Genetic Improvement of Pigeonpea [*Cajanus cajan* (L.) Millspaugh] for Yield, Earliness and Resistance to *Fusarium* Wilt (*Fusarium udum* Butler) in Malawi

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2020

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A Thesis Submitted in Fulfilment of the Requirements for the Degree of Doctor of Philosophy (PhD) in Plant Breeding

African Center for Crop Improvement

School of Agricultural, Earth and Environmental Sciences

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University of KwaZulu-Natal

Republic of South Africa

November 2020

THESIS ABSTRACT

Pigeonpea [*Cajanus cajan* (L.) Millspaugh, 2n=2x=22] is a one of the important food legumes in Sub-Saharan Africa and Asia. Malawi is a major pigeonpea grower in Africa with production of 403,519 tonnes produced in 248,400 hactares. Pigeonpea is good source of protein and cash income to millions of farmers. Pigeonpea crop residues form excellent animal feed. It serves in atmospheric nitrogen fixation and biomass allocation in the soil. Despite Malawi being the highest pigeonpea producer, grain yield of pigeonpea is low (< 700 kg ha^{-1}) compared with the potential yield of the crop (2000 kg ha^{-1}). The yield gap is due to various production constraints, including *Fusarium* wilt disease, insect pests, and lack of early maturing and high yielding varieties that are photoperiod insensitive. Breeding and deployment of high yielding, early maturing, and *Fusarium* wilt resistant cultivars have the potential to enhance pigeonpea production and productivity. The overall objective of this study was to contribute to food security in Malawi through breeding high yielding and farmer-preferred pigeonpea varieties. The specific objectives were: (1) to determine the production constraints affecting pigeonpea, and to identify farmer-preferred traits in Malawi to guide future breeding of pigeonpea; (2) to determine the diversity among pigeonpea germplasm collections using agro-morphological traits to enable selection of genetically distinct lines for breeding; (3) to determine the genetic diversity among the tested pigeonpea germplasm, using single nucleotide polymorphism (SNP) markers to select genetically distinct lines for breeding; (4) to determine the combining ability effects and gene action controlling agro-morphological traits and resistance to *Fusarium* wilt; and to select the best parents and families from the test population for further breeding.

In the first study, a participatory rural appraisal study was conducted in four major pigeonpea-growing districts in southern Malawi (Chiradzulu, Mulanje, Thyolo and Zomba), using a semi-structured questionnaire, transect walks and focus group discussions (FGDs). The results revealed that a landrace pigeonpea variety, 'Mthawajuni', was preferred by famers due to its positive attributes such as good taste, early to medium maturity, short cooking time and tolerant to pod borer *(Helicoverpa armigera* Hubner). Pigeonpea trait preference was dependent on gender, with female respondents preferring rapid cooking, early maturity, long storage and good pest resistance, whereas men focussed on high yields, large seed size, cream seed colour and disease resistance. The study identified the pod borer *(H. armigera*), *Fusarium* wilt disease

(*Fusarium udum* Butler), low yields of the existing varieties, drought, and unreliable market prices as the leading challenges affecting pigeonpea production in southern Malawi.

A second part of the study focused on phenotypic and genetic diversity and yield stability analyses among pigeonpea accessions in selected target production environments, as a basis to select complementary and unique genotypes for breeding. Eighty-one pigeonpea genotypes were evaluated in six environments in Malawi using a 9 × 9 alpha-lattice design with two replications. Significant genotype variation were recorded for qualitative traits including flower colour, flower streak pattern, pod colour, seed coat colour pattern, seed coat main colour, seed shape and seed eye colour. All evaluated quantitative traits initially were significantly affected by genotype × environment interaction effects except the number of seeds per pod. Genotypes MWPLR 14, ICEAP 01170, ICEAP 871091 and ICEAP 01285 were identified as early maturing varieties, maturing in 125 to 137 days. The genotypes Kachangu, MWPLR 16, TZA 5582, No. 40 and MWPLR 14 had the highest number of pods per plant (NPP) and highest grain yields (GYD). Grain yield was positively and significantly correlated with days to flowering (DTF) ($r=0.23$, $p<0.01$), NPP ($r=0.35$, $p<0.01$) and hundred seed weight (HSWT) ($r=0.50$, $p<0.01$), suggesting the usefulness of these traits for selection to enhance grain yield improvement when assessing pigeonpea populations. Using principal component analysis, three principal components (PCs) accounted for 57.7% of the total variation. The most important traits that reliably discriminated between the test genotypes were DTF, days to maturity (DTM), number of primary (NPB) and secondary branches (NSB), HSWT and GYD. Genotype, environment and genotype × environment interaction accounted for 16.4, 33.5 and 49.6% to the total variation for quantitative traits, respectively. The test environments were delineated into three mega-environments, based on site and seasonal variability. MWPLR 14 (G51), MWPLR 24 (G26) and ICEAP 01155 (G27) were the most stable genotypes for yield across environments, while MWPLR 14, TZA 5582 and MWPLR 4 were the highest yielding genotypes across environments. To broaden the genetic base of the pigeonpea for selection, divergent genotypes such as MWPRL 14, TZA 5582, MWPLR 4, MWPLR 16, Sauma and Kachangu are recommended as parents for targeted crosses.

The fourth part of the study examined genetic relationships among 81 genotypyes using 4122 single nucleotide polymorphism (SNP) markers. The SNP markers also confirmed the genetic diversity among the genotypes. The mean gene diversity and the polymorphic information content (PIC) were 0.14 and 0.11, suggesting moderate genetic differentiation among the

genotypes. The low genetic diversity and PIC could hinder genetic gains in future pigeonpea breeding programs using this population. The genotypes were delineated into three groups based on population structure and the joint analysis of the phenotypic and genotypic data. The analysis of molecular variance (AMOVA) revealed that differences among clusters accounted for only 2.7% of the variation, while within-cluster variation among individuals accounted for 97.3% of the variation. This suggested that unique breeding populations could be created by identifying and selecting divergent individuals as parental lines. There is a need to create new genetic variation or introgress genes from close relatives to increase the genetic base of pigeonpea since the available genetic variability may not meet the demand for improved cultivars. The phenotypic diversity assessment using morphological attributes grouped the genotypes into three distinct clusters. The mean gene diversity and polymorphic information content were 0.14 and 0.11, respectively, suggesting moderate genetic differentiation among the genotypes. The genotypes were delineated into three heterotic groups based on population structure and the joint analysis of the phenotypic and genotypic data, suggesting the possibility of creating unique breeding populations through targeted crosses of parents from divergent heterotic groups.

In a third study, the best and most diverse genotypes from the diversity studies with early maturity, *Fusarium* wilt (FW) resistance from previous studies and farmer-preferred traits were selected for crosses. Finally, the ten selected parental lines were crossed using a factorial mating design and 25 progenies were successfully developed. The parents and progenies were field evaluated at two locations; 1) Chitedze Agricultural Research Station and 2) Makoka Agricultural Research Station in Malawi. The trial design was 7 × 5 alpha lattice design with two replications. The test genotypes were evaluated for FW resistance through a root dip inoculation technique. There was significant genetic variation among parental lines and families for days to 50% flowering (DTF), days to 75% maturity (DTM), plant height (PH), 100 seed weight (HSWT), FW resistance, and grain yield (GYD). Parental lines, ICEAP 87105, and ICEAP 01285 had desirable general combining ability (GCA) (-32.90 and -14.16 respectively) for days to 75% maturity (DTM), parental lines, MWPLR 16, Sauma and Mwaiwathualimi had desirable GCA (319.11, 168.8 and 46.45 respectively) for grain yield (GYD) and parental lines, TZA 5582, ICEAP 00554, Mwayiwathualimi and Sauma had desirable GCA effects (-3.16, -0.54, -0.24 and 0.17 respectively) for FW resistance. Hybrids such as TZA 5582 × MWPLR 22, TZA 5582 × MWPLR 14, and Mwayiwathualimi × MWPLR 22 had desirable specific combining

ability (SCA) effects for DTM (-1.22 -1.51 and -0.91 respectively), GYD (80.93, 42.67 and 79.55 respectively) and FW resistance (-1.10, -0.15, and -1.66 respectively). The study further revealed that additive gene effects were important in inheritance of DTF, DTM and PH traits and non-additive gene effects were important in inheritance of GYD, 100 seed weight (HSWT) and FW resistance. This suggest that both pedigree and recurrent selection are important to achieve pigeonpea improvement. Overall, this study determined the present pigeonpea production constraints and farmer-preferred traits in Malawi. Further, significant genetic variations were detected among a diverse set of pigeonpea germplasm for breeding early maturing/short-duration, high yielding and FW resistant varieties. The study developed new breeding populations based on selected complementary parents for variety development and release in Malawi.

DECLARATION

I, Esnart Nyirenda Yohane, declare the following

1. The research reported in this thesis, except where otherwise indicated, is my original research.
2. This thesis has not been submitted for any degree or examination at any other University.
3. This thesis does not contain other persons' data, pictures, graphs or other information, unless specifically acknowledged as being sourced from other persons.
4. This thesis does not contain other persons' writing, unless specifically acknowledged as being sourced from other researchers. Where other written sources have been quoted, then:

a) Their words have been re-written but the general information attributed to them has been referenced.

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Signed:

[redacted]

[redacted]……

Esnart Nyirenda Yohane

As the candidate's supervis[redacted]r, I agree to the submission of this dissertation

[redacted]

……

Prof. H. Shimelis (Supervisor)

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Prof. M. Laing (Co-supervisor)

ACKNOWLEDGEMENTS

I would like to thank God Almighty for providing me the opportunity to attain PhD qualification. I owe it all to God. To Him be the glory forever.

My sincerely thanks goes to my supervisors, Professor Hussein Shimelis Hussein and Professor Mark Laing for their guidance, insightful criticisms and encouragement throughout my study. I have gained a lot of knowledge from their constructive comments, suggestions and review of my work. I am forever indebted to you. I would also like to acknowledge Dr Albert Changaya, my in-country co-supervisor, and Drs Admire Shayanowako and Isack Mathew for their support, proof reading, advice and encouragement during the development and write-up of this thesis.

I am grateful to the Alliance for a Green Revolution in Africa for funding of this study through the African Centre for Crop Improvement (ACCI). The entire ACCI staff is acknowledged for their esteemed support. I would especially like to thank Rowelda Donnelly and Lyndre Joan Anderson who provided administration support beyond expectations. You will be in my heart forever.

I sincere thank the Ministry of Agriculture/Malawi, through the Director of Agricultural Research Services, for granting me paid study leave and institutional support to pursue my PhD studies and for providing research facilities for my research. My sincere thanks goes to the Station Managers and staff at the Chitedze and Makoka Research Stations, and staff at the International Crops Research Institute for the Semi-Arid Tropics (ICRISAT)/Malawi for providing research facilities to conduct my research work. My heartfelt gratitude goes to Mrs Florence Ngwira and the entire Other Grain Legumes (OGL) team for the unlimited support that they rendered to me during my study period. You made my four years of study so easy. May the good Lord richly bless you.

Much appreciation goes to ICRISAT pathology lab specifically Dr Sam Njoroge and Mr Joseph Maruwo for the support and guidance in the *Fusarium* wilt work, and for allowing me to conduct

studies in their laboratory and glasshouse. I must also mention the technicians, Liness Dama and Nowa Sendeza, who assisted me in the laboratory and glasshouse.

Many thanks to Dr Gangarao and Dr Didas Kimaro for providing the pigeonpea germplasm used in this study.

I would like to acknowledge my fellow postgraduate students with whom I have shared both good and bad times with, the overall PhD research experience, and of course endless discussions. Special thanks to Mulu Fetahi, Nelly Phiri, Chapwa Kasoma, Wilson Nkhata, William Suvi, Yared Samahegn, Happy Daudi, Ruth Magareta, John Lobulu and Chimwemwe Josiah: you made my life easy and enjoyable at UKZN.

My heartfelt gratitude goes to my husband, Ivan Yohane, for taking both the fatherly and motherly role of our children during my absence. I know it was not an easy job leaving you with an 8 months old child but you managed. I am forever indebted to you but I am sure that God will repay you. I love you so dearly. My sweet daughters, Mayamiko, Michelle and Mafuno, thanks for your perseverance during my study period. I know you missed the motherly love from me. There was a time you needed me most but I was not there for you. Much love to you my babies. Many thanks to my parents Mike and Florence, brother and sisters for their prayers and moral support.

DEDICATION

This thesis is dedicated to my parents, Mike and Florence Nyirenda, my husband, Ivan Yohane, and my daughters, Mayamiko, Michelle and Mafuno: your presence means a lot to my success.

TABLE OF CONTENTS

ABBREVIATIONS AND ACRONYMS

ADD	Agricultural Development Division
AMMI	Additive main effects and multiplicative interaction
ANOVA	Analysis of variance
CV	Coefficient of variation
DArTseq	Diversity Arrays Technology sequencing
DARS	Department of Agricultural Research Services
DF	Degrees of freedom
DTF	Days to 50% flowering
DTM	Days to 75% maturity
EPA	Extension planning area
FMC	Flower main colour
FSP	Flower streak pattern
GCA	General combining ability
G×E	Genotype x environment interaction
GGE	Genotype main effects and genotype × environment interaction
GH	Growth habit
GYD	Grain yield
H^2	Broad sense heritability
h^2	Narrow sense heritability
Ha^{-1}	Per hectare
HSWT	100 seed weight
IPCA	Interaction principal component analysis
Kg	Kilogram
LSD	Least significant difference
N	Nitrogen
NPB	Number of primary branches
NPP	Number of pods per plant
NRP	Number of racemes per plant
NSB	Number of secondary branches
NSP	Number of seeds per pod

PCA	Principal component analysis
PC	Pod colour
PH	Plant height
PRA	Participatory rural appraisal
SCA	Specific combining ability
SCP	Seed colour pattern
SEC	Seed eye colour
SED	Standard error of difference
SMC	Seed main colour
SNP	Single nucleotide polymorphism
SS	Seed shape
SSIPCA	Sum of squares interaction principal component analysis
TARI	Tanzania Agricultural Research Institute
UKZN	University of KwaZulu-Natal

INTRODUCTION TO THESIS

Background

Pigeonpea [*Cajanus cajan* (L) Millspaugh), 2n=2x=22] is one of the most important legume crops in the tropics and sub-tropics. In sub-Saharan Africa (SSA) and Asia, the crop is cultivated mostly by smallholder farmers for food and cash income. The seed is rich in protein (26.60%) (Dabhi *et al.* 2019), complimenting the cereal-based diets notably in SSA (Simtowe *et al.* 2010). Pigeonpea serves as a good source of feed for livestock. In addition, anti-nutritional factors such as protease inhibitors (trypsin and chymotrypsin), amylase inhibitors and polyphenols are found at lower levels in pigeonpea than other legumes such as soybean, garden pea and field bean (Singh *et al.* 1990). The crop also improves soil fertility and structure through biological nitrogen fixation and organic matter accumulation, enhancing recycling of plant nutrients (Saidia *et al.* 2019). Further, the crop has medicinal values because of its roots, leaves and flowers are used to treat a wide range of liver, skin, lung and kidney diseases. Despite its diverse uses in the food and feed industry, and local and regional markets, pigeonpea has not received as much research and development support as other commodity crops such as maize, wheat and rice. Consequently, the majority of farmers in SSA, including those in Malawi, largely cultivate landrace varieties of pigeonpea, which have low yield potential. The average grain yields in Asia and Africa are estimated at 866.2 and 736.2 kg ha^{-1}, respectively, compared to the potential yield of 2500 kg ha^{-1} (Saxena 2008). There is a need to develop new and improved cultivars to enhance the productivity of pigeonpea in Africa.

Constraints to pigeopea production

Pigeonpea accounts for almost 5% of the world's pulse production (Mula and Saxena 2010). India is the largest producer of pigeonpea, accounting for 25% of world's production, followed by Myanmar and Malawi (FAOSTAT 2017). In Malawi, pigeonpea accounts for more than 22% of total legume production and ranks as the 3rd most important legume crop after groundnut and common beans (Dzanja *et al.* 2016). Major pigeonpea growing agricultural divisions in the country are Machinga and Blantyre, located in southern Malawi, constituting about 93% of the total production area (Kananji *et al.* 2016). According to Kimaro *et al.* (2020), the low productivity of pigeonpea is attributable to various insect pests and diseases, a lack of early

maturing/short duration varieties, and drought, among other factors. In Malawi, *Fusarium* wilt (FW) caused by the fungus *Fusarium udum* (Butler) is among the most serious challenges affecting pigeonpea production (Changaya *et al.* 2012). The disease is destructive in most pigeonpea growing countries, including Kenya, Tanzania and India (Hillocks *et al.* 2000, Gwata *et al.* 2006, Reddy *et al.* 2012). *Fusarium* wilt caused crop losses estimated at USD 71 million in India in 2011. In eastern Africa FW caused yield loss with a monetary value of USD 5 million (Reddy *et al.* 2012). FW can cause yield losses of 50 to 100%, depending on cultivar susceptibility (Soko 1992).

Importance of farmer involvement in cultivar development

Understanding farmer and market preferred traits, and identification and prioritization of their production constraints, are crucial for successful variety design, development and deployment. This is directly related to the adoption rate of new varieties along the value chain of each crop (Daudi *et al.* 2018). Therefore, there is a need to involve farmers and their clients in trait identification, priority setting, product profiling and participation in the technology evaluation process in the development of new crop varieties. The views and preferences of farmers during variety development and evaluation are necessary pre-conditions for plant breeders to design and prioritize their research goal in order to achieve high adoption levels of new varieties. Ceccarelli and Grando (2007) observed that farmers have the same selection abilities as breeders for quality traits. Close collaboration between farmers and breeders is necessary to speed up the breeding process and to respond to the demands of stakeholders.

Phenotypic and genetic diversity analyses in deciphering genetic variation for cultivar development

The development of new cultivars requires an understanding of the genetic diversity in the available germplasm, to inform breeding programs and germplasm management strategies. Limited information is available on the magnitude of genetic diversity within the cultivated pigeonpea gene pool (Saxena and Sawargaonkar 2014). Knowledge of genetic diversity facilitates identification of heterotic groups and best parents for breeding. Morphological, biochemical and molecular markers have been used in genetic diversity assessments in crop improvement. Morphological traits such as days to 50% flowering, days to 75% maturity,

number of primary secondary branches, hundred seed weight and grain yield are the most important traits in pigeonpea phenotypic diversity studies (Yohane *et al.* 2020). Molecular markers are more robust than morphological and biochemical markers because they are not affected by environmental conditions, which can confound genotype selection efforts (Zavinon *et al.* 2018). Several molecular markers have been used in genetic diversity analysis of pigeonpea, such as restriction fragment length polymorphisms (RFLP) (Sivaramakrishnan *et al.* 2002), amplified fragment length polymorphisms (AFLP) (Pati *et al.* 2014), random amplified polymorphic DNA (RAPD) (Malviya and Yadav 2010), simple sequence repeats (SSR) or microsatellites (Sarkar *et al.* 2017) and single nucleotide polymorphisms (SNP) (Saxena *et al.* 2014). SNP markers derived from next generation sequencing have been widely used because they provide high-density and whole-genome profiles at a relatively low cost (Jaccoud *et al.* 2001). Thousands of SNPs have been detected across the pigeonpea genome that are useful for characterizing germplasm and marker-trait association mapping.

Combining ability analyses in cultivar development

Combining ability analysis provides useful information on the gene action controlling trait inheritance and to identify superior genotypes as donor parents, and the best performing crosses, that can be used to develop breeding populations (Griffing 1956). Combining ability effects are broadly categorised into two categories: the general combining ability (GCA) effect and the specific combining ability (SCA) effect. A high GCA effect of a parent relates to additive gene action, while SCA effects in crosses is due to non-additive gene action (Griffing, 1956; Acquaah, 2009). Information on the GCA effects is crucial to select superior parental genotypes that will produce desirable offspring in subsequent crosses. Information on the SCA effects are useful to select the best cross combinations or families derived from favourable allelic combinations (Pandey *et al.* 2014). Different mating designs, including factorial or North Carolina designs, and diallel designs, among others, are used to analyse combining ability effects and deduce gene action controlling quantitative traits inheritance (Falconer *et al.* 1996).

The North Carolina mating designs were first developed by Comstock and Robinson (1948) to estimate combining ability effects, variance components, and heritability of quantitative traits. The North Carolina Design II or factorial mating design partitions the variance components into

additive and dominance variances to discern the magnitude of heritability of quantitative traits to guide selection.

Rationale of the study

Pigeonpea is one of the most important legume crops in SSA and Asia that is cultivated for food security, regional and global markets. In Malawi, pigeonpea accounts for more than 22% of total legume production. Despite the growing demand for pigeonpea in Malawi, farmers cultivate pigeonpea using old landrace varieties, which have low yield potential. The International Crops Research Institute for the Semi-Arid Tropics (ICRISAT) introduced some "improved" pigeonpea varieties that were bred in India and Kenya. The introduced varieties are relatively high yielding, with cream seed colours but have long cooking times, poor eating quality and are highly susceptible to key pests and diseases. As a result of these flaws, farmers have not adopted these varieties. Landraces are characterized by many excellent traits, including good taste, short cooking times and resistance to pod borer *(Helicoverpa armigera* Hubner). Other important traits are performance stability despite climate variability in high latitude and altitude areas, early maturity and *Fusarium* wilt resistance. Furthermore, any new cultivar must meet the trait requirements of farmers, grain traders and consumers. In order to develop superior new cultivars, selection of genetically diverse parents with excellent phenotypic traits is essential.

Overall Objective

The aim of this study was to contribute to food security in Malawi through breeding high performing and farmer-preferred pigeonpea varieties.

Specific Objectives

i. To determine production constraints and farmer-preferred traits affecting pigeonpea in Malawi to guide future breeding.
ii. To determine the diversity among pigeonpea germplasm collections using agro-morphological traits to select genetically distinct lines for breeding.
iii. To determine the genetic diversity among pigeonpea germplasm using single nucleotide polymorphism (SNP) markers to select genetically distinct lines for breeding.

iv. To determine the combining ability and gene action controlling early maturity, yield gains and resistance to *Fusarium* wilt, and to select the best parents and families for further breeding.

Hypotheses

i. Farmers are not aware of the constraints to pigeonpea production and they do not have specific cultivar preferences.

ii. Phenotypic traits do not vary significantly among the available pigeonpea germplasm.

iii. Agronomic traits and SNP markers do not significantly associated in explaining the genetic variation present in pigeonpea genotypes.

iv. The selected pigeonpea parents and the new families do not show good combining ability effects for earliness to maturity, grain yield, and for *Fusarium* wilt resistance.

Outline of thesis

This thesis consists of a total of five chapters as outlined below (Table 0.1). Chapter 1 is written as a separate review paper, while Chapters 2 to 5 are written as discrete research papers in the form of stand-alone research papers, followed by a general overview of the study. Chapters 2 to 5 were written following the University of KwaZulu-Natal format with abstract, introduction, materials and methods, discussion and conclusion sections. Due to their interdependence, the chapters contain some unavoidable overlaps and repetitions of references and introduction sections. Chapter 2 is under review in the South African Journal of Plant and Soil. The first part of Chapter 3 on phenotypic divergence analysis was published in Agronomy 10 (11), 1682. doi.org/10.3390/agronomy10111682. The second part of Chapter 3 on genotype – by – environment interaction and stability analyses was published in Acta Agriculturae Scandanavia, Section B – Plant Soil Science. doi;10.1080/09064710.2020.1859608. The fourth chapter on determining the genetic diversity among pigeonpea germplasm using single nucleotide polymorphism (SNP) markers to select genetically distinct lines for breeding is under review with PLOS one Journal. The fifth chapter on determining the combining ability and gene action controlling early maturity, yield gains and resistance to *Fusarium* wilt, and to select the best parents and families for further breeding is under review with Journal for Crop Improvement.

Table 0.1. Structure of thesis showing chapter number and title

Chapter	Title
----	Introduction to thesis
1	Literature review
2	Farmers' perceptions of the primary constraints to pigeonpea production in Malawi, and their variety choice and preferred traits: implications for variety design
3	Phenotypic divergence and grain yield stability analysis in pigeonpea [*Cajanus cajan* (L) Millspaugh] germplasm accessions
4	Genetic diversity and population structure analyses of pigeonpea genotypes using morphological traits and SNP markers
5	Combining ability, gene action and heritability for agronomic traits and *Fusarium* wilt (*Fusarium udum* Butler) resistance in pigeonpea
----	An overview of research findings

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CHAPTER 1. REVIEW OF THE LITERATURE

Abstract

Pigeonpea belonging to the primary gene pool of *Cajanus* species, and the only cultivated food crop in the family of *Cajaninae,* a sub-tribe of *Phaseolus*. The other gene pools (secondary, tertiary and quaternary) have desirable traits such as *Fusarium* wilt (*Fusarium udum* Butler) resistance, early maturity, resistance to *Helicoverpa armigera* and high pod set. Transferring such traits into the cultivated pigeonpea has been a challenge due to the incompatibility of *C. cajan* to other *Cajanus* species. Conventional breeding is widely used in transferring important traits from wild species to adapted cultivars. *Fusarium* wilt disease, pest damage and a lack of early maturing varieties that are photo-period insensitive are key constraints to pigeonpea production in Malawi. Efforts were previously made to develop early maturing cultivars that were determinate, photo-period insensitive and *Fusarium* wilt resistant varieties. However, these varieties were introductions from ICRISAT that did not include essential farmers' preferred traits such as flavor and short cooking time, hence they were not adopted by farmers. Understanding the inheritance of *Fusarium* wilt disease and genes governing the early maturity are key in a successful pigeonpea breeding program, in order to optimize the breeding strategy to be employed. To enhance pigeonpea production and productivity, it is important to explore additional variability for key traits such as short duration (early maturity), *Fusarium* wilt resistance and high yields in an effort to develop cultivars that will take into account the preferred traits of farmers, grain traders and consumers, including adaptation to climate variability in the high latitude and altitude areas. This chapter summarizes pigeonpea production, origin and diversity, advances in pigeonpea breeding, combining ability, genetics of earliness *Fusarium* wilt resistance, genotype × environment interactions, and the role of farmers in pigeonpea breeding.

Key words: early maturity, combining ability, cultivars, diversity, resistance, pigeonpea

1.1 Introduction

Pigeonpea is a diploid species (2n=2x=22), with a genome size of 833 Mbb (Varshney *et al.* 2012). It belongs to the genus *Cajanus* and tribe Phaseoleae clustered into four gene pools: primary (1 species, *C. cajan*), secondary (10 species), tertiary (24 species), and quaternary (9 species). It is one of the most important legume crops in the tropics and sub-tropics, and is considered to have originated in India (Van der Maesen 1990, Kassa *et al.* 2012, Saxena *et al.* 2014). It is believed to have been taken to Africa before 2000 BC (Van der Maesen 1990, Songok *et al.* 2010). Globally, pigeonpea production accounts for almost 5% of the world pulse production (Hillocks *et al.* 2000). India remains the largest grower and producer of pigeonpea, accounting for three-quarters of the world's pigeonpea production, followed by Myanmar and Malawi (Nedumaran *et al.* 2015). In Malawi, the crop accounts for more than 22% of the total legume production. However, it is ranked as the third most important legume crop after groundnut (*Arachis hypogaea* L.) and common bean (*Phaseolus vulgaris* L.). In Malawi, the Machinga and Blantyre Agricultural Development Divisions (ADDs), which are located in the southern region, remain the major pigeonpea growing ADDs, contributing to 93% of the total production area (Kananji *et al.* 2016). The southern region of Malawi used to receive additional rain during the cold, dry season, which was locally known as the 'Chiperoni' rains. Normally, these light rains would come after the main rainy season, when the crop was not fully mature, hence facilitating the maturation process of late maturity varieties. However, in recent years, the region has not received enough Chiperoni rains, which has reduced the yields of long maturity pigeonpea varieties. Attempts to extend pigeonpea production to other ADDs has proved futile because the medium to late maturing varieties grown in Malawi are affected by end-of-season droughts, coupled with declining temperatures and shortening day-lengths, adversely affecting yield potential and productivity. In the areas where pigeonpea is produced, diseases are a major biological constraint to pigeonpea production. The crop is attacked by about 60 pathogens including fungi, bacteria, viruses, mycoplasma and nematodes (Reddy *et al.* 2012). *Fusarium* wilt caused by the fungus *Fusarium udum* (Butler) is an important disease of the crop, which is both seed and soil-borne. It is therefore, important to explore additional variability for key traits such as short duration (early maturity) and *Fusarium* wilt resistance to develop cultivars that will take into account farmers, market and consumers' preferred traits including adaptation to climate variability in the high latitude and altitude areas to enhance production and productivity. This

could be achieved through phenotypic and genotypic characterization of germplasm to select the best parents with farmers' preferred traits combined with genotypes carrying early maturity and *Fusarium* wilt traits from wild relatives through modern biotechnology techniques (Saxena 2008, Mallikarjuna *et al.* 2011).

Global pigeonpea breeding has generated determinate genotypes with short maturity periods that are relatively photo- and thermo-insensitive. Some extra early maturity lines have also been reported by Vales *et al.* (2012). Good progress has been made in breeding for disease resistance in pigeonpea. Different sources of resistance to diseases such as *Fusarium* wilt and sterility mosaic disease have been identified and lines/cultivars resistant to these diseases have been developed globally(Sharma *et al.* 2012). Breeders have sought to develop early maturing varieties that are determinate and photo-period insensitive and *Fusarium* wilt resistant varieties. Two cultivars namely, Sauma (ICPL 9145) and Kachangu (ICEAP 00040) with *Fusarium* resistance and two short duration (early maturing) cultivars, and good adaptability to local climatic and edaphic conditions were released in Malawi out of introductions from ICRISAT. However, adoption of these cultivars by farmers was low due to their high susceptibility to diseases and insect pests, poor taste quality and long cooking time. Hence, there is a need to develop cultivars that have early maturity, *Fusarium* wilt resistance and farmer preferred quality traits by involving farmers in the breeding process. The chapter presents a review of the literature on the importance of pigeonpea, production and production constraints, origin, diversity and diversity analysis, advances in pigeonpea breeding, combining ability, genetics of earliness and *Fusarium* wilt and genotype - by - environment interactions, and the potential role of farmers in pigeonpea breeding.

1.2 Importance of Pigeonpea

Pigeonpea [*Cajanus cajan* (L) Millspaugh), 2n=2x=22] is one of the most important legume crops in the tropics and sub-tropics. In sub-Saharan Africa (SSA) and Asia, the crop is cultivated mostly by smallholder farmers for food and cash income. The grain is rich in protein (26.60%) (Dabhi *et al.* 2019), complimenting cereal-based diets, notably in SSA (Simtowe *et al.* 2010). Pigeonpea also serves as a good source of feed for livestock. In addition, anti-nutritional factors such as protease inhibitors (trypsin and chymotrypsin), amylase inhibitors and polyphenols are

found in relatively limited quantities in pigeonpea compared with other legumes such as soybean, peas and field beans (Singh *et al.* 1990). The crop improves soil fertility and structure through biological nitrogen fixation and organic matter accumulation, enhancing recycling of plant nutrients (Saidia *et al.* 2019). Further, the crop has medicinal values because its roots, leaves and flowers are used to treat a wide range of liver, skin, lung, and kidney diseases (Rahmatullah *et al.* 2009). Pigeonpea has a better tolerance to drought stresses compared to other legumes, and hence widely grown in low rainfall areas (Saida *et al*. 2019).

1.3 Global Pigeonpea Production

Globally, pigeonpea accounts for almost 5% of the world's pulse production (Hillocks *et al.* 2000). Pigeonpea production is mainly concentrated in South and Southeast Asia, followed by Sub-Saharan Africa. Global pigeonpea cultivation increased at an annual rate of 1.3% from 2.7 million ha in 1961 to about 4.6 million ha in 2007 (Simtowe *et al.* 2010). Between 2008 and 2010, South and South East Asia reported the highest production area of 1.4 million ha, yielding an output of 3.3 million tons. India is the largest grower and producer of pigeonpea (Nedumaran *et al.* 2015). In 2008, it accounted for 75% of the world's pigeonpea production, followed by Myanmar (15%), Malawi (2.6%), Kenya (2.5%), Uganda (2%), and Tanzania (1.5%) (FAOSTAT 2008). In 2014, India produced 3,290,000 tons followed by Myanmar with 575,100 tons and Malawi with 335,165 tons (FAOSTAT 2017). In 2019, India produced 3,315,440 tons, followed by Malawi (464,787 tons), Myanmar (347,395 tons) and Kenya (87,912 tons) (FAOSTAT, 2020).

1.3.1 Pigeonpea Production in Malawi

Malawi currently has a world pigeonpea production share of of over 6.9% (FAOSTAT 2020). The crop accounts for more than 22% of the total legume production. It has been ranked as the third most important legume crop after groundnut (*Arachis hypogaea* L.) and common bean (*Phaseolus vulgaris* L.) (Simtowe *et al.* 2010), or second after groundnuts in terms of production (Dzanja *et al.* 2016). The crop is grown in almost all the Agricultural Development Divisions (ADDs) in Malawi, of which Machinga and Blantyre, located in the southern region, remain the major pigeonpea growing ADDs contributing to 93% of the total production area (Kananji *et al.*

2016). About 65% of the pigeonpea produced in Malawi is consumed on-farm by farm households either as cooked dry or green peas, or as immature pods (Simtowe *et al.* 2010). The consumption rate is similar to that of Kenya (65%) but slightly higher than that of Tanzania (35%). An estimated 10% of Malawi's pigeonpea produce is sold locally while 25% is exported. This contributes 20% to household incomes (Orr and Orr 2002). Pigeonpea yield had remained constant for several years, and then it has drastically increased by 59% since 2011. The increase in production quantity from 2011 (Figure 1.1) was because of the release of medium duration varieties that are high yielding, and adaptable to diverse growing conditions. This allowed for the expansion of pigeonpea production, and increased yields.

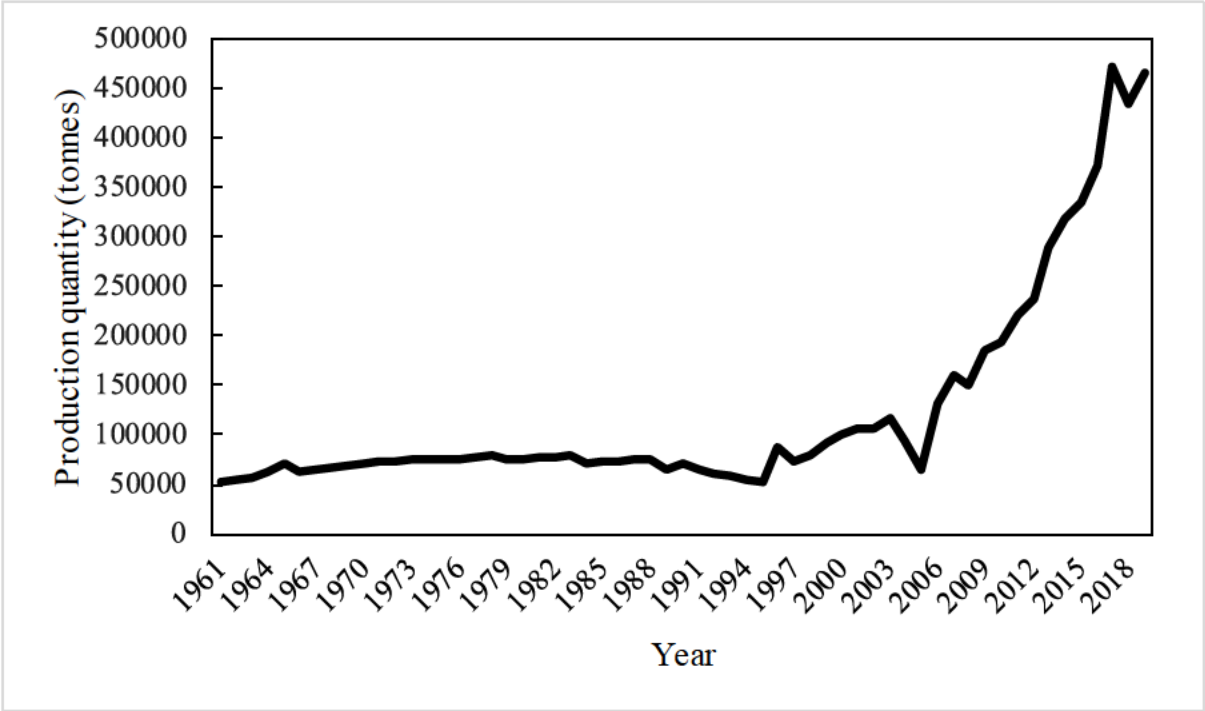

Figure 1.1. Pigeonpea total production trend from 1961 to 2018 in Malawi (FAOSTAT 2020)

1.3.2 Constraints to Pigeonpea Production in Malawi

The current farmers' average yield for pigeonpea is 700 kg ha^{-1} in Malawi, which is far below the potential yield of the crop, which can reach 2500 kg ha^{-1} (Kananji *et al.* 2016). This is attributed to several challenges faced by pigeonpea farmers.

Diseases are a major biological constraint to pigeonpea production. The crop is attacked by about 60 pathogens including fungi, bacteria, viruses, mycoplasma and nematodes (Reddy *et al.* 2012). *Fusarium* wilt caused by the fungus *Fusarium udum* (Butler) is an important disease of pigeonpea both seed and soil borne. The fungus survives in infected plant debris in the soil for about 10 years (Reddy *et al.* 2012). The disease is reported to be destructive in a number of countries including Malawi (Hillocks *et al.* 2000, Gwata *et al.* 2006, Reddy *et al.* 2012). *Fusarium* wilt reportedly led to annual economic losses in 2011 of USD 71 million in India. In eastern Africa FW caused yield losses with a monetary value of USD 5 million (Reddy *et al.* 2012). FW can cause grain yield losses of 50 to 100%, depending on cultivar susceptibility (Soko 1992). The other biotic constraint to pigeonpea production in Malawi is insect pests (Kananji *et al.* 2016). Pigeonpea is a host of up to 200 species of insects (Reed and Lateef 1990). Chewing or sucking insects such as jassids (*Empoasca kerri* Pruthi), aphids (*Aphis craccivora* Kosh), whitefly (*Bemisia tabaci* Genn), leaf web (*Grepholita critica* Meyer) and stem fly (*Ophiomyia centrosematis* de Meisere) attack the crop from seedling to vegetative stage. Flower and pod damaging insects, pod borers (*Helicoverpa armigera* Hub, *Maruca testulalis* Geyer), blister beetles (*Mylbris pustulata* Thunberg) and, pod flies (*Melanagromyza obtuse* Malloch). Pod borers (*H. armigera*, *M. testulalis*) are the most important pigeonpea pests in the tropics and sub-tropics because of their diverse host range, destructiveness, and wide distribution. It is estimated that *H. armigera* causes yield loss of up to 60% (Shanower *et al.* 1999). In addition, birds, white grubs and rats can cause damage to the seed in the soil, hence affecting germination.

1.4 Origin and Genetic Diversity of Pigeonpea

Pigeonpea is one of the most ancient crops originating from India (Saxena *et al.* 2014). It is believed to have been taken to Africa before 2000 BC (Van der Maesen 1990, Songok *et al.* 2010). Today pigeonpea is widely cultivated in all tropical and semi-tropical regions. The crop is cultivated in more than 25 tropical and subtropical countries either as a sole crop or is intercropped with cereals or legumes. Pigeonpea has a somatic chromosome number of 2n=2x=22), with a genome size of 833 Mbp (Varshney *et al.* 2012). It belongs to the genus of *Cajanus* and tribe Phaseoleae, clustered into four gene pools: primary (1 species, *Cajanus cajan*), secondary (10 species), tertiary (24 species), and quaternary (9 species) based on their genetic crossability, cyto-morphological behaviour and exchange of genetic materials (Mallikarjuna *et*

al. 2011). Significant studies have been made in making crosses between *C. cajan* belonging to the primary gene pool with species from other gene pools. The secondary gene pool is known to be cross-compatible with wild relatives. Some species (*C. aculifolius* and *C. lanceolatus*) within this gene pool have been discovered to have good pest and disease resistance, the A5 cytoplasmic male sterility system, high seed weight, and beige seed colour. The tertiary gene pool is incompatible with *C. cajan* species. However, *C. platycarpus,* which has *Fusarium* wilt resistance traits, extra early flowering/maturing traits, photo-period insensitivity traits, and A7 cytoplasmic male sterility, as well as *C. volubilis*, with dwarf plant type, early maturity, and high pod number, have been utilized in pigeonpea breeding (Saxena 2008, Mallikarjuna *et al.* 2011). The quaternary gene pool is incompatible with *C. cajan*. However, among the species under this gene pool, the crossing of pigeonpea with *Rhynchosia* species has been reported successful and superior hybrids have been developed (Mallikarjuna *et al.* 2011). Natural selection, domestication and breeding for desirable traits have resulted in the loss of genetic diversity in most annual crop species such as pigeonpea (Saxena *et al.* 2014). However, to broaden the genetic diversity of the cultivated pigeonpea, there is need to cross cultivated-pigeonpea with wild relatives and landraces that possess desirable traits that can contribute to crop improvement (Saxena *et al.* 2014). To conserve pigeonpea genetic resources, collections have been done worldwide and the materials have been kept in various gene banks. According to Upadhyaya *et al.* (2016), large pools of accessions are available at different gene banks, including ICRISAT India (13,771), the National Bureau of Plant Genetic Resources (NBPGR) India (11,221), United States Department of Agriculture (USDA), USA (4,116), Kenya Agricultural Research Institute, National Genebank of Kenya (KARI-NGBK), Kenya (1,288), National Plant Genetic Resource Laboratory in Philippines, (433) and at ICRISAT's regional genebank in Nairobi, Kenya (8,869). As a safety measure, 80% of the accessions are deposited in Svalbard Global Seed Vault in Norway. These are the reservoirs of genetic resources for the present and future pigeonpea improvement programs (Pazhamala *et al.* 2015).

1.5 Genetic Diversity Analysis

Genetic analysis studies are of great importance in crop improvement programs as they help in analysing genetic variability and identification of parents for crossing to develop populations. Furthermore, they provide information useful during introgression of desirable genes available in

diverse populations into cultivated germplasm and facilitates the grouping of accessions, genotypes or breeding populations into heterotic groups (Mohammadi and Prasanna 2003). The level of genetic diversity in germplasm accessions, breeding lines, and populations can be analysed using different methods such as morphological markers, biochemical markers and molecular or DNA-based markers (Casassola *et al.* 2013). These methods rely on morphological, biochemical and molecular marker data. Since each of these data sets provide different types of information, the choice of method (s) to use depend on the study objective, level of resolution required, resources, and technological infrastructure available and time (Collard and Mackill 2008). Several pigeonpea diversity studies (Chanda Venkata *et al.* 2019, Qutadah *et al.* 2019, Reddy and Jayamani 2019, Zavinon *et al.* 2019, Zavinon *et al.* 2020) have been done, and proved to be very useful in pigeonpea breeding programs.

1.5.1 Diversity Analysis using Morphological Traits

Morphological or agronomic traits such as flower colour, growth habit, seed colour, number of pods per plant, number of primary and secondary branches, grain yield, plant height, seed shape, and pod colour among others are used to assess the genetic diversity among germplasm materials (Kumara *et al.* 2013). This method involves growing plants in the field using statistically sound replicated designs during a full growing season cycle and recording the plants' characters. With morphological genetic diversity analysis, genotypes are grouped into clusters according to their traits, indicating the degree of their genetic diversity (Pandey *et al.* 2016). This helps to select parents for crossings as crosses made between diverse genotypes belonging to clusters separated by high inter-cluster distances with desired means are likely to produce transgressive segregants that may developed into high yielding pigeonpea cultivars (Pandey *et al.* 2016). A diversity study by Kumara *et al.* (2013) reported high levels of genetic diversity and variability among pigeonpea genotypes in all the morphological traits studied. In a similar study, Yohane *et al.* (2020) reported significant genotype variation for qualitative traits including flower colour, flower streak pattern, pod colour, seed coat colour pattern, seed coat main colour, seed shape, seed eye colour, and quantitative traits such as grain yield, hundred seed weight, number of branches per plant, plant height, days to 50% flowering, days to maturity, number of pods per plant and number of seeds per pod among 81 pigeonpea genotypes. This shows that

characterization of genotypes using morphological traits is of great importance since those that are stable across environments owing to the oligogenic nature of quantitative traits, hence they serve as morphological markers in breeding programs, and can be used in varietal or genotypic identification, varietal purification, and seed production (Muniswamy 2014). Even though phenotypic characterization provides a range of information about the genetic variability among accessions, it is affected by the environment and measurement errors, hence the need for a combination of morphological and molecular markers so as to verify the findings.

1.5.2 Genetic Diversity Analysis using Molecular Markers

Plant breeding programs depend on a high level of genetic diversity for achieving progress from selection. Broadening the genetic base of core breeding material requires the identification of diverse germplasm. DNA markers are used to characterize genetic resources to provide breeders with more detailed information to assist in selecting parents (Collard and Mackill 2008). There are several types of DNA markers namely; restriction fragment length polymorphism (RFLP), amplified fragment length polymorphism (AFLP), random amplified polymorphic DNA (RAPD), simple sequence repeats (SSR/microsatellites), and single nucleotide polymorphism (SNP).

The development of new cultivars requires a clear understanding of the existing diversity to inform breeding programs and management strategies. Limited information is available on the magnitude of genetic diversity within the cultivated pigeonpea gene pool (Saxena and Sawargaonkar 2014). Genetic diversity studies help to identify heterotic groups for breeding. Morphological markers, biochemical markers, and molecular or DNA based markers have been used in genetic diversity assessments in crop improvement. However, molecular markers are preferred to morphological and biochemical markers since they are not affected by environmental effects. In the past, molecular markers such as RAPD (Walunjkar *et al.* 2015), SSR/microsatellites (Bohra *et al.* 2011), SNP (Zavinon *et al.* 2018), and diversity array technology (DArT) (Yang *et al.* 2006a), have been used in pigeonpea genetic diversity studies. In particular, SSR and SNP markers have been used for genetic studies and breeding application in pigeonpea (Varshney *et al.* 2007). SSR markers have the advantage of multi-allelic and co-

codominant inheritance (Dutta *et al.* 2011) while SNP markers allow for high throughput and are cost-effective. Recently, Zavinon *et al.* (2020) reported a high level of genetic variability among 77 pigeonpea landraces using SNP and SSR markers. Despite the wide use of SSR markers in pigeonpea diversity studies, very few markers are available for use. DArT makers have been used in many crop species, including pigeonpea, since no sequence information is needed to develop the markers. DArT was initially developed in rice (Jaccoud *et al.* 2001). In pigeonpea, (Yang *et al.* 2006b) developed a pilot DArT array comprising of 5,376 features. This array was used to analyse 96 genotypes representing 20 species of *Cajanus*, and the results indicated a narrow variation in the cultivated genotypes. Recently, the DArT array for pigeonpea has been upgraded with > 15,000 features (Varshney 2015). These DArT markers have been used in genotyping mapping populations, and developing integrated and high-density genetic maps of pigeonpea.

1.6 Pigeonpea Breeding

1.6.1 Advances in Pigeonpea Breeding

Breeding progress depend on the nature and magnitude of genetic variation, the crop's reproductive behaviour, usage, adaptation to the environments and cropping systems (Changaya 2007). Pigeonpea is a self- pollinating crop, which also exhibits some levels of outcrossing. The natural outcrossing level varies from 5 to 70%, depending on the prevailing weather conditions and insect pollinators availability (Choudhary *et al.* 2015). Genetic mechanisms such as protogyny and self-incompatibility operate in pigeonpea to promote natural outcrossing (Choudhary 2011, Choudhary *et al.* 2012). Pigeonpea breeding efforts have been directed towards development of genotypes with zero percent outcrossing. To date, cleisto type genotypes have been developed by ICRISAT. Such genotypes have twisted flowers with tightly wrapped wings, enlarged keels, and free stamens (Sameer Kumar *et al.* 2015). Research efforts have also been directed towards hybrid breeding to increase productivity and adaptation. So far, cytoplasm from various wild relatives have been transferred into cultivated pigeonpea. Stable cytoplasmic male sterility systems have been developed from wild relatives *C. cajanifolius* A4 and *C. scaraboides* A2 (Saxena *et al.* 2005). Currently, cytoplasmic male sterility systems are being

used by pigeonpea breeders for diversification of A- lines and to produce commercial hybrids (Saxena 2008).

