., 2017; Mwadzingeni et al., 2016). The higher impact of drought stress on biomass production revealed that drought stress imposed earlier during pre-anthesis vegetative stages impacts on all biomass and growth parameters, whereas other studies focused on terminal drought imposed at anthesis or post-anthesis (e.g. Foulkes et al., 2007; Mwadzingeni et al., 2016). However, the changes in ranking of genotypes in biomass accumulation across water regimes disagreed with Foulkes et al., (2007) who asserted that genotypes with high performance under optimal soil water conditions still perform well under drought stress.

Biomass allocation to roots (R:S) was reduced by 14% due to drought stress, unlike previous studies which supported the optimal partitioning theory (Poorter et al., 2012; Eziz et al., 2017). The present findings agree with Vanaja et al., (2011) who also found that drought reduced R:S ratios by 7% in sunflower and 60% in maize. The reason for decreased R:S ratios can be explained by the fact that the prolonged drought stress may have exceeded a threshold level causing the plant to completely lose its biomass allocation regulatory ability (Xu et al., 2010). Nonetheless, the higher impact of drought on biomass production (>30%) compared to 14% on R:S ratio agreed with previous report by Lopez-Castaneda and Richards, (1994) who indicated that drought stress has more impact on dry matter production than its allocation to organs.

5.4.3. Associations between biomass and agronomic traits under contrasting water regimes

Drought stress affected the relationship among agronomic traits. Similarly, Mwadzingeni et al. (2016) found differences in trait relationship explained by PC under different water regimes. Under both drought stress and non-stress conditions, variations in yield were largely explained by changes in shoot biomass than any other parameter. This agreed with Dodig et al. (2012) and Sareen et al. (2014), who both reported moderate to high correlations ($r > 0.3$) between grain yield and biomass production under stressed conditions. Reynolds et al., (2009) reckoned that the association between grain yield and shoot biomass is vital since future yield improvement in wheat will be accountable to increase in above ground biomass rather than shifts in biomass partitioning. This presents a conflict in efforts to increase soil C input via root biomass while attempting to maintain or increase yield potential simultaneously.

Root biomass and biomass allocation between roots and shoots (R:S) were more important under drought stressed conditions. This is vital because under stressed conditions root biomass is pivotal in accessing water resources (Poorter et al., 2012; Tatar 2016; Eziz et al., 2017). In concurrence, Atta et al. (2013) attributed 45% of variance in grain yield to root traits. Similarly, Fang et al. (2017) reported significant correlation of 0.24 between root biomass and grain yield under irrigation and even stronger correlation of 0.78 under lower soil water availability in a wheat rain-fed system.

5.5. Conclusion

Wide intra-specific variation in biomass production and its allocation to roots and shoots was found among the 100 genotypes of wheat, showing that these are vital genetic resources for development of drought tolerant and enhanced C sequestering varieties. The results from correlation and diversity analyses show that it is possible to simultaneously select for high grain yield and root biomass production to satisfy both food production and C sequestration needs. The study also concluded that R:S is inadequate as a sole predictor for drought tolerance, biomass productivity or soil C input by wheat due to the low correlations between R:S and biomass variables (GY, PB and SB) under both water regimes. The reduction in R:S due to drought stress contradicted with widely accepted optimal partitioning theory, but supports the theory that prolonged drought stress can lead to the collapse of biomass allocation regulatory ability in plants. These results provide valuable information for investigating C sequestration potential and revealing the genetic basis of drought tolerance in wheat. Meanwhile, 10 genotypes were identified and selected for their diversity, high yield and total biomass production under drought stress conditions. These genotypes will be investigated for C allocation using isotopic labelling and assessment of genetic basis and heritability of biomass allocation, which will lay the foundation for breeding C efficient, drought tolerant and high yielding varieties.

Chapter 6 WATER USE EFFICIENCY OF MAIZE AND SORGHUM UNDER FIELD CONDITIONS

Abstract

Sequestration of atmospheric carbon (C) into plants and ultimately to soils is becoming a credible strategy to mitigate against climate change and alternatively to restore land productivity as soil carbon is key for soil fertility and water storage. While some studies have compared the ability of existing crops to store carbon and allocate it to the soil through roots, yet the variations between crop genotypes have received less attention. The objective of this study was to compare the atmospheric carbon sequestration of selected genotypes under different scenarios of soil water availability. The experiments were set up under field conditions with 10 genotypes each of maize, and sorghum grown at 25% field capacity (drought stressed) and 75% (non-stressed). The variables considered were as follows: shoot (SB) and root (RB) biomass, grain yield (GY), root to shoot ratios (R:S), and water use efficiency (WUE) for GY and biomass production. Genotypic variation existed for all measured traits useful for improving drought tolerance, while the calculated R:S values can improve accuracy in estimating C sequestration potential of wheat. Under well-watered (WW) conditions, average shoot biomass for sorghum was 610 g m^{-2} and average RB 328 g m^{-2}. The average root: shoot ratio was 0.55. The WUEsb for sorghum under WW conditions was 0.2 g mm^{-2} m^{-2}, while WUErb was 0.11 g mm^{-2} m^{-2}. For maize, the average shoot biomass was mean value of 900 g m^{-2} but average RB was 164 g m^{-2}. The mean root: shoot biomass ratios was 0.18). On average, the WUEsb for maize was mean 0.29 g mm^{-2} m^{-2}, while WUErb was 0.05 g mm^{-2} m^{-2} under well-watered conditions. Sorghum showed that it was more efficient compared to maize in water productivity.

Keywords: agronomic traits, grain yield, genotype by environment interaction, root to shoot ratio, water stress

6.1. Introduction

Maize (*Zea mays*) and sorghum (*Sorghum bicolor*) are important crops to the economy of South Africa as they contribute to food security and overall GDP of the country. As such it is important to explore their carbon sequestration potential and efficiency to utilise available water resources.

Since water is often limited in most smallholder farming areas of South Africa, it is worthwhile to screen for varieties of these crops that have the potential to produce high biomass yield under water stress. Field crops are generally known to differ in the way they use carbon, based on their chemical composition as well as environmental conditions prevailing at the time of their growth (Sheahan et al., 2014). Tropical cereals (sorghum, maize and rice) for example, have higher biomass production than temperate (wheat, barley) crops because of their C_4 photosynthesis system. C_4 plants have higher photosynthesis capacity (Mathew et al., 2017), and are known to delay their stomatal closure which enables them to be more tolerate of water stress than their temperate C_3 counterparts. As a result, C_4 plants prolong carbon dioxide exchange, thereby producing more biomass and carbon (Sheahan et al., 2014). Generally, cereals are known to have heavier and longer root systems than legumes, thereby amassing more C in their roots (Gregory et al., 1995). It is for this reason that it is necessary to compare field crops for their water use efficiencies (WUE) and C sequestration potential to make more informed decisions about which varieties to select under marginal field conditions.

South African soils found under smallholder agricultural production are often characterised by poor soil fertility and very low C levels. Good shoot and root biomass production in a crop is essential as it adds the much need soil carbon as the plant residues decompose. Sorghum in particular has been reported to sequester high amounts of carbon, tolerate water stress and alkalinity (Brauteseth, 2009), hence it is worthwhile to assess its relatively unexplored genetic capacity, in an effort to explore its ease of decomposition and contribution towards C sequestration once the biomass is incorporated into the soil. Since a lot of work has been done on wheat, ten varieties each of sorghum and maize were further assessed for their WUE potential under field conditions as detailed below.

6.2. Materials and methods

6.2.1. Plant material

Ten genotypes per species of maize and sorghum obtained from various sources were evaluated in this study (Tables 6.1 and 6.2). Some commercial varieties of maize were also included as possible control checks as they are already in production across various environments in South Africa. The maize genotypes used were previously evaluated for pest resistance under field conditions and will provide important genetic resources to improve local varieties. The sorghum accessions were still in developmental stages and they were previously evaluated for drought tolerance and diversity.

Table 6.1. A list of maize genotypes used in the study

	Genotype	Code	Source	Attributes
1	NC QPM NPGRC	NELSON QPM CPSTN	South Africa	Commercial QPM
2	PAN4P-228	PAN4P-228	South Africa	Commercial
3	Shesha NPGRC	SHESHA CPSTN	South Africa	Commercial
4	N3XCML444	N3XCML444	South Africa	Developmental hybrid
5	M/Pearl NPGRC	MAC MEDIUM PEARL	South Africa	Commercial
6	Kep NPGRC	KEP SELECT	South Africa	Commercial
7	ZM1421	1421/DT-STR	Zimbabwe	Drought tolerant
8	ZM1423	ZM1423	Zimbabwe	Drought tolerant
9	10 Obanapta	OBANAPTA	CSIR/Ghana	QPM
10	R201	R201	South Africa	Developmental hybrid

CSIR=The Council for Scientific Industrial Research in South Africa, QPM=quality protein maize

Table 6.2. A list of sorghum genotypes used in the study

No.	Genotype	Source	Attributes
1	AS8	ACCI	Experimental hybrid
2	AS15	ACCI	Experimental hybrid
3	AS16	ACCI	Experimental hybrid
4	AS18	ACCI	Experimental hybrid
5	05-POTCH-138	ARC	Experimental hybrid
6	Mamolokwane	ARC	Landrace
7	LP4303	Limpopo	Landrace
8	NW5430	North West	Landrace
9	KZ5246	KwaZulu-Natal	Landrace
10	NW5393	North West	Landrace

ACCI=African Centre for Crop Improvement, ARC=Agricultural Research Council of South Africa.

6.2.2. Site description, experimental layout and design

The field experiment was carried out at the University of KwaZulu-Natal Ukulinga Research farm (LAT: 29.667° LON: 30.406° and ALT: 811 m) using a complete randomized block design with 3 blocks and 3 replications per block. Long-term average temperature and rainfall for Ukulinga are 18°C and 738 mm, respectively. Table 6.3 below shows the properties of the soil at the study site after characterization for its physico-chemical properties using standard laboratory techniques. The field was ploughed in April 2018 during the winter season. Basal fertilizer NPK (2:3:4) was applied at a rate of 250 kg/ha. Top dressing was applied four weeks after crop emergence and supplied in the form of lime ammonium nitrate (LAN) (28%) kg/ha as a single application.

6.2.3. Maize and sorghum field experiments

The field experiment was carried out at the University of KwaZulu-Natal Ukulinga Research farm (LAT: 29.667^{o} LON: 30.406^{o} and ALT: 811 m) using a complete randomized block design with 3 blocks and 3 replications per block. Long-term average temperature and rainfall for Ukulinga are 18°C and 738 mm, respectively. Table 6.3 shows the properties of the soil at the study site after characterization for physico-chemical properties using standard laboratory techniques. The field was ploughed in April 2018 during the winter season. Basal fertilizer NPK (2:3:4) was applied at a rate of 250 kg/ha. Top dressing was applied four weeks after crop emergence and supplied in the form of lime ammonium nitrate (LAN) (28%) kg/ha as a single application.

6.2.3.1. Maize planting

Two seeds were planted per station at 15 cm intra-row spacing and 45 cm between rows soon after ploughing. Each genotype was planted per 3 m row plot in a block consisting of 20 rows with randomly assigned genotypes. The plants were thinned to one plant per station 2 weeks after emergence to leave 20 plants per row.

6.2.3.2. Sorghum planting

Five seeds were planted per station at 20 cm intra-row spacing and 45 cm between rows soon after ploughing. Each genotype was planted per 3 m row plot in a block consisting of 20 rows with randomly assigned genotypes. The plants were thinned to one plant per station 2 weeks after emergence to leave 15 plants per row.

6.2.3.3. Weed and pest control

Weeds were controlled by a combination of Basagran, Gramoxone and Troopers for the control of annual grasses and broadleaf weeds. Herbicides were complemented by manual weed control where necessary. A combination of pesticides Lambda and Carbofuran was applied at recommended rates both as preventative and curative measures at six weeks after crop emergence.

6.3 Irrigation

Irrigation was applied through a drip irrigation system so as to maintain soil water content at FC in the well-watered regime. All the plots were maintained at adequate moisture regime for the first 6 weeks after emergence. Under the drought stress treatment, irrigation was withheld until just before signs of permanent wilting were observed upon which irrigation was resumed. The amount of water applied after 6 weeks was recorded.

Table 6.3. Soil properties and mean temperature for the Ukulinga study site

Property	Field experiment
Bulk density	1.04
Phosphorous (mg/L)	39.00
Potassium (mg/L)	241.00
Calcium (mg/L)	1453.00
Magnesium (mg/L)	369.00
Electrical Conductivity (cmol/L)	11.02
pH (KCl)	4.56
Organic carbon (%)	2.60
Nitrogen (%)	0.23
Clay (%)	28.00
Mean Temperature (°C)	16.63

pH (KCl)=pH measured in potassium chloride solution

6.3.1. Measurements

Agronomic traits were recorded during the growth period. At maturity, 8 plants (half the plot) were harvested from each plot. Plant parts for each plot were separated at maturity into shoot and root. The above ground biomass was cut off at the soil surface to separate from below ground biomass. All the roots to a depth of 60 cm within the soil volume were collected per genotype per plot. The larger roots were manually separated from the soil and the finer roots were collected by wet sieving. The separated plant parts were oven dried at 60°C for 72 hours to measure the dry weight. The weight was converted to gram per plot (3×0.45 m). Root: shoot (R:S) ratio was computed accordingly.

6.3.2. Data Analysis

6.3.2.1 Analysis of variance

Analysis of variance (ANOVA) was conducted using the lattice procedure using Genstat 18th edition (Payne et al., 2017). In addition, the means of genotypes and the different water regimes were separated by Fischer's unprotected least significant difference (LSD) at 0.05 significance level to quantify the effects of genotype, environment and water regime.

6.3.2.2. Water use efficiency (WUE)

The cumulative amount of irrigation water was calculated at the end of the growing period. WUE was calculated as the ratio of the amount of water used by the plant to amount of biomass accumulated yield or carbon in the soil at the end of experimentation.

$$\text{WUE} = \text{DM}/\text{TIW}$$

where DM is plant dry mass and TIW is total amount of irrigation water.

6.3. Results

6.3.1. Biomass productivity and WUE of sorghum under different watering regimes

A closer look at the water regimes showed that under well-watered (WW) conditions, shoot biomass for sorghum ranged from 363 to 1191 g m^{-2} with a mean value of 610 g m^{-2} for the 10 varieties (Table 6.4). RB on the other hand under WW condition was still lower than SB as it ranged from 183 to 660 g m^{-2}, giving a mean value of 328 g m^{-2} across varieties. This resulted in root: shoot biomass ratios of 0.34 to 0.77 (mean 0.55) while the total sorghum plant biomass ranged from 591 to 1612 g m^{-2} (mean 938 g m^{-2}) across genotypes in the WW regime.

WUEsb for the different sorghum varieties under WW conditions ranged from 0.12 to 0.38 g mm^{-2} m^{-2} (mean 0.2 g mm^{-2} m^{-2}), while WUErb was 0.06 to 0.21 g mm^{-2} m^{-2} (mean 0.11 g mm^{-2} m^{-2}). The total biomass plant WUE ranged from 0.19 to 0.52 g mm^{-2} m^{-2} (mean 0.3 g mm^{-2} m^{-2}) across genotypes in the WW regime (Table 6.4).

Table 6.4. Summary statistics for selected plant morphological variables for 10 selected sorghum varieties the field under well-watered conditions

	SB	RB	RS	PB	WUEsb	WUErb	WUEpb
Mean	610	328	0,55	938	0,20	0,11	0,30
Median	574	294	0,54	876	0,18	0,09	0,28
Minimum	363	183	0,34	591	0,12	0,06	0,19
Maximum	1191	660	0,77	1612	0,38	0,21	0,52
Quartile 1	463	265	0,46	724	0,39	0,66	0,64
Quartile 3	687	405	0,66	1062	0,58	1,01	1,05
CV%	33	31	21,52	30	29,56	27,98	22,25
SEM	26	13	0,02	37	0,02	0,03	0,04
Skew	1	1	0,20	1	0,25	0,29	0,37
Kurt	1	2	-0,92	0	-0,71	-0,26	-0,32

CV=coefficient of variation, SEM=standard error of mean, SB=shoot biomass, RB=root biomass, PB=total plant biomass, RS=root to shoot ratio; WUEsb, WUErb and WUEpb=water use efficiency for shoot, root and total plant biomass, respectively.

Under water-stressed conditions, shoot biomass for sorghum ranged from 239 to 784 g m^{-2} (mean 414 g m^{-2}) for the 10 varieties (Table 6.5). While RB was still lower than SB as it ranged from 152 to 444 g m^{-2}, (mean 328 g m^{-2}) across varieties. This resulted in root: shoot biomass ratios of 0.43 to 0.85 (mean 0.63) while the total sorghum plant biomass ranged from 391 to 1228 g m^{-2} (mean 668 g m^{-2}) across genotypes (Table 6.5).

WUEsb for the different sorghum varieties under water-stressed conditions ranged from 0.21 to 0.71 g mm^{-2} m^{-2} (mean 0.38 g mm^{-2} m^{-2}), while WUErb was 0.14 to 0.4 g mm^{-2} m^{-2} (mean 0.23 g mm^{-2} m^{-2}). The total plant biomass WUE ranged from 0.35 to 1.11 g mm^{-2} m^{-2} (mean 0.61 g mm^{-2} m^{-2}) across genotypes under water stress (Table 6.5).

Table 6.5. Summary statistics for selected plant morphological variables for 10 sorghum varieties grown in the field under water stressed conditions

	SB	RB	RS	PB	WUEsb	WUErb	WUEpb
Mean	414	254	0,63	668	0,38	0,23	0,61
Median	353	249	0,63	621	0,32	0,23	0,56
Minimum	239	152	0,43	391	0,21	0,14	0,35
Maximum	784	444	0,85	1228	0,71	0,40	1,11
Quartile 1	304	206	0,55	513	0,17	0,32	0,23
Quartile 3	520	283	0,73	798	0,30	0,44	0,46
CV%	33	27	19,31	30	28,89	25,88	18,91
SEM	18	9	0,02	26	0,01	0,01	0,03
Skewness	1	1	0,47	1	-0,09	0,53	0,27
Kurtosis	0	1	-0,69	1	-1,04	0,01	-0,36

CV=coefficient of variation; SEM=standard error of mean; SB=shoot biomass; RB=root biomass; PB=total plant biomass; RS=root to shoot ratio; WUEsb, WUErb and WUEpb=water use efficiency for shoot, root and total plant biomass, respectively.

Comparisons between sorghum genotypes showed that AS15 consistently gave higher biomass yields in the shoot, root and total plant biomass in both watering regimes (Fig 6.1). It was also the most water-use efficient genotype for all plant parts and under both watering regimes (Fig 6.2). Other high yielding sorghum genotypes were 05-POTCH-138, NW5430 and AS8, for both roots and shoots, while NW5393 and AS16 gave the lowest shoot and root biomass under water-stressed conditions. In WW conditions, AS18, NW5393 and AS16 were poor performers for biomass accumulation and WUE in both plant parts.

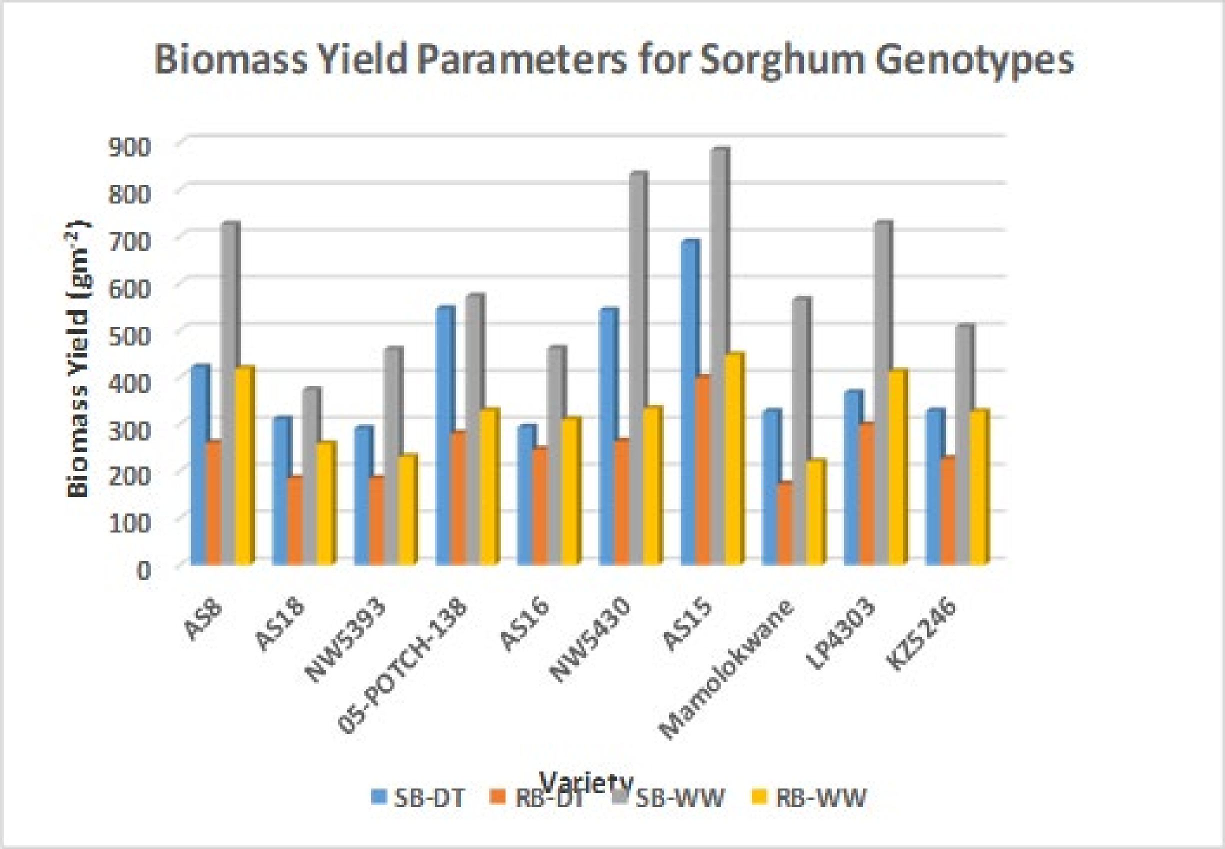


Figure 6.1. Biomass yield parameters for sorghum genotypes

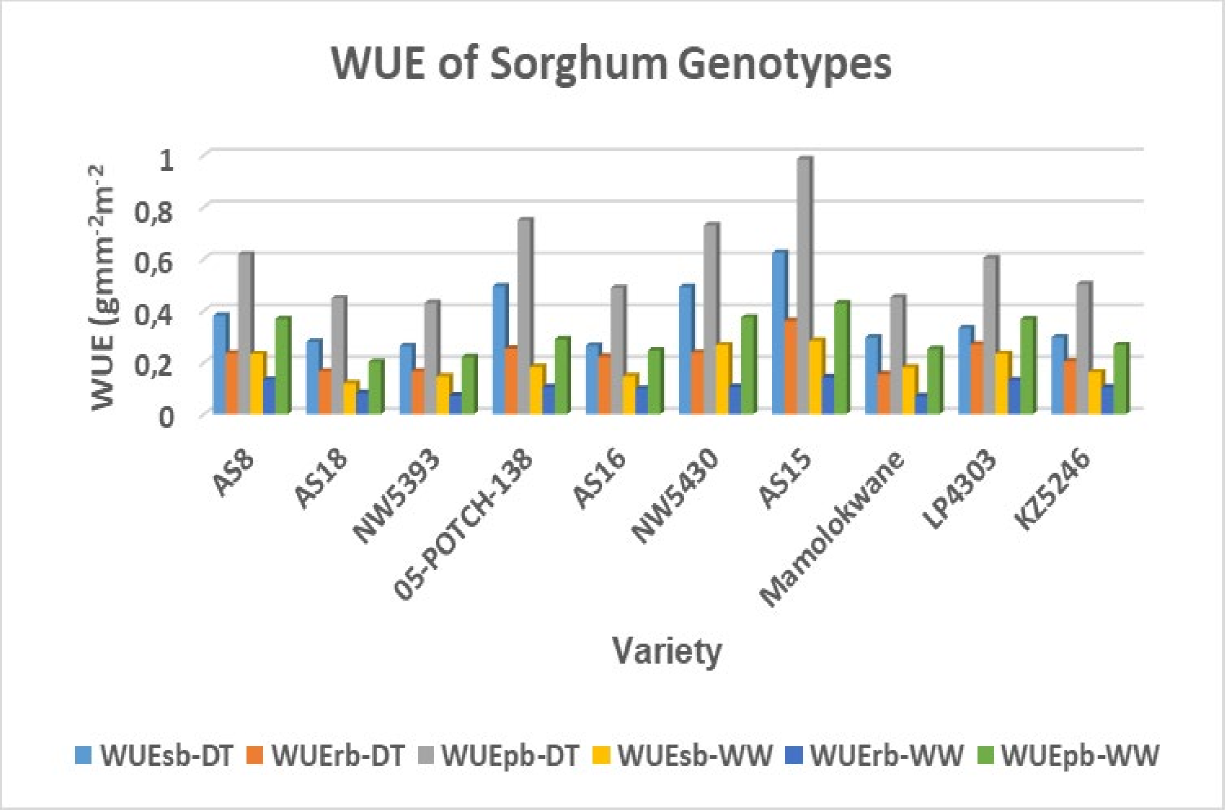


Figure 6.2. Water use efficiency parameters for sorghum genotypes

6.3.2. Biomass productivity and WUE of maize under different watering regimes

Under well-water (WW) conditions, shoot biomass for maize ranged from 535 to 1612 g m^{-2} with a mean value of 900 g m^{-2} for the 10 varieties (Table 6.6). RB under WW conditions was still lower than SB as it ranged from 35 to 379 g m^{-2}, (mean 164 g m^{-2}) across varieties. This resulted in root: shoot biomass ratios of 0.04 to 0.43 (mean 0.18), while the total maize plant biomass ranged from 619 to 1943 g m^{-2} (mean 1069 g m^{-2}) across genotypes in the WW regime.

WUEsb for the different maize varieties under WW conditions ranged from 0.17 to 0.51 g mm^{-2} m^{-2} (mean 0.29 g mm^{-2} m^{-2}), while WUErb was 0.01 to 0.12 g mm^{-2} m^{-2} (mean 0.05 g mm^{-2} m^{-2}) under well-watered conditions. The total plant biomass WUE then ranged from 0.2 to 0.63 g mm^{-2} m^{-2} (mean 0.34 g mm^{-2} m^{-2}) across genotypes in the WW regime (Table 6.6).

Table 6.6. Summary statistics for selected plant morphological variables for 10 maize varieties grown in the field under well-watered conditions

	SB	RB	RS	PB	WUEsb	WUErb	WUEpb
Mean	900	164	0,18	1064	0,29	0,05	0,34
Median	851	130	0,16	1069	0,27	0,04	0,34
Minimum	535	35	0,04	619	0,17	0,01	0,20
Maximum	1612	379	0,43	1943	0,51	0,12	0,63
Quartile 1	756	102	0,13	861	0,26	0,31	0,29
Quartile 3	1027	204	0,21	1206	0,35	0,26	0,19
CV%	23	56	54,35	25	21,94	20,66	26,09
SEM	27	12	0,01	34	0,01	0,02	0,15
Skewness	1	1	1,21	1	0,48	0,57	0,78
Kurtosis	3	0	1,18	3	0,79	0,81	0,79

CV=coefficient of variation, SEM=standard error of mean, SB=shoot biomass, RB=root biomass, PB=total plant biomass, RS=root to shoot ratio; WUEsb, WUErb and WUEpb=water use efficiency for shoot, root and total plant biomass, respectively.

Under water-stressed conditions, shoot biomass for maize ranged from 608 to 2930 g m^{-2} with a mean value of 1261 g m^{-2} for the 10 varieties (Table 6.7). RB under water stress was lower than SB, ranging from 33 to 562 g m^{-2}, (mean 219 g m^{-2}) across varieties. This resulted in root: shoot biomass ratios of 0.03 to 0.73 (mean 0.19) while the total maize plant biomass ranged from 754 to 3492 g m^{-2} (mean 1488 g m^{-2}) across genotypes in the water-stressed regime.

WUEsb for the different maize varieties under water stress ranged from 0.55 to 2.66 g mm^{-2} m^{-2} (mean 1.14 g mm^{-2} m^{-2}), while WUErb was 0.03 to 0.51 g mm^{-2} m^{-2} (mean 0.2 g mm^{-2} m^{-2}) under water stress. The total plant biomass WUE ranged from 0.68 to 3.17 g mm^{-2} m^{-2} (mean 1.34 g mm^{-2} m^{-2}) across genotypes under water stress (Table 6.7).

Table 6.7. Summary statistics for selected plant morphological variables for 10 maize varieties grown in the field under water stressed conditions

	SB	RB	RS	PB	WUEsb	WUErb	WUEpb
Mean	1261	219	0,19	1488	1,14	0,20	1,34
Median	1109	183	0,18	1359	1,0	0,17	1,19
Minimum	608	33	0,03	754	0,55	0,03	0,68
Maximum	2930	562	0,73	3492	2,66	0,51	3,17
Quartile 1	853	134	0,11	1077	0,06	0,03	0.04
Quartile 3	1435	297	0,23	1644	0,10	0,16	0,18
CV%	42	57	66,88	41	35,40	80,41	74
SEM	69	16	0,02	79	0,00	0,06	0.07
Skewness	1	1	2,83	1	0,34	1,07	1,13
Kurtosis	2	1	11,60	3	-0,34	1,79	1,53

CV=coefficient of variation, SEM=standard error of mean, SB=shoot biomass, RB=root biomass, PB=total plant biomass, RS=root to shoot ratio; WUEsb, WUErb and WUEpb=water use efficiency for shoot, root and total plant biomass, respectively.

Maize genotype comparisons showed that Kep Select, R201, Nelson QPM CPSTN and 1421/DT-STR had higher root and shoot biomass yields while Obanpata performed poorly under water stress (Fig 6.3). Under WW conditions R201 gave higher RB and SB, while Kep Select, 1421/DT-STR and Nelson QPM CPSTN resulted in lower RB and SB.

WUE trends showed that the maize genotypes Kep Select, R201, Nelson QPM CPSTN and 1421/DT-STR gave higher WUE in roots, shoots and total plant biomass under water stress (Fig 6.4). When WW, R201 resulted in higher WUE in shoot, root and total plant biomass, while Kep Select and 1421/DT-STR had lower WUE in the different plant parts.

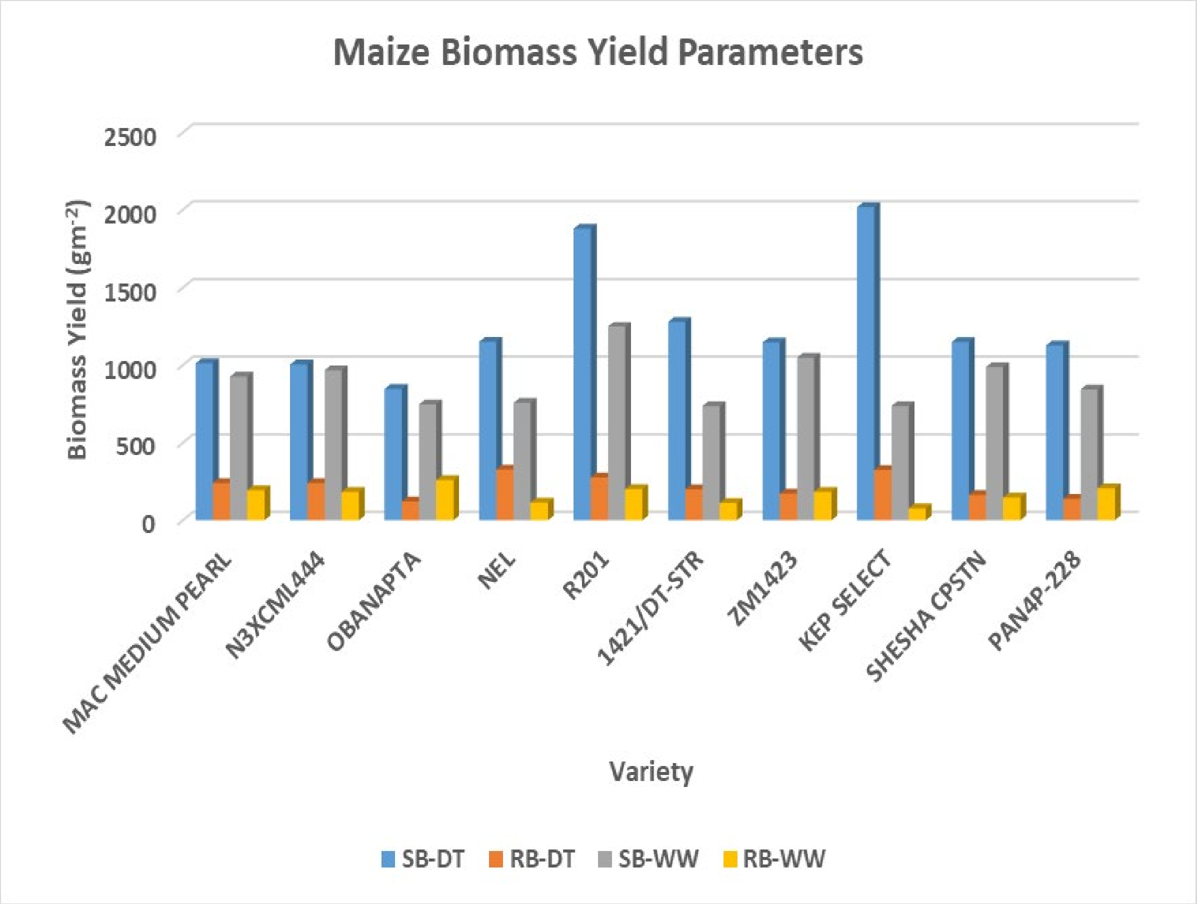


Figure 6.3. Biomass yield parameters for maize genotypes

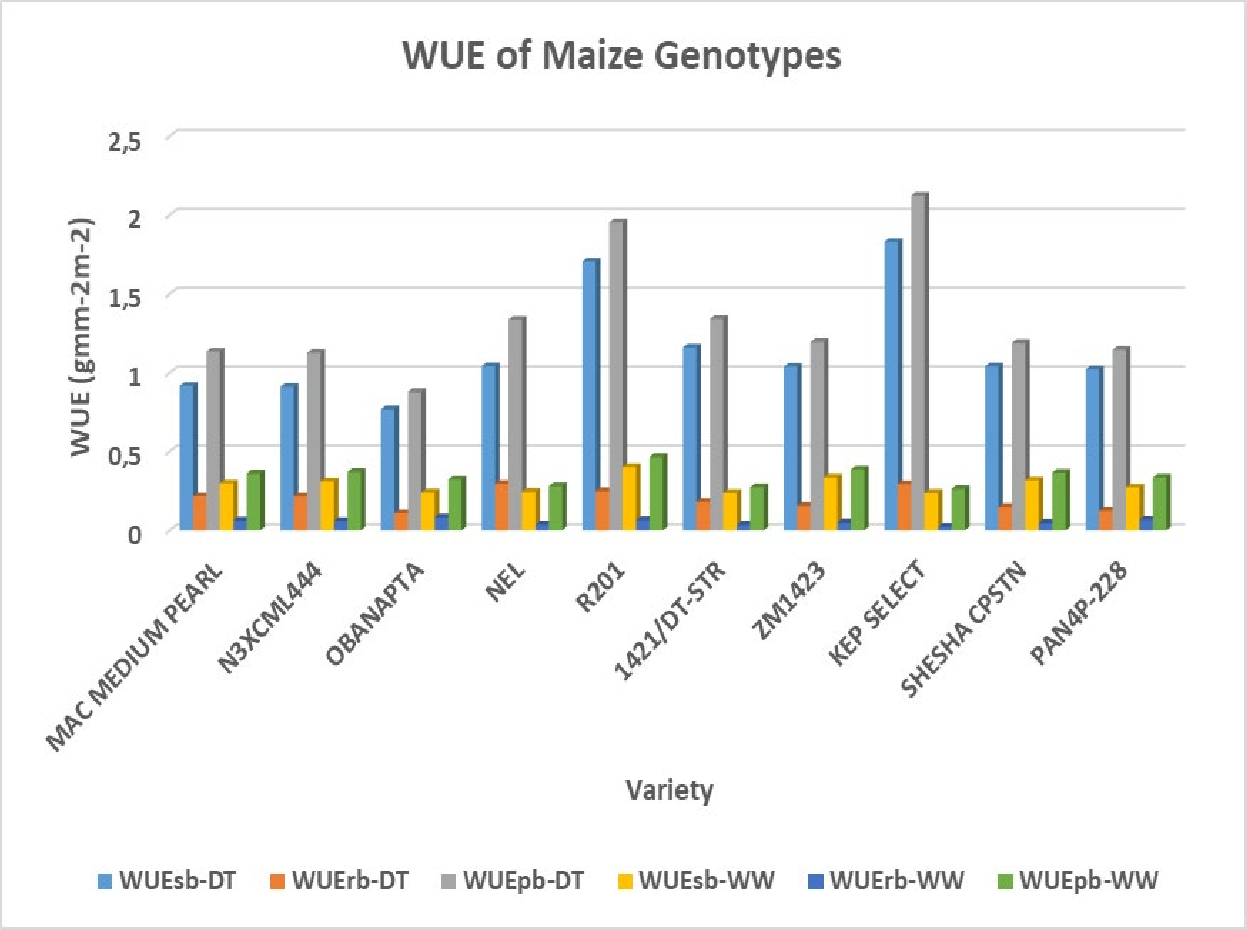


Figure 6.4. WUE parameters for maize genotypes

6.4. Discussion

6.4.1. Biomass and grain yield production

The high biomass and grain production exhibited by maize compared to sorghum shows its high potential to accumulate biomass. The mean biomass production by maize of between 5.35 to 16.12 t ha^{-1}, which was similar to a range between 7.00 and 20.00 t ha^{-1} reported previously for maize (Langholtz et al., 2016; Wilhelm et al., 2011). Previously, Stewart and Amanullah (2013) attributed the high biomass and grain productivity of maize to its high efficiency in solar radiation interception and maximum utilization of resources. However, the variation in productivity depends on soil amendments such as fertilizer application, climatic conditions and nature of germplasm used in different studies and water availability. Tropical maize and silage maize accumulate comparably higher biomass than temperate and grain maize, respectively. Infante et al. (2018)

reported up to 40% increase in biomass in non-grain biomass than temperate maize. There are also differences among open pollinated varieties, hybrids or inbred lines that contributed to the observed variation. In general, hybrids accumulate large amounts of grain and biomass yield compared to inbred lines or open pollinated varieties. The average maize yield found in this study was not significantly different from 9.2 t ha^{-1} reported by Muchow (1989). The maximum attainable biomass for sorghum were 11.91 t ha^{-1}, showing that it also has high potential for biomass production. Sorghum is particularly more productive than maize under water limited conditions. Its advantages under drought stress are rendered by an extensive root system that is efficient at water and nutrient mobilization. There are different types of sorghum for instance, biomass sorghum is bred specifically for producing large quantities of biomass for silage or biofuel production (Rooney, 2014). However, other researchers have reported higher biomass production in sorghum compared to maize or millet (Hallam et al., 2001). This variation exhibited by maize and sorghum is also affected by whether the varieties grown are bred for grain or biomass production. Varieties bred for silage or biofuels have higher potential for non-grain biomass production.

6.4.2. Impact of drought stress on biomass and grain yield production

Water stress significantly reduced biomass and grain yield production in all the crops and genotypes, which corroborates assertions that drought stress is detrimental to plant growth and development. For instance, Khayatnezhad et al. (2010) asserted that water stress negative affects germination, seedling establishment, shoot and root growth and flowering in plants. Maize suffered higher reduction in biomass and yield production due to drought stress. Drought sensitivity has been previous reported to be higher in maize (Rooney, 2014). Leaf water potential, stomatal opening and cell turgidity are reduced under drought stress conditions leading to poor plant growth and development (Riboldi et al., 2016). Stunted growth and development in turn reduce biomass production potential. Sorghum has an extensive root system with higher capacity for water capture, which supports its drought resilience. In addition to rooting ability, its small leaf area minimizes transpiration loss. This increases its water use efficiency and conserves water for biomass and yield production. The impact of environmental conditions on crop performance in biomass and grain yield production is also affected by their pre-existing acclimation and photosynthetic pathway (Berry and Björkman, 1980). Thus, variation in biomass accumulation among the different crops point to their different acclimation, genetic constitution and potential, which govern their efficiency in utilizing water resources under the different water availability scenarios. Reports have shown that crops such as sorghum, or maize vary in biomass and grain yield accumulation depending on their genetic constitution and agronomic practices, varieties and water availability (Zegada-Lizarazu et al., 2012; Oikawa et al., 2014). Dimkpa et al., (2019) reported a 39% decrease in sorghum shoot biomass under water stress.

6.4.3. Variation in water use efficiency among the crops

Water use efficiency (WUE) was affected by genotypic factors and water availability. Thus, water stress induced higher water use efficiency in both maize and sorghum in agreement with other reports (Hasan et al., 2017; Jabereldar et al., 2017; Bhattarai et al., 2020; Mbava et al., 2019). Hasan et al., (2017) found that sorghum exhibited higher water use efficiency compared to maize, which was attributed to high drought sensitivity in maize while sorghum is known to reduce transpiration rates at vegetative stage and maximize water use (Hasan et al., 2017; Ajeigbe et al., 2018). However, Maroco et al., (2000) argued that high WUE does not mean the crop has high biomass and grain yield production under water stress. In all cases, biomass and grain yield production declines with water shortage but biomass accumulation per amount of water consumed increases significantly due to invoking of drought tolerance mechanisms (Hasan et al., 2017).

6.5. Conclusion

The variation in response to water stress by the sorghum and maize genotypes showed that genetic differences are important in plant adaptation to water availability. Drought stress is detrimental to plant growth, thus drought tolerant crops and genotypes must be identified for recommendation to farmers to reduce the impact of climate change on crop production. Different crops and genotypes invoke drought tolerance mechanisms to adapt to water shortage. These mechanisms increase water use efficiency under drought stress but lead to reduction in photosynthetic capacity and biomass production.

Chapter 7 CARBON SEQUESTRATION POTENTIAL OF CROPS

Abstract

Quantifying the ability of plants to store atmospheric carbon in their biomass and ultimately to the soil for climate change mitigation and soil fertility improvement is essential. We performed a meta-analysis of 227 research trials reporting C fluxes from plant to soil in different crops. On average, crops assimilated 4.54 Mg C ha^{-1} yr^{-1} from the atmosphere with values averaging 1.70 and 5.23 Mg C ha^{-1} yr^{-1} for barley and maize, respectively. On average, 61% of the assimilated C was allocated to shoots, 20% to roots, 7% to soils while 12% was respired back to the atmosphere. Maize and ryegrass had the greatest allocation to the soil (1.0 Mg C ha^{-1} yr^{-1} or 19% total assimilation) followed by wheat (0.8 Mg C ha^{-1} yr^{-1}, 23%) and rice (0.7 Mg C ha^{-1} yr^{-1}, 20%). Carbon transfer to soil correlated the most to root biomass C. The variation in C transfer to the soil within crop types opens possibilities for selecting and breeding varieties with superior C transfer capacity. The question of the long-term stability of the newly transferred C in the soil remains asked.

Keywords: Carbon sequestration, C emission, plant C, C labelling, C flux

7.1. Introduction

In 2015, 192 countries ratified the COP21 in Paris to reduce global temperatures to 2°C below pre-industrial levels. At that occasion, the 4 per mille Initiative: Soils for food security and climate (4p1000) was launched by the French government with the aspiration to increase soil organic carbon stock by 0.4% per year (Minasny et al., 2017). The convention envisaged that spillover benefits such as reduced global temperature, improved soil fertility, good soil structure, high soil biodiversity and reduced risk of soil erosion will accrue (Lal, 2016) to sustain higher crop productivity. These benefits have positive impacts on the environment as they improve soil and plant health.

However, some practices proposed by Paustian et al. (2016) to meet the 4p1000 objectives, such as reduced tillage and land use conversion, are subjects of debate. For example, it has been argued that reduced tillage only causes redistribution of soil C within the soil profile with no net gain in the soil organic carbon (SOC) (Dimassi et al., 2014), while land use conversion from cropland to grassland or forest can have negative implications on food security. Furthermore, there is a high tendency to rely on best agronomic practices for carbon sequestration in the soil, while neglecting the effects of plants on the global C cycle despite abundance of evidence that genetic variation, especially in root traits, influences C allocation in ecosystems (Warembourg et al., 2003). Therefore, there is a need to also look into the plant components for fulfilling the 4p1000 objectives.

Plant assimilated C is either incorporated as structural C, released as exudates or respired as CO_2 (Ostle et al., 2003). These processes vary with crops' genetic make-up, leading to differences in soil C contributions among genotypes (Kuzyakov and Domanski, 2000). Quantifying soil C input by a genotype is integral to understanding the terrestrial C cycle. However, the underlying genetic factors, the mechanisms involved in C sequestration and how plant breeding programs can improve C sequestration are still poorly understood (Wegener et al., 2015). The first step in developing protocols for increased C sequestration by crop genotypes lies in understanding the C fluxes in different pools, i.e. biomass, soil and rhizospheric respiration, and how they are inter-related in different genotypes because the fate of C in each of the different pools differ with variable implications on soil C dynamics.

There have been concerted efforts to understand C fluxes in the plant/soil system (e.g. Remus and Augustin 2016; Studer et al., 2014). Commonly, the C fluxes are quantified by successive harvesting of plant biomass (Hemminga et al., 1996) and measuring changes in SOCs over time. This method lacks precision and cannot account for C in other pools, and fails to distinguish C from diverse sources (Remus and Augustin, 2016). With the advent of isotopic C labelling, which exploits the differences in atomic signature of atmospheric CO_2 and commercially labelled CO_2, it is now possible to understand the C dynamics among the different C pools (Studer et al., 2014). The relative proportions of C recovered in each of the pools are then used to estimate potential C sequestration (Nguyen, 2009). Based on C labelling, it has been reported that plants transfer 30-50% of photosynthetic C to below ground pools (Buyanovsky and Wagner, 1997). Furthermore, C labelling has enabled tracing of 'real time' C movement and it has been reported that translocation of C to roots occurs immediately after assimilation in above ground organs with up to 10% being detected within 2 hours (Kaštovská and Šantrůčková, 2007).

Carbon assimilated by plants is allocated to above ground (shoots and reproductive organs) and below ground (roots) biomass depending on plant genetic constitution (de Neergaard and Gorissen, 2004), plant growth pattern, environmental conditions and interactions among these factors (Rangel-Castro et al., 2005). However, it is the root C fluxes which are particularly important in soil C balance. Root C is integral to soil C accumulation due to its higher stability and mineralization rate compared to shoot-derived C (Rasse et al., 2005). Labile and unstable C inputs such as those derived from above ground biomass are easily decomposed by autotrophic and heterotrophic respiration leading to C losses of between 15-45% in perennial plants and 27-60% in annuals (e.g. Warembourg and Paul, 1977; Swinnen et al., 1995a). In light of the above background, this paper aimed at comparing C fluxes in different plant-soil systems using data from multiple trials across the world, to evaluate the C sequestration potential of different crop species and to foresee better ways to manage crop residues. The hypothesis tested in this study was that C fluxes in roots are the key drivers of C transfer into the soil.

7.2. Materials and Methods

7.2.1. Isotopic C enrichment metadata collection

The study was a meta-analysis based on data collated from isotopic C enrichment experiments conducted across the world. The data was obtained from peer-reviewed journal articles published between 1985 and 2016. The journal articles were obtained from academic databases (Google Scholar, Refseek, Science Direct, SciFinder, Scopus, Springer Link and Web of Science) using key words and phrases, singly and/or in combination, such as “isotopic pulse labelling”, “C allocation”, “plant carbon sequestration”, “rhizodeposition” and “plants/soil/microbial respiration”. The literature survey considered studies reporting the allocation of C to plant shoots and roots, soil pools and proportions of the respired C either as percentages or absolute quantities. Thirty-three journal articles (Table 7.1) detailing different studies were obtained using the above criteria providing 227 observations from different experiments, globally (Fig 7.1). A database was constructed to capture the names of authors, year of publication, type of C isotope, reference crop, quantitative information on C allocation variables and the controlling factors.

Table 7.1. List of authors, countries, crop types studied and type of experiment carried out using an identified carbon isotope

No.	Author	Country	Isotope	Crop	Experiment
1	Aljazairi et al., 2015	Spain	13C	Cereal	Greenhouse
2	An et al., 2015	China	13C	Cereal	Field
3	Aranjuelo et al., 2009	Spain	12C	Cereal	Greenhouse
4	Bazot et al., 2006	Switzerland	14C	Grass	Field
5	Butler et al., 2004	USA	13C	Grass	Greenhouse
6	Chaudhary et al., 2012	USA	13C	Grass	Greenhouse
7	Davenport and Thomas 1988	Canada	14C	Cereal	Greenhouse
				Grass	Greenhouse
8	de Graaf et al., 2009	USA	13C	Cereal	Greenhouse
9	de Neergaard and Gorissen 2004	Denmark	14C	Grass	Greenhouse
				Legume	Greenhouse
10	Domanski et al., 2001	Germany	14C	Grass	Greenhouse
11	Fan et al., 2008	China	13C	Cereal	Greenhouse
				Legume	Greenhouse
12	Fang et al., 2016	Australia	13C	Cereal	Field
13	Fernández et al., 2003	Spain	14C	Cereal	Field
14	Gocke et al., 2011	Germany	14C	Cereal	Field
				Grass	
15	Gregory and Tawell 1991	Australia	14C	Cereal	Field
16	Harris et al., 1985	USA	14C	Legume	Greenhouse
17	Hodge and Millard 1998	UK	14C	Non-legume	Greenhouse
18	Hodge et al., 1997	UK	14C	Grass	Greenhouse
19	Holland et al., 1995	USA	14C	Cereal	Greenhouse
20	Kaiser et al., 2015	Australia	13C	Cereal	Greenhouse
21	Kakiuchi and Kobata 2008	Japan	14C	Legume	Greenhouse
22	Lodhi et al., 2009	Pakistan	14C	Cereal	Greenhouse
23	Meng et al., 2013	China	13C	Cereal	Greenhouse
24	Mutegi et al., 2011	Denmark	14C	Legume	Field

No.	Author	Country	Isotope	Crop	Experiment
25	Qiao et al., 2012	China	13C	Legume	Field
26	Schmitt et al., 2013	Germany	14C	Grass	Greenhouse
27	Schulze and Poschel 2004	Germany	14C	Cereal	Greenhouse
28	Swinnen et al., 1995	Netherlands	14C	Cereal	Field
29	Tian et al., 2013	China	14C	Cereal	Greenhouse
				Grass	
30	Warembourg et al., 2003	France	14C	Legume	Greenhouse
				Non-legume	
31	Werth and Kuzyakov 2006	Germany	14C	Cereal	Lab
32	Wu et al., 2008	China	13C	Cereal	Greenhouse
33	Wu et al., 2010	China	13C	Grass	Field

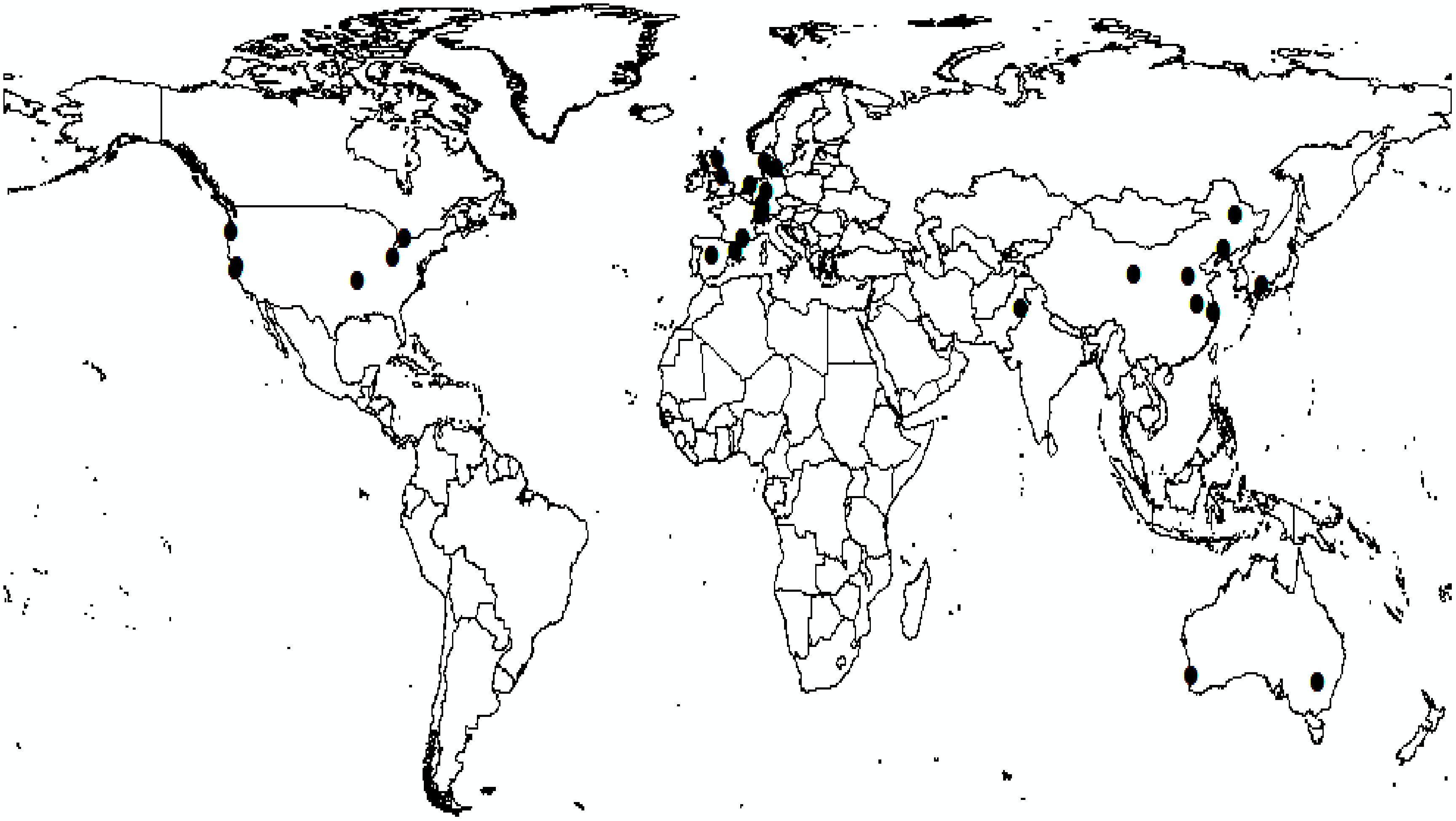


Figure 7.1. Global distribution of the study sites shown by shaded circles on the map

7.2.2. Definition of C allocation variables in meta-study

Data on proportions of C assimilated in shoots, roots, soil and losses to the atmosphere due to respiration were considered. The allocation variables used in this study are defined in Table 7.2. Carbon fluxes in shoot (S_c), root (R_c), respiration (Re_c) and soil C sequestered (C_{seq}) were defined as net C changes in total C stocks calculated in the shoot, root, respired CO_2 pools and soil, respectively, after C enrichment per unit time. Respiration is the emission of C fixed by plants from the soil back to the atmosphere and represents a negative loop in the plant/soil system. Given the difficulty in partitioning below ground respiration (Kuzyakov and Larionova, 2005; Trumbore, 2006) all CO_2 emitted from the soil was assumed to emanate from root respiration. Soil C pool was the total of both rhizospheric and bulk soil C and was the balance of C in the soil after respiration and other C losses.