Photo- and thermo-sensitivity have been major issues in pigeonpea production, restricting expansion to different cropping systems in various agro-ecologies (Sameer Kumar *et al.* 2015). However, pigeonpea breeders have developed determinate genotypes with short maturity periods that are relatively photo- and thermo-insensitive. Some super early lines have also been reported by Vales *et al.* (2012), which have led to the expansion of pigeonpea production to non-traditional areas. Pigeonpea breeding has also focused on abiotic stresses such as drought, water logging, salinity and high temperatures that reduce crop production by 30% in most tropical and sub-tropical regions (Choudhary *et al.* 2015). Concurrently, good progress has been made in breeding for disease resistance in pigeonpea. Different sources of resistance to diseases such as *Fusarium* wilt and sterility mosaic disease have been identified and lines/cultivars resistant to these diseases have been developed(Sharma *et al.* 2012).

In Malawi, there is no active pigeonpea breeding program. However, seven introductions from ICRISAT have been released as pigeonpea cultivars (Kananji *et al.* 2016). Two *Fusarium* wilt resistant cultivars, Sauma (ICPL 9145) and Kachangu (ICEAP 00040), were released in 1987 and 2000. These cultivars are long duration varieties with photo- and thermo-sensitivity. The two cultivars have already lost their resistance due to the development of a new virulent race of the causative pathogen (Changaya 2007). Two early maturing cultivars, ICPL 87105 and ICPL 93026, bred by ICRISAT Nairobi were also released in 2003 but their adoption by farmers was poor due to undesirable traits such as poor taste and cookability, as well as susceptibility to pests and diseases (Changaya 2007). Since 2009-2014, three medium-duration varieties, Mwayiwathualimi (ICEAP 00557), Chitedze Pigeonpea 1 (ICEAP 01415/14) and Chitedze pigeonpea 2 (ICEAP 01485/3) bred by ICRISAT Nairobi were released. The release of the medium-duration cultivars increased pigeonpea production since the production has extended to non-traditional pigeonpea growing areas (Kananji *et al.* 2016). Despite the release of all these cultivars, farmers are still growing landraces that are slow maturing and *Fusarium* wilt susceptible (Changaya 2007). This is a clear indication that landraces possess desirable traits that are not available in released cultivars. To date, no pigeonpea cultivar has been bred in Malawi.

Developing new cultivars by introgressing earliness and *Fusarium* wilt traits into the landraces would improve pigeonpea production in Malawi.

1.6.2 Breeding for Earliness in Pigeonpea

Early maturity is an important agronomic trait in plant breeding for better adaptation to climate change. It enables farmers to harvest 2-3 times a year, hence increasing production and productivity (Yohannes *et al.* 2016). With pigeonpea, early maturing varieties are relatively short and produce relatively little biomass. Pigeonpea has a strong photo-period requirement, and its flowering is induced by long periods of darkness. The photo-period sensitive reaction is positively linked to its time of flowering and biomass production. Early maturing pigeonpeas are relatively less sensitive to photo-period responses (Srivastava *et al.* 2012). The development of such cultivars allows pigeonpea production in new latitudes and altitudes. This provides alternative cropping options and promotes market-oriented agriculture (Vales *et al.* 2012). Therefore, it is important to breed for the earliness trait in Malawi due to differences in agro-ecologies that restricts expansion of pigeonpea production to the central and northern regions. The southern region of Malawi used to receive additional drizzle rain during cold, dry season locally known as 'Chiperoni' rains. Normally, the light rains would fall after the main rainy season when the crop was not fully mature, hence facilitating the maturity process. However, in recent years, the region has not received adequate Chiperoni rains, reducing yields of long maturity pigeonpea varieties. With the development of early maturing cultivars, pigeonpea production and productivity will increase resulting into increased food and income security. Earliness is measured as the time that cultivars take to flower from the date of seeding to physiological maturity. The flowering genes may influence maturity date through their effects on the onset of reproduction and duration of the reproductive phase (Vales *et al.* 2012). In pigeonpea, days to flowering and days to maturity determine earliness. Early flowering helps to prolong the reproductive period, which is a major yield determinant (Vales *et al.* 2012), thus early flowering enhances early maturity.

1.6.3 Combining Ability and Genetics of Earliness in Pigeonpea

Combining ability is the capacity of an individual to transmit superior traits to its offspring (Pandey et al., 2014). Combining ability studies provide useful information on the gene action

controlling inheritance of traits. They help to identify general combiners to be used as a donor parent to improve specific traits and classify parental lines in terms of their hybrid performance (Griffing, 1956). Combining ability effects are broadly categorised into two, the general combining ability (GCA) effect and the specific combining ability (SCA) effect. A GCA effect of parents relates to additive gene action, while an SCA effect of crosses is due to non-additive gene action (Griffing, 1956; Acquaah, 2009). Information on the GCA effects is crucial to select superior parental genotypes that would produce desirable offspring in subsequent crosses. Information on the SCA effects are useful in exploiting heterosis during hybrid breeding to select the best cross combinations or families derived from favourable allelic combinations (Pandey *et al.* 2014). Different mating designs, including factorial or North Carolina and diallel designs, among others, are used to analyse for combining ability and deduce gene action controlling quantitative traits inheritance (Falconer *et al.* 1996).

Negative GCA and SCA effects for days to maturity have been reported for some pigeonpea parents, indicating that the parents could be utilized for exploiting early maturity genes (Baskaran and Muthiah 2007, Patil *et al.* 2014). Previous studies show that, days to 50% flowering are controlled by additive gene action and days to maturity are highly heritable and influenced by both additive and non-additive genes, with partial dominance for earliness (Kandalkar, 2005; Srivastava et al., 2012). However, some authors reported that days to 50% flowering is controlled by additive gene action (Thiruvengadam and Muthiah 2012, Patil *et al.* 2014). To quantify the number and position of genes controlling earliness in pigeonpea, QTLs for plant height (qPH5.1), number of secondary branches per plant (qSB5.1), number of pods per plant (qPD5.1), days to flowering (qFL5.1) and days to maturity (qMT5.1) were identified (Kumawat *et al.* 2012). Identification of QTLs controlling early maturity in pigeonpea is advantageous to early maturing breeding since it is easy to make introgression of this trait.

1.7 Breeding for Fusarium Wilt Resistance

1.7.1 Fusarium Wilt Disease

Fusarium wilt is caused by *Fusarium udum* Butler, a fungal pathogen. It is the most important soil-borne disease of pigeonpea, and has been reported as a major constraint to pigeonpea production in many pigeonpea growing areas and, the pathogen survives in the plant debris in the

soil for three years (Reddy *et al.* 2012, Sharma *et al.* 2012). The. More wilt inocula are found in sandy soils (94%) than in heavy soils (18%), and the fungal population is found more (30%) in soils with a low water holding capacity and soil temperature of between 20-30°C (Reddy *et al.* 2012). Root rot nematode (*Meloidogyne spp*) infections increase wilt incidence, and cyst nematodes enhance the pathogenicity of *F. udum* in wilt susceptible genotypes (Reddy *et al.* 1990). Early sowing, good weed management, good crop growth encourage wilt development. In addition, long and medium maturing types suffer the disease more than short duration or early maturing types, and ratooning renders the plant susceptible to wilt attack (Reddy *et al.* 2012). *Fusarium* wilt incidence has been reported from 30 to 60% at the flowering and full maturity stages, respectively (Reddy *et al.* 2012). It can cause yield losses of up to 100% in susceptible cultivars (Dhar *et al.* 2005). In Malawi, yield losses of more than 50% has been reported in the Thyolo and Mulanje districts (Soko 1992), and wilt incidence have ranged from 0-90% (Karimi *et al.* 2012). The annual crop losses due to pigeonpea wilt in Eastern Africa in 2011 was estimated at US$5 million, while in India it was US$36 million (Reddy *et al.* 2012). However, the loss can be reduced through the use of wilt resistant cultivars. The disease was first reported in India in 1906 but has now distributed in the following countries: Malawi, Kenya, Tanzania, Bangladesh, Ghana, Grenada, Mauritius, Myanmar, Nepal, Nevis, Trinidad, Tobago, Uganda and Venezuela (Reddy *et al.* 2012). The pathogen has also been reported in Mozambique (Southern Zambezia province) (Gwata *et al.* 2006).

1.7.2 Symptoms of *Fusarium* Wilt Disease

Since *F. udum* is a soil-borne pathogen, fungus enters the host vascular system at the root tips through wounds leading to progressive chlorosis of leaves and branches, wilting, and the collapse of the root system (Pande *et al.* 2013). Despite infection occurring at the early stage, the disease become visible later at the developmental stages (Reddy *et al.* 2012). The most obvious symptoms of *Fusarium* wilt are the loss of turgidity in plants and slight interveinal chlorosis. The most obvious internal symptom in the bark is a purple band of discoloured vascular bundles, extending upwards from the base of the main stem. According to Reddy *et al.* (2012) and Pande *et al.* (2013), the discoloured vascular band is more easily seen in varieties with green stems than purple stems. Partial wilting of the plant, resulting from taproot infection, is a definite indication of wilt disease and distinguishes the disease from termite damage, drought, and *Phytophthora*

blight that all kill the whole plant. Browning of the stem tissue and browning or blackening of the xylem visible when the stem or primary branches barks are peeled off is another characteristic symptom of the *Fusarium* wilt disease. The intensity of browning or blackening decreases from the base to the tip of the plant leading the plant to die-back (Reddy et al, 2012).

1.7.3 Sources of Resistance to *Fusarium* Wilt Disease

The existence of races of *F. udum* is a major challenge in breeding for *Fusarium* wilt in pigeonpea (Singh *et al.* 2011). *F. udum* isolates from the same site or diverse geographic origins have shown to exhibit high variability in cultural characteristics and virulence (Mishra and Dhar 2005). Therefore, it is important to search for additional sources of resistance to *Fusarium* wilt resistance. Genetic diversity in the fungus can be due to sexual genetic changes, segregation, mutation, recombination of genes during meiotic division or heterokaryosis, whereby cells of the fungal hyphae contain two or more nuclei that are genetically different as a result of fertilization (Agrios 2005). Variation in cultural and morphological characteristics of *F. udum* could also be due to environmental conditions, age of the isolates, sub-culturing, method of storage, and culturing conditions (Kiprop *et al.* 2005). It has been reported that the wide variations in virulence or pathogenicity to different genotypes of pigeonpea among *F. udum* isolates can be affected by environmental conditions and the inoculation techniques used (Kiprop *et al.* 2005), hence need for more research to confirm these findings.

1.7.4 Methods of controlling *Fusarium* Wilt Disease

Cultural practices such as rotation with crops such as sorghum [*Sorghum bicolour* (L) Moench], tobacco (*Nicotiana tobacum* L.) in every three years has been found to eliminate the pathogen from the field (Reddy *et al.* 2012). According to Natarajan *et al.* (1985), pigeonpea rotation with tobacco is recommended due to the toxic effects of tobacco root exudates on the pathogen such that a one year break with sorghum or fallow reduces wilt in the following pigeonpea crop by 20%. However, in Malawi, especially in the southern part where 93% of pigeonpea is produced, 58% of farmers have a land holding size of less than one hectare. In this case, rotation as a control measure is not a viable option since farmers depend on the small piece of land to produce a number of crops. In addition, application of nitrogen in the form of farm yard manure or green

manuring with *Crotalaria juncea* L. has also been reported to reduce the incidence of *Fusarium* wilt disease (Rao *et al.* 2014). However, there is a need for more studies to understand the effect of cultural practices on the disease to develop integrated pest management.

Several control measures have been reported to control *Fusarium* wilt disease, such as seed treatments with benomyl, thiram, a combination of carbendazim and thiophanate-methyl, application of a biocontrol strain of *Trichoderma,* and trace elements, among others (Prasad *et al.* 2012, Reddy *et al.* 2012). However, none of the fungicides have been reported to give adequate protection against *Fusarium* wilt disease since the pathogen is soil-borne(Prasad *et al.* 2012). The use of chemicals is harmful to the environment and ecosystem since it causes water and soil pollution and the killing of beneficial microorganisms (Devi and Chetry 2012). This calls for other alternatives like host plant resistance.

Biological control measures have attracted much attention worldwide in controlling soil-borne diseases such as *Fusarium* wilt since it is a safer control option than chemicals. With biological control, soils are supplemented with fungal or bacterial antagonists (e.g., *Serratia, Azotobacter, Clostridium, Bacillus, Arthrobacter, Alcarigens, Agrobacterium, Bradyrhizobium*) (Maisuria *et al.* 2008). In a tomato (*Lycopersicon esculentum* L.) study, Khan and Khan (2002) reported that root –dip applications of strains of *Bacillus subtilis, Pseudomonas fluorescens, Aspergillus avamori, Aspergillus niger, and Penicillum digitatum* resulted in significant declines of *Fusarium oxysporum f. sp lycopersici* populations in the rhizosphere. Anjaiah *et al.* (2003) also reported a reduction of *Fusarium* wilt disease incidence in pigeonpea and chickpea when the seeds were treated with *Pseudomonas aeruginosa*. However, due to lack of broad-spectrum activity, inconsistent performance, costs and limited availability, smallholder farmers have not adopted biocontrol options.

Use of resistant cultivars remains the most viable option for managing Fusarium wilt disease, especially in Malawi where most farmers are resource constrained. Several resistant pigeonpea lines and or cultivars have been identified and released with various levels of resistance in India, Kenya, Tanzania, and Malawi (Gwata *et al.* 2006, Singh *et al.* 2011, Sharma *et al.* 2012). These cultivars/genotypes are sources of Fusarium wilt resistance in pigeonpea breeding programs.

However, despite having resistant cultivars in some countries like Malawi, *Fusarium* wilt disease is still a challenge. This is because of different virulence levels that are environmentally specific, which may overcome resistance in certain environments (Patel *et al.* 2011). Therefore, it is important to have a clear understanding of the inheritance of the resistance, particularly in the belief that genotypes show different levels of resistance under field conditions. Since there is a strong chance of the resistance being matched by a new race of *F. udum*, it is important to focus on horizontal resistance in the breeding program. According to Agrios (2005), host plant resistance can either be physical, mechanical or a combination of the two. The physical resistance includes the plant structural characteristics such as thick cell wall, cuticles, waxy leaves, and other species that inhibit entry and spreading of the pathogen through the plant. For chemical resistance, pigeonpea wilt resistant cultivars produce cajanol, chlorogenic acid, caffeic acid, and phenolic acid (Marley and Hillocks 1993). These chemicals are known to inhibit germination and germ tube growth of conidia of *F. udum*. For durable resistance, it is important to breed for both physical and chemical resistance.

Hillocks *et al.* (2000), reported that the resurgence of pigeonpea wilt as a problem in Malawi has been due to a lack of a vibrant seed industry to make available seed of *Fusarium* wilt resistant cultivars to farmers. No breeding effort has been made in Malawi to introgress the genes of resistance into wilt susceptible local cultivars with consumer-preferred traits such as good cookability and taste. Therefore, it is important to test the resistant cultivars (Sauma and Kachangu) for their resistance levels so that they can be used as donor parents in development of early maturing and *Fusarium* wilt resistance cultivars.

1.7.5 Screening Techniques for *Fusarium* Wilt Resistance

Many screening techniques have been developed and modified over time for screening pigeonpea genotypes against *F. udum* in field, greenhouse, or controlled environments (Reddy *et al.* 2012). Field screening is done in a “sick plot”, whereby stubbles of wilted plants are collected and incorporated into the plot, and pigeonpea genotypes are planted in the inoculated plot. In this case, the susceptible genotypes will succumb to the disease at various growth stages, while resistant genotypes will remain healthy during the entire growing season (Changaya *et al.* 2012). However, the technique has some shortfalls because the distribution of the pathogen in the soil is

not even, leading to disease escape in some susceptible genotypes (Reddy et al. 2012). To ensure uniform pathogen distribution, the cereal seed inoculation technique can be used to infest the sick plot. Changaya *et al.* (2012), reported that autoclaved seed of finger millet, sorghum and wheat are equally effective for rapid multiplication of *F. udum* isolates.

Greenhouse screening is another technique used in screening for resistance to *Fusarium* wilt. This technique is done to confirm the resistance of the genotypes identified during large scale resistance screening in sick plots (Reddy *et al.* 2012). The most common greenhouse screening technique is root-dip screening. This is a method whereby seedlings are raised in sterilized river sand. The root tips of seedlings are later cut off and dipped in an inoculum suspension for 1-2 minutes (Patel *et al.* 2011). The inoculum concentration of 1 x 10^6 colony forming units (CFU) ml^{-1} in sterile water is ideal for inoculations (Patel *et al.* 2011). The cutting off seedling tips is done to facilitate the entry of the pathogen into the host. The inoculated seedlings are then transplanted in sterilized vertisol and sand in the ratio of 3:1 (Reddy *et al.* 2012). Odeny *et al.* (2009) reported that the root dip technique proved to be a relatively quick and reliable procedure for screening pigeonpea genotypes for *Fusarium* wilt resistance.

The use of a sick pot technique is commonly used in the greenhouse. This method is similar to the natural method. In this method, a known concentration of propagules for inoculation is applied to the sterilized sand or soil with no *Fusarium* wilt history in the pots in a greenhouse. In this method, fungus infested –pigeonpea flour medium is mixed with autoclaved field soil in the pots where genotypes are planted (Reddy *et al.* 2012). However, Changaya *et al.* (2012), reported a modified sort of sick pot method whereby the seedling root tips were bruised, and *F. udum* infested cereal seeds were placed on the bruised roots to obtain optimum inoculation concentration of the *F. udum* and later the seedlings were planted in the pots with autoclaved soils. This method was more effective in screening pigeonpea germplasm and filial generations in a breeding program without any disease escape (Changaya *et al.* 2012). In addition, the method is less expensive, and more suitable for developing countries like Malawi.

Other inoculation techniques include the water culture method, spore suspension, stem inoculation, and seed inoculation (Mishra and Dhar 2005).

1.7.6 Marker-assisted Selection (MAS) for *Fusarium* Wilt Resistance

Screening large number of genotypes for wilt resistance is time-consuming and labor demanding, hence the need for biotechnological techniques. The use of molecular markers enables breeders/geneticists to locate specific genes and QTLs governing trait of interest (Collard and Mackill 2008). This makes it possible to improve the efficiency of breeding through marker-assisted selection. The detection of genes or QTLs controlling a trait is possible due to genetic linkage analysis, which is based on the genetic recombination during meiosis (Malviya and Yadav 2010). Assessment of resistance or susceptibility of *Fusarium* wilt disease is done at the early stage of crop development, eliminating the need of maintaining the early stage of crop development, eliminating the need to maintain virulent isolates of wilt pathogen development of sick plots or artificial screening (Magadum *et al.* 2013). Several studies have been done to find markers linked to disease resistance (Saxena *et al.* 2010, Bohra *et al.* 2012, Kumawat *et al.* 2012, Saxena *et al.* 2012, Saxena *et al.* 2017), and markers linked to *Fusarium* wilt resistance have been identified for use in pigeonpea improvement programs. However, these are likely to be associated with monogenic resistance which may not be stable.

1.7.7 Genetics of Inheritance to *Fusarium* Wilt Resistance

Knowledge of genetic inheritance is essential for developing strategies on how to transfer the genes into well-adopted susceptible varieties. A number of studies reported that dominant genes control *Fusarium* wilt. This has been observed in various combinations involving resistant and susceptible parents (Kotresh *et al.* 2006, Chaithanya *et al.* 2011, Changaya *et al.* 2012). The dominant nature of inheritance allows for the easy transfer of resistance to susceptible cultivars using any selection method. Odeny *et al.* (2009) reported contrasting results, finding that multiple recessive genes controlled *Fusarium* wilt resistance. The control of resistance by recessive genes suggests greater mechanistic complexity largely due to mutations (Deslandes *et al.* 2002). Other studies have indicated that *Fusarium* wilt resistance is controlled by two complementary genes (one dominant and one recessive) (Kimani *et al.* 1994, Okiror 2002, Karimi *et al.* 2012, Saxena *et al.* 2012). The information from previous studies regarding the genetics and inheritance of *Fusarium* wilt is conflicting. This can be attributed to differences in experimental methodologies, the test population of pigeonpea, the pathogen isolates used,

inoculation techniques used, and environmental conditions. Therefore there is need to have a better understanding of inheritance of *Fusarium* wilt in pigeonpea.

Studies on QTL mapping for *Fusarium* wilt resistance have not been done extensively. Recently, (Saxena *et al.* 2017) reported a total of fourteen QTLs, including six major loci explaining >10% of the phenotypic variance observed. Comparative analysis between two recombinant inbred lines populations and F2 population generated three important QTLs: qFW11.1, qFW11.2 and qFW11.3 with up to 56.45% phenotypic variance explained (PVE) on CcLG11 genome region. This region can be considered the first choice for breeders aiming to introduce *Fusarium* wilt in susceptible cultivars.

1.8 Genotype-by-Environment Interaction and Stability Analysis for Grain Yield

Cultivar performance is a function of genotype, environment, and genotype × environment interaction (Acquaah 2009). Environmental factors have a greater effect on the quantitative traits than qualitative traits; hence there is a need to evaluate genotypes in multiple locations and years. Genotype × environment interaction (G × E) analysis measures genotypes' relative performance across two or more environments (Annicchiarico 2002). Genotype × environment interactions occur in two different ways: 1) when the difference between genotypes varies without alteration in their performance ranking (non-crossover interaction); and 2) when the ranking between cultivars changes across environments (crossover interaction) (Russell *et al.* 2003). Genotype × environment interactions reduce the association between phenotypic and genotypic values (Annicchiarico 2002). Strong G × E interactions for quantitative traits such as seed yield limit gains from selecting superior genotypes for improved cultivar development (Lynch and Walsh 1998). G × E analyses are done to breed superior genotypes that have both high mean performance and stability across environments and seasons (general adaptability), to breed varieties that perform consistently well in particular environments (specific adaptation), identify ideal environment(s) for genotype evaluation and to delineate environments into different mega environments(Yan and Hunt 2001). Understanding the magnitude of genotype × environment interaction helps in the identification of test conditions and recommendation of genotypes to areas of adaptation.

Yield stability of genotypes across various environments is important in plant breeding programs to make specific or wide area recommendations. An ideal genotype should have the genetic potential for superior performance under target growing conditions and should produce acceptable yields under less favourable conditions (Yan *et al.* 2007). Plant breeders often apply G × E stability statistics to assess the performance of their crosses or advanced genotypes across environments.

1.8.1 Additive Main Effects and Multiplicative Interaction (AMMI) Analysis

The AMMI analysis uses analysis variance (ANOVA) followed by principal component analysis (PCA) applied to the sum of squares allocated by the ANOVA to the genotype × environment interaction. The analyses partitions effects of genotype (G) and environment (E), additive main effects and their interaction as a multiplicative interaction component separately and submits to PCA for partitioning (Gauch *et al.* 2008, Amare *et al.* 2015). PCA is a generation of linear regression that can overcome the pattern of univariate analysis by giving more than one statistic to describe the pattern of genotype by environment interaction (Crossa 1990). Genotypes or environments with large interaction PCA values negative or positive have high interaction while those with smaller scores are considered stable (Gauch *et al.* 2008). Integrating biplot displays and genotypic stability statistics allows genotypes to be grouped based on their similarities in appearance across diverse environments (Amare *et al.* 2015). Several authors have reported on G × E in pigeonpea using AMMI analyses (Wamatu and Thomas 2002, Singh *et al.* 2017).

1.8.2 Genotype, and Genotype × Environment Interaction (GGE) Bioplot Analysis

A biplot is a scatter plot that approximates and graphically displays two-way “bi” data. A GGE biplot is a graphical tool that allows visualization of the interrelationship between environments and genotypes (Yan *et al.* 2000). GGE biplot analysis is useful for: 1) mega environment identification (which one where pattern that facilitates the identification of specific genotypes for their mega environment); 2) evaluation of genotype performance; and 3) environmental site evaluation based on their power to discriminate among genotypes in target environments (Yan and Tinker 2006, Yan and Holland 2010). The construction of GGE biplot is based on the first two principal components, PC1 and PC2, also referred to as primary and secondary effects,

respectively. These principal components are derived from subjecting environment centred yield data to dimension reduction. Biplots are also used to identify discriminating land representative test locations (Yan and Holland 2010). According to Yan *et al.* (2007), the angle between the vectors of two environments is measuring their correlation. An acute angle (< $90^{0)}$ implies positive correlation, an obtuse angle (> 90^{0}) implies negative correlation and a right angle (= 90^{0}) implies no correlation. Ideal environments are both representative (close to the average environmental axis) and discriminating. When ranking genotype based on mean performance and stability, lines perpendicular to the average environment axis (AEA) measure stability of genotypes in either direction. Genotypes with the smallest perpendicular lines and close to the AEA are stable. Therefore, ideal genotypes are located at the centre of concentric circles (high mean and stable). Good genotypes are located closer to the ideal genotype. With mega environment analysis (which one where), a polygon connects all the furthest genotypes, and perpendicular lines divide the polygon into sectors. The sectors helps to visualize mega environments, and the winning genotypes for each sector are located at the vertex. Several studies have been done to analyse G × E interaction in pigeonpea using GGE biplots, and cultivars have been recommended for general and specific adaptability (Chand *et al.* 2014, Sharma *et al.* 2015, Sharma *et al.* 2016, Pagi *et al.* 2017).

A number of G × E interaction studies have been done worldwide in pigeonpea improvement, however, very few studies have been conducted in Malawi. It is therefore important to evaluate the available pigeonpea genotypes in Malawi across environments over the years using the AMMI and GGE bioplot. It is expected that pigeonpea genotypes will rank differently in different environments; thus, evaluation of G × E interaction among genotypes of interest will identify early maturing, high yielding, *Fusarium* wilt resistant, and stable genotypes. This will make pigeonpea production more profitable to farmers, hence, improving food and income security.

1.9 Role of Farmers in Pigeonpea Breeding

The adoption of modern cultivars of food crops by small scale farmers is low (≤ 35%) in Sub-Saharan Africa (Walker and Alwang 2015) because the new plant varieties that are developed do not adequately meet the needs and preferences of farmers', processors, retailers, and consumers.

According to Walker and Alwang (2015), a demand-led plant breeding approach is the best way to ensure that the development of high-performing and quality crop varieties actually meet consumers requirements and market demands, hence, increasing the adoption rate. The approach follows the principles and processes of stakeholder involvement during cultivar development and commercialization.

Participatory Rural Appraisal (PRA) offers a rapid and cost-effective strategy for developing and selecting farmers preferred varieties for large scale production (Ceccarelli 2012). It enables local people to share, enhance, and analyse their knowledge of life and conditions. PRA is a popular and effective approaches that is used to gather information regarding farmers' knowledge in plant breeding (Dorward *et al.* 2007). Plant breeders use PRA to understand farmers' production constraints, perception, and preferences that can be accommodated in breeding programs to develop cultivars accepted by farmers (Ceccarelli 2012, Machida *et al.* 2014). A number of PRA tools are used to generate information, including focus group discussions and semi-structured interviews. Through focus group discussions and individual interviews, Ayenan *et al.* (2017) collected information on pigeonpea production constraints and preferred traits in Benin. They reported a lack of improved varieties and quality seed as major factors constraining pigeonpea production. High yields, early maturity and resistance to pod borers were ranked as the most preferred traits. With similar PRA tools applied in Tanzania, insect pests were reported to be the major constraint to pigeonpea production while high yields, early maturity, drought tolerance, short cooking period, indeterminate type, cream seed color, and large seed size were reported as the traits most important to farmers (Kimaro *et al.* 2017). Semi-structured interview and focus group discussion have proved to be important PRA tools to understand pigeonpea production constraints, perceptions and preferred traits, hence they will be used in the present study.

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CHAPTER 2. FARMERS' PERCEPTIONS OF THE PRIMARY CONSTRAINTS TO PIGEONPEA PRODUCTION IN MALAWI, AND THEIR VARIETY CHOICE AND PREFERRED TRAITS: IMPLICATIONS

Abstract

Pigeonpea is one of the most important grain legumes in Malawi, accounting for 22% of its legume production. However, the productivity of pigeonpea is low (< 1 ton ha^{-1}) due to various biotic and abiotic stresses and socio-economic constraints. There are no current information on farmers' perceptions of the primary production constraints, and their variety choice and preferred traits in Malawi for pigeonpea cultivar development and deployment. The objective of the present study was to determine farmers' perception of production constraints, and their variety and trait preferences in pigeonpea varieties in southern Malawi, as a first step of market research, leading to improved variety design and release. Participatory rural appraisal study was conducted in four main pigeonpea-growing districts in southern Malawi (Chiradzulu, Mulanje, Thyolo and Zomba). Data were collected using a semi-structured questionnaire, transect walks and focus group discussions (FGDs). A total of 304 individuals were interviewed using a semi-structured questionnaire, while 60 individuals participated in the FGDs. Maize (*Zea mays* L.) and pigeonpea were the major important crops, grown by 27.3 and 20.3% of the respondents, respectively. About 71 and 16.3% of the respondents intercropped pigeonpea with maize and sorghum, respectively. A landrace pigeonpea variety, 'Mthawajuni', was grown by 44.5% of the respondents for its positive attributes such as good taste, early to medium maturity, short cooking time and resistance to pod borer. Pigeonpea trait preference was dependent on gender, with female respondents preferring short cooking time (25% of the respondents), early maturity (15%), longer storage (3%) and pest resistance (10%), whereas men preferred high yielding (25%), large seeded (10%), cream seed colour (6%) and disease resistance (14%). All respondent farmers identified pod borer *(Helicoverpa armigera* Hubner) and *Fusarium* wilt disease (*Fusarium udum* Butler) incidence, low yields of their existing varieties, drought, and unreliable market prices as the leading challenges affecting pigeonpea production in southern Malawi. When designing new pigeonpea varieties, breeding for these farmer-preferred traits will enhance the adoption of newly released varieties, which should enhance pigeonpea production in Malawi.

Key words; Farmer preference, focus group discussion, landrace variety, Malawi, pigeonpea, participatory rural appraisal, variety design

Chapter 2 is under review in the South African Journal of Plant and Soil.

2.1 Introduction

Pigeonpea is one of the most important legume crops in Sub-Saharan Africa (SSA) and Asia, and it is cultivated for food security, and as a commercial grain for regional and global markets. The seed has a protein content of at least 21%, which is valuable in complimenting the predominantly cereal-based diets in SSA (Simtowe et al. 2010). Pigeonpea cultivation as a sole crop or as an intercrop improves soil fertility through biological nitrogen fixation and organic matter accumulation (Snapp et al. 2002). Globally, Malawi is the third largest producer of pigeonpea with 8.3% production after India (63.5%) and Myanmar (14%). Malawi ranked 5th in global exports of pigeonpea (FAOSTAT 2017). In the country, pigeonpea is grown in almost all the eight Agricultural Development Divisions (ADDs). However, 93% of the total production area is situated in the Machinga and the Blantyre Agricultural Development Divisions (ADDs) in the southern region (Kananji et al. 2016). However, the productivity of pigeonpea is low (< 1 ton ha^{-1}) in Malawi and Sub-Saharan African countries due to various biotic and abiotic stresses, and socioeconomic constraints.

Several constraints limit pigeonpea production and productivity globally, including a lack of access to breeder-released and high yielding varieties, diseases, pests, low soil fertility and erratic climatic conditions (Kaoneka et al. 2016). In Malawi, the International Crops Research Institute for the Semi-Arid Tropics (ICRISAT) has introduced some "improved" pigeonpea varieties that were bred in India. The introduced varieties are relatively high yielding and cream seed colour, but have long cooking times, poor eating quality and are highly susceptibility to key pests and diseases, hence farmers have not adopted these varieties. Instead, local farmers continue to grow landraces because of their many excellent traits, including good taste, short cooking time and resistance to pod borer (*(Helicoverpa armigera* Hubner). Therefore, there is need to develop high yielding pigeonpea varieties that meet the needs and requirements of local farmers' and their value chains. This is a common situation globally. Walker et al. (2015) reported low adoption rates (≤ 35%) of newly released cultivars of food crops in sub-Saharan Africa. The reasons for their rejection by farmers included the unsuitability of the breeder-released plant varieties to adequately meet farmers' needs and preferences, the unsuitability of the varieties to meet current and changing market demands, farmers' limited access to the seeds, and a lack of credit and production inputs, among others. In Zimbabwe in the 2000's, farmers' were still growing old maize varieties that were developed in the early 1960's -1970's, along

with even older landraces, although new and high yielding maize hybrids were available. The active rejection of the new maize varieties in Zimbabwe was attributed to their poor grain milling quality and grain palatability (Derera et al. 2006).

Understanding farmer and market preferred traits, and the identification and prioritization of their production constraints, are crucial for successful variety design, development and deployment. This is directly related to the adoption rate of new varieties along the value chain of each crop (Daudi et al. 2018). Therefore, there is a need to involve farmers and their clients in trait identification, priority setting, product profiling and participation in the technology evaluation process in the development of new crop varieties. The views and preferences of farmers during variety development and evaluation are necessary pre-conditions for plant breeders to design and prioritize their research goal in order to achieve high adoption levels of new varieties. Ceccarelli and Grando (2007) observed that farmers have the same selection abilities as breeders for quality traits, hence close collaboration between farmers and breeders is necessary to speed up the breeding process and respond to the demands of stakeholders.

Participatory Rural Appraisal (PRA) is a multi- disciplinary tool that has been used to capture farmers' perceptions regarding production constraints, variety choices and trait preferences (Ceccarelli 2012; Machida et al. 2014). PRA processes enable farmers to conduct their own analyses, planning and implementation, and directly inform plant breeders of their requirements. PRA studies have been successfully used in Malawi, Benin and Tanzania to guide pigeonpea breeding programs through the identification of challenges, variety choices and trait preferences (Ayenan et al. 2017; Changaya 2007; Kimaro et al. 2017). Despite the increasing economic significance of pigeonpea, currently there are no studies documenting farmers' perceptions of production constraints, varietal choice and preferred traits in pigeonpea in Malawi, as a basis for cultivar development and deployment. Therefore, the objective of the present study was to collect this information from farmers in southern Malawi using PRA tools, as a basis for a pigeonpea breeding program based on farmers' and market preferences.

2.2 Materials and Methods

2.2.1 Study Sites

The study was conducted in 2018 in the Chiradzulu, Zomba, Mulanje and Thyolo districts in southern region of Malawi (Figure 2.1). The districts were selected because they are major pigeonpea producing areas. Geographic coordinates, weather characteristics and altitude of the study districts are summarised in Table 2.1.

Table 2.1. Geographic coordinates, weather characteristics and altitude of the study districts

District	Coordinates	Altitude (m.a.s.l.)	Daily mean temperature (^{0}C)	Mean annual rainfall (mm yr^{-1})
Mulanje	16º 01' 53.87" S, 35º 30' 0.00" E	812	16.1 - 31.1	1626
Zomba	15º 09' 60.00" S, 35º 29' 59.99" E	741	22.0 - 24.0	1282
Chiradzulu	15º 40' 33.42" S, 35º 08' 26.26" E	889	20.0 - 29.0	890
Thyolo	-16º 06' 20.99" S, 35º 09' 2.16" E	885	11.0 - 30.0	1125

m.a.s.l. = meters above sea level, ^{0}C = degrees Celsius, yr^{-1} =per year

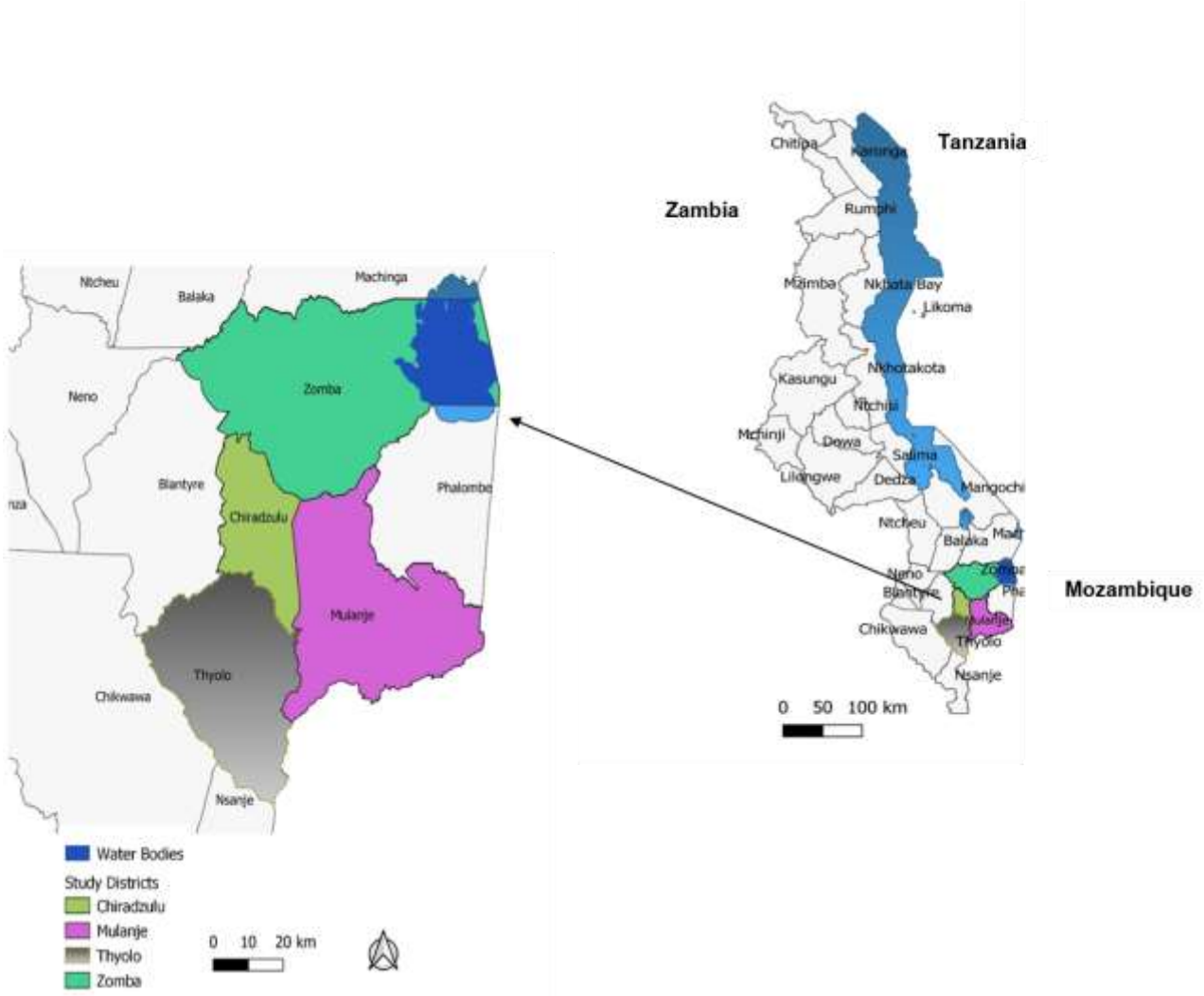

Figure 2.1. Maps of Malawi showing the four study districts

2.2.2 Sampling Procedure

A purposive sampling procedure was followed to select the following four major pigeonpea growing districts: Zomba, Chiradzulu, Thyolo and Mulanje, which are situated in the south of Malawi. In each district, two extension-planning areas (EPAs) were sub-sampled. EPAs are the smallest agricultural unit in Malawi. In each EPA, one village was sampled and 34 to 40 farmers were selected, based on their experience in pigeonpea production. This provided a total of 304 farmers for the semi-structured interviews (Table 2.2). A further four focus groups were established with 60 farmers for focus group discussions (FGD). One focus group discussion was done per district. Each FGD had a total of 15 farmers selected by local leaders in each village. Participants for FGDs were selected to represent the spectrum of farmers including from various pigeonpea cooperatives and taking into consideration gender balance (Table 2.2).

Table 2.2. Total number of farmers participated in the structured interviews and focus group discussion in the selected districts, EPAs and villages.

District	EPA	Village	Sample size (interviews)	Sample size (FGD)
Chiradzulu	Mombezi	Namasalima	40	15
	Thumbwe	Sumani	40	
Mulanje	Boma	Ngothima	40	15
	Msikawanjala	Mpenemuno	40	
Thyolo	Masambanjati	Namalanga	38	15
	Thyolo central	Lipulo	34	
Zomba	Dzaone	Moleni	38	15
	Mpokwa	Mathombo	34	
Total			304	60

EPA= Extension planning area, FGD= Focus group discussion

2.2.3 Data Collection

Data were collected using established participatory rural appraisal (PRA) tools such as a semi-structured questionnaire, focus group discussions (FGD), transect walks and direct observations. A semi-structured questionnaire (Appendix 1) was designed on topics related to the general socio-economic characteristics of the households, variety preference, cropping systems and production constraints. Pairwise and ranking matrices (Ceccarelli 2012) were used to identify the importance of pigeonpea as a food security and income generation crop, whereby a matrix scoring method was used to rank the farmers' preferences and the

perceived constraints affecting pigeonpea production. Participants mentioned their preferences and the perceived constraints, and these were listed on the flip charts, followed by ranking using a fixed number of votes. After voting, the percentage values of each parameter were calculated. A survey was conducted in April 2018 and individual interviews were held at each EPA where farmers converged. A total of 304 individuals were interviewed.

For the focus group discussion, each discussion was guided by a check-list that focused on the pigeonpea -based cropping systems, pigeonpea production constraints and variety preference. All the information from the focus group discussions was recorded and documented. Complimentary information was recorded through personal observation made on the transect walk through each of the sampled villages. During transect walks, observations were made on land size, crops grown, cropping systems, pigeonpea varieties grown, and the pigeonpea pests and diseases that were prevalent.

2.2.4 Data Analysis

Both qualitative and quantitative data collected through the questionnaire were coded and subjected to statistical analysis using Statistical Package for the Social Sciences (SPSS) version 25 (SPSS software, 2018). Cross tabulations were used to summarize the data and determine relationships between the variables. Pearson chi-square test procedure was used to draw significant tests and statistical inferences.

2.3 Results

2.3.1 Socio-Economic Description of the Study Areas

The socio-economic characteristics of the study areas including gender, age, education level and land holding size are summarised in Table 2.3. Out of 304 interviewees, 17.8% of the respondents were women, while 82.2% were men. There was significant difference ($P < 0.05$; $X^2 = 9.57$) in the number of women and men participated in the four districts. Participation of women was relatively higher (28%) in the Mulanje district and lower (10%) in the Thyolo district. Among the interviewees, 56.2% of the respondents were middle aged (31-50 years), whereas only 10% were young adults (21 and 30 years).

In the present study, 65% of the interviewees were illiterate and only 1.5% of the respondents attended tertiary education. There were significant difference ($P < 0.05$; $X^2 = 19.9$) in the education status among the four districts. The Thyolo district had more illiterate farmers (76%) than the Mulanje district (53%).

Most of the respondents (57.3%) had a land size of between 0.2-0.5 hectares (ha), 1.0% had a land size < 0.1 ha and 0.7% had > 2.0 ha. There was a significant difference ($P>0.5$; $X^2 = 27.9$) in the land holding size among the four districts. More farmers in the Mulanje district (36%) had land holding size of between 0.6 -1.0 ha, whereas in the Zomba district only 25% of the farmers had an equivalent land size.

Table 2.3. Socio-demographic information of households (%) in the study districts

		District							
		Chiradzulu	Mulanje	Thyolo	Zomba				
Variable	Category	(%)	(%)	(%)	(%)	Mean	df	X^2	*P* value
Gender	Female	15	28	10	18	17.8	3	9.57	0.023
	Male	85	72	90	82	82.2			
Age (years)	21-30	9	11	11	9	10.0	6	11.30	0.023
	31-50	56	57	53	59	56.2			
	≥51	35	32	36	32	33.8			
Education level	Illiterate	68	53	76	65	65.5	9	19.90	0.018
	Primary	25	44	20	30	29.5			
	Secondary	4	3	3	4	3.5			
	Tertiary	4	0	1	1	1.5			
Farm land size (ha)	<0.1	1	1	1	1	1.0	12	27.90	0.006
	0.2-0.5	58	52	59	60	57.3			
	0.6-1.0	32	36	27	25	30.0			
	1.1-2.0	8	11	12	13	11.0			
	>2.0	1	0	1	1	0.7			

X^2= Chi-square, df= degrees of freedom, P value= Probability value

2.3.2 Major Crops Grown, Cropping Systems and Pigeonpea Production Status

In all four districts maize (27.3%) was identified as an important crop, followed by pigeonpea (20.3%), cassava (10.9%) and sorghum (9.5%) (Figure 2.2). Tobacco was predominantly grown in the Zomba district (10.5% of respondents), and cowpea in the Mulanje district (12.1%).

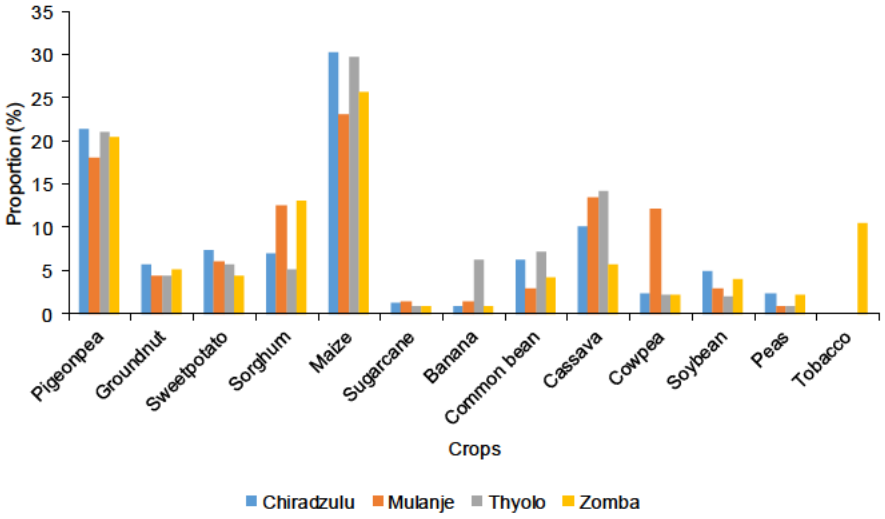

Figure 2.2. Proportion (%) of crops grown in Chiradzulu, Mulanje, Thyolo, and Zomba districts in Malawi

Table 2.4 summarizes the cropping systems, crops intercropped with pigeonpea and pigeonpea production status. Most of the interviewees (83.3%) practiced intercropping (maize-legume), some practiced sole cropping (13.3%), whereas few (3.5%) used a crop rotation system. Maize, sorghum, cassava, groundnuts and common beans were intercropped with pigeonpea, with 71% intercropped pigeonpea with maize, and 16.3% with sorghum. During transect walk, it was noted that some farmers grew a number of crops in one field (mixed cropping) to minimise crop losses during drought spells.

In the present study, 71.25% of the respondents indicated that pigeonpea production remained constant, whereas some 20.8% believed that production was decreasing. There was a significant difference ($P < 0.05$; $X^2 = 9.57$) in pigeonpea production status among the four

study districts. In the Mulanje district, 25% of the respondents believe that there was a decrease in pigeonpea production, while 4% believe that there was an increase in production. In the Chiradzulu district, 14% of the respondents believed there to be a decrease in pigeonpea production, while 11% believe there to be an increase in production.

Table 2.4. Pigeonpea cropping systems and farmers perceptions on production trends in four selected districts in Malawi

Variable	Category	District				Mean	Df	X^2	*P* value
		Chiradzulu (%)	Mulanje (%)	Thyolo (%)	Zomba (%)				
Cropping system	Sole cropping	20	12	12	9	13.3	6	13.15	0.041
	Intercropping	76	84	83	90	83.3			
	Crop rotation	4	4	5	1	3.5			
	Total	100	100	100	100	100			
Crops intercropped with pigeonpea	Maize	51	65	59	79	71.0	12	11.99	0.446
	Sorghum	25	20	7	13	16.3			
	Cassava	15	11	1	6	8.3			
	Groundnut	3	1	3	0	2.0			
	Common beans	6	0	0	3	2.4			
	Total	100	100	100	100	100			
Pigeonpea production status	Constant	74	66	73	72	71.2	6	12.91	0.044
	Increasing	11	9	4	7	8.0			
	Decreasing	14	25	23	21	20.8			
	Total	100	100	100	100	100			

X^2= Chi-square, df= degrees of freedom, P value= Probability value

2.3.3 Predominantly Cultivated Pigeonpea Varieties

The popularity of pigeonpea landraces and introduced varieties grown in the four study districts with their traits of importance are presented in Table 2.5. FGD participants perceived that the landrace variety 'Mthawajuni' was the most important and widely grown variety in the study areas. Mthawajuni has brown, black or reddish brown colour, early to medium maturity, resistance to pod borer, a rapid cooking time with a good taste. However, it is susceptible to *Fusarium* wilt [caused by *Fusarium udum* Butler]. Other important landrace varieties commonly grown in the four districts include 'Namanjo' and 'Rozikhuthula'. These landraces have good taste, good pod set, and resistant to pod borer, an important pigeonpea pest. The latter landraces are susceptible to *Fusarium* wilt and they have late maturity. Chi-square analysis revealed the presence of a significant difference ($P<0.01$; $X^2 = 267.36$) in landraces/varieties grown in the four districts. In the Chiradzulu district, most farmers (64.6%) grew 'Mthawajuni'. In the Zomba district, 40.8% of the respondents reported 'Rozikhuthula' important, whereas in the Mulanje district 37.7% of the respondents reported 'Namanjo' as the major pigeonpea variety grown.