Table 7.2. Definition of C allocation variables and controlling factors as used in this paper

Parameter	Units	Definition	Categories	Remarks
Shoot carbon fluxes (Sc)	$MgCha^{-1}yr^{-1}$	C fluxes in the shoot calculated from isotopic C in the shoot	Shoot	All crops used in the study
Root carbon fluxes (Rc)	$MgCha^{-1}yr^{-1}$	C fluxes in the root calculated from isotopic C in the root	Root	All crops used in the study
Plant C fluxes (Pc)	$MgCha^{-1}yr^{-1}$	Sum of shoot and root C fluxes	Plant	All crops used in the study
Soil carbon fluxes C_{seq})	$MgCha^{-1}yr^{-1}$	C fluxes in the soil calculated isotopic C in the soil	Soil	All crops used in the study
Respired carbon fluxes	$MgCha^{-1}yr^{-1}$	CO_2 emission calculated from isotopic C in the respired CO_2	Respiration	All crops used in the study
Crop type		The broad group to which a plant belongs	Cereals	All monocot grain crops
			Legumes	All N fixing plants
			Grasses	All pasture grasses
Shoot biomass (Sb)	$Mgha^{-1}yr^{-1}$	The biomass in the shoot per annual basis	All	All crops used in the study
Root biomass (Rb)	$Mgha^{-1}yr^{-1}$	The biomass in the root per annual basis	All	All crops used in the study
Plant biomass (Pb)	$Mgha^{-1}yr^{-1}$	The sum of root and shoot biomass	All	All crops used in the study
Clay content (Cl%)	%	The clay fraction of the soil	Sand	below 20%
			Loam	between 20-30%
			Clay	above 30%
Soil Organic Carbon content (SOCc)	$g\ C\ kg^{-1}$	The C content of the soil at the start of the experiment	Low	below 10 $g\ C\ kg^{-1}$
			Medium	between 10 and 15 $g\ C\ kg^{-1}$
			High	above 15 $g\ C\ kg^{-1}$
Temperature (Temp)	ºC	The average temperature during CO_2 enrichment	Cool	up to 10ºC
			Warm	between 10 and 20°C
			Hot	above 20°C

7.2.3. Definition of the plant/soil system

A plant/soil system refers to the close relationship existing between a plant and its soil environment, including the soil microbes that facilitate nutrient cycling. Any exogenous amendments that bypass the main effects of the plant component such as mulching and fertilizer application were not considered part of the plant/soil system. The plant/soil system is divided into four pools, which are shoots, roots, soil and respired C. The shoot and root C pools are the amount of C contained in the plant biomass. The soil C pool is the amount of C measured in the soil after experimentation while respired C pool is the total amount of C respired by the plant after enrichment.

7.2.4. Data normalization and analyses

The data from the different experiments was normalized for unbiased comparison. This was done by dividing the reported C fluxes by the respective CO_2 concentration used during enrichment, duration of exposure during enrichment or the duration of that particular experiment. The sample size for each variable was determined and stratified by crop type and environmental variable (Table 7.3). Summary statistics were derived including minimum, maximum, median, standard deviation (SD), skewness, 25th quartile (Q1) and 75th quartile (Q3), kurtosis and coefficient of variation for the C variables. Afterwards, the data were summarized using box plots to elucidate the variability and distribution after checking out the outliers. The boxplots presented are only of six major staple crops due to scarcity of data in the other crops. Bivariate and multivariate analyses, using Spearman rank correlations and principal component analysis (PCA), respectively, were conducted to depict the general relationships between C variables and some controlling factors.

Table 7.3. Sample size of C allocation variables stratified by different factors

Variable		Sc	Rc	REc	C_{seq}	C_{seq}/Sc	C_{seq}/Rc	REc/Sc	REc/Rc	C_{seq}/Sb	C_{seq}/Rb
	Description	200	194	142	153	153	153	142	136	138	138
Crop type	Cereal	130	124	95	96	96	96	95	89	96	96
	Grass	47	47	36	36	36	36	36	36	24	24
	Legume	22	22	10	20	20	20	10	10	18	18
	Non-legume	1	1	1	1	1	1	1	1		
Soil texture	Clay	19	19	7	11	11	11	7	7	4	4
	Loam	39	39	23	31	31	31	23	23	31	31
	Sand	14	14	14	14	14	14	14	14	14	14
	Unclassified	128	122	98	97	97	97	98	92	89	89
SOCc (g C kg^{-1})	High	35	35	21	27	27	27	21	21	27	27
	Low	16	16	6	10	10	10	6	6	10	10
	Medium	17	17	4	6	6	6	4	4	6	6
	Unclassified	132	126	111	110	110	110	111	105	95	95
Temperature (°C)	Cool	49	49	37	49	49	49	37	37	38	38
	Hot	36	36	10	20	20	20	10	10	20	20
	Warm	89	83	73	74	74	74	73	67	70	70
	Unclassified	26	26	22	10	10	10	22	22	10	10

Sc=shoot C fluxes; Rc=root C flux; SOc=soil C flux; REc=respiration C flux; SOc:Sc, SOc:Rc, REc/Scs, REc:Rcs, SOc/Sb and SOc/Rb are ratios of the respective variables

7.3. Results

7.3.1. Variability in C fluxes

The global distribution of the study sites, and variations in plant species and treatments applied (e.g. fertilizer rates and environmental conditions) resulted in large variability and skewness in the data (Table 7.4). The shoot C fluxes ranged from 0.20 Mg C ha^{-1} yr^{-1} recorded by Swinnen et al. (1995b) for barley in the Netherlands to 5.60 Mg C ha^{-1} yr^{-1} for wheat in Spain (Aljazairi et al., 2015); while the mean shoot C flux for all plant species was 2.5± 0.09 Mg C ha^{-1} yr^{-1}. The mean C fluxes in the root were 1.0± 0.04 Mg C ha^{-1} yr^{-1} with a lowest of 0.1 Mg C ha^{-1} yr^{-1} calculated for sub-tropical wheat in Australia (Fang et al., 2016) and a highest of 2.8 Mg C ha^{-1} yr^{-1} calculated for ryegrass in a temperate region of USA (Butler et al., 2004). The lowest total C fixed by plants was 0.2 Mg C ha^{-1} yr^{-1}, while the highest was 22.6 Mg C ha^{-1} yr^{-1} by a grass *Kobresia* spp. in temperate China (Wu et al., 2010). Mean C emission from a planted field (REc) was 0.6±0.3 Mg C ha^{-1} yr^{-1}. Greenhouse grown soyabean recorded the highest C emissions of 1.8 Mg C ha^{-1} yr^{-1} (Harris et al., 1985). An average of 0.90±0.05 Mg C ha^{-1} yr^{-1} was stabilized in the soil (C_{seq}), while the maximum was 3.00 Mg C ha^{-1} yr^{-1} calculated for temperate maize in the USA (Holland et al., 1995). Environmental factors also showed wide variability. The soil clay content ranged between 6.1 and 36%, while the SOCc ranged between 2.70 and 37 g C kg^{-1}. Temperatures varied from 5°C in the Netherlands (Swinnen et al., 1995) to 34.7°C in Spain (Fernández et al., 2003).

Table 7.4. Summary statistics of C allocation in different fractions of the plant-soil system for all crops

	C fluxes					C flux ratios							Plant factors		Environmental factors		
	Sc	Rc	C Fix	REc	C_{seq}	REc/Sc	C_{seq}/Sc	REc/Rc	C_{seq}/Rc	C_{seq}/REc	C_{seq}/Sb	C_{seq}/Rb	Sb	Rb	SOCc	Clay%	Temp
Obs.	195	201	192	152	146	141	127	136	127	113	138	141	77	91	68	53	199
Mean	2.5	1	4.8	0.6	0.9	35.1	91.9	77.2	91.9	158.7	13	41	15.4	6.3	15.1	21.6	20.3
Median	2.2	0.9	3.9	0.6	0.8	26.9	70	53.3	70	124.4	8.8	31	13.8	4.8	15.4	21	22
Min	0.2	0.1	0.2	0.1	0.1	2.6	4	1.2	4	12.3	0.4	1.6	2.5	0.9	2.7	6.1	5
Max	5.6	2.8	22.6	1.8	3	143	450	525	450	1210	97.4	129	34.4	17.9	37	36	34.7
Q1	1.5	0.6	2.8	0.4	0.4	15.8	39.8	28.5	39.8	56.8	6	15.7	8.6	3.3	10.3	15	19.5
Q3	3.6	1.4	5.7	0.8	1.1	50.5	132	92.4	131	210	14.6	59.6	21.8	8.3	19	26.9	24.5
SEM	0.1	0.1	0.3	0	0	2.3	6.2	6.9	6.2	14.6	1.1	2.7	1	0.4	0.9	1.3	0.5
%CV	52	71	79	55	64	78	76	104	76	98	103	79	55	67	47	43	35
Skew	0.4	2.8	2.4	0.8	1	1.6	1.8	2.6	1.8	3.4	3.2	1.1	0.5	1.1	1.4	0.2	-0.7
Kurtosis	-0.9	14.7	6.8	0.9	1.6	3.5	5.4	9.2	5.4	17.8	13.8	0.2	-0.8	0.5	2.9	-1	-0.1

Obs.=number of observations: Min.=minimum; Max.=maximum; Q1=first and third quartile, respectively; SEM=standard error of mean; CV=coefficient of variation, C fix=C fixation; Sc=shoot C fluxes; Rc=root C flux; C_{seq}=soil C flux; REc=respiration C flux; SOCc=soil organic C content; Clay%=soil clay content; Temp=temperature; SOc:Sc, SOc:Rc, REc/Scs, REc:Rcs, SOc/Sb and SOc/Rb are ratios of the respective variables

7.3.2. Partitioning of C between plant parts

Figure 7.2 shows the C fluxes in different pools (shoot, root, respiration and soil) of the plant-soil system of six selected crops. The highest mean shoot C fluxes were observed in maize (4.10±0.12 Mg C ha^{-1} y^{-1}), which recorded 44% higher shoot C fluxes than ryegrass (Fig. 7.2a). The shoot C fluxes in maize were also 191% more compared to shoot C fluxes in barley. The shoot C fluxes in wheat (2.52±0.20 Mg C ha^{-1} yr^{-1}) and rice (2.30±0.15 Mg C ha^{-1} yr^{-1}) were not significantly different although wheat showed wider variation. Ryegrass had at least 17% higher shoot C stocks compared to all the other crops except maize. On the other hand, the highest mean root C fluxes were found in ryegrass (1.50±0.18 Mg C ha^{-1} y^{-1}), rice (1.48±0.17 Mg C ha^{-1} y^{-1}) and maize (1.40±0.21 Mg C ha^{-1} y^{-1}), which showed non-significant differences (Fig 7.2b). Further, barley exhibited significantly ($p \leq 0.05$) lower root C fluxes compared to all the other crop species, while wheat and soyabean expressed non-significant differences in the root C fluxes.

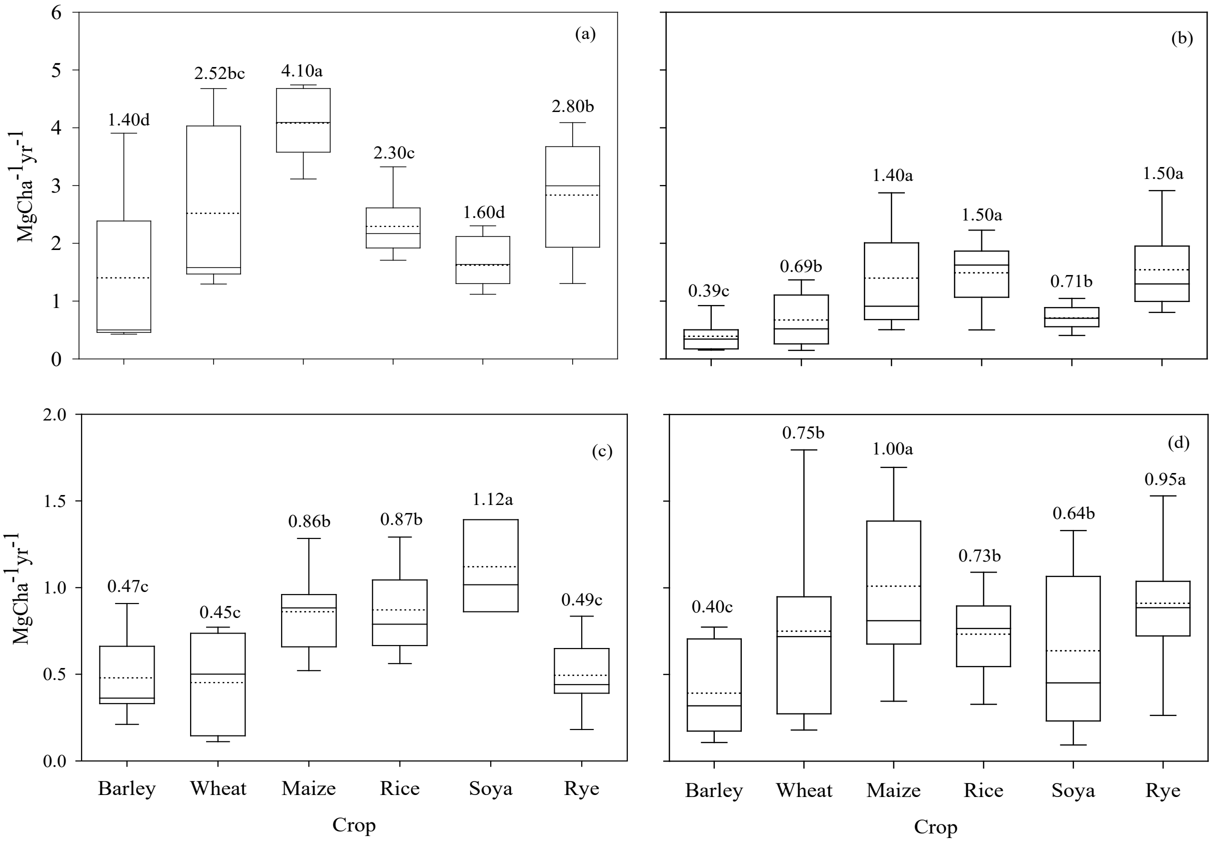


Figure 7.2. C fluxes into (a) Sc: shoot (b) Rc: roots c) REc: respiration and (d) Cseq: soil of the plant/soil system as affected by crop species.

Rye=ryegrass and Soya=soyabean. Values above the top whisker represent mean values of each crop. Means followed by the same letters are not significantly different at p≤0.05

7.3.3. Soil C respiration and sequestration

Soils under soyabean lost the highest mean C (1.12±0.24 Mg C ha^{-1} yr^{-1}) through respiration (REc), and this was 33 and 140% higher than soils under maize and wheat, respectively (Fig 7.2c). Barley, wheat and ryegrass had considerably low REc with a mean value of 0.47 Mg C ha^{-1}. Maize and rice, with a mean of 0.85 Mg C ha^{-1} yr^{-1} C, lost nearly double the amount of C respired by wheat and barley (0.47 Mg C ha^{-1} yr^{-1}). The C sequestered in soils (C_{seq}) by the different crops however varied from 0.39 Mg C ha^{-1} $yr^{-1}$for barley to 1.00 Mg C ha^{-1} yr^{-1} for maize (Fig 7.2d). Maize sequestered the highest amount of C in the soil being about 5% more than the amount sequestered by ryegrass, which had the second highest mean C_{seq}.

Figure 7.3 shows the comparison of C fluxes in one pool to another under the different crops. Soyabean had the largest proportion (0.32±0.07) of unit C transfer to the soil (C_{seq}) per total plant C fluxes (C_{fix}) (Fig 7.3a). Barley was the second most efficient crop because it sequestered 0.29±0.06 units of C per unit of C fixed by the plant. Wheat, maize and rice showed similar efficiencies in terms of C sequestration relative to total C fixed in the plants (between 0.18 and 0.20). Carbon sequestration in the soil relative to shoot C fluxes exhibited a similar trend with soyabean having the highest efficiency followed by barley (Fig. 7.3b). Barley and wheat had the highest C sequestration rate relative to root C fluxes reaching 1.22 units of C transferred to soil per unit C retained in the root (Fig 7.3c). The least C sequestration per unit root C flux was recorded in rice with 0.48 units of the root C reaching the soil. Maize and soybean showed non-significant difference with means of 0.96 and 0.98 for C_{seq}: root C flux, respectively. Figure 7.3d shows the Cseq relative to C emission from the soil under different crops. The highest C seq per unit C emission was observed in ryegrass which recorded 1.88±0.17 units of C transfer to the soil per unit of biomass C respired. Wheat was second with 1.48±0.11 units C sequestration to soil followed by maize (1.34±0.13 units) and barley (1.00±0.23 units).

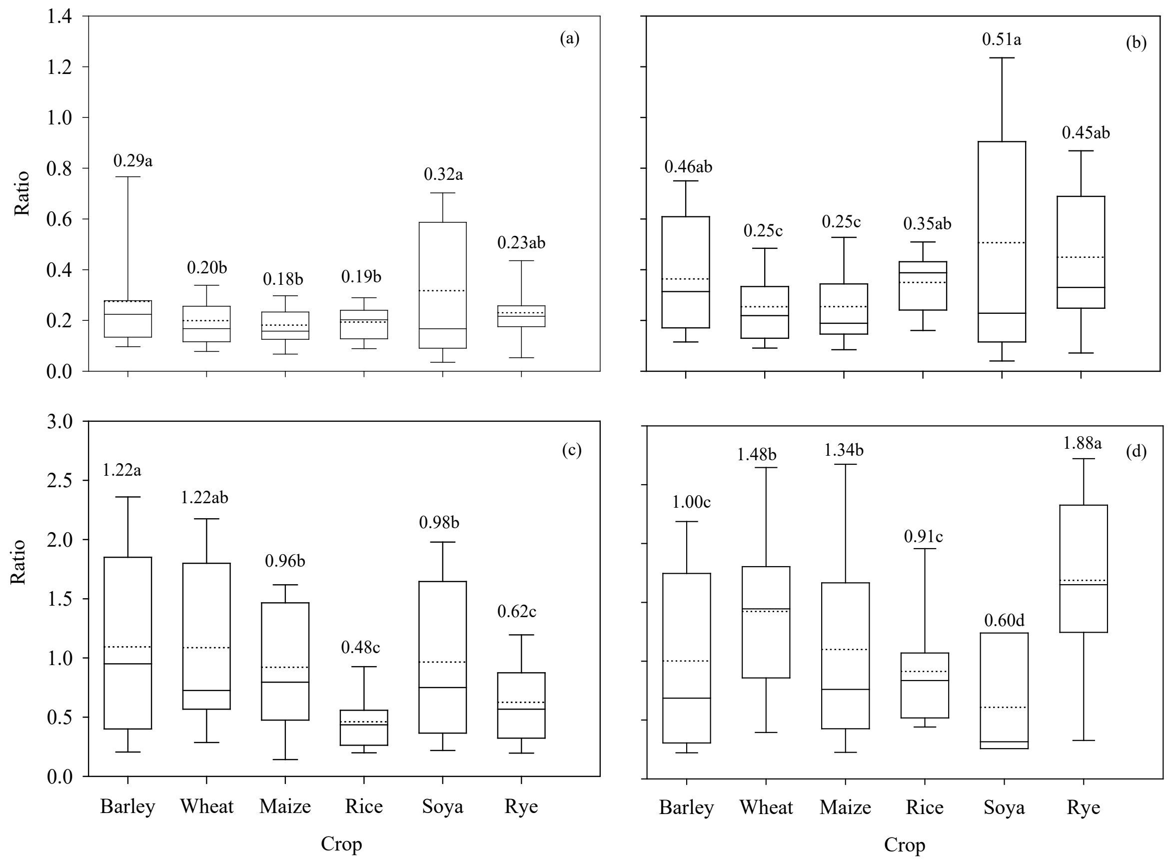


Figure 7.3. Proportion of C sequestered in the soil after isotopic C enrichment to (a) total plant C (b) Sc: shoot C; (c) Rc: root C, and (d) REc: respired C.

Rye=ryegrass; Soya=soyabean. Values above the top whisker represent mean values of each crop. Means followed by the same letters are not significantly different at $p \leq 0.05$

Figure 7.4 compares C fluxes in the plant to C emission (Rec). Ryegrass lost the least amount of C through respiration relative to total plant C fixation as shown by its REc/C_{fix} ratio of 0.11±0.01 (Fig 7.4a). For soyabean and barley, respiration C constituted above 40% when compared to the total plant C fixation while for the rest of the species the ratio was less than 20%. A similar trend for Rec was observed relative to shoot C assimilation. Soyabean had the highest REc/Sc at 0.85±0.13 which was 24% higher than ryegrass and wheat which had the least mean REc/Sc (Fig 7.4b). The trend was different in relation to root C fluxes where barley and soyabean had the highest relative loss of C at 1.49±0.26 and 1.38±0.17, respectively (Fig 7.4c).

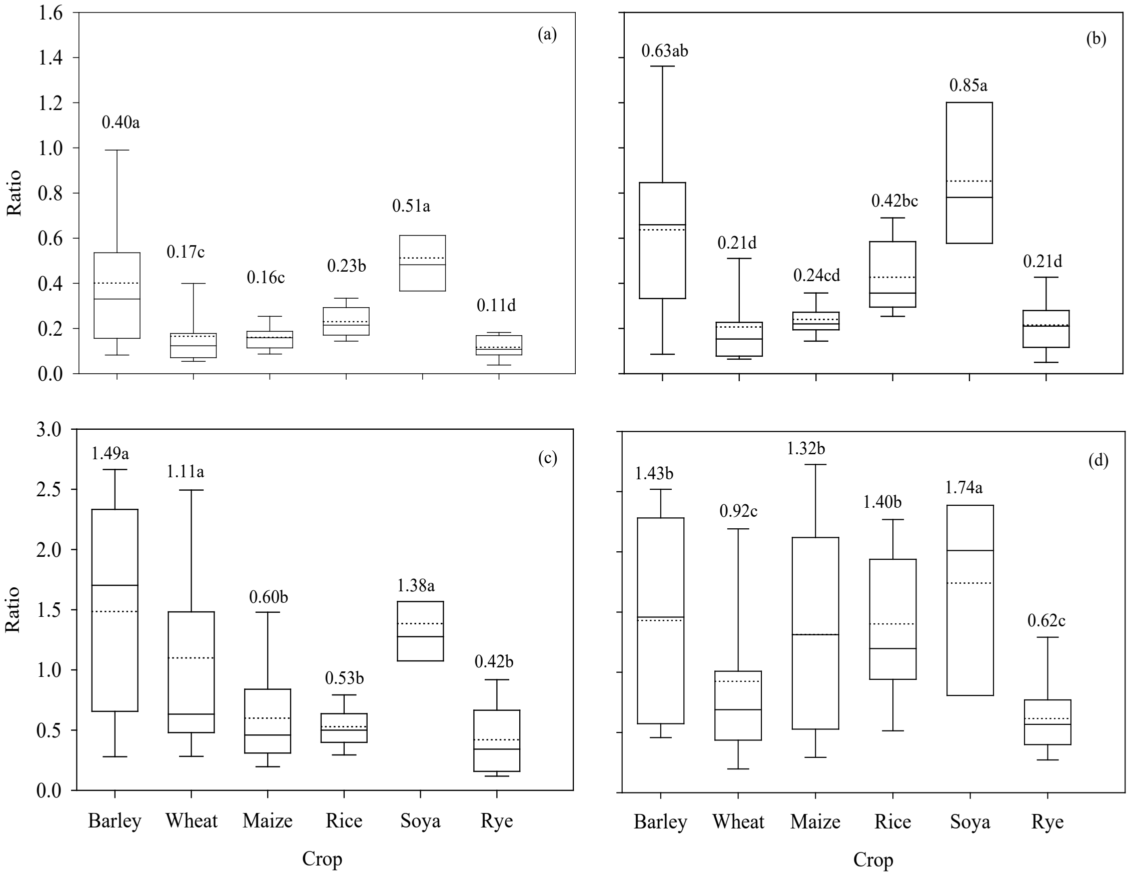


Figure 7.4. Respired C as a fraction of a) total plant C fixation (b) Sc: shoot C (c) Rc: root C and (d) C_{seq}: soil C.

Rye=ryegrass; Soya=soyabean. Values above the top whisker represent mean values of each crop. Means followed by the same are not significantly different at $p \leq 0.05$

7.3.4. Changes in plant C allocation with soil properties and climate

7.3.4.1. Soil texture

Figure 7.5 shows changes in C fluxes in response to soil texture. Under clay soils, the mean shoot C flux was 2.36±0.23 Mg C ha^{-1} yr^{-1} (Fig 7.5a). This was not significantly different from that of loam-textured soils with a mean of 2.70±0.25 Mg C ha^{-1} yr^{-1}. The shoot C fluxes exhibited the widest variability under loam soils ranging between 0.5 and 4.6 Mg C ha^{-1} yr^{-1}. Root C fluxes showed a positive association with clay content ($r=0.21$; $p \leq 0.05$) (Table 7.5). The positive association was depicted in Fig 7.5b where root C fluxes under clayey (1.30±0.13 Mg C ha^{-1} yr^{-1}) and loamy (1.36±0.16 Mg C ha^{-1} yr-1) soils were significantly ($p \leq 0.05$) higher than under sandy soils (0.91 Mg C ha^{-1} yr^{-}1). Conversely, respiration C fluxes were negatively associated with clay content ($r=-0.28$, $p \leq 0.05$). Carbon emission decreased by 60% from 0.75±0.12 Mg C ha^{-1} yr^{-1} in sandy soils to 0.47±0.03 Mg C ha^{-1} yr^{-1} in clayey and (0.46±0.04

Mg C ha^{-1} yr^{-1}) in loamy textured soils (Fig 7.5c). Soil C sequestration was positively associated with soil clay content ($r=0.46$; $p\leq0.05$) (Table 7.5). Figure 8.5d shows a general increase in soil C fluxes from 0.43±0.09 Mg C ha^{-1} yr^{-1} in sandy soils to 1.04±0.07 Mg C ha^{-1} yr^{-1} under loamy and 1.08±0.06 Mg C ha^{-1} yr^{-1} under clayey soils. The soil C fluxes in clay soils were nearly 200% higher than soil C fluxes under sandy soils.

Table 7.5. Correlation coefficients showing pair-wise relationship among C flux variables and controlling factors used in the study for all crops

Variables	Temp	Cl%	SOCc	Sb	Rb	Sc	Rc	C_{seq}	REc	C_{seq}:Sc	C_{seq}:Rc	REc/Sc	REc:Rc	C_{seq}/Sb	C_{seq}/Rb
Temp	1														
Cl%	0.66*	1													
SOCc	0.23*	0.91*	1												
Sb	-0.15	-0.79*	-0.66*	1											
Rb	-0.03	-0.61*	-0.37*	0.80*	1										
Sc	-0.04	-0.11	0.2	0.02	-0.20*	1									
Rc	0.04	0.21*	0.44*	-0.12	0.04	-0.01	1								
C_{seq}	-0.01	0.46*	0.11	-0.13	-0.18	0.11	0.38*	1							
REc	0.34*	-0.28*	0.20*	0.09	0.09	0.19*	0.33*	-0.12	1						
C_{seq}:Sc	0.04	0.40*	0.48*	-0.17	-0.05	-0.51*	0.33*	0.75*	0.01	1					
C_{seq}:Rc	-0.14	0.07	-0.23	-0.04	-0.14	0.22*	-0.67*	0.35*	-0.23*	0.18*	1				
REc/Sc	0.11	-0.26	0.49*	0.05	0.12	-0.59*	0.23*	-0.02	0.57*	0.41*	-0.31*	1			
REc:Rc	0.02	-0.19	-0.16	0.11	0.24*	0.03	-0.78*	-0.34*	0.25*	-0.31*	0.62*	0.18*	1		
C_{seq}/Sb	0.01	0.04	0.01	0.1	0.12	0.02	0.03	0.72*	0.13	0.69*	0.58*	0.13	0.05	1	
C_{seq}/Rb	0.04	-0.35*	-0.1	0.24*	0.22*	0.11	-0.08	0.59*	0.19	0.48*	0.58*	-0.03	0.16	0.88*	1

*=significance at $p \leq 0.05$, Temp=temperature; Cl%=soil clay content; SOCc=soil organic carbon content; Sb=shoot biomass; Rb=root biomass; Sc=shoot C fluxes; Rc=root C fluxes; Soc=soil C fluxes; REc=respiration C fluxes; SOc:Sc, SOc:Rc, REc/Scs, REc:Rcs, SOc/Sb and SOc/Rb are ratios of the respective variables

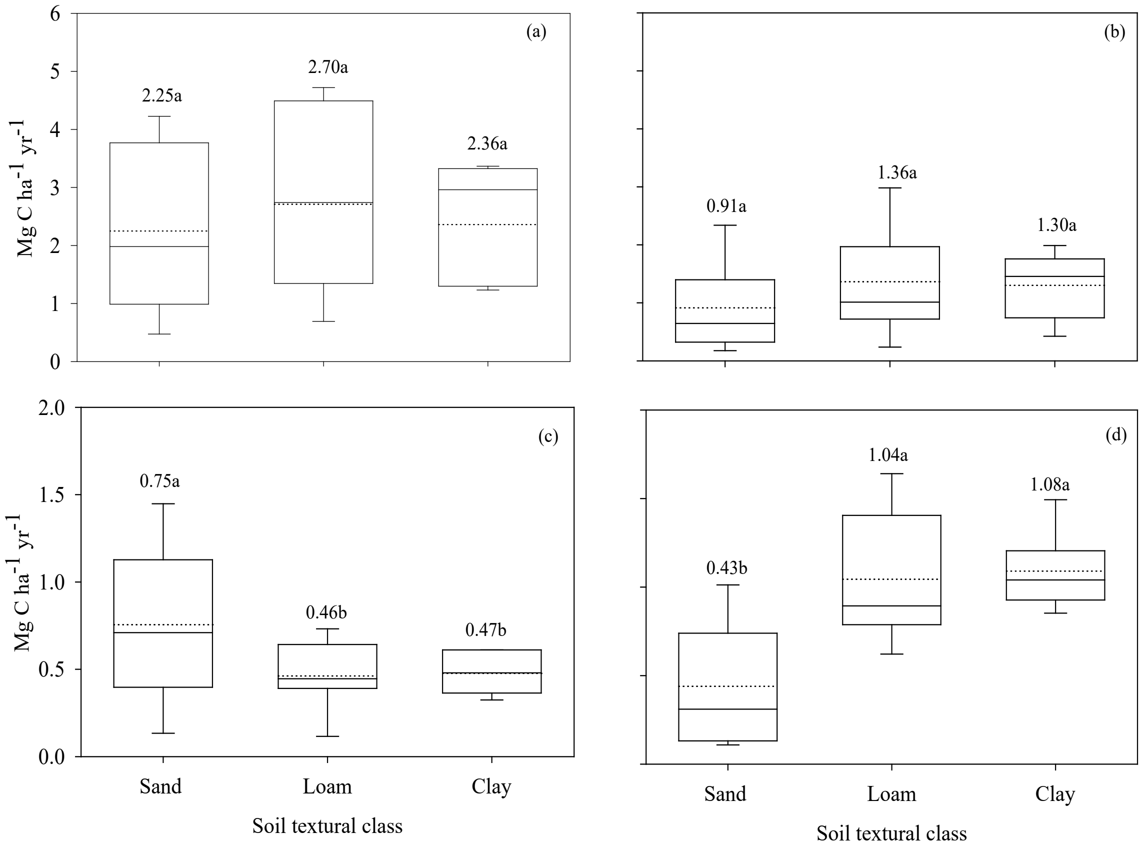


Figure 7.5. Comparison of C fluxes (Mg C ha^{-1}yr^{-1}): a) Sc: shoot C b) Rc: root C c) REc: respiration C and d) Cseq: soil C pools across different soil clay classes.

Values above the top whisker represent mean values for respective soil texture class. Means followed by the same letter are not significantly different at $p \leq 0.05$.

7.3.4.2. Soil carbon content

Soil carbon content exhibited a positive association with shoot C fluxes although the relationship was non-significant ($p>0.05$) (Table 7.5). Thus, the highest mean shoot C fluxes of 2.92±0.11 Mg C ha^{-1} yr^{-1} were recorded in soils with large SOCc, while the least was recorded in small SOCc soils with a mean of 1.83±0.27 Mg C ha^{-1} yr^{-1} (Fig 7.6a). Soil OC content also showed a positive effect on root C fluxes ($r=0.44$; $p\leq0.05$). The increase in root C fluxes was gradual, showing a significant increase of only 33% from 0.79±0.08 Mg C ha^{-1} yr^{-1} under low SOCc to 1.07±0.08 Mg C ha^{-1} yr^{-1} under medium SOCc soils (Fig 7.6b). However, root C fluxes under medium and high SOCc soils (1.01 Mg C ha^{-1} yr^{-1}) were not significantly different ($p>0.05$). Respiration C flux exhibited a positive correlation with SOCc ($r=0.20$; $p\leq0.05$) (Table 7.5). The C fluxes due to respiration were not significantly different under low and medium SOCc soils at 0.56±0.13 and 0.59±0.19 Mg C ha^{-1} yr^{-1}, respectively, while they were significantly higher under high SOCs (0.87±0.06 Mg C ha^{-1} yr^{-1}) soils ($p \leq 0.05$), (Fig 7.6c). The soil sequestered C increased by 60% from than 0.56±0.12 Mg C ha^{-1} yr^{-1} under low SOCc soils to medium and high SOCc soils (combined mean 0.93 Mg C ha^{-1} yr^{-1}), which were not significantly different (0.84±0.06 and 1.01±0.16 Mg C ha^{-1} yr^{-1} under high and medium SOCc soils, respectively) (Fig 7.6d).

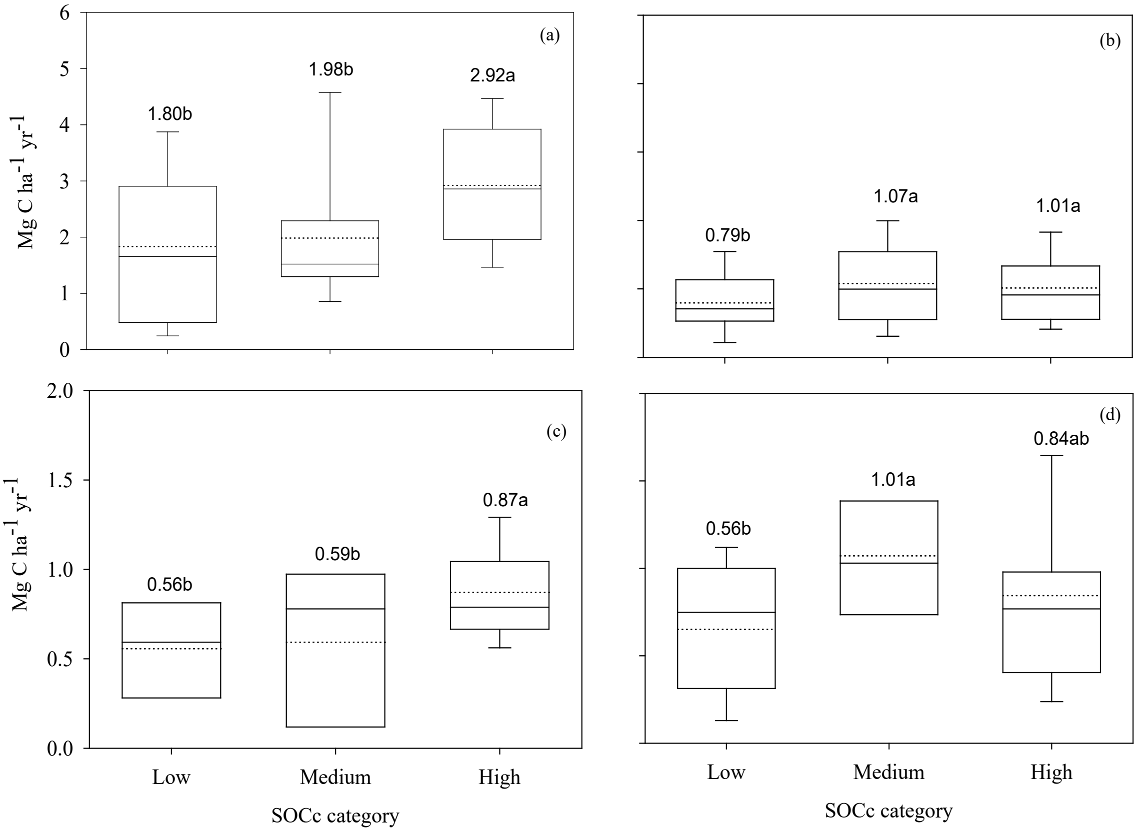


Figure 7.6. Comparison of C fluxes (Mg C ha^{-1}yr^{-1}) in response to soil organic carbon content (SOCc) in a) Sc: shoot C flux b) Rc: root C flux c) REc: respired C and d) C_{seq}: sequestered C in soil.

Values above the top whisker represent mean values for respective SOCc categories. Means followed by the same letter are not significantly different at $p \leq 0.05$.

7.3.4.3. Climate

Bivariate analysis revealed a weak negative correlation between temperature and shoot C fluxes (Table 7.5). Figure 7.7a supports this as there were no significant differences in shoot C among the different climate zones. Correlation also showed that it was not significant for root C fluxes and temperature (Table 7.5). Thus, root C fluxes were not significantly different under cool (0.75±0.14 Mg C ha^{-1} yr^{-1}) and warm temperatures (0.99±0.12 Mg C ha^{-1} yr^{-1}), but there was a sharp increase of 200% from warm to hot temperatures (1.51±0.15 Mg C ha^{-1} yr^{-1}) (Fig 7.7b).

The association between temperature and C emission was relatively strong and positive ($r=0.34$; $p \leq 0.05$) (Table 7.5). Carbon emissions under warm (0.63±0.01 Mg C ha^{-1} yr^{-1}) and hot (0.62±0.02 Mg C ha^{-1} yr^{-1}) temperatures were similar, while they were significantly lower under cool temperatures (0.54±0.05 Mg C ha^{-1} yr^{-1}) (Fig 7.7c). Soil C fluxes exhibited a negative correlation with temperature (Fig. 7.7d). However, the differences in soil C fluxes between cool (0.90±0.08 Mg C ha^{-1} yr^{-1}) and warm (0.86±0.05 Mg C ha^{-1} yr^{-1}) temperature zones were not significant, while they were lowest under hot temperatures (Fig 7.7d).

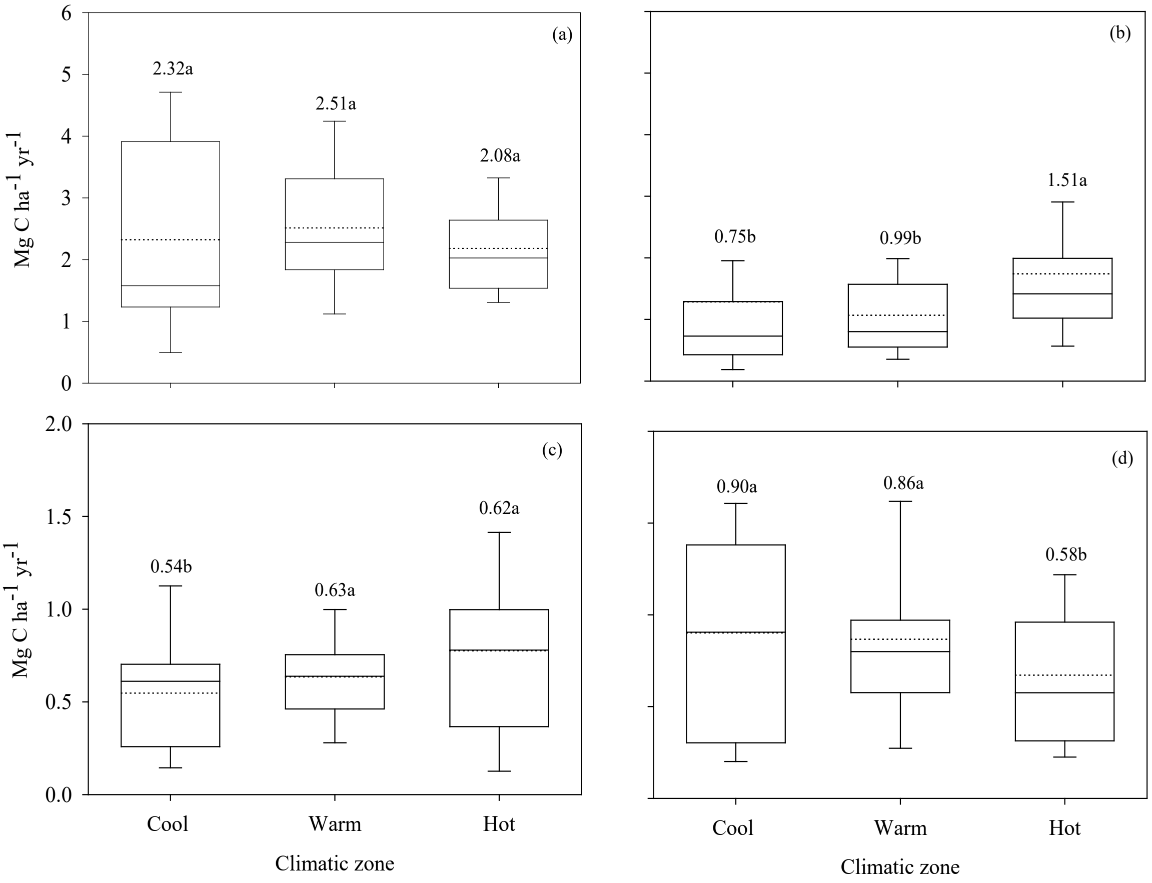


Figure 7.7. Comparison of C fluxes (Mg C $ha^{-1}yr^{-1}$) under different temperature ranges in a) shoots b) roots c) respiration and d) soil pools.

Values above the top whisker represent mean values for respective temperature class. Means followed by the same letter are not significantly different at $p \leq 0.05$.

7.3.5. Multivariate analysis

Principal component analysis (PCA) was conducted using data from all crop species. The first two principal components (PCs) (Fig 7.8) accounted for 68% of the data variability. The first principal component (PC1), which accounted for 40%, correlated the most with soil organic C content (SOCc) with positive coordinates. Therefore, it can be inferred that soil carbon respiration (REc) increased the most with SOCc because both REc and SOCc increased in the same direction. There was also an increasing trend for REc with shoot and total plant C allocation. Principal component 2 (PC2), accounting for 28% of the variability in data, correlated the most with the shoot (Sb) and plant (Pb) biomass with positive coordinates, and temperature (Temp) and soil clay content (Clay) on the negative side. Therefore, carbon fluxes to both the roots and soil media seemed to increase with soil clay content and temperature. Interestingly, C inputs to the roots and soil appeared to decline with increasing shoot and total plant biomass. These observations are in general agreement with the results of correlation analysis presented in Table 7.5.

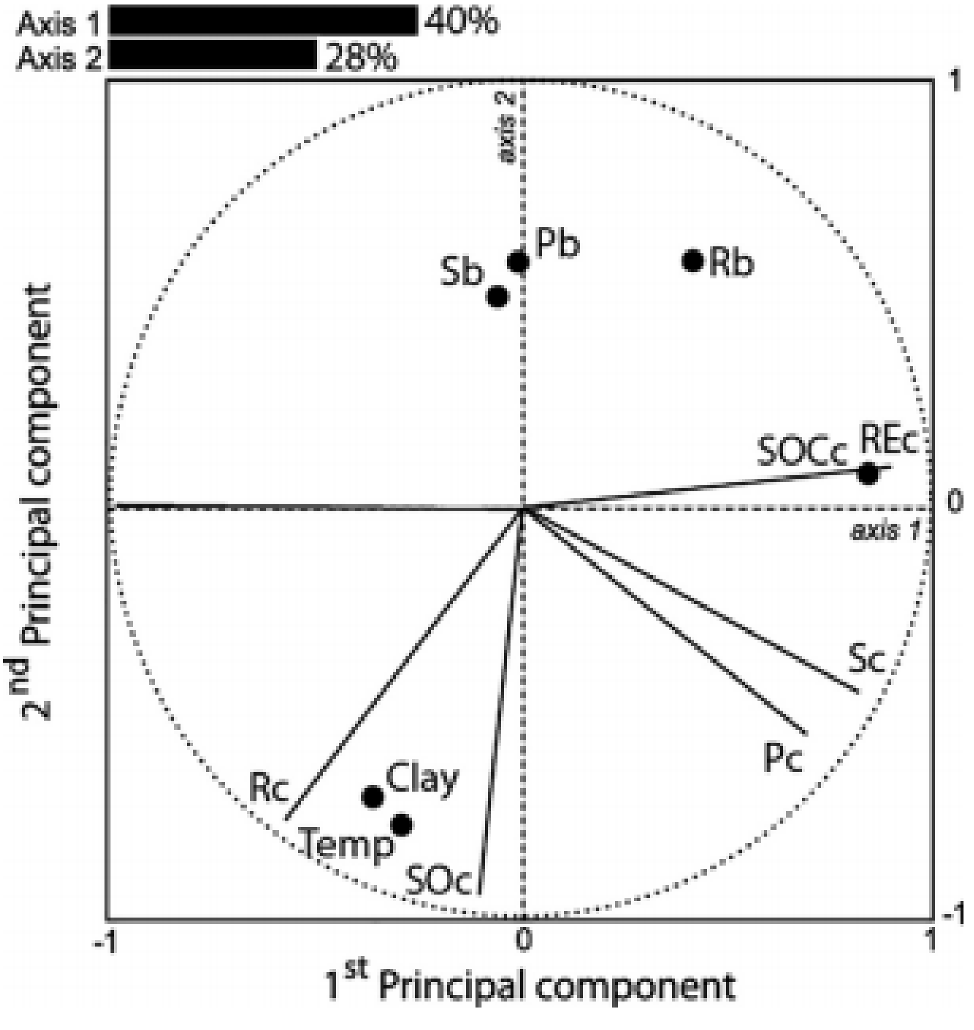


Figure 7.8. PCA showing multivariate relationship among variables across all crop types.

Sc=shoot C fluxes, Rc=root C fluxes, C_{seq}=soil C sequestration, REc=respiration C fluxes, Rb=root biomass, Pb=plant biomass, Rb/Sb=biomass root:shoot, Temp=temperature and SOCc=soil organic C content.

7.4. Discussion

7.4.1. The impact of crop species on C assimilation from the atmosphere to plant biomass

The wide variations in C fluxes from the atmosphere to plant shoots of 0.2 to 5.6 Mg C $ha^{-1}yr^{-1}$ was expected and are likely to result from genetic differences and variable environmental conditions (Staddon et al., 2004; Warembourg et al, 2003). Carbon assimilation in shoots is affected by growth pattern and internal C metabolism, which are under genetic control. Maize exhibited a 44% higher assimilation of atmospheric C into shoots than ryegrass confirming previous reports by Pausch and Kuzyakov (2017) and Atkinson et al. (2014). The fact that maize assimilated greater C amounts shows that annual crops retain more above ground C than perennial grasses in short growth cycles due to accelerated growth rates. The differences in shoot C fluxes among annual crops could be partly due to variations in C capture and internal C metabolism. For instance, maize has a higher leaf area and C4 photosynthetic pathway, which render it more efficient in atmospheric C capture, while its high concentration of lignin and hemicellulose result in the retention of more C in the stover (shoots) than in the straw of C3 crops (wheat or rice) (Adapa et al., 2009; Li et al., 2013).

The flux of C to roots in maize, rice and ryegrass was above the mean of 1.0 Mg C ha^{-1} yr^{-1}across all crop types, which agreed with results from Huang and Johnson (1995) and Mathew et al. (2017) showing that these plants had higher efficiency in C translocation to roots. Genetic variations and growth habit influenced root C fluxes. For example, ryegrass has a perennial growth habit that increases the root C fluxes for energy storage (Purdy et al., 2015). The high root C fluxes in maize are associated with its high photosynthetic capacity which avails large amount of assimilates for translocation to the roots (Amanullah and Stewart, 2013; Pausch and Kuzyakov, 2017) after meeting demands in the above ground biomass (Ludewig and Flügge, 2013). In rice, the high root C fluxes are a result of the plant's response to maintain root development under hypoxic conditions (Huang and Johnson, 1995). Increased C allocation to organs under stress such as hypoxia is in accordance with the 'functional equilibrium' hypothesis which states that plants will allocate more C resources to an organ under stress in order to reduce the impact of the stress on plant growth (Poorter and Nagel, 2000). In general, crop species and genotypes which translocate higher quantities of C to the roots have higher potential for soil C sequestration to fulfil the ambitions of the 4p1000 initiative provided the roots exude large amounts of C. The root debris is also left to breakdown in the soil and soil aggregates protect the deposited C.

7.4.2. The link between plant C fluxes and C transfer to the soil

The lack of significant correlation between shoot C fluxes and soil C sequestration concurs with several studies that found no impact of shoot residues on SOC (Rasse et al., 2005). The contribution of shoot C to soil C is known to be negligible due to high temperatures and humidity at the soil surface which favour litter decomposition (Heal et al., 1997; Palm et al., 2001). The existence of a significant correlation between root C fluxes and soil C sequestration ($r=0.38$; $p \leq 0.05$) confirms prior knowledge that root derived C constitutes a substantial proportion of soil C (Rasse et al., 2005). Root C contributes to soil C through various ways, e.g. provision of C exudates, which are eventually stabilized in the soil, physical protection of soils against erosion, which keeps C deposits intact, formation of associations with mycorrhizae for C immobilization and providing exchange site for chemical interactions between plant and soil ions (Rasse et al., 2005). Intuitively, higher C allocation to roots increases the potential for higher C transfer to soils as more exudates are released into the soil profile.

Crop species differ in the amounts of C allocation to the roots and in their root characteristics, which subsequently results in differences for C transferred to soil by the different crop species. Ryegrass and rice exhibited high soil C sequestration, concomitant with higher C allocation to the roots. However, maize also showed high soil C sequestration (1.0 Mg C ha^{-1} yr^{-1}) since it had similar root C fluxes to ryegrass. Soil C sequestration under wheat (0.8 Mg C ha^{-1} yr^{-1}) was also relatively high despite low root C fluxes and this suggests that wheat probably experience higher exudation rate per unit of root C that contributes to soil C. It was also interesting to observe that soyabean had low soil C sequestration ability (0.64 Mg C ha^{-1} yr^{-1}), despite its high root C fluxes and high C sequestration relative to total plant C fixation. This suggests that a large proportion of C translocated to legume roots is used up during microbial respiration, which is released into the atmosphere as shown by the high REc. In the short term, soil C sequestration under annual crops such as maize may be higher than perennials such as ryegrass because C exudation below ground in annual crops peaks earlier than in perennials (Pausch and Kuzyakov, 2017). On the contrary, ryegrass is likely to contribute more stable C to soils through root turnover rather than exudation (Mota et al., 2010; Menichetti et al., 2015). In addition, chemical, physical and biochemical processes influence the amount of C stabilized in the soil. Thus the C: N ratio is one of the biochemical parameters which has a great influence on the mean residence time of the plant deposited C in the soil. Hence plant matter with low C: N ratio, like soyabean stover, are easily decomposed by microbes but have rapid effect on soil aggregation which protects deposited C, while those with high C: N are more recalcitrant and have longer term impact on soil C reserves (Abiven et al., 2009). On the other hand, cereals will likely take longer to decompose due to their considerably higher C: N but do not provide adequate soil aggregation for C protection. Among the cereals, wheat could have a long-term impact due to its higher C: N ratio compared to maize.