Table 2.5. Pigeonpea landraces and improved varieties cultivated by farmers (%) with their distinguishing traits in the study districts.

	District						
Variety name	Chiradzulu (%)	Mulanje (%)	Thyolo (%)	Zomba (%)	Mean	Preferred traits	Drawback
Mthawajuni	64 6	38 8	28 2	46 5	44 5	Early/medium maturity, short cooking time, good eating quality, resistant to pod borers	Susceptible to *Fusarium* wilt, brown/purple seed colour
Rozikhuthula	8 9	0 0	0 0	40 8	12 4	High yielding, good eating quality, high pod set	Late maturity
Manyazi/Chinziri	0 0	18 3	0 0	0 0	4 6	Large seeded, heavy seed weight	Susceptible to bruchids, poor flavor, medium maturity
Chitedze pigeonpea1	1 3	0 0	0 0	0 0	0 3	Cream seed colour, short cooking time, good eating quality	Susceptible to pests and *Fusarium* wilt, poor eating quality
Mwaiwathualimi	13 9	11 3	2 8	7 0	8 7	High yielding, cream seed colour, tolerant to *Fusarium* wilt	Susceptible to pests, poor eating quality
Nadzombe	2 5	0 0	24 0	1 4	7 0	Cream seed colour, short cooking time, good eating quality	Susceptible to *Fusarium* wilt, late maturity
Sauma	0 0	0 0	2 8	0 0	0 7	Resistant to *Fusarium* wilt	Poor eating quality, late maturity, hard seed coat
Manjalende	0 0	5 0	1 4	0 0	1 6	Short cooking time, medium maturity, high pod set	Susceptible to *Fusarium* wilt
Namanjo	1 3	37 5	12 7	0 0	12 9	High yielding, good eating quality, resistant to pests	Late maturity
Njati	0 0	0 0	4 2	0 0	1 1	Good eating quality, short cooking time	Late maturity, susceptible to *Fusarium* wilt
Dawa	5 0	0 0	0 0	0 0	1 3	Very good eating quality, fast cooking, cream seed colour, tolerant to pests and diseases	Late maturity
Nangondo	2 5	7 5	4 2	4 2	4 6	Good for fresh pod consumption	Late maturity and Susceptible to *Fusarium* wilt
Kafula	0 0	0 0	1 4	0 0	0 4	Early maturity	Poor eating quality
Total	100 0	100 0	100 0	100 0	100 0		
df	36						
X^2	267 36						
P-value	0 000						

X^2= Chi-square, df= degrees of freedom, P value= Probability value

2.3.4 Farmer' Awareness of Available Introduced Pigeonpea Varieties and Seed Sources

The majority (66.2%) of the respondents knew of 'Mwaiwathualimi' as an introduced and popular variety, whereas few (4.8%) knew of 'Chitedze pigeonpea 2' as a new introduced variety (Table 2.6). In the Mulanje and Thyolo districts most respondents (91 and 52%, respectively) knew of 'Mwaiwathualimi'.

No significant difference ($P>0.05$; $X^2 = 25.23$) (Table 2.6) was reported on the source of information regarding new pigeonpea varieties among the four districts. However, the focus group discussion revealed that farmers mostly got the information from the EPA through extension officers. Information on new varieties was also passed on to farmers by researchers through research trials and demonstration plots.

Chi-square analysis showed significant difference ($P<0.01$; $X^2 = 49.71$) in seed sources among the four districts. Most of the farmers (36.5%) accessed pigeonpea seed by buying it from the local market. The other most common source of the pigeonpea seed as perceived by farmers were farm-saved seed (27.8% of the respondents) and sharing amongst the farmers (14.5% of the respondents).

Table 2.6. Farmer's awareness of introduced pigeonpea varieties and seed sources (%) in the study districts

Variable	Variety	District				Mean	Df	X^2	P value
		Chiradzulu (%)	Mulanje (%)	Thyolo (%)	Zomba (%)				
Awareness of new varieties	ICPL87105	8	0	5	5	4.5	15	35.56	0.002
	Chitedze Pigeonpea 1	16	7	14	5	10.5			
	Mwaiwathualimi	66	91	52	56	66.2			
	Chitedze Pigeonpea 2	4	0	10	5	4.8			
	Sauma	0	0	14	21	8.8			
	Kachangu	6	2	5	8	5.2			
	Total	100	100	100	100	100			
Source of information for new varieties	Extension planning area (EPA) office	36	40	31	35	35.5	18	25.23	0.112
	Cooperatives	10	14	19	18	15.3			
	NGOs	19	14	10	6	12.3			
	Buyers	2	5	0	6	3.3			
	Research (trials/demos)	15	14	22	22	18.3			
	Relatives/neighbours	15	7	12	12	11.5			
	Radio/newspaper/television	3	7	5	1	4.0			
	Total	100	100	100	100	100			
Source of pigeonpea seed	Local market	50	38	28	21	36.5	21	49.71	0.000
	Friend/neighbour	9	11	18	14	14.5			
	NGOs	11	4	1	4	5.4			
	EPA	1	7	6	4	5.0			
	Agricultural development and marketing corporation (ADMARC)	1	1	1	0	0.8			
	Agro-dealer	0	1	0	0	0.5			
	Cooperative	6	13	13	4	9.5			
	Farm saved seed	21	25	32	23	27.8			
	Total	100	100	100	100	100			

X^2= Chi-square, df= degrees of freedom

2.3.5 Farmer Perception of Constraints affecting Pigeonpea Production

The main constraints of pigeonpea production and their relative importance as reported by farmers during the focus group discussions are presented in Figure 2.3. In the Zomba district, pests and diseases, small land holdings and late maturity varieties were identified as the three top constraints affecting pigeonpea production. In the Chiradzulu district, pests and diseases, late maturity varieties and unreliable market conditions were regarded as the major constraints to pigeonpea production. In the Mulanje district, farmers prioritized pests and diseases, late maturity varieties and small land holding size as three major constraints. Farmers in the Thyolo district reported that pests and diseases, late maturity varieties and small land holdings were three main constraints to pigeonpea production. Overall, in the surveyed districts, pests and disease, late-maturity varieties, small land holdings and unreliable market conditions were the four main constraints to pigeonpea production, reported by the farmers.

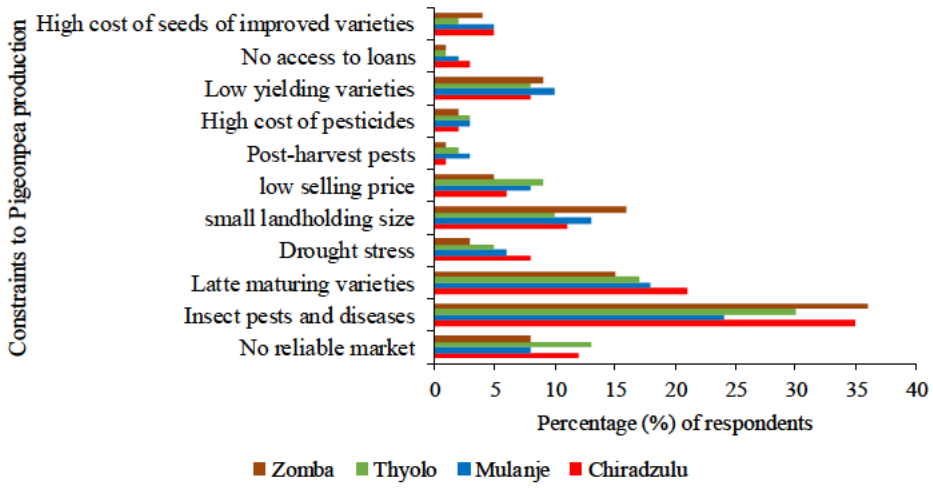

Figure 2.3. Farmer perceived constraints to pigeonpea production and their relative importance (%) in the four study districts

2.3.6 Pigeonpea Pest and Disease Management Options

The major pigeonpea diseases as perceived by farmers from the focus group discussions were *Cercospora* leaf spot (*Cercospora cajani* Hennings), *Fusarium* wilt and powdery mildew (*Leveillula taurica* (Lev.) Arnaud) (Table 2.7). Farmers reported that *Fusarium* wilt was the most important pigeonpea disease across the study districts. Chi-square revealed significant differences ($P < 0.05$); $X^2 = 24.02$) in management of *Fusarium* wilt across the four districts. 59.8% of the respondents reported to uproot and burn the infected plants, 24.0% of the respondents used no remedy and 14.3% used resistant varieties as a management options.

Aphids (*Aphis craccivora*), pod borers (*Helicoverpa armigera*), blister beetles (*Mylabris spp*) and pod sucking bugs (*Clavigralla spp*) were reported as the major pests for pigeonpea across the four districts (Table 2.7). Most respondents (72%) reported pod borer as the major pest limiting pigeonpea production across the four districts. However, most respondents (76.2%) did not use any remedy to manage pigeonpea pests. Farmers prioritized three main management options to control the pests: botanical pesticides, hand picking and crushing, and use of synthetic chemicals (14, 5.3 and 4.5%, respectively). Chi-square analysis revealed significant differences ($P <0.05$); $X^2 = 26.34$) in the management options adopted across the four districts. The Mulanje district had the most respondents (84%) that did not use any remedy to manage pigeonpea pests. None (0%) of the respondents used synthetic pesticides and only 10% used botanical pesticides. In the Zomba district, 14 and 17% used botanical and synthetic pesticides, respectively, to manage pigeonpea pests.

Table 2.7. Major pests and diseases of pigeonpea and their management strategies reported by farmers in the study area

Variable	Category	District				Mean	df	X^2	P value
		Chiradzulu (%)	Mulanje (%)	Thyolo (%)	Zomba (%)				
Pigeonpea diseases	*Cercospora* leaf spot	31	22	17	24	23.5	6	5 50	0.481
	Fusarium wilt	63	64	78	73	69.5			
	Powdery mildew	6	14	5	3	7.0			
	Total	100	100	100	100	100.0			
Management options for *Fusarium* wilt	Uprooting and burning diseased plants	60	70	63	46	59.8	12	24.02	0.020
	Use of resistant varieties	17	13	4	23	14.3			
	Crop rotation	3	2	0	1	1.5			
	Use of synthetic pesticides	1	1	0	0	0.5			
	No option	19	14	33	30	24.0			
	Total	100	100	100	100	100.0			
Common pests	Aphids	10	10	4	10	8.5	9	27.28	0.001
	Pod borers	73	69	74	72	72.0			
	Blister beetles	13	12	13	14	13.0			
	Pod sucking bugs	4	9	9	4	6.5			
	Total	100	100	100	100	100.0			
Pest management options	Hand picking	3	6	9	3	5.3	9	26.34	0.002
	Use of synthetic pesticides	1	0	3	14	4.5			
	Use of botanicals	15	10	14	17	14.0			
	No option	81	84	74	66	76.2			
	Total	100	100	100	100	100.0			

X^2= Chi-square, df= degrees of freedom, P value= Probability

2.3.7 Farmer-Preferred Traits of Pigeonpea

During the focus group discussions, respondents highlighted a number of traits that they would prefer to be incorporated in any new pigeonpea varieties. The important traits were high yield (19%), good taste (18%), a short cooking time and early maturity (14%), disease resistance (12%), pest resistance (9%), large seeded (6%) drought tolerant and cream colour (4%) (Table 2.4). It was observed that female trait preferences were different from male preferences. Women preferred short cooking time, early maturity, long storage and pest resistance whereas men focused on high yields, large seeds, cream colour and disease resistance. Good taste was equally important to men and women.

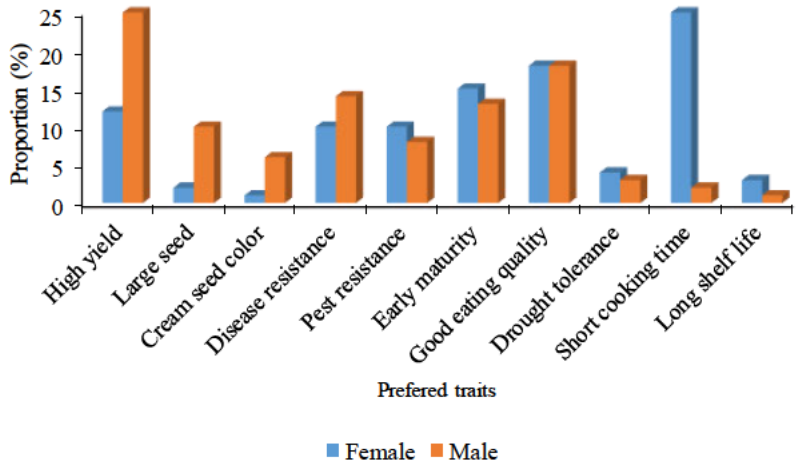

Figure 2.4. Preferred traits of pigeonpea aggregated by gender (%) in the study areas.

2.4 Discussion

2.4.1 Socio-Economic Description of the Study Areas

The present study revealed that more men participated in pigeonpea production than women (Table 2.3). This could be strongly attributed to traditional culture in Africa whereby husbands or men of have a greater decision-making power and authority on crop production and family matters. Also, men are custodians of common household wealth (Me-Nsope N and Larkins M 2016). However, in the south Malawi, matrilineal land inheritance is common whereby women have greater powers in family leadership and land holding. This suggests that women would have dominated pigeonpea production and marketing decisions, contrary to the data suggested in this study. Conversely, Simtowe et al. (2010) reported that pigeonpea production is dominated by women, making it a woman's crop in Malawi.

In the present study the majority of the sampled farmers consisted of middle-aged adult males (Table 2.3). Age is related with accumulated knowledge in crop production and management through years of cultivation (Dixit 2011). The low participation by youth in pigeonpea production suggests that youths are more involved in non-agricultural activities in local urban areas and or neighbouring countries. Youths migrate to urban areas in search of employment, engaging in businesses such as in barbershops, salons, and selling second-hand clothes, among others. In agreement with the present findings, Daudi et al. (2018) reported a low level of participation of youth in groundnut farming in Tanzania.

Despite free primary education program in Malawi, 65% of the interviewees were found to be illiterate (Table 2.3). This could be attributed to the age of the respondents. Free primary education program started in the multi-party era (1994) when most of the individuals who participated in the present study had passed the primary school age. However, studies have shown that a moderate level of education is a pre-requisite to the adoption of novel agricultural technology and efficient farm production (Abraha et al. 2017). Pigeonpea farming was based on land sizes of less than a hectare due to extensive estate farming and a high population growth in the southern Malawi (Chinsinga and Chasukwa 2012).

2.4.2. Cropping Systems and Pigeonpea Production Status

Pigeonpea is the second most important crop after maize, the leading food security crop in the Southern Malawi (Table 2.4). It is estimated that 65% of the pigeonpea produced in Malawi

is used for local consumption in the form of snacks, vegetables or dry seed (Simtowe et al. 2010). It is a source of cash to farmers who sell the surplus grain (Snapp et al. 2002). Pigeonpea is viewed as a low cost crop because it is cultivated without the use of fertilizers.

In this study, intercropping was perceived as a common cropping practice (Table 2.4) due to the shortage of land for crop production. Maize/pigeonpea intercropping was a common practice among households, citing several benefits such as nutrient build up, maintenance of soil fertility, efficient utilization of the available resources, and weed and pest control. With intercropping, there is an insurance of the harvest against crop damage or failure due to weather extremes such as drought. Furthermore, there is minimal competition between maize and pigeonpea due to slow growth of pigeonpea during the early developmental stages and rapid growth after the maize harvest (Kimani 2001). Contradictory findings have been reported on yield responses from intercropping enterprises. Rusinamhodzi et al. (2011) reported significantly higher yields of pigeonpea and maize in an intercrop than sole cropping, associated with a complementary interaction between the maize and pigeonpea crop. On the other hand, Saxena et al. (1998) reported maize yields were reduced by 5-23% and pigeonpea yields were reduced by 11-78%. However, significantly higher land equivalent ratio is anticipated in the maize/pigeonpea intercrop.

2.4.3. Predominantly Cultivated Pigeonpea Varieties

The present study found variability in farmer variety preference across sites (Table 2.5). This may be influenced by both historical and social factors. Landrace pigeonpea varieties are predominantly cultivated in the South Malawi, despite the availability of some introduced varieties. Landraces such as 'Mthawajuni', 'Namanjo' and 'Rozikhuthula' are characterised by early-medium maturity, good eating quality, short cooking time and pest tolerance, which are lacking in the introduced varieties. All the released varieties in Malawi are ICRISAT introductions from India, and most of these varieties did not meet farmers' needs and requirements. Most of the respondents (66.2%) were aware of the introduced varieties, the best-known being 'Mwaiwathualimi'. The popularity of this variety is because it was the first medium maturity (150-180 days) variety released in Malawi as an alternative to long maturity introduced varieties and landraces, which are yielding poorly due to climate change. The southern region of Malawi used to receive additional light rain during cold, dry season, locally known as the 'Chiperoni' rains. Normally, this rain would fall after the main rainy season when the crop is not fully mature, hence facilitating the maturation process. However,

in recent years, the region has not received enough Chiperoni rains, reducing yield of long maturity pigeonpea varieties. Mwaiwathualimi is high yielding and suitable for all agro-ecological zones of Malawi. As a result, it has expanded pigeonpea production to new areas, hence increasing national production (Gumma et al. 2019).

2.4.4. Farmers' Awareness of Available Introduced Pigeonpea Varieties and Seed Sources

The major source of information for the improved pigeonpea varieties and related agronomic practices is the extension planning area office (Table 2.6). Extension agents are provided with extension circulars of the newly released variety to be disseminated to the farmers. However, the major challenge is the limited human resource to provide adequate, effective agricultural extension and advisory services to the famers. The current ratio of government workers to farmers is 1:3000 against the recommended ratio of 1:1500 (GoM 2011). It is recommended that the Malawi government should train, recruit and deploy more extension agents in rural areas in order to effectively disseminate agricultural information to farmers for increased production and productivity. The present findings contradicts Ngwira and Majawa (2017) and Isaya et al. (2018) who reported that radio is the major source of agricultural information and extension agents are the second most important source of information. The present findings could be attributed to easy accessibility of the extension agents since the EPA offices are within their localities.

Farmers in the study areas access pigeonpea seed from the local market, save seed from previous crops, or source seed from friend/neighbours (36.5, 27.8 and 14.5%, respectively) (Table 2.6). The present findings shows that there is informal pigeonpea seed system in Malawi. Similar findings were reported in several countries in SSA (Abady et al. 2019; Ayenan et al. 2017; Kimaro et al. 2017; Manyasa et al. 2009; Mula 2012).

2.4.5. Pigeonpea Pest and Disease and Management Options

Pest and diseases are the major biotic constraint to pigeonpea production in SSA. *Fusarium* wilt was considered to be the most serious disease affecting pigeonpea yields (Table 2.7). In severe attacks, the disease can cause yield losses up to 100% (Changaya 2007; Gwata et al. 2006; Hillocks et al. 2000; Karimi et al. 2010; Kimaro et al. 2017; Reddy et al. 2012). In the present study, high incidences of the *Fusarium* wilt disease were reported by 69.5% of respondent farmers (Table 2.7). This was exacerbated by continuous growing of susceptible

varieties on the same land due to limited access to agricultural lands. This would increase the disease inoculum in the soil. Most respondent farmers (59.8%) tried to control *Fusarium* wilt by uprooting and burning the infected plants (Table 2.7), which is labor and energy intensive activity, and would not eliminate inoculum in the soil.

Among the major pests that attack pigeonpea in the study districts, pod borer was the most dangerous pest (Table 2.7). Shanower et al. (1999) also reported that pod borer was the most dangerous pest in India due to its diverse host range, destructiveness and wide distribution. The pest damages immature pods, reducing the quality and quantity of pigeonpea grain (Reed and Lateef 1990). Controlling the pest is difficult, especially because once larvae get inside the pod, the symptoms only occur when the damage has already occurred.

Use of long maturity varieties was perceived as a constraint to pigeonpea production (Table 2.7) because most of landraces took more than six months to mature. With climate change, rainfall has become unpredictable in Malawi, hence the need for varieties that mature quickly. This would allow farmers to harvest twice in one growing season, hence improving pigeonpea production in Malawi.

2.4.6. Farmer-preferred Traits of Pigeonpea

High yield, good eating quality, short cooking time, early maturity and disease resistance were the most desirable attributes across the study districts (Figure 2.4). Also, FGDs revealed that gender differences were observed in the choice of the traits of interest (Figure 2.4). Organoleptic aspects of pigeonpea varieties such as good eating quality and short cooking time were the most important traits of woman-preference followed by early maturity, pest resistance and longer storage time. This means that women's choices of traits were influenced by production and use of the grain, and food security at the household level. The need for early maturing variety is driven by the desire to get quick produce, as a mechanism to cope with climate change, and to limit pigeonpea competition with intercropped species (Ayenan et al. 2017). Men's preferences were based on production and marketing aspects. Male-preferred traits included: high yield, large seeds and resistance to disease (*Fusarium* wilt). Similar findings were reported by Weltzien et al. (2019), who reported that trait preference by men and women in a crop variety is influenced by different needs. Hence, it is important to note that gender is a key issue for variety development and that inclusion of complimentary womens' and mens' trait preference in a given pigeonpea variety facilitate responding to the

full range of households. The above findings justify the need to initiate and revitalise pigeonpea breeding in Malawi to develop client-preferred cultivars for increased adoption, production and productivity.

2.5 Conclusions

The present study confirmed that pigeonpea is the second most important crop in Malawi. The study identified pests and disease, late-maturity varieties, small land holdings and unreliable market conditions were the four main constraints to pigeonpea production. From the study, it can be concluded that landrace pigeonpea varieties such as 'Mthawajuni', 'Namanjo' and 'Rozikhuthula' are predominantly cultivated in the South Malawi, despite the availability of some introduced varieties because they are characterised by early-medium maturity, good eating quality, short cooking time and pest tolerance. The study also revealed that pigeonpea trait preferences are gender-based. Women trait preferences are influenced by production and use of grain, and food security at the household level. The men's trait preferences were influenced by production and marketing. Overall, the study identified; short cooking time, good eating quality, high yield, early maturity, long shelf-life, pest resistance, large seed, cream colour and disease resistance as major farmer preferred traits to be considered in new pigeonpea varieties. Focusing on these farmer-preferred traits while designing new pigeonpea varieties will ensure their adoption, and will increase pigeonpea production in Malawi.

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CHAPTER 3. PHENOTYPIC DIVERGENCE AND GRAIN YIELD STABILITY ANALYSIS IN PIGEONPEA GERMPLASM ACCESSIONS

Abstract

Knowledge of the genetic diversity and yield stability in pigeonpea is essential for effective breeding, genetic conservation and variety recommendation. The objectives of this study were to assess the genetic diversity and yield stability among pigeonpea accessions in selected target production environments to select complementary and unique genotypes for breeding. Eighty-one pigeonpea accessions were evaluated in six environments in Malawi using a 9 × 9 alpha-lattice design with two replications. Significant genotype variation were recorded for qualitative traits including flower colour, flower streak pattern, pod colour, seed coat colour pattern, seed coat main colour, seed shape and seed eye colour. All assessed quantitative traits were significantly affected by genotype × environment interaction effects except for the number of seeds per pod. Genotypes MWPLR 14, ICEAP 01170, ICEAP 871091 and ICEAP 01285 were identified as early maturing varieties, maturing in 125 to 137 days. Furthermore, test genotypes such as Kachangu, MWPLR 16, TZA 5582, No. 40 and MWPLR 14 had the most pods per plant (NPP) and highest grain yields (GYD). Grain yield was positively and significantly correlated with days to flowering (DTF) ($r=0.23$, $p<0.01$), NPP ($r=0.35$, $p<0.01$) and HSWT ($r=0.50$, $p<0.01$), suggesting the usefulness of these traits for selection to enhance grain yield improvement when assessing pigeonpea populations. From principal component analysis, three principal components (PCs) accounted for 57.7% of the total variation. The most important traits that reliably discriminated between the test genotypes were DTF, days to 75% maturity (DTM), number of primary (NPB) and secondary branches (NSB), HSWT and GYD. Genotype, environment and genotype × environment interaction (GEI) accounted for 16.4, 33.5 and 49.6% to the total variation for quantitative traits, respectively. The test environments were delineated into three mega-environments based on site and seasonal variability. MWPLR 14 (G51), MWPLR 24 (G26) and ICEAP 01155 (G27) were the most yield stable genotypes across environments, while MWPLR 14, TZA 5582 and MWPLR 4 were the highest yielding genotypes across environments. The selected high yielding and stable genotypes may be recommended as parental lines for breeding and grain yield improvement in Malawi or similar agro-ecologies.

Key words: Agronomic performance, correlation analysis, AMMI model, GGE bi-plot, Malawi, pigeonpea, yield stability

The first part of chapter 3 on phenotypic divergence analysis was published in Agronomy 10(11) 1982.doi.org/10.3390/agronomy10111682. The second part of chapter 3 on genotype by – environment interaction and stability analyses was accepted for publication in Acta Agriculturae Scandanavia, Section B –Plant Soil Science pending copy editing and typesetting doi;10.1080/09064710 2020.1859608.

3.1 Introduction

Pigeonpea is a multi-purpose crop, cultivated mainly for its edible grains that are high in dietary protein and essential amino acids such as leucine (16.48g/kg), tyrosine (14.77 g/kg) and arginine (13.51 g/kg) (Ade-Omowaye et al. 2015). Pigeonpea is an important component of the agriculture systems in semi-arid ecologies due to its adaptation to grow with relatively little rainfall and with poor soil fertility. It has a deep root system and a unique ability to maintain optimal osmotic adjustment under limited water condition (Subbarao et al. 2000). Pigeonpea is capable of fixing atmospheric nitrogen in the soils through symbiosis with species of *Rhizobium* bacteria depositing up to 200 kg of nitrogen per hectare in agricultural lands (Giller 2001; Kwena et al. 2019). Thus, pigeonpea has important roles of enhancing food security and livelihoods, especially during drought years, and providing ecosystem services through nitrogen fixation and soil health improvement.

Pigeonpea accounts for almost 5% of the world's pulse production (Mula and Saxena 2010). India is the largest producer of pigeonpea, accounting for over 75% of world's production, followed by Malawi (11%) and Myamnar (8%) (FAOSTAT 2020). In Malawi, pigeonpea accounts for more than 22% of total legume production and ranks as the 3rd most important legume crop after groundnut and common beans. The grain productivity of pigeonpea in Malawi is low (~700 kgha^{-1}) compared to the potential yield of 2500 kg ha^{-1} (Kananji et al. 2016). The yield gap is due to a various constraints, including insect pests and diseases, drought stress and a lack of improved cultivars. Breeding and deployment of improved cultivars has the potential to enhance pigeonpea production and productivity. Successful development of improved cultivars with client and market-preferred traits depends on the availability of adequate genetic variation. Reportedly, modern pigeonpea cultivars and varieties exhibit relatively low levels of genetic diversity (Bohla et al., 2011). The loss of the genetic diversity is due to continuous artificial selection and breeding for a few targeted economic traits to meet the market requirements (Saxena et al. 2014). Hence, there is a need to initiate pre-breeding programs in the target production environments through divergence breeding involving modern and obsolete cultivars, landraces and wild relatives that possess desirable traits. This will broaden the genetic diversity through gene recombination and effective selection (Saxena et al. 2014). The International Crops Research Institute for the Semi-Arid Tropics (ICRISAT) and various national and regional improvement programs are actively involved in genetic improvement and conservation of the pigeonpea. Diverse

pigeonpea collections are preserved globally, including by ICRISAT, the International Institute of Tropical Agriculture (IITA) and the Svalbard Global Seed Vault in Norway. These genetic resources can be used for pigeonpea improvement and breeding programs globally (Pazhamala et al. 2015; Upadhyaya et al. (2016).

To date only seven pigeonpea cultivars have been released in Malawi. These cultivars were introductions from ICRISAT (Kananji et al. 2016), which were developed in Kenya with germplasm from the Eastern and southern Africa (ESA). The ESA region is recognised as a secondary centre of genetic diversity for pigeonpea. The introduced cultivars were poorly adapted to local farming conditions in Malawi and lacked farmer-preferred traits such as good cooking quality, resistance to pod borers and high yield potential. The introduced cultivars have not yielded well probably because they were not adapted to the local growing conditions, or they had poor yield stability. Therefore, development of high performance, locally adapted pigeonpea cultivars is an important target in Malawi. This requires a range of genetic resources and crosses to integrate adaptive and functional traits, according to the needs and preferences of farmers and the value chain. Introduced germplasm can provide useful genetic resources that can be introgressed into locally adapted germplasm to improve economic traits such as high yield, early maturity, and pest and disease resistance, among others. Evaluating accessions maintained by the public and private breeding sectors within the ESA region provides an opportunity to identify stable and high yielding genotypes for selection.

Screening in several agro-ecological zones allows for the determination of genetic diversity present in the germplasm collections and to identify distinct genotypes for breeding or variety recommendation. Genotypes exhibit differential responses to a range of environmental conditions such as soil, temperature, moisture, and disease pressure, which provides opportunities for identifying superior and adapted genotypes. The differential response of a genotype to varying environmental conditions is caused by the genotype × environment interaction (GEI). The GEI provides opportunities and challenges during breeding. The GEI confounds the selection process during breeding, making it difficult to identify the best and most stable genotypes for cultivar recommendation (Cucolotto et al. 2007). Furthermore, the GEI reduces the correlation between genotype and phenotypic expression, leading to low genetic gains achieved from selection of traits with quantitative inheritance such as grain

yield (Bustos-Korts et al. 2018). Ultimately, GEI reduces the ability to predict genotype performance for quantitative traits using the genotype and environment main effects only. Consequently, GEI requires advanced statistical analysis tools to adequately separate genotype and environment and their interaction effects on phenotypic expression (Gauch et al. 2008). Conversely, GEI enables the identification of superior genotypes adapted to specific environments or genotype selection with static or dynamic stability adapted to a number of environments. Genotypes with dynamic stability have the ability to significantly improve their yield with improvements in environmental conditions, while those with static stability maintain a relatively similar performance across different environments (Sabaghnia et al. 2015; Yan 2016). Thus, it is important to evaluate the magnitude of the GEI effect and identify genotypes with known stability type to enable cultivar selection and recommendation for broad and specific environments.

The additive main effect and multiplicative interaction (AMMI) and genotype plus genotype × environment (GGE) bi-plot methods are used widely to evaluate GEI (Gauch 2006). The AMMI model is a statistical tool that combines analysis of variance with principal component analysis and interprets the main effects as additive and GEI effects as multiplicative (Gauch et al. 2008; Amare et al. 2015). After separating the additive main effects, the AMMI model subjects the multiplicative effects to principal component analysis to decompose the GEI into two principal axes and estimate trait means using the least square method (Thillainathan and Fernandez 2001). In addition, Purchase et al. (2000), developed the concept of the AMMI stability value (ASV) to identify genotypes that are relatively stable across a number of environments. The ASV is a parametric measure based on the interaction principal component analysis 1 (IPCA1) and interaction principal component analysis 2 (IPCA2) scores for each genotype derived from the AMMI model. The ASV is widely used in applied plant breeding. The GGE bi-plot analysis graphically represents the relationship between genotypes and test environments (Yan et al., 2000). The GGE biplot complements the AMMI method by identifying genotype similarities in different environments. With the GGE-biplots, grain yield potential and stability are evaluated using an average environment coordination (AEC), which is defined by the average principal component scores for all the environments (Dehghani et al. 2009). In addition, the GGE biplot enables to identify genotype discriminating environments to improve selection efficiency and in deployment of genotypes

adapted to specific environments. Also GGE is a useful tool in decision-making for managing resources for cost-effective breeding programs (Mitrović et al. 2011).

A considerable number of pigeonpea genotypes have been collected and maintained at the Department of Agricultural Services in Malawi for breeding purposes. The genotypes are adapted to the ESA region, and possess valuable attributes including good cooking quality, insect pests and disease resistance, but are limited by their poor yield performance. The key traits present in the local and introduced germplasm should be assessed for pre-breeding and breeding purposes. Hence, the objectives of the study were to assess the genetic diversity and yield stability among pigeonpea accessions in selected target production environments in Malawi to select complementary and unique genotypes for breeding.

3.2 Materials and Methods

3.2.1 Plant Materials

The germplasm used in this study comprised of 81 pigeonpea genotypes, including 28 landraces, 6 released cultivars, and 47 advanced elite lines sourced from three gene banks (Table 3.1). The landraces and cultivars were collected from the Department of Agricultural Research Services (DARS) in Malawi and the Tanzania Agriculture Research Institute (TARI). The elite lines were obtained from the International Crop Research Institute for the Semi-Arid Tropics (ICRISAT) in Kenya. The landraces were included as germplasm with adaptation to local conditions and quality traits such as good palatability and short-cooking time that meet farmers' demands. Whereas the elite lines would provide important genetic resources since Tanzania and Kenya have advanced pigeonpea-breeding programs. The released cultivars provided a benchmark against commercial standards that are currently in production.

Table 3.1. Description of the pigeonpea genotypes used in the study

Code	Genotype designation/.name	Source	Origin	Code	Genotype designation/.name	Source	Origin
G1	ICEAP 0673/1	ICRISAT	Kenya	G42	ICEAP 87105	ICRISAT	Kenya
G2	ICEAP 00554	ICRISAT	Kenya	G43	MWPLR 16	MPGRC	Malawi
G3	ICEAP 01164/1	ICRISAT	Kenya	G44	TZA 2496	TARI	Tanzania
G4	MWPLR 19	MPGRC	Malawi	G45	TZA 5582	TARI	Tanzania
G5	MWPLR 22	MPGRC	Malawi	G46	TZA 5596	TARI	Tanzania
G6	ICEAP 01170	ICRISAT	Kenya	G47	Chitedze Pigeonpea 2	DARS	Malawi
G7	ICEAP 01169	ICRISAT	Tanzania	G48	MWPLR 7	MPGRC	Malawi
G8	TZA 2439	TARI	Tanzania	G49	Babati	TARI	Tanzania
G9	MWPLR 9	MPGRC	Malawi	G50	TZA 5557	TARI	Tanzania
G10	MWPLR 6	MPGRC	Malawi	G51	MWPLR 14	ICRISAT	Kenya
G11	MWPLR 17	MPGRC	Malawi	G52	ICEAP 01101/1	ICRISAT	Kenya
G12	TZA 253	TARI	Tanzania	G53	TZA 2456	TARI	Tanzania
G13	MWPLR 1	MPGRC	Malawi	G54	TZA 5464	TARI	Tanzania
G14	MWPLR 18	MPGRC	Malawi	G55	ICEAP 01101/2	ICRISAT	Kenya
G15	TZA 2464	TARI	Tanzania	G56	ICEAP 01285	ICRISAT	Kenya
G16	ICEAP 00604	ICRISAT	Kenya	G57	MWPLR 25	MPGRC	Malawi
G17	TZA 2509	MPGRC	Malawi	G58	ICEAP 87091	ICRISAT	Kenya
G18	ICEAP 01146/1	ICRISAT	Kenya	G59	TZA 2692	TARI	Tanzania
G19	MWPLR 11	MPGRC	Malawi	G60	TZA 2807	TARI	Tanzania
G20	TZA 5555	TARI	Tanzania	G61	ICEAP 00068	ICRISAT	Kenya
G21	No. 40	TARI	Tanzania	G62	TZA 2785	TARI	Tanzania
G22	ICEAP 01150	ICRISAT	Kenya	G63	MWPLR 10	MPGRC	Malawi
G23	MZ2/9	TARI	Tanzania	G64	ICEAP 00612	ICRISAT	Kenya
G24	ICEAP 01172/1	ICRISAT	Kenya	G65	MWPLR 21	MPGRC	Malawi
G25	ICEAP 01103/1	ICRISAT	Kenya	G66	TZA 2514	TARI	Tanzania
G26	MWPLR 24	MPGRC	Malawi	G67	TZA 2466	TARI	Tanzania
G27	ICEAP 01155	ICRISAT	Kenya	G68	ICEAP 01179	ICRISAT	Kenya
G28	ICEAP 01180/2	ICRISAT	Malawi	G69	MWPLR 13	MPGRC	Malawi
G29	MWPLR 4	MPGRC	Malawi	G70	MWPLR 2	MPGRC	Malawi
G30	Kachangu	DARS	Malawi	G71	TZA 250	DARS	Malawi
G31	Mwayiwathualimi	DARS	Kenya	G72	MWPLR 3	MPGRC	Malawi
G32	MWPLR 8	ICRISAT	Malawi	G73	TZA 5541	TARI	Tanzania
G33	ICEAP 01154/2	ICRISAT	Kenya	G74	MWPLR 23	MPGRC	Malawi
G34	Chitedze Pigeonpea 1	DARS	Malawi	G75	ICEAP 00979/1	ICRISAT	Kenya
G35	ICEAP 01164	ICRISAT	Kenya	G76	TZA 197	TARI	Tanzania
G36	Bangili	TARI	Tanzania	G77	MWPLR 20	MPGRC	Malawi
G37	ICEAP 00053	ICRISAT	Kenya	G78	HOMBOLO	TARI	Tanzania
G38	MWPLR 12	MPGRC	Malawi	G79	ICEAP 86012	ICRISAT	Kenya
G39	TZA5463	TARI	Tanzania	G80	ICEAP 01106/1	ICRISAT	Kenya
G40	MWPLR 5	MPGRC	Malawi	G81	Sauma	DARS	Malawi
G41	MWPLR 15	MPGRC	Malawi				

ICRISAT=International Crops Research Institute for the Semi-Arid Tropics, MPGRC= Malawi Plant Genetic Resource Centre, DARS= Department of Agricultural Research Services, TARI= Tanzania Agricultural Research Institute

3.2.2 Study Sites

Field experiments were conducted at three selected sites in Malawi, namely at the Bvumbwe, Chitedze and Makoka Research Stations, during the 2017/18 and 2018/19 cropping seasons. The geographic location, altitude, weather and soil characteristics of the study locations are presented in Table 3.2. Each season and site combination presented unique environmental conditions due to variations in temperature, rainfall and agronomic practices. Therefore, due to site × season combinations, a total of six environments were identified for evaluating the genotypes. The conditions prevailing in Bvumbwe during 2017/18 season was considered as Environment 1 (E1), while Bvumbwe in 2018/19 season was E2, Chitedze in 2017/18 season was E3, Chitedze in 2018/19 season was E4, Makoka in 2017/18 season was E5 and Makoka in 2018/19 season was E6.

Table 3.2. Physical and weather characteristics of the study locations

Site	Latitude	Longitude	Altitude (masl)	Soil texture	Rainfall (mm)		Min Temp (^{O}C)		Max Temp (^{O}C)	
					2017/18	2018/19	2017/18	2018/19	2017/18	2018/19
Bvumbwe	15 ^{O} 55' S	35 ^{O} 04' E	1228	Sandy clay loam	975.2	1442	16.2	17.9	22.6	24.9
Chitedze	13 ^{O} 59' S	33 ^{O} 38' E	1146	Sandy clay	929.8	693.4	18.5	20.2	24.7	29.4
Makoka	15 ^{O} 32' S	35 ^{O} 11' E	1029	Sandy clay loam	566.6	1184.8	16.3	15.6	23.2	28.2

Masl= metres above sea level, mm= millimetres, min= minimum, max= maximum, temp= temperature, ºC= degrees Celsius

3.2.3 Experimental Design and Data Collection

The experiment at each site was laid out in a 9 × 9 alpha-lattice design with two replications. Each genotype was planted in a plot consisting of two rows. The rows were 5m in length and 0.90 m apart, giving a plot size of 4.5 m^2. Seeds were planted at 0.75 m apart within a row. Three seeds were planted per planting hole and thinned to one plant two weeks after emergence. All agronomic practices including weeding and insect pest management, were carried out following standard procedures for pigeonpea production in Malawi (Kananji et al., 2016). Grain yield data was collected following descriptors outlined by The International Board for Plant Genetic Resources (IBPGR, 1993).

Table 3.3. Descriptors for the pigeonpea qualitative and quantitative traits

Traits	Code	Description
Qualitative traits		
Growth habit	GH	1=Compact (erect), 2=semi-spreading (semi-erect) or 3=spreading
Flower streak pattern	FSP	0= no streaks, 1=Sparse, 2=medium and 3=dense streaks, 4= uniform coverage of second colour
Flower base/main colour	FMC	1=Ivory (green white), 2= light yellow, 3= yellow, 4= orange, 5= red, 6= purple
Pod colour	PC	1=Green, 2=purple, 3=mixed (green +purple) and 4=dark purple
Seed colour pattern	SCP	1= Plain, 2= mottled, 3=speckled, 4=Mottled and speckled, 5=ringed
Seed main colour	SMC	1= white (yellow white), 2= cream (grey white), 3= orange, 4=brown, 5=grey, 6= purple, 7= black
Seed eye colour	SEC	1= Purple, 2= light brown, 3= reddish brown, 4= grey/dark, 5= cream/white
Seed shape	SS	1=Oval, 2=pea-shape, 3= square/angular, 4= elongate
Quantitative traits		
Plant height	PH	Measured in cm from plant base to the tip of the main stem
Days to 50% flowering	DTF	Number of days from sowing until when 50% of the plants have at least one open flower
Primary branches	NPB	Average number of primary branches of 10 randomly selected and tagged plants
Secondary branches	NSB	Average number of secondary branches of 10 randomly selected and tagged plants
Days to 75% maturity	DTM	Number of days from sowing until when 75% of the pods in a plot turn brown
Number of seeds per pod	NSP	Average number of pods per plant from 10 randomly selected and tagged pods
Number of pods per plant	NPP	Average number of pods from 10 randomly selected and tagged plants
Number of racemes per plant	NRP	Average number of racemes from 10 randomly selected and tagged plants
Grain yield (t/ha)	GYD	Weight of the grain harvested in a plot extrapolated to t/ha
100 seed weight (g)	HSW T	Weight of a random sample of 100 grain

Grain yield was converted to kg ha^{-1} using on the following formula:

$$\left(\frac{Plot\ weight\ (g)}{Plot\ area\ (m)} \times \frac{100-14}{100-mc}\right) \times 10{,}000$$

Equation [1]

where; mc is moisture content measured at harvesting, 14% is standard constant moisture content for legumes (Parker A and Namuth-Covert D 2017) and 10,000 is the conversion factor for a hectare.

3.2.4 Statistical Analysis

Data collected on qualitative traits (Table 3.3) were subjected to frequency distribution and cross tabulation analyses using SPSS for Windows 25.0 (SPSS, 2018).

The quantitative data from each variable was tested for homogeneity using the Bartlett's test and normality using the Shapiro-Wilks test prior to analysis of variance (ANOVA). Subsequently, the data was pooled across sites and subjected to a combined analysis of variance following the alpha lattice procedure in Genstat 18th edition (Payne et al. 2017). The total variance was partitioned into genotype (σ^2g), environment (σ^2e) and genotype by environment (σ^2ge) components based on the mean squares derived from the partial analysis of variance adapted from (Shimelis and Shiringani 2010). Correlation and principal component analyses were performed using Genstat 18th edition (Payne et al. 2017) to determine influential components and trait relationships. Subsequently, the grain yield data was subjected to AMMI analysis of variance to partition genotype and genotype × environment interaction effects following the model presented by (Gauch Jr 1988):

$$Y_{ij} = \mu + G_i + E_j + \sum_{n=1}^{n} \lambda_k \alpha_{ik} \gamma_{jk} + \varepsilon ij \qquad [2]$$

where Y_{ij} is the yield of the i^{th} genotype in the j^{th} environment; μ is the grand mean; Gi is the genotype mean deviation; Ej is the environment mean deviation; λ_k is the eigenvalue of the principal component (PCA) axis; α_{ik} and γ_{jk} are the genotype and environment PCA scores for the PCA$_k$ axis of the i^{th} genotype and j^{th} environment respectively; and εij is the residual error.

The stability of the genotypes across locations was tested by calculating the AMMI stability values for each genotype according to Purchase et al. (2000):

$$AMMI\ Stability\ Value\ (ASV) = \sqrt{\left[\left(\frac{\text{SSIPCA1}}{SSIPCA2}(IPCA1_{score})\right)^2 + [IPCA2_{score}]^2\right]} \quad [3]$$

Where ASV is the AMMI stability value, IPCA1 and IPCA2 are the first and second interaction principal component axes, and SSIPCA1 and SSPCA2 are the sum of squares for IPCA1 and IPCA2, respectively.

The relationships among genotypes, environments, and between genotypes and environments were further illustrated graphically using the GGE biplot based on the following model (Yan et al. (2000):

$$Y_{ij} - \mu - \beta_j = \gamma_1\delta_{i1}\lambda_{j1} + \gamma_2\delta_{i2}\lambda_{j2} + \varepsilon_{ij} \quad [4]$$

Where, Y_{ij} is the measured mean yield of genotype i in environment *j;* μ is the grand mean; β_j is the main effect of environment; j, γ_1 and γ_2 are singular values for the first and second principal components, respectively; δ_{i1} and δ_{i2} are eigenvectors of genotype i and the first and second principal components, respectively; λ_{j1} and λ_{j2} are eigenvectors of environment j and the first and second principal components, respectively; and ε_{ij} is the residual associated with genotype i, in environment j.

3.3 Results

3.3.1 Variation Based on Qualitative Traits

Significant variations were exhibited among genotypes for all assessed qualitative traits ($p<0.001$) such as growth habit, flower main colour, flower streak pattern, pod colour and seed traits (Table 3.4, Figure 3.1 A to D). A large proportion of test genotypes (61.9%) were semi-spreading, followed by spreading (26.6%) and compact (11.5%) in growth habits. A majority of the test genotypes (64.9%) had yellow flower colour (Table 3.4, Figure 3.1 A), while 16.8% had purple flowers, 13.6% had ivory flowers and 7.4% had light yellow flowers (Table 3.4, Figure 3.1 A). A large population of the genotypes (60.5%) had no flower streaks and the rest of the genotypes had sparse, medium, dense and uniform coverage streaks at 8.1, 1.9, 14.5 and 15%, respectively (Table 3.4, Figure 3.1 B). About 48.7% of the genotypes had a green pod colour, while 33.9% had a mixed pod colour and 7.1% had purple pods (Table 3.4, Figure 3.1 C). A majority of the genotypes (76.8%) had a cream seed coat colour, while

11% had a brown seed coat colour and the rest had grey, orange and purple seed coat colours (Table 3.4, Figure 3.1D). About 70.2% of the test genotypes had a brown seed eye and 20.7% had a purple seed eye, while the remainder had grey or cream seed eyes. The most common seed shape was square or angular shapes, exhibited by 69.3% of the test genotypes.