Ideally, a plant must have high C root flux and total plant C fixation capacity to sequester high amounts of C into the soil. However, high C transfer to the soil may not translate to long-term stabilization since the mean residence time of the deposited C will depend on its quality, microbial activity and soil properties. In general, only up to 5% of the C assimilated by plants is eventually stabilized in the soil after accounting for respiration (Hütsch et al., 2002). All these sequestration rates were very high when compared against the overall average of 7% which could be a result of controlled environmental factors under greenhouse conditions. The C sequestration rates exhibited by the six crops (from 0.4-1.0 Mg C ha^{-1} yr^{-1}) are comparable to C sequestration rates reported for land management practices (0.1-1.0 Mg C ha^{-1} yr-^{1}) (Paustian et al., 2016), which means that focusing on the C sequestration potential of crops is equally important for mitigating global warming. Against this backdrop, ryegrass, maize, rice and wheat appear to fit the objectives of 4p1000 due to their high soil C sequestration rates coupled with low returns of CO_2 to the atmosphere. Soyabean may not be a good candidate crop for 4p1000 due to higher losses of C to the atmosphere through respiration.

7.4.3. The link between C transfer to the soil and soil CO_2 emissions

The meta-analysis showed no significant correlation between soil C sequestration and CO_2 emissions suggesting that C losses through respiration were negligible to cause detectable shifts in amount of C in soils in the short term. However, extremes were still noted with, for example, soyabean emitting high amounts of C (1.12 Mg C ha^{-1} yr^{-1}) while ryegrass emitted low amounts (0.49 Mg C ha^{-1} yr^{-1}). The high amount of carbon emission under soyabean was linked to high microbial respiration due to low C: N ratio (Schmitt et al., 2013). Contrastingly, wheat, barley and ryegrass have higher C: N ratios which discourage rapid microbial decomposition. In addition, ryegrass and other perennial grasses have evolved over time through natural selection to reduce C emission and conserve C for energy during overwintering periods making them more efficient at transferring C to the soil (Pausch and Kuzyakov, 2017). Their contribution to soil C is largely through root turnover rather than exudates. Root C tends to be more stable than exudate C (Mota et al., 2010; Menichetti et al., 2015). The higher CO_2 emissions under maize (0.9 Mg C ha^{-1} yr^{-1}) than wheat and barley (0.5 Mg C $ha^{-1}yr^{-1}$) indicate differences in the internal

C cycling influenced by C: N ratio mentioned above and also, possibly, shallower soil C allocation. Maize incurs higher C emissions due to synthesis of the lignin and hemicellulose-rich biomass (Adapa et al., 2009; Li et al., 2013) and lower C: N ratio than barley and wheat (Velthof et al., 2002). Again, maize has been reported to have shallower rooting depth (118 cm) compared to wheat (150 cm) or barley (146 cm) (Fan et al., 2016), which means it deposits C closer to the soil surface than wheat or barley, which could have contributed to high CO_2 effluxes under maize. Despite higher CO_2 emission, maize still sequestered more C to the soil than wheat, barley and rice showing that it is more efficient in atmospheric C fixation (Amanullah and Stewart, 2013) which render large amounts of C for translocation to the soil. This shows that soil C accumulation can still be driven by root C fluxes provided they are high enough to offset CO_2 emissions. The foregoing clearly demonstrates that maize and ryegrass had superior ability for C sequestration into the soil than the other plant species, while wheat seems to be the most efficient among the winter crops.

7.4.4. The effect of environmental conditions on soil C transfer

The positive correlation between soil C sequestration and clay content ($r=0.46$; $p\leq0.05$), and the fact that clayey soils stabilized 200% more soil C than sandy soils show the importance of soil clay fractions in C dynamics. Clay particles provide adsorption and aggregation sites for the stability of C compounds and protection from microbial decomposition which increases persistence of organic C in fine-textured soils in comparison with coarse texture soils (Hütsch et al., 2002). There is therefore a need to complement plant C sequestration in the soil by adopting good land management practices in order to maintain or improve soil physico-chemical properties. The lack of a significant correlation between soil C sequestration and temperature was rather surprising and contrary to other study reports (e.g. Jones et al., 2005; Davidson and Janssens, 2006). Giardina et al. (2014) reported that high temperatures promoted root C fluxes but reduced exudation because the roots would retain their C with increasing temperatures to maintain vital biological processes for stress avoidance.

7.5. Conclusion

This study used data from 227 research trials and evaluated the allocation of assimilated carbon (C) to plant shoots and roots, and ultimately to the soil. The main conclusions drawn from this study were:

(i) C transfer was highest in maize which yielded the greatest soil C sequestration rate (1.00 Mg C ha^{-1} yr^{-1}) followed by ryegrass (0.95 Mg C ha^{-1} yr^{-1}), rice (0.70 Mg C ha^{-1} yr^{-1}), and wheat (0.80 Mg C ha^{-1} yr^{-1}). These rates are comparable to potential C sequestration of many land management practices.

(ii) Only 7% of the assimilated C was stabilized in the soil but specifically wheat transferred 23% of its total C allocation to the soil followed by rice with 20%, and maize and ryegrass at 19%.

(iii) Higher C transfer to soils occurred under clayey soils and warmer climates provided that exudation is high enough to offset respiration C losses.

These results could be used in selecting crop species and ideotypes for high soil C sequestration, while the variability within species could be useful in breeding efforts to enhance the genetic capacity of existing crops to sequester C. However, there is need to identify genetic markers associated with the key traits for C sequestration to enhance marker-assisted breeding and trait introgression into agronomically desirable genotypes. The main limitation of the study was that the C fluxes were estimated from planting to harvest season, while multiple cycle or across year experiments should possibly provide a more accurate estimation of soil C sequestration by crops.

Chapter 8 BIOMASS PRODUCTION AND CARBON SEQUESTRATION OF MAIZE, SORGHUM AND WHEAT UNDER GLASSHOUSE CONDITION

Abstract

Biomass productivity and its root/shoot ratios are important traits for estimating C fluxes and potential C sequestration in crops. However, investigations of biomass and C partitioning in field crops is still lagging behind, which hinders efforts to estimate their contribution to the C global cycle or their potential to offset C emissions from agricultural soils. Against this background, this study evaluated 10 genotypes each of maize, sorghum and wheat under greenhouse conditions, to elucidate their biomass production and potential for C accumulation using a completely randomized block design experiment. Significant differences in genotypic effects in biomass and C stocking potential were observed among the genotypes and treatments for shoot and root biomass and root: shoot ratio ($p \leq 0.05$). However, root and shoot carbon and nitrogen content were not significantly different within the crops. The aboveground biomass of maize ranged from 231 g m^{-2} in genotype Shesha Capstone under drought stressed to 1401 g m^{-2} for genotype Kep Select under well-watered conditions. AS15 had the highest shoot biomass of 2356 g m^{-2} among the sorghum genotypes while the lowest was 1120 g m^{-2} recorded in AS16. In wheat the shoot biomass ranged from 638 g m^{-2} in LM48 under drought stress to 1196 g m^{-2} in BW140 under well-watered conditions. Below ground biomass followed the same trend with respective means of 379, 497 and 1403 g m^{-2} for maize, wheat and sorghum respectively under well-watered conditions. Carbon content and C stocks were higher in sorghum followed by maize and wheat with respective root C stocks of 370, 150 and 100 g m^{-2} under drought stress conditions. Sorghum exhibited the highest potential to sequester C into roots and possibly to the soil. However, maize and wheat have potential for longer C residence time in the soil as they have higher C: N ratio in the root residues which increases C recalcitrance. The C stocking capacity of maize and wheat was limited by their low biomass production capacity. Biomass productivity is the key driver of C sequestration. Root biomass C in particular can be potentially transferred to the soil for organic matter stabilization under adequate moisture.

Keywords: biomass; carbon sequestration; root: shoot; drought; maize, sorghum, wheat

8.1. Introduction

Atmospheric C concentration has increased dramatically over the years due to a combination of industrialization and land mismanagement. Agricultural activities contribute 22-31% of the total anthropogenic carbon (C) emissions (Smith et al., 2014; Tubiello et al., 2015). Rising temperatures across the world and the subsequent changes in climate curtail crop production prompting the search for strategies to mitigate against this. The accumulation of C in the atmosphere and its loss from the soil are reversible processes. One of the strategies to cope with a variable climate is harnessing the capability of crops to foster transfer of C to the soil during their life cycle. Reducing atmospheric carbon (C) by fostering its transfer to the soil through phyto-sequestration is a sustainable intervention to reduce global warming. Assimilation of atmospheric C during plant growth and its subsequent transfer to soil by plant roots has the potential to offset 20% of CO_2 emissions from agricultural lands. Globally, plants can assimilate up to 123±8 pentagrams of carbon per year (Pg C yr^{-1}) (Beer et al., 2010) during biomass synthesis. A study by Hütsch et al., (2002) estimates that 2-8% of plant assimilated C can be stabilized in the soil. Plants can then transfer between 3 and 10 Pg C yr^{-1} to the soil which is sufficient to offset annual C emission from agricultural lands (Jannson et al., 2010). On a global scale, the soil sink has capacity to absorb more than 120 GigaTons C (GtC yr^{-1}) (Jansson et al., 2010) that is sufficient to absorb the entire potential C fixed by plants.

Carbon sequestration by plants hinges on biomass productivity and differential C investment between shoots and roots. This means the amount of crop residues and their quality are important determinants of soil C fluxes (Lynch and Wojciechowski, 2015; Dou et al., 2007). High biomass productivity increases the potential for C sequestration. Biomass and C allocation between roots and shoots also influence the sequestration potential of a crop. Above ground residues represent a small pool of C and their impact on C sequestration in the soil is minimal (Kumar et al., 2006). In contrast, root derived C accounts for up to 80% of C stocks in ecosystems (Yang et al., 2012). Intuitively, higher root biomass and high root/shoot ratios increase the potential for stable C sequestration in the soil by plants. Deeper and denser plant root systems potentially stabilize more C in the soil by releasing exudates into deeper horizons, which physically protect the C from immediate microbial decomposition back to the atmosphere. However, information on below ground C fluxes is still limited (Mokany et al., 2006), and there exist a lot of uncertainties in estimating crop C budgets due to complexity of measuring root parameters.

The partitioning of biomass and C between roots and shoots differs widely among crop species and thus they have different C sequestration capacity. There are few published studies on biomass and C allocation in relation to C sequestration potential of annual crops (Paustian et al., 2016). Information on C sequestration of annual crops will assist in efforts to improve soil C status of communal farming ecosystems and contribute to offsetting C emission for climate change mitigation. The majority of soils in smallholder farming communities have very low nutrient status and the retention of crop residues will contribute to improving soil nutrient status. Information on biomass productivity, biomass root: shoot ratios, C and N content and C: N ratio in the plant residues will help to quantify the potential C contribution from crop residues. Therefore, a study was conducted to quantify biomass productivity, and C and N content in maize and sorghum, which are commonly grown by smallholder farmers (Zegada-Lizarazu et al., 2012) as well as wheat, which is usually grown under commercial production in South Africa. Maize and sorghum are ranked the first and second, respectively, in extent of production area and food for the majority of people in Sub-Saharan Africa (Botha and Viljoen, 2008). In South Africa, sorghum is grown at smallholder scale in various provinces including Mpumalanga, North West, Northern Cape, Eastern Cape, and KwaZulu-Natal while in the Free State it is being commercialized (DAFF, 2010). It is also used extensively as livestock feed and is a potential biofuel. There is wide genetic diversity of sorghum in South Africa including sweet and grain sorghum but they have not been adequately characterized for breeding (Mofokeng et al., 2014), especially for C sequestration potential. Maize is a staple food in many countries around the world. It has been identified as having the potential to sequester more C among annual cereals in a meta-analysis study (Mathew et al., 2017). On average, maize allocates 52% and 7% of its total C to shoots and roots respectively, (Johnson et al., 2006) and stabilizes 8-20% of biomass C into the soil (Bolinder et al., 1999). Wheat is an important food crop in the world. However, its production is being challenged by unfavorable climate. Drought is the single most climate related challenge constraining wheat production in the South Africa, causing widespread food insecurity. Identifying genotypes with high carbon sequestration and high biomass production potential under water limited conditions will help to develop drought resilient cultivars for food security. Hence, the aim of this study was to quantify biomass production, biomass allocation, C and N accumulation capacity and C: N ratios in different maize, sorghum and wheat genotypes to elucidate C allocation and estimate C sequestration potential.

8.2. Materials and methods

8.2.1. Plant materials

Ten genotypes each of maize, sorghum and wheat obtained from various sources were evaluated in this study. Four commercial varieties of maize namely PAN4P-228, Shesha, Mac Medium Pearl and Kep Select were included among the ten as possible control checks since they are already in production across South Africa. The sorghum and wheat accessions are still in the developmental stages and were previously evaluated for drought tolerance and diversity.

8.2.2. Site description

The experiments were carried out at the Controlled Environment Facility of the University of KwaZulu-Natal Pietermaritzburg Campus between January and July 2018.

8.2.3. Agronomy and soil moisture treatments

The potting media consisted of soil sourced from Cedara agricultural research station in Pietermaritzburg. It was analysed for physico-chemical properties using standard analytical methods, and results are shown in Table 8.1. Five seeds of sorghum were sown around the perimeter of the pot in a glasshouse, and were later thinned to two plants per pot (equivalent to 18 plants m^{-2}) after 4 weeks of establishment. For maize, three seeds were sown and later thinned to two plants per pot (equivalent to 18 plants m^{-2}) after 4 weeks of establishment. For wheat, ten seeds of a single genotype were sown in a 25 cm diameter plastic pot. The number of plants was thinned to five per pot three weeks after emergence providing about 80 plants per square meter. Fertilizer was applied through automated drip irrigation at a rate of 300 kg N ha^{-1} and 200 kg P_2O_5 ha^{-1} for all crops. The average day/night temperatures in the greenhouse were 25/12°C, with natural photoperiod of about 11 hours. Weeds were manually removed while pests and diseases where chemically controlled by a combination of Tilt fungicide for powdery mildew and Aphox for the control of aphids. The plants were grown under drought-stressed and non-stressed (well-watered) conditions. In the non-stressed regime, the plants were watered to field capacity (FC) whenever average soil moisture content fell to 80% of FC. The volumetric soil moisture content was allowed to drop to 30% of FC in the drought stressed treatment before watering back to FC. The two watering treatments were maintained until maturity (~120 days).

Table 8.1. Soil properties and glasshouse temperature used in this study

Property	Value
Bulk density (gcm^{-3})	0.75
Phosphorous (mg/L)	122
Potassium (mg/L)	289
Calcium (mg/L)	1906
Magnesium (mg/L)	404
Electrical Conductivity ($Cmol_c/L$)	13.72
pH (KCl)	5.09
Organic carbon (%)	5.5
Nitrogen (%)	0.48
Clay (%)	16
Mean Temperature (°C)	25.65

pH (KCl) = pH measured in 1: 5 potassium chloride solution

8.2.4. Data collection

Plant, shoot and root biomass and their respective ratios were determined after the plants were cut at the ground level in each pot to separate roots from shoots. Plant parts for each plot were separated, oven dried at 60°C for 72 hours and then weighed to record shoot biomass (SB) and root (RB) biomass. Root: shoot (RS) ratio was computed after determining RB, and SB. All biomass variables were normalized to grams per square meter based on a population density of 18, 18 and 128 plants m^{-2} for maize, sorghum and wheat, respectively. The different plant parts (root and shoots) were dried in the oven at 60°C for 72hrs to constant weight. The total plant biomass was determined as the sum of shoot and root biomass weights. The C and N contents of the plant roots and shoots for the different crops were measured using the LECO CNS analyzer in the laboratory, after grinding the residue in a mill with a 40 µm mesh screen. The respective C: N ratios were computed for each plant component.

8.2.5. Data analyses

The experiment was set up in a randomized complete block design with two replications per genotype for each crop. Statistical analysis was conducted using Genstat 18 (Payne et al., 2017). A two-way ANOVA was done using procedure for balanced designs, and genotypes were treated as factors. The means of genotypes are presented to describe genotype main effects. The least significant difference (LSD) test at the 0.05 probability level was used to compare significant differences among treatment means.

8.3. Results

8.3.1. Genotypic variation in biomass and C content among the different crops

8.3.1.1. Maize shoot biomass

Biomass accumulation in the shoot was significantly different ($p<0.05$) among maize genotypes and across water regimes (Table 8.2) ranging between 231 to 805 g m^{-2} under drought stressed and 759 to 1401 g m^{-2} under non-stressed conditions (Table 8.3). Under drought stress conditions, Mac Medium Pearl had the highest SB production of 805 g m^{-2} while Shesha CPSTN had the least (231 g m^{-2}). In contrast, Kep Select had the highest SB (1401 g m^{-2}) under well-watered conditions while 1421/DT-STR and R201 had the least SB (759 g m^{-2}). Overall, drought stress reduced SB by 51% relative to well-watered conditions.

8.3.1.2. Maize root biomass

Root biomass accumulation was significantly different ($p<0.05$) among maize genotypes and between the water regimes (Table 8.2). The variety ZM1423 had the highest RB of 684 g m^{-2} while R201 had the least at 164 g m^{-2} attained under well-watered conditions (Table 8.3). The variety Nelson QPM CPSTN produced the highest root biomass (427 g m^{-2}) under water-stress compared to Obatanpa, which produced only 32 g m^{-2}. Increasing water availability, improved root biomass production by 46%. Genotype R201 consistently ranked low in biomass production under both water regimes.

Table 8.2. Mean squares and significant tests for selected traits of 10 maize genotypes

Source of variation	d.f.	SB	RB	RS	RC	SC	C:N
Replicates	1	73	21166	0.071	60.43	0.3456	586.4
Genotypes (G)	9	127588**	79663***	0.0878*	12.77	0.5038	158.7
Water regime (WR)	1	3110962***	143204**	0.173*	125.11	1.7406	141.6
G*WR	9	24874	26794	0.04	8.64	0.3503	113.3
Residual	18	33158	13765	0.025	29.65	0.8326	205.2

SB=shoot biomass, RB=root biomass, RS=root to shoot ratio, RC=root carbon content, SC=shoot carbon content, C: N=carbon to nitrogen ratio, df=degrees of freedom

8.3.1.3. ***Maize root to shoot ratio***

The R: S ratio for maize ranged between 0.09 and 0.75 under drought stressed conditions and 0.18 and 0.61 under well-watered conditions (Table 8.3). The genotypes showed variation with PAN4P-228 and Obatanpa exhibiting the highest and lowest R: S, respectively, under drought stress conditions. Under well-watered conditions, the highest and lowest R: S were attained by genotypes ZM1423 and R201, respectively. Overall, drought stress increased RS by 38% compared to non-stress conditions.

Table 8.3. Biomass accumulation in maize genotypes under two watering regimes

	SB g m^{-2}		RB g m^{-2}		R:S	
Genotype	DT	WW	DT	WW	DT	WW
Kep Select	594^{abc}	1401^{a}	263^{bc}	460^{abc}	0.45^{abc}	0.33^{bc}
Nelson QPM CPSTN	603.9^{abc}	1357^{a}	427^{ab}	564^{ab}	0.74^{a}	0.42^{ab}
Mac Medium Pearl	805^{a}	1123^{ab}	230^{bcd}	260^{bc}	0.30^{bc}	0.23^{bc}
N3XCML444	621^{abc}	1165^{ab}	275^{bc}	445^{abc}	0.44^{abc}	0.38^{abc}
PAN4P-228	708^{ab}	1077^{ab}	389^{ab}	507^{abc}	0.75^{a}	0.36^{bc}
ZM1423	497^{abc}	1094^{ab}	315^{abc}	684^{a}	0.58^{ab}	0.61^{a}
Obatanpa	357^{bc}	1031^{ab}	32^{d}	280^{bc}	0.09^{c}	0.27^{bc}
Shesha CPSTN	231^{c}	1060^{ab}	99^{cd}	393^{abc}	0.38^{abc}	0.37^{bc}
1421/DT-STR	456^{abc}	759^{b}	176^{bc}	280^{c}	0.58^{ab}	0.24^{bc}
R201	376^{bc}	759^{b}	134^{cd}	164^{c}	0.39^{abc}	0.18^{bc}
Mean	525	1083	259	379	0.47	0.34
LSD (Genotypes) (5%)	271.7	271.7	210.41	210.41	0.23	0.23
LSD (WR) (5%)	384.2	384.2	78.28	78.28	0.105	0.105
CV%	22.09	22.09	44.42	44.42	38.83	38.83

SB=shoot biomass, RB=root biomass, RS=root to shoot ratio, DT=drought stressed conditions, WW=well-watered conditions, LSD=least significant difference at 0.05, WR=water regime, CV=coefficient of variation. Different letters in a column show significant differences

8.3.1.4. ***Sorghum shoot biomass***

Similarly, genotypic and water regime effects on shoot biomass accumulation were significant among sorghum genotypes ($p<0.05$) (Table 8.4). The mean shoot biomass was 1546 g m^{-2} under drought stress whereas it was 2253 g m^{-2} under well-watered conditions (Table 8.5). Genotype AS15 (as well as NW5430 under WW only) exhibited the highest shoot biomass under both water regimes. AS16 recorded the lowest SB of 1120 g m^{-2} under drought stress while AS18 had the least SB of 1360 g m^{-2} under non-stress conditions. Biomass production was also significantly affected by water availability in sorghum (Table 8.5), with a 31% reduction in SB due to drought stress.

8.3.1.5. Sorghum root biomass

Genotypic differences and water regime main effects had significant impact on root biomass accumulation ($p<0.05$) in sorghum genotypes (Table 8.4). The overall mean RB were 1102 and 1403 g m^{-2} under drought-stressed and well-watered conditions respectively (Table 8.5). Genotype AS15 produced the highest root biomass under both drought-stressed (1572 g m^{-2}) and well-watered (1868 g m^{-2}) conditions. On the other hand, LP4303 produced the lowest root biomass of 130 g m^{-2} under drought stressed conditions while NW5393 had the lowest RB of 988 g m^{-2} under non-stressed conditions. On average, drought stress significantly reduced root biomass by 21% compared to non-stressed condition.

Table 8.4. Mean squares after analysis of variance for phenotypic traits of 10 sorghum genotypes

Source of variation	d.f.	SB	RB	RS	RC	SC	C:N
Replicates	1	553567	231040	0.0009	1.5	7.035	3.81
Genotypes (G)	9	1039633***	357943**	0.045**	16.98	1.872	17.48
Water regime (WR)	1	5002450***	904806**	0.077*	2.18	0.059	8.03
G*WR	9	177454	22396	0.014	7.81	2.191	34.31
Residual	17	136086	69886	0.009	26.17	4.345	18.94

SB=shoot biomass, RB=root biomass, RS=root to shoot ratio, RC=root carbon content, SC=shoot carbon content, C:N=carbon to nitrogen ratio

8.3.1.6. Sorghum root to shoot ratio

For sorghum, both water regime and variety had significant impact on R: S ratio (Table 8.4). The mean R: S ratio under drought stress was 0.73 compared to 0.64 under non-stressed conditions (Table 8.5). Genotypes AS16 and NW5430 consistently exhibited the highest and lowest R: S ratio, respectively, under both water regimes. Mamolokwane also had one of the lowest R: S ratio (0.47) under well-watered conditions. While drought stress increased R: S ratio by 14%.

Table 8.5. Biomass accumulation in sorghum genotypes under two watering regimes

	SB g m^{-2}		RB g m^{-2}		R: S	
Genotype	DT	WW	DT	WW	DT	WW
05-POTCH-138	1928^{ab}	2052^{bc}	1168^{cd}	1464^{abc}	0.61^{d}	0.71^{ab}
AS15	2356^{a}	3268^{a}	1572^{a}	1868^{ab}	0.67^{cd}	0.58^{ab}
AS16	1120^{c}	1768^{bc}	1088de	1388^{abc}	0.97^{a}	0.79^{a}
AS18	1196^{c}	1360^{c}	808^{g}	1036^{bc}	0.68^{cd}	0.76^{a}
AS8	1928^{ab}	2468^{abc}	1372^{b}	1692^{abc}	0.71^{cd}	0.69^{ab}
KZ5246	1264^{c}	1560^{c}	988^{ef}	1128^{abc}	0.79^{bc}	0.72^{ab}
LP4303	1411^{c}	2800^{ab}	130^{8bc}	1960^{a}	0.93^{ab}	0.67^{ab}
Mamolokwane	1180^{c}	2212^{abc}	724^{g}	1032^{bc}	0.62^{d}	0.47^{b}
NW5393	1168^{c}	1740^{bc}	864^{fg}	988^{c}	0.74^{cd}	0.58^{ab}
NW5430	1908^{b}	3304^{a}	1132de	1476^{abc}	0.60^{d}	0.47^{b}
Mean	1546	2253	1102	1403	0.73	0.64
LSD (Genotypes) (5%)	566	566	393	393	0.14	0.14
LSD (WR) (5%)	246	246	176	176	0.06	0.06
CV%	20.07	20.07	21.13	21.13	20.65	20.65

SB=shoot biomass, RB=root biomass, RS=root to shoot ratio, DT=drought stressed conditions, WW=well-watered conditions, LSD=least significant difference at 0.05, WR=water regime, CV=coefficient of variation. Different letters in a column show significant differences

8.3.1.7. Wheat shoot biomass

There was significant genotypic variation for shoot biomass production in wheat ($p<0.05$) as exhibited by significant mean squares for SB (Table 8.6). Average SB production in wheat was 783 and 959 g m^{-2} under drought-stressed and well-watered conditions, respectively (Table 8.7). Genotype BW140 exhibited the highest shoot biomass of 1196 g m^{-2} while LM48 had the lowest of 783 g m^{-2} under well-watered conditions. Similarly, LM48 had the lowest SB of 638 g m^{-2} under drought stressed conditions. On average, drought stress reduced shoot biomass production in wheat by 18% relative to well-watered condition. BW141 attained the highest SB (994 g m^{-2}) under drought stress, suffering only 10% reduction compared to non-stress conditions.

8.3.1.8. *Wheat root biomass*

Wheat exhibited significant differences among the genotypes and watering regimes for root biomass production (Table 8.6). The range for root biomass was between 352 g m^{-2} in LM47 to 500 g m^{-2} in BW140 under drought stress (Table 8.7). The range changed to between 368 (in LM71) to 615 g m^{-2} (in BW140) under well-watered conditions. Under non-stressed conditions the mean RB was 497 g m^{-2}, which was 11% higher than the mean RB attained under drought stress. BW140 produced among the highest root biomass under both conditions while LM47 and LM71 exhibited the least RB under drought stressed and well-watered conditions, respectively.

Table 8.6. Mean squares after analysis of variance for phenotypic traits of 10 wheat genotypes

Source of variation	DF	SB	RB	RS	RC	SC	C:N
Replicates	1	89	207	0.000015	24.72	7.035	3.81
Block	3	36940***	17413**	0.030*	16.55	3.426	9.47
Genotypes (G)	9	45248***	16788***	0.030**	8.08	1.872	17.48
Water regime (WR)	1	311734***	26228**	0.034*	58.89	0.059	8.03
G*WR	9	3049	3275	0.005	11.79	2.191	34.31
Residual	16	1825	3017	0.006	22.37	4.345	18.94

SB=shoot biomass, RB=root biomass, RS=root to shoot ratio, RC=root carbon content, SC=shoot carbon content, C:N=carbon to nitrogen ratio

8.3.1.9. *Wheat root to shoot ratio*

Biomass allocation between roots and shoots (R: S) was significantly different between stressed and non-stressed conditions (Table 8.7). Under well-watered condition, R: S ranged between 0.39 and 0.75; compared to 0.40 to 0.79 under drought stressed conditions. Drought stress increased mean R: S ratios across genotypes by 11% compared to non-stressed condition. Genotype LM26 attained the highest R: S in both watering regimes.

Table 8.7. Biomass accumulation in wheat genotypes under two watering regimes

	SB g m^{-2}		RB g m^{-2}		R:S	
Genotype	DT	WW	DT	WW	DT	WW
BW140	936^{a}	1196^{a}	500^{ab}	615^{a}	0.53^{cd}	0.52^{ab}
BW141	994^{a}	1108^{a}	398^{cd}	525^{abc}	0.40^{e}	0.47^{ab}
BW152	852^{b}	1100^{a}	542^{cd}	409^{abc}	0.48^{de}	0.49^{ab}
BW162	791^{bc}	874^{bc}	491^{ab}	564^{ab}	0.62^{bc}	0.64^{ab}
LM26	657^{e}	861^{bc}	518^{a}	646^{a}	0.79^{a}	0.75^{a}
LM47	706de	902^{bc}	352^{d}	416^{c}	0.59^{bc}	0.39^{b}
LM48	638^{e}	783^{c}	368^{cd}	405^{bc}	0.64^{b}	0.48^{ab}
LM70	757^{cd}	901^{bc}	490^{bcd}	453^{abc}	0.60^{bc}	0.54^{ab}
LM71	747^{cd}	942^{b}	406^{cd}	368^{c}	0.54^{cd}	0.39^{b}
LM75	748^{cd}	924^{bc}	463^{abc}	502^{abc}	0.62^{bc}	0.54^{ab}
Mean	782.7	959.3	445.9	497.1	0.58	0.52
LSD (Genotypes) (5%)	60.98	60.98	79.5	79.5	0.11	0.11
LSD (WR) (5%)	28.64	28.64	36.82	36.82	0.05	0.05
CV%	4.7	4.7	11.01	11.01	13.22	13.22

SB=shoot biomass, RB=root biomass, RS=root to shoot ratio, DT=drought stressed conditions, WW=well-watered conditions, LSD=least significant difference at 0.05, WR=water regime, CV=coefficient of variation. Different letters in a column show significant differences

Analysis of variance revealed that carbon content was not different within each crop species and was not affected by water regime in maize (Table 8.8), sorghum (Table 8.9) and wheat (Table 8.10). Variation in C content among the maize genotypes was narrow and insignificant even between water regimes (Table 8.8). Root C content was lower compared to shoot C and decreased under drought stress conditions for all genotypes. Kep Select accumulated the least root C concentration under both water regimes.

Table 8.8. Carbon content in maize under different water regimes

	Shoot C %		Root C %	
Genotype	WW	DT	WW	DT
1421/DT-STR	41.4	41.7	41.2	39.1
Kep Select	42.0	42.4	37.1	34.2
Mac Medium Pearl	42.0	41.8	39.3	36.7
N3XCML444	42.3	42.1	38.3	38.0
Nelson QPM CPSTN	43.2	41.5	40.0	37.2
Obatanpa	42.4	42.6	39.7	34.2
PAN4P-228	42.3	41.3	47.4	37.4
R201	41.9	41.5	41.4	35.6
Shesha CPSTN	42.9	41.6	39.7	38.0
ZM1423	41.5	41.6	39.2	37.5
Mean	42.2	41.8	40.3	36.8
Lsd (0.05)	1.35	1.12	2.62	1.63
se	0.93	0.87	1.21	0.81

C=carbon, WW=well-watered conditions and DT=drought stress conditions, lsd=least significant difference at 5%, se=standard error of mean

Sorghum had similar C trends as maize in that it also showed a narrow range in shoot C content (Table 8.9). AS18 had the highest shoot C at 45.0% under well-watered condition while KZ5346 had the highest shoot C (44.3%) content under drought conditions. However, the mean root C content in sorghum under drought was significantly lower compared to well-watered conditions. The highest root C content was 39.6% recorded in genotype AS15 under well-watered conditions compared to 38.7% in AS8 under drought conditions.

Table 8.9. Carbon content in sorghum under different water regimes

	Shoot C %		Root C %	
Genotype	WW	DT	WW	DT
05-POTCH-138	42.0	40.6	35.8	35.6
AS15	41.6	41.6	39.6	34.9
AS16	41.3	41.6	36.3	35.1
AS18	45.0	41.7	32.5	35.9
AS8	40.8	42.3	36.9	38.7
KZ5246	42.2	44.3	33.8	35.7
LP4303	41.2	41.3	36.1	35.3
Mamolokwane	42.2	42.2	34.3	33.5
NW5393	42.2	42.1	32.5	28.4
NW5430	42.3	42.3	36.8	36.9
Mean	42.1	42.0	35.4	35.0
Lsd (0.05)	1.64	1.41	2.10	1.83
se	1.03	0.91	1.02	1.10

C=carbon, WW=well-watered conditions and DT=drought stress conditions, lsd=least significant difference at 5%, se=standard error of mean

The mean shoot C content was lowest in wheat compared to the other cereals (Table 8.10). There was wide variation in shoot C content within the wheat genotypes ranging between 29% in LM70 to 34.3% in BW152 under well-watered conditions. Under drought stress, shoot C ranged from 27.2% in LM48 to 34.4% in LM26. The carbon concentration in wheat roots varied between 30.1% and 38.2% of dry matter with a mean at 33% under well-watered conditions (Table 9.10). The range was slightly wider under drought stress conditions from 26.9% to 36.2% with an average of 32%. LM75 and BW140 had the highest root C content under well-watered and drought conditions, respectively.

Table 8.10. Carbon content in wheat genotypes under different water regimes

	Shoot C %		Root C %	
Genotype	WW	DT	WW	DT
BW140	33.1	30.3	32.6	36.2
BW141	32.3	33.3	34.3	26.9
BW152	34.3	33.3	31.8	30.9
BW162	30.9	28.9	34.6	34.9
LM26	30.7	34.4	32.5	33.5
LM47	30.8	32.0	33.0	28.5
LM48	32.3	27.2	30.1	34.4
LM70	29.0	32.3	31.1	29.7
LM71	29.8	34.0	32.2	32.6
LM75	33.5	32.7	38.2	32.4
Mean	31.7	31.8	33.0	32.0
lsd	1.65	1.24	2.11	3.79
se	0.78	0.68	0.71	0.86

C=carbon, WW=well-watered conditions and DT=drought stress conditions, lsd=least significant difference at 5%, se=standard error of mean

8.3.2. C stocks in maize, sorghum and wheat

Sorghum accumulated the highest C stocks in roots and shoots, while maize and wheat showed no significant differences (Fig 8.1). Sorghum accumulated on average 950 g C m^{-2} of shoot C stocks compared to 450 and 375 g C m^{-2} in maize and wheat, respectively under well-watered conditions. Similarly, sorghum accumulated more shoot C stocks compared to maize and wheat under drought stress, although the mean was significantly lower than under well-watered conditions. A similar trend was observed in the root C stocks under Well-watered regimes. The C stocks were concomitant with biomass production since the intra-and inter specific C concentration did not vary significantly.

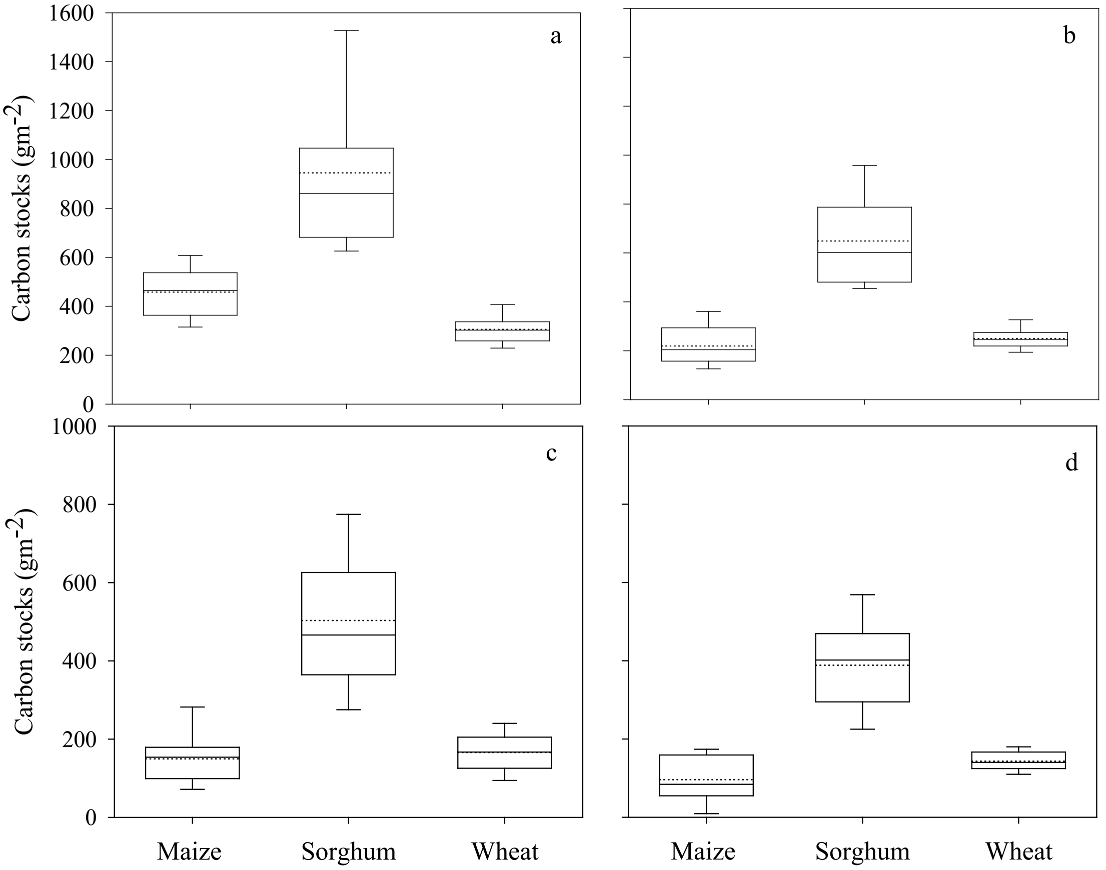


Figure 8.1. Variation in shoot C stocks under (a) well-watered and (b) drought stressed conditions and root C stocks under (c) well-watered conditions and (d) drought stressed conditions in the different crops. Dotted lines represent means of variables

8.3.3. Potential C sequestration

Based on its lower root C: N ratio of less than 30 under both water regimes, sorghum root biomass is less recalcitrant and would decompose more readily in soil compared to maize or wheat (Fig. 8.2). Maize exhibited a wider variation in root C: N suggesting that the genotypes accumulated different amounts of C and N in the roots; while a narrow variation in the shoot C: N shows that the differences in C and N accumulation in the shoots were not different. The C: N ratio in wheat straw suggests that its recalcitrance is similar to that of maize and significantly higher than that of sorghum (Fig. 8.2).

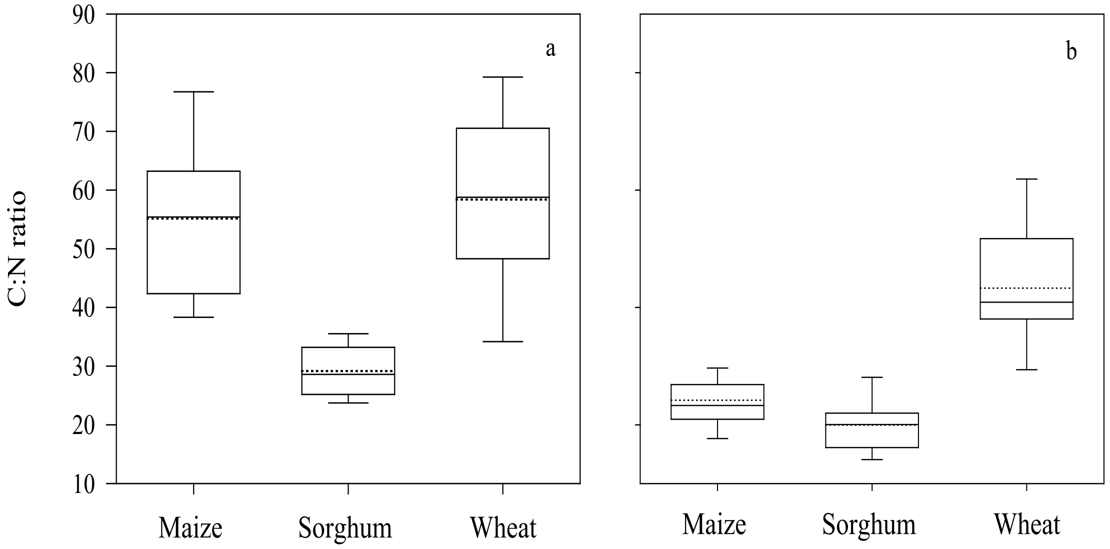


Figure 8.2. The C: N ratio of (a) root and (b) shoot biomass of three cereals

8.4. Discussion

8.4.1. Genotype variation in biomass and C content

The glasshouse study showed that biomass production was significantly different within and between cereal crops, in agreement with other findings by other researchers (Shewry et al. (2007); Amanullah and Stewart (2013). Under well-watered conditions, maize and sorghum accumulated higher shoot biomass compared to wheat, possibly due to higher photosynthetic capacity rendered by the C4 photosynthesis system. Maize and sorghum maximize C assimilation and light capture compared to wheat (Amanullah and Stewart, 2013) when other inputs are not limiting. The high biomass accumulation (both shoot and root) in sorghum under drought stress conditions compared to the other crops was expected due to the inherent drought tolerance capability of sorghum rendered by an extensive root system. The low performance of maize under drought stress compared to that of wheat could have been exacerbated by low temperatures which were more favourable during wheat growth. Within crop variability resulted from genotypic differences. For maize, Mac Medium Pearl suffered the least reduction in SB due to drought stress showing that it is relatively the most drought tolerant maize variety in terms of shoot production. For sorghum, 05-POTCH-138 incurred the least shoot biomass loss of 6% due to drought stress pointing to a higher degree of drought tolerance. This has implications on C assimilation from the atmosphere as these genotypes would suffer the least reduction in the potential to extract C from the atmosphere. In general, relative C extraction from the atmosphere would be highest in sorghum followed by maize and lastly wheat based on their shoot biomass production capabilities.

The higher root biomass in sorghum compared to the other crops is because sorghum possesses a deeper and more profuse root system (Salih et al., 1999; Machado et al., 2001). The lower root biomass recorded in wheat is concordant with its shallow rooting capacity typical of most C3 plants compared to C4 plants such as sorghum and maize. However, the low root biomass found in maize compared to wheat was unexpected but probably resulted from the effect of low temperature during the growing season which favoured wheat production. The impact of drought stress on all crops resulted in a reduction of root biomass, implying that drought would reduce potential for C sequestration. However, there are genotypes that showed a tendency to maintain rooting capacity even under drought stress. Sorghum genotype AS15 exhibited relative stability by losing only 15% of its rooting capacity due to drought stress. In maize, Mac Medium Pearl, which is an open pollinated variety, suffered the least reduction in rooting capacity of 11% due to drought stress, while Nelson QPM CPSTN, a commercial quality protein maize variety, had the highest RB under drought stress conditions and incurred only a 24% reduction. In wheat, BW140 which had the highest root biomass under both water regimes suffered an 18% reduction in root biomass due to drought stress compared to only 7% reduction in genotype LM70. Due to its high root biomass, which is strongly linked to SOC sequestration (Paustian et al., 2016), sorghum can sequester more atmospheric C to the soil compared to maize and wheat. The sorghum genotype AS15 would be ideal to sequester the highest amount of C.

Maize had the lowest R: S ratio among the cereals because the relative growth rate of its above ground biomass is faster than the other cereals. Sorghum attained the highest R: S ratio among all the cereals due to its inherent profuse root system that creates a substantial sink for biomass relative to shoot biomass (Machado et al., 2001). The R: S ratio of wheat and maize would be expected to be lower than sorghum due to their relatively inferior root system. PAN4P-228 would allocate more biomass to root due to drought, a response which is beneficial for C sequestration in maize. Sorghum genotype AS15 had relatively higher biomass compared to AS16 which had the highest R: S under drought stress. In the same regard, LM26 of wheat had the highest allocation of biomass to roots due to drought which makes it ideal for drought tolerance and C sequestration.

The concentration of C in a particular plant part is assumed to be driven by the C requirements of that specific organ (Kundu et al., 2007). Plants primarily allocate more carbon for development in the above ground structures compared to roots. The differences in shoot C content among the crops could be partly due to variations in efficiency of internal C metabolism. Sorghum and maize had higher plant C concentration compared to wheat, which could be related to high C demand for heat stress tolerance (Rattey et al., 2009). For maize, its high concentration of lignin and hemicellulose result in the retention of more C in the stover (shoots) than in the straw of wheat (McKendry, 2002; Adapa et al., 2009; Li et al., 2013). However, there was no definite trend in the data, pointing to a limited impact of plant type or water regime on the overall plant C concentration. Genotype effects on C concentration were not significantly possibly showing that C concentration is predetermined and may not be easily altered through selection under different water regimes. Other researchers have argued that the environment rather than genetic factors influence C allocation more. Poorter et al. (2011) reported significant differences in C concentration between species but non-significant differences within species indicating that C content is determined at species level rather than genotype.

8.4.2. Carbon sequestration potential of maize, sorghum and wheat

The general decrease of total plant carbon stocks from sorghum, to maize and to wheat was concomitant with decrease in biomass. Sorghum and maize were shown to accumulate higher biomass, which increased their capacity to sequester more C while wheat showed limited C stocks as a direct result of low biomass production. C sequestration was a direct consequence of biomass production because the range of C concentration was narrow and insignificant within each crop species. However, sorghum had a superior per unit C sequestration potential compared to the other two cereals. For every unit of C accumulated in the shoots, sorghum transferred 35% to the roots increasing the potential for C transfer to the soil. In comparison, wheat transferred 33% and maize 25% C to root. Given the lower C: N ratio, sorghum straw is more likely to be mineralized in the soil and provide a substrate for microbial respiration. The decomposability of the residues imply that they will have a short mean residence time in the soil, which reduces the C sequestration potential of the crop. Wheat and maize have higher C: N ratios in roots and shoots suggesting that their biomass would have longer residence time in the soil. However, their potential will be restricted by low biomass production compared to sorghum.

8.4.3. Impact of water regime on C sequestration and biomass

The variations in biomass production under different water regimes confirms previous studies on the effects of environment on biomass production (Anderson-Teixeira et al., 2013). Maize suffered the highest reduction in biomass production due to water stress compared to sorghum and wheat. The difference between sorghum and maize was because sorghum is relatively more tolerant to water stress than maize. Drought tolerance in sorghum is rendered by extensive rooting and reduced surface area in above ground parts (compared to maize) for transpirational losses (Salih et al., 1999). Wheat is grown in winter where temperatures are low, and as a result suffered lower biomass losses due to drought. The impact of drought stress on sorghum and maize is exacerbated by higher temperatures during their summer growth period. All crop species accumulated more biomass in shoots compared to roots, leading to an R: S ratio of below 1, showing that plants prioritize shoots over roots especially under sufficient water supply (Poorter and Nagel, 2000). However, the prioritization changes under limited moisture, where R: S ratio increase. This shows that plants re-arrange biomass allocation to balance between survival and productivity under moisture stress (Ludewig and Flügge, 2013).

Although the optimal partitioning theory stresses that drought stress induces and increases biomass and C allocation to roots in all the crops (Poorter et al., 2011), this study found otherwise. The reduction in biomass production is due to water limitation. The low R: S and C accumulation under drought can be attributed to reduced ability to acquire C resources from the atmosphere and increased hydraulic stress as the soil dries out. These factors cause a significant reduction in biomass and C accumulation resulting in low availability of assimilates to translocate below ground, and the trade off in biomass production would outweigh the benefits of increased R: S ratio and root C content. The decrease in the C sequestration potential of maize would be significantly higher under drought stress compared to sorghum due to low biomass production. In comparison, wheat would also experience huge decrease in C sequestration potential under drought stress due to a combination of poor rooting system and reduction in total biomass.

8.5. Conclusion

The study highlighted C allocation within plants parts in maize, sorghum and wheat, which can be used as a basis for identifying crops and genotypes with high C sequestration potential. Sorghum showed the highest potential for C sequestration based on high root biomass productivity. Among sorghum, genotype AS15 exhibited the highest potential for rooting biomass and C stocks. For maize, Nel and Obanapta would be superior in shoot C sequestration under well-watered and drought stressed conditions respectively. BW140 is the most suitable genotype of wheat to promote C sequestration in water limited environments. The challenge for promoting wheat as a candidate for C sequestration would be water deficit in communal farming systems where it is envisaged that it would suffer huge losses in C sequestration potential due to lack of irrigation facilities. Maize is widely grown among communal farmers and can be grown concurrently with sorghum particularly in water limited environments, resulting in an increase in C sequestration potential in their agro-systems while still maintaining food security.

Chapter 9 CARBON SUPERIOR WHEAT CULTIVARS FOR ENHANCED SOIL CARBON SEQUESTRATION

Abstract

The sequestration of atmospheric carbon (C) by plants and ultimately by soils is a possible strategy for climate change mitigation, which is imperative for restoring land productivity. While some studies have compared the ability of existing crops to store C and allocate it to the soil, the genetic variations between crop genotypes have received less attention. The objective of this study was to compare the sequestration of atmospheric C by genetically diverse wheat genotypes under different scenarios of soil water availability. The experiments were set up under open field and greenhouse conditions with 100 wheat genotypes sourced from the International Maize and Wheat Improvement Centre and grown at 25% (drought stressed) and 75% (non-stressed) field capacity using an alpha lattice design with 10 incomplete blocks and 10 genotypes per block, replicated twice. The following data were collected and analyzed: grain yield (GY), plant shoot and root biomass (SB and RB in that order) and C content and stocks in plant parts. Also, ^{13}C pulse labelling was performed during the crop growth period under greenhouse conditions to determine the genotypic differences in C transfer from the atmosphere to the soil. The average GY varied from 75 to 4696 g m^{-2} and total plant biomass (PB) between 1967 to 13528 g m^{-2}. The plant C stocks ranged from 592 to 1109 g C m^{-2} (i.e. an 87% difference) under drought condition and between 1324 and 2881 g C m^{-2} (i.e. 117%) under well-watered condition. There was a trend for the atmospheric C transfer to the soils to increase with the increase in the root to shoot allocation of biomass and C, with the highest values found for the genotypes BW140 and BW162 and LM47. LM47 not only exhibited the highest C flux to the soil (39% higher than the worst performing genotype) but also ranked second in terms of grain yield; therefore, making it an ideal candidate for attaining the twin objectives of enhanced food security and C sequestration into soils. Future research studies need to identify genomic regions associated with grain yield and soil C sequestration to enable breeding of "carbon-superior" cultivars.

Keywords: agronomic traits, cultivars, cereals, water stress, global change

9.1. Introduction

Climate change is mainly driven by human-induced release of large quantities of carbon (C), as greenhouse gases (GHGs), to the atmosphere. The C was initially sequestered in soils and rocks by plants through photosynthesis and other organisms. Evidence points to agriculture as a major source of GHGs starting several thousand years ago through conversion of natural ecosystems (e.g. forests and grasslands) into croplands. Following the conversion, most of the C from the biomass and soil pools, the latter consisting of plant and animal residues at various stages of decomposition, is emitted to the atmosphere through oxidation by microbes. A total amount of 133 Pg of soil C is estimated to have been lost to the atmosphere (Sanderman et al., 2018).

The subsequent rise in atmospheric CO_2 concentrations has caused an increase in global temperatures, which has significantly affected precipitation patterns, leading to intermittent warmer and drier environments. This, in turn, has negative affected agriculture. For instance, rising surface temperatures curtails crop production because crops are adapted to specific optimal temperature ranges and deviations from the optimal range negatively affects biomass production, grain yield and quality (Pandey et al., 2018). Specifically, higher temperatures exacerbate drought stress by increasing evaporation and reducing crop water use efficiency (Sinclair, 2018). In addition, higher atmospheric CO_2 concentrations reduce the nutritional quality of grains, thus impacting the dietary requirements of billions of people across the world (Pandey et al., 2018).

Restoring the lost carbon to soils is increasingly considered a potential climate change mitigation strategy and a necessary step for adapting to future climate situations. For instance, higher soil carbon contents enhance soil quality and its water holding capacity. This option looks credible (Minasny et al, 2017) because soil C stocks have largely depleted over time. In addition, there are large C exchanges between the atmosphere, plants and soils (Raich and Potter, 1995). The C flux between these pools is about 60 Gt C per year, which is about one fifth of the atmospheric C pool. Therefore, even a relatively small increase in soil C stocks could thus be a *game changer* for climate change. Minasy et al. (2017) estimated that an increase of soil C stocks by 0.4% per year is possibly achieved in several countries for partially mitigating all human-induced CO_2 emissions from fossil carbon (estimated at 8.9 giga tonnes C). Such an increase in soil C is not only expected to mitigate climate change, but to also provide additional benefits as soil organic matter, of which carbon is a main component, and accrual of numerous positive effects on soil health (quality, fertility, water holding capacity and biodiversity).

Several propositions to mitigate climate change, such as agroforestry and changes in farming practices, e.g. from conventional to reduced tillage systems, have been made but they are shrouded in controversy and not easy to adopt (Paustian et al., 2017). The low adoption rates could be partly because farmers, in their large majority, do not necessarily want to change their production methods. Therefore, pragmatic solutions for transferring and storing the excess atmospheric C into soils that fit well into farmers' already established production systems are still needed. One such solutions could be identification of genetic resources that are useful to farmers, while at the same time enhancing the amount of soil C. In particular, special attention on the ability of food and cash crops to allocate C in their rooting system is needed. Studies reported that 50-80% of soil C comes from root activity during growth and decomposition after senescence (Rasse et al., 2005; Yang et al., 2012). Moreover, a larger root system, as expressed by an elevated root to shoot (R:S) ratio, is not only expected to increase root C exchanges with the soil but also to increase the water extraction capacity of plants (Lynch and Wojciechowski, 2015), which is a key defense trait to counter potential climate-change driven water scarcity.

Mathew et al. (2017), in their analysis of different crop types from 389 trials worldwide, pointed that greatest C allocation to roots was in grasses (R:S = 1.19), followed by cereals (0.95), legumes (0.86), oil crops (0.85), and fibre crops (0.50). Several studies have shown that R:S for annual cereals vary greatly. For example, Amanullah and Stewart (2013) reported values of 0.41 and 0.29 for sorghum and maize, respectively. Yang et al. (2010) reported a mean of 0.25 for maize and wheat. Variations of R:S within single species have also been reported. Fang et al. (2017) reported a ratio of 1.13 for landrace wheat compared to 0.61 and 0.81 for two modern cultivars. Similarly, Siddique et al. (1990) reported ratios ranging from 0.72 to 0.84 for modern wheat cultivars.

Despite several studies on plant C stocks, little is known about the potential ability of numerous genotypes of individual crops to transfer C to soils. The objective of the current study was to evaluate and compare the capacity of selected wheat genotypes to sequester atmospheric C. The wheat genotypes were bred for sub-Saharan Africa, one of the regions in the world most affected by climate change, food insecurity and land degradation. Selection of genotypes with high C storage ability constitutes a pragmatic solution for climate change mitigation because farmers would provide positive impact on food security and reduce land degradation without any need to change their practices. Here we considered 100 wheat genotypes from the

International Maize and Wheat Improvement Centre (CIMMYT), grown in the field and in glasshouses and subjected to water stressed and non-stressed conditions. The results could be helpful to crop breeders in evaluating diversity in biomass allocation and agronomic performance, which is important for developing varieties with higher water use efficiency, drought tolerance, grain yield and potential for C sequestration into soils. The results would also be beneficial to farmers who seek solutions for storing more C into soils without changing their current practices.

9.2. Materials and methods

9.2.1. Plant material and trial management

One hundred genotypes from the International Maize and Wheat Improvement Centre (CIMMYT) were evaluated in this trial. Two experiments were carried out at the University of KwaZulu-Natal (UKZN), Pietermaritzburg (South Africa); one from October 2016 to February 2017 in a glasshouse, and another one was carried out from May to September 2017 in the open field. The glasshouse experiment was a 10 × 10 alpha lattice design with two replications and with ten pots allocated per incomplete block. The genotypes were randomly assigned to the pots to minimize experimental errors associated with water discharge from the drip irrigation. Each pot received ten seeds and the plants were thinned to eight individuals three weeks after emergence. Fertilizer was applied through automated drip irrigation at a rate of 300 kg N ha^{-1} and 200 kg P_2O_5 ha^{-1}. Two water regimes were applied 6 weeks after sowing. Plants in the ‘well-watered’ treatment were irrigated to field capacity (FC) whenever average soil water content fell to 80% of the FC. Plants in the ‘water stress’ treatment were irrigated to FC after soil water content had dropped to 30% of the FC. The two watering treatments were maintained until crop maturity. The pots were weighed daily to estimate soil water content. The field experiment was carried out at the UKZN Research Farm (latitude 29.667°, longitude 30.406°). The experiment layout was also a 10 × 10 alpha lattice design with two replications. The field was first ploughed to a depth of 0.30 m and a custom-made plastic mulch installed on the drought stressed area to exclude rainwater. Seeds were sown along rows spaced 0.3 m apart with three seeds every 0.1 m. Each row was treated as an incomplete block consisting of 10 genotypes. Fertilization was applied at rates of 120 kg N ha^{-1}, 30 kg P ha^{-1} and 30 kg K ha^{-1}. Other agronomic practices followed the standard practices in South Africa (DAFF, 2010). In the well-watered treatment, FC was maintained using a drip irrigation system controlled by soil water probes. Under the drought stress treatment, irrigation was withheld to 30% field capacity or just before signs of permanent wilting.

9.2.2. Data collection and laboratory analysis

9.2.2.1. Agronomic traits

Several agronomic traits were measured. These included (i) number of days from sowing to heading (DTH) when 50% of the plants had fully-emerged spikes, (ii) number of days to maturity (DTM) when 50% of the plants were dry, (iii) number of productive tillers (NPT), (iv) dry grain (GY), (v) shoot and root biomass (SB, RB) at harvest after oven drying at 60°C for 72 hours and the resulting root to shoot (R:S) ratio for biomass, and (vi) thousand kernel weight (TKW). These variables were used to select a sub-set of 10 genotypes for further investigations on C. The 100 genotypes were grouped into 10 sub-groups using hierarchical clustering based on phenotypic data combined across water regimes and sites. A dendrogram of similarities was derived using the Unweighted Pair Group Method with Arithmetic mean algorithm (UPGMA) and, based on this, the 10 genotypes with the highest grain yield and plant biomass were selected for further C studies.

9.2.3. ^{13}C pulse labelling

Pulse labelling was performed in order to estimate the transfer of atmospheric C to the soil. The labelling was performed on the 10 genotypes selected from the initial population of 100 on the basis of biomass production and drought sensitivity index. The selected genotypes were planted on 13 January 2018 with 3 pots per genotype and per water regime (well-watered: 75% FC; water stressed: 25% FC) and laid out in a randomized complete block design in the glasshouse. The 10 genotypes were randomly assigned to the pots to minimize the experimental errors associated with water discharge from the drip irrigation. Each pot received ten seeds and plants were thinned to eight individuals to mimic the optimal plant density three weeks after emergence. Fertilizer was applied through the automated drip irrigation system at a rate of 300 kg N ha^{-1} and 200 kg P_2O_5 ha^{-1}. Drip irrigation was used to establish the crops for 4 weeks before the different water regimes and pulse labeling commenced.

The pulse labelling was performed using a closed system, which consisted of an upper chamber made of plastic sheeting. The chamber was installed in the late afternoon of the day before isotope labelling. Extra fine earth was packed firmly around the base of the chamber to reduce gas leakage. The airtight chamber was 1.2 m long, 0,6 m large and 1 m high and was built in a greenhouse where the air temperature was 25±5°C. The inner surface of the chamber was smeared with anti-fog agent to reduce water vapour condensation during labelling, thus maintaining light intensity and reducing the dissolution of $^{13}CO_2$ in water drops forming on the chamber's inner plastic sheet.

Pulse labelling was carried out once a week, starting from 4 weeks after emergence (9 February) until maturity (110days). The ^{13}C-labelling was achieved by replacing the ambient CO_2 in the chamber by $^{13}CO_2$. The ambient CO_2 from the chamber was trapped in a 1M NaOH solution in a beaker placed in the centre of the chamber. This was performed until the CO_2 concentration, monitored by an infrared gas analyser (EGM-1; Environmental Gas Monitor, PP Systems, Hitchin, UK), reached 10 ppmv. When the ambient CO_2 concentration reached 10 ppmv, $Na_2{}^{13}CO_3$ (~97 atom %) powder was added into a second beaker containing 1 M H_2SO_4 solution to generate $^{13}CO^2$ to bring the carbon dioxide concentration back to 450 ppmv the chamber carbon dioxide concentration to 450 ppmv. More $Na_2{}^{13}CO_3$ powder was added to the H_2SO_4 solution when necessary to maintain the chamber carbon dioxide concentration at about 450 ppmv. Each labelling process lasted 6 hours to ensure sufficiently high ^{13}C abundance in both shoot and root samples compared with unlabelled control samples. The air inside the chamber was circulated by a vertically mounted electric (12V, 0.21A) fan placed in the center of the chamber to ensure good distribution of the $^{13}CO_2$ within the chamber throughout the labelling session. All the chamber carbon dioxide was trapped into the NaOH solution at the end of labelling session before opening the chamber to prevent $^{13}CO_2$ assimilation by the non-labelled neighbouring plants.

9.2.4. Plant biomass and soil sampling

Plant shoots were harvested by clipping at the soil surface after crop maturity. Stems and leaves were pooled, and used for C and ^{13}C analysis. Soil and roots from the pots were separated manually, by stirring the soil-root mixture in a bucket to ensure complete disaggregation of the soil followed by sieving. All plant and soil materials were dried in an oven at 60°c for 72 hours before weighing and grinding in ceramic mortar. The ground materials were passed through 1 mm sieves. The sieved samples were stored at room temperature prior to analysis for total C and ^{13}C contents.

Grain, shoot and root samples of the 10 selected genotypes were grounded to <0.5 mm and analyzed for total C and N in triplicates using a LECO CNS-2000 Dumas dry matter combustion analyzer (LECO Corp., St. Joseph, MI). The organic C content (OC_C) was used to estimate C stocks in the different plant parts as follows:

$$\boldsymbol{GC_S = GY \times GC_C \times b} \tag{1}$$

where GC_S was the grain C stock (kg C m^{-2}), GY was grain yield and GC_C was the C concentration in the grain (g C kg^{-1}); and b was a constant equal to 0.001.

$$\boldsymbol{SC_S = SB \times SC_C \times b} \tag{2}$$

where SC_S was the shoot C stock (kg C m^{-2}), SB was the shoot biomass and SC_C was the C concentration in the shoots (g C kg^{-1}); and b was a constant equal to 0.001.

$$\boldsymbol{RC_S = SR \times RC_C \times b} \tag{3}$$

where RC_S was the root C stock (kg C m^{-2}), RB was the root biomass and RC_C was the C concentration in the roots (g C kg^{-1}); and b was a constant equal to 0.001.

The plant carbon stocks (PC_S) corresponded to the sum of the stocks from the different plant parts (e.g. GC_S+SC_S+RC_S for C stocks).

The $^{13}C/^{12}C$ ratios in soil and plant samples were analyzed using an isotope ratio mass spectrometer (Deltaplus, Finnigan MAT GmbH, Bremen, Germany) coupled with an elemental analyzer (NC 2500, ThermoQuest Italia S.p.A., Milan, Italy) by an interface (ConFlo II, Finnigan MAT GmbH, Bremen, Germany). The natural abundance of ^{13}C in the soil and plant samples was expressed as $d^{13}C$ (%) relative to Pee Dee Belemnite following Equation 1 and 2:

$$\boldsymbol{R_{sample}} = \left(\frac{\boldsymbol{\delta^{13}C}}{\boldsymbol{1000}} + \boldsymbol{1}\right) \times \boldsymbol{0.011237} \tag{4}$$

$$\boldsymbol{^{13}C}(\%) = \left(\frac{\boldsymbol{R_{sample}}}{\boldsymbol{R_{sample}+1}}\right) \times \boldsymbol{100} \tag{5}$$

where R_{sample} was the isotope ratio ($^{13}C/^{12}C$) of a sample and 0.011237 was the ratio of $^{13}C/^{12}C$ in Pee Dee Belemnite, and ^{13}C (%) represented the percent of ^{13}C atom in total carbon atoms in a given sample.

9.2.5. Water-use efficiency for grain yield and carbon storage

The water-use efficiency of productivity, typically defined as the ratio of mass produced to the rate of evapotranspiration, was calculated for the selected variables: GY, SB, RB, GC_S, SC_S, RC_S, PC_S and ^{13}C (%). Meteorological data, such as air temperature, air humidity and wind speed, and quantity of water applied through irrigation or natural rains were recorded using a standard weather station located in the vicinity of the pots. These data were recorded each day to estimate crop evapotranspiration as follows:

$$ET = \Delta W + I + P + CR - D - R \qquad (6)$$

where ET was crop evapotranspiration, ΔW was the change in soil or pot water mass between two consecutive measurements of soil water content, I was amount irrigation water applied, P was amount of rainfall, CR was capillary rise from the water table to the crop root zone, D was downward drainage from the crop root zone and R was surface runoff. The soils at the open field experiment exhibited high infiltration by water and the water table was below 20 m, hence runoff and capillary contribution from groundwater were ignored. Crop water use efficiency for biomass, C stocks and ^{13}C (%) was calculated as biomass or C stocks divided by crop total evapotranspiration.

9.2.6. Data analysis

Descriptive statistics were estimated for the selected variables. In addition, the means of the genotype variables and the different water regimes were separated by Fischer's unprotected least significant difference (LSD) at 0.05 significance level to quantify the effects of genotype, environment and water regime. In addition, Principal Component Analysis (PCA) was performed to depict the multiple relationships between plant biomass, C allocation and stocks, and C transfer to the soil. The PCA axes were generated using ^{13}C content in the different plant parts, and the other variables of plant biomass and plant C were used as display.

9.3. Results

9.3.1. Variations in grain yield and plant biomass amongst genotypes

The overall average grain yield (GY) amongst the initial set of 100 genotypes, across the open field and glasshouse trials for both water regimes, was 1387 g m^{-2} with a standard error of ± 84 g m^{-2} (Table 9.1). GY varied by a 62 factor from 75 to 4696 g m^{-2} and showed a positively skewed distribution (Skew=1.53). Total plant biomass (PB) was much less variable than GY as values ranged from 1967 to 13528 g m^{-2}, a difference of only 6.8 times, which was significant at $P<0.001$. The 100 genotypes had an average root to shoot ratio for biomass of 0.12, meaning that they allocated an average 12% of their total biomass to their roots, with values from 3 to 38% (Table 9.1). The sub-set of 10 selected genotypes showed much less average variation in GY, from 621 to 4383 g m^{-2} (a 7.0-fold increase *vs* 62 times for the 100 genotypes), but similar variations in PB with values between 2475 and 13529 g m^{2} (5.4 *vs* 6.8) (Table 9.1).

We learn from Table 9.2 that the mean GY in the glasshouse increased from 326 g m^{-2} at 25% FC to 414 g m^{-2} at 75% FC. In the open field, the increase was from 1700 g m^{-2} at 25% FC to 2062 g m^{-2} at 75% FC. A similar trend of greater values in the open field and also at 75% FC as compared to the glasshouse and 25% FC was observed for all the biomass variables studied. For instance, PB decreased in the glasshouse from 6289 g m^{-2} at 75% FC to 2992 g m^{-2} at 25% FC as did the R:S from 0.57 to 0.43 (Table 9.2).

On average, LM75 was the best grain yielding genotype, followed by LM47, LM26 and BW141 while BW152 was the least performing one (Table 9.3). Under 25% FC, LM75 ranked first (1390 g m^{-2}) and was immediately followed by LM47 (1360 g m^{-2}), while the other genotypes exhibited GY below 1100 g m^{-2} (Figure 9.1). Under 75% FC, LM75 also ranked first at 1617 g m^{-2} and LM26 was second at 1369 g m^{-2}. Five other genotypes had GY over 1100 g m^{-2}.

BW140 had the highest PB, followed by LM75 and BW162, the latter being characterized by the highest R:S ratio for biomass allocation and the highest root C stocks (Table 9.3). Figure 9.1 indicates much higher total biomass values under 75% FC than 25% FC. LM75 exhibited the lowest R:S ratio for plant biomass under drought conditions and was followed by LM47, the two more grain yielding genotypes. BW162 had the highest root biomass allocation under the two water regimes. We also learn from Table 9.3 that LM75 was the most water efficient genotype in terms of grain yield, shoot and plant biomass production, but ranked second in terms of water use efficiency for root biomass. In contrast, LM47 that ranked second for WUE in terms of GY, was the least efficient genotype for root biomass. BW162 was the most water efficient for root biomass.

Table 9.1. Summary statistics for grain yield and selected morphological variables for the 100 wheat genotypes grown in the field and in the glasshouse under different water regimes.

	GY	SB	RB	PB	R:S
	----------------------g m^2 -----------------------				
Mean	1387	2498	305	4189	0,12
Median	1309	2332	263	3930	0,11
Min	75	1179	65	1976	0,03
Max	4696	8658	1219	13529	0,38
Q1	959	1827	189	3026	0,09
Q3	1644	2908	365	4893	0,15
CV%	47	37	57	37	41
SEM	84	121	22	200	0,01
Skew	1,53	2,04	2,03	1,82	1,67
Kurt	4,35	7,15	5,76	5,66	4,39

min=minimum: max=maximum; Q1=1st quartile; Q3=3rd quartile; CV= coefficient of variation; SEM=standard mean error; skew=skewness; kurt=kurtosis, GY: grain yield, SB: shoot biomass, RB: Root Biomass, PB: Total Plant Biomass, R:S Root to Shoot

Table 9.2. Summary statistics for grain yield and selected morphological for 10 selected wheat varieties grown in the field and glasshouse and across water regimes.

	GY	SB	RB	PB	R:S	SCC	RCC	SCS	RCS	PCS
	-----------------------g m^2 -----------------------					----------%--------		--------------g m^2 ----------------		
					Glasshouse					
25% Field capacity										
Mean	326	1873	792	2992	0,43	33	31	625	239	865
Median	266	1826	762	2918	0,44	34	31	621	225	854
Min	58	1349	368	2156	0,15	25	21	409	103	592
Max	869	2415	1263	3983	0,70	37	39	805	379	1109
CV%	68	24	36	24	34	19	23	25	34	23
SEM	28	57	37	94	0,02	1	1	20	10	26
75% Field capacity										
Mean	414	3726	2149	6289	0,57	33	31	1243	647	1890
Median	397	3440	1823	5508	0,57	34	31	1186	605	1716
Min	136	2779	1290	4531	0,37	25	21	851	333	1324
Max	811	5192	3575	9305	0,78	37	39	1822	1059	2881
CV%	50	26	38	30	26	19	23	27	35	28
SEM	27	127	104	247	0,02	1	1	44	29	69
					Field					
25% Field capacity										
Mean	1700	2507	302	4508	0.12	33	31	832	95	927
Median	1401	2534	277	4042	0.12	34	31	827	80	968
Min	621	1598	152	2475	0.08	25	21	534	45	582
Max	4488	3775	642	8100	0.19	37	39	1215	214	1409
CV%	53	27	39	34	24	8	16	26	47	27

SEM	116	87	15	199	0.004	0.36	0.62	28.10	5.79	32.30
75% Field capacity										
Mean	2062	3714	403	6179	0.12	34	31	1254	121	1375
Median	1720	3154	382	5462	0.10	35	31	1052	112	1170
Min	1118	1954	135	3555	0.06	30	21	675	53	728
Max	4383	8658	1006	13529	0.26	37	39	2869	302	3022
CV%	44	42	44	39	43	6	15	42	43	39
SEM	117	202	23	313	0.01	0.26	0.61	67.57	6.69	69.93

min=minimum: max=maximum; Q1=1st quartile; Q3=3rd quartile; CV= coefficient of variation; SEM=standard mean error; skew=skewness; kurt=kurtosis, GY: grain yield, SB: shoot biomass, RB: Root Biomass, PB: Total Plant Biomass, R:S Root to Shoot

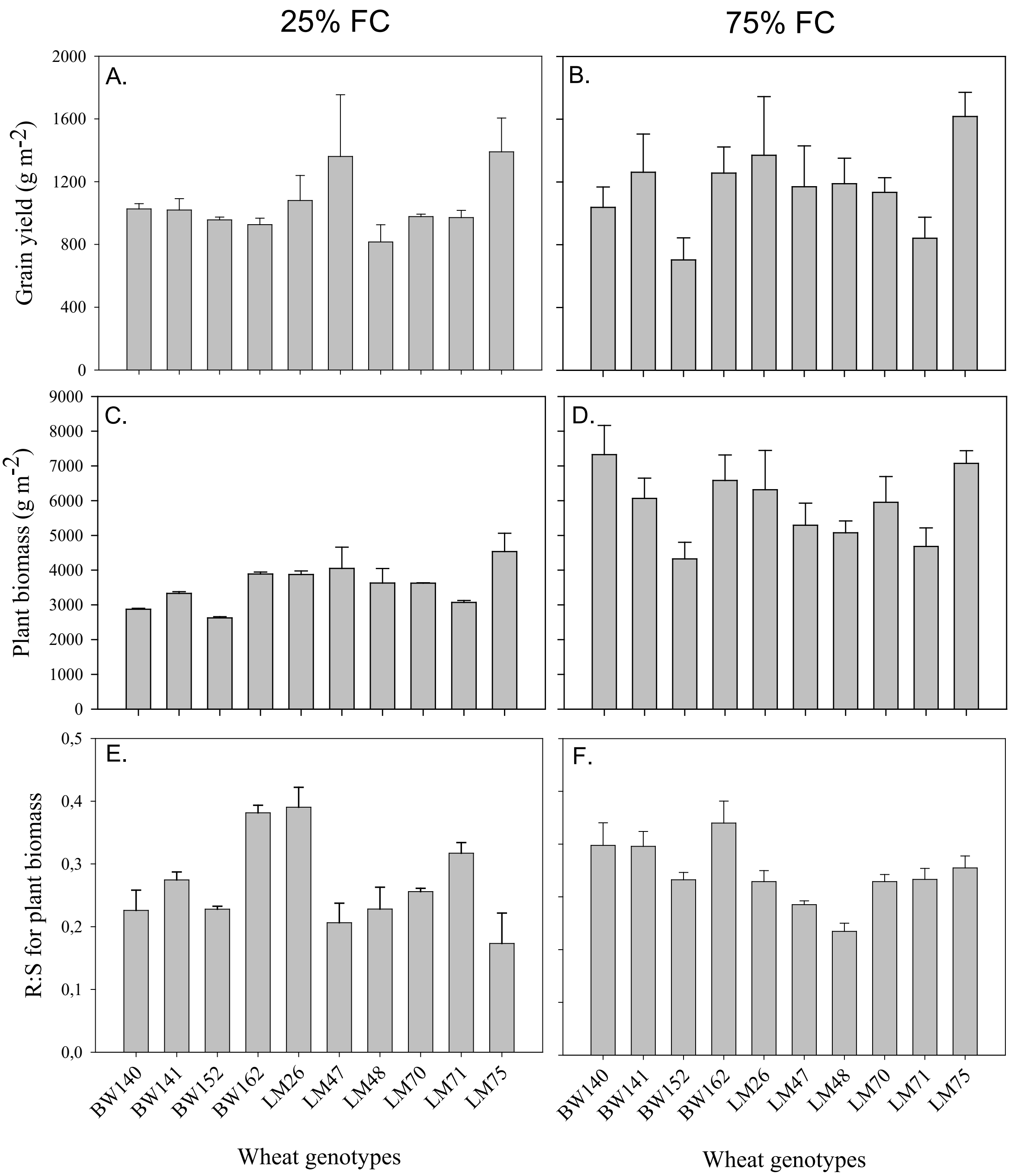


Figure 9.1. Wheat genotypes and soil water availability (25 and 75% field capacity) impacts on grain yield (A, B), plant biomass (B, C) root to shoot ratio for plant biomass (D, E).

Data are Mean and Standard Errors (n=18) from field and glasshouse experiments.

9.3.2. Variations in ability of genotypes to store C

The average C content of the selected 10 genotypes was 34±0.9% in the shoots (SC_C) and decreased to 30±1.1% in the roots (RC_C), which corresponded to a significant difference at $P<0.05$. SC_C ranged from 30 to 37%, while RC_C varied from 21 to 39%, with all the differences significant at $P<0.05$. Plant C stocks (PCs) exhibited large variations between the test genotypes, with a mean of 1174± 64 g m^{-2}. The values ranged from 582 to 3022 g m^{-2}, a 5-fold difference between the lowest and highest value. BW140, which had the highest GY, also showed the highest Root C stocks at 75% FC (Figure 9.2C), but one of the two lowest at 25% FC. Finally, LM75 root C content was one of the lowest in both water regimes, while LM71, LM47 and LM48 had the highest.

The average ^{13}C content of the shoots was 64.81±14.5% and higher than the average content in the roots (58.58±14.8%) and the soils (55.05±6.8%). The ^{13}C content in both the roots and soils was higher under well-watered than under dry conditions (55.43±15.5% vs 61.73±14.06% for roots and 52.18±0.18% vs 57.92±13.49% for soils, respectively). However, the ^{13}C content in shoots was highest under drought conditions (72.14±21.0% vs 57.47±8.49%).

In water stressed conditions, soil ^{13}C content only differed slightly among the genotypes, with values ranging between 52.16±0.16% for LM75 and 52.29±0.78% for LM48, which corresponded to a 0.27% maximum difference. The difference increased to 21% under well-watered conditions with ^{13}C ranging between 53.86±7.76% for LM71 and 65.17±5.09% for LM47. The second highest value was for LM48 (63.77±32.4%). The high ^{13}C assimilation into the soil for LM47 corresponded to one of the highest ^{13}C root assimilation rates. LM48 exhibited one of the lowest ^{13}C root enrichment. LM47, which ranked overall first for C storage within the soil, ranked second for GY and third for root C content. In contrast, LM75, the best genotype for GY under both water regimes, had one of the lowest root C content but the highest plant C stock (Figure 9.2).

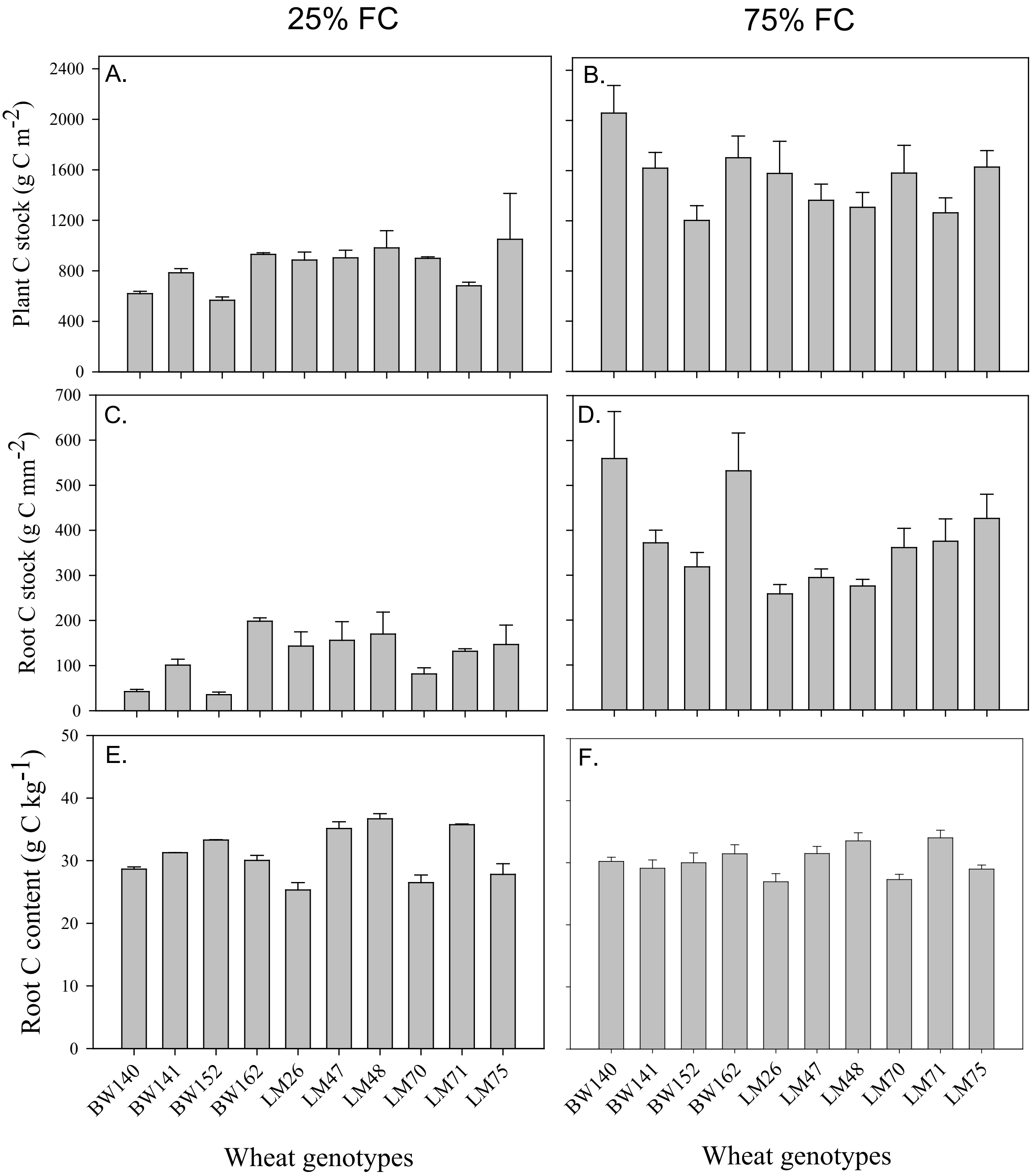


Figure 9.2. Wheat genotype and soil water availability (25 and 75% field capacity) impacts on plant carbon stocks (A, B), root C stocks (B, C) and root C content (D, E).

Data are Mean and Standard Errors (n=18) from field and glasshouse experiments.

9.4. Discussion

9.4.1. Variation of biomass allocation in the genotypes

The study genotypes were characterized by high variations in their ability to produce grain and total biomass, and to allocate C to shoots and roots, as indicated by the high coefficients of variation for these variables. This large variability agrees with reports of Akman et al. (2017), Nevo and Chen (2010) and Waines et al., (2012). Based on field observations, the authors pointed to wide genetic variability in root biomass production in wheat. In particular, the R:S values ranging between 0.15 and 0.78 were much more variable than the 1.00-1.36 range reported by Fang et al., (2017). This is probably due to the fact that some of the CIMMYT genotypes were developed for heat and drought tolerance, which influence them to allocate more biomass to their roots.

9.4.2. Genotype C storage

The present results pointed to large differences in the ability of the selected genotypes to store atmospheric C in their tissues, with total plant C stocks ranging from 582 to 3022 g m^{-2}, a 5.2-fold difference. The variation in root and shoot C stocks was also surprisingly high with for instance a 3-fold difference for root C stocks. Such a wide range in the ability of cultivated genotypes to allocate C into the roots was unexpected. It was, however, observed when comparing a wild against a cultivated genotype (Graaff et al., 2009) that the wild genotype has higher R:S than the cultivated one. Graaff et al. (2009) reported R:S ratio of 0.6 for the wild genotype as compared to 0.3 for the cultivated one, which was less than within our set of cultivated ones. Furthermore, Aljazairi, et al, (2015) indicated that modern genotypes tend to invest more C in their spikes than do the traditional genotypes, which invested more C in non-reproductive shoot tissues. A large variability in grain yield, shoot and root biomass amongst cultivated genotypes was observed in the present study.

9.4.3. C stabilization into the soil

The accumulation of soil organic C requires a positive balance between inputs and outputs from soil organic matter (SOM) stocks. Genotypes such as LM47 and LM48 showed superior ability to transfer atmospheric C to the soil matrix, which was estimated to be about 30% higher than the least efficient genotypes. The stability of this C is still not known, but the fact that soil sampling was performed weeks after pulse labelling and after harvesting demonstrates a certain level of stability of the C, because the most labile fractions decompose rapidly. Hütsch et al. (2002) showed that cereal root exudates, which are 80% water soluble (64% carbohydrates, 22% amino acids and 14% organic acids), are rapidly (1-2 days) stabilized into water-insoluble forms and bound preferentially to clay particles. Warembourg and Estelrich (2000), in their study of a permanent grass, showed that 56 to 69% of the net C assimilation was below ground with 52-62% into roots, 24-31% being respired back to the atmosphere (most labile exudate fractions) and 13-21% being found in the soil matrix. However, increased production of root exudates by the best C genotypes could, however, stimulate microbial activity and accelerate the turnover of SOM (Phillips et al.,, 2011), a process known as the "priming effect", where soil organic matter (SOM) decomposition is stimulated by addition of labile substrates to the soil (Jenkinson et al.,, 1985; Dalenberg and Jager, 1989). In support, Shahzad et al. (2018) pointed that deep soil layers stimulates mineralization of millennia-old organic carbon. However, this process might be limited because exudate stabilization is quick and a large fraction of root exudates become stable organic matter (Warembourg and Estelrich, 2000). Finally, the large amount of dead root and shoot material produced by LM48 and LM47 might also become important sources of soil organic matter, hence their contribution to total soil organic matter should be further investigated.

9.5. Conclusion

Three main conclusions can be drawn from this study that investigated the abilities of existing wheat genotypes (from CIMMYT) to build soil carbon. The first conclusion was that the ability of the genotypes to store atmospheric carbon into the soil varied highly with differences of up to 87% under drought conditions, and up to 117% under well-watered conditions. The second conclusion was that C transfer to the soil was higher under well-watered than water-stressed conditions. The third conclusion was that the more a genotype invests in allocating carbon into its shoots the less carbon goes to the soil. Such an improved understanding of the links between plant characteristics and soil C storage may open opportunities for wheat breeding with climate change mitigation in mind. Research on stover management strategies that address farmer needs while enhancing soil C is also needed. In addition, further research on factors that control the transfer and storage of atmospheric C into soils through plants is still important.

Chapter 10 CARBON SEQUESTRATION ANALYSIS UNDER FIELD CONDITIONS

Abstract

Field crops have potential to offset agricultural carbon emissions but the impact of plants on carbon sequestration has not been fully investigated. Field experiments were therefore conducted to investigate the potential of maize, sorghum and wheat genotypes to sequester C in below and above ground biomass. Ten genotypes each of sorghum, maize and wheat were cultivated at Cedara and Ukulinga. Two water regimes were used as follows: drought-stressed (DS) and well-watered (WW). Shoot and root biomass (SB and RB), grain yield, carbon and nitrogen contents were measured. Biomass and grain yield significantly varied among the crops and genotypes of the same crop species. Sorghum accumulated the highest plant biomass (31.5 Mg ha^{-1}), followed by maize (24.9 Mg ha^{-1}) and wheat (13.4 Mg ha^{-1}). A similar trend was observed for root biomass with sorghum (12.5 Mg ha^{-1}) followed by maize (7.1 Mg ha^{-1}) and wheat (4.7 Mg ha^{-1}), which resulted in a root to shoot ratio of 0.69 for sorghum, 0.55 for wheat and 0.41 for maize. The crops also differed highly in their ability to store carbon into their biomass with significantly greater carbon contents in sorghum and maize shoots and roots but lower C: N ratio for sorghum (19.9 in shoots; 29.2 in roots) as compared to the other crops. Overall, below ground biomass production was highest in sorghum concomitant with inherent ability to withstand harsher soil conditions, while it was least in wheat. The best cultivars for root biomass and root C stocks were AAS15 for sorghum, NEL and ZM1423 for maize and LM 26 and BW140 for wheat. Sorghum incurred the least grain yield loss of 33% due to drought compared to 42 and 48% loss in wheat and maize. Carbon sequestration in biomass is species-specific and is also influenced by genotype and environmental conditions. The species-specificity allows comparison and selection of suitable crops varieties.

Keywords: biomass production; carbon sequestration; root: shoot ratios; drought; stress, maize, sorghum, wheat

10.1. Introduction

Agriculture is the backbone of almost all developing economies around the world. In sub-Sahara Africa, staple cereal production dominates the agricultural landscape. Maize and sorghum are the most important crops by tonnage (Taylor, 2003) where they are grown for food and fibre for human population, livestock feed and as industrial raw materials. Maize feeds more than 50% of the population in Sub-Saharan Africa (Smale et al., 2011) and is used in industrial extraction of starch, molasses for biofuels and other additives. In South Africa, white maize is produced on 1 643 100 ha while yellow maize occupies 985 500 ha (DAFF, 2017). The major producing provinces are e the Free State, North West and Mpumalanga. Annual production of white maize is estimated at 16,744 million tons, with an average productivity of 6,37 t/ha. The white maize is the most important with the extraction of super sifted maize meal accounting for up to 89% of maize utilization (National Chamber of Milling, South Africa). Similarly, sorghum is used extensively as a source of carbohydrates for human daily food intake. It is also an important ingredient in breweries. Over the years, an average of 42 350 ha was planted to sorghum annually in South Africa with Mpumalanga (41,2%), followed by Limpopo (33,3%), Free State (16,3%) and the North West (6,6%) being the highest producing provinces (DAFF, 2017). Annual production was 147 740 tons, which contributed about R100 million per annum or 0,7% to gross domestic value of crops (DAFF, 2017). The importance of these crops has led to intensification of their production across the region. However, maize and sorghum are commonly cultivated under smallholder agriculture systems characterized by numerous challenges such as drought stress and lack of inputs, which impact negatively on the crops' biomass production and their ability to sequester carbon.

Generally, the over-dependence on agriculture to support national gross domestic product (GDP) and economic growth has resulted in intensification of crop production in many sub-Saharan economies (Diao et al., 2010). The intensification of maize and sorghum production in SSA also reinforces food security status of many countries in this region and increases household food security in communal areas. However, intensification of crop production in sub-Sahara Africa is coupled with negative effects such as environmental pollution and soil C depletion; which have a complex cause-effect relationship (Gregory et al., 2002). Intensive crop production is characterized by soil degradation through improper tillage methods, and environmental pollution from excessive use of inorganic fertilizers and chemicals. This will potentially increase C emissions through improper agronomic practices (Adhya et al., 2000), despite the inherent potential of crops to offset C emissions. Very few studies have been conducted in sub-Saharan Africa, especially South Africa, on the impact of crop production in terms of GHG emissions and soil organic carbon (SOC) storage.

Organic carbon stocks of a particular soil are known to be a balance between carbon inflow and outflow. Although adopting good agronomic practices and the proper management of crops residues helps to reduce carbon emission from agricultural soils, their impact on soil carbon stocks is debatable (Dimassi et al., 2014). In contrast, plants are sole depositors of organic carbon into the soils via their root activity and debris, which is critical in long-term storage of C in plant and soil fractions of agricultural ecosystems (Albrecht et al., 2003). Carbon sequestration depends on soil and climatic conditions as well as plant characteristics. Soil properties such as pH, clay content and microbe populations; and climatic factors, e.g. temperature and precipitation, have profound impact on C sequestration by plants and soils (Davidson & Janssens, 2006). Among the plant factors are species, quantity and quality of deposits and growth habit. Mathew et al. (2017) analyzed global data on C sequestration by plants and posited that biomass productivity was the most important determinant of C sequestration by plants. Plant biomass is largely made up of C, which accounts for 40% on average (Cooley & Manning, 1987). Thus, species or genotypes that produce large quantities of biomass sequester relatively large amounts of C from the atmosphere into their biomass. It also means that, potentially, such genotypes will have large quantities of residues to deposit into the soil to increase the soil C balance. In addition to the quantity of the biomass, quality is also important. The quality can be defined in terms of C composition in relation to other nutrients such as nitrogen. These differences in nutrient composition affect the decomposition rate of plant residues, which influences the accumulation and stabilization of carbon in the soil (Zhou et al., 2014). The ratio of C relative to nitrogen is particularly important as it defines the recalcitrance or degradability of the material. Plant residues with high C: N ratio are highly recalcitrant and cannot be easily decomposed by soil microbes, which means they become long term reservoirs for C (Ghimire et al., 2017). High C content in plant residues will promote high C sequestration potential. Genotype and water availability affect both quantity and quality of plant biomass in sub-Sahara, but there is a lack of studies highlighting the impact of biomass production on soil C sequestration among genotype of species such as maize and sorghum.

Some studies have evaluated interspecific differences in C sequestration potential based on C content and biomass under different agronomic practices. However, variation within species for maize and sorghum are still scantly reported. Variation in C content between cereals such as maize, sorghum, wheat and rice has been reported by McKendry et al., (2002). The higher C content in maize was attributed to its higher hemicellulose content in the stover (Gomez et al., 2008). Biomass was found to be high in maize especially in the tropical varieties which tend to be tall and thick. For sorghum, high biomass was reported in silage and biofuel varieties compared to sweet grain sorghum (Vasilakoglou et al., 2011). Such variation in biomass accumulation and C composition have implications on C sequestration potential under different field management practices. Quantifying genetic variation among genotypes of maize and sorghum for their C sequestration potential under field conditions remains a knowledge gap that needs to be filled. This will assist in decision making for farmers, genotype specific estimation of C budgets and future selection for breeding. Selecting carbon superior genotypes along with good agronomic practices such as reduced tillage and appropriate crop rotations will contribute to reducing C emission, ameliorating soil fertility and improving crop productivity in the long term (Ali et al., 2012).

10.2. Materials and methods

10.2.1. Plant materials

Ten wheat genotypes selected from a previous study for high drought tolerance index were also included in this study. Ten genotypes of maize obtained from various sources were evaluated in this study. The maize genotypes included open pollinated varieties, hybrids and commercial varieties to represent a wide genetic pool. Commercial varieties of maize were included as possible control checks as they are already in production in South Africa. The maize genotypes were selected based on previous evaluation for pest resistance under field conditions and will provide important genetic resources to improve local commercial varieties. The genotype Mac Medium Pearl is an open pollinated variety (OPV) grown in South Africa. Some varieties imported from Zimbabwe were included because of their drought tolerance potential. Zimbabwe and South Africa share a lot of germplasm through commercial seed companies and it is common to test varieties in both countries for adaptation. The genotype from Ghana has been tested locally by the Council for Scientific and Industrial Research (CSIR) of South Africa for possible inclusion in breeding programs to improve quality protein content. It will thus be worthwhile to investigate its C sequestration potential given its high nutritional content. Sorghum accessions on the other hand were still in developmental stages and these were previously evaluated for drought tolerance and diversity. The sorghum accessions used represent a considerable genetic pool as they were collected from different provinces across South Africa and possibly exhibit divergent adaptation.

10.2.2. Growing conditions and trial management

The field experiments were set up at Cedara (LAT: 29°67’S LONG: 30°41’E and ALT: 1076 m) and Ukulinga Research farms (LAT 29° 67′E, LONG 30° 41′S and ALT: 809) (ARC, 2015). Long-term average temperature and rainfall for Cedara are 15°C and 900 mm, respectively. Cedara is located in the Midlands KwaZulu-Natal mist belt and has high humidity, relatively higher rainfall and lower temperatures (Fairbanks and Benn, 2000). It is characterized by sandy clay soils with high organic matter content, which are reasonably fertile and well drained with low risk of flooding due to a good slope and ground cover. Maize was the preceding crop on the site before the trials were established in 2019, so its debris was abundant. The ground cover also provided mulch and helped in moisture conservation. Ukulinga is generally dry and warmer than Cedara, with mean annual temperature and precipitation of 18°C and 738 mm, respectively (Table 10.1). The soil at Ukulinga farm is loam, fertile and friable with good drainage. However, it is susceptible to cracking and crusting under flooding. The previous crop at Ukulinga was maize and the residues were ploughed under giving good organic matter content. The field was ploughed in January 2019 during the rainy season. Supplementary irrigation was applied through a sprinkler irrigation system at both sites when necessary to maintain adequate soil moisture.

10.2.3. Crop establishment

For maize, two seeds were planted per station at 15 cm intra-row spacing and 75 cm between rows soon after ploughing. Each genotype was planted per 5 row plot in a block consisting of 20 rows with randomly assigned genotypes. The plants were thinned to one plant per station 2 weeks after emergence to leave 20 plants per row. Basal fertilizer sufficient for 10-ton grain crop was band-applied at planting. A top-dressing fertilizer was applied at a rate of 120 kg/ha N, 33 kg/ha P, 44 kg/ha K at Cedara while at Ukulinga the top dressing was applied at 120 kg/ha N, 40 kg/ha P, 45 kg/ha K. Basal fertilizer NPK (2:3:4) was applied at a rate of 250 kg/ha at both sites. Top dressing was applied four weeks after crop emergence and supplied in the form of lime ammonium nitrate (LAN) (28%) kg/ha as single application.

For sorghum, five seeds were planted per station at 20 cm intra-row spacing and 80 cm between rows soon after ploughing. Each genotype was planted per 3 m row plot in a block consisting of 20 rows with randomly assigned genotypes. The plants were thinned to one plant per station 2 weeks after emergence. Basal fertilizer NPK (2:3:4) sufficient for an 8-ton grain crop was applied before planting at a rate of 250 kg/ha. Top-dressing fertilizer was applied at the rate of 100 kg/ha N, 24 kg/ha P, 20 kg/ha K at both Cedara and Ukulinga. Top dressing was applied four weeks after crop emergence and supplied in the form of lime ammonium nitrate (LAN) (28%) kg/ha as a single application. Wheat was only planted at Ukulinga. Three seeds were planted per station at 10 cm intra-row spacing and 30 cm between rows soon after ploughing. Each genotype was planted per 3 m row plot in a block consisting of 20 rows with randomly assigned genotypes. The plants were thinned to one plant per station 2 weeks after emergence. Basal fertilizer composed of nitrogen (N), phosphorous (P) and potassium (K) was applied at a rate of 120:30:30 kg ha^{-1} (N: P: K). Other agronomic practices were as per normal wheat production (DAFF 2010).

10.2.4. Weed and pest control

Standard cultural practices, including hand planting, hand weeding and application of herbicides were followed. Weeds and pests were controlled by chemical sprays. Mechanical weed control from just before emergence until just before the piping stage was done for sorghum. Weeds were controlled by a combination of Basagran, Gramoxone and Troopers for the control of annual grasses and broadleaf weeds. A combination of pesticides Lambda and Carbofuran was applied at recommended rates both as preventative and curative measures at six weeks after crop emergence to prevent cutworm in all crops and stalk borer damage in maize and sorghum.

Table 10.1. Soil properties and mean temperatures at Ukulinga and Cedara

Property	Ukulinga	Cedara
pH (KCl)	4.56	5.4
Organic carbon (%)	2.6	6.5
Phosphorous (mg/L)	39	32
Potassium (mg/L)	241	275
Calcium (mg/L)	1453	931
Magnesium (mg/L)	369	249
Nitrogen (%)	0.23	0.68
Clay (%)	28	42
Mean Temperature (°C)	16.6	14.7

10.2.5. Data collection

The plants were grown to maturity, ~4 months, after which 8 plants (half the plot) were harvested from each plot. The shoot biomass was cut off at the soil surface to separate from root biomass. All the roots to a depth of 60 cm within the soil volume were collected per genotype per plot using a monolith box. The larger roots were manually separated from the soil and the finer roots were collected by wet sieving. Plant parts for each plot were then separated into grain, shoot and root, then dried at 60°C for 72 hours in a dry air oven. Afterwards, the dry matter of the separate plant parts was weighed on a digital scale and converted to kilograms per square meter using the plot size (3 m*0.75 m for maize and 3 m*0.75 m for sorghum). Root: shoot (R: S) ratio was computed accordingly. The separated plant parts were then ground into fine powder to pass through a 0.5 mm sieve mesh, after which their carbon and nitrogen contents was analyzed using the LECO (CNS Trumac autoanalyser).

10.2.6. Data analysis

Summary statistics of minimum, maximum, mean, median, Q1, Q3, skewness and kurtosis of the data were derived to ascertain distribution of the data. After checking for normality, the data on biomass, yield, carbon and nitrogen contents was subjected to general analyses of variance for balanced designs.

*Partial destruction of the trial by birds and monkeys occurred at Cedara and Ukulinga. The extent of destruction was terribly high at Cedara, so grain yield and data from Cedara was not reported on.

10.3. Results

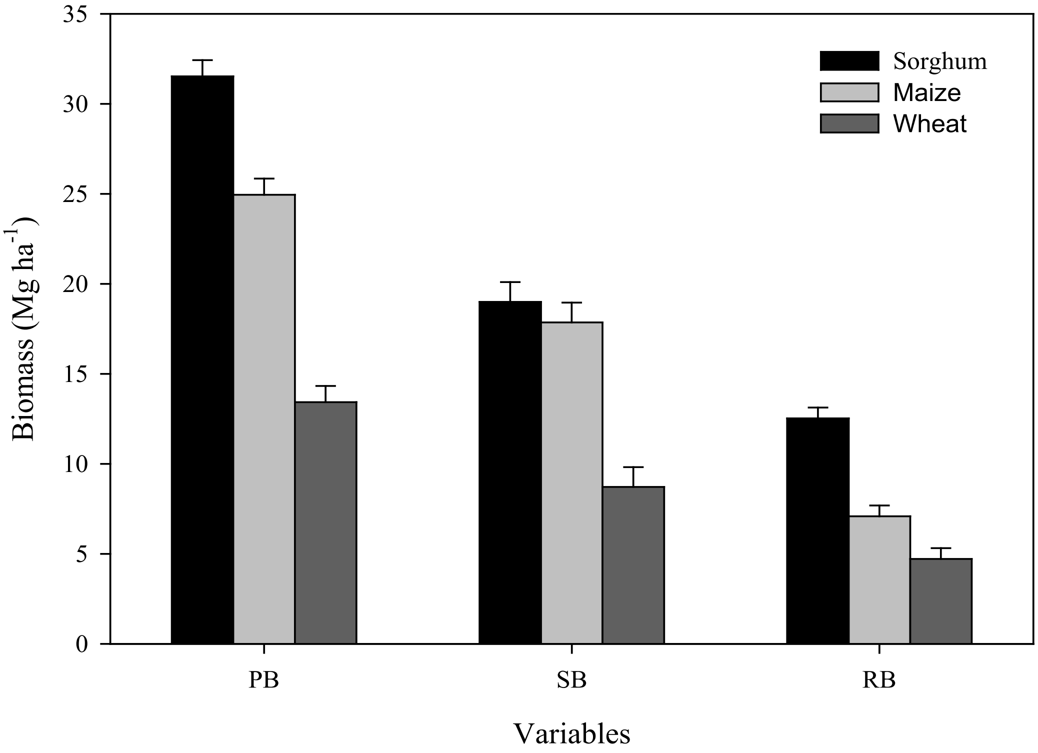
10.3.1. Biomass productivity of different crops

Data on mean performance of different crops under drought stressed (DT) and well-watered (WW) conditions are presented in Table 10.2. The mean plant biomass across the water regimes was highest for sorghum (31.5 Mg ha^{-1}), which was 2.3 times higher than wheat (13.4 Mg ha^{-1}) and 1.3 times higher than maize (24.9 Mg ha^{-1}). Similarly, shoot biomass decreased from 18.99 Mg ha^{-1} for sorghum, 17.86 Mg ha^{-1} for maize to 8.71 Mg ha^{-1} for wheat. A similar trend was observed for root biomass with sorghum (12.53 Mg ha^{-1}), followed by maize (7.09 Mg ha^{-1}) and wheat (4.72 Mg ha^{-1}), which resulted in a root to shoot ratio of 0.69 for sorghum, 0.55 for wheat and 0.41 for maize. These results point to sorghum as the plant producing the highest amount of biomass, with the highest below ground allocation (Fig 10.1). From Figure 10.2, we learn that sorghum and maize showed the greatest differences among cultivars (ranges) as compared with wheat, for total biomass, shoot and root biomass (Fig 10.2A-C). For instance, total plant biomass ranged from 20 to 47.5 Mg ha^{-1}, i.e. a 2.3 times difference for sorghum, between 9 to 40 Mg ha^{-1}, i.e. a 4.4 times difference for maize, as compared to a 1.5 times difference for wheat.