Table 3.4. Frequency distribution and significance tests among 81 pigeonpea genotypes assessed based on qualitative traits

Trait	Description	Frequency (%)	DF	Chi-square	Genotype code[a]
Growth habit	Compact	11 5	160	304 52**	G53, G2, G1, G27, G26
	Semi-spreading	61 9			G63, G50, G28, G70, G76, G80, G51, G78, G49, G32, G62, G39, G67, G5, G8, G13, G72, G24, G74, G3,32, G22, G4, G40, G30, G52, G56, G48, G79, G36, G23,G16, G77, G7, G71, G44, G67, G46, G69, G33, G54, G20, G43, G42, G71, G62, G65,G39, G69, G17, G18, G59
	Spreading	26 6			G45, G41, G29, G49, G56, G64, G37, G60, G15, G11, G65, G75, G81, G44, G67, G11, G46
Flower colour	Ivory	13 6	240	910 08***	G78, G40, G36, G27, G33, G80, G51
	Light yellow	7 4			G13, G5, G31
	Yellow	64 9			G50, G45, G70, G53, G76, G72, G24, G74, G3, G22, G4, G58, G68, G18, G19, G17, G9, G62, G29, G32, G65, G21, G52, G1, G56, G37, G48, G79 G23, G16, G61, G77, G7, G71, G44, G15, G67, G11, G69, G65, G75, G20, G43, G26, G71, G44, G15, G67,G62, G11, G46, G65
	Purple	16 8			G63, G28, G41, G56, G60, G25, G46, G54, G26, G42
Flower streak pattern	No streaks	60 5	320	589 69***	G17, G53, G36, G12, G15, G37, G20, G60, G9, G54, G11, G66, G55, G80, G81, G71, G73, G23, G1, G65, G21, G18, G7, G13,G51, G62, G48, G49, G58, G14, G32, G16, G2, G27, G22, G6, G57G10, G31, G8, G39, G30
	Sparse streaks	8 1			G49, G69, G42, G33, G28, G5, G70
	Medium sparse	1 9			G72, G74
	Dense streaks	14 5			G47, G61, , G29, G60, G34, G40, G45, G67, G45, , G68, G63, G77, G19
	Uniform coverage	15			G79, G50, G76, G59, G25, G46, G78, G38, G51, G75, G26, G35, G52, G56, G41, G43
Pod colour	Green	48 7	240	647 43***	G73, G42, G1, G24, G74, G75, G52, G16, G65, G21, G18, G7, G13, G62, G17, G47, G61, G15, G20, G29, G44, G72, G60, G64, G9, G11, G66, G55, G80, G71, G58, G14, G27, G6, G57, G10, G8, G19
	Purple	7 1			G76, G45, G67, G38
	Mixed (green +purple)	33 9			G81, G70, G53, G36, G61, G43, G37, G34, G54, G79, G50, G40, G25, G33, G46, G42, G51, G4, G68, G26, G49, G3, G35, G32, G69, G2, G63, G22, G56, G77,G41, G30
	Dark purple	10 3			G31, G28, G39, G48, G59, G43
Seed colour pattern	Plain	56 6	240	841 57***	G59, G80, G5, G18, G6, G53, G65, G62, G35, G34, G67, GG4, G60, G66, G21, G70, G36, G42, G40, G14, G50, G66, G20, G79, G49, G2, G3, G69, G56, G81, G47, G72,G15, G44
	Mottled	15 3			G41, G25, G34, G48, G28, G78, G23, G31, G9, G37, G57
	Speckled	22 2			G75, G68, G43, G38, G10, G19, G52, G58, G51, G73, G59, G76, G16, G29, G13, GG3, G17, G8, G54, G1, G24, G7, G71, G27, G12, G22, G55, G77
	Mottled + speckled	5 9			G46, G33, G30, 632, G39, G45, G26
Seed main colour	Cream	76 8	320	1049 31***	G75, G68, G59, G43, G5, G18, G6, G38, G10, G53, G65, G63, G35, G19, G34, , 52, G72, G15, G44, G22, G55, G57, G77, G60, G58, G78, G32, G73, G51, G70, G36, G16, G29, G42, G40, G23, G14, G17, G8, G50, G66, G20, G49, G54, G2, G3, G69, G1, G24, G45, G7, G9, G71, G81, G12, G47
	Orange	3			G4, G46, G25
	Brown	11			G64, G76, G63, G30, G34, G48, G28, G31, G37, G26
	Grey	6 2			G80, G66, G67, G56
	Purple	3			G39, G33, G41
Seed shape	Oval	30 7	80	480 21***	G75, G22, G5, G25, G38, G53, G35, G34, G28, G73, G51, G70, G36, G29, G42, G40, G31, G8, G18, G49, G3, G45, G37, G28, G27, G12, G55, G57
	Square/angular	69 3			G15, G44, G22, G77, G68, G59, G43, G46, G80, G18, G33, G30, G41, G6, G10, G65, G62, G19, G34, G67, G4, G52, G48, G60, G58, G66, G32, G64, G76, G21, G16, G13, G23, G14, G63, G17, G39, G52, G66, G79, G54, G2, G69, G1, G24, G56, G7, G9, G71, G81
Seed eye colour	Purple	20 7	240	848 32***	G68, G5, G34, G25, G60, G78, G51, G64, G76, G21, G16, G29, G42, G40, G31, G50, G49, G2, G69, G24, G81, G55, G57
	Light brown	70 2			G75, G59, G43, G46, G18, G33, G30, G41, G6, G10, G53, G65, G62, G35, G19, G34, G67, G52, G48, G58, G28, G66, G32, G73, G36, G23, G14, G17, G39, G74, G20, G79, G54, G1, G46, G45, G9, G71, G37, G27, G12, G47, G15, G44, G22
	Grey/dark	1 2			G25
	Cream	7 5			G80, G38, G63, G8, G7, G26

DF= degrees of freedom, *, ** and ***= significance at 0.05, 0.01 and 0.001 levels, respectively; For genotype code refer to Table 3.1

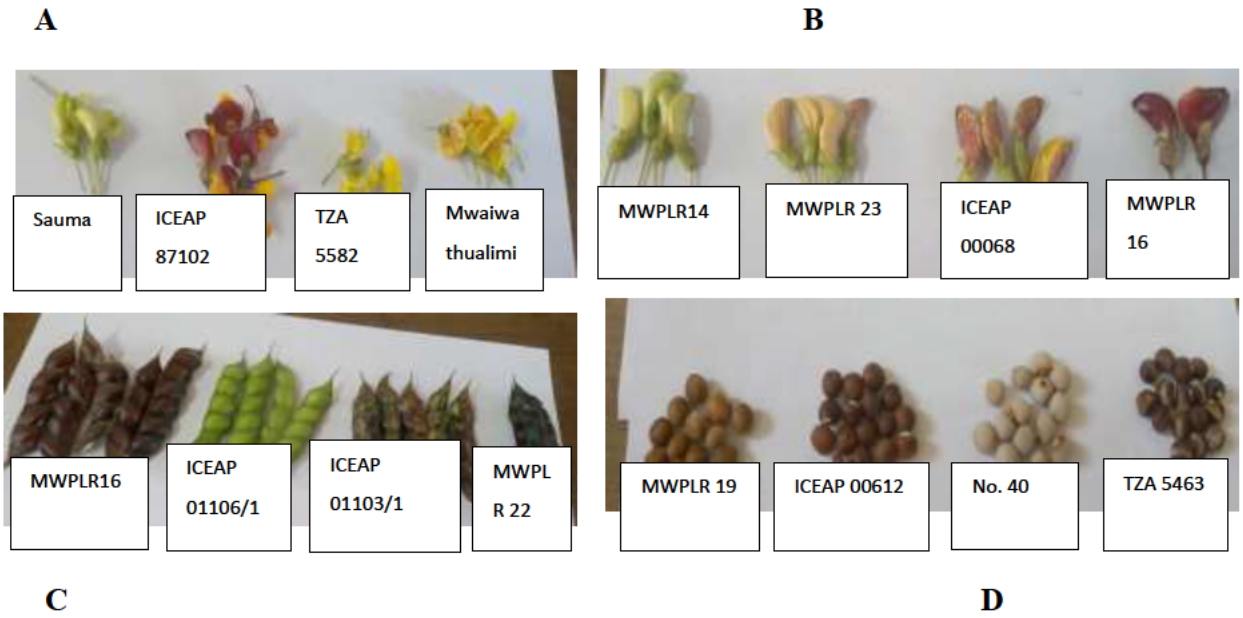

Figure 3.1. Genetic variation for some qualitative traits in pigeonpea genotypes: (A) flower colour: genotype Sauma (ivory), ICEAP 87105 (purple), TZA 5582 (yellow), Mwaiwathualimi (light yellow); (B) flower streak pattern: genotype MWPLR 14 (no streak), MWPLR 23 (medium streaks), ICEAP 00068 (dense streaks), MWPLR 16 (uniform coverage); (C) pod colour: genotype MWPLR 16 (purple), ICEAP 01106/1 (green), ICEAP 01103/1 (mixed), MWPRL 22 (dark purple) and (D) seed coat colour: genotype MWPLR 19 (orange), ICEAP 00612 (brown) No. 40 (cream) and TZA 5463 (purple).

3.3.2 Genotype and Environment Variances for Quantitative Traits

The quantitative agronomic data was pooled across sites after applying tests for homogeneity of variance and normality. The genotype × environment interaction effects were significant ($P<0.001$) for GYD, DTF, DTM, PH, NPB, NPP, NRP HSWT and NSB (Table 3.5). The genotype and environment had significant ($P<0.001$) effects on all assessed traits except the NSP.

3.3.3 Mean Performance of Pigeonpea Genotypes Across the Test Environments

Table 3.6 summarizes the mean values and statistics for eight quantitative traits that were recorded from three locations in two seasons (six environments). The table presents the best ten and bottom five genotypes ranked on grain yield response. The raw data of all test genotypes across the environments is presented in Appendix 2. The mean DTF and DTM were 112 and 157 days, respectively. Genotype MWPLR 14 was the earlier to attaining 50%

flowering and maturity at 74 and 113 days, followed by ICEAP 01170 at 85 and 125 days, ICEAP 87091 at 85 and 132 days, ICEAP 01285 at 87 and 133 days and ICEAP 01169 at 91 and 137 days, respectively. Sauma was among the slowest genotype to flower and mature at 145 and 205 days, respectively. There were marked genotype difference in plant height that varied from 125.3 and 202.4 cm. The mean plant height of the test genotypes was 167.5 cm. The shortest and most desired genotype across the testing environments was ICEAP 87105 (125.3 cm). The tallest genotypes recorded were Kachangu, No. 40, ICEAP 01106/3, ICEAP 00068, TZA 5596, MWPLR 6, Sauma and ICEAP 00053, which had above 180 cm plant height. The mean number of the primary branch of the test genotypes was 15. The most productive genotypes with many primary branches per plant were MWPLR 12, MWPLR 20, ICEAP 01170 and MWPLR 23, with 19, 18, 17 and 17 primary branches per plant, in that order. The mean number of pods per plant varied from 67 to 144, with a grand mean of 94 pods per plant. The highest number of pods per plant were 144, 134, 126, 124 and 123 for the genotypes Kachangu, MWPLR 16, TZA 5582, No. 40 and MWPLR 14, in that order. The number of seeds per pod exhibited non-significant differences among the assessed genotypes. The mean number of grain per pod was five. There was a wide genetic variation for grain yield that ranged from 0.5 to 1.8 t ha^{-1} with a mean of 1.1 t ha^{-1}. Accessions No. 40, MWPLR 14 and MWPLR 16 were the three best performing genotypes with mean yields of 1.8, 1.7 and 1.7 t ha^{-1}, respectively. The lowest grain yield response was 0.5 t ha^{-1} recorded for the genotypes ICEAP 00604 and ICEAP 01285. The 100 seed weight ranged from 11.0 to 17.3 g/100 seed. Accessions MWPLR 22, TZA 5582 and MWPLR 14 expressed the highest HSWT ≥17 g/100 seed.

Table 3.5. Mean squares and significant tests for grain yield and yield components measured in 81 pigeonpea genotypes across six environments in Malawi.

Source of variation	DF	DTF	DTM	PH	NPB	NSB	NRP	NPP	NSP	GYD	HSWT
Environment (E)	5	5202.1***	3755.2***	44162.9***	499.0***	1069.6***	1335956.0***	194398.0***	29.9***	16.7***	475.7***
Replication	6	287.2^{ns}	1560.7***	2452.0***	39.6***	78.7*	10242.0****	4062.0**	2.5**	0.2^{ns}	50.5**
Block (Rep)	96	438.7***	794.3***	25.98.6***	20.3**	50.7***	9822.0***	2951.0***	1.1.*	0.3***	32.9***
Genotype (G)	80	1038.5***	1440.1***	1906.0***	17.3*	24.1*	7226.0***	2400.0***	0.7^{ns}	0.5***	16.9*
G × E	400	356.5***	525.2***	1097.8***	15.8*	27.3*	7624.0***	2.1***	0.8^{ns}	0.3***	15.1*
Residual	384	245.4	379.3	755.5	13.2	29.9	2585.0	1.68	0.8	0.2	14.7

DF= degrees of freedom, Rep= replication, DTF= days to 50% flowering, DTM= days to 75% maturity, PH= plant height, NPB= number of pods per plant, NSB= number of secondary branches per plant, NRP = number of racemes per plant, NPP= number of primary branches per plant, GYD= grain yield, HSWT= 100 seed weight, *, ** and ***= significance at 0.05, 0.01 and 0.001 probability levels, respectively

Table 3.6. Mean values for 10 quantitative traits among the ten top best and five bottom performing genotypes after evaluating 81 genotypes in six environments in Malawi.

	DTF							DTM						
	Y1			YII				YI			YII			
Genotype	S1	S2	S3	S1	S2	S3	Mean	S1	S2	S3	S1	S2	S3	Mean
Top ten genotypes														
21	129	131	141	124	131	132	131	173	191	211	158	176	176	181
43	125	105	119	117	105	105	113	177	166	172	156	161	154	164
51	63	65	64	87	67	98	74	95	105	102	127	116	132	113
30	100	97	118	128	116	118	113	133	150	164	159	159	164	155
45	107	96	91	128	101	124	108	143	158	146	170	153	165	156
81	163	127	155	132	165	130	145	215	201	254	171	211	178	205
17	147	120	125	109	120	106	121	182	167	174	156	160	147	164
66	120	95	115	116	108	116	111	155	151	170	154	158	161	158
74	118	78	123	113	115	118	110	163	145	166	153	165	163	159
20	116	120	129	122	120	127	122	143	163	175	156	160	172	161
Bottom five genotypes														
39	113	90	131	85	90	88	99	149	144	195	127	150	122	147
13	126	117	109	116	107	115	115	167	166	153	145	154	155	156
50	117	77	107	116	77	115	101	141	136	156	155	137	149	145
42	114	102	127	120	102	120	114	145	154	172	164	166	162	160
79	124	101	122	117	127	119	118	168	153	165	152	179	161	163
Mean	117.8	102.8	115.5	110.6	106.1	113.1	110.6	154.7	156.5	163.2	148.7	155.7	154 3	155.3
STD	17.9	18.2	15.1	13.0	16.9	12.3	10.5	22.0	22.0	21.1	13.7	18.4	14.9	11.9
SED±	2.0	2.0	1.7	1.4	1.9	1.4	1.2	2.4	2.4	2.3	1.5	2.0	1.7	1.3
CV(%)	15.2	17.7	13.1	11.8	15.9	10.8	9.5	14.2	14.0	12.9	9.2	11.8	9.6	7.7

Table 3.6. Continued

	PH							NPB						
	Y1			Y11				Y1			Y11			
Genotype	S1	S2	S3	S1	S2	S3	Mean	S1	S2	S3	S1	S2	S3	Mean
Top ten genotypes														
21	166.5	220.0	193.0	160.0	212.8	193.0	190.9	19	19	17	14	18	12	16
43	113.5	147.5	127.5	96.5	146.7	148.0	163.7	14	15	17	14	17	11	15
51	151.5	109.0	158.0	234.5	209.4	149.0	168.6	13	12	14	18	13	11	13
30	229.5	188.5	204.0	170.0	218.5	204.0	202.4	15	13	18	15	16	15	15
45	139.5	144.5	173.0	161.5	169.4	197.5	164.2	15	13	22	15	17	14	16
81	163.0	222.0	191.0	160.5	168.1	194.5	183.2	13	17	19	18	12	14	15
17	163.5	164.0	163.5	100.0	152.1	156.0	149.9	15	14	21	17	16	13	16
66	181.5	177.5	164.0	161.5	156.8	149.5	165.1	12	13	13	14	16	12	13
74	156.0	195.0	185.5	124.5	178.7	164.0	167.3	15	18	17	20	18	12	17
20	152.5	163.0	168.5	138.5	247.5	166.5	172.8	10	12	20	12	18	11	14
Bottom five genotypes														
39	203	154.5	174	157.5	200	151.5	173.4	16	18	17	15	12	12	15
13	169	171.5	134	134.5	203.3	156.5	161.5	18	12	18	15	10	15	14
50	119	101.5	149.5	130.5	218.5	166.5	147.6	18	13	14	15	17	13	15
42	140	153	175.5	104.5	207.7	120	125.3	14	9	16	14	13	13	13
79	174	165.5	167.5	120.5	201.4	148	162.8	11	18	23	13	14	13	15
Mean	168.0	166.7	166.2	143.4	195.5	166.1	167.3	14.6	13.6	18.0	14.9	14.6	12.8	14.5
STD	23.9	34.5	22.1	23.0	27.0	23.1	12.6	2.7	4.4	2.7	2.4	3.2	2.0	1.3
SED±	2.7	3.8	2.5	2.6	3.0	2.6	1.4	0 3	0 5	0 3	0.3	0.4	0.2	0.1
CV (%)	14.2	20.7	13.3	16.0	13.8	13.9	7.5	18.7	32.1	15.0	16.3	22.0	15.6	9.1

Table 3.6. Continued

	NRP							NPP						
	Y1			Y11				Y1			Y11			
Genotype	S1	S2	S3	S1	S2	S3	Mean	S1	S2	S3	S1	S2	S3	Mean
Top ten genotypes														
21	214	402	71	130	61	47	154	157	270	66	61	92	98	124
43	138	173	97	117	95	58	113	119	315	98	72	110	90	134
51	260	155	146	113	80	51	134	167	231	109	65	76	90	123
30	178	430	134	132	73	52	166	127	362	95	97	83	101	144
45	191	647	160	151	88	83	220	96	261	106	81	92	122	126
81	200	536	85	89	69	40	170	140	240	69	61	70	82	110
17	184	258	96	139	94	61	139	102	158	65	35	112	89	93
66	148	168	108	119	76	49	111	69	186	82	26	78	64	84
74	196	414	98	125	84	81	166	128	112	64	46	40	94	81
20	126	259	106	148	130	73	140	115	177	78	38	157	45	101
Bottom five genotypes														
39	161	465	103	145	55	60	165	128	125	93	38	61	82	88
13	155	228	80	119	99	52	122	98	195	55	37	60	95	90
50	116	321	199	195	81	46	159	79	78	60	59	96	84	76
42	122	150	87	151	80	62	109	99	78	90	62	67	70	78
79	98	552	70	131	163	54	178	53	226	51	26	165	90	102
Mean	174.1	312 3	99.0	161.6	91.8	58.9	149.4	114.6	148.2	80.0	51.0	80.9	86.7	93.4
STD	43.9	146 5	27.7	39.8	30.0	12.1	26.2	30.5	56.7	22.1	16.1	33.4	19.7	14.1
SED±	4.9	16.3	3.1	4.4	3.3	1.3	2.9	3.4	6.3	2.5	1.8	3.7	2.2	1.6
CV (%)	25.2	46.9	28.0	24.7	32.7	20.6	17.5	26.6	38.2	27.7	31.5	41.3	22.8	15.1

Table 3.6. Continued

	GYD							HSWT						
	Y1			Y11				Y1			Y11			
Genotype	S1	S2	S3	S1	S2	S3	Mean	S1	S2	S3	S1	S2	S3	Mean
Top ten genotypes														
21	2.1	0.9	2.3	2.4	1.3	1.7	1.8	16.0	16.5	10.0	10.5	12.5	15.5	13.5
43	1.7	1.7	1.6	1.8	1.6	1.9	1.7	17.0	14.5	14.0	17.0	22.5	13.0	16.3
51	1.8	1.0	2.1	2.1	1.7	1.7	1.7	16.5	17.5	14.5	18.5	21.5	13.5	17.0
30	2.3	1.6	1.2	1.2	1.4	1.8	1.6	17.5	17.0	15.0	16.0	16.0	12.0	15.6
45	1.5	0.9	1.4	1.5	2.3	1.9	1.6	18.4	19.0	15.5	16.0	16.5	18.0	17.2
81	1.3	0.5	1.5	1.6	2.3	2.3	1.6	19.5	16.0	15.5	19.0	15.0	11.0	16.0
17	1.1	0.5	0.7	1.4	2.5	3.0	1.5	18.5	14.0	11.0	17.5	20.0	15.5	16.1
66	2.4	1.5	1.2	1.2	1.4	1.5	1.5	15.5	15.5	15.0	17.5	17.5	13.5	15.8
74	2.2	1.6	1.1	1.0	1.1	1.8	1.5	14.5	14.5	15.5	16.9	20.0	13.5	15.8
20	1.2	0.9	1.7	1.7	1.7	1.2	1.4	16.0	12.5	15.0	18.5	15.0	14.0	15.2
Bottom five genotypes														
39	0.4	0.4	1.1	1.1	1.2	0.9	0.8	15.5	14.5	14.5	16	15	16	15.3
13	0.8	0.2	0.5	1.4	0.5	0.3	0.6	12.5	15	14	15	16.5	16	14.8
50	0.9	0.5	0.4	0.5	0.4	0.7	0.6	13	10.5	17.5	21	19	14.5	15.9
42	0.6	0.4	0.9	0.5	0.4	0.4	0.5	12	12.5	14	19	20	14.5	15.3
79	0.8	0.3	0.4	0.5	0.3	0.5	0.5	13	16.5	14.5	17.5	17.5	14	15.5
Mean	1.1	0.6	1.3	1.3	1.5	1.3	1.2	15.9	13.9	13.5	17.6	12.9	14.2	14.7
STD	0.4	0.3	0.4	0.4	0.5	0.4	0.2	2.4	3.2	2.4	2.3	4.5	2.5	1.3
SED±	0.0	0.0	0.0	0.0	0.1	0.0	0.0	0.3	0.4	0.3	0.3	0.5	0.3	0.1
CV (%)	37.3	43.3	32.8	32.1	31.3	33.5	20.5	15.1	22.9	18.0	13.2	35.1	17.5	8.9

STD= standard deviation, SED= standard error of difference, CV= coefficient of variation, S1= site 1 (Bvumbwe), S2= site 2 (Chitedze), S3= site 3 (Makoka), Y1= year 1 (2017/18), Y11= year 2 (2018/19), DTF= days to flowering, DTM= days to 75% maturity, PH= plant height (cm), NPB= number of primary branches, NRP= number of racemes per plant, NPP= number of pods per plant, GYD= grain yield (t ha^{-1}), HSWT= 100 seed weight (g), See genotype codes (G1-G81) in Table 3.1.

3.3.4 Correlation Analysis Among Phenotypic Traits

Assessed traits exhibited variable degree of associations with grain yield (Table 3.7). Grain yield was moderately correlated with HSWT (r=0.50, p<0.01), NPP (r=0.35, p<0.01) and DTF (r=0.23, p<0.05). A number of secondary traits exhibited variable pairwise correlations. DTF and DTM exhibited the strongest correlation (r=0.79, p<0.01). There were moderate correlations between DTF and PH (r=0.44, p<0.01), NPB and NSP (r=0.41, p<0.01) and, DTM and PH (r=0.41, p<0.01). Relatively, HSWT exhibited weak correlations (r<0.30) with NPB and NPP.

Table 3.7. Phenotypic correlation coefficients among the ten quantitative traits of 81 pigeonpea genotypes evaluated in six environments

Trait	DTF	DTM	PH	NPB	NSB	NRP	NPP	NSP	GYD	HSWT
DTF	1	0.787**	0.442**	0.069	0.006	0.063	0.121	-0.134	0.232*	-0.021
DTM		1	0.409**	0.066	0.037	0.034	0.121	-0.020	0.131	0.023
PH			1	0.057	0.149	0.249*	0.190	-0.123	0.123	0.021
NPB				1	0.044	0.261*	0.145	0.406**	0.174	0.350**
NSB					1	0.024	0.152	-0.101	0.214	0.090
NRP						1	0.191	0.262*	0.177	0.124
NPP							1	0.099	0.354**	0.307**
NSP								1	0.051	0.173
GYD									1	0.498**
HSWT										1

**. Correlation is significant at the 0.01 level, *. Correlation is significant at the 0.05 level (2-tailed), DTF= days to 50% flowering, DTM= days to 75% maturity, PH= plant height, NPB= number of pods per plant, NSB= number of secondary branches per plant, NRP = number of racemes per plant, NPP= number of primary branches per plant, GYD= grain yield, HSWT= 100 seed weight

3.3.5 Principal Component (PC) Analysis

The principal component analysis of agronomic traits among the pigeonpea genotypes revealed that the three most important principal components (PCs) accounted for 25.3, 44.7 and 57.7% of the total variation, respectively (Table 3.8). Days to flowering with a loading score of 0.91 and days to maturity (0.88) contributed the most to the variation on the first PC. The NSP and HSWT exhibited negative associations with the first PC. The variation on the second PC was contributed the most by HSWT and GYD with loading scores of 0.68 and 0.77, respectively. Only NSP exhibited negative loading to PC2. The major contributors of the variation explained by PC3 were the number of primary branches and number of seeds per pod. The DTF and NRP were negatively associated with PC3. The relationship between the measured traits and the genotypes is depicted by the principal component bi-plot, where

PC1 and PC2 contributed to 74.99% of the total variation (Figure 3.2). The following traits: DTF, DTM, PH, NRP, NPP and GYD, were highly correlated due to their unidirectional line vectors with small angles between them. Genotypes 81 and 71 performed better in terms of grain yield, with positive trait correlations with NRP, NPP, DTF and DTM. The following genotypes: G24, G69, G45 and G79, were the best in HSWT, NPB, NRB and NSB, respectively. Such genotypes will be useful for introgression of genes into superior genotypes.

Table 3.8. Principal components showing variation and contribution by 10 quantitative traits among 81 pigeonpea genotypes assessed in six environments in Malawi

Parameter	Principal components		
	PC 1	PC 2	PC 3
Eigen value	2.525	1.948	1.298
Variance (%)	25.254	19.492	12.98
Cumulative variance (%)	25.254	44.746	57.726
DTF	0.908	0.042	-0.004
DTM	0.881	0.003	0.052
PH	0.689	0.164	0.011
NPB	0.041	0.192	0.732
NSB	0.164	0.133	0.573
NRP	0.022	0.564	-0.271
NPP	0.146	0.619	0.17
NSP	-0.154	-0.069	0.799
GYD	0.148	0.771	0.129
HSWT	-0.094	0.68	0.346

DTF= days to 50% flowering, DTM= days to 75% maturity, PH= plant height, NPB= number of pods per plant, NSB= number of secondary branches per plant, NRP = number of racemes per plant, NPP= number of primary branches per plant, GYD= grain yield, HSWT= 100 seed weight

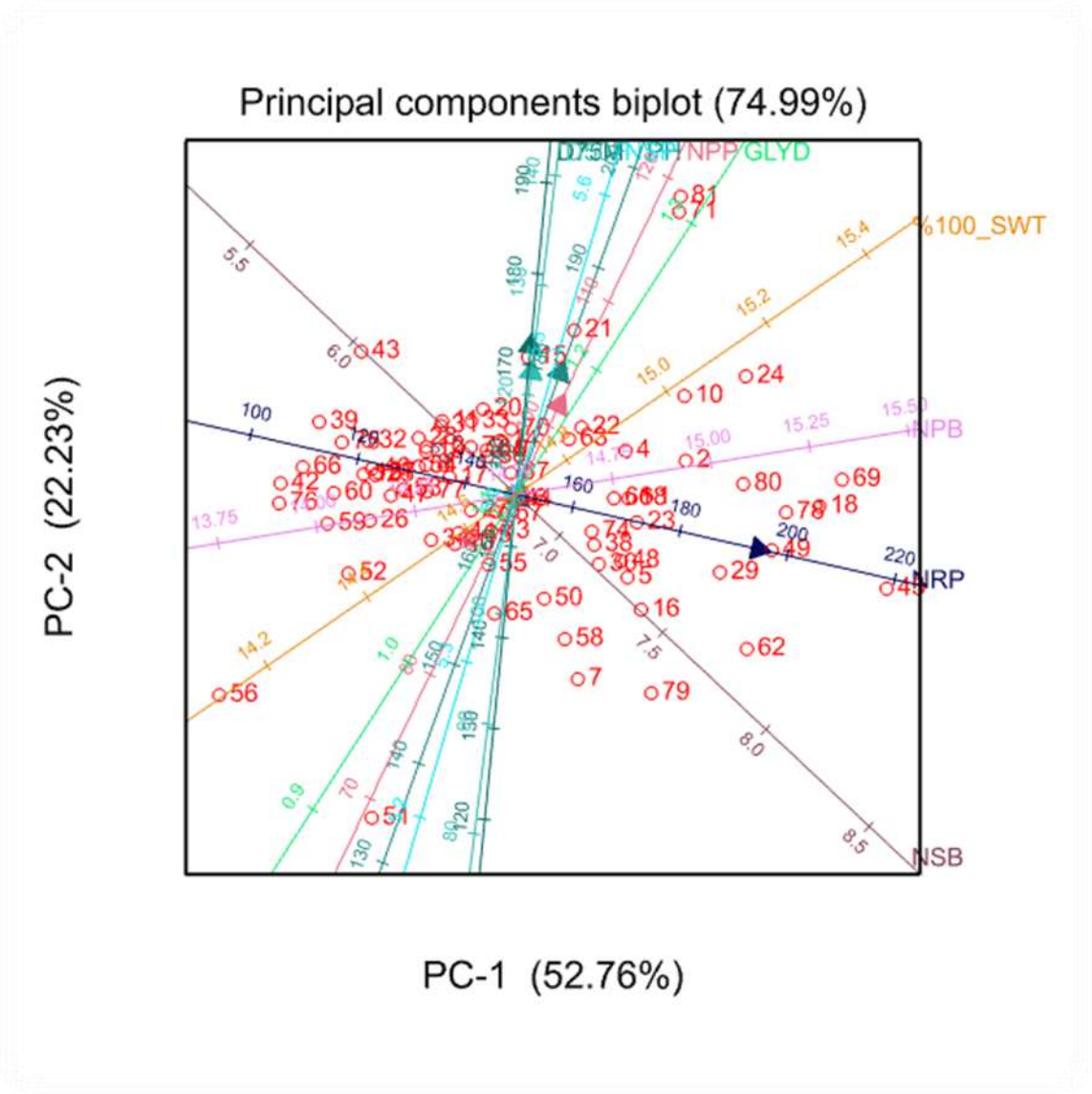

Figure 3.2. Genotype-trait biplot showing relationship of 10 quantitative agronomic traits in 81 pigeonpea genotypes evaluated in six environments in Malawi

See Table 3.1 for codes of genotypes and Table 3.3 for traits description. PC= principal component

3.3.6 Grain Yield Stability Analysis Based on the Additive Main Effect and Multiplicative Interaction (AMMI) Model

The AMMI analysis of variance revealed that environments, genotypes, and their interactions had significant effects on grain yield (Table 3.9). The environment accounted for 33.5% of the total observed variance, while genotype and genotype × environment interaction accounted for 16.4 and 49.6%, respectively. The mean squares of interaction principal component analysis 1 (IPCA1) and interaction principal component analysis 2 (IPCA2) were

highly significant (P < 0.001) and explained 47.3 and 27.4% of the total variation, respectively. The interaction principal component analysis 3 (IPCA3) was significant and accounted for 20.5% of the variance explained by the GEI.

Table 3.9. AMMI analysis of variance for grain yield of 81 pigeonpea genotypes evaluated in six environments in Malawi

Source of variation	DF	MS	Total variation explained (%)	G × E explained (%)
Genotypes	80	0.52***	16.35	
Environments	5	16.96***	33.49	
Block	6	0.22	0.41	
Interactions	400	0.31***	49.64	
IPCA1	84	0.71***		47.33
IPCA2	82	0.42***		27.45
IPCA3	80	0.32***		20.50
IPCA4	78	0.06		3.70
IPCA5	76	0.02		1.10
Residuals	234	0.14		
Error	480	0.18		

DF= degreed of freedom, MS= mean squares, G= genotype, E= environment, G×E=genotype by environment interaction, IPCA= interaction principal component analysis, *** = significant at the 0.001 level

3.3.7 Biplot Analysis of Genotype-by-Environment Interaction for Grain Yield Based on AMMI 1 Model

Environments E1 (2017/18 season, Bvumbwe) and E2 (2018/19 season, Bvumbwe) had the lowest IPCA1 scores, which contributed most to the stability of genotypes (Figure 3.3). In terms of contribution to GEI, environments E3 (2017/18 season, Chitedze) and E6 (2018/19 season, Makoka) had high IPCA1 scores, corresponding to the highest contribution to the GEI component. The lowest mean grain yield was attained in E2 while the highest yielding environments were E1, E5 (2017/18 season, Makoka) and E6. Furthermore, environments E3 and E4 (2018/19 season, Chitedze), and E5 and E6, were highly correlated with similar signs of their IPCA1 scores. The AMMI biplot showed that genotypes with positive IPCA1 scores, including G30, G2, G43, G57, G79, G46 and G72, had positive interactions with E1, E2, E3 and E4. On the other hand, genotypes with negative IPCA1 scores, which included G24, G14, G32, G16, G12, G22 and G69, had positive interactions with E5 and E6. The stable genotypes with IPCA1 scores close to zero included G26, G51 and G27. In terms of high yield, genotypes such as G24, G29, G43 and G45 were superior (with mean grain yield of ≥1.5 t/ha^{-1}) across all the test environments.

3.3.8 Biplot Analysis of Genotype-by-Environment Interaction for Grain Yield Based on AMMI 2 Model

The first principal component axis (PCA1) scores were plotted against the second principal component axis (PCA2) scores of genotypes and environments to demonstrate the magnitude of the G × E interaction (Figure 3.4). The vector length of each environment can be used to indicate the ability of each environment to discriminate between the genotypes. The environment with the longest vector was E1 concomitant with its high capability to discriminate the genotypes. The least discriminatory environment was E2, while E3 and E4 had similar discriminatory capability. The vectors for genotypes such as G79, G46, G77, G9, G74 and G43 were correlated with vectors for environments E3 and E4. Genotypes G64, G57, G21 were correlated with environment E2. In addition, genotypes G55, 31 and 47 were associated with environment E1, while G24 associated more with environments E5 and E6. Genotypes that are associated with a particular environment have specific adaptation to that environment. Genotypes such as G29, G27, G45 and G51 that were plotted near the origin were the most stable genotypes.

3.3.9 Genotype Stability for Grain Yield Response

The mean grain yield (ton ha-^{1}) and AMMI stability values (ASV) for 20 selected pigeonpea (15 most stable and 5 least stable) genotypes are presented in Table 3.10. Genotypes G51, G27 and G26 were the most stable genotypes with respective ASVs of 0.02, 0.05 and 0.06. These genotypes were stable, although they exhibited low mean yields across the environments. On the contrary, genotypes G24, G43, G12, G21 and G40 had higher ASVs, denoting their lack of stability across the environments. However, G45 and G29 had the highest grain yield and were among the most stable genotypes, while G51 was the most stable genotype.

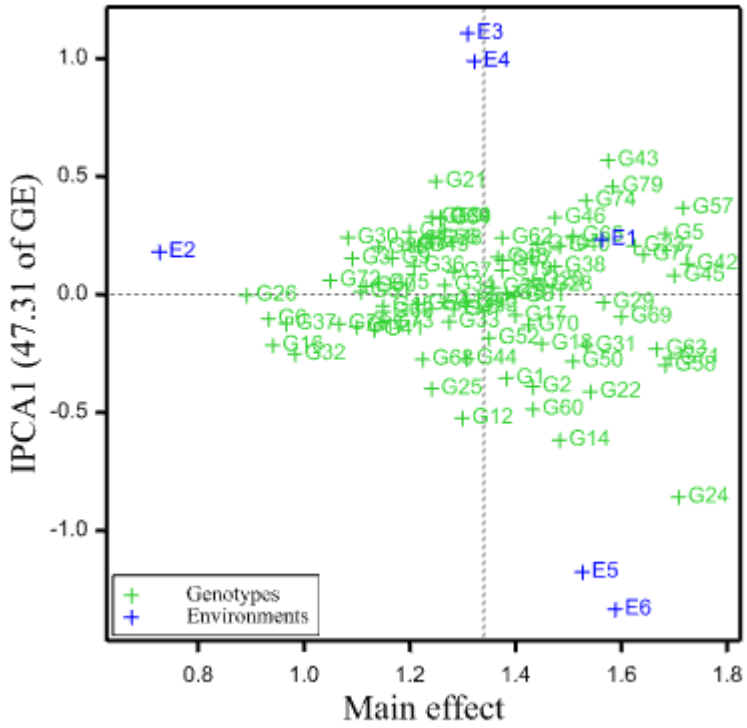

Figure 3.3. GEI biplot based on AMMI 1 model for the PCA1 scores and grain yield (t ha^{-1}) of 81 pigeonpea genotypes evaluated in six environments in Malawi

E1= Bvumbwe, 2017/18, E2= Bvumbwe in 2018/19, E3=Chitedze 2017/18, E4=Chitedze 2018/19, E5= Makoka 2017/18 and E6= Makoka in 2018/19. Dotted vertical and horizontal lines indicate zero coordinate for *x* and *y* axes, respectively. See genotype codes (G1-G81) in Table 3.1.

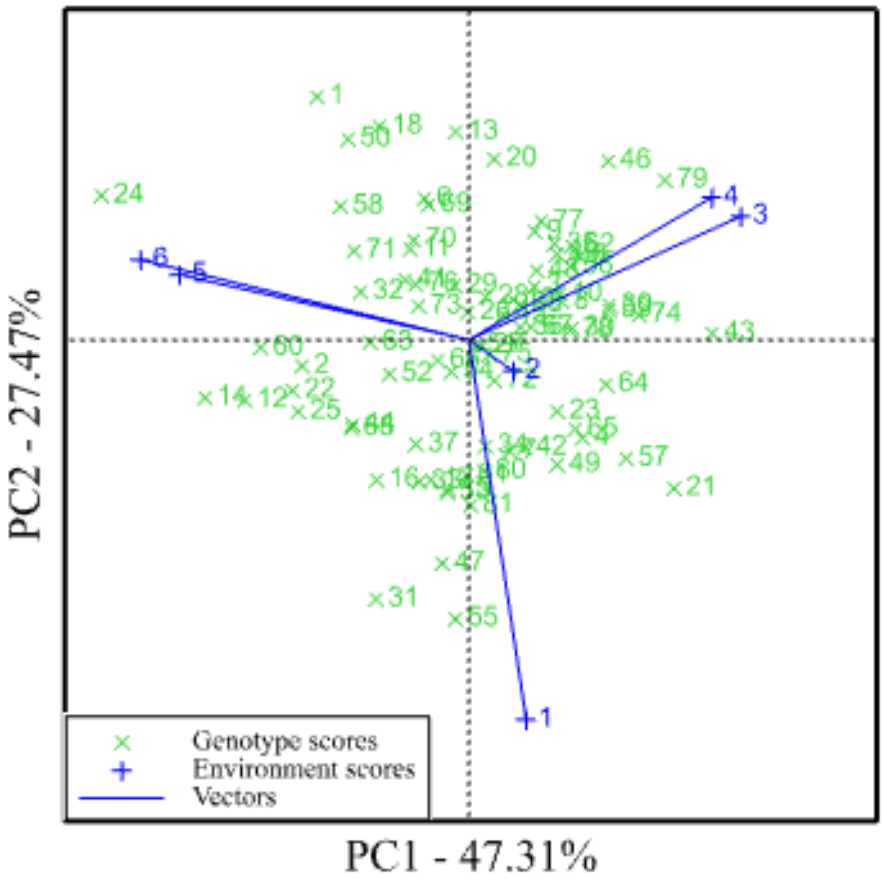

Figure 3.4. AMMI 2 model biplot for grain yield for 81 pigeonpea genotypes evaluated in six environments.

E1= Bvumbwe, 2017/18, E2= Bvumbwe in 2018/19, E3=Chitedze 2017/18, E4=Chitedze 2018/19, E5= Makoka 2017/18 and E6= Makoka in 2018/19. Dotted vertical and horizontal lines indicate zero coordinate for x and y axes, respectively. See genotype codes (G1-G81) in Table 3.1.

Table 3.10. Mean grain yield (ton ha-[1]) and AMMI stability values (ASV) for 20 selected pigeonpea genotypes

Genotype code	Mean	IPCA1	IPCA2	ASV
15 most stable genotypes				
G51	1.11	0.01	-0.01	0.02
G27	1.36	0.03	-0.01	0.05
G26	0.89	0.00	0.06	0.06
G35	1.14	0.20	0.20	0.09
G49	1.22	0.21	-0.25	0.10
G18	1.45	-0.21	0.44	0.11
G29	1.50	-0.03	0.11	0.13
G73	1.15	-0.12	0.07	0.13
G72	1.05	0.06	-0.08	0.13
G45	1.50	0.08	0.05	0.15
G19	1.38	0.10	0.08	0.19
G36	1.21	0.12	0.03	0.21
G15	1.15	-0.05	-0.30	0.21
G38	1.48	0.12	0.07	0.22
G13	1.27	-0.03	0.42	0.23
5 least stable genotypes				
G21	1.25	0.48	-0.30	0.88
G12	1.30	-0.52	-0.12	0.91
G43	1.50	0.57	0.01	0.98
G40	1.48	0.21	0.17	1.07
G24	1.50	-0.86	0.29	1.51

IPCA1 and IPCA2= first and second interaction principal component axes, respectively; ASV= AMMI stability value. See genotype codes (G1-G81) in Table 3.1.

3.3.10 Ideal Environment for High Grain Yield Response

Figure 3.5 presents the average environment coordination (AEC) view, comparing environments relative to an ideal environment. The ideal environment is a hypothetical and highly discriminative environment represented by the point at the centre of the concentric circles of the environment-centred biplot. Test environments in close proximity to the ideal environment are highly capable of differentiating the tested genotypes and are a good representative of the target location. Environments E1 and E2 were located closest to the ideal environment, showing that they were better environments for evaluating the pigeonpea genotypes, followed by environments E3 and E4. On the other hand, E5 and E6 were plotted farthest from the ideal environment, with large PC2 and smaller PC1 scores, showing that they were neither representative nor highly discriminative. Environments E1, E2, E3 and E4 were identified with high mean yields, while E5 and E6 were identified as low yielding environments, from the AMMI biplot (Figure 3.4).

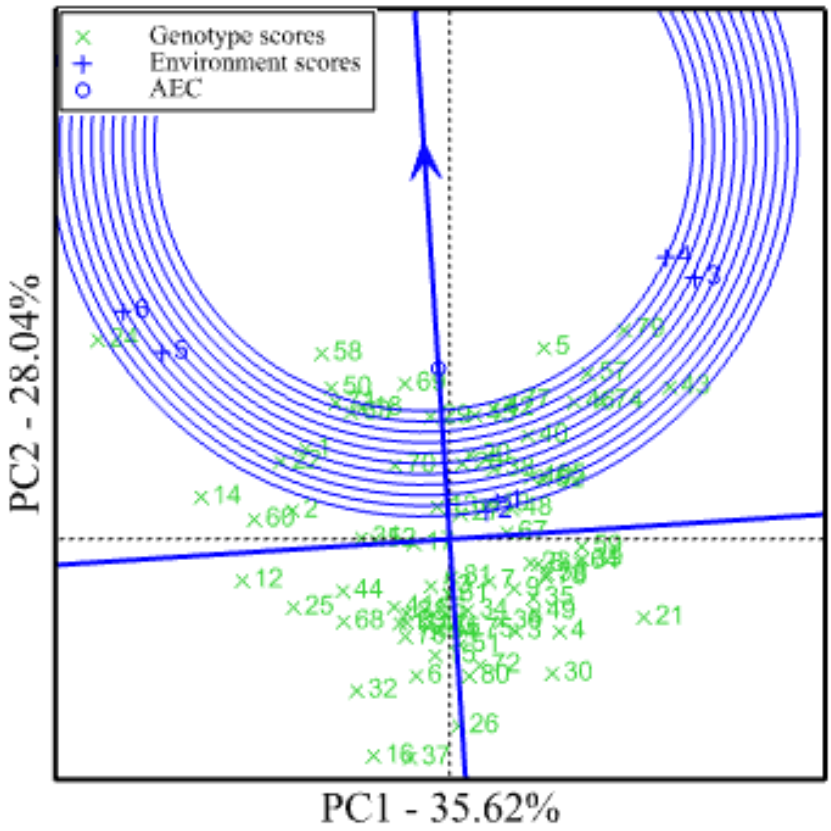

Figure 3.5. GGE biplot comparing the six test environments with the ideal environment based on grain yield of 81 pigeonpea genotypes.

E1= Bvumbwe, 2017/18, E2= Bvumbwe in 2018/19, E3=Chitedze 2017/18, E4=Chitedze 2018/19, E5= Makoka 2017/18 and E6= Makoka in 2018/19. Dotted vertical and horizontal lines indicate zero coordinate for x and y axes, respectively. The small circle on the arrowed line indicates the average environment, the arrow indicates the ideal environment, and concentric circles indicate the distances of genotypes and environments from the ideal environment. See genotype codes (G1-G81) in Table 3.1.

3.3.11 Ideal Genotype

Genotype yield performance and stability were depicted using the AEC (Figures 3.6 and 3.7). According to Yan and Tinker (2006), the AEC is the line that passes through the biplot origin and is defined by PC1 (mean yield) and PC2 (stability) scores for all the environments. The line that passes the biplot origin and is perpendicular to AEC represents stability of the genotypes. Genotypes located away from the biplot origin in either direction of the AEC indicate greater GEI and reduced stability. An ideal genotype is regarded as one that has a high mean yield (PC1) and a low GEI or high stability (PC2). In a GGE biplot, concentric circles are drawn to visualize the distance between each genotype and the ideal genotype.

About 60 genotypes, including G21, G45, G30, G57 and G43, had above average yields in all the environments, while the remaining genotypes, including G12, 24, G25, G14 and G60, attained below average yield. The highest yielding genotypes were G43, G21 and G79. Genotypes G36, G27, G45, G29, G72 and G51 were plotted in close proximity to the stability axis, showing that they were stable. However, stable genotypes are desirable when they have high mean performances and are located close to the ideal genotype in the GGE biplot. In this case, G51 was the most desirable genotype since it had a high yield (above mean yield) and was stable, with a PC2 score close to zero. The stability results for GGE were almost similar to AMMI (Figures 3.3 and 3.4), which identified G51, G27 and G26 as the most stable genotypes across the environments.

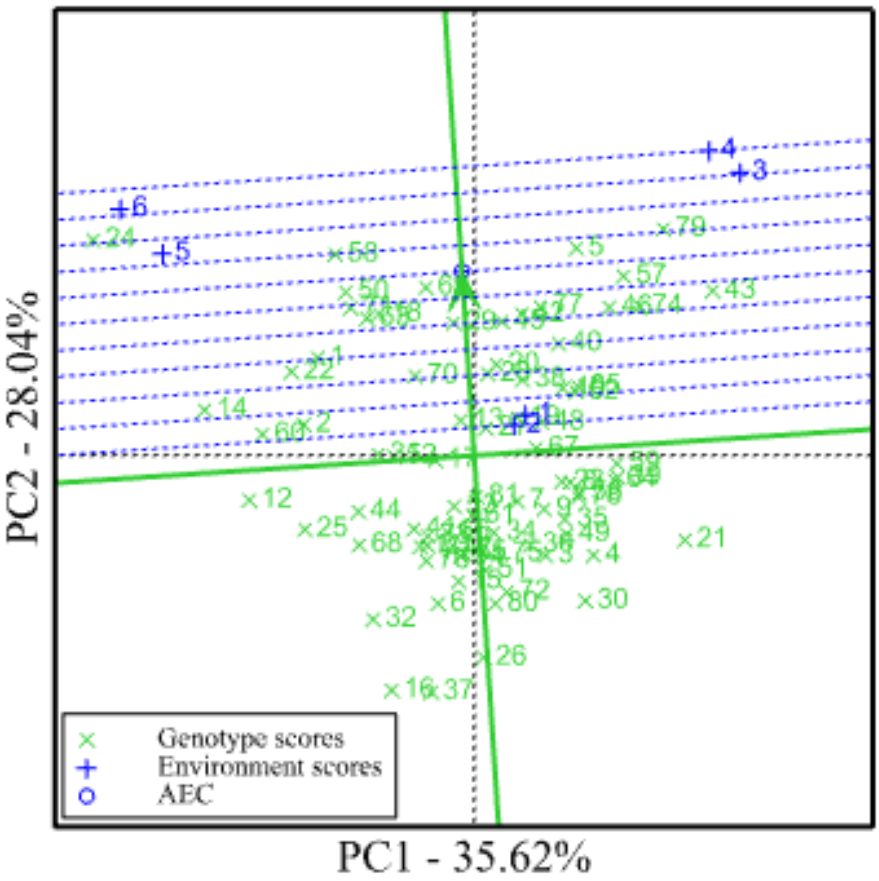

Figure 3.6. GGE biplot showing of distribution of 81 pigeonpea genotypes for mean yield and stability performance when assessed in six environments in Malawi.

E1= Bvumbwe, 2017/18, E2= Bvumbwe in 2018/19, E3=Chitedze 2017/18, E4=Chitedze 2018/19, E5= Makoka 2017/18 and E6= Makoka in 2018/19. Dotted vertical and horizontal lines indicate zero coordinate for x and y axes, respectively. The small circle on the arrowed line indicates the average environment and orthogonal dotted lines indicate the distance of genotypes and environments from the biplot origin. See genotype codes (G1-G81) in Table 3.1.

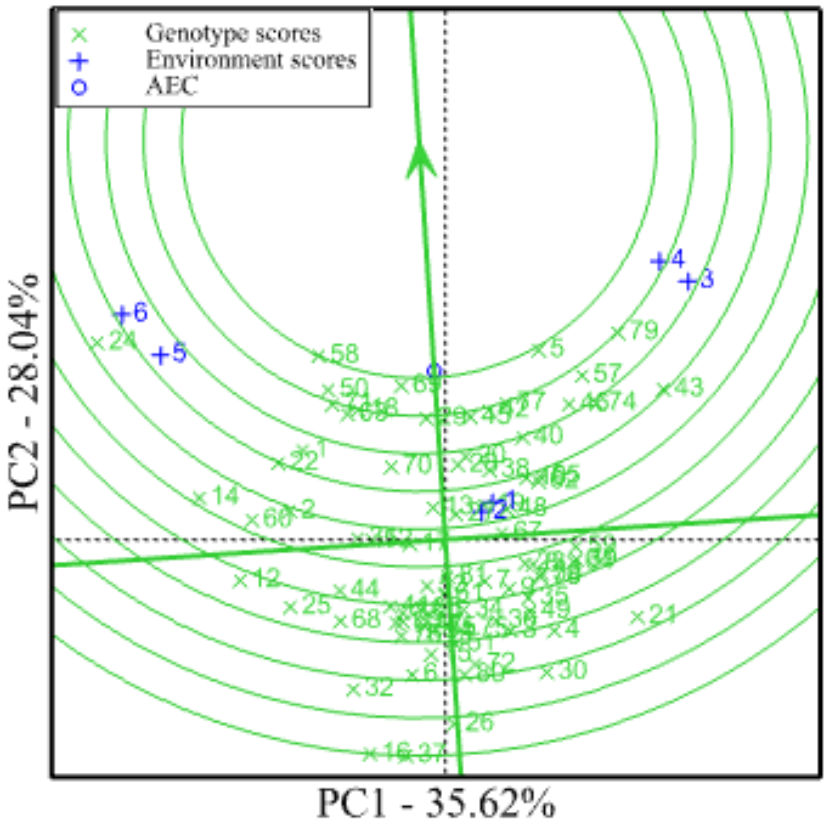

Figure 3.7. GGE biplot comparing 81 pigeonpea genotypes to the ideal genotype based on grain yield assessed in six environments in Malawi.

E1= Bvumbwe, 2017/18, E2= Bvumbwe in 2018/19, E3=Chitedze 2017/18, E4=Chitedze 2018/19, E5= Makoka 2017/18 and E6= Makoka in 2018/19. Dotted vertical and horizontal lines indicate zero coordinate for *x* and *y* axes, respectively. The small circle on the arrowed line indicates the average genotype, the arrow indicates the ideal genotype, and concentric circles indicate the distances of genotypes and environments from the ideal genotype. See genotype codes (G1-G81) in Table 3.1.

3.3.12 Delineation of Mega Environments and Genotype Adaptation

Figure 3.8 presents the polygon view that depicts the relationship between genotypes and environments. The biplot showed the outstanding genotypes in their respective environments. From the biplot, the vertex genotypes were G79, G43, G21, G16, G37, G12, G14, G26, G30 and G37, showing that they excelled in the respective environments bound within sectors in the biplot. All the six test environments were grouped into three mega environments. The first mega-environment was comprised of two environments, E1, and E2. The second mega-environment was comprised of two environments, E3 and E4 and the third mega-environment

consisted of environment E5 and E6. The highest performing genotypes in mega environment 1 were G48 and G67, while G79 and G24 performed well in mega environments 2 and 3.

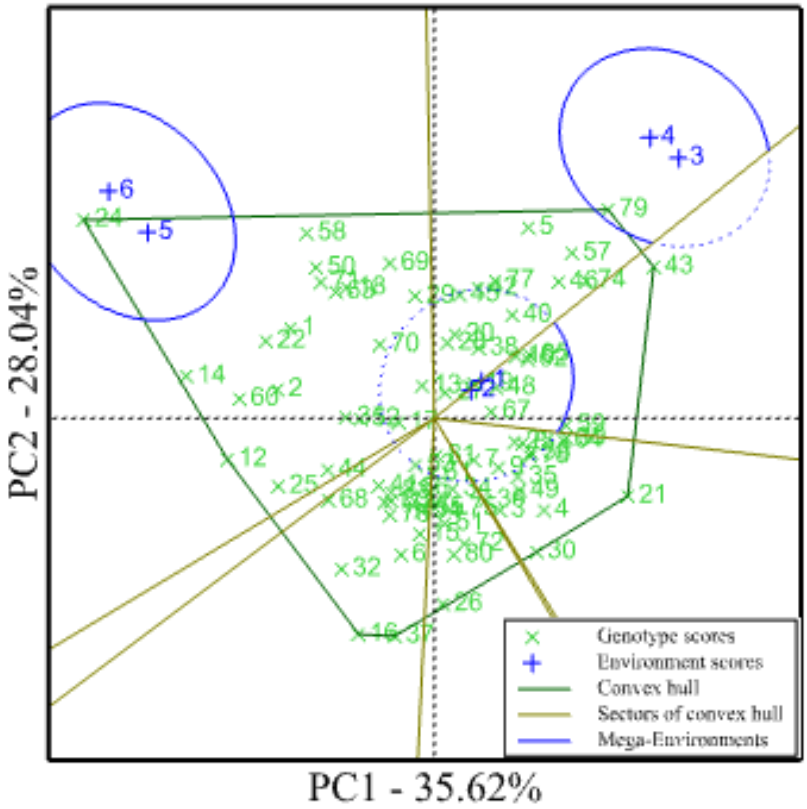

Figure 3.8. The polygon view of GGE biplot to the identification of winning genotypes and their related mega-environments.

E1= Bvumbwe, 2017/18, E2= Bvumbwe in 2018/19, E3=Chitedze 2017/18, E4=Chitedze 2018/19, E5= Makoka 2017/18 and E6= Makoka in 2018/19. Dotted vertical and horizontal lines indicate zero coordinate for *x* and *y* axes, respectively. Vertices of the polygon indicate superior genotypes in each sector. See genotype codes (G1-G81) in Table 3.1.

3.4 Discussion

3.4.1 Variation Based on Qualitative Traits

The current study evaluated 81 pigeonpea genotypes across six environments to assess the genetic diversity and yield stability, and to select complementary and unique genotypes for breeding. The genotypes exhibited wide and significant variation in qualitative traits, which indicated that the tested germplasm could harbour important genetic variation that underpins the morphological variation. Similarly, Upadhyaya et al. (2007) reported significant variation in qualitative traits among pigeonpea accessions sourced from ICRISAT's international genebank. The variation in qualitative traits such as growth habit and seed colour is important for breeding cultivars that meet farmer expectations and environmental constraints. For instance, the variation present in growth habit is important to identify genotypes with compact growth habit for intercropping to maximize farm space and productivity in moisture-limited environments. It is reported that pigeonpea cultivars of spreading growth habit are not suitable for the intercropping system in which pigeonpea is often produced, in association with cereal crops such as maize, sorghum and legumes such as groundnut (Manyasa et al. 2009). The diversity in pigeonpea seed colour helps to identify genotypes that supports farmers' preference. For instance, farmers in Malawi prefer pigeonpea varieties with cream seed colour, which they associate with good cooking quality. Similar findings were reported by Ayenan et al. (2017) who reported a predominance of cream and light greyed pigeonpea, which reflects the farmers' colour preferences in Benin. Knowledge of variability in qualitative traits among the accessions and understanding farmer preferences are important as basis for the development of direct breeding objectives and appropriate breeding strategies.

3.4.2 Genotype and Environment Variances for Quantitative Traits

The significant genetic variation exhibited in the quantitative traits highlights the genetic diversity available for exploitation during cultivar development. The genotype performances were also affected by significant genotype × environment interactions, suggesting that genotype performances were not consistent in all the environments. Genotypic variation is underpinned by differences in genetic constitution among the genotypes, which is important for crop improvement (Grausgruber et al. 2004). The environment influences phenotypic expression

through variation in factors such as temperature, humidity and soil fertility. The significant impact of the environment on phenotypic expression is known to reduce genotype-phenotype correlation (Bustos-Korts et al 2018), which complicates the identification of stable and superior genotypes. Significant genotype × environment interaction on yield and yield components of legumes such as common bean, cowpea and pigeonpea has been previously reported (Vales et al. 2012; Kimaro 2016; Gerrano et al. 2020). In the present study, the genotypes that matured early were short with low numbers of branches and pods per plant, and low grain yields when compared to the medium to late maturing genotypes that grew tall, produced more branches and pods per plant, and higher grain yields. Similarly, Rekha et al. (2013) reported that cultivars with higher numbers of primary branches, secondary branches, number of pods per plant, taller plant height had higher grain yields. The early maturity exhibited by the ICRISAT genotypes could be a result of selection for earliness at ICRISAT in Kenya, which has advanced pigeonpea breeding programs and has developed a number of elite breeding lines that have been distributed in several East and Southern African countries for evaluation trials. The TARI and DARS genotypes comprised of landraces and cultivars that are medium to late maturing, as they have not been selected for earliness. Vales et al. (2012) also reported that traditionally grown pigeonpea cultivars and landraces are represented by varieties from medium to long maturity groups (150 to 280 days), which are high yielding but are very sensitive to photoperiod.

3.4.3 Correlations Analysis Among Phenotypic Traits

The positive correlations of GYD with HSWT, indicated that these traits could be used for direct selection for GYD. The positive correlation between GYD and DTF shows that selection for earliness would compromise grain yield production in pigeonpea. Although pigeonpea is relatively drought tolerant, there is a need to develop early flowering and maturing cultivars to fit in the cropping cycles of sub-Saharan Africa, which are becoming progressively shorter due to climate change. The positive correlations exhibited by most secondary traits show that multiple trait selection would be possible. However, the weak correlations among the traits would result in inefficient selection or low genetic gains. Saroj et al. (2013) reported a strong correlations (r = 0.858) between grain yield and number of pods per plant, while Sreelakshmi et al. (2010) reported moderate to weak correlations between grain yield and days to 50% flowering (r =0.58), days to maturity (r =0.59) and plant height (r = 0.42). Conversely, Hemavathy et al. (2017) and

Narayanan et al. (2018) reported a negative association between 100 seed weight and grain yield. The significant relationship between DTF, DTM, HSWT, PH, NPP and GYD are useful when selecting for high grain yield (Upadhyaya et al. 2007). Direct selection for these traits would result in yield improvement in pigeonpea.

3.4.4 Principal Component (PC) Analysis

The PCA enabled the identification of important traits with high variability among the genotypes. In this study, DTF, DTM, HSWT, GYD, NPB and NSB were identified as the most important traits due to their high contribution on PCs (Table 3.8), which are useful selection criteria in conservation or improvement programs. Accessions that exhibit high and desirable mean performances in the target traits would be selected for improvement. Other reports indicated that trait contribution to different PCs varies with genetic diversity within the test germplasm and the number of traits evaluated (Upadhyaya et al. 2007; Saroj et al. 2013). The following quantitative traits: DTF, NPP, NPB, NSB and HSWT, are important secondary traits for indirect selection for GYD due to their favourable correlations with GYD and their high contribution on the PCs.

3.4.5 Additive Main Effect and Multiplicative Interaction (AMMI) Analysis

The significant impact of GEI on grain yield confounds selection for superior genotypes for yield improvement. Hence, it is essential to quantify the GEI to enable breeders to devise suitable and effective breeding strategies to manage and circumvent challenges presented by GEI. It is important to determine the GEI, especially for complex and highly quantitative traits such as grain yield in pigeonpea, which has largely been neglected in terms of yield improvement. Genotype by environment interaction effects arise when genotype performances are not consistent across different environments. The differential response of genotypes indicate that each environment exerted significant and different selection pressure emanating from edaphic or climatic conditions in the particular environment. This suggest that selection of genotypes based on the overall mean would be misleading since genotypes that exhibited specific adaptation to a particular environment may be discarded. Therefore, selection should also consider genotype performance in individual environments. The present findings are in agreement with Aina et al.

(2009) and Kamau et al. (2013) who both found significant G × E interaction effects on grain yield among pigeonpea genotypes. Conversely, Singh et al. (2018) suggested that it is impractical to select varieties for specific adaptation, based on the significance of G × E effects, and proposed that selection should target genotypes with broader adaptation. In this study, both specific and broad adaptation were pursued to increase chances of identifying suitable parental genotypes for pigeonpea improvement. Partitioning the GEI revealed that GEI accounted for 95.3% of the variation in grain yield, and that out of three principal component axes, three had significant interaction effects (Table 3.9). However, IPCA1 and IPCA2 explained 75% of the total GEI, suggesting that the interactions among the 81 genotypes and the six environments were best predicted by the first two principal components.

An environmental variance of 33.5% indicates that this variable was an important determinant of pigeonpea yields in the present study. The three test locations have different soil characteristics and experienced different weather conditions during the two different cropping seasons, leading to the large environmental variance, accounted for in the AMMI analysis. Use of homogenous environments would exert similar selection pressures during genotype evaluation, and would result in low environmental variance in the AMMI analysis. The genotype component accounted for 16.4% of the variation, indicating that genotypic expression was masked by environmental factors. The masking of genotypic expression will lead to inefficient selection over a large number of environments. It would be prudent to integrate other techniques such as molecular techniques for marker-assisted selection to circumvent the environmental influence to improve selection in this germplasm collection.

3.4.5.1 Genotype Stability for Grain Yield Response

The present study revealed that Bvumbwe site (E1 and E2) (Figure 3.3) was the largest contributor to the stability of the genotypes due to its ability to provide suitable conditions for the genotypes to express their genetic potential compared to the other environments. The stability provided by E1 and E2 could be due to the combination of good rainfall and moderate temperature experienced in Bvumbwe (Table 3.2). The stable genotypes had static stability, which allowed them to maintain relative similar performance across the environments, although the performance was not necessarily high. Genotypes such as G51, G26 and G27 may be

recommended for a number of environments because they are likely to attain reasonable economic yield and have a lower risk of failure under unfavourable conditions due to their static stability. The high yielding and stable genotypes included G45 and G29 were medium to late maturing landraces and had extended periods of dry matter accumulation, which means they could be recommended for production in environments that experience extended rainy seasons to support their genetic potential of high yield accumulation. In addition, the landraces have under gone many generations of selection for resistance to pests and diseases by local farmers, making them adapted to the local conditions. Adaptation is essential to withstand environmental stresses and allows the genotypes to exhibit phenotypic plasticity in response to environmental stimuli that curtail the growth of non-adapted genotypes. The AMMI 1 biplot further revealed that Bvumbwe and Makoka were the two best environments. This could be attributed to the effects of the off-season rainfall, locally known as 'Chiperoni', which usually occurs in southern Malawi where the Bvumbwe and Makoka sites are located. The off-season rains prolong the growing season, providing the crop with essential moisture during the late season when most other environments experience terminal droughts. The AMMI 2 model showed differential response of genotypes to different environmental conditions, which confirms the importance of multi-environment trials in plant breeding to identify suitable genotypes for selection.

3.4.6 GGE Biplot Analysis

3.4.5.1 Genotype Stability for Grain Yield Response

GGE biplots are useful tools used to explore multi-environment trial data. The biplots allow the visualization of relationships among the test genotypes and environments, and to investigate the pattern of GEI since they help to identify outstanding genotypes that are adapted to a particular environment, and to group environments into mega environments (Yan et al. 2000). According to (Yan and Tinker 2006), the environment whose vector has the smallest angle with the AEA represents the most representative environment. The most informative and discriminating environment is the one farthest from the origin of the biplot. The environmental-focused scaling GGE biplot revealed that E1 and E2 were the most discriminating environments among the six test environments, indicating that these sites should be used for future pigeonpea evaluations (Figure 3.5). The genotype-focused scaling GGE biplot revealed that the genotypes G36, G26, G27, G72, G29 and G51 were the most stable genotypes across the test environments, although

some did not necessarily attain high yield. Among the stable genotypes, genotype G51 can be recommended for widespread adoption since it exhibited high yield potential, coupled with high stability (Figure 3.6) Genotype G51 is a landrace commonly grown in Bvumbwe and Makoka, where local farmers call it as “Rozikhuthula” or “Zalalende”, which translate to “high yielding”, showing that it is popular among local farmers. The best performing genotypes in terms of stability and adaptability can be selected as parents in yield improvement programs to develop new cultivars.

3.4.5.2 Delineation of Mega Environments and Genotype Adaptation

The use of the polygon view biplot “which-won-where” is a key component of the GGE, which helps to visualize the interaction patterns between genotypes and environments, to show the presence of crossover GEI, mega-environment differentiation and specific adaptation (Yan and Tinker 2006). It further helps to identify the representativeness of environments and their discriminating ability, which enables breeders to detect locations that can be discarded from further evaluation trials without losing important information about genotypes. Grouping of the environments into mega-environments also helps to have fewer test environments that reduce the cost of evaluation and increase breeding efficiency. In the present study, the test environments were grouped into three mega-environments. However, the Bvumbwe and Chitedze sites were correlated and similar. These two sites can substitute each other in future evaluations of pigeonpea genotypes to reduce costs and to increase breeding efficiency without loss of information. On the other hand, Makoka can be used as an additional site for future pigeonpea evaluations in Malawi because it provides extra discriminatory capacity to complement the other sites. Genotypes MWPLR 25 (G57), MWPLR 16 (G43), ICEAP 86012 (G79) and MWPLR 22 (G5) can be recommended for production in both Bvumbwe and Chitedze, while ICEAP 01172/1(G24) would be targeted for Makoka. However, there is a need to subject these genotypes to molecular analysis and to document the phenotypic traits that are preferred by the local farmers to design marker-assisted and demand-led breeding strategies to develop new cultivars and to enhance potential adoption of the new cultivars among local farmers.

3.5 Conclusions

The current study examined 81 pigeonpea genotypes for their phenotypic diversity and yield stability. The genotypes exhibited a wide genetic variation ($p<0.001$) in qualitative traits such as growth habit, flower main colour, flower streak pattern, pod colour and seed traits. The combined analysis revealed that all quantitative traits were significantly affected by genotype × environment interaction effects, except for the number of seeds per pod, suggesting that both the genotype and environment had an influence on the traits measured. Genotypes MWPLR 14, ICEAP 01170, ICEAP 871091 and ICEAP 01285 were selected for their early maturity, varying from 125 to 137 days. The genotypes Kachangu, MWPLR 16, TZA 5582, No. 40 and MWPLR 14 had the highest number of pods per plant and high grain yield. The positive and significant association between days to flower, number of pods per plant, 100 seed weight and grain yield suggest that these traits can be used in selecting high yielding pigeonpea genotypes. The presence of crossover GEI effects on pigeonpea grain yield indicated the need to breed for specific adaptation. The environments were delineated into three mega-environments based on site × season interaction. The stability analysis in AMMI and GGE indicated that the genotypes MWPLR 14 (G51), MWPLR 24 (G26) and ICEAP 01155 (G27) were stable across environments, while MWPLR 14, TZA 5582 and MWPLR 4 were the highest yielding genotypes across environments. The high yielding genotypes were selected as parental lines for breeding to introgress their high yield and stability genes into popular varieties in future yield improvement programs. In the future, there is a need to incorporate molecular techniques to guide selection and to circumvent the strong environmental influence.

3.6 References

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CHAPTER 4. GENETIC DIVERSITY AND POPULATION STRUCTURE ANALYSES OF PIGEONPEA GENOTYPES USING MORPHOLOGICAL TRAITS AND SNP MARKERS

Abstract

Knowledge of population structure and genetic interrelationships among pigeonpea germplasm collections is fundamental to select breeding parents with unique genetic constitution. The objectives of this study were to assess the genetic diversity and population structure present among 81 pigeonpea genotypes using 24 morphological traits and 4,122 single nucleotide polymorphism (SNP) markers. Genotype × environment interaction effects were significant ($P<0.001$) for grain yield (GYD), days to 50% flowering (DTF), days to 75% maturity (DTM), plant height (PH), number of primary branches per plant (NPB), number of pods per plant (NPP), number of racemes per plant (NRP), 100 seed weight (HSWT), and number of secondary branches per plant. The principal component analysis identified eight components that explained 67.57% of the total phenotypic variation. Traits including DTF, DTM, growth habit (GH), second flower colour (FSC), pod colour, seed shape (SSH), HSWT, and GYD were identified as the most important for discriminating among the test genotypes. The phenotypic diversity assessment using morphological attributes grouped the genotypes into three distinct clusters. The mean gene diversity and polymorphic information content were 0.14 and 0.11, respectively, suggesting moderate genetic differentiation among the genotypes. The genotypes were delineated into three heterotic groups based on population structure and the joint analysis based on the phenotypic and genotypic data, suggesting the possibility of creating unique breeding populations through targeted crosses of parents from divergent heterotic groups. To broaden the genetic base of the pigeonpea for selection, divergent genotypes such as MWPRL 14, TZA 5582, MWPLR 4, MWPLR 16, Sauma, Kachangu are recommended.

Key words: divergence, genetic differentiation, heterotic groups, genetic analysis, Malawi, pigeonpea, phenotypic traits

4.1 Introduction

Pigeonpea is protein-rich legume crop cultivated in more than 25 tropical and sub-tropical countries either as a sole crop or intercropped with cereals or other legumes. Pigeonpea is also a major income source for many small scale farmers in Africa and Asia (Mergeai *et al.* 2001). Pigeonpea has high biomass productivity, making it suitable as a fodder crop (Odeny 2007). Like other legume crops, pigeonpea forms symbiotic associations with nitrogen fixing bacteria and can potentially fix between 69 to 100 kg ha^{-1} atmospheric nitrogen (N) (Rao *et al.* 1987) with a net contribution of 2 to 28 kg N ha^{-1} depending on genotype and environmental factors (Myaka *et al.* 2006, Egbe 2007). Furthermore, its roots help release soil-bound phosphorus to make it available for plant growth (Noriharu *et al.* (1990). Despite its diverse economic importance, pigeonpea is classified as an underutilized and orphan crop species. Consequently, the production and productivity of pigeonpea varieties are still too low to attract interest from commercial and large-scale farming enterprises. The neglect of orphan crops such as pigeonpea by crop improvement research programs compared to other commodity crops such as maize, wheat and rice has contributed to a lack of improved and high yielding pigeonpea cultivars in sub-Saharan Africa (SSA) that meet farmer and market preferences in SSA. Nonetheless, the crop has substantial market potential if the quantity and quality of grain were to be enhanced (Odeny 2007). Sustainable promotion and advancement of pigeonpea will require developing and deploying improved cultivars acceptable to farmers and the entire value-chain.

The development of new cultivars requires an understanding of the existing diversity to inform breeding programs and germplasm management strategies. Limited information is available on the magnitude of genetic diversity within the cultivated pigeonpea gene pool (Saxena and Sawargaonkar 2014). Knowledge of genetic diversity facilitates identification of heterotic groups and the best parents for breeding. Morphological, biochemical and molecular markers have been used in genetic diversity assessments in crop improvement. Molecular markers are robust compared to morphological and biochemical markers in genetic diversity analysis. Molecular markers are not affected by environmental conditions that can confound genotype selection efforts (Zavinon *et al.* 2018). Several molecular markers have been used in genetic diversity analysis of pigeonpea, including markers based on restriction fragment length polymorphisms (RFLP) (Sivaramakrishnan *et al.* 2002), amplified fragment length polymorphisms (AFLP) (Pati *et al.* 2014), random amplified polymorphic DNA (RAPD)

(Malviya and Yadav 2010), simple sequence repeats (SSR/microsatellites) (Sarkar *et al.* 2017) and single nucleotide polymorphisms (SNP) (Saxena *et al.* 2014). SNP markers derived from next generation sequencing have been widely used because they provide high-density and whole-genome profiles at a relatively low cost (Jaccoud *et al.* 2001). Thousands of SNPs detected across the genome are useful for characterizing germplasm and maker-trait association mapping. Yang *et al.* (2006) developed a pilot diversity array technology (DArT) library for pigeonpea comprising 5,376 SNPs to analyse 96 genotypes representing 20 species of *C. cajan*. The authors reported a narrow range of genetic diversity among the tested genotypes. More than 15,000 SNPs were discovered recently across the pigeonpea genome (Varshney 2016).

The recently compiled DArT library on pigeonpea genome provide opportunities for gene discovery and for developing strategies for marker-assisted selection to accelerate breeding progress in pigeonpea. Pigeonpea breeding in Malawi is lagging behind and is mainly focussed on conventional breeding methods. Conventional breeding is slow to respond to the rapidly changing environment. This will result in delayed cultivar development and release. In order to expedite breeding progress and potentially achieve higher genetic gains, there is a need to integrate conventional and molecular breeding approaches. Yohane *et al.* (2020) evaluated the phenotypic divergence present in a panel of pigeonpea with phenotypic traits and reported the existence of significant genetic variation. The authors indicated that selection efforts were confounded by the high environmental variance. It was imperative to complement phenotypic data with data derived from molecular markers to reduce the impact of environmental variance and improve selection efficiency for cultivar development. Therefore, the objectives of this study were to assess the genetic diversity and population structure among 81 pigeonpea genotypes using 24 morphological traits and 4,122 single nucleotide polymorphism markers. The results will assist in parental selection to initiate pigeonpea pre-breeding in Malawi.

4.2 Materials and Methods

4.2.1 Plant Materials and Phenotyping

The study used a population of 81 pigeonpea genotypes presented in Chapter 3 (Table 3.1). The 81 pigeonpea accessions were evaluated at three sites in Malawi, namely at the Bvumbwe, Chitedze, and Makoka Research Stations, in two cropping seasons (2017/18 and

2018/19). The geographic location, altitude, weather, and soil characteristics of the study locations are presented in Chapter 3 Table 3.2. Treatments were laid out using a 9 × 9 alpha-lattice design at each testing location. Each genotype was planted on a plot consisting of two rows. Each row was 5m in length spaced at 0.90 m apart, giving a plot size of 4.5 m^2. Seeds were planted at 0.75 m apart within a row. Three seeds were planted per planting station and thinned to one plant after two weeks emergence. The phenotypic data collected included qualitative and quantitative attributes which are described using the IBPGR (1993) as presented in chapter 3 (Table 3.3).

4.2.2 Phenotypic Data Analysis

Quantitative data was subjected to analysis of variance (ANOVA) using Genstat 18th edition. The means from the genotype × environment analysis were used for principal component analysis (PCA) using the "factorMiner" and "missMDA" procedures in the R statistical package (Team 2014). Phenotypic clusters based on the dissimilarity matrix were generated using the Gower method implemented in the "cluster" and "graphics" procedures in the R statistical package. The final hierarchical cluster was constructed using the ward D2 method in "cluster" in R package (Maechler *et al.* 2019). The correlations among quantitative and qualitative phenotypic traits were determined using the Spearman's rank correlation in SPSS version 25 (Wagner III 2019).

4.2.3 DNA Extraction and DArT Sequencing

Ten seeds of each pigeonpea genotype were planted in plastic pots for three weeks. Fresh leaf samples were collected from three-week old seedlings for each genotype and stored in a deep freezer at – 80^{o}C. Deoxyribonucleic acid (DNA) extraction was performed following the Diversity Arrays Technology Sequencing (DArTseq) protocol (https://www.diversityarrays.com/files/DArT). Fifty milligrams of total genomic DNA were extracted from the well developed trifoliate leaves using the NucleoSpin Plant II kit (Macherrey-Nagel, Duren, Germany) with the Lysis Buffer I (based on the cetyl trimethylammonium bromide (CTAB) method). The DNA quality and quantity of each sample were determined on 2% agarose gel followed by quantification using a NanoDrop 2000 Spectrophotometer (ND-2000 v3.5 NanoDrop, Technologies, Inc). The DNA samples were sent to the Biosciences eastern and central Africa International Livestock Research Institution (BecA-ILRI-hub in Kenya (https://hub.africabiosciences.org/) for genotyping.

4.2.4 SNP Calling and Filtering

For quality control, DArTseq SNP delivered markers were filtered to remove bad SNPs and genotypes using the "impute" package in R software (Hastie *et al.* 2017). Markers and genotypes with > 20% missing data, 20% of heterozygosity, and the minor allele frequency less than 0.05 were removed, resulting in 4,122 informative SNP markers and 81 genotypes that were used for analysis.

4.2.5 Analysis of Genetic Diversity Parameters

The gene diversity, minor allele frequency (MAF), polymorphic information content (PIC), and heterozygosity (Ho) were calculated using the "diveRsity" procedure in R software (Keenan *et al.* 2013). The analysis of molecular variance (AMOVA) was conducted using the GenAlex version 6.5 (Peakall and Smouse 2006).

4.2.6 Population Structure, and Cluster Analysis

The population structure of 81 genotypes was determined using the admixture model-based clustering method in STRUCTURE Harvester (Earl 2012). The burn-in period and Markov Chain Monte Carlo (MCMC) iterations were set at 10,000 to derive the population structure based on 4,124 SNP markers distributed across the pigeonpea genome. The K-value was set between 1 and 10 to generate the number of sub-populations in the accessions. The best K-value with the highest likelihood for estimating suitable population size for the data set was determined using the Evanno procedure (Evanno *et al.* 2005). The accessions with a membership probability ≤ 0.70 of a sub-population were assigned to an admixture group, and those ≥ 0.70 were assigned to a distinct population. The dendrograms were generated using the genetic dissimilarity matrix using the "phylogenetics" and "evolution" procedures in R (Paradis *et al.* 2004).

4.2.7 Joint Analysis of Phenotypic and Molecular Data

Genetic groups were defined using a combination of the phenotypic and genotypic dissimilarity matrices. The joint matrix was generated by the summation of the genotypic and phenotypic dissimilarity matrices. The phenotypic dissimilarity matrix was generated using Gower's distance matrix, while genotypic dissimilarity matrix was based on Jaccard's

coefficients. The clusters generated from the phenotypic and genotypic sets were compared using the "viridis" procedure in R (Garnier *et al.* 2018) and the similarity of the two dendrograms was assessed using tanglegram function developed by the "dendextend" R package (Galili 2015).

4.3 Results

4.3.1 Diversity and Differentiation Based on Phenotypic Traits

The quantitative agronomic data were pooled across sites after testing for homogeneity of variance and normality. The genotype × environment interaction effects were significant ($P<0.001$) for grain yield , days to 50% flowering, days to maturity, plant height, number of primary branches , number of pods per plant , number of racemes per plant , 100 seed weight , and number of secondary branches (Table 4.1). The genotype and environment had significant ($P<0.001$) effects on all the assessed traits except the number of seeds per pod.

The principal component analysis was performed to identify the most discriminative variables among the pigeonpea genotypes. The first eight principal components cumulatively explained 67.57% of the total phenotypic variation (Table 4.2). The first principal component accounted for 16.58% of the total phenotypic variation. Traits including DTF, DTM, FSC, PH, and FP had positive loadings on the first principal component (PC1). In contrast, PC, SEC, SCP and SC had negative loading on PC1. The second principal component (PC2) accounted for 28.55% of the total variation, with FSP and FP being the highest positive contributors. Conversely, traits including HSWT (-0.57), LS (-0.55), GYD (-0.55) and NPB (-0.43) exhibited negative correlations with PC2. Traits such as NPP, SMC, HSWT, FSC, and SCP contributed much to the observed variation on the third principal component (PC3), with PC loadings ranging from 0.40 to 0.52. However, FSC had a negative (-0.42) PC loading. The fourth principal component accounted for 44.80% variation contributed by two traits, GH and FMC. PC4, PC5 and P6 explained 57.83, 62.92, and 67.92% to the total variation, in that order.

Correlation analysis revealed a positive correlation between GYD and quantitative traits, including HSWT, PH, NPP, NSP, DTF, DTM, PL, and qualitative traits such as LS, SMC, SCP, and SSH. The GYD also exhibited negative correlations with FMC and SC (Appendix 3).

The phenotypic diversity assessment using morphological attributes grouped the genotypes into three distinct clusters (Figure 4.1). Cluster 2 recorded the highest number (51) of genotypes, followed by Cluster 1 (27) and Cluster 3 (3). The genotypes in Cluster 1 included two landraces from Malawi; MWPLR 14 (G41) and MWPLR 24 (G26), and one collection from Tanzania, TZA 197 (G76), both with medium maturity. The genotypes in Clusters 1 and 2 were a mixture of landraces, breeding lines, and cultivars. However, genotypes in Cluster 2 were mostly medium to late maturing, which included Babati (G49), Hombolo (G78), Sauma (G81), TZA 5557 (G50), ICEAP 0673/1 (G1), MZ2/9 (G23), among others. Cluster 1 had most of the early maturing genotypes such as ICEAP 87105 (G42), ICEAP 01170 (G6), ICEAP 87091 (G58), ICEAP 01150 (G22), ICEAP 00612 (G64), ICEAP 01172/1 (G24), ICEAP 01146/01 (G18).

Table 4.1. Mean squares for grain yield and yield components computed from 81 pigeonpea genotypes evaluated in six environments in Malawi.

Source of variation	DF	DTF	DTM	PH	NPB	NSB	NRP	NPP	NSP	GYD	HSWT
Location (L)	2	9024.2 ***	8735.4 ***	54965.0 ***	114.4 ***	93.7 *	226.9 ***	3236 **	22.5 ***	5968860.0 ***	1008.1 ***
Replication (L)	1	701.9 ns	289.0 ns	118.0 ns	1.2 ns	105.4 *	14646.0 ns	9810 *	0.45 ns	1663232.0 *	9.5 ns
Block (Rep)	8	3168.5 ***	5703.4 ***	7710.9 ns	52.9 *	93.7 *	9099 .0*	6433.6 **	2.4 *	16534356.5 ***	72.2 **
Genotype (G)	80	879.2 ***	1234.9 ***	2137.0 ***	12.5 *	30.9 *	5004.9 *	1990.3 *	0.8 ns	351745.3 *	16.8 *
Season (S)	1	3370.5 **	2945.3 *	447.0 ns	409.6 ***	650.1 ***	2023492.0 ***	437.5 ***	31.5 ***	30308789.0 ***	50.2 *
G × L	160	243.0 *	361.9 *	1106.0 *	18.0 *	35.6 *	6150.9 *	1916.1 *	0.9 ns	360816.9 *	20.7 **
G × S	80	3610.3 ns	606.9 ns	1198.0 ns	17.9 *	34.7 *	4642.7 ns	1060.3 *	0.9 ns	400468.2 *	14.9 ns
G × L × S	160	330.6 ns	484.9 ns	744.0 ns	15.2*	34.5 *	6110.9 ns	1502.8 *	0.7 ns	919105.3 ns	16.2 ns
Residual	469	345.4	585.8	1243.1	14.5	11.8	5822.9	5667.2	0.8	313554.0	15.4

DF= degrees of freedom, Rep= replication, DTF= days to 50% flowering, DTM= days to 75% maturity, PH= plant height, NPB= number of primary branches, NSB= number of secondary branches per plant, NRP = number of racemes per plant, NPP= number of pods per plant, NSP= number of seeds per pod, GYD= grain yield, HSWT= 100 seed weight, *, ** and ***= significance at 0.05, 0.01 and 0.001 probability levels, respectively

Table 4.2. Principal component analysis (PCA) based on qualitative and quantitative traits among 81 pigeonpea genotypes assessed in six environments in Malawi

Variables	PC1	PC2	PC3	PC4	PC5	PC6	PC7	PC8
Flower main colour	-0.39	0.27	0.22	**0.55**	-0.01	0.27	-0.30	-0.07
Flowering pattern	0.45	0.46	0.01	0.15	-0.29	-0.27	0.24	0.00
Flower second colour	0.57	0.30	-0.42	0.34	0.09	0.13	-0.06	0.25
Flower streak pattern	0.31	0.59	-0.22	0.26	0.09	0.39	0.04	0.31
Growth habit	0.24	0.02	0.20	0.62	-0.29	0.11	0.42	0.06
Leaf hairiness	-0.02	0.03	-0.21	0.31	0.44	0.31	0.22	-0.42
Leaf shape	-0.12	-0.55	-0.12	-0.23	-0.11	0.46	0.16	0.29
Pod colour	-0.63	0.20	0.28	-0.18	0.06	0.36	0.02	0.02
Stem colour	-0.41	0.16	0.20	0.13	-0.56	-0.19	-0.15	0.30
Seed eye colour	-0.53	0.36	0.31	0.08	0.05	0.15	0.09	-0.11
Seed main colour	-0.06	0.24	0.42	0.30	0.10	-0.38	-0.26	-0.07
Seed shape	0.25	0.13	0.22	-0.03	0.58	-0.12	-0.34	0.40
Seed colour pattern	-0.48	0.33	0.40	-0.14	0.09	0.12	0.28	-0.07
Days to flowering	0.72	0.15	0.25	-0.28	-0.25	0.27	-0.24	-0.20
Days to maturity	0.69	0.22	0.26	-0.25	-0.25	0.23	-0.27	-0.25
Plant height	0.48	0.35	0.12	-0.19	0.00	-0.03	0.24	-0.21
Number of primary branches	0.25	-0.43	0.15	0.14	-0.05	-0.30	0.29	0.03
Number of pods/plant	0.37	-0.18	0.52	0.07	0.26	-0.05	0.11	-0.04
Number of racemes/plant	0.36	-0.27	0.36	0.05	0.43	-0.07	0.13	0.10
Pod length	0.13	0.35	0.37	-0.39	0.02	0.11	0.31	0.41
100 seed weight	0.07	-0.57	0.42	0.29	-0.17	0.17	-0.11	0.05
Grain yield	0.31	-0.55	0.21	0.19	-0.08	0.35	-0.11	0.05
Eigenvalue	3.65	2.63	1.90	1.68	1.48	1.39	1.12	1.02
Variance (%)	16.58	11.97	8.62	7.63	6.72	6.31	5.09	4.65
Cumulative variance (%)	16.58	28.55	37.17	44.80	51.52	57.83	62.92	67.57

PC= principal component

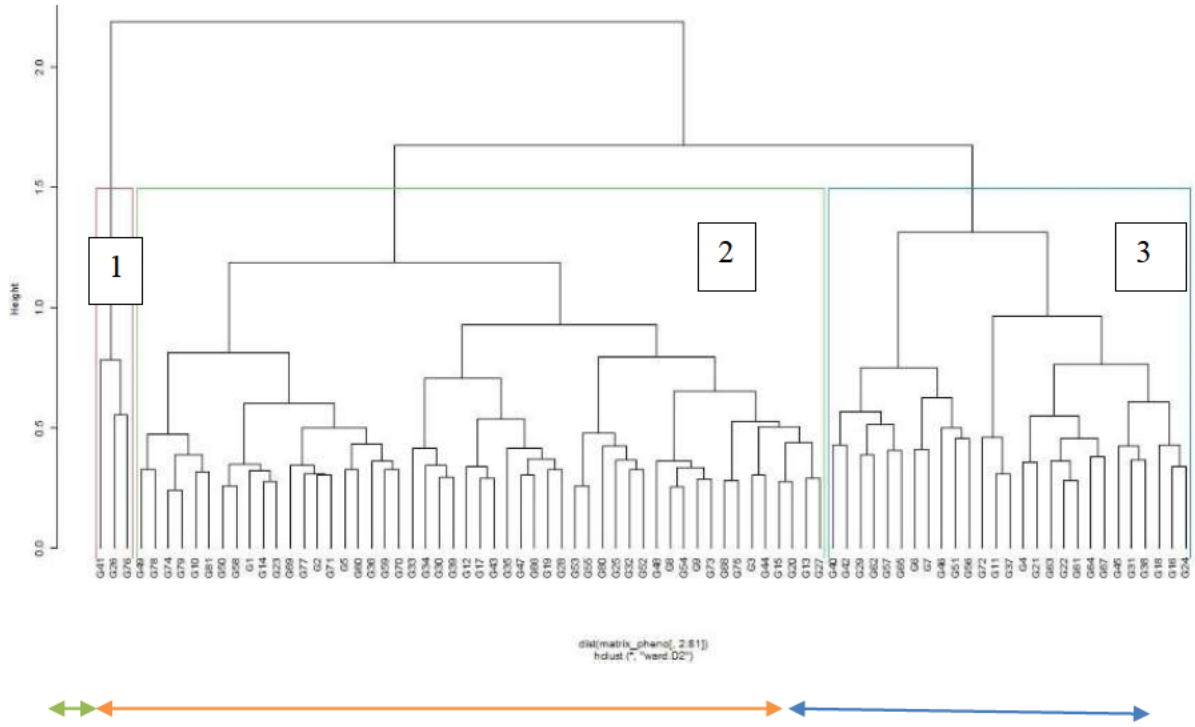

Figure 4.1. Hierarchical cluster dendrogram showing genetic similarity matrix among 81 pigeonpea genotypes evaluated in six environments in Malawi based on phenotypic traits. Numbers 1, 2 and 3 denote clusters, See Table 3.1, Chapter 3 for code of genotypes.

4.3.2 Genetic Diversity and Population Structure Based on SNP Markers

4.3.2.1 Population Genetic Parameters

Heterozygosity values varied from 0.21 to 0.23, with a mean of 0.22 (Table 4.3). Gene diversity ranged from 0.00 to 0.50, with a mean of 0.14. The SNP markers were moderately polymorphic, with PIC values ranging from 0.00 to 0.38, with a mean value of 0.11. The markers included the rare variants with a minimum MAF of 0.00 and common variants with a maximum MAF of 0.50 and a mean of 0.12. The inbreeding coefficient averaged -0.56, showing a high level of inbreeding.

4.3.2.2 Population Structure Analysis

Based on 4122 SNPs, population structure analysis revealed three distinct sub-populations among the 81 accessions (Figures 4.2A and 4.2B) based on the highest ΔK value at K = 3

following the Evanno method. Sub-population 1 consisted of 15% of genotypes and comprised breeding lines. Sub-population 2 had 5% of the genotypes, mainly cultivars, while sub-population 3 consisted of mainly landraces. The genetic differentiation among the populations ranged from -0.011 to 0.002 (Table 4.3). The highest (0.002) genetic differentiation (Fst) was observed between sub-population 1 (breeding lines) and sub-population 2 (cultivars), while the lowest (-0.011) Fst value was observed between sub-population 2 (cultivars) and sub-population 3 (landraces) (Table 4.4). The analysis of molecular variance showed significant variation ($P < 0.01$) within the populations, while non-significant variation were found among the populations (Table 4.5). The within population variation accounted for 97.3% of the total variation exhibited by the presently assessed pigeonpea genotypes.

4.3.2.3 Cluster Analysis Based on Molecular Data

Results based on the population structure analysis were confirmed by the phylogenetic tree, which resolved the 81 genotypes into three clusters (Figure 4.3). The clustering was independent of geographical sources of collection. Genotypes in cluster 1 were early maturing, while clusters 2 and 3 comprised medium and late maturing accessions, respectively.

Table 4.3. Diversity parameters of 81 pigeonpea genotypes based on 4122 SNP markers

Parameter	GD	PIC	MAF	Ho	F
Minimum	0.00	0.00	0.00	0.21	-0.65
Maximum	0.50	0.38	0.50	0.23	-0.49
Mean	0.14	0.11	0.12	0.22	-0.56

GD= genetic diversity, PIC= polymorphic information content, MAF= minor allele frequency, Ho= observed heterozygosity, F= inbreeding coefficient

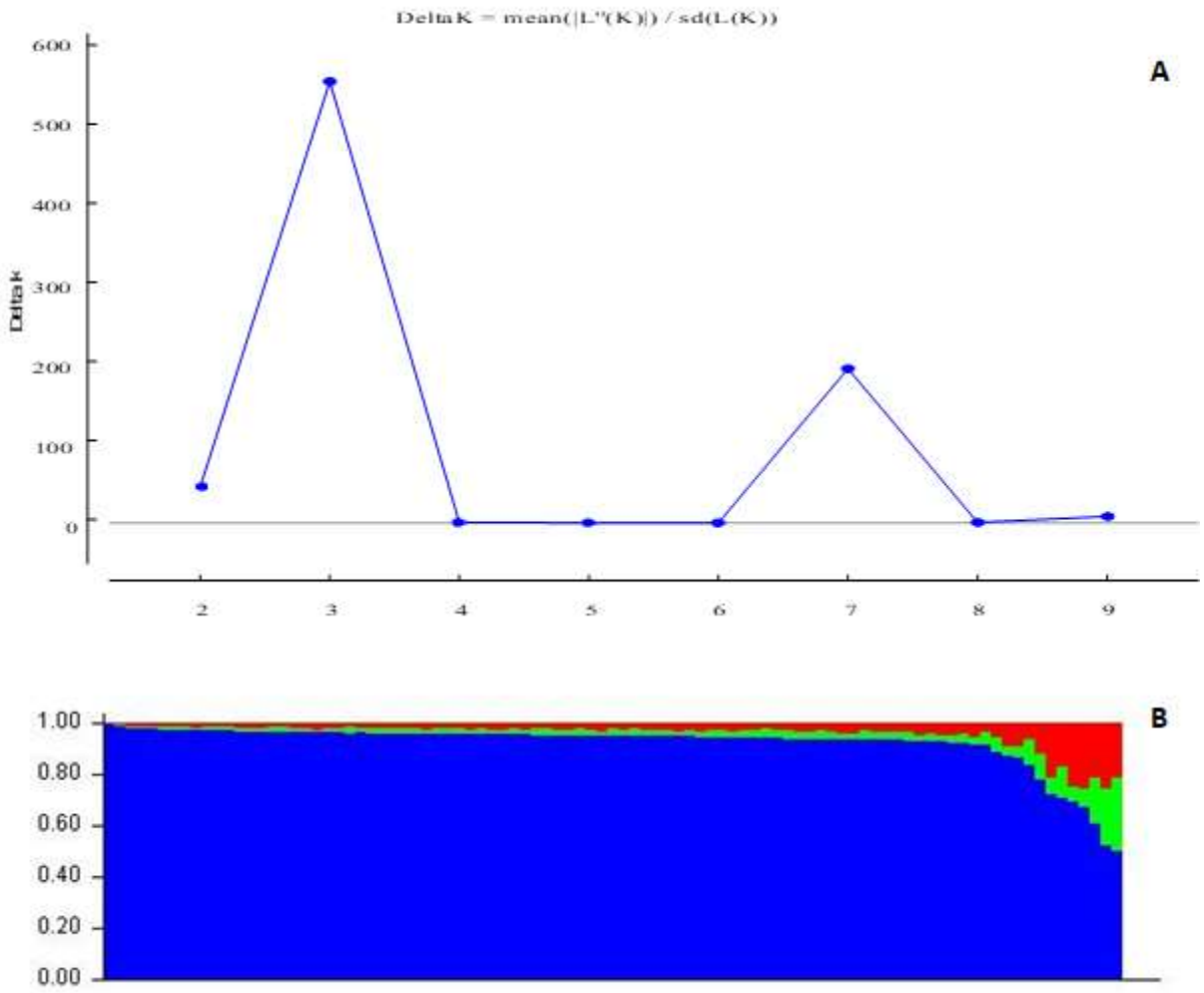

Figure 4.2. Sub-population inference among the 81 pigeonpea genotypes based on 4122 SNP markers showing (A) likelihood and delta K values for the number of assumed clusters (B) population structure at K = 3

Table 4.4. Population genetic differentiation/distance (Fst) for 81 pigeonpea genotypes

Population	**G1**	**G2**	**G3**
G1	-		
G2	0.002	-	
G3	-0.011	-0.014	-

G1= breeding lines, G2= cultivars, G3= breeding landraces

Table 4.5. Analysis of molecular variance based on three sub-populations detected among 81 pigeonpea genotypes

Source	DF	SS	MS	Estimated variance	Variance (%)	P-value
Among populations	2	229.25	114.63	0.34	2.70	0.300
Within population	78	8377.19	107.40	107.40	97.30	0.003
Total	**80**	**8606.44**		**107.74**	**100**	

DF = degrees of freedom, SS = sum of squares, MS = mean square, P-value = significance level

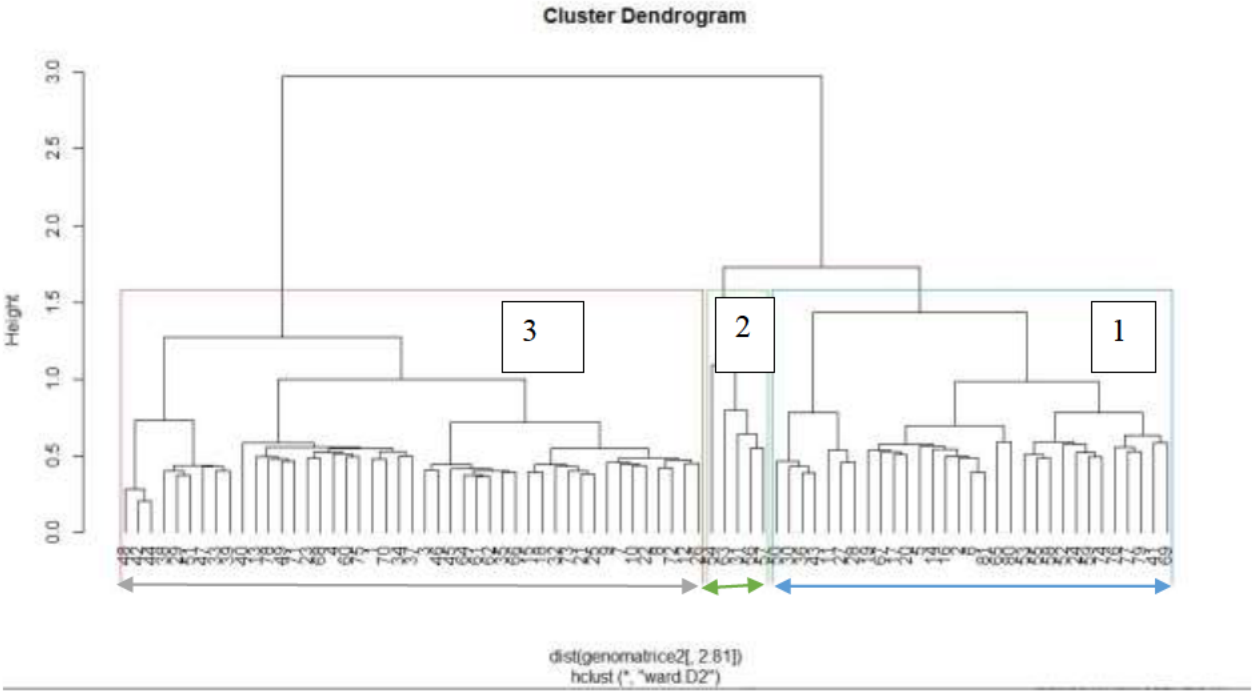

Figure 4.3. Hierarchical cluster dendrogram showing the genetic relationships among 81 pigeonpea accessions using 4122 SNP markers.