Table 10.2. Performance of different crops under drought stressed (DT) and well-watered (WW) conditions

	Mean			Sorghum		Maize		Wheat	
Variable	Sorghum	Maize	Wheat	DT	WW	DT	WW	DT	WW
PB (Mg ha^{-1})	31.52	24.95	13.43	26.48	36.56	17.42	32.47	12.29	14.56
SB (Mg ha^{-1})	18.99	17.86	8.71	15.46	22.53	11.66	24.06	7.83	9.59
RB (Mg ha^{-1})	12.53	7.09	4.72	11.02	14.03	5.76	8.42	4.46	4.97
RS (Mg ha^{-1})	0.69	0.41	0.55	0.73	0.64	0.47	0.34	0.58	0.52
SC%	42.03	42.02	31.75	41.99	42.07	41.81	42.23	31.84	31.66
RC%	35.21	38.57	32.51	34.98	35.44	36.8	40.34	31.98	33.04
SC:N	19.98	24.2	40.79	19.68	20.29	23.63	24.77	39.86	41.71
RC:N	29.16	55.14	39.22	28.71	29.61	53.26	57.02	41.59	36.85
PCs (Mg ha^{-1})	12.22	10.09	7.36	10.22	14.22	6.84	13.35	6.22	8.5
SCs (Mg ha^{-1})	7.97	7.52	2.77	6.48	9.45	4.87	10.17	2.49	3.01
RCs (Mg ha^{-1})	4.45	2.72	1.54	3.88	5.02	2.13	3.32	1.43	1.67

PB=total biomass (shoot + root), SB=shoot biomass, RB=root biomass, RS=root to shoot ratio, SC%=shoot carbon content, RC%=root carbon content, SC:N=carbon to nitrogen ratio in shoot, RC:N=carbon to nitrogen ratio in root, PCs=total carbon stocks (shoot + root) calculated as product of biomass and average carbon content, SCs=shoot carbon stocks; i.e. SB*SC%, RCs=root carbon stocks i.e. RB*RC%



*lines above each bar represent standard error

Figure 10.1. Average biomass production by the different crops

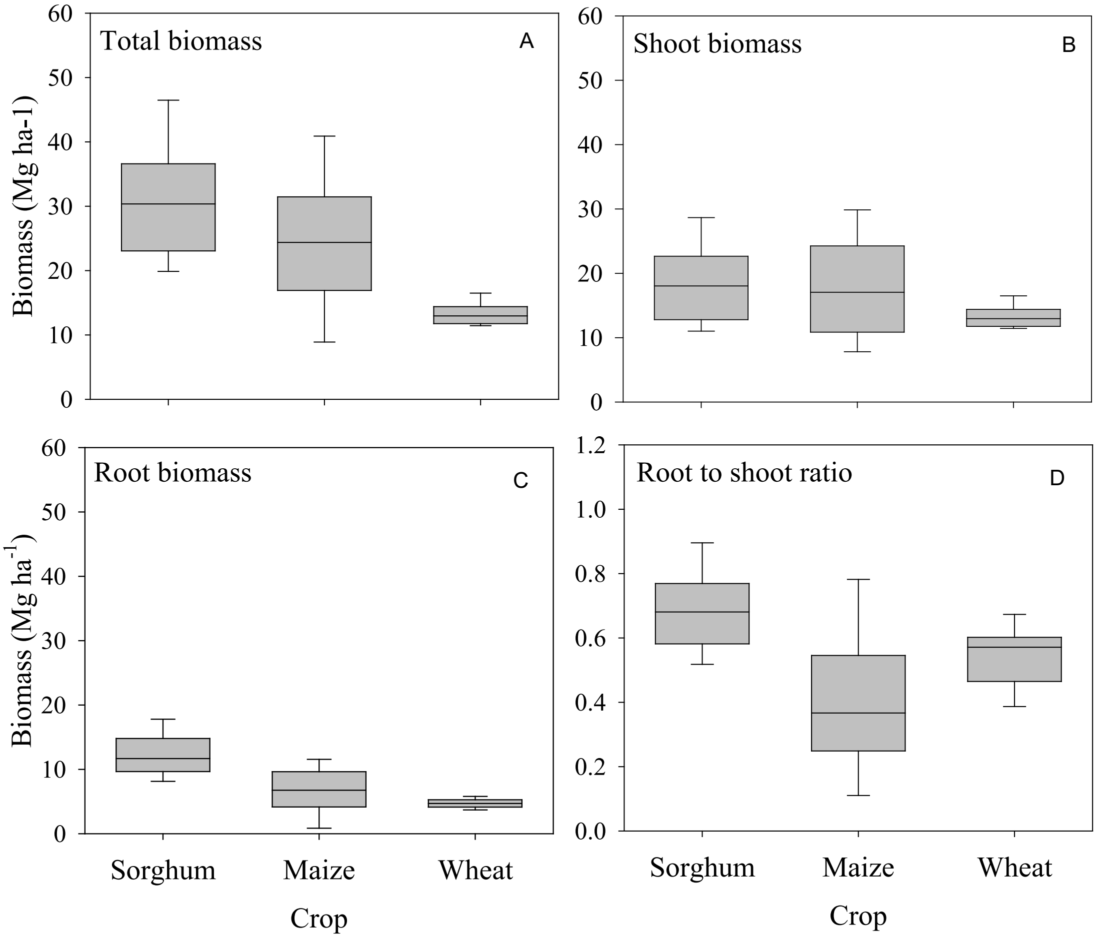
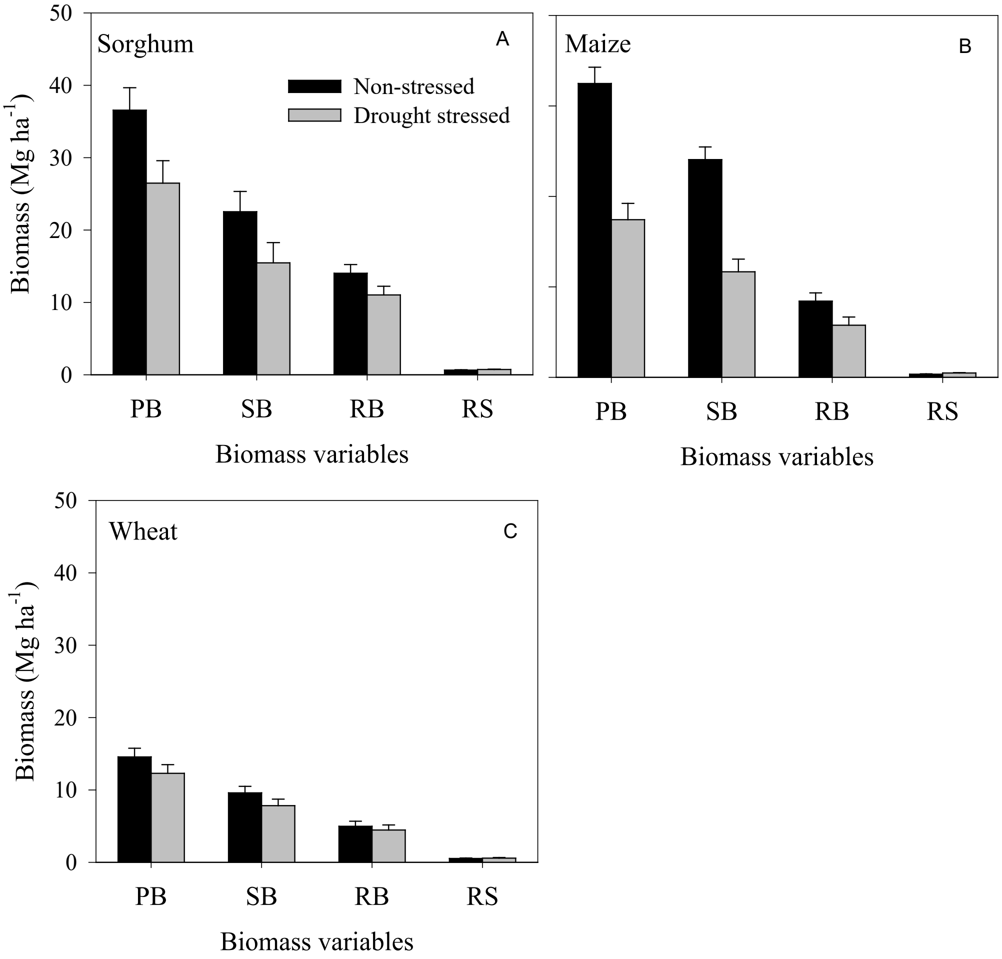


Figure 10.2. Variation in biomass production of different crops

10.3.2. Impact of water regimes on biomass productivity of different crops

Biomass production was also significantly affected by the water regime (Table 10.2, Figure 10.3). For instance, sorghum total biomass decreased from 36.56 Mg ha^{-1} under well-watered to 26.48 Mg ha^{-1} under stressed conditions, which corresponded to a 27.6% decrease. Maize plant biomass decreased from 32.47 to 17.42 Mg ha^{-1} (i.e. a 46.4% decrease) while wheat biomass decreased the least (15.6%: from 14.56 to 12.29 Mg ha^{-1}). Shoot biomass was the most affected by drought stress with biomass reduction values being 51.5% for maize, 31.4% for sorghum to 18.4% for wheat, and plants tended to allocate more of their biomass into their shoots as expressed by a R: S of 0.73 for stressed sorghum as compared to 0.64 for non-stressed conditions.

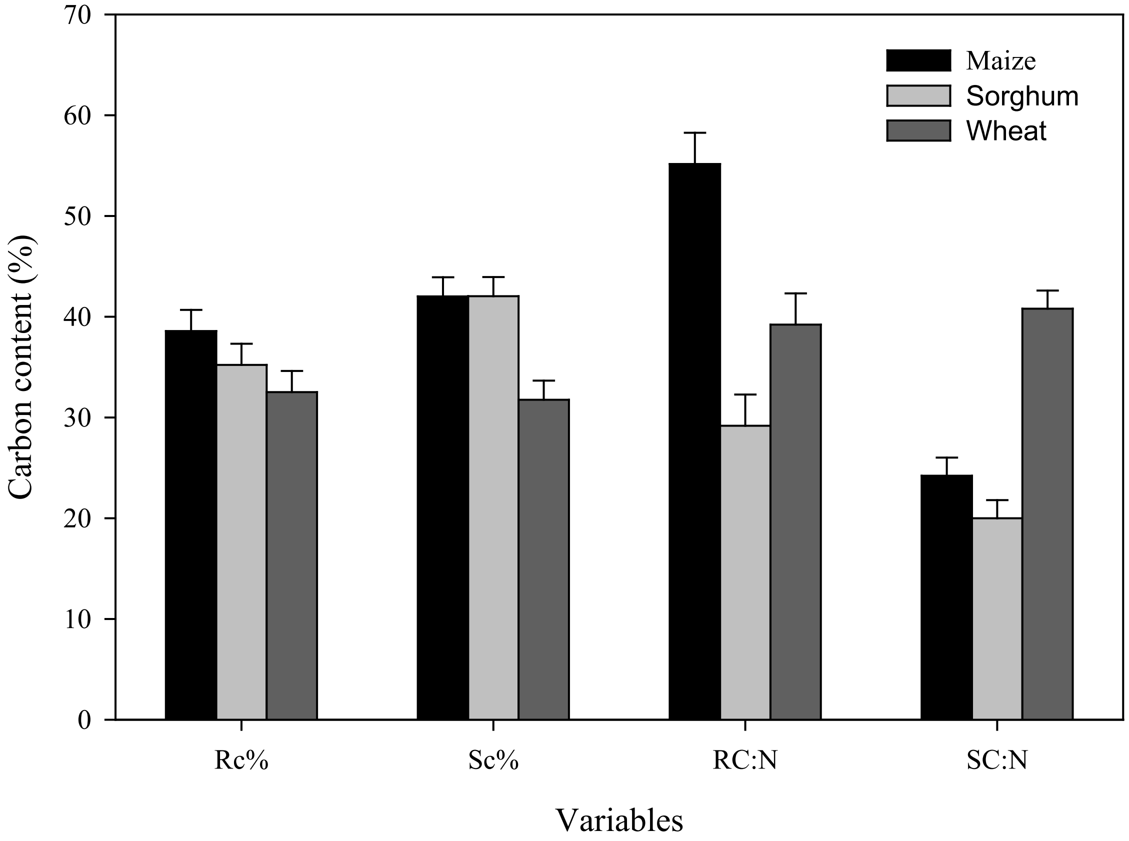


*lines above each bar represent standard error

Figure 10.3. Biomass production by different crops under different water regimes

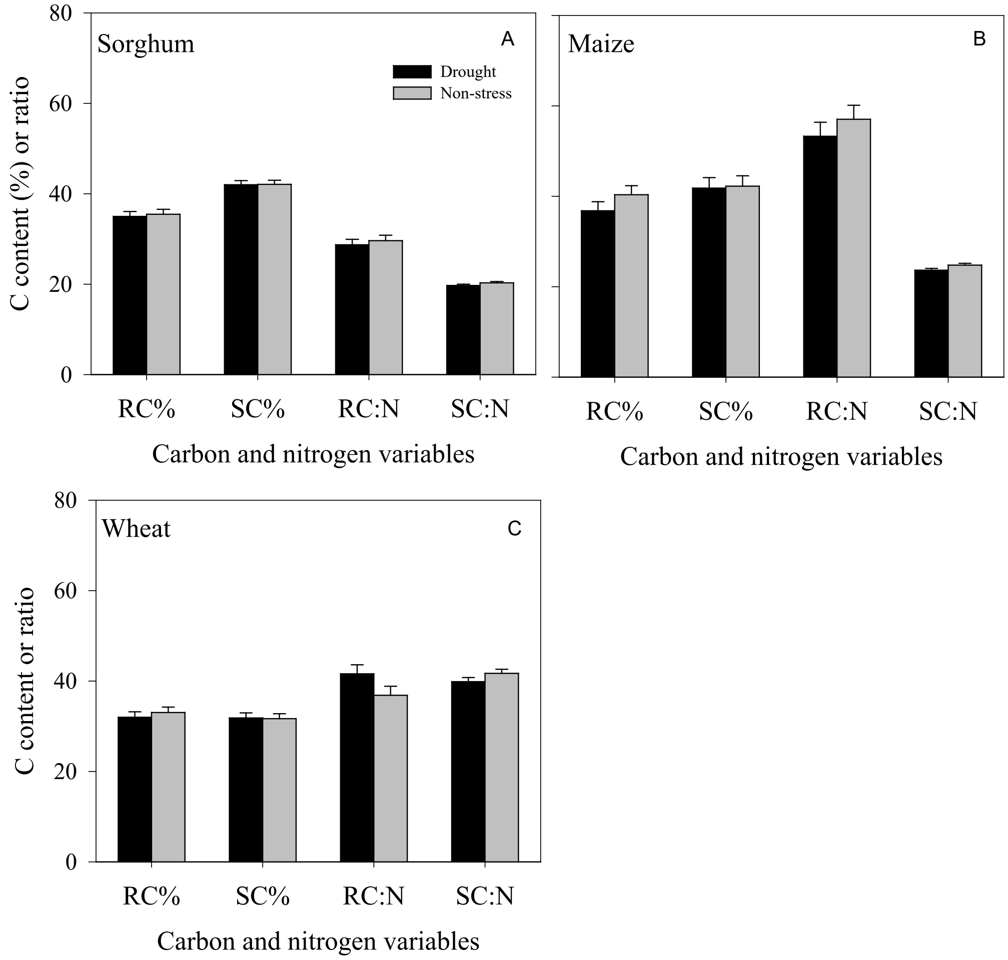
10.3.3. Carbon composition of different crops

Table 11.2 also informs about the differences between crops in the ability to store carbon into their biomass. The carbon content of plant shoots ranged between 42% for sorghum and maize to 31.75% for wheat. For roots, the highest C content was observed for maize (38.57%) followed by sorghum (35.21%) and wheat (32.51%) (Fig 10.4, Table 10.2). While shoot carbon content was less affected by water regime; root carbon content significantly increased with increasing water availability for maize by 9.3% and for wheat by 3.2% (Table 10.2, Fig 10.5). Figure 10.6 revealed that the range in root C content between the study cultivars was similar for the three crops with an interquartile from 6 to 8% (Fig 10.6A). In contrast, the interquartile of cultivars' shoot C content was below 2% for sorghum and maize versus 8% for wheat. The resulting plant carbon stocks thus decreased from 12.22 Mg ha^{-1} on average for sorghum, to 10.09 Mg ha^{-1} for maize and to 4.36 Mg ha^{-1} for wheat, with the greatest below-ground stocks also recorded for sorghum (4.45 Mg ha^{-1}), followed by maize (2.72 Mg ha^{-1}) and wheat (1.54 Mg ha^{-1}) (Table 10.2, Figure 10.7). The variability of carbon stocks was smallest for wheat shoots and roots as compared with sorghum and maize (Figure 10.7).



*lines above each bar represent standard error

Figure 10.4. Average C content in the plant residues of the different crops. Rc and Sc%=root carbon content RC:N=root C to root N ratio SC:N=shoot C to shoot N ratio



*lines above each bar represent standard error

Figure 10.5. Average C and N composition of plant residues under different water regimes.

Rc and Sc%=root carbon content RC:N=root C to root N ratio SC:N=shoot C to shoot N ratio

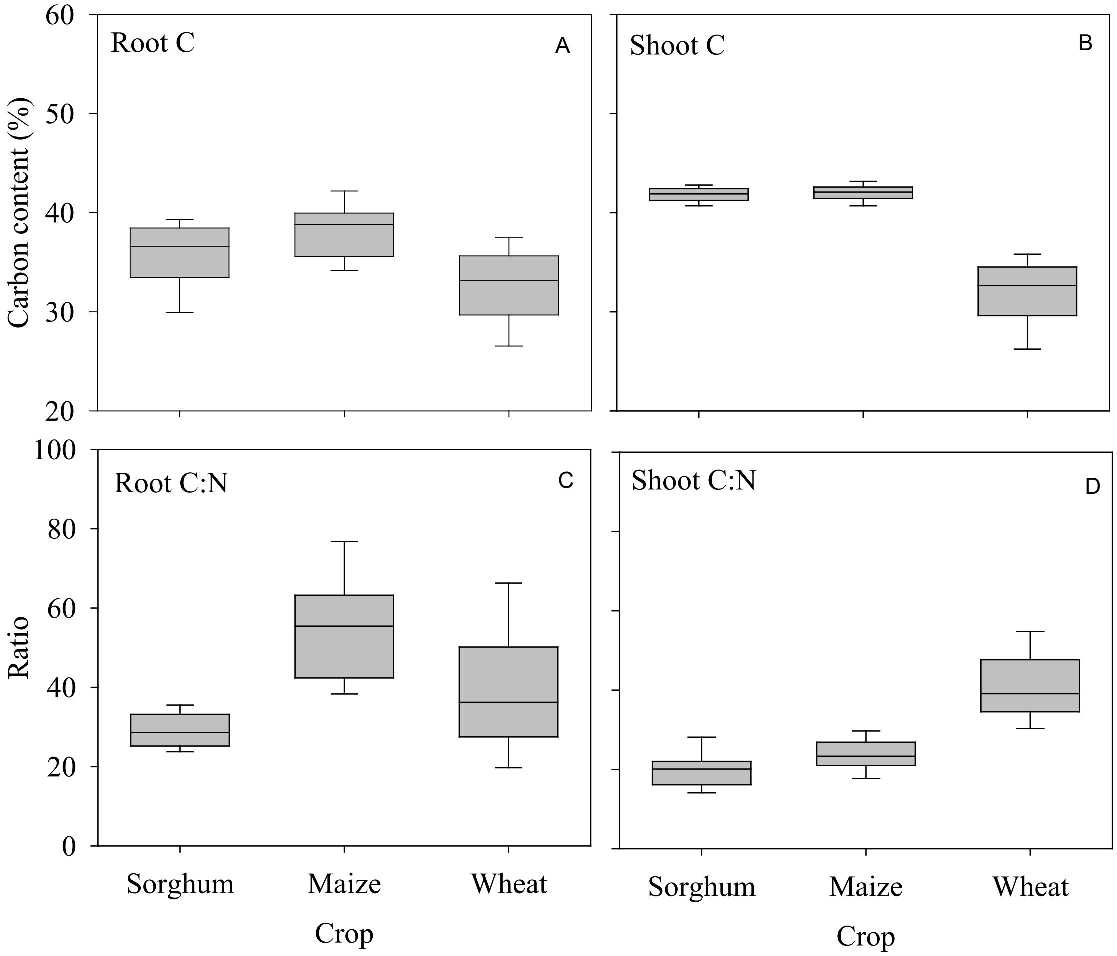
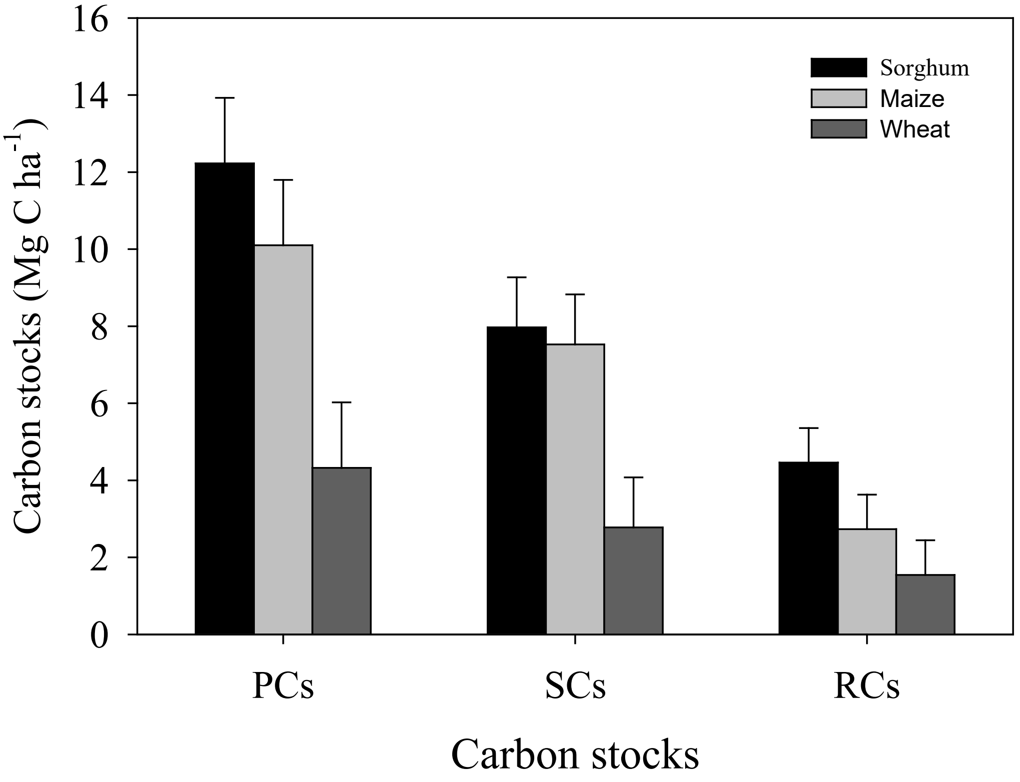


Figure 10.6. Variations in C content and C: N ratios of roots and shoots of different crops

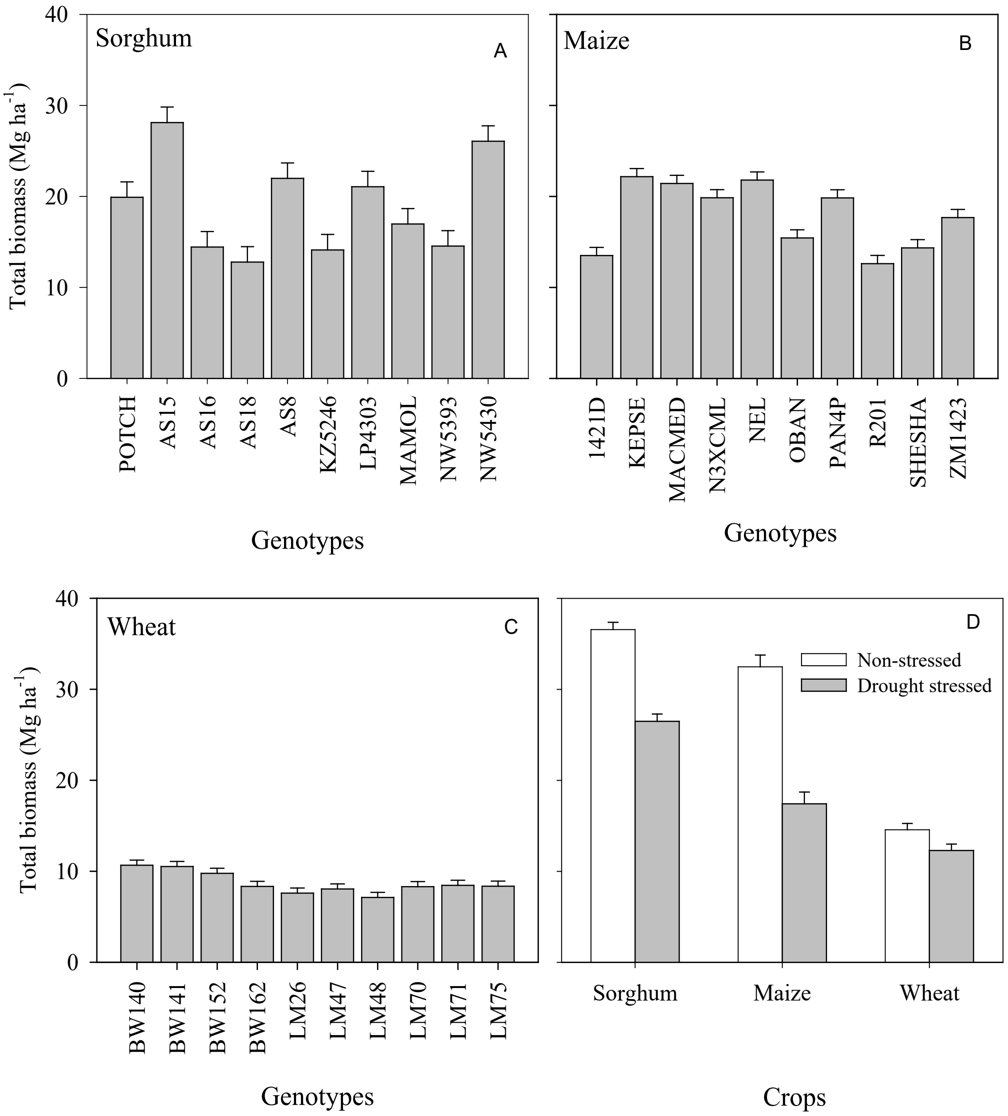


*lines above each bar represent standard error

Figure 10.7. Average total plant, shoot and root carbon stocks in the different crops

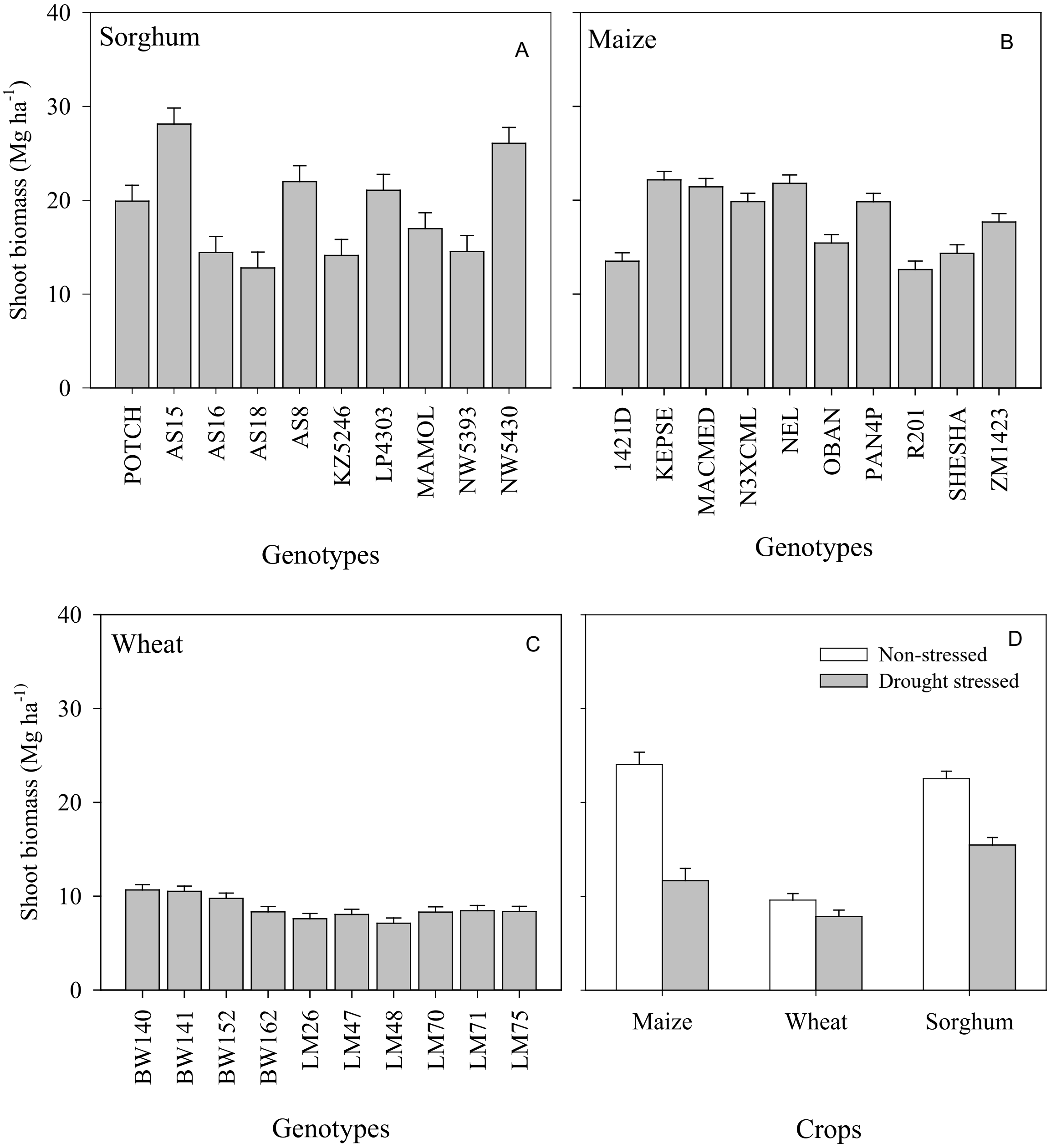
10.3.4. Biomass productivity of different crop cultivars

Overall, the cultivars producing the highest total biomass were AS15 for sorghum, Nelson QPM CPSTN for maize and BW140 for wheat (Figure 10.8). AS15 (sorghum), Nelson QPM CPSTN (maize) BW140 and 141 (wheat) were also the best varieties for shoot and root biomass production (Figs 10.9 and 10.10).



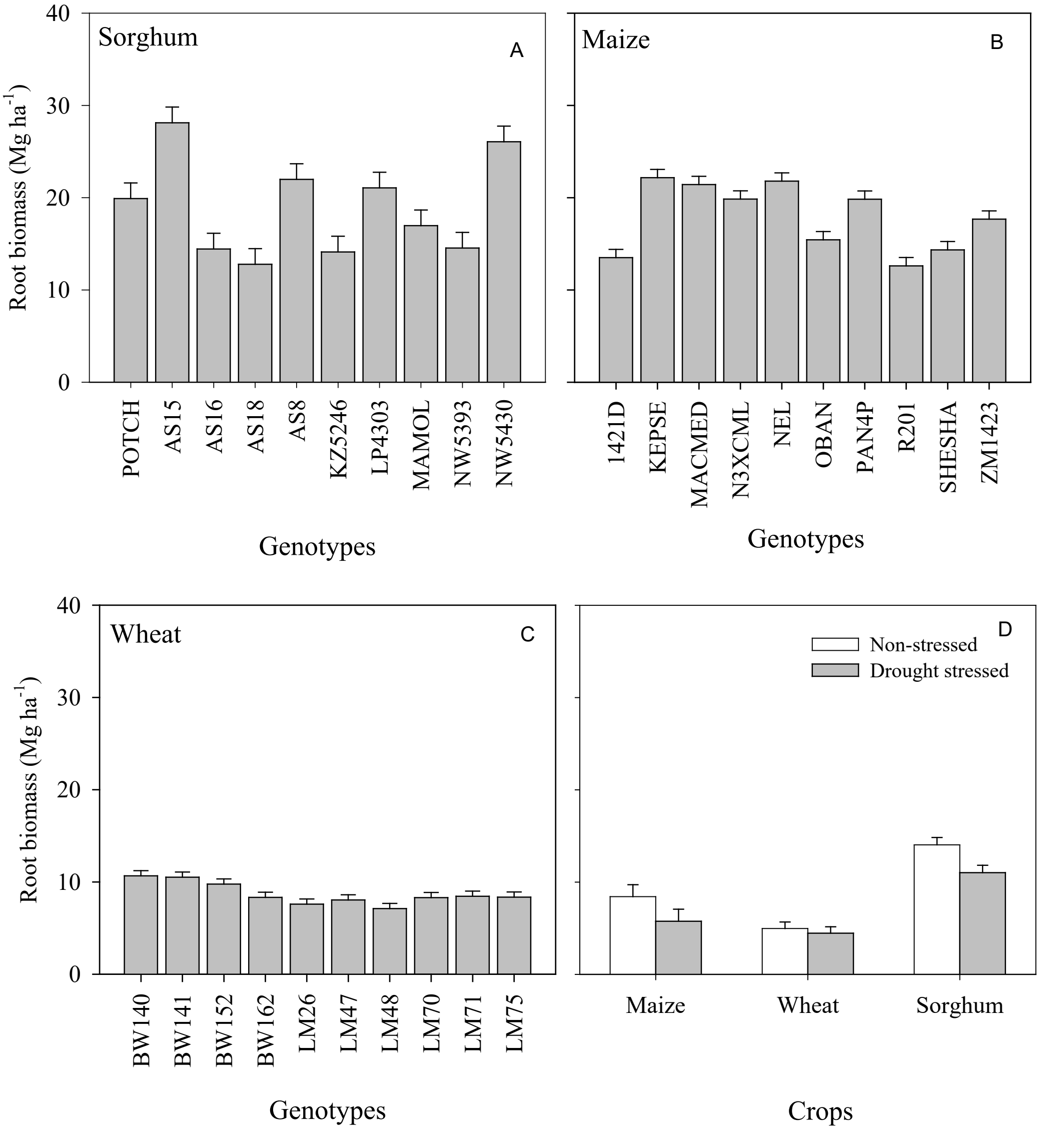
*lines above each bar represent standard error

Figure 10.8. Variation in total plant biomass production among different crop genotypes



*lines above each bar represent standard error

Figure 10.9. Variation in shoot biomass production among different crop genotypes

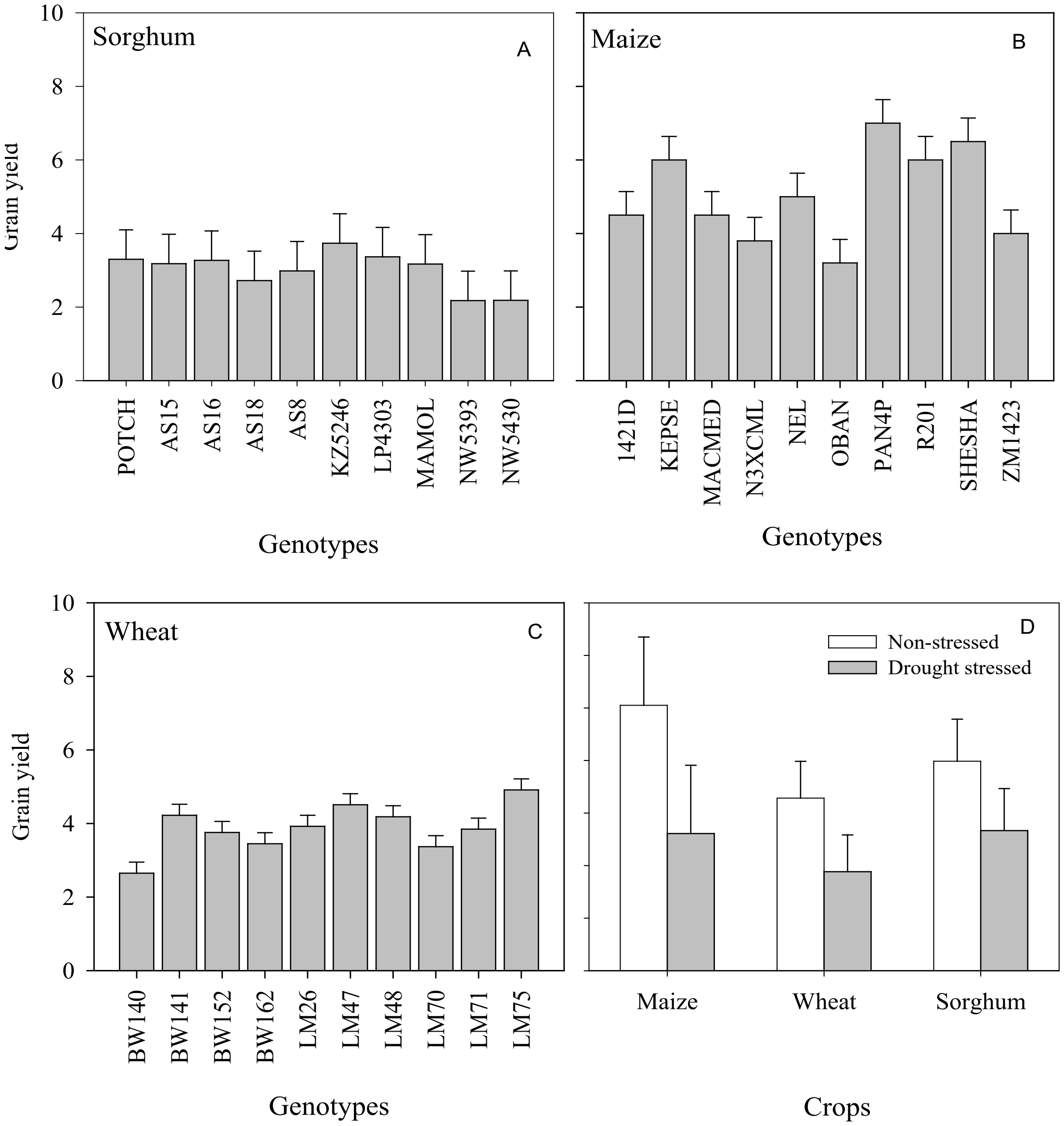


*lines above each bar represent standard error

Figure 10.10. Variation in root biomass production among different crop genotypes

10.3.5. Grain yield productivity of different crops

Genotype KZ5246 had the highest grain yield among the sorghum genotypes, with mean yield of 3.7 Mg ha^{-1}, although there was little variation among the genotypes (range 2.2 to 3.7 Mg ha^{-1}) (Fig 10.11A). Maize genotypes exhibited significant variation in grain yield ranging between 3.2 to 7.0 Mg ha^{-1}. The commercial variety PAN4P-228 had the highest yield of 7.0 Mg ha^{-1} compared to 3.2 Mg ha^{-1} attained by Obanpta (Fig 10.11B). Wheat also showed significant variation (2.6 to 4.9 Mgha^{-1}) with LM75 being the most grain productive genotype (Figure 10.11C). Water availability affected grain productivity in all the crops, with drought stress inducing 48, 42 and 33% grain yield reduction in maize, wheat and sorghum, respectively. In general, wheat had the lowest grain yield production under all water conditions compared to the maize and sorghum (Fig 10.11D).



*lines above each bar represent standard error

Figure 10.11. Variation in grain yield production among different crop genotypes

10.4. Discussion

10.4.1 Crop variation in biomass productivity and carbon accumulation

The field study concluded that biomass production and therefore carbon accumulation differed among the different crop types. There were further variations within a crop type due to genotype variation. These differences have implications on carbon sequestration since it is achieved primarily via two processes, i.e. decomposition of plant matter and rhizodeposition. The decomposition of plant matter is influenced by quantity and quality of biomass deposited into the soil. In this regard, sorghum produced large amounts of biomass compared to the other crops. Similar findings have been reported elsewhere and this could possibly be due to the non-differentiation of sorghum germplasm into grain or silage genotypes (Maw et al., 2017). Silage sorghum genotypes, which are mainly cultivated for biofuel production are known to be prolific biomass producers. In comparison, maize can also be used in silage and biofuel production as it is comparatively competitive in biomass production. Wheat is not known for being a prolific biomass producer compared to the C4 crops (maize and sorghum) under temperate conditions (Evans and von Caemmerer, 2000).

10.4.2. Crop variation in chemical composition of biomass

Based on biomass, especially root biomass, sorghum appears to have the highest potential to improve C stocks in the soil. However, the quality of biomass, defined by the C: N ratio, also influences the C sequestration potential of crop residues (Chen et al., 2014; Romero-Olivares et al., 2017). This is important for microbial activity and thus controls the recalcitrance of plant matter. Thus, plant residues with high C: N ratio are more recalcitrant as they slow down microbial activity (Zhang et al., 2017). Sorghum exhibited the least C: N (both root and shoot) while maize had the highest C: N ratio in the roots and wheat straw appeared to be potentially more recalcitrant (highest C: N in shoots). Thus, further investigation is required to elucidate the cost benefit of accumulating higher biomass in sorghum compared to maize considering both their C: N ratios.

10.4.3. Crop variation in chemical composition of root biomass and exudates

The C: N ratio of exudates is also important during growth as it supports mycorrhizae development in the rhizosphere. Differences in root composition of the crops indicates that they most likely deposit exudates of different composition. Differences in deposits of crops such as maize, barley, wheat and rice have been reported (Velthof et al., 2002; Adapa et al., 2009; Li et al., 2013). Soil mycorrhiza are important in the agglomeration of C into SOM (Zhang et al., 2017).

10.5. Conclusion

Overall, the identified genotypes with high biomass, high C: N and grain yield will be recommended for production and for breeding. Differences in the growth habit and utilization of each crop (maize, sorghum and wheat) means that there will be no competing effects among the crops. Wheat is grown as a winter crop, thus providing C sequestration and ecosystem services during the minor season in SSA. Although maize and sorghum are both summer crops, they can complement each other. The drought tolerance exhibited by sorghum means that it is more favourable to increase C stocks in marginal areas. In contrast, maize is more suitable to replenish C stocks under high potential and intensively cultivated systems.

Chapter 11 CARBON SEQUESTRATION BASED ON CROP RESIDUES

Abstract

Soil management techniques such as crop residue incorporation are good strategies used to enhancing soil organic carbon sequestration. In addition, the decomposition of crop residues releases soil essential nutrients such as nitrogen, phosphorus, potassium and sulphur among others. Wheat and sorghum are important food crops commonly grown in South Africa. However, there is inadequate information on their potential to sequester carbon (C) and release nutrients once incorporated into the soil. The objective of this study was thus to assess carbon sequestration potential and mineralization patterns of wheat and sorghum residues from different genotypes upon incorporation into the soil. Laboratory experiments were conducted where about 0.25 g each of wheat root (RT) or shoot (ST) from 5 wheat genotypes namely LM70, LM75, BW140, BW152 and BW162 and 5 sorghum genotypes namely 05-POTCH-138, MAMOLOKWANE, KZ5245, AS8 and LP 4403 were mixed with 100 g of soil then transferred into an air tight PVC pot. NaOH solution was also placed inside the incubation pot to trap CO_2 released during decomposition, and this was measured on day 0, 7, 15, 23, 31, 39, 47, 55, 63, 77, 91,105, and 120 of incubation. Moist soil from each pot was also analysed for NH_4^+-N, NO_3^--N and extractable P mineralized during incubation. In general, wheat and sorghum shoots evolved higher net CO_2-C and mineralized more net amounts of N and extractable P compared to roots. Net CO_2-C evolved from shoot residues was 183.1 and 63.1 mg CO_2-C kg^{-1} soil for wheat and sorghum respectively while it was 71.8 mg CO_2-C kg^{-1} and 62.9 mg CO_2-C kg^{-1} for wheat and sorghum roots. Net total N mineralized was 28.78 and 20.23 mg N kg^{-1} soil for wheat and sorghum shoots while it was 19.6 and 15.91 mg N kg^{-1} for wheat and sorghum roots respectively. Net extractable P released was 5.148 and 4.939 mg P kg^{-1} soil for sorghum shoots and roots while it was 1.527 and 2.685 mg P kg^{-1} soil for wheat roots and shoots respectively. The best genotypes for carbon sequestration were LM70 roots for wheat, while it was AS8 roots as well as KZ5246 roots and shoots for sorghum. N mineralization was mineralised most after incorporating shoots of wheat genotypes BW162, BW140, BW152, while roots and shoots of the sorghum genotype 05-POTCH-138 were better N sources. Lastly, the best genotypes for P mineralization were BW162, BW152 and LM75 shoots for wheat as well as roots of the sorghum genotypes AS8. These results showed the potential and variation of wheat and sorghum genotypes on C sequestration and nutrient recycling.

Keywords: carbon sequestration, decomposition, incubation, mineralization, residues

11.1. Introduction

Poor soil fertility has been identified as one of the major constraints to crop production and agricultural productivity in Sub-Saharan Africa (Henao and Baanante, 2006). This is due to the inherent variability of soils in Sub-Saharan Africa (SSA). South Africa is characterized by soils with very low organic matter levels. Du Preez et al. (2011) reported that about 58% of South African soils exhibit <0.5% organic C, 38% contain about 0.5-2% organic C whilst about 4% contain about >2% organic C. Anthropogenic activities such as continuous cultivation, intensive tillage and crop residue removal have exacerbated soil organic matter deterioration and nutrients depletion of most of these soils (Henao and Baanante, 2006).

To manage poor soil fertility and improve crop production, inorganic fertilizers have been used over the years. Their use has achieved a high level of success through increased crop production. However, inorganic fertilizers have also imposed some limitations due to their high costs and lack of knowledge about proper application methods. Excessive use of mineral fertilizers has induced soil acidification, leading to death of soil microorganisms and declining soil quality. Therefore, the need to develop strategies for improving soil fertility without damaging the environment is increasing. Organic sources have been identified as safe alternatives to prevent continued use of inorganic fertilizers, due to their easy access and availability in the local environments. Common organic sources of nutrients in SSA include crop residues, green manures, animal manures and household wastes (Hossner and Juo, 1999). Organic materials aid in building up soil organic matter (SOM) which helps in recycling mineral nutrients thus improving soil fertility (Kumwenda et al., 1996). Crop residues organic resources that are available in abundance in our local environments. Their incorporation into the soil improve fertility through the release of essential soil nutrients (N, P, K) from plant tissues upon mineralization. They also aid in carbon sequestration through stabilisation of organic C upon decomposition.

Murungu et al. (2010) assessed decomposition and nutrient mineralization from winter-grown cover crops (i.e. grazing vetch, forage peas and oat). Their findings showed that oat had a slow decomposition rate compared to grazing vetch and forage peas therefore it was useful in improving soil organic matter. Maximum net mineral N and P were also higher in grazing vetch (84.8 mg N kg^{-1}; 3.6 mg P ha^{-1}) compared to forage peas (66.3 mg N kg^{-1}; 2.7 mg P ha^{-1}) and oats (13.7 mg N kg^{-1}; 2.8 mg P ha^{-1}). This was because oats had significantly higher C: N ratio and lignin content compared to grazing vetch and forage peas which caused its slow decomposition and low mineralization. In a similar study by Gezahegn et al. (2006), decomposition and nitrogen mineralization patterns of maize and soybean residues as well as their mixture was assessed. Results showed that soybean residues with lower C: N ratio and lignin content had higher decomposition rate, and mineralized more N (7.4 to 98.4 mg N kg-1 soil) compared to maize residues which induced N immobilization (-10.75 to -3.69 mg N kg^{-1} soil) due to higher C:N ratio.

Wheat (*Triticum aestivum*) and sorghum (*Sorghum bicolor*) are staple food crops to many in SSA. Wheat is the 2nd important grain crop after maize in South Africa (Gbetibouo and Hassan, 2005). It is mainly used for human consumption (making bread, biscuits, breakfast cereals, etc.) as well as animal feed (DAFF 2010). It is produced throughout South Africa, with Western Cape and Free State provinces being the largest producers. However, smaller quantities of wheat are also produced in KwaZulu-Natal, Eastern Cape, Gauteng and Mpumalanga provinces (DAFF, 2010). Similarly, sorghum is used as a source of carbohydrates for human consumption. It is mainly cultivated in dry areas due to its drought tolerance. The production of sorghum in South Africa varies from 13 000 ha to 150 000 ha per annum. The Free State, Mpumalanga and Limpopo province are the largest contributors of the area planted to sorghum (DAFF, 2010). Both wheat and sorghum have high biomass allocation to their roots and shoots therefore have potential for building up carbon stocks when their residues are incorporated into the soil. However, not much is known about the potential of their residues to sequester soil C or recycle essential nutrients (N and P) once incorporated into the soil. A gap also exists on variability of this potential among residues of different genotypes from the same plant.

11.2. Materials and methods

11.2.1. Soil Sampling

Soil used in the incubation was collected from an arable field located at Ukulinga Research Farm of the University of KwaZulu-Natal, South Africa. The farm is located at latitude 29.667’S and longitude 30.406’E at an elevation of 811 m above sea level. The soil in the study site was loam in texture classified as Chromic Luvisols (FAO, soil Classification). Soil samples were collected from a depth of 0-15 cm at random points using a soil auger, then mixed thoroughly to form a composite sample. This was then air-dried and ground to pass through a 2 mm sieve. A sub-sample of 0.5 kg was taken for determination of soil physical and chemical properties.

11.2.2. Collection, characterization and selection of crop residues

Residues of maize, sorghum and wheat were used in this study to assess their decomposition patterns and impact on soil properties. The residues were chosen from five varieties of each crop that proved to have high biomass production from a previous biomass allocation field study (Mathew et al., 2019). In addition to high biomass production, the residues were also chosen based on their C: N ratios and lignin contents. Thus, in the end, we had two varieties with high, two with low and one with medium C: N ratios for each crop. Ultimately, we used five genotypes of sorghum namely AS8, KZ5246, LP4403, Mamolokwane and 05-POTCH-138; five maize genotypes, i.e. R201, Mac-Medium Pearl, Nelson QPM CPSTN, PAN4P-228 and Shesha CPSTN; and five wheat genotypes namely LM70, BW152, LM75, BW162 and BW140. These were then used in residue incubation experiments that were conducted in a laboratory at the University of KwaZulu-Natal, as well as an additional litterbag study done on sorghum residues at Ukulinga university research farm both located in Pietermaritzburg, South Africa.

The crop residues were separated into roots and shoots, oven dried at 70°C for 48 hrs, then ground to pass through a 1 mm sieve before chemical analysis. Triplicate samples of plant residues were then taken and analysed for C, N and lignin concentrations. Total N and C was analysed using the LECO Trumac CNS auto analyser version 1.1x (LECO Corporation, 2012). The lignin content was determined using Van Soest methods (Van Soest et al., 1991). Total P was determined by digestion (sulphuric acid + hydrogen peroxide) with an electric hot plate without pH adjustment then using ascorbic acid (Okalebo, 1993).

11.2.3. Crop residue incubation

11.2.3.1 CO_2-emission determination

The residue incubation experiment was set up using a completely randomized design with 11 treatments replicated 3 times per crop. The root (RT) or shoot (ST) residues of five genotypes of either sorghum AS8, KZ5246, LP4403, Mamolokwane and 05-POTCH-138; maize genotypes R201, Mac-Medium Pearl, Nelson QPM CPSTN, PAN4P-228 and Shesha; or wheat genotypes LM70, BW152, LM75, BW162 and BW140 were used in this study. Ground root or shoot residues (0.25 g) of each genotype was mixed with 100 g of soil in a 100 ml plastic pot, slowly wetted to fill up 50% pore space and placed in a 500 ml airtight plastic pot. A vial containing 25 ml of 1 M NaOH solution was also placed inside the plastic pot to trap CO_2. The pots were closed so that they were airtight and incubated in the dark in a constant temperature room set at 25°C. The CO_2-C evolved was measured at 0, 7, 15, 23, 31, 39, 47, 55, 63, 77, 91, 105 and 120th day after incubation, where NaOH was titrated with 0.5M HCl, using phenolphthalein as an indicator, after precipitating carbonates with $BaCl_2$. The crop residues had different amounts of C in their tissues, therefore the results of CO_2-C emitted were normalised by expressing them as mg CO_2- C g^{-1} C added. The net CO_2-C emitted was obtained by calculating the differences in the values of the biomass treated soil and control, while cumulative mineralised CO_2-C was calculated as the sum of all previous measurements.

11.2.3.2. Mineral nitrogen and phosphorus determination

After titrating for CO_2 emission, the incubated treatments were analysed for NH_4^+ -N, NO_3 -N and extractable phosphorus. Samples (2.0 g) from each treatment were removed from the plastic pots at different incubation times and suspended in 20 ml of 2M KCl, shaken for 1 hr at 400pm for 10 minutes followed by filtration. The concentration of NH_4 $^+$-N and NO_3^--N in the extract were analysed using Thermo Scientific Gallery Discrete Autoanalyzer (Scientific Thermo Fisher 2014). Extractable P was assessed following Ambic-2 extraction and determined using Gallery Discrete Auto analyser. Different residues contained different concentrations of N and P in their tissues therefore, the results of N and P mineralized were normalised and represented as mg N g^{-1} N added and mg P g^{-1} P added. Net NH_4^+ -N, NO_3 -N and extractable P were obtained by difference between values of the control and the biomass treated soil. The net mineralized N was calculated as the sum of NH_4^+ -N and NO_3 -N concentrations released from that particular treatment after subtracting the control.

11.2.3.3. Microbial population counts of soil treated with maize residues

Microbial (bacteria and fungi) population counts were only done for the maize residues in a separate incubation experiment that ran for 120 days (with counts done at days 0, 7, 14, 28, 56, 84 and 120 of incubation). One gram of soil from each incubation period was transferred to a sterilised 50 ml bottle, then 9 ml of sterile distilled water was added and homogenized by shaking to make a dilution of 10^{-1}. About 1 ml of this dilution was then aseptically transferred to another 9 ml of sterile distilled water using a sterile pipette to make another dilution of 10^{-2}. Serial dilutions were then done up to the 10^{-5} dilution, after which a 1 ml aliquot each of dilutions 10^{-3} to 10^{-5} was transferred aseptically into a labelled sterile petri plate. One ml of penicillin, streptomycin, and tetracycline (50 µg/ml) were then added to the plates labelled for fungi. While molten nutrient agar and malt extract agar was poured into the plates for bacterial and fungal culture respectively. The plates were swirled before incubating at 37° C for 7 days, after which they were checked for growth of colonies which were then counted and colony properties (colour, texture and size) recorded.

11.2.4. Sorghum field litterbag experiment

Residues of five sorghum varieties (used in the incubation experiment above) were sorted into roots and shoots, air-dried, then chopped into 3-5 mm pieces before packing in 20 cm x 20 cm nylon mesh bags. The residue litter were spread evenly in each bag and closed by stapling, then each bag weighed before burying. This litterbag experiment was set up at the University of KwaZulu-Natal's Ukulinga farm in a wheat field, and was initiated two weeks after planting the wheat crop. This area has a mean maximum temperature of 26°C, and mean minimum temperature of 8.8°C (Swemmer et al., 2007 and Zeglin et al., 2007). It receives about 694 mm mean annual precipitation (Zeglin et al., 2007). The soil at this site was classified as Plintic Paleustalfs (FAO), that had clay content of 30.7%, organic matter content of 2.5%, with pH (KCl) of 4.8, (Chikuvire et al., 2018; Mutondwa et al., 2017).

The sorghum litterbag experiment was then set up as a completely randomised block design with 3 blocks, 5 sorghum varieties, 2 plant parts (root or shoot) and 9 sampling times (Day 0, 14, 28, 42, 56, 84, 112, 140 and 168), giving a total of 270 litterbags. A total of 10 wheat rows made up each block, with 45 cm inter-row and 10 cm intra-row spacing. Each block (5 x 5 m in size) with 1 m inter-block spacing, was divided into 2 subplots (to cater for the root-vs-shoot treatments) and had 9 sampling positions in each sub-plot to cater for the 9 sampling times. Each sampling positions was dug to a 10 cm depth below the surface, before burying 5 bags of either sorghum root or shoot representing the 5 varieties. The surrounding soil was used to bury and surface-level each sampling slot/position, to create contact between the litterbags and soil. Prior to planting, the land was tilled, disked then the crop supplemented with a basal fertilizer recommended for normal wheat growth. Additionally, supplementary irrigation was done using a sprinkler, while hand-hoeing was used for weed removal where necessary. At each sampling time, litterbags were recovered, then gently brushed to ensure that adhering soil was removed, before transporting to the laboratory for weighing to monitor weight loss. Thus, the measured weight of the litterbag after burial was expressed as a % of the initial weight of the litterbag prior to burial. Thereafter, litterbag samples were analysed for total C and N using the LECO auto-analyser. In addition, mineral P was analysed using AMBIC extraction before reading at 880 nm wavelength of spectrophotometry absorbance. The disappearance rate of litter was determined by getting a difference of weight of litter between two successive sampling intervals.

11.2.5. Data Analysis

A two-way analysis of variance (ANOVA) was done on all parameters studied, the means of parameters were grouped together for comparisons and differences were separated by least significant differences (LSD) using GenStat 18th edition (Payne et al., 2017). Significant differences were determined at $p = 0.05$. Bivariate correlations among CO_2-C emissions, N and P mineralization and biochemical properties of wheat and sorghum genotypes were done using the Spearman rank correlations procedure.

11.3. Results

11.3.1. Properties of soil and crop residues

The characteristics of soil used in the incubation study are shown in Table 11.1, while the properties of the crop residues are shown in Tables 45-47. Maize residue characterisation showed that among the root residues, Mac Medium Pearl had the highest C: N ratio and lignin content, while R201 roots had the lowest lignin content and lignin: N ratio (Table 11.2). In the case of the shoot residues, Mac Medium Pearl had the highest lignin content, C: N and lignin: N ratios; while Shesha CPSTN had among the lowest C: N and lignin: N values. Sorghum characterisation results showed that among the roots, KZ5246 had the highest while had the lowest C: N and Lignin: N ratios (Table 11.3). Genotype 05-POTCH-138shoots also had the lowest lignin content, C: N and lignin: N ratios; while AS8 shoots had the highest C: N and lignin: N ratios. In the case of wheat, roots of BW152 exhibited the highest C: N (89.4) and lignin: N (75) ratios, while shoots of BW140 and BW162 had lowest C: N and lignin: N ratios, and were the only genotypes with C: N ratio below 30 (Table 11.4). All the wheat genotypes exhibited C: P ratio < 200, except for BW140 roots that had the highest C: P ratio (217), while BW152 shoots had the lowest C: P ratio (98.6).

Table 11.1. Physico-chemical properties of the soil used in the residue incubation study

Soil Property	Values ±SD
Bulk density (g cm^{-3})	1.24±0.006
Sand (%)	30.0±0.061
Silt (%)	34.9±0.058
Clay (%)	24.4±0.015
Texture	Loam
pH (KCl)	4.73±0.015
Phosphorous (mg L^{-1})	11±0.051
Potassium (mg L^{-1})	114±0.168
Calcium (mg L^{-1})	1294±0.176
Magnesium (mg L^{-1})	389±0.156
Exchangeable Acidity (cmol L^{-1})	0.047±0.001
Zinc (mg L^{-1})	3.6±0.01
Manganese (mg L^{-1})	15±0.01
Copper (mg L^{-1})	7.2±0.202
Total carbon %	1.9±0.067
Total nitrogen %	0.17±0.01

Table 11.2. Biochemical properties of selected maize residues used in the incubation

Treatment	Genotype	Total C%	Total N%	Total P%	Lignin	C:N	Lignin: N
Root	Shesha CPSTN	17.2^{a}	0.41^{a}	4.51^{a}	7.03^{cd}	42.23^{bc}	17.18^{f}
Root	R201	36.84^{d}	0.79^{b}	5.13^{abc}	6.4^{c}	46.38^{c}	8.05^{c}
Root	Nelson QPM CPSTN	42.32^{g}	0.89^{bc}	6.03^{c}	7.65^{d}	47.70^{c}	8.63^{c}
Root	PAN4P-228	40.52^{f}	1.05^{c}	4.60^{ab}	9.58^{e}	38.48^{b}	9.12^{cd}
Root	Mac Medium Pearl	39.10^{e}	0.49^{a}	7.57^{d}	17.94^{g}	79.80^{d}	12.30^{e}
Shoot	Shesha CPSTN	41.92^{g}	1.46^{d}	4.52^{ab}	6.96^{cd}	28.71^{a}	5.18^{b}
Shoot	R201	40.32^{ef}	0.98^{c}	4.72^{ab}	2.79^{a}	41.32^{bc}	2.85^{a}
Shoot	Nelson QPM CPSTN	22.08^{b}	0.50^{a}	5.29^{abc}	2.95^{a}	44.32^{bc}	5.94^{b}
Shoot	PAN4P-228	28.29^{c}	1.35^{d}	5.61^{bc}	5.31^{b}	20.96^{a}	10.76^{de}
Shoot	Mac Medium Pearl	21.77^{b}	0.50^{a}	4.68^{ab}	16.31^{f}	43.22^{bc}	32.34^{g}

Note: different letters in each column show significant differences among treatments at ($p < 0.05$)

Table 11.3. Biochemical properties of selected sorghum residues used in the incubation

Treatment	Genotype	Total C%	Total N%	Lignin%	C: N	Lignin: N
Root	AS8	38.88^{abcd}	1.430^{bcdef}	28.77^{a}	27.22^{bcde}	20.10^{abcd}
Root	05-POTCH-138	41.31^{ef}	2.298^{def}	25.56^{a}	18.06^{ab}	11.15^{a}
Root	LP 4403	41.79^{bcdef}	1.991^{ef}	30.57^{a}	21.84^{abc}	16.21^{abc}
Root	KZ5246	36.02^{ab}	1.149^{b}	29.31^{a}	31.43^{cde}	25.64^{d}
Root	Mamolokwane	42.09^{def}	2.246^{f}	26.54^{a}	18.84^{ab}	11.80^{a}
Shoot	AS8	36.50^{abc}	1.237^{bcd}	29.91^{a}	29.75^{cde}	24.44^{cd}
Shoot	OS-POTC	41.35^{ef}	2.367^{def}	26.12^{a}	17.87^{ab}	11.40^{a}
Shoot	LP 4403	42.14^{bcdef}	1.985^{f}	29.05^{a}	21.26^{abc}	14.64^{ab}
Shoot	KZ5246	30.33^{abc}	1.357^{a}	32.40^{a}	$22.47a^{bcd}$	24.30^{cd}
Shoot	Mamolokwane	42.12^{cdef}	2.050^{f}	28.18^{a}	22.18^{abcd}	14.21^{ab}

Note: different letters in each column show significant differences among treatments at ($p < 0.05$)

Table 11.4. Biochemical properties of selected wheat residues used in the incubation

Treatment	Genotype	Total C%	Total N%	Total P %	Lignin%	C: N	C: P	Lignin: N
Root	LM70	23.9^{a}	0.45^{b}	0.20^{c}	28.4^{b}	53.0^{d}	119^{ab}	63.1^{f}
Root	BW162	33.1^{ab}	0.64^{c}	0.21^{d}	29.3^{b}	51.8^{d}	158^{f}	45.8^{e}
Root	BW152	30.4^{ab}	0.34^{a}	0.16^{b}	25.5^{b}	89.4^{g}	191^{g}	75.0^{g}
Root	LM75	33.4^{ab}	0.41^{ab}	0.17^{b}	29.8^{b}	81.5^{f}	197^{h}	72.6^{g}
Root	BW140	28.2^{ab}	0.83^{d}	0.13^{a}	30.1^{b}	34.0^{b}	217^{i}	36.3^{d}
Shoot	LM70	34.0^{b}	0.65^{c}	0.26^{ef}	15.2^{a}	52.4^{d}	131^{c}	23.4^{b}
Shoot	BW162	31.6^{b}	1.60^{e}	0.27^{f}	14.2^{a}	19.7^{a}	117^{ab}	8.85^{a}
Shoot	BW152	34.5^{b}	0.88^{d}	0.35^{h}	10.7^{a}	39.2^{c}	98.6^{a}	12.2^{a}
Shoot	LM75	36.2^{b}	0.57^{c}	0.30^{g}	17.4^{a}	63.5^{e}	121^{b}	30.4^{c}
Shoot	BW140	34.9^{b}	1.64^{e}	0.25^{e}	16.3^{a}	21.3^{a}	139^{d}	9.91^{a}

Note: different letters in each column show significant differences among treatments at ($p < 0.05$)

11.3.2. Variation in properties of soil incubated with different crop residues

11.3.2.1. Properties of incubated maize residues

There were significant differences in net carbon dioxide (CO_2) emission among maize treatments ($p < 0.001$, Table 11.5), with shoots (average 212.23 mg-CO_2/kg) generally having higher emissions than roots (average 94.5 mg-CO_2/kg). Among the roots, Mac Medium Pearl had the lowest (of all treatments) while Shesha CPSTN had the highest net CO_2 release. Whereas among the shoots, Mac Medium Pearl had the lowest while Nelson QPM CPSTN and R201 had the highest net CO_2 release. The net cumulative CO_2-C evolution also followed this same trend in the maize residue treatments. In terms of nitrate mineralisation, there were no significant differences in net NO_3^-N among the roots, while Shesha CPSTN shoots released the highest, and Mac Medium Pearl shoots had the lowest net NO_3^-N of all treatments (Table 11.5). Again, the shoots generally mineralised more net NO_3^-N than the root treatments. NH_4^+-N mineralisation showed roots of R201, (together with Mac Medium Pearl and Nelson QPM CPSTN) to release high net NH_4^+-N with PAN4P-228 releasing very low net NH_4^+-N (Table 11.5). In the shoots however, Shesha CPSTN had the highest (of all treatments) while Mac Medium Pearl had the lowest net NH_4^+-N. These mineralisation patterns resulted in Nelson QPM CPSTN having the highest net total mineral N, while PAN4P-228was lowest among the roots. Shesha shoots however released the highest net mineral N of all treatments, while Mac Medium Pearl shoots had very low overall net mineral N release (of all maize residue treatments). Mac Medium Pearl roots released the highest, while Shesha CPSTN shoots had the lowest net mineral P of all treatments. R201 shoots also had relatively high net mineral P compared to other shoot treatments (Table 11.5).

Table 11.5. Net mineral N, P and CO_2 emission in soil amended with different maize residues

Treatment	Genotype	CO_2 -C	Net NO_3^--N	Net NH_4^+-N	Net N	Net P
		mg/kg				
Root	Shesha CPSTN	133.90^{def}	5.035^{ab}	2.573^{bc}	7.608^{bc}	7.737^{b}
Root	R201	100.70^{bcd}	3.095^{ab}	5.119^{c}	8.214^{bc}	5.142^{ab}
Root	Nelson QPM CPSTN	95.9^{bc}	5.421^{ab}	4.506^{c}	9.927^{c}	6.675^{ab}
Root	PAN4P-228	84.4^{abc}	2.276^{ab}	1.348^{ab}	3.623^{b}	6.274^{ab}
Root	Mac-Med	57.6^{a}	3.269^{ab}	4.954^{c}	8.223^{bc}	11.487^{c}
Shoot	Shesha CPSTN	115.2^{cde}	13.139^{c}	8.874^{d}	22.013^{d}	3.832^{a}
Shoot	R201	153.2^{f}	5.565^{b}	4.001^{bc}	9.566^{bc}	7.982^{bc}
Shoot	Nelson QPM CPSTN	156.3^{f}	3.182^{ab}	4.440^{c}	7.622^{bc}	5.282^{ab}
Shoot	PAN4P-228	140.7^{ef}	6.049^{b}	3.498^{bc}	9.547^{bc}	4.860^{ab}
Shoot	Mac-Med	71.3^{ab}	1.342^{a}	1.350^{a}	2.692^{a}	5.028^{ab}
LSD		23.23	4.245	1.735	3.811	2.282

LSD=least significant difference at 5%; figures with same letter within a column are not different at 5% lsd.

11.3.2.2. Properties of incubated sorghum residues

Significant net CO_2 emissions were also observed among the sorghum residues ($p < 0.001$, Table 11.6). Thus, 05-POTCH-138 released the highest while KZ5246 and AS8 had lower net CO_2 emissions in both roots and shoots. This same trend was replicated in the net cumulative CO_2 emissions. Net ammonium, nitrate, and total mineral N of sorghum residues also followed the same trend with KZ5246 and AS8 showing the lowest, while 05-POTCH-138 had the highest N parameters in both roots and shoots (Table 11.6). An opposite trend was however observed for Net mineral P, with AS8 releasing the highest while 05-POTCH-138 released the lowest net P in both roots and shoots (Table 11.6). KZ5246 roots also had very low net P release.

Table 11.6. Net mineral N, P and CO$_2$ emission in soil amended with different sorghum residues

Treatment	Genotype	Net CO$_2$-C	Net Cum CO$_2$-C	Net NH$_4^+$-N	Net NO$_3^-$ -N	Net tot min N	Net min P
		……………………………………………mg kg^{-1} soil…………………………………………					
Root	AS8	38.97^{a}	305.6^{a}	4.91^{ab}	3.33^{ab}	8.25^{b}	6.34^{b}
Root	KZ5246	38.85^{a}	319.5^{a}	2.67^{a}	1.92^{a}	4.59^{a}	4.42^{a}
Root	LP4403	61.99^{abc}	487.5^{b}	11.82^{cd}	9.15^{bcd}	20.97^{bcd}	5.89^{ab}
Root	05-POTCH-138	92.97^{d}	734.5^{c}	14.67^{de}	11.01^{cd}	25.68^{d}	4.34^{a}
Root	Mamolokwane	81.52^{cd}	645.4^{c}	12.07^{cde}	8.02^{abcd}	20.09^{bcd}	4.75^{ab}
Shoot	AS8	47.23^{ab}	378.6^{a}	8.38^{bc}	6.47^{abcd}	14.85^{abc}	5.74^{ab}
Shoot	KZ5246	35.53^{a}	282.5^{a}	6.03^{ab}	4.86^{abc}	10.89^{ab}	4.60^{ab}
Shoot	LP4403	63.90^{abcd}	509.5^{b}	12.38^{cde}	10.56^{cd}	22.94^{cd}	5.28^{ab}
Shoot	05-POTCH-138	93.62^{d}	771.5^{d}	17.12^{e}	12.56^{d}	29.68^{d}	4.34^{a}
Shoot	Mamolokwane	75.04^{bcd}	649.7^{c}	11.92^{cd}	10.88^{cd}	22.80^{cd}	4.76^{ab}
	LSD	5.87	108.3	3.18	4.04	6.56	1.17

LSD=least significant difference at 5%; figures with same letter within a column are not different at 5% lsd.

11.3.2.3. Properties of incubated wheat residues

Table 11.7 shows incubation variables measured for wheat, and significant differences were also noted among treatments ($p < 0.001$). Among the roots, LM70 released the lowest net CO_2, while the other varieties did not significantly differ. In the shoots, however, LM70 and 75 had the lowest while BW162 and 140 had the highest net CO_2 emissions. The cumulative emissions also followed more or less the same trend. Nitrate, ammonium and total N mineralisation saw BW152 roots giving the lowest (of all treatments), while BW140 had the highest net NH_4^+-N, NO_3^- -N and total N release among the roots (Table 11.7). In the shoots, LM75 (as well as LM70) also released the lowest while BW162 and 140 mineralised the highest net NH_4^+-N, NO_3^--N and total N of all treatments. P mineralisation saw BW140 having the lowest (of all treatments) while BW162 had the highest net mineral P among the roots; and BW140 also had the lowest while BW152 released the highest (of all treatments) net mineral P among the shoots.

Table 11.7. Net mineral N, P and CO_2 emission in soil amended with different wheat residues

Treatment	Genotype	Net CO_2-C	Net Cum CO_2-C	Net NH_4^+-N	Net NO_3^- -N	Net tot min N	Net min P
		………………………………	….mg kg^{-1} soil……	………………………	………………………	…...	
RT	LM70	48.8a	383.3a	10.53b	12.13b	22.66b	1.96bc
RT	LM75	71.8b	526.7a	8.21b	13.26b	21.47b	1.77bc
RT	BW152	74.2b	488.0a	3.56a	4.83a	8.39a	1.50ab
RT	BW162	86.4b	644.6ab	10.47b	12.56b	23.02b	2.14bcd
RT	BW140	86.6b	602.6ab	17.76d	16.74bcd	34.50d	0.82a
ST	LM70	141.5c	1052.1c	11.84bc	14.14bcd	25.98bc	2.87de
ST	LM75	135.6c	955.9bc	9.78b	13.81bc	23.59bc	3.15ef
ST	BW152	189.3d	1318.3cd	15.41cd	15.81bcd	31.22cd	3.69f
ST	BW162	218.7e	1616.4d	18.58d	20.22d	38.80d	2.91def
ST	BW140	223.8e	1641.4d	18.30d	19.60cd	37.90d	2.47cde
	LSD	13.37	240.3	2.384	3.836	5.056	0.5004

LSD=least significant difference at 5%; figures with same letter within a column are not different at 5% lsd.

11.3.3. Comparison of mineralisation patterns among different crop residues

A comparison of net cumulative CO_2 emissions of all crops saw shoots releasing more CO_2 than roots in most of crops (Fig 11.1). Again, average net cumulative CO_2 emissions saw wheat having the highest emission (especially among the shoots), while maize had the lowest emissions in the roots, and sorghum had moderate overall emissions. Again, as with CO_2 emissions, shoot residues of all crops mineralised more net total mineral N than roots, thereby making shoot residues better sources of mineral N than roots (Fig 11.2). A comparison of crop residues however showed net mineral N release patterns of the order wheat > sorghum > maize in both roots and shoots ($p < 0.001$), making wheat residues the best, while maize residues were poorer sources of mineral N (Fig 11.2). The order was however different for net mineral P release with maize residues being better sources of P than wheat residues, while sorghum residues were moderate. Roots also released slightly higher mineral P than shoots for maize and sorghum, while the opposite was true for wheat residues.

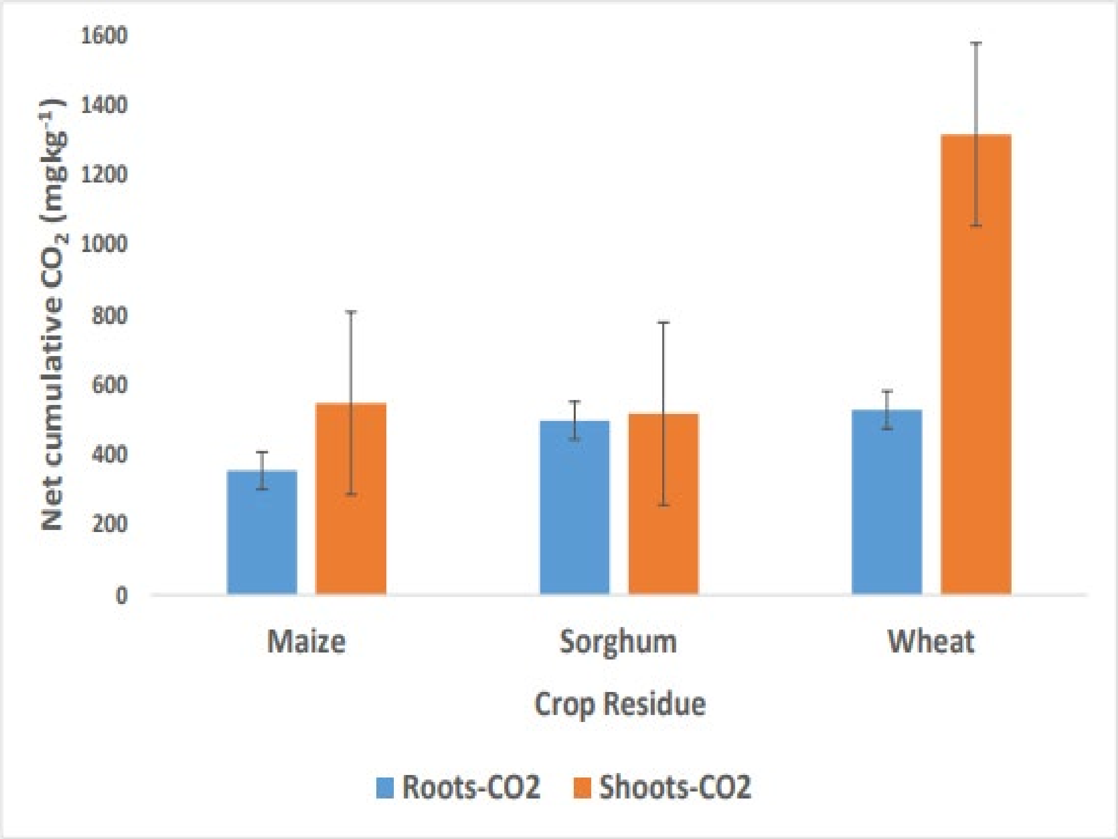


Figure 11.1. Variation in net cumulative CO_2 emissions of incubated crop residues

Figure 11.2. Variation in N and P mineralisation of incubated crop residues

11.3.4. Variation in CO_2 emission among different crop residues

Net carbon dioxide (CO_2) emission varied among the maize varieties and between plant parts throughout the incubation period (Fig 11.3). Maize shoots (212.23 mg-CO_2/kg) generally produced higher CO_2 emissions than roots (94.5 mg-CO_2/kg). The root residues for all maize cultivars evolved the highest CO_2-C on day 28 and while shoots emitted the most CO_2-C on day 42. The shoots for cultivar Nelson QPM CPSTN and roots of Shesha CPSTN had the highest CO_2-C evolution on days 42 and 28, respectively. While the variety Mac Medium Pearl generally had the lowest CO_2-C emissions from both roots and shoots throughout the incubation.

Similarly, there were variations in CO_2 emission among the sorghum varieties and between their different parts (Fig 11.4). The net CO_2 emitted was higher for shoots compared to root residues throughout the incubation period. There was a sharp increase in CO_2 emission up to about 3 weeks after incubation followed by a steady decline thereafter for both shoots and root residues. Wheat genotypes also varied widely in CO_2 emission, with shoots also releasing more CO_2 than roots treatments (Fig 11.5). A comparison of all 10 treatments showed that root tissues of LM70 emitted the lowest net CO_2, while shoot tissues of genotypes BW162 and BW140 emitted the highest.

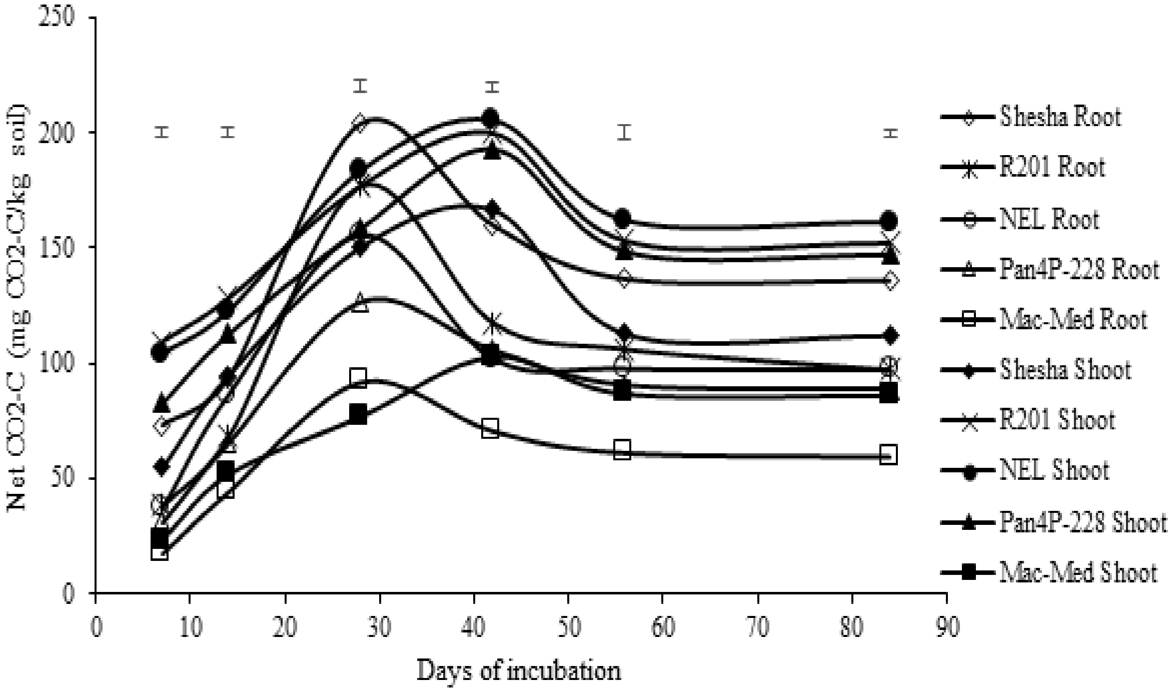


Figure 11.3. Carbon dioxide emission from soil amended with different cultivars of maize residues (bars represent $LSD_{0.05}$)

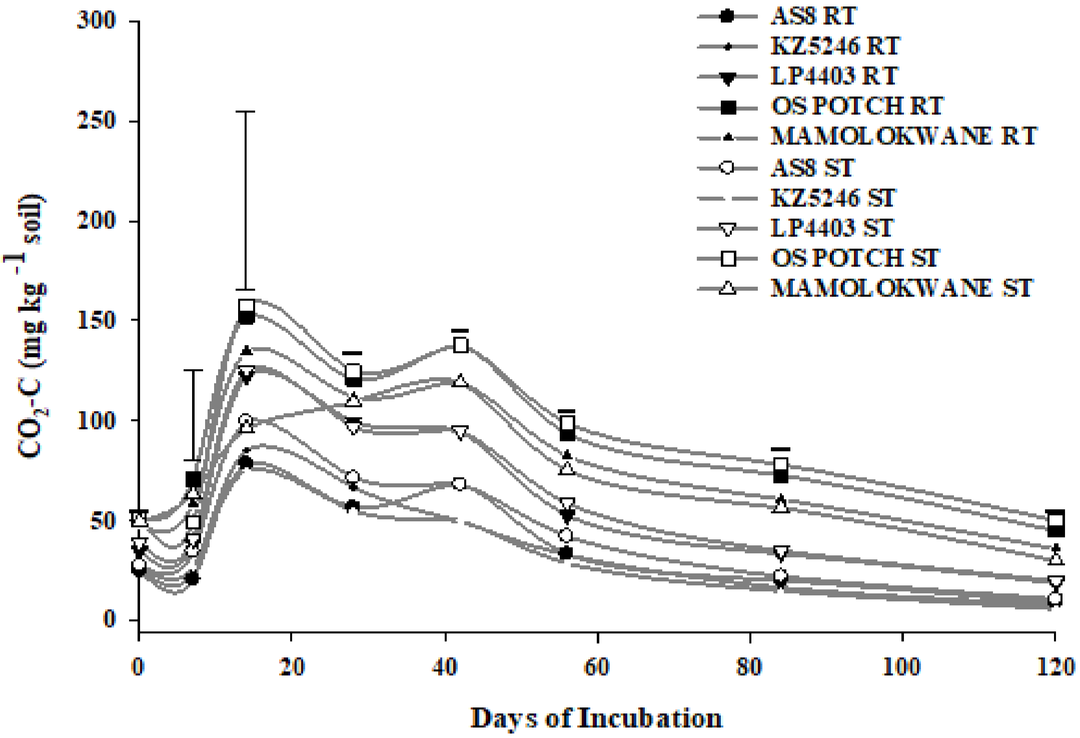


Figure 11.4. Net CO_2-C emission from roots (RT) and shoots (ST) of different sorghum varieties incubated over a 120-day period. Bars represent the LSD ($p<0.001$)

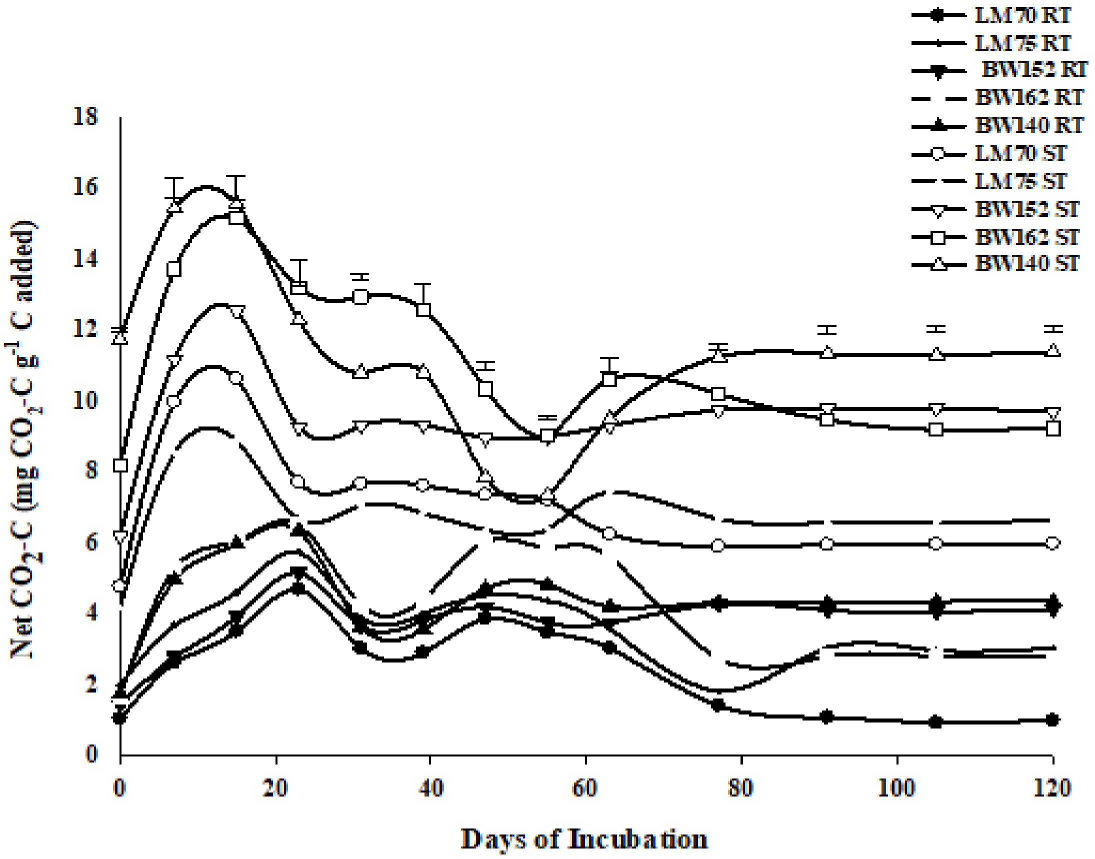


Figure 11.5. Net CO_2-C emission from roots (RT) and shoots (ST) of different wheat varieties incubated over a 120 days period. Bars represent the LSD ($p<0.001$)

The shoots mineralized more N than root residues for all the crops (Fig 11.6). The net mineral N averaged 19.6 soil and 28.78 mg N kg^{-1} soil for wheat roots and shoots, respectively, while it was 15.91 and 20.23 mg N kg^{-1} soil for sorghum roots and shoots. The shoot residues of maize mineralized 10.29 mg N Kg^{-1} compared to 7.52 mg N Kg^{-1} mineralized by roots (Fig 11.7). In general, when comparing the three crops, wheat mineralized higher net mineral N compared to sorghum and maize residues (Fig 11.8).

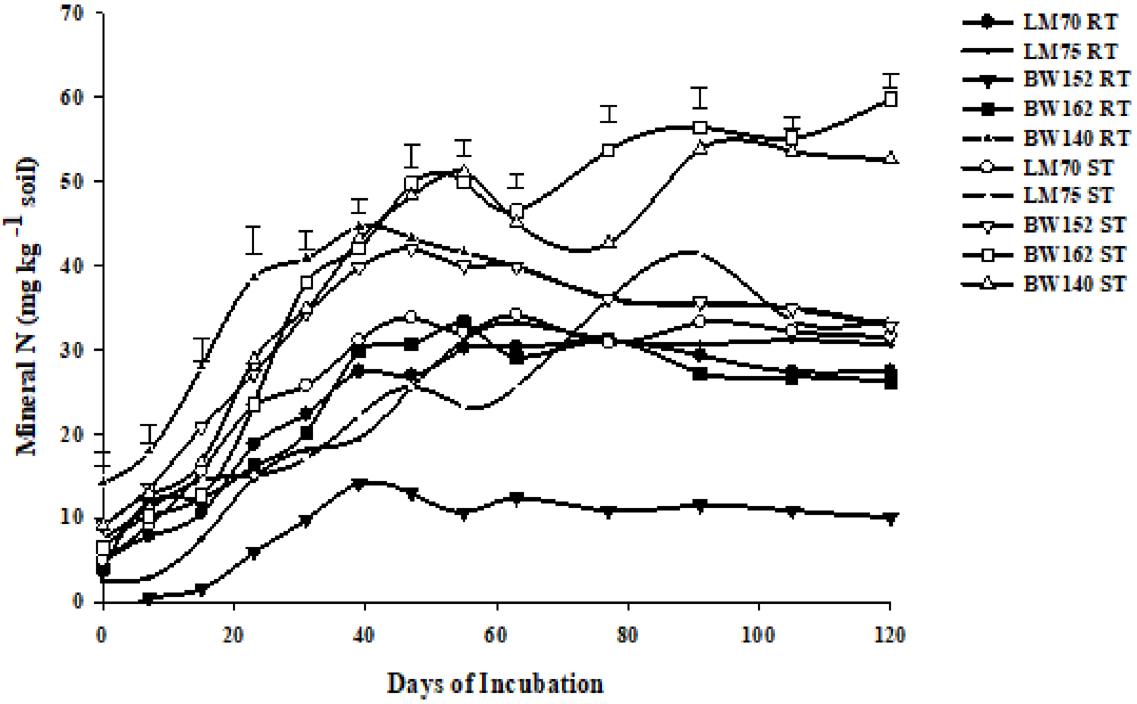


Figure 11.6. Net N mineralized (NH_4^+-N $+NO_3^-$-N) from roots (RT) and shoots (ST) of different wheat varieties incubated over a 120-day period. Bars represent the LSD ($p<0.001$)

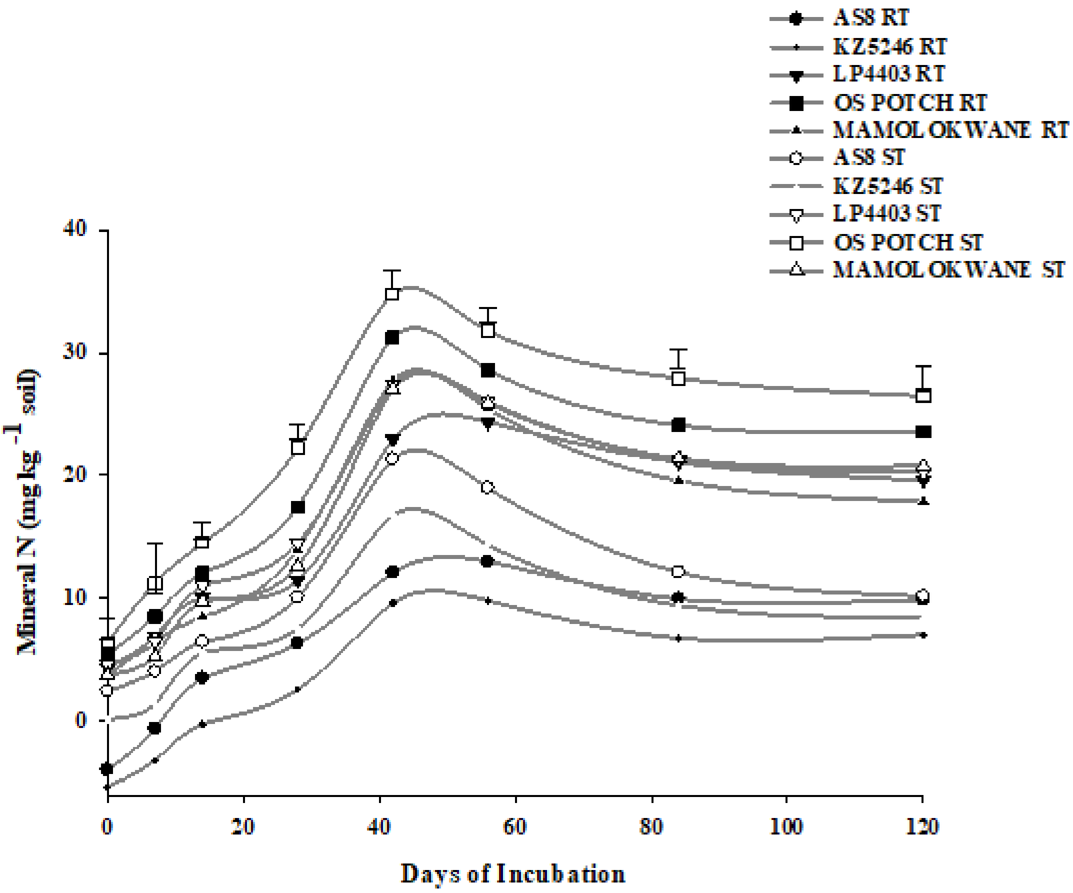


Figure 11.7. Net N mineralized (NH_4^+-N +NO_3^--N) from roots (RT) and shoots (ST) of different sorghum varieties incubated over a 120-day period. Bars represent the LSD ($p<0.001$)

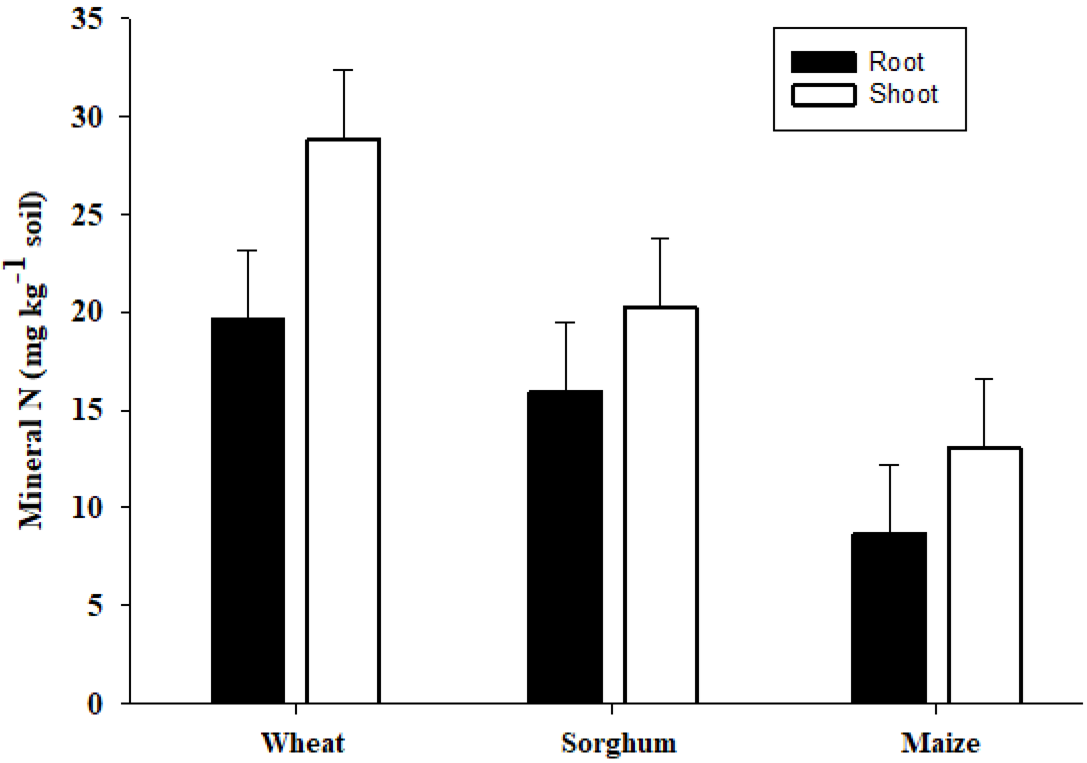


Figure 11.8. Net N mineralized (NH_4^+-N +NO_3^--N) from roots and shoots of different wheat and sorghum varieties incubated over a 120-day period. Bars represent the LSD ($p<0.001$)

11.3.5. Relationship between CO_2-C emissions and mineralized nutrients with residue quality

Table 11.8 shows that significant positive correlation existed between residue total C and CO_2-C emitted (r= 0.58, $p<0.05$), NH_4^+-N (r= 0.24, $p<0.05$), net total N mineralized (r= 0.33, $p<0.05$) as well as total P (r= 0.63, $p<0.001$). C: N correlated positively with C: P (r= 0.22, $p<0.05$), lignin (r= 0.28, $p<0.05$), lignin: N (r= 0.82, $p<0.001$); while C: P positively correlated with lignin (r= 0.22, $p<0.05$) and lignin: N (r=0.36, $p<0.05$). Lignin positively correlated with lignin: N (r=0.82, $p<0.001$); while CO_2-C correlated positively with NH_4^+-N (r= 0.42, $p<0.05$), NO_3^--N (r=0.63, $p<0.001$), total N (r=0.52, $p<0.05$), net total N mineralized (r=0.43, $p<0.05$) and total P (r= 0.72, $p<0.001$). NH_4^+-N positively correlated with NO_3^--N (r= 0.92, $p<0.001$), total N (r=0.51, $p<0.05$), net total N mineralized (r=0.87, $p<0.001$) and total P (r=0.44, $p<0.05$) while NO_3^--N correlated positively with total N (r= 0.35, $p<0.05$), net total N mineralized (r= 0.76, $p<0.001$) and total P (r= 0.43, $p<0.05$), and net total N mineralized positively correlated with total P (r= 0.56, $p<0.05$). Total N positively correlated with net total N mineralized ($r = 0.68$, $p<0.05$) and total P ($r=0.56$, $p<0.05$). Lastly, a positive significant correlation was observed between net extractable P mineralized and total P ($r = 0.73$, $p<0.05$). On the other hand, significant negative correlations were observed between total C and C: P ($r = -0.52$, $p<0.05$), lignin ($r = -0.36$, $p<0.05$), lignin: N ($r = -0.42$, $p<0.05$). C: N correlated negatively with NH_4^+-N ($r = -0.46$, $p<0.05$), NO_3^--N ($r = -0.52$, $p<0.05$), total N ($r = -0.87$, $p<0.001$), net total N mineralized ($r = -0.69$, $p<0.05$) and total P ($r = -0.32$, $p<0.05$). C: P negatively correlated with CO_2-C ($r = -0.52$, $p<0.05$), NO_3^-- ($r = -0.31$, $p<0.05$), total N ($r = -0.57$, $p<0.05$), net total N mineralized ($r=-0.43$, $p<0.05$), net extractable P mineralized ($r = -0.87$, $p<0.05$) and total P ($r = -0.84$, $p<0.001$). Lignin negatively correlated with CO_2-C ($r = -0.65$. $p<0.05$), total N ($r = -0.35$, $p<0.05$) and total P ($r = -0.24$, $p<0.05$) while lignin: N correlated negatively with CO_2-C ($r = -0.63$, $p<0.001$), NH_4^+-N ($r = -0.36$, $p<0.05$), total N ($r = -0.92$, $p<0.001$) and total P ($r = -0.62$, $p<0.001$) (Table 11.8).

Table 11.8. Pearson's linear correlation coefficients showing bivariate correlations between selected biochemical properties of wheat and sorghum residues

	TC	C: N	C: P	Lignin	Lignin: N	CO_2-C	NH_4^+-N	NO_3^--	TN	N Min	P Min	TP
C	1.00											
C:N	0.28	1.00										
C:P	-0.52*	0.22*	1.00									
Lignin	-0.36*	0.28*	0.22*	1.00								
Lignin: N	-0.42*	0.82**	0.36*	0.82**	1.00							
CO_2-C	0.58*	-0.35	-0.52*	-0.65*	-0.63**	1.00						
NH_4^+-N	0.24*	-0.46*	-0.23	-0.14	-0.36*	0.42*	1.00					
NO_3^--N	0.19	-0.52*	-0.31*	-0.08	-0.19	0.63**	0.92**	1.00				
N_%	0.15	-0.87**	-0.57*	-0.35*	-0.92**	0.52*	0.51*	0.35*	1.00			
N Min	0.33*	-0.69*	-0.43*	0.06	-0.19	0.43*	0.87**	0.76**	0.68*	1.00		
P Min	0.22	-0.15	-0.87*	0.12	-0.11	0.06	-0.03	-0.09	0.31	-0.06	1.00	
TP	0.63**	-0.32*	-0.84**	-0.24*	-0.62**	0.72**	0.44*	0.43*	0.56*	0.48*	0.73*	1.00

TC=total carbon, C:N=carbon:nitrogen ratio, C:P=carbon: phosphorous ratio, CO2=carbon dioxide, C=carbon, NH^{4+}=ammonium, NO^{3-}=nitrate, TN=total nitrogen, N-Min=mineralized nitrogen, P-Min=mineralized phosphorous, TP=total phosphorous

11.3.6. Microbial population counts of soil incubated with maize residues

An additional experiment to quantify microbial populations of incubated maize residues showed that there were more bacteria (range of 1.1-1 111.8 x 10^3 CFU/ g moist soil) than fungi (range of 1.1-308.3 x 10^3 CFU/ g moist soil) in all residue treatments; with the control treatments also showing lower counts for both bacteria (≤126 x 10^3 CFU/ g moist soil) and fungi (< 80 x 10^3 CFU/ g moist soil) than most residue treatments (Table 12.9). Again, both bacterial and fungal populations were highest at day 0, and lowest at day 120 (i.e. generally decreased as time progressed, though there was a slight increase in populations at day 84 than 56 for both organisms). However, there were no clear differences in populations of both organisms between root and shoot residue treatments. More specific trends of bacterial populations of residue genotypes showed that Mac Medium Pearl residues (and sometimes PAN4P-228) of both roots (126.3 – 509.9 x 10^3 CFU/ g moist soil) and shoots (174 – 482.4 x 10^3 CFU/ g moist soil) resulted in the lowest; while R201 (and sometimes Nelson QPM CPSTN) residues gave the highest bacterial populations (roots = 279.1 – 919.1 x 10^3 CFU/ g moist soil; shoots = 355 – 1 111.8 x 10^3 CFU/ g moist soil) from day 0 to day 28. At day 56 however, R201 roots gave the highest bacterial populations (161.4 x 10^3 CFU/ g moist soil) while the rest of the treatments did not significantly differ. This trend changed as from day 84 to 120 with Nelson QPM CPSTN, and sometimes PAN4P-228 giving lower, while Mac Medium Pearl and sometimes Shesha CPSTN residues gave the highest bacterial populations in both roots and shoots.

No significant differences in fungal populations however were recorded at days 0 and 14 (Table 11.9). On day 7 however, Shesha CPSTN (285.1 x 10^3 CFU/g moist soil) had the highest fungal populations in roots while the other treatments did not significantly differ. In the shoot treatments, Mac Medium Pearl had the lowest (84.2 x 10^3 CFU g moist soil /), while PAN4P-228 (as well as Shesha CPSTN) had the highest (261.6 x 10^3 CFU/ g moist soil) fungal populations. Days 28-120 saw either Mac Medium Pearl, PAN4P-228or Nelson QPM CPSTN having the lowest while mostly R201 and Shesha CPSTN had higher fungal populations in both residue treatments.

Table 11.9. Microbial population counts in a maize residue incubation experiment

	Days of Incubation						
Treatment	0	7	14	28	56	84	120
			Bacterial Counts (CFU x 10^{3})/ g moist soil				
Shesha Root	712^{c}	515.6^{bc}	464^{c}	180^{a}	51.6^{a}	201.7^{b}	18.5^{ba}
R201 Root	919.1^{dc}	803.1^{c}	396.4^{cb}	279.1^{b}	161.4^{b}	160.8^{ba}	22.9^{ba}
NEL Root	946.8^{dc}	779.6^{c}	385^{bc}	251.8^{b}	26.7^{a}	144.3^{a}	5.3^{a}
Pan4P-228 Root	532.2^{b}	384.4^{b}	393.6^{cb}	241.1^{b}	9.8^{a}	161.9^{ab}	1.1^{a}
Mac med Root	509.9^{b}	399.7^{b}	342.4^{b}	126.3^{a}	17.7^{a}	205.2^{b}	25.3^{ba}
Shesha Shoot	618.1^{cb}	463^{b}	401.7^{cb}	187^{ab}	72.3^{a}	243^{b}	21.2^{ba}
R201 Shoot	1 111.8^{d}	797^{c}	422.7^{c}	355^{b}	38^{a}	154.6^{a}	30.4^{b}
NEL Shoot	1 025.3^{d}	655.2^{c}	387.9^{b}	289.3^{b}	46^{a}	176.4^{ba}	17.5^{ba}
Pan4P-228 Shoot	603^{cb}	434.2^{b}	402.7^{cb}	144.1^{a}	33.3^{a}	231.6^{b}	2.9^{a}
Mac med Shoot	482.4^{b}	410.7^{b}	308.3^{b}	174^{a}	15.9^{a}	103.6^{a}	34.1^{b}
Control	126^{a}	125.6^{a}	122.3^{a}	40.6^{a}	15.7^{a}	71^{a}	1.5^{a}
LSD($p<0.05$)	281.8	161.7	97.5	166.6	78.6	113.6	24.5
			Fungal Counts (x 10^{3}) g moist soil				
Shesha Root	175^{a}	285.1^{b}	158.9^{a}	96.9^{b}	36.2^{ba}	61.4^{ab}	29.2^{b}
R201 Root	233.6^{a}	184.2^{ba}	128.7^{a}	101.7^{b}	23.3^{ab}	69^{ab}	32.2^{b}
NEL Root	128.6^{a}	119.8^{a}	182.7^{a}	66.1^{ba}	12.4^{ab}	11.2^{a}	2.3^{a}
Pan4P-228 Root	166.8^{a}	221.3^{ba}	167^{a}	48.3^{ab}	1.2^{a}	16.2^{a}	3.3^{a}
Mac med Root	172.2^{a}	157.9^{a}	154.3^{a}	34.6^{ab}	26.7^{ab}	45^{ab}	3.1^{a}
Shesha Shoot	51.8^{a}	234.9^{b}	131.1^{a}	61.7^{ba}	27.8^{ab}	258.1^{b}	42.6^{c}
R201 Shoot	177.6^{a}	185.6^{ab}	212^{a}	97.3^{b}	22.9^{ab}	39.3^{a}	3.1^{a}
NEL Shoot	308.3^{a}	178.7^{ba}	222.2^{a}	165.1^{b}	55.7^{b}	13.6^{a}	3.7^{a}
Pan4P-228 Shoot	149^{a}	261.6^{b}	116.4^{a}	59.1^{ab}	1.1^{a}	81.9^{ab}	2.3^{a}
Mac med Shoot	160.3^{a}	84.2^{a}	87.4^{a}	51.1^{ab}	26.7^{ab}	12.3^{a}	2.4^{a}
Control	49^{a}	73.9^{a}	60.7^{a}	13.4^{a}	13.3^{a}	1.2^{a}	0.1^{a}
LSD($p<0.05$)	202.7	138.9	169.6	81.1	53.5	205.3	8.6

LSD=least significant difference at 5%; figures with same letter within a column are not different at 5% lsd.

11.3.7. Sorghum litterbag experiment

11.3.7.1 The effect of decomposition on dry weight of different sorghum residues

The decomposition of different sorghum residues, as measured by percentage of weight remaining was influenced by crop genotype, and also varied with plant part and residue burial time (Fig 11.9). Thus, the remaining dry weight (DW) progressively decreased with increase in burial time throughout the duration of the experiment ($p < 0.01$). Hence between days 0 to 28, all treatments had more than 50% remaining DW, but by day 168, most treatments had less than 20% remaining DW. At day 14, AS8 root showed the highest remaining DW (97%), followed by KZ5642 root (84%), while AS8 and KZ5642 also had higher remaining DW among the shoots. All root treatments generally had higher DW than their respective shoot treatments (Fig 11.9). This was more or less the same trend for the rest of the time periods, with AS8 root generally having the highest DW throughout the duration of the experiment.

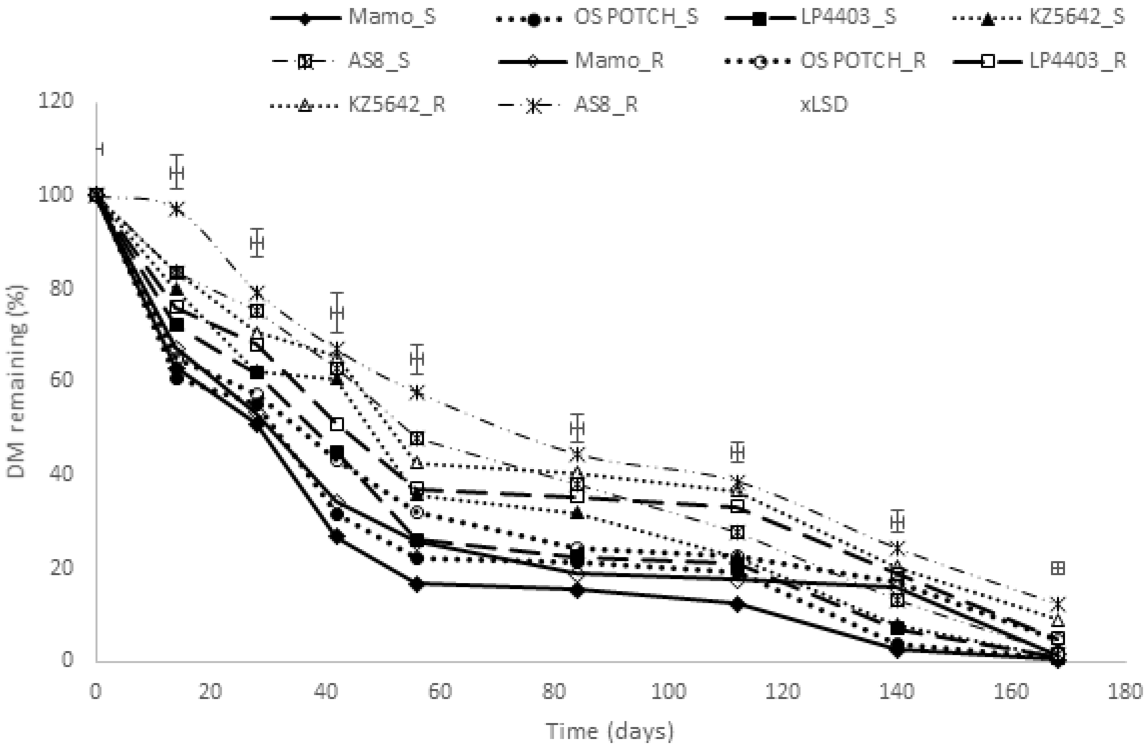


Figure 11.9. Weight remaining (%) of different sorghum residues. (Mamo is Mamolokwane: “R” is root and “S” is shoot)

11.3.7.2. The impact of chemical composition on decomposition rates of sorghum residues

The initial chemical composition of sorghum residues significantly differed, among the root treatments, Mamolokwane (similar to 05-POTCH-138 root) had the lowest initial C: N ratio (18.8), while KZ5642 root had the highest (31) followed by AS8 root (27). In the shoot treatments however, AS8 had the highest (29.8) while 05-POTCH-138 had the lowest (17.9), initial C: N ratio (Figure 12.10). There was a sharp increase in C:N ratio at day 14, with AS8 shoot having the highest ratio (53), (which was significantly higher than its root (50.7), while 05-POTCH-138 shoot showed the lowest ratio (34) followed by Mamolokwane shoot (40) (Fig 11.10). Thereafter, a decreasing trend in C: N ratio was observed with time, with AS8 root showing the greatest decrease in C: N ratio compared to its shoot and other treatments, at day 112 (11.6) to 4 at day 140, while Mamolokwane shoot showed higher C: N ratio compared to other treatments at day 140 (21.8). All treatments were observed to have a C: N ratio of less than 30 from day 84.