Numbers 1, 2, 3 denote the three clusters, See Table 3.1, Chapter 3 for code of genotypes

4.3.2.4 Combined Analysis using Phenotypic Traits and SNPs Markers

The phylogenetic tree generated from the phenotypic data was compared to the cluster generated from the SNP data (Figure 4.4). The results show that only 13.5% of the accessions maintained the same position across the hierarchical clusters. There was a clear indication of the grouping patterns and membership delineated by the phenotypic and genotypic datasets. A total of 37

accessions representing 45.7% of the accessions maintained their groups across the phenotypic and genotypic hierarchical clusters.

Genetic diversity assessment using the combined phenotypic and molecular data clustered the accessions into three groups (Figure 4.5). Clusters 1, 2 and 3 composed of 34, 7 and 40 accessions, in that order. The clusters represented a mixture of landraces, breeding lines, and cultivars.

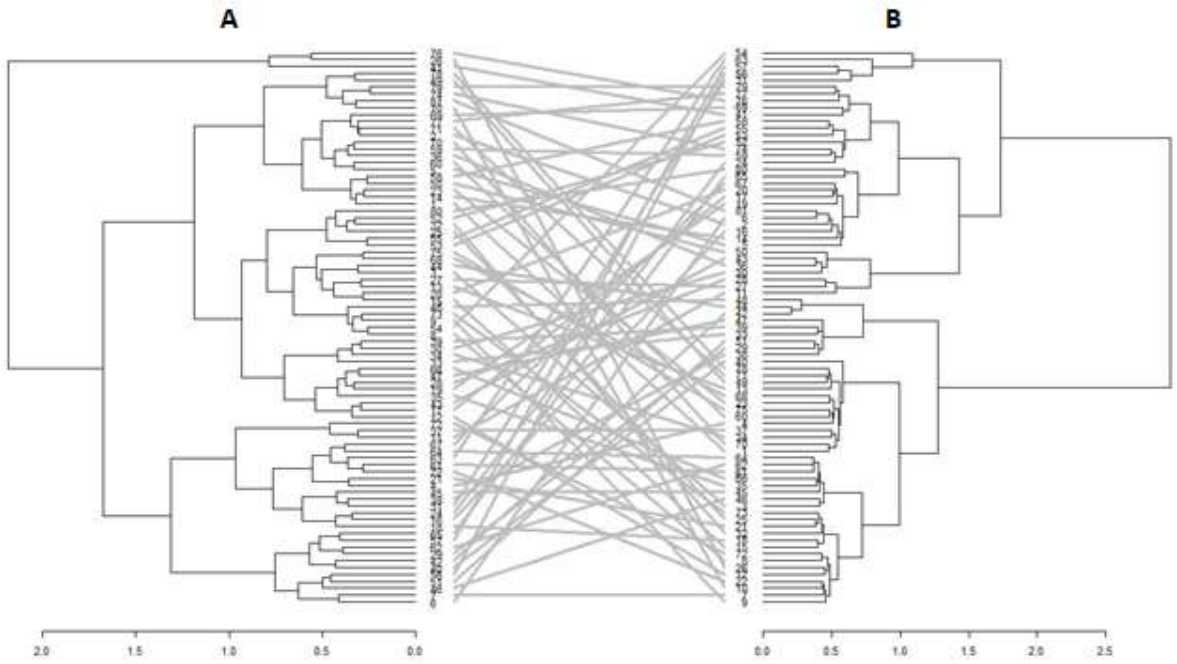

Figure 4.4. Comparison of hierarchical cluster dendrograms based on phenotypic traits (A) and SNPs data (B) in 81 pigeonpea genotypes. See Table 3.1, Chapter 3 for code of genotypes

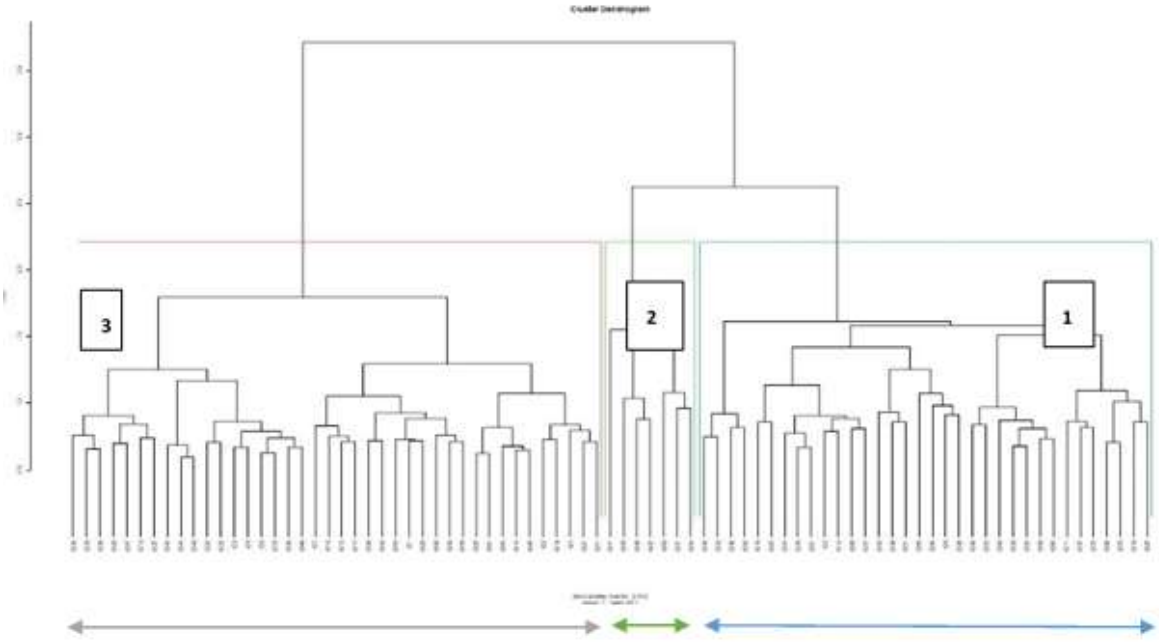

Figure 4.5. Hierarchical cluster based on the combined phenotypic and molecular data in 81 pigeonpea genotypes.

Numbers 1, 2, and 3 denote Clusters 1, 2 and 3, respectively, See Table 3.1, Chapter 3 for code of genotypes

4.4 Discussion

4.4.1 Diversity and Differentiation Based on Phenotypic Traits

Determination of genetic diversity present among genotypes, populations and gene pools is essential to identify unique individuals as sources of genes influencing quantitative or qualitative traits. Morphological and agronomic traits are essential in preliminary description and classification of germplasm for plant breeding programs (Zavinon *et al.* 2018). Several studies assessed the genetic diversity in pigeonpea using morphological descriptors (Upadhyaya *et al.* 2007, Manyasa *et al.* 2009), biochemical markers (Yang *et al.* 2006, Bohra *et al.* 2017, Zavinon *et al.* 2018) and DNA based molecular markers (Varshney *et al.* 2012). The present study revealed significant genetic variation for quantitative traits among 81 pigeonpea genotypes assessed in six environments in Malawi (Table 4.1) indicating the presence of genetic variation for breeding. The genotype performances were also affected by significant genotype × environment interactions, suggesting that genotype performances were not consistent in all the environments. The impact of environmental variance will reduce selection efficiency based on quantitative traits alone. Hence, there is need to further evaluate the available diversity using molecular markers that are not influenced by environmental variance.

The first two principal components accounted for only 28.55% of the morphological variation among the tested genotypes (Table 4.2), indicating that there was a need for more components to differentiate the genotypes. The failure to differentiate the genotypes using few principal components could be due to the high level of relatedness among the genotypes since there was shared parentage among the breeding lines and cultivars. The genotypes were developed by ICRISAT with common parentage from a few landraces or elite lines collected within east and southern Africa. Conversely, Zavinon *et al.* (2018) reported relatively higher cumulative variance (76.9%) explained by the first two PCs in genetic diversity studies of pigeonpea landraces collected from Uganda. In the present study, the PCA identified traits such as GYD, DTF, DTM, PH, NPB, NPP, NRP HSWT and NSB that had very high contributions to the principal components as the most useful in differentiating the pigeonpea genotypes.

4.4.2 Genetic Diversity Based on SNP Markers

The expected heterozygosity and polymorphic information content (PIC) measure genetic diversity among genotypes in a breeding population (Kumar and Abbo 2001). The PIC values

indicate the allelic diversity within individuals and the usefulness of markers for tracking between offspring and parental genotypes, while the gene diversity for the haploid markers provides an estimate of the average genetic distance among individuals in the population (Nei 1978). In the present study, the PIC values ranged from 0.00 to 0.38 showing that the germplasm displayed various levels of allelic diversity. However, the observed average PIC value of 0.11 indicate that the diversity was moderately low (Table 4.3). The average PIC value observed in this study was comparable to 0.16 reported by Saxena *et al.* (2014), who evaluated 184 pigeonpea accessions obtained from the ICRISAT genebank. The low PIC value indicate that the individuals in the germplasm had a narrow genetic base. Conversely, Yang *et al.* (2006) reported moderately informative DArT and SSR markers, with average PIC values of 0.34 and 0.41, for 232 and 48 pigeonpea accessions obtained from the ICRISAT genebank and Tanzania, respectively. The low levels of observed heterozygosity (0.22), gene diversity (GD) (0.14) and minor allele frequency (MAF) (012) indicated that the majority of individuals were homozygous and shared common alleles. Pigeonpea is predominantly self-pollinating species and the low heterozygosity levels is concomitant with its autogamous mating system. Similarly, (Kimaro *et al.* 2020) reported low levels of observed heterozygosity (0.27) among Tanzanian pigeonpea accessions. The lack of high heterozygosity, low genetic diversity among individuals and rare variants in the population could present bottlenecks for breeding. Adequate genetic diversity facilitates the adaptation of populations to changes in environmental conditions (Markert *et al.* 2010). High heterozygosity and rare variants provide opportunities for optimal gene recombinations during cultivar development (Imai-Okazaki *et al.* 2019). Hence, the current population will need to be harnessed to undergo recombination events to increase heterozygosity, frequency of rare variants and genetic diversity to enhance prospects of pigeonpea improvement in Malawi.

4.4.3 Population Structure Analysis Using SNP Markers

The population structure analysis identified three groups among the 81-pigeonpea genotypes (Figure 4.2) that included genotypes from different selections showing that clustering was irrespective of source of collection. The lack of a clear variation among the three groups showed that there were possibly many admixtures in the groups that reduced genetic differentiation (Fst) between the groups (Table 4.4). Pigeonpea is an orphan crop suffering from a lack of dedicated improvement programs and as such landraces are still commonly

grown among smallholder farmers leading to admixtures and germplasm collections that are not well characterised. The Fst is a measure of population differentiation due to genetic structure. It is reported that an Fst value greater than 0.15 is considered as high (Frankham *et al.* 2002). Consistent with clustering using the phenotypic data, the 81 genotypes were grouped into three clusters that were overlapping between the breeding lines, cultivars, and landraces confirming the existence of admixtures within these groups.

The grouping of genotypes into three clusters (Figures 4.1 and 4.3) using morphological attributes and SNP markers revealed a mixture of breeding lines, landraces, and cultivars in each group. This could be attributed to the geographical proximity between the two countries, Malawi and Tanzania, where the landraces were collected. Farmers between the two countries have a long history of sharing germplasm. In addition, the breeding lines from ICRISAT were developed using some parents selected from the landraces from Tanzania and Malawi, because east Africa is believed to be a centre of diversity for pigeonpea. Hence, the genotypes in the germplasm were likely to be related. These findings corroborated with that of Yang *et al.* (2006), who reported that there was little variation among the cultivated pigeonpea varieties collected in Africa.

4.4.4 Combined Analysis Using Phenotypic Traits and SNPs Markers

A joint analysis of phenotypic and genotypic data was conducted to capture the genetic variability in the population. The comparison between the phenotypic and genotypic information showed that 45.7% (Figure 4.5) of the accessions evaluated maintained their membership both across the phenotypic and molecular clustering, showing that the phenotypic and molecular matrices were different but complementary. Expectedly, the phenotypic and genotypic clusters were different due to the impact of environmental variance on phenotypic expression. The use of both derived clusters would increase precision in the selection of divergent parents from the groups for breeding. Increased precision in selection using a combination of genotypic and phenotypic data have been previously reported in legumes such as cowpea (Nkhoma *et al.* 2020). The present findings have a positive implication for pigeonpea improvement. New breeding populations can be developed by hybridization among the three divergent genetic groups, especially those that have maintained their groups, in order to broaden the genetic base as part of a pigeonpea pre-breeding program in Malawi.

4.5 Conclusions

The present study assessed the genetic diversity among the 81-pigeonpea accessions sourced from Malawi, Tanzania, and ICRISAT/Kenya. Phenotypic analysis using the qualitative and quantitative traits showed that there was significant variation among the accessions. The genetic diversity present in the test populations were confirmed using morphological traits, SNPs data, and the joint analysis. The population structure and hierarchical clustering methods grouped the accessions into three clusters, enabling the selection of divergent individuals as parents for hybridization in pigeonpea improvement programs. Genotypes such as MWPRL 14, TZA 5582, MWPLR 4, MWPLR 16, Sauma (ICPL 9145) and Kachangu (ICEAP 00040) were divergent and recommended as parental lines for development of new breeding populations. This study provided insights on pigeonpea genetic profile and identified promising genotypic resources for effective breeding and conservation in Malawi.

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CHAPTER 5. COMBINING ABILITY, GENE ACTION AND HERITABILITY FOR AGRONOMIC TRAITS AND *FUSARIUM* WILT (*Fusarium* udum Butler) RESISTANCE IN PIGEONPEA

Abstract

Combining ability analysis is fundamental in breeding programs to select desirable parents and progenies. The objectives of the study were to estimate the combining ability effects and determine the gene action controlling agronomic traits and *Fusarium* wilt (FW) resistance in pigeonpea. Twenty-five progenies developed from ten parental lines using a factorial mating design were evaluated for agronomic traits together with their parents in two sites in Malawi using a 7 × 5 alpha lattice design. FW resistance was assessed using a root dip inoculation technique in a glasshouse. Parents, ICEAP 87105, and ICEAP 01285 had desirable general combining ability (GCA) (-32.90 and -14.16 respectively) for days to 75% maturity (DTM), parental lines, MWPLR 16, Sauma and Mwaiwathualimi had desirable GCA (319.11, 168.8 and 46.45 respectively) for grain yield (GYD) and parental lines, TZA 5582, ICEAP 00554, Mwayiwathualimi and Sauma had desirable GCA effects (-3.16, -0.54, -0.24 and 0.17 respectively) for FW resistance. Hybrids such as TZA 5582 × MWPLR 22, TZA 5582 × MWPLR 14, and Mwayiwathualimi × MWPLR 22 had desirable specific combining ability (SCA) effects for DTM (-1.22 -1.51 and -0.91 respectively), GYD (80.93, 42.67 and 79.55 respectively) and FW resistance (-1.10, -0.15, and -1.66 respectively). The study further revealed that additive gene effects were important in inheritance of DTF, DTM and PH traits and non-additive gene effects were important in inheritance of GYD, 100 seed weight (HSWT) and FW resistance. This suggest that both pedigree and recurrent selection are important to achieve pigeonpea improvement. The new population developed from this study is a valuable genetic resource for pigeonpea improvement in Malawi.

Key words: combining ability, early maturity, host resistance, high yielding, pigeonpea

5.1 Introduction

Pigeonpea [*Cajanus cajan* (L.) Millspaugh, 2n=2x=22] is an important legume crop in the semi-arid tropics of Africa, Asia and Latin America. Pigeonpea grain is rich in protein (21-25%) content suitable to complement cereal based diets in Africa (Saxena *et al.* 2010). Pigeonpea has a high biomass productivity, making it an ideal fodder crop. Furthermore, it is used as a companion crop in intercropping systems to restore soil fertility through its symbiosis with nitrogen-fixing bacteria, and as a medicinal crop (Mula and Saxena (2010). Despite its diverse uses in the food and feed industry, and local and regional markets, pigeonpea has not received as much research and development support as compared to other legumes such as common bean and groundnut. Consequently, the majority of farmers in sub-Saharan Africa, including Malawi, cultivate pigeonpea using landrace varieties, which have low yield potential. The average grain yields in Asia and Africa are estimated at 866.2 and 736.2 kg ha^{-1}, respectively, compared to the potential yield of 2,500 kg ha^{-1} (Saxena 2008). There is a need to develop new and improved cultivars to enhance the productivity of pigeonpea. The low productivity of pigeonpea is also caused by various insect pests and diseases, and drought and heat stress, among other factors. In Malawi, *Fusarium* wilt (FW) caused by the fungus *Fusarium udum* (Butler) is among the major challenges of pigeonpea production. The disease is destructive in most pigeonpea growing countries, including Kenya, Tanzania and India (Hillocks *et al.* 2000, Gwata *et al.* 2006, Reddy *et al.* 2012). *Fusarium* wilt reportedly led to annual economic losses at US$ 71 million in India in 2011. In eastern Africa FW causes yield loss with a monetary value of US$ 5 million (Reddy *et al.* 2012). FW can cause 50 to 100% grain yield losses, depending on cultivar susceptibility (Soko 1992). In addition to FW, drought stress in Malawi has significantly reduced the growing season of the crop. There has not been much emphasis on pigeonpea breeding and variety deployment in Malawi. Hence, low yielding landraces are dominantly used in the country. Therefore, it is an overriding consideration to develop high yielding, FW resistant, and early maturing varieties through exploiting the genetic variation available in various gene pools. Combining ability analysis or progeny testing is fundamental to select desirable parents and progenies to breed early maturing pigeonpea varieties with high yield potential and *Fusarium* wilt (FW) resistance.

Combining ability analysis provides useful information on the gene action controlling trait inheritance and to identify superior genotypes as donor parents and best performing crosses

to develop breeding populations (Griffing, 1956). Combining ability effects are broadly categorised in general combining ability (GCA) and specific combining ability (SCA) effects. A higher GCA effect of parents relates to additive gene action, while SCA effects in crosses is due to non-additive gene action (Griffing, 1956; Acquaah, 2009). Information on the GCA effects is crucial to select superior parental genotypes that would produce desirable offspring in subsequent crosses. Information on the SCA effects are useful to select the best cross combinations or families derived from favourable allelic combinations (Pandey *et al.* 2014). Different mating designs, including factorial or North Carolina designs and diallels, among others, are used to analyse combining ability and to deduce gene action controlling quantitative traits inheritance (Falconer *et al.* 1996).

The North Carolina mating designs were first developed by Comstock and Robinson (1948) to estimate combining ability effects, variance components, and heritability of quantitative traits. The North Carolina Design II or factorial mating design partitions the variance components into additive and dominance variances to discern the magnitude of heritability of quantitative traits to guide selection. Yohane *et al.* (2020) evaluated a diverse set of pigeonpea germplasm collections and identified superior genotypes for pre-breeding programs in Malawi. The selected lines displayed complementary and farmer-preferred attributes which are useful genetic resources for variety development after designed crosses and strategic selection. Hence, the objective of this study was to assess the combining ability effects and to determine the gene actions controlling agronomic traits and FW resistance in pigeonpea parents and derived progenies for breeding.

5.2 Materials and Methods

5.2.1 Parental Lines, Crosses and Mating Design

The study used 10 genotypes selected from a previous germplasm evaluation study (Yohane *et al.* 2020). The selected parents were genetically distinct, based on *Fusarium* wilt resistance (Kimaro, 2016), maturity period, cooking and eating quality, yield, and yield-related traits (Table 5.1). The parents included landraces, introduced varieties, and advanced breeding lines sourced from Malawi, Tanzania, and Kenya.

Table 5.1. Descriptors and source of parental genotypes used in the study

Genotype name/designation	Role in cross	Attribute(s)	Breeding history	Source
MWPLR 14	Female	Short cooking time, pod borer resistant, high yielding, cream seed colour	Landrace	DARS, Malawi
MWPLR 22	Female	Short cooking time, pod borer resistant	Landrace	DARS, Malawi
ICEAP 00554	Female	Resistant to *Fusarium* wilt, medium maturing	Advanced line	ICRISAT, Kenya
MWPLR 16	Female	Good eating quality, short cooking time and large grain size	Landrace	DARS, Malawi
ICEAP 87105	Female	Early maturing, white/cream colour	Variety	DARS, Malawi
TZA 5582	Male	High yielding, *Fusarium* wilt resistant	Landrace	TARI, Tanzania
Mwayiwathualimi	Male	Medium maturing, high yielding, *Fusarium* wilt tolerant	Variety	DARS, Malawi
MWPLR 4	Male	Good for fresh pods, very good eating quality, fast cooking, cream seed colour	Landrace	DARS, Malawi
Sauma	Male	*Fusarium* wilt resistant, high yielding	Variety	DARS, Malawi
ICEAP 01285	Male	Early maturing	Advanced line	ICRISAT, Kenya

DARS= Department of Agricultural Research Services, TARI= Tanzania Agricultural Research Institute, ICRISAT= International Crops Research Institute for the Semi-Arid Tropics

5.2.2 Generation of Crosses and Phenotypic Evaluation

The two sets of parents (five male and five female) were crossed using a 5 × 5 North Carolina mating design II which provided 25 crosses. Crosses were carried out manually by removing anthers from the staminal column using fine forceps before flowers open up. Ten buds per branch were emasculated, and small buds were removed from the branch to prevent competition with the inflorescence. Pollination was conducted 24 hours after emasculation to allow the stigma to recover from disturbances during emasculation and increase its receptiveness to pollen. Both emasculation and pollination were carried out in the morning before 10:00 am to avoid heat stress, which can cause the stigma of the emasculated plants to rupture. Each pollinated flower was tagged and labelled, indicating the parents involved in the cross and crossing date. Since the success rate of crosses in pigeonpea is generally low (20%), as many flowers as possible were crossed to produce a minimum of 60 seeds per cross (Sharma *et al.* 1980). The F_1 seeds were harvested at maturity and retained for subsequent evaluation.

The 25 F_1 and 10 parents were evaluated in two locations (Chitedze and Makoka Agricultural Research Stations) in the 2019/20 crop season. The geographic location, altitude, weather, and soil characteristics of the study locations are presented in Table 5.2. The experiments were established using a 7 × 5 alpha lattice design with two replications. Plot area consisted of two rows measuring 3m each, with inter- and intra-row spacing of 0.75m and 0.90 m, respectively. All agronomic practices were applied following the standard practices for pigeonpea production in Malawi (Kananji *et al.* 2016).

Table 5.2. Physical and weather characteristics of the study locations

Site	Latitude	Longitude	Altitude (masl)	Soil texture	Rainfall (mm)	Min Temp (^{O}C)	Max Temp (^{O}C)
Chitedze	13 ^{O} 59' S	33 ^{O} 38' E	1146	Sandy clay	841 3	17 9	32 4
Makoka	15 ^{O} 32' S	35 ^{O} 11' E	1029	Sandy clay loam	1001 8	16	25 1

Masl= metres above sea level, mm= millimetres, min= minimum, max= maximum, temp= temperature, ^{o}C= degrees Celsius

5.2.3 Agronomic Data Collection

Data on days to 50% flowering, days to 75% maturity, plant height, number of primary branches, number of secondary branches, number of pods per plant, number of seeds, grain yield, and 100 seed weight were collected according to pigeonpea descriptors of the International Board for Plant Genetic Resources (IBPGR 1993) (Table 5.3). Grain yield was converted to kg ha^{-1} using the following formula:

$$\left(\frac{Plot\ weight\ (g)}{Plot\ area\ (m)} \times \frac{100-14}{100-mc}\right) \times 10{,}000$$

where; mc is moisture content measured at harvesting, 14% is standard constant moisture content for legumes (Parker A and Namuth-Covert D 2017), and 10,000 is the conversion factor for a hectare.

Table 5.3. Descriptors for the pigeonpea quantitative traits recorded in this study

Trait	Code	Description
Plant height	PH	Measured in cm from plant base to the tip of the main stem
Days to 50% flowering	DTF	Number of days from sowing until when 50% of the plants have at least one open flower
Primary branches	PBR	The average number of primary branches of 10 randomly selected and tagged plants
Secondary branches	NSB	The average number of secondary branches of 10 randomly selected and tagged plants
Days to 75% maturity	DTM	Number of days from sowing until when 75% of the pods in a plot turn brown
Number of seeds per pod	NSP	The average number of pods per plant from 10 randomly selected and tagged pods
Number of pods per plant	NPP	The average number of pods from 10 randomly selected and tagged plants
Grain yield (t/ha)	GY	Weight of the grain harvested in a plot extrapolated to t/ha
100 seed weight (g)	SWT	Weight of a random sample of 100 grain

5.2.4 Evaluation of F_1 Progenies and Parental Lines for *Fusarium* Wilt Resistance Under Glasshouse Conditions

5.2.4.1 Preparation of Inoculum and Pathogenicity Test

Both laboratory and glasshouse experiments were conducted at ICRISAT- Malawi, Chitedze Agricultural Research Station. *Fusarium udum* was isolated from infected pigeonpea plants collected from Zomba and Chiradzulu districts of southern Malawi, where *Fusarium* wilt is most prevalent. The pathogen was isolated from cut pieces of the inner stem, which was surface sterilized with 3% sodium hypochlorite (NaOCl) for five minutes and rinsed three times in distilled sterile water. The sterilized plant tissues were then blotted dry on filter paper and placed aseptically on potato dextrose agar (PDA) media. The plates were sealed with parafilm and incubated at 28ºC for 3 days. Pure cultures of *F. udum* were developed using the single spore technique (Agrios 2005). Cultures were prepared on fresh PDA in order to check for mycelial growth and sporulation. The cultures were preserved by the repeated culturing method (Tuite, 1969).

Before inoculation, pathogenicity tests were carried out using KAT60/8, a *Fusarium* wilt susceptible cultivar. Three isolates EN 156, EN 140 and, EN 119 were used in the pathogenicity test. Isolate EN156 was selected as the best based on high severity/mortality rate and morphological characteristics (Booth 1971). To prepare the inoculum suspension, mycelia from the edge of *F. udum* cultures were aseptically placed on 100 ml PDA broth in 250 ml conical flasks and sealed with parafilm. The flasks were shaken four to six times daily for 10 days at room temperatures (25 - 30 C). The mycelial mat growth in the flasks were

macerated in a Waring blender for one minute in distilled sterile water, and filtered through a cheesecloth. The concentration of suspension was adjusted to 3-4 x 10^6 spore ml^{-1} by adding distilled sterile water.

5.2.3.2. Glasshouse Evaluation for *Fusarium* Wilt Resistance

The 10 parents and 25 crosses were evaluated in the glasshouse for *Fusarium* wilt resistance through artificial inoculation using the root dip method. Ten-day-old seedlings were inoculated by dipping the roots in the inoculum suspension as per Karimi et al. (2010). The roots of the seedlings were trimmed with a sterile scissor and submerged into the tubes containing 30 ml of *F. udum* spore suspension for 30 minutes. The inoculated seedlings were transplanted into mini pots of 15 cm diameter, which were surface sterilized with 0.1% mercuric chloride. The trial was laid out as a completely randomized design with two replications. Two seedlings were planted per pot. The number of wilting plants showing *Fusarium* wilt symptoms was recorded weekly, starting from two days after inoculation up to three months. Assessment of reaction to the *Fusarium* wilt disease was based on Kannaiyan *et al.* (1984) where genotypes with 0 – 20% plant mortality rate were categorised as resistant, 21 - 40% mortality rate = moderately resistant, 41 – 60% mortality rate = susceptible, 61-80% = moderately susceptible, and 81 – 100% = highly susceptible.

5.2.3 Data Analysis

Data on agronomic traits were subjected to analysis of variance (ANOVA) in Genstat18th Edition (Payne *et al.* 2017). The disease incidence data were normalized by arcsine transformation before being subject to ANOVA (Gomez and Gomez, 1984). General combining ability (GCA) and specific combining ability (SCA) effects were deduced in AGD-R version 4.0 (Rodríguez et al., 2015) based on a North Carolina II design (Garretsen and Keuls 1978). The following model was used:

$$Y_{ijk} = \mu + g_i + \varepsilon_j + r_{ij} + e_{ijk}$$

where Y_{ijk} is the observed measurement for the cross made between i^{th} and j^{th} parents grown in the k^{th} replication or environment; μ is the population mean; g_i and ε_j are the additive component of the i^{th} female and j^{th} male parents; r_{ij} the interaction component of the cross

between the i^{th} female and j^{th} male parents and e_{ijk} is the error term associated with the ij^{th} cross evaluated in the k^{th} replication or environment.

Variance components attributable to general and specific combining ability male and female effects were estimated using the following formulas as suggested by Burton and Devane (1953).

$\sigma^2_m = \sigma^2_f = COV_{H.S} = ¼\ \sigma^2_A$

$\sigma^2_{mf} = COV_{F.S} - 2\ COV_{H.S} = ¼\ \sigma^2_D$

Where σ^2_m = male variance, σ^2_{mf} = male × female variance, σ^2_A = additive variance, σ^2_D = dominant variance, COV = covariance

$$\sigma_g^2 = \frac{MSg - MSe}{r}$$

$$\sigma_p^2 = \sigma_g^2 + \sigma_e^2$$

Where; σ_g^2 is the genotypic variance, σ_p^2 is the phenotypic variance, σ_e^2 is the environmental variance, MSg is the mean square due to genotypes, MSe is the environmental mean square and r is the number of replications.

Predictability ratio $=2\sigma^2_{gca}/ (2\sigma^2_{gca}+ \sigma^2_{sca})$ was calculated as suggested by (Baker 1978).
Heritability estimates were derived through variance components (Allard 1999) as follows:

Broad sense heritability $(h^2_b) = \frac{\sigma_g^2}{\sigma_p^2}$ x100

Where σ_g^2 is genetic variance and σ_p^2 is phenotypic variance.

Narrow sense heritability $(h^2_n) = \frac{\sigma_A^2}{\sigma_p^2}$

Where σ_A^2 = additive variance and σ_p^2 is phenotypic variance.

5.3 Results

5.3.1 Analysis of Variance (ANOVA) for Agronomic Traits and FW

The genotypes exhibited significant ($P \leq 0.001$) variation (Table 5.4) for DTF and DTM, PH, GYD, and HSWT. Conversely, non-significant differences were observed among the genotypes for NPB, NSB, and NSP traits. The genotype × location interaction was non-

significant for all the studied traits except for PH and HSWT. The genotypes exhibited significant ($P \leq 0.01$) variation for FW mortality (Table 5.5).

The parents ICEAP 87105 (female), and ICEAP 01285 (male) were the earliest to flower in 73 days and matured in 106 and 109 days, respectively (Table 5.6). Among the crosses, ICEAP 01285 × ICEAP 00554 flowered in 66 days and matured in 96 days. ICEAP 01285 × MWPLR 22 flowered in 75 days and matured in 104 days, and ICEAP 01285 × ICEAP 87105 flowered in 78 days and matured in 116 days. Plant height also varied widely among the parental lines, with genotype ICEAP 87105 (124 cm) being the shortest, and Sauma (223 cm) was the tallest. Crosses ICEAP 01285 × ICEAP 00554 (141 cm), ICEAP 01285 × MWPLR 22 (158 cm) and ICEAP 01285 × ICEAP 87105 (165 cm) were the shortest crosses. The tallest families among the crosses were derived from TZA 5582 × MWPLR 16 (244 cm) and Sauma × ICEAP 00554 (236 cm). The female parental lines MWPLR 14, MWPLR 16, and MWPLR 22 exhibited the biggest seed sizes with a mean 100-seed weight of 24.00, 23.00, and 21.00 respectively. Crosses such as TZA 5582 × MWPLR 22, Sauma× ICEAP 00554, and Mwayiwathualimi × MWPLR 22 had the biggest seed sizes with 100-seed weight of 20.00 g. Among the parental lines, MWPLR 16 and Sauma exhibited the highest grain yield potential with mean GYD of 2807 and 2143 kg ha^{-1}, respectively. Cross ICEAP 01285 × MWPLR 22 had the highest GYD (3847 kg ha^{-1}) followed by Mwayiwathualimi × MWPLR 22 (2029 kg ha^{-1}), and MWPLR 4 × MWPLR 14 (1732 Kg ha^{-1}) compared to other crosses and parental lines (Table 5.6).

Table 5.4. Mean squares for grain yield and yield components computed among 35 pigeonpea genotypes evaluated in two locations in Malawi

Source of variation	DF	DTF	DTM	PH	NPB	NSB	NPP	NSP	GYD	HSWT
Location	1	30.18	2799.10 ***	10638.40 ***	835.46 ***	3713.15 ***	527058.00 ***	19.31 ***	132974.00	108.06 ***
Replication	1	75.78	749.80 *	1279.30	35.00 *	38.06	5582.00	0.00	5.00	1.21
Block (Replication)	4	300.93 **	595.50*	3999.90 ***	6.98	22.13	14740.00	0.36	1024550.00 ***	21.33***
Genotype	34	1329.93 ***	2668.40 ***	3168.30 ***	15.05	92.09	12607.00 *	0.22	1185447.00 ***	27.80 ***
Genotype * Location	34	43.71	86.50	1232.00 **	11.76	73.56	10488.00	0.35	111055.00	3.78 ***
Residual	65	59.26	149.4	548.9	11	99.4	8065	0.35	125830	1.35

DF = degrees of freedom, DTF = days to 50% flowering, DTM = days to 75% maturity, PH = plant height, NPB = number of primary branches per plant, NSB = number of secondary branches per plant, NPP = number of pods per plant, NSP = number of seeds per pod, GYD = grain yield, HSWT = 100 seed weight, *, ** and *** = significance at 0.05, 0.01 and 0.001 probability levels, respectively.

Table 5.5. Analysis of variance for FW resistance computed among 35 pigeonpea genotypes evaluated in glasshouse in Malawi

Source of variation	DF	FW mortality (%)
Replication	1	978.00
Genotype	34	1939.00 *
Residual	34	1016.00

DF = degrees of freedom, FW = *Fusarium* wilt mortality rate, ** = significance at 0.01 probability level

Table 5.6. Mean values for agronomic traits among 25 F_1 population and 10 parents of pigeonpea evaluated at Chitedze and Makoka in Malawi

Genotype	DTF	DTM	PH (cm)	NPB	NSB	NPP	NSP	GYD (kg ha^{-1})	HSWT (g/100 seed)
Female parents									
MWPLR 14	106	148	206	15	19	231	5	1545	24
MWPLR 22	112	156	189	14	19	209	6	1610	21
ICEAP 00554	113	157	146	15	16	292	6	1391	19
MWPLR 16	111	147	171	15	22	210	5	2807	23
ICEAP 87105	73	106	124	17	31	267	6	1246	14
Male parents									
TZA 5582	125	179	193	19	19	220	6	1408	13
Mwayiwathualimi	113	162	199	14	17	186	6	1599	17
MWPLR 4	101	139	182	13	18	224	6	1023	15
Sauma	153	225	223	13	16	208	6	2143	20
ICEAP 01285	73	109	139	18	20	259	6	1182	14
Crosses									
TZA 5582 × MWPLR 14	78	117	166	18	30	338	6	1631	17
TZA 5582 × MWPLR 22	117	167	222	16	29	301	6	3847	20
TZA 5582 × ICEAP 00554	108	145	230	18	21	217	6	1307	17
TZA 5582 × MWPLR 16	105	141	244	19	25	297	6	1291	15
TZA 5582 × ICEAP 87105	103	131	204	17	26	370	6	1453	19
Mwayiwathualimi × MWPLR 14	110	155	225	18	30	372	6	1600	19
Mwayiwathualimi × MWPLR 22	116	156	205	16	21	299	6	2029	20
Mwayiwathualimi × ICEAP 00554	119	172	206	18	24	232	6	1444	17
Mwayiwathualimi × MWPLR 16	103	140	218	13	24	383	6	1109	19
Mwayiwathualimi × ICEAP 87105	112	147	222	15	17	280	6	1368	19
MWPLR 4 × MWPLR 14	114	158	230	13	25	392	6	1732	19
MWPLR 4 × MWPLR 22	105	148	200	13	24	377	6	1221	13
MWPLR 4 × ICEAP 00554	113	154	201	17	16	288	6	1151	15
MWPLR 4 × MWPLR 16	114	159	196	13	18	259	6	923	15
MWPLR 4 × 87105	100	134	179	16	19	303	6	683	17

Table 5.6. Continued

Sauma × MWPLR 14	111	150	197	14	28	243	5	973	16
Sauma × MWPLR 22	121	169	215	16	26	308	6	1082	21
Sauma × ICEAP 00554	128	195	236	18	23	250	6	1463	20
Sauma × MWPLR 16	100	133	229	15	22	269	6	1170	18
Sauma × ICEAP 87105	112	158	207	16	21	257	6	1416	16
ICEAP 01285 × MWPLR 14	88	126	171	14	19	208	5	1577	17
ICEAP 01285 × MWPLR 22	75	104	158	12	27	345	6	1427	15
ICEAP 01285 × ICEAP 00554	66	96	141	17	24	304	5	947	15
ICEAP 01285 × MWPLR 16	79	118	188	15	14	281	6	1218	16
ICEAP 01285 × ICEAP 87105	78	116	165	14	26	250	6	1114	17
Mean	104	146	195	16	22	278	6	1461	18
LSD (0.05)	10.1***	14.9***	39.7***	4.9ns	13.1ns	134.4*	0.8ns	494.0***	2.2***
CV (%)	6.9	7.3	14.5	22.3	42.3	34.4	10.8	23.9	9.1

CV = coefficient of variation, LSD = least significant difference, DTF = days to 50% flowering, DTM = days to 75% maturity, PH = plant height, NPB = number of primary branches per plant, NSB = number of secondary branches per plant, NPP = number of pods per plant, NSP = number of seeds per pod, GYD = grain yield, HSWT = 100 seed weight, *, ** and *** = significance at 0.05, 0.01 and 0.001 probability levels, respectively.

5.3.2 *Fusarium* Wilt Resistance

There were significant differences ($P \leq 0.05$) among the test genotypes for FW mortality (Table 5.7). FW mortality ranged from 0 to 75%. Two parents, TZA 5582 and Sauma, were highly resistant to FW, exhibiting 0% mortality, while Mwayiwathualimi, ICEAP 00554, and ICEAP 87105 were moderately resistant, with mortality rates of 25%. Crosses including TZA 5582 × MWPLR 14, Mwayiwathualimi × ICEAP 00554, Mwayiwathualimi × MWPLR 16, MWPLR 4 × ICEAP 00554, MWPLR 4 × ICEAP 87105, Sauma × MWPLR 14, Sauma × MWPLR 22, Sauma × ICEAP 00554 were resistant to FW, displaying mortality rates of 0%. In comparison, crosses TZA 5582× ICEAP 00554, TZA 5582× MWPLR 16, TZA 5582× MWPLR 14, Sauma × MWPLR 16, and Sauma × ICEAP 87105 were moderately resistant to FW with mortality rates of 25%.

Table 5.7. Mortality rates among 25 hybrids and 10 parents of pigeonpea when inoculated with *Fusarium udum* isolates in Malawi

Genotype	FW Mortality rate (%)
Female parents	
MWPLR 14	75
MWPLR 22	75
ICEAP 00554	25
MWPLR 16	75
ICEAP 87105	25
Male parents	
TZA 5582	0
Mwayiwathualimi	25
MWPLR 4	75
Sauma	0
ICEAP 01285	50
Crosses	
TZA 5582 × MWPLR 14	0
TZA 5582 × MWPLR 22	75
TZA 5582 × ICEAP 00554	25
TZA 5582 × MWPLR 16	25
TZA 5582 × ICEAP 87105	50
Mwayiwathualimi × MWPLR 14	25
Mwayiwathualimi × MWPLR 22	50
Mwayiwathualimi × ICEAP 00554	0
Mwayiwathualimi × MWPLR 16	0

Table 5.7. Continued

Mwayiwathualimi × ICEAP 87105	50
MWPLR 4 × MWPLR 14	25
MWPLR 4 × MWPLR 22	50
MWPLR 4 × ICEAP 00554	0
MWPLR 4 × MWPLR 16	50
MWPLR 4 × 87105	0
Sauma × MWPLR 14	0
Sauma × MWPLR 22	0
Sauma × ICEAP 00554	0
Sauma × MWPLR 16	25
Sauma × ICEAP 87105	25
ICEAP 01285 × MWPLR 14	50
ICEAP 01285 × MWPLR 22	75
ICEAP 01285 × ICEAP 00554	50
ICEAP 01285 × MWPLR 16	50
ICEAP 01285 × ICEAP 87105	50
Minimum	0
Maximum	75
Mean	33
LSD (0.05)	11.6 *
CV (%)	24.5

CV = coefficient of variation, LSD = least significant difference, * = significance at 0.05 probability level, FW = *Fusarium* wilt mortality rate

5.3.3 Combining Ability Effects

The analysis of variance (ANOVA) showing the combining ability effects for yield and yield components and FW resistance are presented in Tables 5.8 and 5.9, respectively. There existed significant ($P \leq 0.05$) differences among male and female GCA effects for DTF, DTM, NPB, HSWT, GYD, and FW resistance. The female GCA effects were significant ($P<0.05$) for DTF and DTM. The SCA effects of the crosses for FW resistance and most agronomic traits varied significantly ($P<0.05$) except for NPB, NSB, NPP, and NSP. There were significant ($P<0.05$) SCA × environment interaction effects for PH and HSWT. Due to non-significance differences in GCA_{male}, GCA_{female}, and SCA_{cross} for NPB, NSB and NSP (Table 5.8), the traits are excluded for general combining ability, specific combining ability and variance components computation.

Table 5.8. Combining ability analysis of variance for agronomic traits of 25 pigeonpea hybrids derived from a 5 × 5 North Carolina design II

Source of variation	DF	DTF	DTM	PH	NPB	NSB	NPP	NSP	GYD	HSWT
Location (E)	1	30.3	8735.4 ***	54965.0 ***	114.4 ***	93.7 *	3236.0 **	22.5 ***	5968860.0 ***	1008.1 ***
Replication	1	0.1	625.0*	76.0	17.6	566.4*	128.0	0.0	68388.0	7.3 *
Block (Replication)	4	515.73 ***	1033.0***	3293.0 ***	7.73	60.0	18214.00	0.4	333396.0 ***	19.2 ***
GCA_{male}	4	927.1 ***	3130.3 ***	7779.0 *	31.8 *	64.2	11103.0	0.2	2072443.0 ***	28.6 ***
GCA_{female}	4	5014.7 ***	7839.2 ***	2120.2 *	21.9*	140.5	13105.0	0.2	3443310.0 ***	67.5 ***
SCA_{cross}	16	136.8*	364.1 *	1937.4 *	9.0	86.5	12027.4	0.3	79774.1***	3.9 *
$GCA_{male} \times E$	4	26.0	71.2	11.61.5	5.8	17.3	13233.0	0.2	30695.0	5.2
$GCA_{female} \times E$	4	183.2 *	255.2 *	1330.6	12.9	140.5	12388.0	0.3	123671.0	0.5
$SCA_{cross} \times E$	16	40.6	69.5	1552.5 *	9.0	89.2	9215.0	0.4	100367.0	3.6 *
Residual	106	345.4	585.8	1243.1	14.5	11.8	5667.2	0.8	313554.0	15.4

DF= degrees of freedom, DTF = days to 50% flowering, DTM = days to 75% maturity, PH = plant height, NPB = number of primary branches, NSB = number of secondary branches, NPP = number of pods per plant, NSP = number of seeds per pod, GYD = grain yield, HSWT = 100 seed weight, *, ** and *** = significance at 0.05, 0.01 and 0.001 probability levels, respectively, GCA_{male} = general combining ability effect of males, GCA_{female}= general combining ability of females, SCA_{cross} = specific combining ability effect of males × females, $GCA_{male} \times E$ = interaction between general combining ability effect of males and environment, $GCA_{female} \times E$ = interaction between general combining ability effect of females and environment, $SCA_{cross} \times E$ = interaction between specific combining ability effect of the cross and the environment.

Table 5.9. Combining ability analysis of variance for FW resistance of 25 pigeonpea hybrids derived from a 5 × 5 North Carolina design II

Source of variation	DF	FW mortality (%)
Replication	1	1444
GCA_{male}	4	1000.0 ***
GCA_{female}	4	478.0 ***
SCA_{cross}	16	1104.0*
Residual	24	1367

*, ** and *** = significance at 0.05, 0.01 and 0.001 probability levels, respectively, GCA_{male} = general combining ability effect of males, GCA_{female}= general combining ability of females, SCA_{cross} = specific combining ability effect of males × females, DF = degrees of freedom, FW = *Fusarium* wilt mortality rate

5.3.4 General Combining Ability Effects

The parental lines exhibited variable GCA effects for the different traits (Table 5.10). Negative GCA effects for DTF, DTM, PH, and FW susceptibility were desirable. Among the parents, male parent ICEAP 01285 and female parent ICEAP 87105 exhibited the most desirable GCA effects for DTF, DTM, and PH. However, these lines had undesirable GCA effects for HSWT, FW susceptibility, and GYD. Male parent Sauma and female parent MWPLR 16 exhibited desirable GCA effects for HSWT and GYD, but had positive GCA effects for DTF and DTM, which were undesirable. Parental lines TZA 5582, Sauma, and ICEAP 00554 exhibited negative GCA effects for FW susceptibility, which was desirable.

Table 5.10. General combining ability effects for yield-related traits and *Fusarium* wilt susceptibility in pigeonpea parental lines

Genotype	DTF	DTM	PH	GYD	HSWT	FW susceptibility (%)
Male parents						
TZA 5582	-0.07	-4.05	18.97*	-33.51	-0.67	-3.16**
Mwayiwathualimi	2.00	-4.10	1.00	46.45	0.40	-0.24
MWPLR 4	-2.84	-5.62	-1.64	-115.79	0.20	2.10
Sauma	8.79*	18.61*	10.72	168.8*	1.4*	-0.17*
ICEAP 01285	-7.87*	-12.04*	-14.16*	-65.96	-1.33*	1.67
Female parents						
MWPLR 14	3.59	0.93	0.56	-123.59	1.28*	0.2
MWPLR 22	9.55*	13.96*	-0.09	-61.87	0.71	1.94*
ICEAP 00554	8.97*	12.81*	1.98	-85.12	-2.03*	-0.54*
MWPLR 16	4.7	5.19	-0.81	319.11*	1.87*	1.00
ICEAP 87105	-26.59**	-32.90**	-16.53*	-48.52	-1.82*	0.83
SE± (male)	3.40	5.65	8.60	92.0	0.55	4.79
SE± (females)	4.08	5.92	3.58	102.5	0.56	2.48

DTF = days to 50% flowering, DTM = days to 75% maturity, PH = plant height, GYD = grain yield, HSWT = 100 seed weight, FW = Fusarium wilt susceptibility (mortality), *, ** and *** = significance at 0.05, 0.01 and 0.001 probability levels, respectively, SE = standard error

5.3.5 Specific Combining Ability Effects

The SCA effects varied widely among the 25 hybrids (Table 11). Hyibrids, Sauma × ICEAP 87105, TZA 5582 × MWPLR 14, ICEAP 01285 × MWPLR 16, Mwayiwathualimi × ICEAP 00554, TZA 5582 × ICEAP 00554, and TZA 5582 × MWPLR 14 had negative SCA effects for DTF and DTM. Hybrid ICEAP 01285 × MWPLR 14 had desirable SCA effects for 100 seed weight. The most desirable negative SCA effects associated with FW susceptibility were computed in hybrids TZA 5582 × 00554, Mwayiwathualimi × MWPLR 22, and Mwayiwathualimi × MWPLR 14. The highest positive SCA effects for grain yield were recorded in the following hybrids; Sauma × MWPLR 16, TZA 5582 × MWPLR 22, and Mwayiwathualimi × MWPLR 22. Overall, the following hybrids; TZA 5582 × MWPLR 22, TZA 5582 × MWPLR 14, and Mwayiwathualimi × MWPLR 22 had consistently desirable SCA effect for early maturity, *Fusarium* wilt resistance, and grain yield.

Table 5.11. Specific combining ability effects for agronomic traits and *Fusarium* wilt susceptibility in 25 newly developed hybrids of pigeonpea

Crosses	DTF	DTM	PH	GYD	HSWT	FW susceptibility
TZA 5582 × MWPLR 14	-1.44	-1.51	1.75	42.67*	-0.09	-0.15
TZA 5582 × MWPLR 22	-0.478	-1.22	4.19	80.93*	-0.11	-1.10*
TZA 5582 × ICEAP 00554	-1.94**	-2.51**	-2.57***	-66.29	0.1	-2.05**
TZA 5582 × MWPLR 16	2.72*	2.33	-3.16**	-195.33*	-0.02	-0.61
TZA 5582 × ICEAP 87105	1.35	3.01	3.16	2.41	0.14*	-0.62
Mwayiwathualimi × MWPLR 14	-0.86*	-2.01*	0.54	-130.19*	-0.02	-1.63*
Mwayiwathualimi × MWPLR 22	-0.55	-0.91	-0.43	79.55*	0.04	-1.66*
Mwayiwathualimi × ICEAP 00554	-3.22***	-5.52***	1.96	0.90	0.06	0.53
Mwayiwathualimi × MWPLR 16	3.1	8.90	0.94	-69.76	0.09	0.14
Mwayiwathualimi × ICEAP 87105	2.09	0.80	-3.57**	51.62	-0.21**	2.21
MWPLR 4 × MWPLR 14	0.55	0.62	1.36	-28.37	-0.24	0.38
MWPLR 4 × MWPLR 22	-1.21	-1.11	-0.35	28.84	0.09	0.70
MWPLR 4 × ICEAP 00554	1.5	2.81	2.35	-66.12	-0.04	1.90
MWPLR 4 × MWPLR 16	-0.68*	-1.83*	3.71	-58.97	0.14*	-0.79
MWPLR 4 × 87105	0.38	0.80	4.74	31.24	0.07	1.14
Sauma × MWPLR 14	2.48	4.44	4.36	-32.26	0.15	3.00*
Sauma × MWPLR 22	3.09	3.12	-7.36	-21.08	-0.02	0.43
Sauma × ICEAP 00554	0.21	1.71	-5.58	-180.23	0.00	-1.39*
Sauma × MWPLR 16	0.58	0.81	2.81	721.90***	-0.09	1.93
Sauma × ICEAP 87105	-6.08***	-6.13***	0.94	-138.19	-0.08	1.51
ICEAP 01285 × MWPLR 14	-0.74*	-2.51*	-2.67*	69.71	0.19*	1.17
ICEAP 01285 × MWPLR 22	-0.20*	-0.88*	4.23	-59.47	0.02	0.09
ICEAP 01285 × ICEAP 00554	2.51	5.72	-0.82	40.63	-0.23*	-0.89
ICEAP 01285 × MWPLR 16	-2.83**	-5.84	-1.28*	-2.59	-0.08	-0.08
ICEAP 01285 × ICEAP 87105	-0.34	-1.33	-9.26**	-101.52	0.08	-0.40
SE±	3.11	4.83	8.55	133.07	0.28	1.67

DTF = days to 50% flowering, DTM = days to 75% maturity, PH = plant height, GYD = grain yield, HSWT = 100 seed weight, FW = *Fusarium* wilt susceptibility, *, ** and *** = significance at 0.05, 0.01 and 0.001 probability levels, respectively, SE = standard error

5.3.6 Variance Components for Agronomic Traits and *Fusarium* Wilt Resistance in Pigeonpea

The GCA variances (σ^2 GCA) for FW, HSWT, and GYD were lower than the corresponding SCA variances (σ^2 SCA) (Table 5.12). Conversely, the σ^2 GCA for DTF, DTM, and PH were higher than their corresponding σ^2 SCA. The σ^2 gca/σ^2 sca ratios varied from 0.24 (FW

susceptibility) to 54.58 (DTF). The additive (σ^2 A) variance was greater than the dominance (σ^2 D) variance in DTF, DTM, and PH. All the traits except FW susceptibility exhibited larger broad and narrow heritability estimates.

Table 5.12. Estimates of variance components, degree of dominance and heritability for agronomic traits and *Fusarium* wilt susceptibility in pigeonpea

Component	DTF	DTM	PH	GYD	HSWT	FW susceptibility
σ^2 Female	2.01	8.62	31.12	16.22	0.15	0.02
σ^2 Male	4.54	7.95	22.74	38.22	0.24	0.10
σ^2 GCA	6.55	16.57	53.86	54.44	0.39	0.12
σ^2 SCA	0.12	1.47	1.55	124.44	0.91	0.51
σ^2 GCA /σ^2 SCA	54.58	11.27	34.75	0.44	0.43	0.24
σ^2 Additive	6.55	16.57	53.86	54.44	0.39	0.12
σ^2 Dominance	0.48	5.88	6.2	497.76	3.64	2.04
H^2	0.98	0.98	0.83	0.98	0.95	0.36
h^2	0.93	0.92	0.66	0.67	0.93	0.23

DTF = days to 50% flowering, DTM = days to 75% maturity, PH = plant height, GYD = grain yield, HSWT = 100 seed weight, FW = Fusarium wilt susceptibility, σ^2 Female = female variance, σ^2 male =male variance, σ^2 GCA = general combining ability variance, σ^2 SCA = specific combining ability variance, σ^2 GCA /σ^2 SCA =ratio of general combining ability variance and specific combining ability variance, σ^2 additive = additive variance, σ^2 dominance = dominance variance, H^2 = broad sense heritability, h^2 = narrow sense heritability

5.4 Discussion

5.4.1 Analysis of Variance and Mean Performance of Genotypes

The presence of significant genotypes variation revealed genetic divergence among the parents and their progenies. These findings suggest the presence of sufficient genetic variation for selection of superior genotypes and families for further breeding. The differences in the assessed parental lines and new families show the variable genetic composition underpinning variable agronomic performance and FW reaction. The parental lines composed of short, medium, and long-maturity groups. Differences in maturity period determine the variation in vegetative growth patterns, agronomic performance and grain yield potential. Chattopadhyay and Dhiman (2006) reported a wide range of variability for the number of branches per plant, days to flowering and maturity, plant height, number of seeds per pod, and 100 seed weight in pigeonpea after evaluating parental lines and their crosses. The presence of significant genotype ×

environment interaction effects suggested that genotype performance was not consistent across the environments due to variability in soil properties, temperature conditions, among others. Yield and yield-related traits and disease resistance are polygenic traits highly influenced by environmental variance (Houle 1992), leading to variable genotype ranking across environments. The changes in genotypes' ranking across environments complicate the breeding process by masking genotype superiority and confounding selection.

Parental genotypes such as ICEAP 87105 and ICEAP 01285 and crosses such as ICEAP 01285 × ICEAP 00554, ICEAP 01285 × ICEAP MWPLR 22 and ICEAP 01285 × ICEAP 87105 had early maturity period and would be selected as sources of genes for early flowering and maturity. Sub-Saharan Africa (SSA) is challenged by short-season duration due to climate change (Srivastava and Saxena 2019). Identifying genotypes with the ability to escape terminal drought stress would contribute to the increasing productivity of pigeonpea. Furthermore, development of early maturing pigeonpea lines that are photo-period insensitive could allow for pigeonpea expansion to wider latitudes and altitudes and provide alternative cropping systems (Vales *et al.* 2012). Parental lines such as MWPLR 16 and Sauma and hybrids such as TZA 5582 × MWPLR 22 and Mwayiwathualimi × MWPLR 22 expressed higher grain yield. Similarly, parental lines such as TZA 5582, Sauma, and hybrids such as TZA 5582 × MWPLR 14, Mwayiwathualimi × ICEAP 00554, and Sauma × MWPLR 14 had desirable resistance to FW (Table 5.7). These genotypes are useful sources of genetic variation for FW resistance breeding in pigeonpea. Changaya *et al.* (2012) and Kimaro *et al.* (2020) identified some sources of FW resistance for pigeonpea. FW disease is a destructive disease in most pigeonpea growing countries in the SSA (Gwata *et al.* 2006). The disease causes a yield loss of up to 100% (Reddy *et al.* 2012); hence developing FW resistance is imperative for SSA. Parental lines with a high mean performance for agronomic traits and FW resistance would be suitable for selection to maintain pure breeding lines. On the other hand, crosses that exhibit desirable agronomic performance and disease resistance would be selected for developing breeding populations.

5.4.2 Combining Ability Effects of Parents and Progenies

There existed significant variation in GCA_{male} and GCA_{female} effects for assessed agronomic traits and FW resistance. The observed variability indicates differential heritability and hence response to selection. The SCA_{cross} effects were diverse among the crosses indicating that allelic interactions are dependent on the parent's genetic constitution. Favourable interaction between alleles from two different parents in a cross results in desirable and high SCA_{cross} effects. The significant GCA_{male}, GCA_{female} and SCA_{cross} effects exhibited by most traits show that these traits were under the control of additive and non-additive genes. However, the ability to transfer genes, the allelic interaction in a cross, and the effects of additive and non-additive genes on DTF, DTM, PH, and HSWT were influenced by environmental variance, which is concomitant with the quantitative nature of agronomic traits and FW resistance. Mazer and Gorchov (1996) found that the environment was integral in conditioning trait heritability to offspring. The environment had no impact on the GCA_{male} effects. This implies that selection should focus on identifying specifically adapted female rather than male parental lines.

Parental lines exhibiting negative GCA effects for days to flowering, maturity, and plant height are required for breeding for earliness. In this regard, parents ICEAP 01285 and ICEAP 87105 exhibited desirable GCA effects for DTF, DTM, and PH (Table 5.10). For yield and HSWT, NPP, the number of branches, and number of seeds per plant, parents with positive GCA effects are important since they transmit additive genes during selection (Saroj *et al.* 2014). Two parents, Sauma and MWPLR 16, were good general combiners for GYD and HSWT and therefore they can be utilized in breeding for high yielding pigeonpea genotypes. The GCA effects for FW revealed that TZA 5582, Mwayiwathualimi and Sauma were good general combiners, responsible for increased FW resistance, hence can be utilized in breeding for FW resistance in pigeonpea.

Specific combining ability is the deviation from the performance predicted based on GCA, which is controlled by non-additive gene action (Allard 1960). In plant breeding, crosses with high SCA arising from good general combiners are important for selecting transgressive segregants in subsequent generations (Rieseberg *et al.* 1999). Furthermore, Agaba *et al.* (2017) reported that crosses with superior performance over their parents lead enhanced genetic gain achievable during the development of the progenies through favourable recombination. Such genetic gain would need to be fixed in subsequent generations by identifying crosses that may potentially

provide transgressive individuals. The emergence of such transgressive segregants in breeding populations may be attributed to the unique recombination events where the diverse, desirable alleles from two parents combined in a single individual. Such incidences arise due to the presence of additive, epistatic, or complementary gene action. Sometimes, it could be attributed to chromosomal rearrangements, mobilization of transposable elements, or DNA-methylation (Rieseberg *et al.* 1999). A study by Srivastava and Saxena (2019) reported a transgressive segregant pigeonpea line that matured in 90 days, which was nicknamed "Super Early". Similar findings were reported by Ajay *et al.* (2014), who reported transgressive segregants in three pigeonpea crosses for yield and related traits. For traits such as days to flowering, days to maturity, plant height, and FW susceptibility, the hybrids with negative SCA effects are desirable. The negative SCA effects observed in the hybrids, TZA 5582 × 00554, Mwayiwathualimi × MWPLR 22, and Mwayiwathualimi × MWPLR 14, indicate the capacity of the FW resistant parents such as Mwayiwathualimi to transmit their characters to progenies in cross combinations. For instance, Mwayiwathualimi and crosses such as, Mwayiwathualimi × MWPLR 22 and Mwayiwathualimi × MWPLR 14 could be the best general and specific combiners for FW resistance in pigeonpea. High SCA effects for yield and related traits are desirable as they transmit additive genes during selection (Dholariya *et al.* 2014). In this study, high SCA effects recorded in Sauma × MWPLR 16, TZA 5582 × MWPLR 22 and Mwayiwathualimi × MWPLR 22 hybrids, indicate that the parents are good combiners hence their crosses are sources of genes for improving yield and related traits. In order to improve early maturity, *Fusarium* wilt resistance and high yield in pigeonpea, hybrids such as TZA 5582 × ICEAP 00554, TZA 5582 × MWPLR 14, Mwayiwathualimi × MWPLR 22 and ICEAP 01285 × MWPLR 14 are recommended to develop breeding populations.

5.4.3 Variance Components and Heritability Estimates

The high σ^2 GCA for DTF, DTM, and PH compared to their respective σ^2 SCA, shows the prevalence of additive gene action governing the respective traits. In contrast, the lower σ^2 GCA for GYD, HSWT, and FW shows that the traits were under the control of non-additive gene action since their σ^2 SCA were larger. Hence, the environment would have had a significant influence on GYD, HSWT, and *Fusarium* wilt incidence compared to DTF, DTM, and PH.

Similar results have been previously reported (Changaya *et al.* 2012, Saroj *et al.* 2013, Pandey *et al.* 2014, Kimaro *et al.* 2020). However, Tikle *et al.* (2016) reported both additive and non-additive gene effects for DTM, number of branches per plant, number of pods per plant, and grain yield in pigeonpea, with a predominance of non-additive gene action affecting the inheritance of seed yield and its components. Furthermore, Mayomba (2018), reported a higher magnitude of additive variance (σ^2 A) than dominance variance (σ^2 D) in days to 50% flowering, days to 75% maturity and *Fusarium* wilt incidence, signifying the presence of additive gene action for the inheritance of these traits. On the other hand, a higher magnitude of dominance variance (σ^2 D) in grain yield, 100 seed weight and *Fusarium* wilt resistance indicate that dominance gene action is prevalent. Traits such as days to 50% flowering, days to 75% maturity, and plant height controlled by additive gene action can be selected in early generations for improvement. In contrast, traits such as 100-seed weight, FW resistance, and grain yield controlled by dominance gene effects should be selected in advanced generations using recurrent breeding methods. In a similar study, Saroj *et al.* (2013) reported higher σ^2 A than σ^2 D for DTM and HSWT and concluded that these traits were controlled by additive genes.

Heritability is a measure of the proportion of variance observed among genotypes due to genetic differences and is expressed in a broad and narrow sense (Oppong-Sekyere *et al.* 2019). The broad sense heritability (H^2) is responsible for providing the proportion of genetic variance present in the phenotypic or total variance. Higher broad sense estimates may be caused by greater additive genetic variance, lower environmental variance, and the environment (Acquaah 2012). On the other hand, narrow sense heritability estimates show the proportion of a trait transmitted from parents to their progenies. Dabholkar (1999) classified heritability estimate as low (5-10%), medium (10 – 30%) and high (> 30%). The broad-sense heritability estimates obtained for the studied traits were high (> 30%) (Table 5.12), signifying a small environmental influence on the traits, and high breeding values. This means that there is a greater additive genetic effect, which is paramount for crop improvement. The high narrow sense estimate (> 0.60) observed for DTF, DTM, GYD and HSWT signified the presence of additive gene effects and suggest a high level of gene transmission from parents to the progenies. Hence, a strong response to selection is expected in the early generations. Oppong-Sekyere *et al.* (2019) reported high narrow-sense heritability estimates for grain yield (90.0%), plant height (76.0%), days to 50% flowering (91.0%), for groundnut genotypes. Similarly, Techale *et al.* (2013) reported high

heritability estimates for plant height, 100 seed weight, and harvest count for a pigeonpea population. High heritability values for seed yield for pigeonpea were also reported by Venkateswarlu (2006). Contrary to the present findings, Mwale *et al.* (2017) reported low heritability estimates for grain yield and 100 seed weight in cowpea. The lower narrow sense heritability for the *Fusarium* wilt susceptibility (Table 5.12) signifies non-additive gene action and suggests that total variance was largely influenced by the environment, hence the need to delay selection to later generations.

5.5 Conclusions

The present study revealed that tested germplasm is a vital source of genetic variation for breeding for early maturity, *Fusarium* wilt resistance and high yielding cultivars. The most promising parents for early maturing were ICEAP 01285 and ICEAP 87105 due to their significantly higher GCA effects for days to flowering, days to maturity, and plant height. The best high yielding parents were Sauma and MWPLR 16 due to their significant positive GCA effects. In addition, parents Sauma, TZA 5582, and Mwayiwathualimi were the best combiners for *Fusarium* wilt resistance due to their significantly higher negative GCA effect for FW susceptibility. Furthermore, crosses, TZA 5582 × MWPLR 22, TZA 5582 × MWPLR 14 andMwayiwathualimi × MWPLR 22 were identified as the best specific combiners that combined early maturity, *Fusarium* wilt resistance, and high grain yield. The study further revealed that non-additive gene effects had a greater influence on the grain yield, 100 seed weight, and *Fusarium* wilt resistance, while additive gene action had a more significant influence on days to 50% flowering, days to 75% maturity, and plant height. This implies that selection for days to 50% flowering, days to 75% maturity, and plant height can be carried out in the early generation using pedigree-breeding method while selection for grain yield, 100 seed weight, and *Fusarium* wilt resistance can be delayed to a latter generation. Recurrent selection method is the most suitable option for traits with non-additive gene effects.

5.6 References

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GENERAL OVERVIEW AND IMPLICATIONS OF THE STUDY

Introduction and objectives of the study

Pigeonpea [*Cajanus cajan* (L.) Millspaugh, 2n=2x=22] is one of the most important grain legume crops in Malawi. It is a key food security crop and the grain is marketed in local, regional and international outlets. In addition, pigeonpea biomass is a source of fuelwood, and plant parts offer medicinal benefits to the smallholder farmers. Malawi is one of the main producers of pigeonpea in Africa. The southern region is the key producer of pigeonpea in the country. The mean grain yield of the crop is < 1,000 kg ha^{-1} in Africa, including Malawi, which is below the potential yield of the crop of 2,500 kg ha^{-1}. The low yield levels are attributable to various constraints, including a lack of high yielding and early maturing varieties, *Fusarium* wilt disease (*Fusarium udum* Butler) and insect pests such as pod borers *(Helicoverpa armigera* Hubner). *Fusarium* wilt causes up to 100% yield losses in susceptible cultivars. Hence, this study aimed to contribute to food security in Malawi through breeding high performing and farmer-preferred pigeonpea varieties. This section highlights the study objectives, the summary of research findings and the implications of the findings for demand-led pigeonpea breeding. The specific objectives were:

- To determine pigeonpea production constraints and farmer-preferred traits in Malawi to guide future breeding;
- To determine the genetic diversity among pigeonpea germplasm collections using agro-morphological traits to select genetically distinct lines for breeding;
- To determine the genetic diversity among pigeonpea germplasm using the single nucleotide polymorphism (SNP) markers to select genetically distinct lines for breeding;
- To determine the combining ability and gene action controlling early maturity, yield and resistance to *Fusarium* wilt, and to select the best parents and families for further breeding.

Research findings in brief

Farmers' perceptions of the primary constraints to pigeonpea production in Malawi, and their variety choice and preferred traits: implications for variety design

A participatory rural appraisal study was conducted in four main pigeonpea-growing districts in southern Malawi (Chiradzulu, Mulanje, Thyolo and Zomba) using a semi-structured questionnaire, transect walks and focus group discussions (FGDs).The main findings of the study were:

- Pigeonpea is the second most important crop after maize, the leading food security crop in the Southern Malawi.
- Maize/pigeonpea intercropping was the common practice among households, with the perception that it provided several benefits such as nutrient build up, maintenance of soil fertility, efficient utilization of the available resources, and weed and pest control.
- A landrace pigeonpea variety, 'Mthawajuni', was preferred by farmers due to its positive attributes such as good taste, early to medium maturity, short cooking time and resistance to pod borer *(H. armigera*).
- Pigeonpea trait preference was dependent on gender, with female respondents preferring short cooking, early maturity, long storage and pest resistance, whereas men preferred high yielding, large seeds, cream seed colour and disease resistance.
- Pod borer, *Fusarium* wilt disease, the low yields of their existing landrace varieties, drought, and unreliable market prices were the leading challenges affecting pigeonpea production in southern Malawi.

Phenotypic divergence and grain yield stability analysis in pigeonpea germplasm accessions

Eighty-one pigeonpea genotypes were evaluated in six environments in Malawi using a 9 × 9 alpha-lattice design with two replications. The main findings of the study were:

- Significant genotype variation were recorded for qualitative traits including flower colour, flower streak pattern, pod colour, seed coat colour pattern, seed coat main colour, seed shape and seed eye colour.
- All assessed quantitative traits were significantly affected by genotype × environment interaction effects except the number of seeds per pod.

- Genotypes MWPLR 14, ICEAP 01170, ICEAP 871091 and ICEAP 01285 were identified as early maturing varieties, maturing in 125 to 137 days;
- Grain yield was positively and significantly correlated with days to flowering (DTF) ($r=0.23$, $p<0.01$), number of pods per plant ($r=0.35$, $p<0.01$) and 100 seed weight ($r=0.50$, $p<0.01$), suggesting the usefulness of these traits for selection to enhance grain yield improvement when assessing pigeonpea populations;
- The principal component analysis identified three principal components (PCs) that accounted for 57.7% of the total variation;
- The most important traits that reliably discriminated between the test genotypes were days to 50% flowering, days to 75% maturity, number of primary and secondary branches, 100 seed weight and grain yield;
- Genotype, environment and genotype × environment interaction (GEI) accounted for 16.4, 33.5 and 49.6% to the total variation for quantitative traits;
- The GGE analysis delineated the test environments into three mega-environments based on site and seasonal variability;
- The AMMI and GGE biplot analysis revealed that MWPLR 14 (G51), MWPLR 24 (G26) and ICEAP 01155 (G27) were the most yield stable genotypes across environments, while MWPLR 14, TZA 5582 and MWPLR 4 were the highest yielding genotypes across environments.

Genetic diversity and population structure analyses of pigeonpea genotypes using morphological and SNP markers

The genetic diversity and population structure present among 81 pigeonpea genotypes were done using 24 morphological traits and 4,122 single nucleotide polymorphism (SNP) markers. The main findings of the study were:

- Significant ($P<0.001$) genotype × environment interaction effects for grain yield (GYD), days to 50% flowering (DTF), days to 75% maturity (DTM), plant height (PH), number of primary branches per plant (NPB), number of pods per plant (NPP), number of racemes per plant (NRP), 100 seed weight (HSWT), and number of secondary branches per plant (NSB);
- The principal component analysis identified eight components that explained 67.57% of the total phenotypic variation. Traits including DTF, DTM, growth habit (GH), second flower colour (FSC), pod colour, seed shape (SSH), HSWT, and GYD were identified as the most important for discriminating among the test genotypes;

- The phenotypic diversity assessment using morphological attributes grouped the genotypes into three distinct clusters;
- The mean gene diversity and polymorphic information content were 0.14 and 0.11, respectively, suggesting moderately low genetic differentiation among the genotypes;
- The genotypes were delineated into three heterotic groups based on the population structure using a combined analysis based on the phenotypic and genotypic data, suggesting the possibility of creating unique breeding populations through targeted crosses of parents from discrete heterotic groups.

Combining ability, gene action and heritability for agronomic traits and *Fusarium* wilt (*Fusarium udum* Butler) resistance in pigeonpea

Ten selected parental lines were crossed using a factorial mating design and 25 progenies were successfully developed. The parents and progenies were field evaluated in two locations (Chitedze and Makoka Agricultural Research Stations) in Malawi using a 7 × 5 alpha lattice design with two replications. Also, the test genotypes were evaluated for FW resistance through the root dip inoculation technique. The main findings of the study were:

- Non-additive gene effects were more significant for the inheritance of GYD, HSWT and *Fusarium* wilt resistance genes that would be improved through recurrent or pure line selection in the advanced inbred line generations;
- Additive gene effects were more significant for the inheritance of DTF, DTM and PH that would be improved through early generation selection;
- Parental genotypes ICEAP 01285, TZA 5582, Mwayiwathualimi, Sauma, and MWPLR 16 exhibited desirable general combining ability (GCA) effects for DTF, DTM, FW, and GYD;
- The families TZA 5582 × ICEAP 00554, TZA 5582 × MWPLR 14, Mwayiwathualimi × MWPLR 22, and ICEAP 01285 × MWPLR 14 showed favourable specific combining ability (SCA) effects for DTM, GYD, and FW resistance, making them suitable families to develop early maturing and high yielding varieties with FW resistance.

Implications of the research findings for breeding for yield, earliness and resistance to *Fusarium* wilt (*Fusarium udum* Butler) in pigeonpea

- From the participatory rural appraisal (PRA), breeding priorities were identified based on the farmer preferred traits and farmers perceived constraints to pigeonpea production.

Involvement of farmers in the process of cultivar development is important as it enhance the adoption of the new improved cultivars. Farmer preference should be considered in future pigeonpea breeding.

- The phenotypic and genetic variation existed among the genotypes using morphological and SNP markers gives an opportunity for the improvement of important traits in pigeonpea. Crossing of divergent parents enable the selection of superior progenies.
- Non-additive genetic effects observed in controlling GYD, HSWT and FW resistance providing an opportunity for improvement of the population through recurrent or pure line selection in the advanced inbred line generations. In addition, the dominant gene action present in the GYD, HSWT and FW resistance suggests that hybrid breeding is a better option to improve pigeonpea production.
- Parental genotypes ICEAP 01285, TZA 5582, Mwayiwathualimi, Sauma, and MWPLR 16 exhibited desirable general combining ability (GCA) effects for DTF, DTM, FW, and GYD. The parents are recommended in pigeonpea breeding for early maturing, high yield and *Fusarium* wilt resistance.
- The families TZA 5582 × ICEAP 00554, TZA 5582 × MWPLR 14, Mwayiwathualimi × MWPLR 22, and ICEAP 01285 × MWPLR 14 showed favourable specific combining ability (SCA) effects for DTM, GYD, and FW resistance indicating that they are suitable families to develop early maturing and high yielding varieties with FW resistance pigeonpea varieties.
- The families developed from this study should be further evaluated in multi-environment for morphological traits and under controlled environments for *Fusarium* wilt resistance to select the best performing and stable families for variety release in Malawi.

APPENDICES

Appendix 1. Status of pigeonpea production among smallholder farmers in Malawi: household questionnaire (2017)

Introductory Remark:

Dear Sir/Madam, I work for the Ministry of Agriculture, Irrigation and Water Development in the Department of Agricultural Research Services based at Chitedze Research Station. I am conducting out this survey to study the production constraints and farmers' preferred quality traits of pigeonpea so as to improve the varieties. Your response to these questions would remain anonymous. Thank you for your kind cooperation.

MODULE 1: IDENTIFICATION

Household Identification	Code
1.District	
2.Region	
3.EPA	
4.Section	
6.Village Name	
7. Name of VH T/A	
7.Name of household head	
8.Sex of household head **1=Male** **0=Female**	
9.Name of the respondent	
10. Sex of the respondent **1= Male** **0=Female**	

Interview details	Code
18. Date of interview	/ /2017
19. Time started (24 HR)	
20. Name of Enumerator	
21. Name of supervisor	
22. Household type	
[1]Dual(male and female spouses)	
[2]Female headed with another adult male decision maker	
[3]Male headed with another adult female decision maker	
[4]Female headed ,without any adult male decision maker in the household	
[5]Male headed ,without any adult female decision maker in the household	
[6]Male household with more than one wife	
[7]Child headed HH (specify)	
23. Education level of the HH **1= Haven't attended school** **2= Attended adult school (School yakwacha)** **3= Primary (Std 5 and below)** **4= Primary (above Std 5)**	

11. Marital status **1= Married** **0= Single**	
12. Age of the HH **1= 20 yrs and below** **2= 21-35 yrs** **3= 36-49 yrs** **4= 50-65 yrs** **5= Above 65 yrs**	
12. Name of the respondent's spouse	
Homestead GPS Reading	
13. Way point Number	
14. Latitude(South)	
15. Longitude(East)	
16. Elevation (meter above sea level)	
17. Measurement error (±m)	

5= Secondary school **6=Tertiary**
24. Does your household normally grow pigeon peas? **1=Yes** **0=No**

MODULE 2.CURRENT HOUSEHOLD COMPOSITION AND CHARACTERISTICS

PART A. Household size and Composition

A0. How many people currently belong to this household?**.....**

How many of these household members are……		Number (total across A01-A05 must equal A0)
A01	0-5 years old in this household?	
A02	More than 5 and less than 15 years old Enrolled at school?	
A03	More than 5 and less than 15 years old not Enrolled at school?	
A04	More than 15 and less than 65 years?	
A05	Aged 65 and older?	

MODULE 3: LAND HOLDING AND LAND CROPPNG IN THE 2017/2018 AGRICULTURAL SEASON

PART A: Total amount of land and cropping area

Total amount of land owned by your household in the 2016/2017 agricultural year?		**Total amount of land cropped in the 2016/2017 agricultural season?**		**Using the same units……how much of the area cropped in 2016/2017 season was** ***(sum of these should equal A3a)***			
Area*	**Unit CODE 1**	**Area**	**Unit CODE 1**	**Owned?**	**Rented in?**	**Borrowed?**	**Shared?**
A1a	**A2b**	**A3a**	**A4b**	**A5a**	**A5b**	**A5c**	**A5d**

*In case of land fragmentation of owned land use one row per parcel of land.

CODE 1

1=Ha

2=Square meter

3=Acre

4=Yards **99=Other** **(specify)……..**

PART B: FIELD LEVEL DATA ON PIGEONPEA AREA AND PRODUCTION DURING THE LAST PIGEONPEA SEASON 2016/17

Did your household grow pigeonpea this past growing season (2015/2016)? **1=Yes** **0=No =>B3**	**In how many fields did you or any member of your household grow pigeonpea this last season?**<*Enumerator: define a field as a continuous piece of land*>	**What was the main reason for not growing?** **CODE 1**	**Did you have the following problems more in the last season compared to the previous two years?** 1=Yes 0=No 99=Don't know			
			Insect pests	**Diseases**	**Drought/dry spell**	**Too much Rain**
B1	**B2**	**B3**	**B4.1**	**B4.2**	**B4.3**	**B4.4**

CODE 1		
1=Seed not available	4=Low yielding varieties	7=Varieities susceptible to diseases/pests
2=Lack of cash to buy seed	5=Low prices received at the market	99=Other (specify)……
3=Lack of access of credit(for seed)	6=No markets	

Enumerator: Please read the following to the farmer: Now I would like to ask you about the pigeon pea area, input use and production on the field reported above. Let's start with the biggest field where pigeon peas were planted this season.
Enumerator: *Please use one row for each field on which the farmer grew pigeon peas and start with the biggest field. The number of rows to be filled should equal the answer in**B2***

Field ID	**Who in the HH managed this field?** **CODE 1**	**What is the size (area) of this field?**		**What is the […..] on this field?**			**Was pigeon pea intercropped?** **1=Yes** **0=No**	**If Yes what crop was intercropped with Pigeonpea** (If 0 in B8.1 fill NA)	**If Yes, proportion of this field planted to pigeonpea?** (If 0 in B8.1 fill NA) **CODE 7**
		Area	**Unit** **CODE 2**	**Slope** **CODE 3**	**Soil quality** **CODE 4**	**Land tenure** **CODE 5**			

								CODE 6	
B5	**B6**	**B7.1**	**B7.2**	**B8.1**	**B8.2**	**B8.3**	**B9.1**	**B9.2**	**B9.3**
F1									
F2									
F3									
F4									
F5									

CODE 1	CODE 2	CODE 3	CODE 4	CODE 5	CODE 6	CODE 7
1=Household head 2=spouse 3=HH head & spouse 4=Son/daughter 5=Other (sp)……	1=Hectares 2=Square meter 3=Acre 4=Yards 99=Others (sp)……	1=Flat 2=Medium 3=Steep	1=Sand 2=Sandy-loam 3=Clay 97=Don't know 99=Other (sp)……	1=Owned 2=Rented in 3=Shared 4=Borrowed 5=Gvt land 99=Other (sp)……	1=Maize 2=Sorghum 3=Cassava 4=Groundnuts 5=Soybean 6=Common beans	1=< a quarter 2=A quarter 3=Between a quarter & half 4=More than half

PART C: QUANTITY OF PIGEON PEAS HARVESTED THIS LAST SEASON AND VARIETY KNOWLEDGE

SECTION 1: Pigeonpea harvested green and dry stage this season. ***Enumerator Read the following***; now I would like to ask you about the quantity of pigeonpea harvested, sold and other uses. First let's start with pigeonpea harvested in each field where pigeonpea were planted… **(Instructions:** *Fields should be recorded in the same order as in B4***)**

Field ID	Did you harvest any pigeon peas from this field as green pods? 1=Yes 0=No (If No go to **C05.1**)	Approximately, what proportion of pigeon pea planted in this field was harvested as green pods? CODE 1	What did you do with the harvested green pods? CODE 2	What was the total quantity of pigeon pea (as dry grain) harvested from this field?		Enumerator: Add C05.1 and then write total quantity of pigeon pea harvested across all fields (if in the same unit)
				Quantity	Unit CODE 3	
C01	**C02**	**C03**	**C04**	**C05.1**	**C05.5**	**C06**
F1						
F2						
F3						

F4						
F5						

CODE 1	**CODE 2**	**CODE 3**
1=Quarter or less	1= Sold all	1=Kilograms
2=Between quarter & half	2=Used all for home consumption	2=Basin(5kgs)
3=Half	3=Partly sold, partly consumed	99=Other (specify)……
4=More than half	99=Other (specify)……	

SECTION 2A: PIGEONPEA VARIETY KNOWLEDGE-Varieties currently grown.

Variety ID	**Name of the Variety planted this last season***(as reported by the farmer)*	**Do you plan to increase, decrease, not to change or not grow this next season on your farm?** **CODE 1**	**How would you rate the yield of this variety?** (In your assessment and experience) **CODE 2**	**Where did you get the seed of the pigeonpea you planted?** **CODE 3**	**What do you use this variety of pigeon pea for?** **CODE 4**	**What do you like most about this variety?** *(List up to three features in descending order of importance)* **CODE 5**			**What don't you like about this variety?** *(List up to three features in the descending order of importance)* **CODE 6**		
C07	**C08**	**C09**	**C10**	**C11**	**C12**	**C13.1**	**C13.2**	**C13.3**	**C14.1**	**C14.2**	**C14.3**
V1											
V2											
V3											
V4											
V5											
V6											
V7											

V8											
V9											

CODE 1	**CODE 2**	**CODE 3**	**CODE 4**	**CODE 5**	**CODE 6**
1=Increase 2=Decrease 3=Not change 4=Not grow 97=Don't know	1=High 2=Moderately high 3=Medium 4=Low 5=Very low 97=Don't know/can't tell	1=Market 2=Neighbour/ relative 3=NGO 4. Extension office 5= Admarc 6= Agrodealer 9=Other (specify)	1=For home consumption of green pods 2=For home consumption of dry grain 3=For both green pods and dry grain consumption 4=For selling dry grain in the market 5=For both home consumption and selling 6=For selling as quality declared seed 7=Green pods for home consumption and sell 99=other (specify)	1=Taste 2=Colour, size and shape 3=Fast cooking 4=Early maturing 5=Resistant to pests and diseases 6=High yielding 7=Good price premium 8=Sells faster 9=Other cooking/processing quality specify)……. 99=Other agronomic characteristics (specify)……	1=Taste 2=Colour, size and shape 3=Slow cooking 4=Late maturing 5=Susceptible to pests and diseases 6=Low yielding 7=Low price premium 8=Does not sell faster 9=Other cooking/processing quality (specify)…… 99=Other agronomic characteristics (specify)……

SECTION 2B:PIGEONPEA VARIETY KNOWLEDGE-Varieties not grown: **Enumerator:** Please ask the farmer to give you names of **all improved pigeonpea varieties** she/he knows or has heard of or has grown in the past **BUT IS NOT CURRENTLY** growing.

Variety code	Name of the improved pigeonpea variety you are aware of (write legibly for post coding)	Year when this variety was first known/heard of (YYYY)	Source of information about this variety CODE1	Have you ever grown this variety on you farm? 1=Yes 0=No=>go to B05	If No, main reason why you have never grown this variety? CODE 2	If Yes, year when you first planted this variety (YYYY)	Reason for abandonment CODE 2	What traits can you recommend the breeders to include in new varieties CODE 3
B00	**B01**	**B02**	**B03**	**B04**	**B05**	**B06**	**B07**	**B08**
V01								
V02								
V03								
V04								
V05								
V06								
V07								

CODE 1	CODE 2	CODE 3
1=Agricultural office	1= Seed not available	1= High yielding
2=Farmer Cooperatives/union	2=Lack of cash to buy seed	2= Large seeded
3=Farmer groups/association	3=Lack of credit for seed	3=Cream color
4=Non-Governmental Organisations (NGOs)/Community Based Organisations (CBOs)	4=Variety was susceptible to diseases /pests	4= Disease resistance
5=Research trials/demos i.e.DARS	5=Low yielding variety	5= Pest resistance
6=Another farmer relative	6=Low price received for the seed of this variety	6= Early maturity
7=Another farmer neighbor	7=No market	7= Taste
8=Radio/newspaper/TV	8=Requires high skills	8=Drought tolerant
9=ICRISAT	9=Prefer other varieties	9= Easy to cook
99=Other (specify)……..	10=Variety matured late	10= Longer storability
	99=Other (specify)……	99= Other (specify)

MODULE 4: FARMERS' KNOWLEDGE AND PERCEPTION ON PIGEONPEA DISEASES AND DISEASES

PART A: AWARENESS OF PIGEONPEA FIELD DISEASES. Enumerator: Read this to the respondent; now I would like to take you through the pigeonpea diseases.

ISSUE	CODE	RESPONSE
A01.Are you aware of the diseases that attack pigeonpea?	**1=Yes** **0=No**	
A02.If yes ,mention the names of pigeonpea field pests you are aware of*(up to 4 diseases)*		
A03. Of these pigeonpea field pests which one is the most devastating?		
A04.Have you experienced any problem of pigeonpea diseases in your field?	**1=Yes** **0=No**	
A05.If yes to A03, what problems have you experienced in your household as a result of pigeonpea disease attack?	1=Defoliation 2=Yellowing 3= Stuntedness 4=Wilting 5=Dyeing 6=Poor pod setting 99=Other (specify)……	
A06. For how long have you being experiencing these problems?		☐ **Years**
A07. At what stage of growth do these diseases usually affect your pigeon peas?	1= Emergence Stage 2=Vegetative stage	

	3=Flowering stage 4=Podding Stage	
A08. How much grain yield loss was experienced in this past season 2016/2017 (% Yield reduction)	1= < Quarter 2=Quarter 3=Half 4=Three quarter 5= >Three quarter 99=Other (specify)……..	
ISSUE	**CODE**	**RESPONSE**
A09. How has your household being affected by pigeonpea diseases	1= Reduced yield 2= Reduced grain consumption 3=No seed for the next season 4= Reduced income 99=Other (specify)…….	
A10. What management option(s)/practice(s) do you use to control pigeonpea diseases?	1=Biological control 2=Chemical control 3= Resistant varieties 4. Cultural methods 99=Other (specify)…….	
A11. What cropping systems/practices are mostly used in your field?	1=Monocropping 2=Mixed cropping 3=Continuous cultivation 4=Crop rotation 5=Zero tillage 99=Other (specify)…….	
A12. How do you rate most of the pigeonpea fields in terms of disease incidence in your village?	1= < Quarter of the fields 2=Quarter of the fields 3=Half of the fields	

	4=Three quarters of the fields 5= >Three quarter of the fields	

PART B: AWARENESS OF PIGEONPEA FIELD PESTS. Enumerator: Read this to the respondent; now I would like to take you through the pigeon pea field pests.

ISSUE	CODE	RESPONSE
A01.Are you aware of the pests that attack pigeonpea in the field?	**1=Yes** **0=No**	
A02.If yes ,mention the names of pigeonpea field pests you are aware of*(up to 4 pests)*		
A03. Of these pigeonpea field pests which one is the most devastating?		
A04.Have you experienced any problem of pigeonpea field pests in your field?	**1=Yes** **0=No**	
A05.If yes to A03, what problems have you experienced in your household as a result of pigeonpea field pest attack?	1=Defoliation 2=Root logging 3=Holed pods 4=Empty pods 5=Damaged flowers 6=Weeviled stems 99=Other (specify)……	
A06. For how long have you being experiencing these problems?		☐ **Years**
A07. At what stage of growth do these pests usually affect your pigeonpeas?	1= Emergence Stage 2=Vegetative stage	

	3=Flowering stage 4=Podding Stage	
A08. How much grain yield loss was experienced in this past season 2016/2017 (% Yield reduction)	1= < Quarter 2=Quarter 3=Half 4=Three quarter 5= >Three quarter 99=Other (specify)……..	
ISSUE	**CODE**	**RESPONSE**
A09. How has your household been affected by pigeonpea field pests	1= Reduced income 2= Low protein consumption 3=No seed for the next season 99=Other (specify)…….	
A10. What cropping system(s)/practice(s) do you use in pigeonpea production?	1=Monocropping 2=Mixed cropping 3=Continuous cultivation 4=Crop rotation 5=Zero tillage 99=Other (specify)…….	
A11. What cropping systems/practices promote pigeonpea pest infestation in your field?	1=Monocropping 2=Mixed cropping 3=Continuous cultivation 4=Crop rotation 5=Zero tillage 99=Other (specify)…….	
A12. What main control measure do you use to control pests in your pigeonpea field?	1=Chemical control 2=Cultural control 3=Physical control	

	4=Biological control 99=Other (specify)……..	
A13. How do you rate most of the pigeonpea fields in terms of pest infestation in your village?	1= < Quarter of the fields 2=Quarter of the fields 3=Half of the fields 4=Three quarters of the fields 5= >Three quarter of the fields	

MODULE 5: UTILISATION AND MARKET INFORMATION OF PIGEON PEA PRODUCTION THIS LAST SEASON (2017/2018)

SECTION 1: UTILISATION

From the **total quantity** of pigeon peas harvested dry in all fields this last season, how much has been [.....] (Report in the same units as in **C05.5**)		**How much of the total quantity of dry pigeonpea you have today do you plan to [.....]** (Report in the same units as in **C05.5**)				**Is there a market place where agricultural produce are sold in your village?** **1=Yes** **0=No**
Lost due to pests	**Sold**	**Keep as seed for planting in the next season?**	**Keep as food grain for home consumption?**	**Sell in the future?**	**Given out as gift to friends/relatives?**	
C06.1	**C06.2**	**C07.1**	**C07.2**	**C07.3**	**C07.4**	**C08**

Distance to the local market from your residence (Report both if possible)*		**What is the main means of transportation do you use to get to this local market?** **CODE 1**	**On average what does a single trip cost (MK/person) using this means of transport?** *(Write zero if nothing)* *(Write 97 if don't know)*	**Distance (km) from the village center to the main all season road?** *(Write zero if nothing)* *(Write 97 if don't know)*	**Distance (km) from the village to main district town?** *(Write zero if nothing)* *(Write 97 if don't know)*
Km	**Walking minutes**				
C09.1	**C09.2**	**C10**	**C11**	**C12**	**C13**

**During normal walking, a person would take 15minutes to walk 1km*

CODE 1

1=Back/head load/walking4=Minibus

2=Bicycle5=Hired truck99=Other (specify……

3=Motorbike6=Oxcart

SECTION 2: MARKETING OF DRY PIGEONPEA HARVESTED THIS LAST SEASON

C14:Since you harvested your pigeonpea this past season ,how many times have you sold your pigeonpeas?.....(cross check with C06.2)
Enumerator: Fill one row per transaction *(different months, different buyers).The number of rows in the following table should match the response in C14*

Transaction ID	Variety Name (If more than one in the same transaction ,write all)	Market where transaction took place **CODE 2**	Month when sale took place [1-12]	Quantity sold in this transaction **(KG)**	Person in HH responsible for this sale **CODE 3**	Price received (MK/kg)	Who was the buyer **CODE 4**	Relationship to the buyer **CODE 5**	Time taken to get to the market	Mode of Transport **CODE1** (sec.2)	Person transport cost (MK/person)	Crop transport cost (Total) **(MK)**(indicate if hired)
C15	**C16**	**C17**	**C18**	**C19**	**C20**	**C21**	**C22**	**C23**	**C24**	**C25**	**C26**	**C27**
T1												
T2												
T3												
T4												
T5												

CODE 2	**CODE 3**	**CODE 4**	**CODE 5**
1= Farmgate/home	1=Head	1=Farmer group 7=Exporter	1=No relation & not a long term buyer
2=Village market	2=Spouse	2=Farmer union/coop.. 99=Other	2=No relation but a long term buyer
3=Main/district market	3=Spouse & head	specify…….	3=Relative
99=Other (specify)…	4=Son/daughter	3=Consumer/other farmer	4=Friend

	5=Other relative 99=Other (specify)…….	4=Rural assembler 5=Broker/middlemen/retailer 6=Urban wholesaler	5=Money lender 99=Other (specify)……..

MODULE 6: ACCESS TO INSTITUTIONALSERVICES, CREDIT, AND AGRICULTURAL INPUTS

In the past 12 months, did you obtain any agricultural inputs through a government or an NGO program? **1=Yes** **0=No=>4**	**Were any of these inputs applied to pigeon pea production?** *1=Yes* *0=No =>4*	**What pigeon pea inputs did you get through a government or an NGO program?** **CODE 1**	**In these 12 months, did you obtain any agricultural credit for your crop production** *(All crops)?* *1=Yes=>6* *0=No*	**If you did not obtain loan please tell me the two main reasons for this** *(Go to 10 after this)* **CODE 2**		**If yes,did you use part or all of this credit in the pigeon pea crop?** [1]Yes all [2]Yes partly [3]No	**For what purpose did you use this credit for the pigeon pea crop?(list up to three)** [1]Buy fertilizer [2]Buy pesticides [3]Buy seed [4]Buy farm equipment [88]No other purpose=>go to 8 [99]Other (specify)			**Source of credit** **CODE 3**	**How much money (Credit) did you get? (MK)**
				1st	2nd		1st	2nd	3rd		
1	**2**	**3**	**4**	**5.1**	**5.2**	**6**	**7.1**	**7.2**	**7.3**	**8**	**9**

CODE 1	CODE 2	CODE 3
1=Seed 2=Fertilizer 3=Seed & fertilizer 99=Other (specify)…….	1=No need 2=Borrowing is too risky 3=Interest rate is too high 4=Too much paper work/ procedures 5=Expected to be rejected so did not try it 6=No assets for collateral 7=No money lender in this area for this purpose 8=Lenders do not provide the amount needed 9=No credit association available 10=No financial institutions in the region 99=Other (specify)……	1=Money lender 2=Farmer groups/cooperatives 3=Neighbour 4=Microfinance 5=Bank 6=Relative 7=Saving group 99=Other (specify)……..