The lignin: N ratio also varied significantly with different sorghum varieties and time. Thus, KZ5246 root had significantly higher initial lignin: N ratio (25.6), followed by AS8 shoot and root (24 and 20) respectively, while other sorghum treatments had a ratio of less than 15, at day zero (Fig 11.11). A peak in lignin: N ratio was observed at day 14, with KZ5642 and AS8 roots showing significantly high values (39.6 and 38.8) compare to their shoots and other sorghum treatments. This was followed by a decrease in lignin: N ratio observed at day 28 for most treatments, followed by a more acute decrease after day 42 that led to lignin: N ratio of less than 15 by day 56. The ratio then fluctuated thereafter, with high maximum lignin: N ratios (of 25 and 24) for 05-POTCH-138 and Mamolokwane roots respectively at day 112, while most of the other treatments had lower ratios. At the end of the experiment (day 168), LP4403 shoot showed the lowest lignin: N ratio (7) compared to its root and other treatments, while 05-POTCH-138 root and shoot (27) followed by AS8 shoot (24) had the highest ratios.

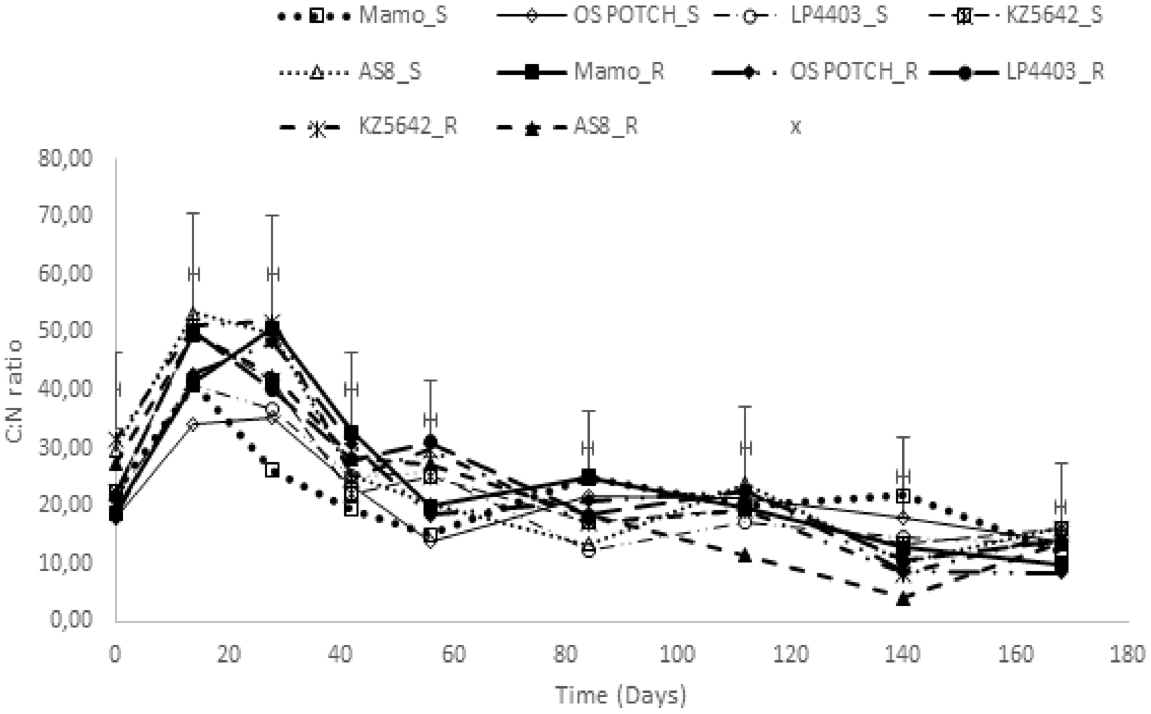


Figure 11.10. Change in C: N ratio of buried sorghum residues. (Mamo is Mamolokwane: "R" is root and "S" is shoot)

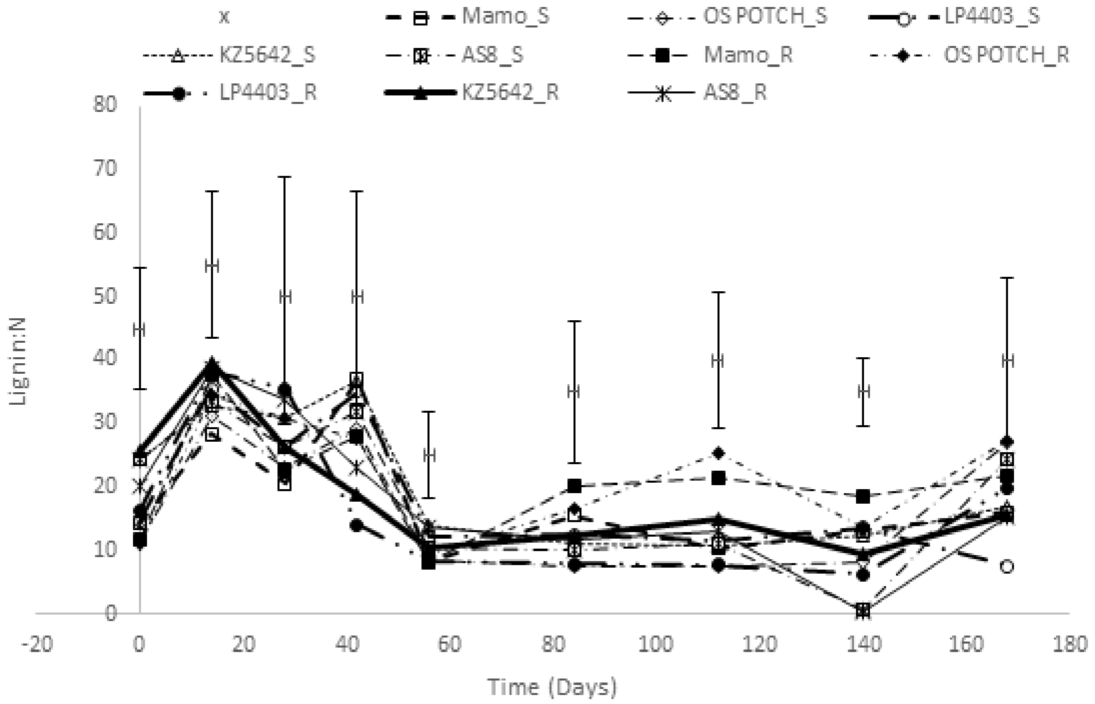


Figure 11.11. Change in lignin: N ratio of different sorghum residues. (Mamo is Mamolokwane: "R" is root and "S" is shoot)

11.3.7.3. The effect of decomposition on extractable P of different sorghum residues

Minimal variation in extractable P was observed with different sorghum varieties and parts throughout the different time periods (Table 11.12). The initial extractable P was a maximum of 3.3 mg/kg soil for LP4033 root (which was similar to LP4033 shoot, KZ5642 shoot and AS8 root), while Mamolokwane root showed the least initial P of 1.92 mg/kg soil, which did not differ from the rest of the other treatments. At day 14, LP4033 shoot (similar to LP4033 root, Mamolokwane shoot and 05-POTCH-138 root) had the highest extractable P than all the other treatments that had lower P. There was an increasing trend of extractable P with time as from day 28 to 56. Thus, at day 28, Mamolokwane shoot (together with 05-POTCH-138 shoot and LP4033 root) showed significantly higher P (9.48 mg/kg soil) compared to it root (5.5 mg/kg soil), while AS8 shoot showed the lowest extractable P (4.6 mg/kg soil), which was also lower than its root (7.1 mg/kg soil). A more or less similar trend of higher P in Mamolokwane and 05-POTCH-138 shoots was observed at day 56, while no significant differences in P were noted between treatments at days 42, 84, 140 and 168. AS8 root had the highest P at day 112 while the other treatments did not significantly differ.

Table 11.10. Change in extractable P (mg/kg soil) of different sorghum residues with time

Day	Mamo_S	05-POTCH-138	LP4403_S	KZ5642_S	AS8_S	Mamo_R	05-POTCH-138	LP4403_R	KZ5642_R	AS8_R	lsd
0	2.4^{a}	2.2^{a}	3.0^{b}	2.95^{b}	2.9^{a}	1.9^{a}	2.3^{a}	3.3^{b}	2.9^{a}	2.9^{b}	0,98
14	5.2^{b}	4.0^{a}	5.3^{b}	3.4^{a}	3.6^{a}	4.8^{a}	5.3^{b}	5.2^{b}	4.3^{a}	4.2^{a}	1.5
28	9.5^{c}	8.0^{bc}	6.8^{a}	5.2^{a}	4.6^{a}	5.5^{a}	8.0^{bc}	9.2^{c}	6.8^{b}	7.1^{b}	1.9
42	11.0^{a}	10.5^{a}	9.6^{a}	6.9^{a}	6.4^{a}	8.6^{a}	7.6^{a}	6.3^{a}	6.1^{a}	5.5^{a}	5
56	11.2^{b}	10.4^{b}	8.8^{a}	8.8^{a}	8.8^{a}	6.9^{a}	8.5^{a}	6.6^{a}	9.7^{a}	4.7^{a}	5.3
84	7.4^{a}	10.0^{a}	10.8^{a}	8.6^{a}	10.6^{a}	8.8^{a}	8.8^{a}	6.6^{a}	7.0^{a}	11.2^{a}	5.1
112	5.8^{a}	7.3^{a}	8.2^{a}	9.1^{a}	11.0^{a}	9.1^{a}	5.5^{a}	6.9^{a}	11.0^{ba}	11.9^{b}	5.7
140	5.2^{a}	6.4^{a}	6.0^{a}	7.5^{a}	6.4^{a}	5.3^{a}	5.0^{a}	6.6^{a}	6.2^{a}	3.3^{a}	8.5
168	6.4^{a}	6.4^{a}	9.6^{a}	7.6^{a}	6.9^{a}	5.5^{a}	8.6^{a}	10.5^{a}	6.1^{a}	11.0^{a}	6.1

Mamo is Mamolokwane: “R” is root and “S” is shoot

11.4. Discussion

11.4.1. C sequestration potential of crop residues

Poor organic matter content and nutrient depletion in soils can be replenished through incorporation of crop residues. The use of crop residues is an inexpensive way for replacing macro- and micronutrients in the soil. In addition, the retention of crop residues will potentially contribute to removal of atmospheric CO_2 and facilitate its long-term storage in the soil. Incorporation of crop residues increased net CO_2-C emission, due to increase in available substrate for microbes compared to soil alone (control). These results agreed with Potthoff et al. (2005), who found that mixing maize straw with soil caused up to 40% increase in CO_2-C production. Variation in the amount of carbon emitted among different crops was attributed to differences in residue quality. In general, residue quality is negatively related to its C: N ratio, lignin: N and lignin content (Zhang et al., 2008; Walli et al., 1988). Thus, low C: N ratio causes rapid mineralization of organic N, whereas high C: N in residues results in immobilization of mineral N as it is locked up in microbial biomass (Mary et al., 1996). Thus, residues with high C: N and lignin: N ratios such as roots and shoots of sorghum genotypes KZ5246 and AS8; roots and shoots of maize genotype Mac Medium Pearl, and roots and shoots of wheat genotypes LM70 and LM75, and roots of wheat genotype BW152 roots decomposed slowly, which contributed to the lower CO_2 emissions and higher carbon sequestration potential compared to other residues. The high C: N and lignin: N ratios in such residues may not have favoured microbial decomposition leading to high recalcitrance and low CO_2 emission. Walli et al. (1988) also found that rice straw and roots with high C: N ratio, lignin, cellulose and hemicellulose content slowed down the decomposition process thereby limiting the amount of CO_2 evolved. Sorghum and maize residues generally evolved lower CO_2 (in both roots and shoots) compared to wheat showing greater potential for these two crops to sequester carbon. This would be beneficial for carbon sequestration and long-term availability of nutrients. The net CO_2 emission for all crops was generally lower for root than shoot residues. Again, this pointed to generally higher lignin, lignin: N and C: N ratios in roots than shoots of most residues, causing shoots to readily decompose. This caused the shoots of wheat genotypes BW140 and BW162, shoots of maize genotypes Nelson QPM CPSTN and R201, as well as shoots of sorghum genotypes 05-POTCH-138 and Mamolokwane to be inferior in terms of C sequestration since they were characterised by very high CO_2 emissions.

11.4.2. Nutrient mineralisation patterns of crop residues

N and P mineralisation patterns also followed the same trend as CO_2 emissions. Thus, AS8 and KZ5246 sorghum roots and shoots (characterised by low initial residue N), that gave low CO_2 emissions, also mineralised the lowest nitrate, ammonium and total mineral N; while 05-POTCH-138 roots and shoots (characterised by very high initial residue N) that had very high emissions, released the highest mineral N parameters. This makes the sorghum variety 05-POTCH-138 a very good source of N that can be applied to a growing crop. Among the maize genotypes, Nelson QPM CPSTN and Mac Medium Pearl roots, as well as Shesha CPSTN shoots (characterised by very high initial residue N) would be better sources of N since they released the highest mineral N; while PAN4P-228 roots and Mac Medium Pearl shoots (with low initial residue N) would be poor sources of N since they had the lowest net N release. Good sources of mineral N among the wheat genotypes would be BW162 and 140 shoots (high initial residue N in the shoot treatment) and roots; while BW152 roots (very low initial residue N) and LM75 shoots are poor sources of N as observed from their poor mineral N release. Maheshwari et al. (2014) attributed rapid increase of mineral N to decomposition of easily degradable nitrogenous substances (amino sugars, nucleic acids and proteins) in organic material. This is why we observed a good link between high initial N of residue with higher mineralisation pattern of N in soil and vice-versa.

Mineral P release trends however were different from that of N. Thus, in maize, Mac Medium Pearl roots (very high initial residue P) and R201 shoots were better sources of P, while Shesha CPSTN shoots (very low initial residue P) resulted in the lowest P release, and hence would not be recommended as a source of mineral P. Sorghum saw the roots and shoots of AS8 being good sources, while KZ5246 and 05-POTCH-138 roots and shoots were poor sources of P. In the case of wheat, BW162 root and BW152 shoot (high initial residue P) were good sources of mineral P; while BW140 roots (very low initial P) were the poorest source of mineral P. Generally, the shoot treatments of wheat were better sources of P than their roots, which is understandable since shoots had higher initial residue P than root treatments. Maize residues on the other hand had generally higher initial residue P in roots than shoots, which also translated to generally higher P mineralisation in root than shoot treatments. When considering organic sources of P, one would thus consider maize residues followed by sorghum, while wheat genotypes would not be good sources of P.

11.4.3. Soil microbial populations after maize residue incorporation

Microbial population studies were only done for maize, and they showed that addition of any maize residue type stimulated microbial population and activity over the control. This is because crop residues serve as ready sources of C, nutrients (N, P, S, etc.) and energy for microbes, that they need for their growth and survival. Again, it was observed that microbial populations were highest soon after maize residue addition, meaning microbes responded quickly to residue addition. The progressive decrease of microbial populations with time could mean that most of the assimilable nutrients had been incorporated by microbes into their bodies leaving little for further mineralisation towards the end of the incubation. Generally, crop residues with higher lignin content, e.g. Mac Medium Pearl roots and shoots of maize, resulted in low bacterial and fungal populations for most of the incubation periods due to low degradation of these residues. Vandecasteele et al. (2018) suggested that decomposers struggle to breakdown residues with high amounts of recalcitrant compounds (e.g. lignin, cellulose and hemicellulose). In the same vain, R201, Nelson QPM CPSTN and Shesha CPSTN roots and shoots that had low lignin amounts (< 10%) resulted in high bacterial and in some instances fungal populations, since they were ready sources of easily degradable nutrients to microbes.

11.4.4. Crop residue decomposition patterns under field conditions

Further decomposition of sorghum residues under field conditions (in a litterbag study), witnessed incorporated residues disappearing progressively with time, showing that resident microbes also readily responded to sorghum residue addition and immediately started degrading it upon incorporation into the soil. The decrease in residue C: N as well as Lignin: N ratio with time also indicates effective degradation of sorghum residues by microbes, as by the end of the study most of the C and lignin had been degraded. Gunnarsson and Marstorp, (2002) also indicated that crop residues chemical composition may change during decomposition, with hemicelluloses and cellulose decreasing with time as they are utilized as carbon and energy sources by microbes. The highest weight loss by Mamolokwane shoot (with high initial residue N and low initial lignin: N ratio) show that it was the easiest to degrade, while AS8 root (with higher lignin: N ratio) had the highest remaining weight throughout the study indicating a greater difficulty by microbes to degrade this genotype. Villegas-Pangga, (2000) also observed differences in decomposition patterns among residues and attributed this to genetic variation and differences in residue quality of different crops. Thus, crop types or/and varieties with high lignin and C: N ratio tend to be more resistant to decomposition and contribute to C sequestration and nutrients cycle balance. C: N ratios were well below 30 for most residue treatments at the end of the study, meaning they had decomposed well by this time. However, LP4403 shoot had the lowest lignin: N ratio (7), while 05-POTCH-138 root and shoot (27) followed by AS8 shoot (24) had the highest ratios at the end of the experiment. This means 05-POTCH-138 root and shoot as well as AS8 shoot would contribute more to C sequestration since they have the highest resistance to degradation under real field conditions. The P release pattern in the field was not that clear however, with just a few residue genotypes releasing P, especially at the early stages of decomposition while most residues did not respond well to incorporation.

11.5. Conclusion

The study concluded that carbon sequestration potential and nutrient mineralization differed among crop types and within genotypes. The incorporation of residues from different sorghum and wheat genotypes had significant effects on CO_2 emission, N and P mineralization. Thus, sorghum residues generally evolved lower CO_2 (in both roots and shoots) compared to wheat residues showing greater potential for sorghum to sequester carbon. The decomposition of residues is largely regulated by microbial activity which is influenced by the residue quality. In general, the residue quality is negatively related to its C:N ratio, lignin: N and lignin content. Low C: N ratio causes rapid mineralization of organic N, whereas high C: N in residues results in immobilization of mineral N as it is locked up in microbial biomass. In the present study, genotypes exhibiting lower C: N ratio, low lignin: N ratio and low lignin content (BW162 shoots and BW140 shoot residues for wheat and 05-POTCH-138 root and shoot residues for sorghum) showed rapid mineralization thereby evolving higher CO_2-C and higher concentration of mineral N while those with high C:N ratio, lignin: N ratio and lignin content (LM70 and BW152 roots for wheat and AS8 roots and KZ5246 root and shoots for sorghum residues) showed slow decomposition, thus releasing low CO_2-C and low mineral N. This would be beneficial for carbon sequestration and longer-term availability of nutrients. Phosphorus mineralization is mainly influenced by the initial P concentration of residues. P mineralization is increased for residues with high initial P content. Again, in this study residues with higher initial P content (BW152 shoots, LM70 shoots and BW162 shoots for wheat residues, and AS8 root residues for sorghum residues) released higher concentrations of extractable P. Overall, the residues which evolved the lowest CO_2-C namely LM70 wheat roots, as well as AS8 and KZ5246 sorghum roots and KZ5246 shoots are recommended to farmers for carbon sequestration and improvement of soil organic carbon (SOC). On the other hand, shoots of wheat genotypes BW162, BW140 and BW152 as well as sorghum roots and shoots from 05-POTCH-138 could be a good source of N recommended to farmers. Lastly shoots from wheat genotypes BW162, BW152 and LM75 as well as sorghum roots from AS8 could be ready sources of mineral P into the soil that can potentially serve as organic P fertilizer.

Chapter 12 MAJOR FINDINGS, GENERAL DISCUSSION AND RECOMMENDATIONS

The major findings of the project were that biomass production, grain yield, water use efficiency and carbon sequestration potential varied significantly among maize, wheat, sorghum and pearl millet, and varieties within each of these crop species. Several highlights were identified from the project:

- **i.** Crops such as sorghum, pearl millet and maize had higher biomass productivity and water use efficiency compared to wheat. This was attributed to genetic differences, with maize, sorghum and pearl millet being C4 species, which have higher capacity to regulate their water potential under harsh environments, and maintain higher biomass productivity and carbon sequestration potential compared to C3 species such as wheat.
- **ii.** Water relations are influenced by the environment, crop species, soil properties and agricultural management, so these should be taken into consideration to ensure sustainable crop production.
- **iii.** Carbon sequestration potential of a crop variety is determined by the amount of biomass produced, the chemical composition of the biomass, the soil and environmental conditions.
- **iv.** Smallholder farming communities in Sub-Saharan Africa, in general, and South Africa, in particular, are a very important stakeholders in formulating intervention strategies for sustainable agriculture. Their production of indigenous crop varieties that have largely been ignored in mainstream agriculture in favour of commercial crops such as maize and wheat offers entry points for carbon emission mitigation, water conservation and food security.

Wide intra-specific variation in biomass production and its allocation to roots and shoots was found among the different crops, showing that there is vital genetic variation for selection and development of drought tolerant varieties with enhanced carbon sequestration capacity. Correlation and diversity analyses showed that it is possible to simultaneously select for high grain yield and root biomass production to satisfy both food production and C sequestration needs. However, root to shoot ratios are inadequate as sole predictors for drought tolerance, biomass productivity or soil C input by crops due to the low correlations between root to shoot ratios and other biomass variables such as grain yield, shoot biomass and total plant biomass under different water availability scenarios. In some instances, drought stress significantly reduced root to shoot ratios in contradiction with the widely accepted optimal partitioning theory. The reduction in root to shoot ratios in response to drought stress supports the theory that prolonged drought stress can lead to the collapse of biomass allocation regulatory ability in plants. Further, investigation of C allocation using isotopic labelling revealed that 61% of the assimilated C was allocated to shoots, 20% to roots, 7% to soils while 12% was respired back to the atmosphere. Maize had the greatest carbon deposition into the soil (1.0 Mg C ha^{-1} yr^{-1} or 19% total assimilation) followed by sorghum (1.0 Mg C ha-1, 17%) and wheat (0.8 Mg C ha^{-1} yr^{-1}, 23%).

Sorghum and maize emitted lower carbon dioxide (CO_2) making them more effective at sequestering C than wheat. The roots of all three crop were also better at sequestering C than shoots, so it is worthwhile to retain roots in the soil to ensure long-term C sequestration. More specific genotypes that had great potential for C sequestration included Mac Medium Pearl roots and shoots in maize; AS8 and KZ5246 roots and shoots in sorghum; then LM70 and LM75 roots and shoots in wheat since they had the least CO_2 emissions. Consequently, Shesha CPSTN roots as well as Nelson QPM CPSTN and R201 shoots (hybrids of maize); 05-POTCH-138 and Mamolokwane roots and shoots (sorghum), and BW162 and BW140 shoots (wheat) were the poorest C sequesters as they decomposed rapidly. A recommendation of R201 roots, Nelson QPM CPSTN roots and Shesha CPSTN shoots (maize) as good sources of mineral N would be ideal. While 05-POTCH-138 shoots and roots (sorghum), as well as BW162 shoots, and BW140 roots and shoots (wheat) would be good sources of mineral N. Better sources of mineral P would be Mac Medium Pearl root and R201 shoot (maize); AS8 root (sorghum); and BW162 root and BW152 shoot in wheat. Crop residue incorporation must also be encouraged as confirmed by the high population counts of microbes induced by maize residues; as well as rapid disappearance of litter in soil that received sorghum residue treatments. Crop residues proved to be good sources of nutrients and energy needed for microbial growth and activity. However, a balance has to be maintained with regards to residues of good quality (Low C: N and lignin) that would easily decompose and provide readily available nutrients to organisms, with crop residues that have lower quality (high C: N, lignin and cellulose) that would be recalcitrant in soil and hence are more effective C sequesters.

The project identified efficient crops and varieties for use in breeding varieties of maize, sorghum and wheat that are water use efficient, drought tolerant and deposit more carbon into the soil. At advanced stages, it was recommended that the project be up-scaled to smallholder farms, where farming communities would provide some of their preferred and locally available varieties for inclusion in the screening and breeding stages. In addition, smallholder farmers would participate in future on-farm trials, workshops and field days to stay reasonably informed on progress and key decisions derived from the project.

The study also identified challenges and limitations that could be encountered in implementing the recommendations proposed in the study. Recommendations for wheat and sorghum production under smallholder farmers in South Africa are limited despite their huge potential for carbon sequestration and water use efficiency. Promoting wheat as a candidate for C sequestration among smallholder farmers in South Africa will be challenged by acute water shortages in communal farming systems where there is a lack of provision for irrigation infrastructure. On the other hand, sorghum is less preferred as a food crop among smallholder farmers in South Africa, which will complicate its promotion for carbon sequestration and water use efficiency because the farmers may find it less useful for food production. Maize is widely grown among communal farmers, but is particularly sensitive to water limitations resulting in reduced C sequestration potential under marginal conditions. For accurate quantification of carbon deposits into the soil, the isotope tracing method is widely used but the method is still largely confined to the greenhouse using a small number of genotypes grown for a relatively short period of time due to huge costs. We adapted the method for use in the greenhouse on a subset of 10 genotypes of wheat and could not extend the experiment to other crops because of the cost. There are methods that have been developed for field experiments using isotopes but these are currently out of reach for many projects in sub-Sahara Africa; due to many technical and financial constraints that prevent their implementation in developing countries. Moreover, this research mostly concentrated on generating scientific data on WUE and C sequestration through on-station experimentation; where facilities such as irrigation, weather data and crop production inputs are available. The results obtained on actual farmers' fields where resources (fertilizers, farming equipment, irrigation, etc.) are limiting might be different. There is therefore need to up-scale this work through on-farm trials to assess performance of the different crops and varieties under real field conditions.

The conclusions derived from this study suggest that more still needs to be understood on the main factors controlling water use efficiency and carbon sequestration in crops. There are many interactions between crop water use efficiency and carbon sequestration in relation to agronomic practices such as fertilizer application, irrigation scheduling, tillage system, planting dates, and plant architecture. Such interactions could not be investigated in the framework of the present study but present important grey areas for further research. Such an improved understanding of the links between plant characteristics, water use efficiency and soil C storage may open opportunities for plant breeding and improved agronomic practices with climate change mitigation in mind. Research on stover management strategies that address farmer needs while enhancing soil C is also needed. There is need to conduct a cost benefit analyses for C sequestration between accumulating high biomass compared to having more favorable biochemistry such as high C: N ratio, high lignin and high cellulose contents to increase recalcitrance of plant residues. The biochemistry of crop residues and carbon deposits have huge implications for microbial populations in the soil that affect the stability and mineralization of the deposited carbon. It will be imperative to conduct assessment on changes in microbial population in the soil following incorporation of different crop residues and determine carbon sequestration potential in that regard. The main limitation of the study was that the carbon fluxes in the soil were estimated from planting to harvest in a single season. Carbon sequestration in the soil is a long-term process and may be difficult to accurately quantify over short periods. Conducting multiple year experiments through on-farm trials should possibly provide a more accurate estimation of soil C sequestration by field crops.

REFERENCES

Abiven, S., Menasseri, S., & Chenu, C. (2009). The effects of organic inputs over time on soil aggregate stability – A literature analysis. Soil Biology and Biochemistry, 41(1), 1-12.

Aboamera, M. (2010). Response of cowpea to water deficit under semi-portable sprinkler irrigation system. *Misr Journal of Agricultural Engineering*, *27*(1), 170-190.

Abuarab, M., Mostafa, E., & Ibrahim, M. (2013). Effect of air injection under subsurface drip irrigation on yield and water use efficiency of corn in a sandy clay loam soil. *Journal of Advanced Research*, *4*(6), 493-499.

Adapa, P., Tabil, L., & Schoenau, G. (2009). Compaction characteristics of barley, canola, oat and wheat straw. *Biosystems Engineering*, *104*(3), 335-344.

Adhya, T. K., Mishra, S. R., Rath, A. K., Bharati, K., Mohanty, S. R., Ramakrishnan, B., ... & Sethunathan, N. (2000). Methane efflux from rice-based cropping systems under humid tropical conditions of eastern India. *Agriculture, Ecosystems & Environment*, *79*(1), 85-90.

Ajeigbe, H. A., Akinseye, F. M., Ayuba, K., & Jonah, J. (2018). Productivity and water use efficiency of sorghum [sorghum bicolor (L.) moench] grown under different nitrogen applications in Sudan savanna zone, Nigeria. *International Journal of Agronomy*, *2018*.

Albrecht, A., & Kandji, S. T. (2003). Carbon sequestration in tropical agroforestry systems. *Agriculture, Ecosystems & Environment*, *99*(1-3), 15-27.

Ali, R. I., Awan, T. H., Ahmad, M., Saleem, M. U., & Akhtar, M. (2012). Diversification of rice-based cropping systems to improve soil fertility, sustainable productivity and economics. *Journal of Animal and Plant Sciences*, *22*(1), 108-12.

Aljazairi, S., Arias, C., & Nogues, S. (2015). Carbon and nitrogen allocation and partitioning in traditional and modern wheat genotypes under pre-industrial and future CO 2 conditions. *Plant Biology*, *17*(3), 647-659.

Al-Naeem, M. A. (2010). Effect of irrigation scheduling on growth parameters and water use efficiency of barley and faba bean crops in Al-Ahsa, Saudi Arabia. *American Journal of Plant Physiology*, *5*(3), 143-152.

Amanullah, S. K. & Stewart, B. A. (2013). Shoot: root differs in warm season C4-cereals when grown alone in pure and mixed stands under low and high water levels. *Pakistan Journal of Botany*, *45*(1), 83-90.

An, T., Schaeffer, S., Li, S., Fu, S., Pei, J., Li, H., ... & Wang, J. (2015). Carbon fluxes from plants to soil and dynamics of microbial immobilization under plastic film mulching and fertilizer application using 13C pulse-labeling. *Soil Biology and Biochemistry*, *80*, 53-61.

Anderson-Teixeira, K. J., Miller, A. D., Mohan, J. E., Hudiburg, T. W., Duval, B. D., & DeLucia, E. H. (2013). Altered dynamics of forest recovery under a changing climate. *Global Change Biology*, *19*(7), 2001-2021.

Angadi, S. V., McConkey, B. G., Cutforth, H. W., Miller, P. R., Ulrich, D., Selles, F., ... & Brandt, S. A. (2008). Adaptation of alternative pulse and oilseed crops to the semiarid Canadian Prairie: Seed yield and water use efficiency. *Canadian Journal of Plant Science*, *88*(3), 425-438.

Aranjuelo, I., Cabrera-Bosquet, L., Mottaleb, S. A., Araus, J. L., & Nogues, S. (2009). 13C/12C isotope labeling to study carbon partitioning and dark respiration in cereals subjected to water stress. *Rapid Communications in Mass Spectrometry: An International Journal Devoted to the Rapid Dissemination of Up-to-the-Minute Research in Mass Spectrometry*, *23*(17), 2819-2828.

Ashraf, M. F. M. R., & Foolad, M. R. (2007). Roles of glycine betaine and proline in improving plant abiotic stress resistance. *Environmental and experimental botany*, *59*(2), 206-216.

Atkinson, J. A., Rasmussen, A., Traini, R., Voß, U., Sturrock, C., Mooney, S. J., ... & Bennett, M. J. (2014). Branching out in roots: uncovering form, function, and regulation. *Plant Physiology*, *166*(2), 538-550.

Bazot S., Ulff L., Blum, H., Nguyen, C., Robin, C. (2006). Effects of elevated CO_2 concentration on rhizodeposition from *Lolium perenne* grown on soil exposed to 9 years of CO_2 enrichment. *Soil Biology and Biochemistry*, 38(4): 729-736

Beer, C., Reichstein, M., Tomelleri, E., Ciais, P., Jung, M., Carvalhais, N., and Bondeau, A. (2010). Terrestrial gross carbon dioxide uptake: global distribution and covariation with climate. *Science*, 329(5993), 834-838.

Bennetzen, J., Hake, S., 2008. Handbook of Maize: Its Biology. Springer Science & Business Media.

Berry J, Björkman, O. (1980). Photosynthetic response and adaptation to temperature in higher plants. *Annual Reviews in Plant Physiology, 31*, 491-543.

Bhattarai, B., Singh, S., West, C. P., and Saini, R. (2019). Forage Potential of Pearl Millet and Forage Sorghum Alternatives to Corn under the Water-Limiting Conditions of the Texas High Plains: A Review. *Crop, Forage and Turfgrass Management, 5(1),* 1-12.

Bhutta, W. M., Akhtar, J., Ibrahim, M., and Shahzad, A. (2006). Genetic variation between Pakistani wheat (*Triticum aestivum* L.) genotypes as revealed by Random Amplified Polymorphic DNA (RAPD) markers. *South African Journal of Botany*, *72*(2), 280-283.

Blum, A. (2005). Plant Breeding for Water-Limited Environments. London: Springer.

Bolinder, M. A., Angers, D. A., Bélanger, G., Michaud, R., & Laverdière, M. R. (2002). Root biomass and shoot to root ratios of perennial forage crops in eastern Canada. *Canadian Journal of Plant Science*, *82*(4), 731-737.

Botha, G. M., & Viljoen, C. D. (2008). Can GM sorghum impact Africa? *Trends in Biotechnology*, *26*(2), 64-69.

Brauteseth, E. M. (2009). *The mutagenesis of Sorghum bicolour (L.) Moench towards improved nutrition and agronomic performance* (Doctoral dissertation).

Busari, M. A., & Salako, F. K. (2013). Effect of tillage, poultry manure and NPK fertilizer on soil chemical properties and maize yield on an Alfisol at Abeokuta, south-western Nigeria. *Nigerian Journal of Soil Science*, *23*(2), 206-218.

Busari, M. A., Kukal, S. S., Kaur, A., Bhatt, R., & Dulazi, A. A. (2015). Conservation tillage impacts on soil, crop and the environment. *International Soil and Water Conservation Research*, *3*(2), 119-129.

Butler, J. L., Bottomley, P. J., Griffith, S. M. & Myrold, D. D. (2004). Distribution and turnover of recently fixed photosynthate in ryegrass rhizospheres. *Soil Biology and Biochemistry, 36(2)*, 371-382.

Buyanovsky, G. A. & Wagner, G. H. (1997) In: Paul, EA et al. (Eds.), Soil Organic Matter in Temperate Ecosystems: Long-term Experiments in North America. CRC Press, Boca Raton, FL, pp 73-83

Cakir, R. (2004). Effect of water stress at different development stages on vegetative and reproductive growth of corn. *Field Crops Research*, *89*(1), 1-16.

Chaudhary, D. R., Saxena, J., Lorenz, N. & Dick, R. P. (2012). Distribution of recently fixed photosynthate in a switchgrass plant-soil system. *Plant Soil Environment, 58(6)*, 249-255.

Chauke, P. K., and Anim, F. D. K. (2013). Predicting Access to credit by smallholder irrigation farmers: A Logistic Regression Approach. *Journal of Human Ecology, 42(3)*, 195-202.

Chemura, A., Mahoya, C., Chidoko, P., & Kutywayo, D. (2014). Effect of soil moisture deficit stress on biomass accumulation of four coffee (*Coffea arabica*) varieties in Zimbabwe. *International Scholarly Research Notices*, *2014*.

Chen, B., Liu, E., Tian, Q., Yan, C., and Zhang, Y. (2014). Soil nitrogen dynamics and crop residues. A review. *Agronomy for Sustainable Development, 34(2)*, 429-442.

Chen, C., Wang, E., Yu, Q. & Zhang, Y. (2010). Quantifying the effects of climate trends in the past 43 years (1961-2003) on crop growth and water demand in the North China Plain. *Climatic Change, 100,* 559-578.

Chen, J., Zhu, M., Liu, R., Zhang, M., Lv, Y., Liu, Y. and Cai, H. (2020). Biomass yield 1 regulates sorghum biomass and grain yield via the shikimate pathway. *Journal of Experimental Botany, 71(8)*, 5506-5520.

Chibarabada, T. P., Modi, A. T. & Mabhaudhi, T. (2015). Water use characteristics of a Bambara groundnut (*Vigna subterranea* L. Verdc) landrace during seedling establishment. *WaterSA, 41*, 472-482.

Chikuvire, T. J., Muchaonyerwa, P. & Zengeni, R. (2018). Decomposition of *Wolffia arrhiza* residues rapidly increases mineral nitrogen and decreases extractable phosphorus in acidic soils. *Environmental Monitoring and Assessment, 190(9)*, 1-11.

Chipanshi, A. C., Chanda, R., & Totolo, O. (2003). Vulnerability assessment of the maize and sorghum crops to climate change in Botswana. *Climatic Change*, *61*(3), 339-360.

Condon, A. G., Richards, R. A., Rebetzke, G. J., & Farquhar, G. D. (2004). Breeding for high water-use efficiency. *Journal of experimental botany*, *55*(407), 2447-2460.

Conley, M. M., Kimball, B. A., Brooks, T. J., Pinter Jr, P. J., Hunsaker, D. J., Wall, G. W., ... & Triggs, J. M. (2001). CO_2 enrichment increases water-use efficiency in sorghum. *New Phytologist*, *151*(2), 407-412.

Cooley, D. R., and Manning, W. J. (1987). The impact of ozone on assimilate partitioning in plants: a review. *Environmental Pollution, 47(2)*, 95-113.

DAFF (2010) *Wheat Production Guideline*. Pretoria: Department of Agriculture, Forestry and Fisheries.

Dağdelen, N., Başal, H., Yılmaz, E., Gürbüz, T., & Akcay, S. (2009). Different drip irrigation regimes affect cotton yield, water use efficiency and fiber quality in western Turkey. *Agricultural Water Management*, *96*(1), 111-120.

Dağdelen, N., Yılmaz, E., Sezgin, F., & Gürbüz, T. (2006). Water-yield relation and water use efficiency of cotton (Gossypium hirsutum L.) and second crop corn (Zea mays L.) in western Turkey. *Agricultural Water Management*, *82*(1-2), 63-85.

Damon, P.M., Bowden, B., Rose, T., & Rengel, Z. (2014). Crop residue contributions to phosphorus pools in agricultural soils: a review. *Soil Biology and Biochemistry, 74*,127-137.

Daryanto, S., Wang, L., and Jacinthe, P. A. (2016). Global synthesis of drought effects on maize and wheat production. *PloS one, 11(5)*, e0156362.

Davenport, J. R. & Thomas, R. L. (1988). Carbon partitioning and rhizodeposition in corn and bromegrass. *Canadian Journal of Soil Science, 68(4)*, 693-701.

Davidson, E. A. & Janssens, I. A. (2006). Temperature sensitivity of soil carbon decomposition and feedbacks to climate change. *Nature, 440*, 165-173.

De Graaff, M.A., Van Kessel. C., & Six, J. (2009). Rhizodeposition-induced decomposition increases N availability to wild and cultivated wheat genotypes under elevated CO_2. *Soil Biology and Biochemistry, 41(6)*, 1094-1103.

De Lima, R. S. N., de Assis, F. A. M. M., Martins, A. O., de Deus, B. C. D. S., Ferraz, T. M., de Assis Gomes, M. D. M., ... & Campostrini, E. (2015). Partial rootzone drying (PRD) and regulated deficit irrigation (RDI) effects on stomatal conductance, growth, photosynthetic capacity, and water-use efficiency of papaya. *Scientia Horticulturae*, *183*, 13-22.

De Neergaard, A. & Gorissen, A. (2004). Carbon allocation to roots, rhizodeposits and soil after pulse labelling: a comparison of white clover (*Trifolium repens* L.) and perennial ryegrass (*Lolium perenne* L.). *Biology and Fertility of Soils, 39(4)*, 228-234.

Dennis, I., & Dennis, R. (2012). Climate change vulnerability index for South African aquifers. *Water SA*, *38*(3), 417-426.

Department of Agriculture, Forestry and Fisheries (DAFF) (2010). *Sorghum production guidelines*. Pretoria, South Africa.

Department of Agriculture, Forestry and Fisheries (DAFF) (2017). Trends in the Agricultural

DerSimonian, R., and Laird, N. (1986). Meta-analysis in clinical trials. *Controlled Clinical Trials, 7(3)*, 177-188.

Di Paolo, E., & Rinaldi, M. (2008). Yield response of corn to irrigation and nitrogen fertilization in a Mediterranean environment. *Field Crops Research*, *105*(3), 202-210.

Diao, X., Hazell, P., and Thurlow, J. (2010). The role of agriculture in African development. *World Development, 38(10)*, 1375-1383.

Dimassi, B., Mary, B., Wylleman, R., Labreuche, J., Couture, D., Piraux, F. & Cohan, J. P. (2014). Long-term effect of contrasted tillage and crop management on soil carbon dynamics during 41 years. *Agriculture Ecosystem and Environment, 188*, 134-146.

Dimkpa, C. O., Singh, U., Bindraban, P. S., Elmer, W. H., Gardea-Torresdey, J. L., & White, J. C. (2019). Zinc oxide nanoparticles alleviate drought-induced alterations in sorghum performance, nutrient acquisition, and grain fortification. *Science of the Total Environment, 688*, 926-934.

Dodig, D., Zorić, M., Kandić, V., Perović, D., and Šurlan-Momirović, G. (2012). Comparison of responses to drought stress of 100 wheat accessions and landraces to identify opportunities for improving wheat drought resistance. *Plant Breeding*, *131*(3), 369-379.

Domanski, G., Kuzyakov, Y., Siniakina, S. & Stahr, K. (2001). Carbon flows in the rhizosphere of ryegrass (*Lolium perenne*). *Journal of Plant Nutrition and Soil Science, 164(4)*, 381-387.

Donnenfeld, Z., Crookes, C., and Hedden, S. (2018). A delicate balance: Water scarcity in South Africa. *ISS Southern Africa Report, 2018(13)*, 1-24.

Dou, F., Soriano, J., Tabien, R. E., & Chen, K. (2016). Soil texture and cultivar effects on rice (Oryza sativa, L.) grain yield, yield components and water productivity in three water regimes. *PloS one*, *11*(3), e0150549.

Du Preez, C. C., Van Huyssteen, C. W. and Mnkeni, P. N. 2011. Land use and soil organic matter in South Africa 2: A review on the influence of arable crop production. *South African Journal of Science,* 107**,** 35-42.

Du, T., Kang, S., Sun, J., Zhang, X., and Zhang, J. (2010). An improved water use efficiency of cereals under temporal and spatial deficit irrigation in north China. *Agricultural Water Management, 97(1)*, 66-74.

Durand, M., Brendel, O., Buré, C., Courtois, P., Lily, J. B., Granier, A., and Le Thiec, D. (2020). Impacts of a partial rainfall exclusion in the field on growth and transpiration: consequences for leaf-level and whole-plant water-use efficiency compared to controlled conditions. *Agricultural and Forest Meteorology, 282*, 107873.

Eliasson, Å., Faurès, J. M., Frenken, K., and Hoogeveen, J. (2003). AQUASTAT – Getting to grips with water information for agriculture. Land and Water Development Division FAO, Rome, Italy (http://www. fao. org/ag/aquastat. Accessed July 2020).

Erdem, T., Delibas, L., & Orta, A. H. (2001). Water-use characteristics of sunflower (Helianthus annuus L.) under deficit irrigation. *Pakistan Journal of Biological Sciences*, *4*(7), 766-769.

Estrada-Campuzano, G., Slafer, G. A., and Miralles, D. J. (2012). Differences in yield, biomass and their components between triticale and wheat grown under contrasting water and nitrogen environments. *Field Crops Research*, *128*, 167-179.

Evans, J. R., and von Caemmerer, S. (2000). Would C4 rice produce more biomass than C3 rice? *Studies in Plant Science, 7*, 53-71.

Evans, R. G., & Sadler, E. J. (2008). Methods and technologies to improve efficiency of water use. *Water Resources Research*, *44*(7).

Eziz, A., Yan, Z., Tian, D., Han, W., Tang, Z., and Fang, J. (2017). Drought effect on plant biomass allocation: A meta-analysis. *Ecology and evolution*, *7*(24), 11002-11010.

Fan, F., Zhang, F., Qu, Z. & Lu, Y. (2008). Plant carbon partitioning below ground in the presence of different neighboring species. *Soil Biology and Biochemistry, 40(9)*, 2266-2272

Fan, J., McConkey, B., Wang, H. & Janzen, H. (2016). Root distribution by depth for temperate agricultural crops. *Field Crops Research, 189*, 68-74.

Fan, T., Wang, S., Xiaoming, T., Luo, J., Stewart, B. A., & Gao, Y. (2005). Grain yield and water use in a long-term fertilization trial in Northwest China. *Agricultural Water Management*, *76*(1), 36-52.

Fang, Y., Du, Y., Wang, J., Wu, A., Qiao, S., Xu, B., and Chen, Y. (2017). Moderate drought stress affected root growth and grain yield in old, modern and newly released cultivars of winter wheat. *Frontiers in Plant Science*, *8*, 672.

Fang, Y., Singh, B. P., Badgery, W. & He, X. (2016). *In situ* assessment of new carbon and nitrogen assimilation and allocation in contrastingly managed dryland wheat crop-soil systems. *Agriculture, Ecosystems and Environment, 235*, 80-90.

Fang, Y., Xu, B., Liu, L., Gu, Y., Liu, Q., Turner, N. C., and Li, F. M. (2014). Does a mixture of old and modern winter wheat cultivars increase yield and water use efficiency in water-limited environments? *Field Crops Research, 156*, 12-21.

Faye, A., Sine, B., Chopart, J. L., Grondin, A., Lucas, M., Diedhiou, A. G., ... and Kane, A. (2019). Development of a model estimating root length density from root impacts on a soil profile in pearl millet (*Pennisetum glaucum* (L.) R. Br). Application to measure root system response to water stress in field conditions. *PloS one, 14(7),* e0214182.

Fernández, J., Curt, M. D., Aguado, P. & Magro, E. (2003). Carbon allocation in a sweet sorghum-soil system using 14C as a tracer. *Journal of Plant Nutrition and Soil Science, 166(1)*, 23-30.

Ferreira, G. (2016). The interaction of drought stress and heat stress as determinant of dry matter yield and nutritional composition of maize (*Zea mays* L) whole-plant for silage. *Maydica, 60(1)*, 1-6.

Ficiciyan, A., Loos, J., Sievers-Glotzbach, S., and Tscharntke, T. (2018). More than yield: Ecosystem services of traditional versus modern crop varieties revisited. *Sustainability, 10(8)*, 2834.

Foulkes, M. J., Sylvester-Bradley, R., Weightman, R., and Snape, J. W. (2007). Identifying physiological traits associated with improved drought resistance in winter wheat. *Field Crops Research*, *103*(1), 11-24.

Gao, Y., Duan, A., Sun, J., Li, F., Liu, Z., Liu, H., & Liu, Z. (2009). Crop coefficient and water-use efficiency of winter wheat/spring maize strip intercropping. *Field Crops Research*, *111*(1-2), 65-73.

Gbetibouo, G.A. and Hassan, R.M. (2005). Measuring the economic impact of climate change on major South African field crops: A Ricardian approach*. Global and Planetary Change 47*, 143-152.

Gelalcha, S., Fantahun, B. A. S. A. Z. E. N., Yaie, B. A. L. C. H. A., and Girma, B. E. D. A. D. A. (2007). Triticale (X Triticosecale wittmack)-a new addition to the Ethiopian cereals. In *African Crop Science Conference Proceedings* (Vol. 8, pp. 1991-1995).

Gezahegn, A. M., Halim, R. A., Yusoff, M. M. and Wahid, S.A. (2016). Decomposition and Nitrogen mineralization of Individual and Mixed Maize and Soybean Residue. *Journal of Agricultural Science, 2*, 28-45.

Ghassemi-Golezani, K., and Tajbakhsh, Z. (2012). Relationship of plant biomass and grainfillingwith grain yield of maize cultivars. *International Journal of Agriculture and Crop Sciences, 4(20*), 1536-1539.

Ghimire, B., Ghimire, R., VanLeeuwen, D., and Mesbah, A. (2017). Cover crop residue amount and quality effects on soil organic carbon mineralization. *Sustainability, 9(12)*, 2316.

Giardina, C. P., Litton, C. M., Crow, S. E. & Asner, G. P. (2014). Warming-related increases in soil CO_2 efflux are explained by increased below-ground carbon flux. *Nature Climate Change 4*, 822-827

Gocke, M., Pustovoytov, K. & Kuzyakov, Y. (2011). Carbonate recrystallization in root-free soil and rhizosphere of Triticum aestivum and *Lolium perenne* estimated by 14C labeling. *Biogeochemistry, 103(1-3)*, 209-222.

Gomez, L. D., Steele-King, C. G., and McQueen-Mason, S. J. (2008). Sustainable liquid biofuels from biomass: the writing is on the walls. *New Phytologist, 178(3)*, 473-485.

Greenway, H., Armstrong, W., & Colmer, T. D. (2006). Conditions leading to high CO2 (> 5 kPa) in waterlogged-flooded soils and possible effects on root growth and metabolism. *Annals of Botany*, *98*(1), 9-32.

Gregory, P. J. & Atwell, B. J. (1991). The fate of carbon in pulse-labelled crops of barley and wheat. *Plant and Soil, 136(2)*, 205-213.

Gregory, P. J., Ingram, J. S., Andersson, R., Betts, R. A., Brovkin, V., Chase, T. N., ... and Howden, S. M. (2002). Environmental consequences of alternative practices for intensifying crop production. *Agriculture, Ecosystems and Environment, 88(3)*, 279-290.

Gunnarsson, S., & Marstorp, H. (2002). Carbohydrate composition of plant materials determines N mineralisation. *Nutrient Cycling in Agroecosystems*, *62*(2), 175-183.

Habyarimana, E., Piccard, I., Catellani, M., De Franceschi, P., and Dall'Agata, M. (2019). Towards predictive modeling of sorghum biomass yields using fraction of absorbed photosynthetically active radiation derived from sentinel-2 satellite imagery and supervised machine learning techniques. *Agronomy, 9(4)*, 203.

Hallam, A., Anderson, I. C. & Buxton, D. R. (2001). Comparative economic analysis of perennial, annual, and intercrops for biomass production. *Biomass and Bioenergy, 21*, 407-424.

Haque, E, Osmani, A.A., Ahmadi, S. H. & Ban, T. (2016). Development of pre-breeding technology for root system study and selection of Kihara Afghan wheat landraces (KAWLR) to enhance wheat breeding in the rain-fed region. *Breeding Science, 66(5)*, 808-822

Harris, D., Pacovsky, R. S. & Paul, E. A. (1985) Carbon economy of soybean-Rhizobium-Glomus associations. *New Phytologist, 101(3),* 427-440.

Hasan, S. A., Rabei, S. H., Nada, R. M., & Abogadallah, G. M. (2017). Water use efficiency in the drought-stressed sorghum and maize in relation to expression of aquaporin genes. *Biologia Plantarum, 61(1),* 127-137.

Heal, O. W., Anderson, J. W. & Swift, M. J. (1997). Plant litter quality and decomposition: an historical overview. In: Cadisch, G., Giller, K.E. (Eds.), Driven by Nature: Plant Litter Quality and Decomposition. CAB International, Wallingford, UK, pp. 3-30.

Hemminga, M. A., Huiskes, A. H. L., Steegstra, M. & Van Soelen, J. (1996). Assessment of carbon allocation and biomass production in a natural stand of the salt marsh plant Spartina anglica using 13 C. *Marine Ecology Progress Series, 130*, 169-178.

Henao, J. and Baanante C. (2006). Agricultural production and soil nutrient mining in Africa: Implication for resource conservation and policy development. IFDC Technical Bulletin.

Hendrickson, L., Chow, W. S., & Furbank, R. T. (2004). Low temperature effects on grapevine photosynthesis: the role of inorganic phosphate. *Functional Plant Biology*, *31*(8), 789-801.

Hodge, A. & Millard, P. (1998). Effect of elevated CO_2 on carbon partitioning and exudate release from *Plantago lanceolata* seedlings. *Physiologia Plantarum, 103(2)*, 280-286.

Hodge, A., Paterson, E., Thornton, B., Millard, P. & Killham, K. (1997). Effects of photon flux density on carbon partitioning and rhizosphere carbon flow of *Lolium perenne*. *Journal of Experimental Botany, 48(10)*, 1797-1805.

Holland, J. N., Cheng, W. & Crossley, D. A. (1995). Herbivore-induced changes in plant carbon allocation: assessment of below-ground C fluxes using carbon-14. *Oecologia, 107(1)*, 87-94.

Hossner, L. R. and Juo A.S.R. (1999). Soil Nutrient Management for Sustained Food crop Production in Upland Farming Systems in the Tropics. Juo Soil and Crop Sciences Department College Station Tennessee 77843, USA. Retrieved from http//www.agnet.org.

Huang, B. and Johnson, J. W. (1995). Root respiration and carbohydrate status of two wheat genotypes in response to hypoxia. *Annals of Botany, 75(4),* 427-432.

Husson, F., Josse, J., Le, S., Mazet, J., and Husson, M. F. (2016). Package 'FactoMineR'. An R package, 96, 698.

Hütsch, B. W., Augustin, J. & Merbach, W. (2002). Plant rhizodeposition – an important source for carbon turnover in soils. *Journal of Plant Nutrition and Soil Science, 165,* 397-407.

Ibragimov, N., Evett, S. R., Esanbekov, Y., Kamilov, B. S., Mirzaev, L., & Lamers, J. P. (2007). Water use efficiency of irrigated cotton in Uzbekistan under drip and furrow irrigation. *Agricultural water management*, *90*(1-2), 112-120.

Infante, P. A., Moore, K., Hurburgh, C., Scott, P., Archontoulis, S., Lenssen, A., and Fei, S. Z. (2018). Biomass production and composition of temperate and tropical maize in Central Iowa. *Agronomy*, *8(6)*, 88.

Intergovernmental Panel on Climate Change (IPCC), Climate change (2014). Impacts, adaptation, and vulnerability, Part B: Regional aspects, Chapter 22: Africa, 1199, www.ipcc.ch/report/ar5/wg2/

Ismail, S. M., & Ozawa, K. (2007). Improvement of crop yield, soil moisture distribution and water use efficiency in sandy soils by clay application. *Applied Clay Science*, *37*(1-2), 81-89.

Israelsen, O. W. (1932). Irrigation principles and practices. Wiley and Sons, New York, USA.

Jabereldar, A. A., El Naim, A. M., Abdalla, A. A., & Dagash, Y. M. (2017). Effect of water stress on yield and water use efficiency of sorghum (*Sorghum bicolor* L. Moench) in semi-arid environment. *International Journal of Agriculture and Forestry, 7(1),* 1-6.

Jones, C., McConnell, C., Coleman, K., Cox, P., Falloon, P., Jenkinson, D. & Powlson, D. (2005). Global climate change and soil carbon stocks; predictions from two contrasting models for the turnover of organic carbon in soil. *Global Change Biology, 11(1)*: 154-166.

Kaiser, C., Kilburn, M. R., Clode, P. L., Fuchslueger, L., Koranda, M., et al. (2015). Exploring the transfer of recent plant photosynthates to soil microbes: mycorrhizal pathway vs direct root exudation. *New Phytologist, 205(4),* 1537-1551.

Kakiuchi, J. & Kobata, T. (2008). High carbon requirements for seed production in soybeans [*Glycine max* (L.) Merr.]. *Plant Production Science, 11(2),* 198-202.

Kanani, E., Dehghanisanij, H., Akhavan, S., 2016. Effects of Different Irrigation Methods

Kang, S., Zhang, L., Liang, Y., Hu, X., Cai, H., & Gu, B. (2002). Effects of limited irrigation on yield and water use efficiency of winter wheat in the Loess Plateau of China. *Agricultural water management*, *55*(3), 203-216.

Kang, Y., Khan, S. & Ma, X. (2001). Climate change impacts on crop yield, crop water productivity and food security – A review. *Progress in Natural Science 19,* 1665-1674.

Kar, G., Kumar, A., & Martha, M. (2007). Water use efficiency and crop coefficients of dry season oilseed crops. *Agricultural Water Management*, *87*(1), 73-82.

Kashiwagi, J., Krishnamurthy, L., Crouch, J. H., and Serraj, R. (2006). Variability of root length density and its contributions to seed yield in chickpea (Cicer arietinum L.) under terminal drought stress. *Field Crops Research, 95*, 171-181

Kassambara, A., & Mundt, F. (2017). Factoextra: extract and visualize the results of multivariate data analyses. *R package version*, *1*(5), 337-354.

Kaštovská, E. & Šantrůčková, H. (2007). Fate and dynamics of recently fixed C in pasture plant-soil system under field conditions. *Plant Soil, 300*, 61-69.

Katerji, N. & Mastrorilli, M. (2014). Water Use Efficiency of Cultivated Crops. *eLS*, 1-13, doi:10.1002/9780470015902.a0025268

Khayatnezhad, M., Gholamin, R., Jamaatie-Somarin, S. H., & Zabihi-Mahmoodabad, R. (2010). Effects of peg stress on corn cultivars (Zea mays L.) at germination stage. *World Applied Science Journal, 11(5)*, 504-506.

Killi, D., Bussotti, F., Raschi, A., & Haworth, M. (2017). Adaptation to high temperature mitigates the impact of water deficit during combined heat and drought stress in C3 sunflower and C4 maize varieties with contrasting drought tolerance. *Physiologia Plantarum*, *159*(2), 130-147.

King, J. S, Giardana, C. P., Pregitzer, K. S. & Friend, A. L. (2007). Biomass partitioning in red pine (Pinus resinosa) along a chronosequence in the Upper Peninsula of Michigan. *Canadian Journal of Forest Research, 37,* 93-102.

Kiziloglu, F. M., Sahin, U., Kuslu, Y., and Tunc, T. (2009). Determining water-yield relationship, water use efficiency, crop and pan coefficients for silage maize in a semiarid region. *Irrigation Science, 27(2)*, 129.

Kontopantelis, E., and Reeves, D. (2009). MetaEasy: A meta-analysis add-in for Microsoft Excel. *Journal of Statistical Software, 30(7)*, 1-25.

Kumwenda, J. D. T., Waddington, S. R., Snapp, S. S., Jones, R. B and Blackie, M. J. (1996). Soil Fertility Management Research for the Maize Cropping Systems in Smallholders in Southern Africa: A review: CIMMYT, Mexico.

Kundu, S., Bhattacharyya, R., Prakash, V., Ghosh, B. Gupta, H. J. S. (2007). Carbon sequestration and relationship between carbon addition and storage under rainfed soybean-wheat rotation in a sandy loam soil of the Indian Himalayas. *Soil and Tillage Research, 92*, 87-95.

Kuscu, H., & Demir, A. O. (2013). Yield and water use efficiency of maize under deficit irrigation regimes in a sub-humid climate. *Philippine Agricultural Scientist*, *96*(1), 32-41.

Kuscu, H., Karasu, A., Mehmet, O., Demir, A.O., Turgut, I. (2013). Effect of irrigation amounts applied with drip irrigation on maize evapotranspiration, yield, water use efficiency, and net return in a sub-humid climate. *Turkish Journal of Field Crops*, *18*(1), 13-19.

Kuzyakov, Y. & Domanski, G. (2000). Carbon input by plants into the soil: Review. *Journal of Plant Nutrition and Soil Science,* 421-431

Kuzyakov, Y. & Larionova, A. A. (2005). Root and rhizomicrobial respiration: a review of approaches to estimate respiration by autotrophic and heterotrophic organisms in soil. *Journal of Plant Nutrition and Soil Science, 168*, 503-520.

Lal, R. (2001). Soil carbon sequestration and climate change. Senate Hearing, Science and Technical Sub-Committee, 24 May, Washington, DC.

Lal, R. (2004). Soil carbon sequestration impacts on global climate change and food security. *Science, 304*, 1623-1627.

Lal, R. (2006). Enhancing crop yields in the developing countries through restoration of the soil organic carbon pool in agricultural lands. *Land Degradation and Development, 17*,197-209.

Lal, R. (2016). Beyond COP 21: potential and challenges of the “4 per Thousand” initiative. *Journal of Soil Water Conservation, 71*, 20A-25A.

Lamptey, S., Nyarko, G., Falon, A., and Yeboah, S. (2014). Assessing the performance of sorghum varieties in the Guinea Savanna Zone of Ghana. *Asian Journal of Agriculture and Food Sciences, 2(1).*

Langholtz, M. H., Stokes, B. J., and Eaton, L. M. (2016). 2016 Billion-ton report: Advancing domestic resources for a thriving bioeconomy, Volume 1: Economic availability of feedstock. Oak Ridge National Laboratory, Oak Ridge, Tennessee, managed by UT-Battelle, LLC for the US Department of Energy, 2016, 1-411.

LECO Corporation. (2012). Truspec CN Carbon /Nitrogen Determinator Instructions Manual. St Joseph, USA: LECO Corporation

Li, Y., Zhang, R., Liu, G., Chen, C., He, Y. & Liu, X. (2013). Comparison of methane production potential, biodegradability, and kinetics of different organic substrates. *Bioresource Technology, 149,* 565-569.

Linderson, M. L., Iritz, Z., and Lindroth, A. (2007). The effect of water availability on stand-level productivity, transpiration, water use efficiency and radiation use efficiency of field-grown willow clones. *Biomass and Bioenergy, 31(7)*, 460-468.

Litton, C. M., Raich, J. W., and Ryan, M. G. (2007). Carbon allocation in forest ecosystems. *Global Change Biology*, *13*(10), 2089-2109.

Lodhi, A. S. M. A., Sajjad, M. H., Mahmood, A. N. S. A. R., Tahir, S. & Azam, F. (2009). Photosynthate partitioning in wheat (*Triticum aestivum* L.) as affected by root-zone salinity and form of N. *Pakistan Journal of Botany, 41*, 1363-1372.

Lopez-Castaneda, C., and Richards, R. A. (1994). Variation in temperate cereals in rain fed environments II. Phasic development and growth. *Field Crops Research, 37(1)*, 63-75.

Lorenz, K. and Lal, R. (2005). The depth distribution of soil organic carbon in relation to land use and management and the potential of carbon sequestration in subsoil horizons. *Advances in Agronomy, 88*, 35-66.

Luca, E. F., Chaplot, V., Mutema, M., Feller, C., Ferreira, M. L., Cerri, C. C., & Couto, H. T. Z. (2018). Effect of conversion from sugarcane preharvest burning to residues green-trashing on SOC stocks and soil fertility status: Results from different soil conditions in Brazil. *Geoderma*, *310*, 238-248.

Ludewig, F. & Flügge, U. I. (2013). Role of metabolite transporters in source-sink carbon allocation. *Frontiers in Plant Science, 4 (231),* 1-16.

Mabhaudhi T, Modi A, Beletse Y. (2013). Growth, phenological and yield responses of a bambara groundnut (*Vigna subterranea* L. Verdc) landrace to imposed water stress: II. rain shelter conditions. *WaterSA* 39: 191-198.

Maheshwari, D. K., Dheeman, S., & Agarwal, M. (2014). Decomposition of organic materials into high value compost for sustainable crop productivity. In *Composting for Sustainable Agriculture* (pp. 245-267). Springer, Cham.

Mahmoud, A. M., & Ahmed, T. A. (2016). Water use efficiency of sunflower genotypes under drip irrigation. *African Journal of Agricultural Research*, *11*(11), 925-929.

Manschadi, A. M., Christopher, J. T., DeVoil, P., and Hammer, G. L. (2006). The role of root architectural traits in adaptation of wheat to water-limited environments. *Functional Plant Biology, 33*, 823-837.

Maroco, J. P., Pereira, J. S., & Chaves, M. M. (2000). Growth, photosynthesis and water-use efficiency of two C4 Sahelian grasses subjected to water deficits. *Journal of Arid Environments, 45(2)*, 119-137.

Mary, B., Recous, S., Darwis, D., & Robin, D. (1996). Interactions between decomposition of plant residues and nitrogen cycling in soil. *Plant and soil*, *181*(1), 71-82.

Mathew, I., Shimelis, H., Mutema, M., & Chaplot, V. (2017). What crop type for atmospheric carbon sequestration: Results from a global data analysis. *Agriculture, Ecosystems & Environment*, *243*, 34-46.

Mathew, I., Shimelis, H., Mutema, M., Clulow, A., Zengeni, R., Mbava, N., and Chaplot, V. (2019). Selection of wheat genotypes for biomass allocation to improve drought tolerance and carbon sequestration into soils. *Journal of Agronomy and Crop Science, 205(4)*, 385-400.

Maw, M. J., Houx III, J. H., and Fritschi, F. B. (2017). Maize, sweet sorghum, and high biomass sorghum ethanol yield comparison on marginal soils in Midwest USA. *Biomass and Bioenergy, 107*, 164-171.

Mbava, N., Mutema, M., Zengeni, R., Shimelis, H., and Chaplot, V. (2020). Factors affecting crop water use efficiency: A worldwide meta-analysis. *Agricultural Water Management, 228*, 105878-105889.

McKendry, P. (2002). Energy production from biomass (part 1): overview of biomass. *Bioresource Technology, 83(1),* 37-46.

Meng, F., Dungait, J. A., Zhang, X., He, M., Guo, Y., & Wu, W. (2013). Investigation of photosynthate-C allocation 27 days after 13 C-pulse labeling of *Zea mays* L. at different growth stages. *Plant and Soil*, *373*(1), 755-764.

Menichetti, L., Ekblad, A., & Kätterer, T. (2015). Contribution of roots and amendments to soil carbon accumulation within the soil profile in a long-term field experiment in Sweden. *Agriculture, Ecosystems & Environment*, *200*, 79-87.

Meskelu, E., Mohammed, M., & Hordofa, T. (2014). Response of maize (Zea mays L.) for moisture stress condition at different growth stages. *International Journal of Recent Research in Life Sciences*, *1*, 12-21.

Mi, N., Cai, F., Zhang, Y., Ji, R., Zhang, S., and Wang, Y. (2018). Differential responses of maize yield to drought at vegetative and reproductive stages. *Plant, Soil and Environment, 64(6)*, 260-267.

Minasny, B., Malone, B. P., McBratney, A. B., Angers, D. A., Arrouays, D., Chambers, A., ... & Winowiecki, L. (2017). Soil carbon 4 per mille. *Geoderma*, *292*, 59-86.

Mofokeng, A., Shimelis, H., Tongoona, P., and Laing, M. (2014). A genetic diversity analysis of South African sorghum genotypes using SSR markers. *South African Journal of Plant and Soil*, *31*(3), 145-152.

Mojid, M. A., Wyseure, G. C. L., & Mustafa, S. M. (2012). Water use efficiency and productivity of wheat as a function of clay amendment. *Environmental Control in Biology*, *50*(4), 347-362.

Mokgope, K., Pollard, S., and Butterworth, J. (2001). Water resources and water supply for rural communities in the Sand River Catchment, South Africa. In 27th WEDC Conference, People and systems for water, sanitation and health, Lusaka, Zambia.

Morales-Olmedo, M., Ortiz, M., & Sellés, G. (2015). Effects of transient soil waterlogging and its importance for rootstock selection. *Chilean Journal of Agricultural Research*, *75*, 45-56.

Morison, J. I. L., Baker, N. R., Mullineaux, P. M., & Davies, W. J. (2008). Improving water use in crop production. *Philosophical Transactions of the Royal Society B: Biological Sciences*, *363*(1491), 639-658.

Mota, C., Alcaraz-López, C., Iglesias, M., Martínez-Ballesta, M. C., Carvajal, M. (2010). Investigation into CO_2 absorption of the most representative agricultural crops of the region of Murcia. Departamento de Nutrición Vegetal, Project report.

Muchow, R. C. (1989). Comparative productivity of maize, sorghum and pearl millet in a semi-arid tropical environment I. Yield potential. *Field Crops Research, 20(3)*, 191-205.

Muhammad, D., Raza, I., Ahmad, S., Afzal, M. N., & Mian, M. A. (2010). Efficiency of drip vs furrow irrigation system with variable plant density on cotton under southern Punjab climatic conditions. *Furrow*, *30*(126), 2-55.

Muhammad, W., Vaughan, S. M., Dalal, R. C., & Menzies, N. W. (2016). Crop residues and fertilizer nitrogen influence residue decomposition and nitrous oxide emission from a Vertisol. *Biology and Fertility of Soils*, *47*(1), 15-23.

Murungu, F. S., Chiduza, C., Muchaonyerwa, P., & Mnkeni, P. N. S. (2011). Mulch effects on soil moisture and nitrogen, weed growth and irrigated maize productivity in a warm-temperate climate of South Africa. *Soil and Tillage Research*, *112*(1), 58-65.

Mutegi, J. K., Petersen, B. M., Munkholm, L. J., & Hansen, E. M. (2011). Belowground carbon input and translocation potential of fodder radish cover-crop. *Plant and Soil*, *344*(1), 159-175.

Mwadzingeni, L., Shimelis, H., Tesfay, S., & Tsilo, T. J. (2016). Screening of bread wheat genotypes for drought tolerance using phenotypic and proline analyses. *Frontiers in Plant Science*, *7*, 1276.

Nagaz, K., Masmoudi, M., & Mechila, N. (2009). Yield and water use-efficiency of pearl millet (Pennisetum glaucum (L.) R. Br.) under deficit irrigation with saline water in arid conditions of Southern Tunisia. *Research Journal of Agronomy*, *3*(1), 9-17.

National Chamber of Milling, South Africa. (2008). Cultivars: Maize. Cited March 23, 2019. www.grainmilling.org.za.

Nevo, E., and Chen, G. (2010). Drought and salt tolerances in wild relatives for wheat and barley improvement. *Plant, Cell and Environment*, *33*(4), 670-685.

Nguyen, C. (2009). Rhizodeposition of organic C by plant: mechanisms and controls. *Sustainable agriculture*, 97-123.

Ni, J. Q., Heber, A. J., Hanni, S. M., Lim, T. T., & Diehl, C. A. (2010). Characteristics of ammonia and carbon dioxide releases from layer hen manure. *British Poultry Science*, *51*(3), 326-334.

Norton, R. M., & Wachsmann, N. G. (2006). Nitrogen use and crop type affect the water use of annual crops in south-eastern Australia. *Australian Journal of Agricultural Research*, *57*(3), 257-267.

Oikawa, P. Y., Jenerette, G. D., & Grantz, D. A. (2014). Offsetting high water demands with high productivity: Sorghum as a biofuel crop in a high irradiance arid ecosystem. *Gcb Bioenergy*, *7*(5), 974-983.

Okalebo, J. R. (1993). Laboratory methods of soil and plant analysis: a working manual, Unesco/Rosta. Nairobi, Kenya.

Onder, D., Akiscan, Y., Onder, S., & Mert, M. (2009). Effect of different irrigation water level on cotton yield and yield components. *African Journal of Biotechnology*, *8*(8), 1536-1544.

Ostle, N., Whiteley, A. S., Bailey, M. J., Sleep, D., Ineson, P., & Manefield, M. (2003). Active microbial RNA turnover in a grassland soil estimated using a 13CO2 spike. *Soil Biology and Biochemistry*, *35*(7), 877-885.

Otieno, F. A., and Ochieng, G. M. (2004). Water management tools as a means of averting a possible water scarcity in South Africa by the year 2025. *Water SA, 30(5),* 120-124.

Oweis, T., Hachum, A., & Pala, M. (2004). Water use efficiency of winter-sown chickpea under supplemental irrigation in a Mediterranean environment. *Agricultural Water Management*, *66*(2), 163-179.

Palm, C. A., Gachengo, C. N., Delve, R. J., Cadisch, G., & Giller, K. E. (2001). Organic inputs for soil fertility management in tropical agroecosystems: application of an organic resource database. *Agriculture, Ecosystems & Environment*, *83*(1-2), 27-42.

Palm, C. A., Giller, K. E., Mafongoya, P. L., & Swift, M. J. (2001). Management of organic matter in the tropics: translating theory into practice. In *Managing Organic Matter in Tropical Soils: Scope and Limitations* (pp. 63-75). Springer, Dordrecht.

Pandey, A. K., Ghosh, A., Agrawal, M., & Agrawal, S. B. (2018). Effect of elevated ozone and varying levels of soil nitrogen in two wheat (*Triticum aestivum* L.) cultivars: Growth, gas-exchange, antioxidant status, grain yield and quality. *Ecotoxicology and Environmental Safety*, *158*, 59-68.

Pausch, J. and Kuzyakov, Y. (2017). Carbon input by roots into the soil: Quantification of rhizodeposition from root to ecosystem scale. *Global Change Biology, 24(1)*, 1-12.

Paustian, K., Lehmann, J., Ogle, S., Reay, D., Robertson, G. P. & Smith, P. (2016). Climate-Smart soils. *Nature, 532*, 49-57.

Payero, J. O., Tarkalson, D. D., Irmak, S., Davison, D., & Petersen, J. L. (2009). Effect of timing of a deficit-irrigation allocation on corn evapotranspiration, yield, water use efficiency and dry mass. *Agricultural Water Management*, *96*(10), 1387-1397.

Payero, J. O., Tarkalson, D. D., Irmak, S., Davison, D., & Petersen, J. L. (2008). Effect of irrigation amounts applied with subsurface drip irrigation on corn evapotranspiration, yield, water use efficiency, and dry matter production in a semiarid climate. *Agricultural Water Management*, *95*(8), 895-908.

Payne, R. W., Murray, D. A., Harding, S. A. (2017). An Introduction to the Genstat Command Language (18th Edition). VSN International Ltd., Hemel, Hempstead, UK. Pp. 1-137.

Peng, S., Krieg, D. R., and Girma, F. S. (1991). Leaf photosynthetic rate is correlated with biomass and grain production in grain sorghum lines. *Photosynthesis Research, 28(1)*, 1-7.

Perdomo, J. A., Conesa, M. À., Medrano, H., Ribas-Carbó, M., and Galmés, J. (2015). Effects of long-term individual and combined water and temperature stress on the growth of rice, wheat and maize: relationship with morphological and physiological acclimation. *Physiologia Plantarum*, *155*(2), 149-165.

Perrier, L., Rouan, L., Jaffuel, S., Clément-Vidal, A., Roques, S., Soutiras, A., ... and Pot, D. (2017). Plasticity of sorghum stem biomass accumulation in response to water deficit: a multiscale analysis from internode tissue to plant level. *Frontiers in Plant Science, 8,* 1516.

Pittelkow, C. M., Liang, X., Linquist, B. A., Van Groenigen, K. J., Lee, J., Lundy, M. E., and van Kessel, C. (2015). Productivity limits and potentials of the principles of conservation agriculture. *Nature*, *517*(7534), 365.

Poorter, H. & Nagel, O. (2000). The role of biomass allocation in the growth response of plants to different levels of light, CO_2, nutrients and water: a quantitative review. *Functional Plant Biology, 27(12),* 1191-1191.

Poorter, H., Niklas, K. J., Reich, P. B., Oleksyn, J., Poot, P., and Mommer, L. (2012). Biomass allocation to leaves, stems and roots: meta-analyses of interspecific variation and environmental control. *New Phytologist*, *193*(1), 30-50.

Porter, D. (2010). Irrigation for small farms Texas AgriLife Research and Extension Service, Amarillo, Texas, United Stated of America. <http://www.twdb.texas.gov/conservation/resources/doc/Irrigation_Manual.pdf > (Accessed 12 July 2020)

Potthoff, M., Dyckmans, J., Flessa, H. & Muhs, A. (2005). Dynamics of maize leaf straw mineralization as affected by the presence of soil and availability of nitrogen. *Soil Biology and Biochemistry, 37*:1259-1266.

Poulton, P., Johnston, J., Macdonald, A., White, R., & Powlson, D. (2018). Major limitations to achieving “4 per 1000” increases in soil organic carbon stock in temperate regions: Evidence from long-term experiments at Rothamsted Research, United Kingdom. *Global Change Biology*, *24*(6), 2563-2584.

Purdy, S. J., Cunniff, J., Maddison, A. L., Jones, L. E., Barraclough, T., Castle, M., ... & Clifton-Brown, J. (2015). Seasonal carbohydrate dynamics and climatic regulation of senescence in the perennial grass, Miscanthus. *BioEnergy Research*, *8*(1), 28-41.

Qiao, Y., Silva, L. C., Han, X., Miao, S., Yan, C. & Horwath, W. R. (2012). Fate of photosynthetically-fixed carbon in soybean crops measured using 13C labelling after long-term fertilization of Phaeozem soils in Northeast China. *Žemdirbystė (Agriculture), 99(4),* 419-424.

Qureshi, A. L., Gadehi, M. A., Mahessar, A. A., Memon, N. A., Soomro, A. G., & Memon, A. H. (2015). Effect of drip and furrow irrigation systems on sunflower yield and water use efficiency in dry area of Pakistan. *American-Eurasian Journal of Agricultural & Environmental Sciences*, *15*(10), 1947-1952.

R Core Development Team (2020). R: A language and environment for statistical computing. R foundation for statistical computing (version 3.6.3). Vienna, Austria. www.cran.r-project.org/ (Accessed 13 January 2020).

Raich, J. W., & Potter, C. S. (1995). Global patterns of carbon dioxide emissions from soils. *Global Biogeochemical Cycles*, *9*(1), 23-36.

Rangel-Castro, J. I., Prosser, J. I., Ostle, N., Scrimgeour, C. M., Killham, K. & Meharg, A. A. (2005). Flux and turnover of fixed carbon in soil microbial biomass of limed and unlimed plots of an upland grassland ecosystem. *Environmental Microbiology, 7(4),* 544-552.

Rasse, D. P., Rumpel, C. & Dignac, M. F. (2005). Is soil carbon mostly root carbon? Mechanisms for a specific stabilization. *Plant and Soil, 269*, 341-356.

Rattey, A., Shorter, R., Chapman, S., Dreccer, F., & van Herwaarden, A. (2009). Variation for and relationships among biomass and grain yield component traits conferring improved yield and grain weight in an elite wheat population grown in variable yield environments. *Crop and Pasture Science*, *60*(8), 717-729.

Reichert, J. M., Albuquerque, J. A., Kaiser, D. R., Reinert, D. J., Urach, F. L., & Carlesso, R. (2009). Estimation of water retention and availability in soils of Rio Grande do Sul. *Revista Brasileira de Ciência do Solo*, *33*(6), 1547-1560.

Remus, R., & Augustin, J. (2016). Dynamic linking of 14C partitioning with shoot growth allows a precise determination of plant-derived C input to soil. *Plant and Soil, 408(1-2),* 493-513.

Reynolds, M., Foulkes, M. J., Slafer, G. A., Berry, P., Parry, M. A., Snape, J. W., and Angus, W. J. (2009). Raising yield potential in wheat. *Journal of Experimental Botany*, *60*(7), 1899-1918.

Rezaei, E. E., Siebert, S., Manderscheid, R., Müller, J., Mahrookashani, A., Ehrenpfordt, B. and Ewert, F. (2018). Quantifying the response of wheat yields to heat stress: The role of the experimental setup. *Field Crops Research, 217*, 93-103.

Riboldi, L. B., Oliveira, R. F., and Angelocci, L. R. (2016). Leaf turgor pressure in maize plants under water stress. *Australian Journal of Crop Science, 10(6)*, 878.

Romero-Olivares, A. L., Allison, S. D., and Treseder, K. K. (2017). Decomposition of recalcitrant carbon under experimental warming in boreal forest. *PloS one, 12(6)*, e0179674.

Rooney, W. L. (2014). Sorghum. Cellulosic Energy Cropping Systems. John Wiley and Sons, UK, 109-129.

Roy, P., Basu, S., Chatterjee, J., Chakraborty, I., Dutta, R., Barik, S., & Goswami, A. (2019). Management of low nitrogen input with potassium and phosphorus fertilizers for cropping system and yield of Sweet Sorghum Crop (Sorghum bicolour L. Moench) in a field experiment at Panskura, East Midnapore, West Bengal. *Indian Journal of Agricultural Research*, *53*(1), 73-77.

Ruggiero, A., Punzo, P., Landi, S., Costa, A., Van Oosten, M. J., & Grillo, S. (2017). Improving plant water use efficiency through molecular genetics. *Horticulturae*, *3*(2), 31.

Rusere, F., Soropa, G., Svubure, O., Gwatibaya, S., Moyo, D., Ndeketeya, A., & Mavima, G. A. (2012). Effects of deficit irrigation on winter silage maize production in Zimbabwe. *International Research Journal of Plant Science*, *3*(9), 188-192.

Russell, J. E. (2002). Soil conditions and plants growth. Daya Books.

Sadras, V. O., McDonald, G., Sadras, V. O., & McDonald, G. (2012). Water use efficiency of grain crops in Australia: principles, benchmarks and management. *Change*, *11*(19), 24.

Sage, R. F. & Monson, R. K. (1998). C4 plant biology. Academic Press, California, USA 92101-4495.

Salih, A. A., Ali, I. A., Lux, A., Luxova, M., Cohen, Y., Sugimoto, Y., & Inanaga, S. (1999). Rooting, water uptake, and xylem structure adaptation to drought of two sorghum cultivars. *Crop Science*, *39*(1), 168-173.

Sánchez, B., Rasmussen, A., and Porter, J. R. (2014). Temperatures and the growth and development of maize and rice: a review. *Global change biology*, *20*(2), 408-417.

Sanderman, J., Hengl, T., & Fiske, G. J. (2017). Soil carbon debt of 12,000 years of human land use. *Proceedings of the National Academy of Sciences*, *114*(36), 9575-9580.

Sareen, S., Tyagi, B. S., Sarial, A. K., Tiwari, V., and Sharma, I. (2014). Trait analysis, diversity, and genotype x environment interaction in some wheat landraces evaluated under drought and heat stress conditions. *Chilean Journal of Agricultural Research*, *74*(2), 135-142.

Schmitt, A., Pausch, J., & Kuzyakov, Y. (2013). C and N allocation in soil under ryegrass and alfalfa estimated by 13 C and 15 N labelling. *Plant and Soil*, *368*(1), 581-590.

Schulze, J., & Pöschel, G. (2004). Bacterial inoculation of maize affects carbon allocation to roots and carbon turnover in the rhizosphere. *Plant and Soil*, *267*(1), 235-241.

Scientific Thermo Fisher (2014). Gallery. Sunnyvale, California. USA.

Shah, N. H., & Paulsen, G. M. (2003). Interaction of drought and high temperature on photosynthesis and grain-filling of wheat. *Plant and Soil*, *257*(1), 219-226.

Shamuyarira, K. W., Shimelis, H. A., Mathew, I., and Tsilo, T. J. (2019). Correlation and path coefficient analyses of yield and yield components in drought-tolerant bread wheat populations. *South African Journal of Plant and Soil, 36(5)*, 367-374.

Sharma, B., Molden, D., & Cook, S. (2015). *Water use efficiency in agriculture: Measurement, current situation and trends* (No. 612-2016-40604).

Sharp, R. E., & Davies, W. J. (1985). Root growth and water uptake by maize plants in drying soil. *Journal of Experimental Botany*, *36*(9), 1441-1456.

Sheehan, J. J., Adler, P. R., Del Grosso, S. J., Easter, M., Parton, W., Paustian, K., & Williams, S. (2014). CO 2 emissions from crop residue-derived biofuels. *Nature Climate Change*, *4*(11), 932-933.

Sherwood, S. C., & Huber, M. (2010). An adaptability limit to climate change due to heat stress. *Proceedings of the National Academy of Sciences*, *107*(21), 9552-9555.

Siddique, K. H. M., Belford, R. K., and Tennant, D. (1990). Root: shoot ratios of old and modern, tall and semi-dwarf wheats in a Mediterranean environment. *Plant and soil*, *121*(1), 89-98.

Sinclair, T. R. (2018). Effective water use required for improving crop growth rather than transpiration efficiency. *Frontiers in Plant Science*, *9*, 1442.

Sivakumar, M. V. K., & Salaam, S. A. (1999). Effect of year and fertilizer on water-use efficiency of pearl millet (*Pennisetum glaucum*) in Niger. *Journal of Agricultural Science*, *132*(2), 139-148.

Sivamani, E., Bahieldin, A., Wraith, J. M., Al-Niemi, T., Dyer, W. E., Ho, T. H. D., & Qu, R. (2000). Improved biomass productivity and water use efficiency under water deficit conditions in transgenic wheat constitutively expressing the barley HVA1 gene. *Plant Science*, *155*(1), 1-9.

Smale, M., Byerlee, D., and Jayne, T. (2011). Maize revolutions in sub-Saharan Africa. The World Bank.

Sorkheh, K., Shiran, B., Gradziel, T. M., Epperson, B. K., Martínez-Gómez, P., and Asadi, E. (2007). Amplified fragment length polymorphism as a tool for molecular characterization of almond germplasm: genetic diversity among cultivated genotypes and related wild species of almond, and its relationships with agronomic traits. *Euphytica*, *156*(3), 327-344.

Srinivasan, V., Lambin, E. F., Gorelick, S. M., Thompson, B. H., & Rozelle, S. (2012). The nature and causes of the global water crisis: Syndromes from a meta-analysis of coupled human-water studies. *Water Resources Research*, *48*(10).

Staddon, P. L. (2004). Carbon isotopes in functional soil ecology. *Trends in Ecology and Evolution, 19(3)*, 148-154.

Stokes, J., Panagioti, M., Alam, R., Checkland, K., Cheraghi-Sohi, S., and Bower, P. (2015). Effectiveness of case management for 'at risk' patients in primary care: a systematic review and meta-analysis. *PloS one, 10(7)*, e0132340.

Stone, L. R., Schlegel, A. J., Gwin Jr, R. E., & Khan, A. H. (1996). Response of corn, grain sorghum, and sunflower to irrigation in the High Plains of Kansas. *Agricultural Water Management*, *30*(3), 251-259.

Studer, M. S., Siegwolf, R. T., & Abiven, S. (2014). Carbon transfer, partitioning and residence time in the plant-soil system: a comparison of two $^{13}CO_2$ labelling techniques. *Biogeosciences*, *11*(6), 1637-1648.

Su, Z., Zhang, J., Wu, W., Cai, D., Lv, J., Jiang, G., ... & Gabriels, D. (2007). Effects of conservation tillage practices on winter wheat water-use efficiency and crop yield on the Loess Plateau, China. *Agricultural Water Management*, *87*(3), 307-314.

Swinnen, J., Van Veen, J. A., & Merckx, R. (1995). Root decay and turnover of rhizodeposits in field-grown winter wheat and spring barley estimated by 14C pulse-labelling. *Soil Biology and Biochemistry*, *27*(2), 211-217.

Swinnen, J., Van Veen, J. A., & Merckx, R. (1995a). Carbon fluxes in the rhizosphere of winter wheat and spring barley with conventional vs integrated farming. *Soil Biology and Biochemistry*, *27*(6), 811-820.

Systat Software. (2008). SigmaPlot for Windows. Version 11.0.

Tambussi, E. A., Bort, J., Guiamet, J. J., Nogués, S., & Araus, J. L. (2007). The photosynthetic role of ears in C3 cereals: metabolism, water use efficiency and contribution to grain yield. *Critical Reviews in Plant Sciences*, *26*(1), 1-16.

Tardieu, F. (2013). Plant response to environmental conditions: assessing potential production, water demand, and negative effects of water deficit. *Frontiers in Physiology*, *4*, 17.

Tari, A. F. (2016). The effects of different deficit irrigation strategies on yield, quality, and water-use efficiencies of wheat under semi-arid conditions. *Agricultural Water Management*, *167*, 1-10.

Tatar, Ö. (2016). Climate change impacts on crop production in Turkey. *Agronomy Series of Scientific Research*, *59*(2), 135-140.

Taylor, J. R. N. (2003, April). Overview: Importance of sorghum in Africa. In *Afripro: Workshop on the Proteins of Sorghum and Millets: Enhancing Nutritional and Functional Properties for Africa, Pretoria* (Vol. 2, No. 4).

TerAvest, D., Carpenter-Boggs, L., Thierfelder, C., & Reganold, J. P. (2015). Crop production and soil water management in conservation agriculture, no-till, and conventional tillage systems in Malawi. *Agriculture, Ecosystems & Environment*, *212*, 285-296.

Thorup-Kristensen, K., Cortasa, M. S., and Loges, R. (2009). Winter wheat roots grow twice as deep as spring wheat roots, is this important for N uptake and N leaching losses? *Plant and Soil*, *322*(1-2), 101-114.

Tian, J., Pausch, J., Fan, M., Li, X., Tang, Q., & Kuzyakov, Y. (2013). Allocation and dynamics of assimilated carbon in rice-soil system depending on water management. *Plant and Soil*, *363*(1), 273-285.

Tilman, D., Cassman, K. G., Matson, P. A., Naylor, R., & Polasky, S. (2002). Agricultural sustainability and intensive production practices. *Nature*, *418*(6898), 671-677.

Tine, F. (2016). Evaluation of Drought Tolerance and Forage Characteristics of Millet Cultivars (Doctoral dissertation, Virginia Tech).

Tolk, J. A., & Howell, T. A. (2003). Water use efficiencies of grain sorghum grown in three USA southern Great Plains soils. *Agricultural Water Management*, *59*(2), 97-111.

Trumbore, S. (2006). Carbon respired by terrestrial ecosystems – recent progress and challenges. *Global Change Biology*, *12*(2), 141-153.

Van Soest, P. V., Robertson, J. B., & Lewis, B. (1991). Methods for dietary fiber, neutral detergent fiber, and nonstarch polysaccharides in relation to animal nutrition. *Journal of dairy science*, *74*(10), 3583-3597.

Vanaja, M., Yadav, S. K., Archana, G., Lakshmi, N. J., Reddy, P. R., Vagheera, P., and Venkateswarlu, B. (2011). Response of C4 (maize) and C3 (sunflower) crop plants to drought stress and enhanced carbon dioxide concentration. *Plant, Soil and Environment*, *57*(5), 207-215.

Vandecasteele, B., Muylle, H., De Windt, I., Van Acker, J., Ameloot, N., Moreaux, K., ... & Debode, J. (2018). Plant fibers for renewable growing media: Potential of defibration, acidification or inoculation with biocontrol fungi to reduce the N drawdown and plant pathogens. *Journal of Cleaner Production*, *203*, 1143-1154.

Vasilakoglou, I., Dhima, K., Karagiannidis, N., and Gatsis, T. (2011). Sweet sorghum productivity for biofuels under increased soil salinity and reduced irrigation. *Field Crops Research*, *120*(1), 38-46.

Velthof, G. L., Kuikman, P. J., & Oenema, O. (2002). Nitrous oxide emission from soils amended with crop residues. *Nutrient Cycling in Agroecosystems*, *62*(3), 249-261.

Viets, F. G. (1962). Fertilizers and the efficient use of water. *Advances in Agronomy,* 14, 223-264.

Villegas-Pangga, G., Blair, G., & Lefroy, R. (2000). Measurement of decomposition and associated nutrient release from straw (Oryza sativa L.) of different rice varieties using a perfusion system. *Plant and Soil*, *223*(1), 1-11.

Waines, J. G. (2012). Determination of optimum root and shoot size in bread wheat for increased water and nutrient use efficiency and grain yield. Report to California Wheat Commission: GH 2011-2012 Experiments. California Wheat Commission.

Walli, T. K., Ørskov, E. R., & Bhargava, P. K. (1988). Rumen degradation of straw 3. Botanical fractions of two rice straw varieties and effects of ammonia treatment. *Animal Science*, *46*(3), 347-352.

Wang, L., Palta, J. A., Chen, W., Chen, Y., & Deng, X. (2018). Nitrogen fertilization improved water-use efficiency of winter wheat through increasing water use during vegetative rather than grain filling. *Agricultural Water Management*, *197*, 41-53.

Wang, W. J., Baldock, J. A., Dalal, R. C., & Moody, P. W. (2018). Decomposition dynamics of plant materials in relation to nitrogen availability and biochemistry determined by NMR and wet-chemical analysis. *Soil Biology and Biochemistry*, *36*(12), 2045-2058.

Warembourg, F. R., & Paul, E. A. (1977). Seasonal transfers of assimilated 14C in grassland: plant production and turnover, soil and plant respiration. *Soil Biology and Biochemistry*, *9*(4), 295-301.

Warembourg, F. R., Roumet, C., & Lafont, F. (2003). Differences in rhizosphere carbon-partitioning among plant species of different families. *Plant and Soil*, *256*(2), 347-357.

Wasson, A. P., Richards, R. A., Chatrath, R., Misra, S. C., Prasad, S. S., Rebetzke, G. J., ... & Watt, M. (2012). Traits and selection strategies to improve root systems and water uptake in water-limited wheat crops. *Journal of Experimental Botany*, *63*(9), 3485-3498.

Water Research Commission (WRC) (2002) Technical Report 1999. Water Research Commission, Pretoria.

Way, D. A., Katul, G. G., Manzoni, S., & Vico, G. (2014). Increasing water use efficiency along the C3 to C4 evolutionary pathway: a stomatal optimization perspective. *Journal of Experimental Botany*, *65*(13), 3683-3693.

Webber, H. A., Madramootoo, C. A., Bourgault, M., Horst, M. G., Stulina, G., & Smith, D. L. (2006). Water use efficiency of common bean and green gram grown using alternate furrow and deficit irrigation. *Agricultural Water Management*, *86*(3), 259-268.

Wegener, F., Beyschlag, W., & Werner, C. (2015). Dynamic carbon allocation into source and sink tissues determine within-plant differences in carbon isotope ratios. *Functional Plant Biology*, *42*(7), 620-629.

Werth, M., & Kuzyakov, Y. (2006). Assimilate partitioning affects 13 C fractionation of recently assimilated carbon in maize. *Plant and Soil*, *284*(1), 319-333.

Wilhelm, W. W., Johnson, J. M., Lightle, D. T., Karlen, D. L., Novak, J. M., Barbour, N. W., ... & Arriaga, F. (2011). Vertical distribution of corn stover dry mass grown at several US locations. *BioEnergy Research*, *4*(1), 11-21.

Wu, W. X., Liu, W., Lu, H. H., Chen, Y. X., Devare, M., & Thies, J. (2008). Use of 13C labeling to assess carbon partitioning in transgenic and nontransgenic (parental) rice and their rhizosphere soil microbial communities. *FEMS microbiology Ecology*, *67*(1), 93-102.

Wu, Y., Tan, H., Deng, Y., Wu, J., Xu, X., Wang, Y., ... & Cui, X. (2010). Partitioning pattern of carbon flux in a Kobresia grassland on the Qinghai-Tibetan Plateau revealed by field 13C pulse-labeling. *Global Change Biology*, *16*(8), 2322-2333.

Xie, Z. K., Wang, Y. J., & Li, F. M. (2005). Effect of plastic mulching on soil water use and spring wheat yield in arid region of northwest China. *Agricultural Water Management*, *75*(1), 71-83.

Xu, Z., Zhou, G., & Shimizu, H. (2010). Plant responses to drought and rewatering. *Plant Signaling & Behavior*, *5*(6), 649-654.

Yamori, W., Hikosaka, K., & Way, D. A. (2014). Temperature response of photosynthesis in C 3, C 4, and CAM plants: temperature acclimation and temperature adaptation. *Photosynthesis Research*, *119*(1), 101-117.

Yang, C. H., Chai, Q., and Huang, G. B. (2010). Root distribution and yield responses of wheat/maize intercropping to alternate irrigation in the arid areas of northwest China. *Plant Soil Environ*, *56*(6), 253-262.

Yang, Y., Fang, J., Ji, C., Ma, W., Mohammat, A., Wang, S. and Smith, P. (2012). Widespread decreases in topsoil inorganic carbon stocks across China's grasslands during 1980s-2000s. *Global Change Biology*, *18*(12), 3672-3680.

Yang, Y., Xu, W., Hou, P., Liu, G., Liu, W., Wang, Y. and Li, S. (2019). Improving maize grain yield by matching maize growth and solar radiation. *Scientific Reports, 9(1)*, 1-11.

Zegada-Lizarazu, W., Zatta, A., & Monti, A. (2012). Water uptake efficiency and above-and belowground biomass development of sweet sorghum and maize under different water regimes. *Plant and Soil*, *351*(1), 47-60.

Zhang, D., Hui, D., Luo, Y., & Zhou, G. (2008). Rates of litter decomposition in terrestrial ecosystems: global patterns and controlling factors. *Journal of Plant Ecology*, *1*(2), 85-93.

Zhang, J., Tang, X., Zhong, S., Yin, G., Gao, Y., & He, X. (2017). Recalcitrant carbon components in glomalin-related soil protein facilitate soil organic carbon preservation in tropical forests. *Scientific Reports*, *7*(1), 1-9.

Zhang, X., Wang, Y., Sun, H., Chen, S., & Shao, L. (2014). Optimizing the yield of winter wheat by regulating water consumption during vegetative and reproductive stages under limited water supply. *Irrigation Science*, *31*(5), 1103-1112.

Zhao, J., Xue, Q., Jessup, K. E., Hao, B., Hou, X., Marek, T. H., ... & Brauer, D. K. (2018). Yield and water use of drought-tolerant maize hybrids in a semiarid environment. *Field Crops Research*, *216*, 1-9.

Zheng, J., Fan, J., Zhang, F., Yan, S., Guo, J., Chen, D., & Li, Z. (2018). Mulching mode and planting density affect canopy interception loss of rainfall and water use efficiency of dryland maize on the Loess Plateau of China. *Journal of Arid Land*, *10*(5), 794-808.

Zhou, W., Lv, T. F., Chen, Y., Westby, A. P., & Ren, W. J. (2014). Soil physicochemical and biological properties of paddy-upland rotation: a review. *The Scientific World Journal*, *2014*. https://doi.org/10.1155/2014/856352.

Zwart, S. J., & Bastiaanssen, W. G. (2004). Review of measured crop water productivity values for irrigated wheat, rice, cotton and maize. *Agricultural Water Management*, *69*(2), 115-133.

APPENDICES

Appendix Table 1 List of 100 wheat genotypes and their pedigree used in the study

ENTRY CODE	PEDIGREE
	Genotypes from CIMMYT Heat Stress Tolerance Nursery
LM01	ACHTAR*3//KANZ/KS85-8-5/4/MILAN/KAUZ//PRINIA/3/BAV92/5/MILAN/KAUZ//PRINIA/3/BAV92
LM12	SOKOLL/ROLF07
LM14	MILAN/KAUZ//PRINIA/3/BAV92/4/WBLL1*2/KUKUNA
LM15	RL6043/4*NAC//PASTOR/3/BAV92/4/ATTILA/BAV92//PASTOR
LM16	PASTOR*2/BAV92/3/FRET2/KUKUNA//FRET2
LM17	ESDA/KKTS
LM18	GOUBARA-1/2*SOKOLL
LM19	SOKOLL*2/4/CHEN/AEGILOPS SQUARROSA (TAUS)//FCT/3/STAR
LM20	PBW343
LM21	PRL/2*PASTOR
LM22	MUNAL #1
LM23	QUAIU
LM24	WBLL1*2/BRAMBLING
LM25	WHEAR//2*PRL/2*PASTOR
LM26	ATTILA*2/PBW65//TAM200/TUI
LM27	YUNMAI 48//2*WBLL1*2/KURUKU
LM28	ATTILA/3*BCN//BAV92/3/TILHI/4/SHA7/VEE#5//ARIV92
LM29	PRL/2*PASTOR*2//SKAUZ/BAV92
LM30	C80.1/3*BATAVIA//2*WBLL1/3/ATTILA/3*BCN*2//BAV92/4/WBLL1*2/KURUKU
LM31	ATTILA*2/HUITES//FINSI/3/ATTILA*2/PBW65
LM32	ATTILA*2//CHIL/BUC*2/3/KUKUNA
LM33	ATTILA*2/PBW65//KACHU
LM35	WBLL1//UP2338*2/VIVITSI
LM36	WBLL1*2/4/SNI/TRAP#1/3/KAUZ*2/TRAP//KAUZ/5/KACHU
LM37	KACHU/SAUAL
LM38	SAUAL/3/MILAN/S87230//BAV92
LM39	ATTILA/3*BCN//BAV92/3/TILHI/5/BAV92/3/PRL/SARA//TSI/VEE#5/4/CROC_1/AE.SQUARROSA (224)//2*OPATA
LM40	WBLL1*2/VIVITSI/6/CNDO/R143//ENTE/MEXI_2/3/AEGILOPS SQUARROSA (TAUS)/4/WEAVER/5/2*JANZ
LM41	C80.1/3*BATAVIA//2*WBLL1/5/REH/HARE//2*BCN/3/CROC_1/AE.SQUARROSA (213)//PGO/4/HUITES
LM42	TRCH/5/REH/HARE//2*BCN/3/CROC_1/AE.SQUARROSA (213)//PGO/4/HUITES
LM43	ROLF07*2/6/PVN//CAR422/ANA/5/BOW/CROW//BUC/PVN/3/YR/4/TRAP#1
LM44	ROLF07/TUKURU/5/WBLL1*2/4/YACO/PBW65/3/KAUZ*2/TRAP//KAUZ
LM46	FRET2/KUKUNA//FRET2/3/PARUS/5/FRET2*2/4/SNI/TRAP#1/3/KAUZ*2/TRAP//KAUZ
LM47	FRET2/KUKUNA//FRET2/3/YANAC/4/FRET2/KIRITATI
LM48	FRET2/KUKUNA//FRET2/3/PASTOR//HXL7573/2*BAU/5/FRET2*2/4/SNI/TRAP#1/3/KAUZ*2/TRAP//KAUZ
LM49	TRCH/SRTU//KACHU
LM50	HUW234+LR34/PRINIA*2//SNLG
LM51	HUW234+LR34/PRINIA*2//YANAC
LM52	HUW234+LR34/PRINIA*2//WHEAR
LM54	PBW343*2/KUKUNA*2//KITE
LM55	PBW343*2/KUKUNA//PARUS/3/PBW343*2/KUKUNA
LM56	PBW343*2/KUKUNA*2//YANAC
LM57	PBW343*2/KUKUNA//SRTU/3/PBW343*2/KHVAKI
LM58	ATTILA*2/PBW65/6/PVN//CAR422/ANA/5/BOW/CROW//BUC/PVN/3/YR/4/TRAP#1/7/ATTILA/2*PASTOR
LM59	FRET2/KUKUNA//FRET2/3/WHEAR/4/FRET2/TUKURU//FRET2

Table S1 Continued

ENTRY CODE	PEDIGREE
LM60	ALD/CEP75630//CEP75234/PT7219/3/BUC/BJY/4/CBRD/5/TNMU/PF85487/6/PBW343*2/KUKUNA/7/CNO79//PF 70354/MUS/3/PASTOR/4/BAV92
	Genotypes from CIMMYT Drought Stress Tolerance Nursery
LM71	BABAX/3/PRL/SARA//TSI/VEE#5/4/CROC_1/AE.SQUARROSA (224)//2*OPATA
LM72	BABAX/3/PRL/SARA//TSI/VEE#5/4/WBLL1
LM75	BUC/MN72253//PASTOR
LM76	MILAN/KAUZ//PRINIA/3/BABAX
LM77	CNDO/R143//ENTE/MEXI_2/3/AEGILOPS SQUARROSA (TAUS)/4/WEAVER/5/2*FRAME
LM79	CROC_1/AE.SQUARROSA (205)//BORL95/3/KENNEDY
LM80	CROC_1/AE.SQUARROSA (205)//KAUZ/3/SLVS
LM81	CROC_1/AE.SQUARROSA (224)//2*OPATA/3/2*RAC655
LM82	HD30/5/CNDO/R143//ENTE/MEXI75/3/AE.SQ/4/2*OCI
LM83	PASTOR/3/VEE#5//DOVE/BUC
LM84	SRN/AE.SQUARROSA (358)//MILAN/SHA7
LM85	SW94.60002/4/KAUZ*2//DOVE/BUC/3/KAUZ/5/SW91-12331
LM86	CHAM 6
LM90	CROC_1/AE.SQUARROSA (205)//BORL95/3/KENNEDY-2
LM91	FRTL/CMH83.2517
LM93	PASTOR/FLORKWA.1//PASTOR
LM96	ALTAR 84/AE.SQ//2*OPATA/3/PIFED
LM97	KRICHAUFF/2*PASTOR
LM98	KABY//2*ALUBUC/BAYA
LM99	ALTAR 84/AEGILOPS SQUARROSA (TAUS)//OCI/3/VEE/MJI//2*TUI
LM100	SW89.5277/BORL95//SKAUZ
	Genotypes from CIMMYT Heat Stress Tolerance Nursery
BW28	CMSA05Y01011T-040M-040ZTP0Y-040ZTM-040SY-14ZTM-03Y-0B
BW48	CMSA04M00346S-040ZTP0Y-040ZTM-040SY-27ZTM-04Y-0B
BW49	CMSA04M00346S-040ZTP0Y-040ZTM-040SY-28ZTM-01Y-0B
BW58	CMSA04M00067S-040ZTB-040ZTY-040ZTM-040SY-2ZTM-02Y-0B
BW63	CMSA04M01020T-050Y-040ZTP0M-040ZTY-040ZTM-040SY-5ZTM-03Y-0B
BW71	CMSA05Y00325S-040ZTP0Y-040ZTM-040SY-7ZTM-01Y-0B
BW103	CMSS05B00581S-099Y-099M-099Y-099ZTM-2WGY-0B
BW111	CMSS05B00663S-099Y-099M-099Y-099ZTM-13WGY-0B
BW116	CMSS05B00742S-099Y-099M-099Y-099ZTM-5WGY-0B
BW124	CGSS05B00153T-099TOPY-099M-099NJ-22WGY-0B
BW127	CGSS05B00162T-099TOPY-099M-099Y-099ZTM-15WGY-0B
BW128	CGSS05B00162T-099TOPY-099M-099NJ-13WGY-0B
BW129	CGSS05B00162T-099TOPY-099M-099NJ-099NJ-7WGY-0B
BW141	CGSS05B00243T-099TOPY-099M-099NJ-099NJ-1WGY-0B
BW142	CGSS05B00243T-099TOPY-099M-099NJ-099NJ-2WGY-0B
BW145	CGSS05B00253T-099TOPY-099M-099Y-099ZTM-8WGY-0B
BW147	CGSS05B00256T-099TOPY-099M-099NJ-099NJ-5WGY-0B
BW148	CGSS05B00258T-099TOPY-099M-099Y-099ZTM-3WGY-0B
BW149	CGSS05B00258T-099TOPY-099M-099Y-099ZTM-11WGY-0B
BW150	CGSS05B00258T-099TOPY-099M-099Y-099ZTM-12WGY-0B
BW151	CGSS05B00258T-099TOPY-099M-099Y-099ZTM-13WGY-0B
BW152	CGSS05B00258T-099TOPY-099M-099NJ-1WGY-0B
BW157	CGSS05B00261T-099TOPY-099M-099NJ-099NJ-8WGY-0B

Table S1 Continued

ENTRY CODE	PEDIGREE
BW159	CGSS05B00290T-099TOPY-099M-099NJ-099NJ-7WGY-0B
BW162	CGSS05B00304T-099TOPY-099M-099NJ-099NJ-3WGY-0B

	Local Checks
LM70	Check
BW80	Check
BW100	Check
BW120	Check
BW140	Check
	Temperate checks
Arenza	Check
Sossognon	Check
Triticale	Check