In general,where do you obtain agriculture-related information(e.g new varieties,production technologies)? *(List up to three sources)* CODE 4			If you have a stress affecting your pigeon pea crop (e.g. pests,diseases),whom do you ask for advice on how to manage this stress? (list the major source) CODE 5	How many times did you get in contact or were visited by agricultural extension worker during the last season 2012/13
First(primary)	Second	Third		(write zero if none)
10.1	10.2	10.3	11	12
CODE 4 1=Never seek information 3=GvtExte.agent 5=Radio/TV/newspaper 2=Other farmer4=NGOs 6=Research Institute 7=Local input stores			CODE 5 1=Never asked for advice 3=Other farmer 5=Gvtext.agent 7=Research institute i.e DARS 2=There is no one I could ask for advise 4=Local input stores 6=NGOs	

Time interview ended …… (HRS)

Thank you very much we value your participation in this study

Appendix 2. Means for 10 quantitative traits of 81 pigeonpea genotypes evaluated in six environments in Malawi

			DTF							DTM						
			Y1			YII			Mean	Y1			Y11			
Genotype code	Pedigree/name	Source	S1	S2	S3	S1	S2	S3		S1	S2	S3	S1	S2	S3	Mean
1	ICEAP 0673/1	ICRISAT	129	111	95	103	99	116	108	177	170	146	144	149	154	157
2	ICEAP 00554	ICRISAT	118	122	102	112	112	115	113	157	167	139	154	153	153	153
3	ICEAP 01164/1	ICRISAT	138	130	107	82	108	91	109	180	172	159	118	153	127	151
4	MWPLR 19	GENEBANK	128	124	109	109	114	107	115	167	189	150	149	166	146	161
5	MWPLR 22	GENEBANK	129	115	112	108	115	114	115	167	162	167	146	156	150	158
6	ICEAP 01170	ICRISAT	64	68	88	101	85	108	85	94	98	128	144	139	148	125
7	ICEAP 01169	ICRISAT	141	118	88	60	85	56	91	190	174	132	92	143	93	137
8	TZA 2439	TARI	123	124	115	111	134	104	118	164	166	159	150	166	144	158
9	MWPLR 9	GENEBANK	131	126	102	112	101	122	115	177	180	157	144	164	159	163
10	MWPLR 6	GENEBANK	122	116	134	123	106	125	121	153	177	199	165	158	169	170
11	MWPLR 17	GENEBANK	133	116	145	109	124	116	124	165	180	205	149	169	152	170
12	TZA 253	TARI	125	116	86	101	102	116	107	159	169	132	141	151	160	152
13	MWPLR 1	GENEBANK	126	117	109	116	107	115	115	167	166	153	145	154	155	156
14	MWPLR 18	GENEBANK	111	127	131	113	131	115	121	148	163	182	151	165	160	161
15	TZA 2464	TARI	126	112	126	117	113	125	120	172	168	171	148	166	172	166
16	ICEAP 00604	ICRISAT	144	113	122	92	109	89	111	192	181	153	125	173	124	158
17	TZA 2509	GENEBANK	147	120	125	109	120	106	121	182	167	174	156	160	147	164
18	ICEAP 01146/1	ICRISAT	134	113	111	125	117	115	119	161	165	161	150	174	156	161
19	MWPLR 11	GENEBANK	125	117	109	111	114	124	116	160	168	152	151	163	164	159
20	TZA 5555	TARI	116	120	129	122	120	127	122	143	163	175	156	160	172	161
21	No. 40	TARI	129	131	141	124	131	132	131	173	191	211	158	176	176	181
22	ICEAP 01150	ICRISAT	122	126	119	117	110	112	118	178	180	166	160	183	153	170
23	MZ2/9	TARI	120	133	117	117	123	112	120	153	186	154	153	73	152	145
24	ICEAP 01172/1	ICRISAT	121	126	116	111	130	126	122	162	186	152	161	173	171	167

Appendix 2. Continued

25	ICEAP 01103/1	ICRISAT	119	100	119	109	92	128	111	149	157	164	143	149	175	156
26	MWPLR 24	GENEBANK	113	87	111	109	96	108	104	146	145	153	146	140	145	146
27	ICEAP 01155	ICRISAT	123	93	115	106	100	115	108	160	153	161	143	154	167	156
28	ICEAP 01180/2	ICRISAT	117	102	123	117	97	117	112	151	156	171	159	159	159	159
29	MWPLR 4	GENEBANK	112	90	111	108	90	111	104	136	154	152	141	150	152	147
30	Kachangu	DARS	100	97	118	128	116	118	113	133	150	164	159	159	164	155
31	Mwayiwathualimi	DARS	114	103	123	111	123	112	114	144	150	171	146	147	164	154
32	MWPLR 8	ICRISAT	125	110	142	118	92	117	117	161	165	193	159	149	161	165
33	ICEAP 01154/2	ICRISAT	140	97	124	128	107	117	119	177	152	170	160	163	157	163
34	Chitedze Pigeonpea 1	DARS	94	92	122	117	92	117	105	125	145	162	160	150	157	150
35	ICEAP 01164/1	ICRISAT	123	107	115	114	87	105	108	157	159	155	141	143	144	149
36	Bangili	TARI	136	101	116	113	142	114	120	167	156	154	151	176	159	160
37	ICEAP 00053	ICRISAT	114	75	131	117	114	118	111	163	139	204	149	143	157	159
38	MWPLR 12	GENEBANK	108	94	119	123	108	120	112	147	139	181	161	164	163	159
39	TZA5463	TARI	113	90	131	85	90	88	99	149	144	195	127	150	122	147
40	MWPLR 5	GENEBANK	125	105	119	121	106	114	115	155	156	162	159	157	156	157
41	MWPLR 15	GENEBANK	100	86	114	90	77	94	93	121	150	163	126	125	136	137
42	ICEAP 87105	ICRISAT	114	102	127	120	102	120	114	145	154	172	164	166	162	160
43	MWPLR 16	GENEBANK	125	105	119	117	105	105	113	177	166	172	156	161	154	164
44	TZA 2496	TARI	110	104	105	115	104	107	107	143	157	145	159	154	144	150
45	TZA 5582	TARI	107	96	91	128	101	124	108	143	158	146	170	153	165	156
46	TZA 5596	TARI	120	94	94	105	94	108	103	164	160	133	140	152	145	149
47	Chitedze Pigeonpea 2	DARS	121	107	94	110	102	119	108	153	164	157	148	157	163	157
48	MWPLR 7	GENEBANK	114	98	111	116	98	125	110	143	158	151	158	158	173	157
49	Babati	TARI	120	106	99	81	106	107	103	163	158	153	120	158	148	150
50	TZA 5557	TARI	117	77	107	116	77	115	101	141	136	156	155	137	149	145
51	MWPLR 14	ICRISAT	63	65	64	87	67	98	74	95	105	102	127	116	132	113
52	ICEAP 01101/2	ICRISAT	85	82	133	128	102	114	107	111	141	162	166	164	155	149
53	TZA 2456	TARI	65	67	142	120	136	130	110	100	99	205	155	180	175	152
54	TZA 5464	TARI	101	63	137	112	106	117	106	139	99	189	152	140	156	146

Appendix 2. Continued

55	TZA 5463	TARI	102	70	101	119	90	131	102	137	105	137	164	138	177	143
56	ICEAP 01285	ICRISAT	62	64	118	100	74	107	87	98	103	165	144	136	152	133
57	MWPLR 25	GENEBANK	118	72	124	116	114	113	109	157	111	171	152	156	156	150
58	ICEAP 87091	ICRISAT	123	91	107	78	73	85	92	152	144	147	120	113	118	132
59	TZA 2692	TARI	138	88	125	108	97	118	112	186	154	171	148	150	168	163
60	TZA 2807	TARI	115	97	119	113	97	116	109	148	155	157	150	146	154	151
61	ICEAP 00068	ICRISAT	114	105	100	115	105	118	109	156	155	146	155	152	160	154
62	TZA 2785	TARI	118	88	106	95	77	92	96	136	147	157	134	128	134	139
63	MWPLR 10	GENEBANK	89	92	142	123	92	120	110	119	139	197	168	142	171	156
64	ICEAP 00612	ICRISAT	123	101	136	118	115	121	119	165	166	187	153	169	160	167
65	MWPLR 21	GENEBANK	113	105	122	91	113	90	105	148	143	172	129	162	128	147
66	TZA 2514	TARI	120	95	115	116	108	116	111	155	151	170	154	158	161	158
67	TZA 2466	TARI	136	98	118	120	98	116	114	180	165	149	159	145	155	159
68	ICEAP 01179	ICRISAT	115	111	113	123	117	134	119	162	167	170	152	171	179	167
69	MWPLR 13	GENEBANK	124	124	113	113	114	115	117	164	186	163	159	155	151	163
70	MWPLR 2	GENEBANK	130	143	104	105	107	106	116	166	193	155	146	166	140	161
71	TZA 250	DARS	131	88	131	125	123	123	120	168	138	186	160	180	163	166
72	MWPLR 3	GENEBANK	113	125	120	116	125	118	119	151	187	161	152	187	156	165
73	TZA 5541	TARI	135	86	117	113	105	113	111	181	157	158	152	176	158	163
74	MWPLR 23	GENEBANK	118	78	123	113	115	118	110	163	145	166	153	165	163	159
75	ICEAP 00979/1	ICRISAT	123	113	113	114	127	124	119	168	165	173	146	167	158	163
76	TZA 197	TARI	101	126	84	79	80	89	81	135	182	126	115	135	125	136
77	MWPLR 20	GENEBANK	115	105	117	116	95	118	111	151	159	159	168	152	156	157
78	HOMBOLO	TARI	111	98	109	114	98	119	108	150	145	153	152	145	164	151
79	ICEAP 86012	ICRISAT	124	101	122	117	127	119	118	168	153	165	152	179	161	163
80	ICEAP 01106/1	ICRISAT	144	129	116	110	125	114	123	194	190	166	145	189	154	173
81	Sauma	DARS	163	127	155	132	165	130	145	215	201	254	171	211	178	205
Mean			118	103	116	111	106	113	111	155	157	164	149	156	154	156
STD			18	18	15	13	17	12	11	22	22	22	14	18	15	12
SED±			2	2	2	1	2	1	1	2	2	2	2	2	2	1
CV (%)			15	18	13	12	16	11	10	14	14	13	9	12	10	8

Appendix 2. Continued

			PH (cm)							NPB						
			Y1			YII			Mean	Y1			Y11			
Genotype code	Pedigree/name	Source	S1	S2	S3	S1	S2	S3		S1	S2	S3	S1	S2	S3	**Mean**
1	ICEAP 0673/1	ICRISAT	158.0	165.0	159.0	147.5	222.1	166.0	169.6	13	17	18	17	12	15	15
2	ICEAP 00554	ICRISAT	138.0	127.0	206.0	158.5	216.5	204.0	175.0	14	13	19	14	15	12	14
3	ICEAP 01164/1	ICRISAT	206.0	210.0	145.0	130.0	167.4	145.0	167.2	13	11	21	19	18	11	15
4	MWPLR 19	GENEBANK	160.0	193.0	164.0	167.0	205.4	172.0	176.9	14	13	20	17	12	11	14
5	MWPLR 22	GENEBANK	156.0	207.0	133.5	118.0	180.4	153.0	158.0	15	9	19	18	13	16	15
6	ICEAP 01170	ICRISAT	142.0	234.5	142.0	167.0	171.9	177.0	172.4	14	15	23	17	18	14	17
7	ICEAP 01169	ICRISAT	170.5	199.0	165.0	109.0	190.6	148.0	163.7	15	16	12	22	15	12	15
8	TZA 2439	TARI	138.5	161.5	185.5	127.5	210.1	162.5	164.3	19	13	16	15	17	12	15
9	MWPLR 9	GENEBANK	214.0	202.0	163.5	120.5	160.4	163.5	170.7	16	16	16	13	12	13	14
10	MWPLR 6	GENEBANK	146.0	213.0	190.5	145.5	249.7	176.0	186.8	14	10	20	17	14	11	14
11	MWPLR 17	GENEBANK	182.5	175.0	163.5	145.0	184.1	163.5	168.9	15	14	19	14	15	16	15
12	TZA 253	TARI	160.5	131.5	188.5	157.0	208.7	196.5	173.8	12	13	18	15	17	16	15
13	MWPLR 1	GENEBANK	169.0	171.5	134.0	134.5	203.3	156.5	161.5	18	12	18	15	10	15	14
14	MWPLR 18	GENEBANK	156.5	132.0	160.5	92.0	183.6	160.5	147.5	19	13	21	16	11	13	15
15	TZA 2464	TARI	158.0	165.5	203.5	152.0	245.0	207.0	188.5	12	16	17	16	18	11	15
16	ICEAP 00604	ICRISAT	179.0	139.0	163.5	138.0	158.0	122.5	150.0	17	12	15	17	18	14	15
17	TZA 2509	GENEBANK	163.5	164.0	163.5	100.0	152.1	156.0	149.9	15	14	21	17	16	13	16
18	ICEAP 01146/1	ICRISAT	185.0	157.5	154.5	131.0	216.4	178.0	170.4	19	15	18	14	18	14	16
19	MWPLR 11	GENEBANK	174.5	138.5	180.5	127.5	205.1	170.5	166.1	14	13	13	15	10	6	12
20	TZA 5555	TARI	152.5	163.0	168.5	138.5	247.5	166.5	172.8	10	12	20	12	18	11	14
21	No. 40	TARI	166.5	220.0	193.0	160.0	212.8	193.0	190.9	19	19	17	14	18	12	16
22	ICEAP 01150	ICRISAT	188.0	147.5	175.0	126.0	174.0	154.0	160.8	14	12	22	13	14	13	15
23	MZ2/9	TARI	172.0	230.0	151.0	110.5	206.4	168.5	173.1	15	14	18	12	16	16	15
24	ICEAP 01172/1	ICRISAT	143.5	228.5	181.0	142.5	198.5	181.0	179.2	13	17	18	16	15	15	16
25	ICEAP 01103/1	ICRISAT	140.0	160.0	172.5	152.5	209.4	192.5	171.2	18	13	18	14	14	11	14
26	MWPLR 24	GENEBANK	157.5	159.0	142.0	114.5	205.6	187.0	160.9	12	17	17	15	17	13	15

Appendix 2. Continued

27	ICEAP 01155	ICRISAT	175.0	152.0	166.0	141.0	224.9	178.5	172.9	15	9	13	13	13	12	12
28	ICEAP 01180/2	ICRISAT	206.0	161.0	173.0	129.0	190.0	149.0	168.0	18	12	14	14	14	12	14
29	MWPLR 4	GENEBANK	185.0	167.0	185.5	116.0	180.5	185.5	169.9	16	18	18	20	14	13	16
30	Kachangu	DARS	229.5	188.5	204.0	170.0	218.5	204.0	202.4	15	13	18	15	16	15	15
31	Mwayiwathualimi	DARS	172.0	216.0	142.0	133.0	174.3	171.5	168.1	19	15	21	15	15	14	16
32	MWPLR 8	ICRISAT	170.5	159.5	166.5	136.5	208.5	201.5	173.8	13	11	16	14	16	14	14
33	ICEAP 01154/2	ICRISAT	174.5	175.0	181.0	143.5	198.9	172.0	174.2	14	15	19	13	11	12	14
34	Chitedze Pigeonpea 1	DARS	169.5	146.5	141.0	157.5	197.9	139.5	158.7	11	12	23	12	13	13	14
35	ICEAP 01164/1	ICRISAT	203.5	188.5	147.5	142.0	191.1	122.0	165.8	21	13	17	19	13	14	16
36	Bangili	TARI	176.0	168.0	148.5	151.0	207.4	170.0	170.2	19	16	15	10	13	12	14
37	ICEAP 00053	ICRISAT	165.5	155.5	175.5	188.0	213.3	202.5	183.4	11	17	18	14	16	12	14
38	MWPLR 12	GENEBANK	214.5	167.0	146.0	153.5	206.1	146.0	172.2	20	42	18	11	12	11	19
39	TZA5463	TARI	203.0	154.5	174.0	157.5	200.0	151.5	173.4	16	18	17	15	12	12	15
40	MWPLR 5	GENEBANK	132.5	171.5	145.0	129.5	217.9	132.0	154.7	13	13	16	18	12	10	13
41	MWPLR 15	GENEBANK	165.0	137.0	156.5	152.0	161.3	182.5	159.1	14	14	16	11	13	9	13
42	ICEAP 87105	ICRISAT	140.0	153.0	175.5	104.5	207.7	120.0	125.3	14	9	16	14	13	13	13
43	MWPLR 16	GENEBANK	113.5	147.5	127.5	96.5	146.7	148.0	163.7	14	15	17	14	17	11	15
44	TZA 2496	TARI	213.5	172.5	168.0	121.5	216.0	155.0	174.4	12	10	17	15	16	15	14
45	TZA 5582	TARI	139.5	144.5	173.0	161.5	169.4	197.5	164.2	15	13	22	15	17	14	16
46	TZA 5596	TARI	183.0	140.5	191.5	181.0	219.1	208.5	187.3	7	17	14	14	12	12	12
47	Chitedze Pigeonpea 2	DARS	176.5	157.0	156.0	170.5	198.6	156.0	169.1	12	9	15	12	12	12	12
48	MWPLR 7	GENEBANK	196.5	179.5	139.5	158.5	183.7	139.5	166.2	13	16	16	11	12	14	14
49	Babati	TARI	175.0	169.5	202.5	115.0	237.4	162.5	177.0	11	15	18	13	16	15	14
50	TZA 5557	TARI	119.0	101.5	149.5	130.5	218.5	166.5	147.6	18	13	14	15	17	13	15
51	MWPLR 14	ICRISAT	151.5	109.0	158.0	234.5	209.4	149.0	168.6	13	12	14	18	13	11	13
52	ICEAP 01101/2	ICRISAT	148.0	111.0	215.0	138.5	238.8	183.0	172.4	11	13	16	16	15	9	13
53	TZA 2456	TARI	185.5	114.0	133.5	147.5	160.0	133.5	145.7	17	8	18	14	13	14	14
54	TZA 5464	TARI	166.0	105.5	186.0	167.0	226.1	204.0	175.8	14	17	19	14	14	14	15
55	TZA 5463	TARI	200.5	105.0	157.5	167.0	181.7	159.0	161.8	16	11	17	16	9	12	13
56	ICEAP 01285	ICRISAT	144.0	108.5	166.5	130.0	224.4	166.5	156.7	12	16	20	16	16	10	15

Appendix 2. Continued

57	MWPLR 25	GENEBANK	184.5	114.5	142.5	168.0	161.1	166.0	156.1	17	10	15	14	15	15	14
58	ICEAP 87091	ICRISAT	203.5	222.0	154.5	114.5	193.1	154.5	173.7	17	13	16	19	13	10	14
59	TZA 2692	TARI	135.5	145.0	141.0	128.5	190.9	134.0	145.8	14	17	23	12	16	16	16
60	TZA 2807	TARI	179.0	203.0	185.5	116.5	204.5	145.0	172.3	13	10	16	16	13	13	13
61	ICEAP 00068	ICRISAT	206.0	159.0	197.5	156.0	209.2	197.5	187.5	12	13	18	12	12	14	13
62	TZA 2785	TARI	186.5	167.5	127.5	147.0	104.4	127.5	143.4	19	15	16	16	16	12	15
63	MWPLR 10	GENEBANK	140.5	173.0	176.5	169.0	156.1	199.0	169.0	18	13	16	15	15	9	14
64	ICEAP 00612	ICRISAT	138.0	138.5	171.5	166.5	244.8	183.5	173.8	18	7	21	13	15	11	14
65	MWPLR 21	GENEBANK	146.0	142.0	99.5	151.5	159.0	118.5	136.1	14	15	19	18	14	11	15
66	TZA 2514	TARI	181.5	177.5	164.0	161.5	156.8	149.5	165.1	12	13	13	14	16	12	13
67	TZA 2466	TARI	142.5	150.0	176.5	166.5	193.4	173.5	167.1	15	9	18	18	8	15	13
68	ICEAP 01179	ICRISAT	160.5	201.5	169.0	174.5	169.9	154.0	171.6	14	5	21	13	12	11	12
69	MWPLR 13	GENEBANK	147.5	193.5	175.0	152.0	192.6	176.0	172.8	13	16	15	17	14	14	15
70	MWPLR 2	GENEBANK	118.0	268.5	192.0	125.5	201.5	182.0	181.3	16	17	24	10	14	10	15
71	TZA 250	DARS	153.0	174.5	189.0	160.0	194.5	206.5	179.6	17	11	23	15	16	11	15
72	MWPLR 3	GENEBANK	142.0	195.5	147.0	151.5	186.7	172.5	165.9	16	9	19	12	14	12	13
73	TZA 5541	TARI	191.0	196.5	126.5	128.0	134.6	133.0	151.6	11	11	20	16	18	13	14
74	MWPLR 23	GENEBANK	156.0	195.0	185.5	124.5	178.7	164.0	167.3	15	18	17	20	18	12	17
75	ICEAP 00979/1	ICRISAT	187.5	202.0	124.5	153.5	197.7	124.5	165.0	11	5	17	17	15	13	13
76	TZA 197	TARI	140.0	95.0	164.5	155.0	150.3	164.5	144.9	15	13	22	14	13	14	15
77	MWPLR 20	GENEBANK	178.0	166.5	182.5	101.0	194.6	164.5	164.5	12	15	19	15	33	18	18
78	HOMBOLO	TARI	151.5	150.0	196.0	151.5	210.1	177.5	172.8	13	14	19	12	23	15	16
79	ICEAP 86012	ICRISAT	174.0	165.5	167.5	120.5	201.4	148.0	162.8	11	18	23	13	14	13	15
80	ICEAP 01106/1	ICRISAT	174.5	201.0	202.5	148.5	234.8	172.5	189.0	20	13	20	12	13	13	15
81	Sauma	DARS	163.0	222.0	191.0	160.5	168.1	194.5	183.2	13	17	19	18	12	14	15
Mean			167.3	167.2	166.0	143.1	195.1	166.2	167.6	15	14	18	15	15	13	15
STD			24.3	34.6	22.5	23.3	27.3	23.1	12.7	2.7	4.4	2.7	2.4	3.2	2.0	1.3
SED±			2.7	3.8	2.5	2.6	3.0	2.6	1.4	0.3	0.5	0 3	0.3	0.4	0.2	0.1
CV (%)			14.5	20.7	13.5	16.3	14.0	13.9	7.6	18.7	31.8	14.8	16.1	21.9	15.5	9.0

Appendix 2. Continued

			NSB							NRP						
			Y1			YII				Y1			Y11			
Genotype code	Pedigree/name	Source	S1	S2	S3	S1	S2	S3	**Mean**	S1	S2	S3	S1	S2	S3	**Mean**
1	ICEAP 0673/1	ICRISAT	4	5	10	6	3	5	5	149	501	90	190	57	48	172
2	ICEAP 00554	ICRISAT	7	6	5	13	2	4	6	194	494	86	144	67	74	176
3	ICEAP 01164/1	ICRISAT	7	12	14	2	4	2	7	144	313	85	171	103	57	146
4	MWPLR 19	GENEBANK	8	6	8	8	2	3	6	207	353	82	232	83	46	167
5	MWPLR 22	GENEBANK	11	8	14	6	6	3	8	150	368	95	233	119	95	176
6	ICEAP 01170	ICRISAT	8	8	10	6	5	5	7	220	219	123	173	86	68	148
7	ICEAP 01169	ICRISAT	4	7	5	17	5	3	7	133	465	83	198	83	43	167
8	TZA 2439	TARI	15	15	5	4	5	6	8	211	219	75	132	110	49	133
9	MWPLR 9	GENEBANK	11	15	7	4	1	5	7	230	291	99	122	78	56	146
10	MWPLR 6	GENEBANK	7	6	7	3	7	3	5	166	514	100	134	92	54	176
11	MWPLR 17	GENEBANK	6	13	8	2	2	8	6	132	339	82	118	92	56	136
12	TZA 253	TARI	8	5	8	3	7	12	7	167	184	80	167	101	73	129
13	MWPLR 1	GENEBANK	11	9	7	2	2	6	6	155	228	80	119	99	52	122
14	MWPLR 18	GENEBANK	5	9	13	11	3	5	8	201	236	63	221	70	73	144
15	TZA 2464	TARI	10	9	8	3	7	9	7	166	276	100	120	138	74	145
16	ICEAP 00604	ICRISAT	13	13	9	12	13	4	10	244	360	157	180	80	45	177
17	TZA 2509	GENEBANK	9	15	7	6	5	5	8	184	258	96	139	94	61	139
18	ICEAP 01146/1	ICRISAT	16	18	14	1	8	8	11	216	582	103	140	108	81	205
19	MWPLR 11	GENEBANK	3	9	12	5	2	3	5	191	196	130	175	59	44	132
20	TZA 5555	TARI	1	10	10	3	15	2	7	126	259	106	148	130	73	140
21	No. 40	TARI	7	13	19	1	1	3	7	214	402	71	130	61	47	154
22	ICEAP 01150	ICRISAT	11	6	8	4	5	3	6	200	416	79	122	82	61	160
23	MZ2/9	TARI	8	6	5	8	2	7	6	193	423	101	154	102	60	172
24	ICEAP 01172/1	ICRISAT	4	7	18	7	3	6	7	113	537	82	199	89	86	184
25	ICEAP 01103/1	ICRISAT	8	8	13	2	4	4	6	245	102	105	141	86	67	124

26	MWPLR 24	GENEBANK	4	18	5	2	4	4	6	132	232	94	146	76	60	123

Appendix 2. Continued

27	ICEAP 01155	ICRISAT	6	4	8	3	4	2	4	147	349	96	115	91	62	143
28	ICEAP 01180/2	ICRISAT	4	12	8	4	4	4	6	192	178	103	147	98	57	129
29	MWPLR 4	GENEBANK	7	20	7	6	4	3	8	174	491	97	231	77	62	188
30	Kachangu	DARS	4	5	12	4	2	4	5	178	430	134	132	73	52	166
31	Mwayiwathualimi	DARS	13	14	8	8	4	4	8	268	164	100	125	71	60	131
32	MWPLR 8	ICRISAT	2	11	14	3	4	5	6	167	90	131	212	87	51	123
33	ICEAP 01154/2	ICRISAT	10	7	6	3	0	7	5	138	280	97	184	66	70	139
34	Chitedze Pigeonpea 1	DARS	2	10	16	5	3	4	6	167	298	88	144	79	39	136
35	ICEAP 01164/1	ICRISAT	14	8	9	8	6	2	8	209	153	126	233	87	45	142
36	Bangili	TARI	4	8	7	4	2	3	4	165	274	97	189	87	54	144
37	ICEAP 00053	ICRISAT	5	18	9	1	9	3	7	121	142	68	128	97	59	102
38	MWPLR 12	GENEBANK	14	6	23	4	2	3	8	136	307	60	118	70	59	125
39	TZA5463	TARI	8	11	8	2	3	4	6	161	465	103	145	55	60	165
40	MWPLR 5	GENEBANK	9	8	7	12	3	1	6	135	112	133	154	78	47	110
41	MWPLR 15	GENEBANK	7	10	13	3	3	1	6	213	158	132	176	99	57	139
42	ICEAP 87105	ICRISAT	7	18	6	6	2	5	7	122	150	87	151	80	62	109
43	MWPLR 16	GENEBANK	7	10	6	6	5	4	6	138	173	97	117	95	58	113
44	TZA 2496	TARI	4	8	6	0	16	4	6	224	310	109	92	90	66	148
45	TZA 5582	TARI	7	3	30	4	3	3	8	191	647	160	151	88	83	220
46	TZA 5596	TARI	4	9	7	4	3	2	5	87	249	66	192	72	63	121
47	Chitedze Pigeonpea 2	DARS	3	8	13	5	2	4	6	134	317	90	111	71	48	128
48	MWPLR 7	GENEBANK	5	9	10	1	2	9	6	191	481	54	203	69	44	173
49	Babati	TARI	5	4	11	5	2	6	5	106	538	82	119	275	58	196
50	TZA 5557	TARI	10	10	11	5	5	4	7	116	321	199	195	81	46	159
51	MWPLR 14	ICRISAT	11	8	12	5	3	2	7	260	155	146	113	80	51	134
52	ICEAP 01101/2	ICRISAT	3	9	6	4	4	1	4	188	171	86	152	89	59	124
53	TZA 2456	TARI	10	12	4	2	7	7	7	207	231	58	122	106	53	129
54	TZA 5464	TARI	5	8	6	10	3	6	6	215	155	65	198	75	62	128
55	TZA 5463	TARI	22	7	8	5	15	4	10	293	164	70	193	106	55	146

56	ICEAP 01285	ICRISAT	5	10	12	12	4	3	8	123	127	62	158	89	74	105
57	MWPLR 25	GENEBANK	9	8	7	5	2	9	6	193	261	102	180	98	46	147

Appendix 2. Continued

58	ICEAP 87091	ICRISAT	7	10	11	6	3	3	6	168	343	74	228	91	70	162
59	TZA 2692	TARI	8	12	13	5	8	5	8	179	173	102	134	81	50	120
60	TZA 2807	TARI	3	16	7	8	7	4	7	116	177	101	170	77	60	116
61	ICEAP 00068	ICRISAT	1	23	6	7	4	4	7	125	479	72	165	90	71	167
62	TZA 2785	TARI	17	7	9	1	3	4	7	165	628	73	169	80	82	199
63	MWPLR 10	GENEBANK	12	10	6	8	4	3	7	209	269	76	231	88	50	154
64	ICEAP 00612	ICRISAT	11	17	20	4	19	2	12	253	104	131	145	146	67	141
65	MWPLR 21	GENEBANK	14	8	16	2	5	3	8	274	185	139	190	76	42	151
66	TZA 2514	TARI	4	8	7	6	2	3	5	148	168	108	119	76	49	111
67	TZA 2466	TARI	8	10	10	5	1	3	6	150	306	107	194	82	48	148
68	ICEAP 01179	ICRISAT	8	12	14	6	9	2	8	155	470	133	129	94	58	173
69	MWPLR 13	GENEBANK	10	9	8	12	7	3	8	174	496	148	265	99	63	207
70	MWPLR 2	GENEBANK	6	7	11	2	2	3	5	159	223	93	274	78	53	146
71	TZA 250	DARS	14	6	6	4	3	3	6	193	398	72	160	90	66	163
72	MWPLR 3	GENEBANK	11	6	10	8	6	3	7	218	175	127	190	99	41	141
73	TZA 5541	TARI	3	6	9	5	4	4	5	104	412	88	149	94	49	149
74	MWPLR 23	GENEBANK	3	6	11	11	9	5	7	196	414	98	125	84	81	166
75	ICEAP 00979/1	ICRISAT	7	12	7	6	3	5	7	113	136	163	184	78	35	118
76	TZA 197	TARI	6	7	14	6	1	6	6	200	123	71	136	73	65	111
77	MWPLR 20	GENEBANK	4	9	19	6	8	8	9	133	271	138	115	81	70	134
78	HOMBOLO	TARI	2	11	9	13	29	7	11	151	574	92	113	196	49	196
79	ICEAP 86012	ICRISAT	3	12	12	3	20	2	8	98	552	70	131	163	54	178
80	ICEAP 01106/1	ICRISAT	15	13	12	1	5	5	8	185	527	84	180	99	77	192
81	Sauma	DARS	9	17	10	4	3	2	7	200	536	85	89	69	40	170
Mean			8	10	10	5	5	4	7	174	312	99	161	91	59	149
STD			4.1	4.0	4.4	3.3	4.7	2.1	1.5	43.8	145.8	27.5	39.8	29.9	12.1	26.2
SED±			0.5	0.4	0 5	0.4	0.5	0.2	0.2	4 9	16.2	3.1	4.4	3.3	1.3	2.9
CV (%)			53.8	40.4	44.1	62.7	90.9	48.6	22.1	25.1	46.8	27.9	24.8	32.6	20.6	17.6

Appendix 2. Continued

			NPP							NSP						
			Y1			YII			Mean	Y1			Y11			
Genotype code	Pedigree/name	Source	S1	S2	S3	S1	S2	S3		S1	S2	S3	S1	S2	S3	**Mean**
1	ICEAP 0673/1	ICRISAT	59	237	99	44	84	110	105	4	5	6	6	6	6	5
2	ICEAP 00554	ICRISAT	91	225	68	76	122	132	119	6	6	6	5	4	6	5
3	ICEAP 01164/1	ICRISAT	120	128	82	49	103	82	94	6	6	5	5	6	6	5
4	MWPLR 19	GENEBANK	105	169	58	78	94	98	100	6	5	6	5	6	7	6
5	MWPLR 22	GENEBANK	76	65	58	48	104	74	71	7	6	5	5	5	6	5
6	ICEAP 01170	ICRISAT	118	142	124	41	36	105	94	6	4	5	5	4	6	5
7	ICEAP 01169	ICRISAT	129	156	59	37	130	58	95	6	6	6	5	6	6	6
8	TZA 2439	TARI	143	117	50	30	100	101	90	7	4	6	5	6	6	5
9	MWPLR 9	GENEBANK	161	79	85	69	77	86	93	7	6	5	5	6	6	6
10	MWPLR 6	GENEBANK	91	175	91	34	143	86	103	7	6	6	5	5	6	6
11	MWPLR 17	GENEBANK	95	85	87	59	55	101	80	6	5	7	6	5	6	6
12	TZA 253	TARI	140	216	63	58	67	114	110	6	6	6	5	6	6	6
13	MWPLR 1	GENEBANK	98	195	55	37	60	95	90	6	6	5	5	6	6	5
14	MWPLR 18	GENEBANK	123	166	57	59	56	110	95	6	6	6	5	5	6	5
15	TZA 2464	TARI	100	156	102	56	123	88	104	6	6	6	4	5	6	5
16	ICEAP 00604	ICRISAT	138	163	72	54	92	59	96	7	5	5	5	5	6	5
17	TZA 2509	GENEBANK	102	158	65	35	112	89	93	7	5	5	5	6	5	5
18	ICEAP 01146/1	ICRISAT	112	123	78	45	158	75	98	6	6	6	5	5	6	5
19	MWPLR 11	GENEBANK	122	112	125	51	99	74	97	6	6	6	5	5	6	6
20	TZA 5555	TARI	115	177	78	38	157	45	101	6	6	6	6	6	6	6
21	No. 40	TARI	157	270	66	61	92	98	124	6	6	5	5	5	7	5
22	ICEAP 01150	ICRISAT	94	152	87	40	44	95	85	6	6	6	6	5	6	6
23	MZ2/9	TARI	69	120	80	100	81	98	91	7	7	6	6	4	6	6
24	ICEAP 01172/1	ICRISAT	144	130	90	65	87	78	99	5	6	6	5	5	7	6
25	ICEAP 01103/1	ICRISAT	101	118	62	54	56	111	83	5	5	7	5	5	6	5

26	MWPLR 24	GENEBANK	104	196	89	26	84	94	99	6	5	6	6	6	5	5
27	ICEAP 01155	ICRISAT	115	86	54	32	89	96	78	7	5	5	5	5	5	5
	Appendix 2. Continued															
28	ICEAP 01180/2	ICRISAT	146	136	112	52	89	85	103	5	6	6	6	5	6	5
29	MWPLR 4	GENEBANK	145	153	111	53	39	89	98	6	6	5	5	2	7	5
30	Kachangu	DARS	127	362	95	97	83	101	144	7	5	5	6	6	6	6
31	Mwayiwathualimi	DARS	185	207	57	50	105	93	116	6	6	6	5	5	6	5
32	MWPLR 8	ICRISAT	77	137	93	29	29	74	73	6	6	5	4	4	6	5
33	ICEAP 01154/2	ICRISAT	128	171	80	35	59	86	93	6	6	6	6	5	7	6
34	Chitedze Pigeonpea 1	DARS	70	237	68	22	74	82	92	6	6	7	5	5	7	6
35	ICEAP 01164/1	ICRISAT	123	123	106	56	103	78	98	6	6	5	5	5	6	5
36	Bangili	TARI	135	109	75	20	93	91	87	6	5	6	6	6	6	6
37	ICEAP 00053	ICRISAT	103	107	60	52	99	38	76	6	6	6	5	5	5	5
38	MWPLR 12	GENEBANK	77	88	67	76	74	84	77	6	6	5	6	6	8	6
39	TZA5463	TARI	128	125	93	38	61	82	88	6	6	5	5	6	6	6
40	MWPLR 5	GENEBANK	115	333	76	46	36	95	117	6	5	6	5	4	6	5
41	MWPLR 15	GENEBANK	145	156	106	47	83	79	102	6	5	6	6	5	7	5
42	ICEAP 87105	ICRISAT	99	78	90	62	67	70	78	6	5	5	5	4	6	5
43	MWPLR 16	GENEBANK	119	315	98	72	110	90	134	6	6	6	5	6	6	6
44	TZA 2496	TARI	141	135	87	30	106	82	96	6	5	6	5	5	3	5
45	TZA 5582	TARI	96	261	106	81	92	122	126	6	5	6	5	5	6	5
46	TZA 5596	TARI	68	140	59	63	81	98	85	6	6	5	6	4	6	5
47	Chitedze Pigeonpea 2	DARS	96	114	90	45	56	82	80	6	6	6	5	5	6	5
48	MWPLR 7	GENEBANK	74	119	61	70	57	72	75	6	5	5	6	5	5	5
49	Babati	TARI	98	151	123	43	97	94	101	6	6	6	5	6	5	5
50	TZA 5557	TARI	79	78	60	59	96	84	76	6	5	6	6	5	6	6
51	MWPLR 14	ICRISAT	167	231	109	65	76	90	123	7	5	5	5	6	6	6
52	ICEAP 01101/2	ICRISAT	134	96	64	55	64	90	84	6	5	5	6	5	6	5
53	TZA 2456	TARI	137	94	48	47	121	66	85	6	6	6	5	4	6	5
54	TZA 5464	TARI	180	96	52	83	106	128	107	6	6	6	6	5	6	6
55	TZA 5463	TARI	187	138	100	56	29	72	97	7	6	7	6	2	6	5

56	ICEAP 01285	ICRISAT	95	75	59	73	64	38	67	6	5	6	5	5	3	5
57	MWPLR 25	GENEBANK	125	193	73	73	112	84	110	7	5	6	5	6	6	6
58	ICEAP 87091	ICRISAT	140	120	62	50	40	95	84	6	4	5	5	4	6	5

Appendix 2. Continued

59	TZA 2692	TARI	146	110	64	46	41	59	78	6	5	6	5	3	6	5
60	TZA 2807	TARI	86	132	62	50	44	102	79	6	5	5	5	5	6	5
61	ICEAP 00068	ICRISAT	69	119	58	66	71	103	81	7	6	5	5	7	6	6
62	TZA 2785	TARI	90	136	84	60	22	64	76	6	6	6	6	2	6	5
63	MWPLR 10	GENEBANK	156	150	87	53	53	72	95	6	6	6	6	5	7	6
64	ICEAP 00612	ICRISAT	116	216	86	61	83	72	105	7	6	6	5	5	6	6
65	MWPLR 21	GENEBANK	182	114	100	38	94	66	99	6	5	6	5	6	6	5
66	TZA 2514	TARI	69	186	82	26	78	64	84	7	6	5	5	5	6	5
67	TZA 2466	TARI	136	174	65	43	66	86	95	7	6	5	5	6	5	5
68	ICEAP 01179	ICRISAT	117	15	144	59	37	58	71	7	5	7	5	5	6	6
69	MWPLR 13	GENEBANK	143	133	126	61	94	97	109	7	6	5	6	6	6	6
70	MWPLR 2	GENEBANK	121	160	73	52	40	103	91	6	6	5	5	6	7	6
71	TZA 250	DARS	140	179	59	48	43	113	97	6	6	6	5	5	7	6
72	MWPLR 3	GENEBANK	105	160	75	52	41	99	89	7	6	6	4	5	6	5
73	TZA 5541	TARI	80	139	70	36	101	80	84	6	5	6	5	6	6	5
74	MWPLR 23	GENEBANK	128	112	64	46	40	94	81	6	6	5	5	3	6	5
75	ICEAP 00979/1	ICRISAT	103	96	142	45	54	56	82	6	5	6	6	5	6	6
76	TZA 197	TARI	111	77	53	37	67	108	75	6	6	6	5	7	6	6
77	MWPLR 20	GENEBANK	100	119	97	37	107	72	89	6	6	6	6	5	6	6
78	HOMBOLO	TARI	64	167	84	53	192	152	118	6	6	5	6	6	6	5
79	ICEAP 86012	ICRISAT	53	226	51	26	165	90	102	6	6	6	6	5	7	6
80	ICEAP 01106/1	ICRISAT	113	139	64	33	83	80	85	7	3	6	5	6	6	5
81	Sauma	DARS	140	240	69	61	70	82	110	6	5	5	5	4	7	5
Mean			115	152	80	51	81	87	94	6	6	6	5	5	6	5
STD			30.5	60.4	22.0	16.1	33.2	19.5	15.0	0 5	0.6	0.6	0.5	1.0	0.7	0.5
SED±			3.4	6.7	2.4	1.8	3.7	2.2	1.7	0 1	0.1	0.1	0.1	0.1	0.1	0.1
CV (%)			26.5	39.8	27.5	31.3	40.7	22.5	15.9	8.8	11.7	10.2	9.9	19.3	11.8	9.1

Appendix 2. Continued

			GYD (t ha^{-1})							HSWT (g)						
			Y1			YII			Mean	Y1			Y11			
Genotype code	Pedigree/name	Source	S1	S2	S3	S1	S2	S3		S1	S2	S3	S1	S2	S3	**Mean**
1	ICEAP 0673/1	ICRISAT	0 9	0.5	1.3	1.3	2.3	1.8	1.3	15.5	16.0	14.5	19.0	11.0	13.5	14.9
2	ICEAP 00554	ICRISAT	1.0	0.7	1.0	1.0	2.2	1.9	1.3	17.5	14.5	12.5	8.5	10.0	17.0	13.3
3	ICEAP 01164/1	ICRISAT	1.0	0.5	1.2	1.4	1.2	1.1	1.0	16.5	14.0	10.5	16.5	12.0	11.0	13.4
4	MWPLR 19	GENEBANK	1.4	0.8	1.3	1.4	1.1	1.1	1.2	18.0	11.5	15.0	16.0	15.0	11.5	14.5
5	MWPLR 22	GENEBANK	0.6	0.4	2.1	2.2	1.3	1.2	1.3	17.0	14.5	13.5	26.5	15.0	17.0	17.3
6	ICEAP 01170	ICRISAT	0.6	0.5	1.8	1.7	1.8	1.3	1.3	15.5	16.5	15.5	19.0	12.5	14.0	15.5
7	ICEAP 01169	ICRISAT	0.6	0.5	1.5	1.1	1.4	0.9	1.0	18.0	15.0	15.0	15.5	9.0	11.0	13.9
8	TZA 2439	TARI	1 2	0.6	1.5	1.5	1.2	1.3	1.2	19.0	11.5	12.5	17.0	17.5	11.5	14.8
9	MWPLR 9	GENEBANK	0.7	0.4	1.4	1.4	1.2	0.7	0.9	14.5	14.0	13.5	15.5	12.5	11.0	13.5
10	MWPLR 6	GENEBANK	0.7	0.5	1.8	1.7	1.4	1.3	1.2	18.5	18.0	15.0	16.0	8.5	14.5	15.1
11	MWPLR 17	GENEBANK	0.6	0.4	1.0	1.0	1.5	1.0	0.9	11.0	7.5	7.5	15.5	10.0	15.5	11.2
12	TZA 253	TARI	1.6	0.5	0.7	0.7	2.1	1.6	1.2	18.0	10.0	14.5	17.5	12.0	17.0	14.8
13	MWPLR 1	GENEBANK	0.8	0.2	0.5	1.4	0.5	0.3	0.6	12.5	15.0	14.0	15.0	16.5	16.0	14.8
14	MWPLR 18	GENEBANK	1.0	0.6	0.8	0.8	2.4	2.2	1.3	17.5	12.5	13.0	19.0	13.5	18.5	15.7
15	TZA 2464	TARI	1 5	0.5	0.9	1.0	1.4	1.4	1.1	15.0	13.0	10.0	16.0	10.0	13.5	12.9
16	ICEAP 00604	ICRISAT	1.0	0.4	0.5	0.5	1.3	1.0	0.8	18.0	10.5	15.0	17.0	13.0	12.5	14.3
17	TZA 2509	GENEBANK	1 1	0.5	0.7	1.4	2.5	3.0	1.5	18.5	14.0	11.0	17.5	20.0	15.5	16.1
18	ICEAP 01146/1	ICRISAT	1.0	0.4	1.5	1.6	2.1	1.2	1.3	16.5	15.5	15.0	18.5	12.5	15.0	15.5
19	MWPLR 11	GENEBANK	1.0	0.4	1.5	1.6	1.5	0.7	1.1	18.0	15.5	15.5	17.0	11.5	16.5	15.7
20	TZA 5555	TARI	1 2	0.9	1.7	1.7	1.7	1.2	1.4	16.0	12.5	15.0	18.5	15.0	14.0	15.2
21	No. 40	TARI	2 1	0.9	2.3	2.4	1.3	1.7	1.8	16.0	16.5	10.0	10.5	12.5	15.5	13.5
22	ICEAP 01150	ICRISAT	0.7	0.3	1.2	1.0	2.3	1.0	1.1	16.0	16.5	19.0	18.5	5.0	14.0	14.8
23	MZ2/9	TARI	1 5	0.9	1.1	1.5	1.1	1.4	1.2	16.5	14.5	15.0	21.0	17.5	15.0	16.6
24	ICEAP 01172/1	ICRISAT	1.4	0.5	1.1	1.3	1.7	1.7	1.3	15.5	10.5	17.5	16.5	11.5	15.0	14.4
25	ICEAP 01103/1	ICRISAT	0 9	0.4	0.8	0.7	1.9	1.2	1.0	13.0	10.5	15.0	16.5	12.5	16.5	14.0
26	MWPLR 24	GENEBANK	1.4	0.7	1.0	1.2	1.5	1.4	1.2	13.0	15.0	7.5	18.0	12.5	14.0	13.3
27	ICEAP 01155	ICRISAT	1.0	0.5	1.3	1.6	1.6	1.2	1.2	15.0	17.0	14.0	14.0	10.5	12.5	13.8

Appendix 2. Continued

28	ICEAP 01180/2	ICRISAT	1 1	0.5	1.5	1.7	1.6	1.2	1.2	11.0	17.0	10.5	18.5	19.5	16.5	15.5
29	MWPLR 4	GENEBANK	1 5	1.0	1.6	1.7	2.0	0.8	1.4	18.5	17.0	12.0	18.0	2.5	15.5	13.9
30	Kachangu	DARS	2 3	1.6	1.2	1.2	1.4	1.8	1.6	17.5	17.0	15.0	16.0	16.0	12.0	15.6
31	Mwayiwathualimi	DARS	1 2	0.8	1.1	1.0	1.9	1.6	1.2	20.5	15.0	14.5	15.5	14.0	14.5	15.7
32	MWPLR 8	ICRISAT	0.6	0.5	0.7	0.7	1.6	0.8	0.8	17.5	14.0	14.5	24.0	11.0	12.5	15.6
33	ICEAP 01154/2	ICRISAT	0.7	0.8	1.0	0.9	1.5	1.1	1.0	15.0	13.5	12.5	17.0	12.5	16.5	14.5
34	Chitedze Pigeonpea 1	DARS	1 1	0.8	1.1	1.2	1.4	1.3	1.1	16.0	15.0	15.0	16.0	10.0	13.0	14.2
35	ICEAP 01164/1	ICRISAT	0.7	0.5	1.4	1.4	1.3	1.1	1.1	15.0	16.0	12.5	17.0	20.0	12.5	15.5
36	Bangili	TARI	1 5	1.1	1.2	1.3	1.2	1.1	1.2	13.0	10.0	12.5	15.0	16.5	13.0	13.3
37	ICEAP 00053	ICRISAT	1 1	0.7	0.6	0.6	1.2	1.3	0.9	19.5	6.5	13.5	20.5	11.5	5.0	12.8
38	MWPLR 12	GENEBANK	1.0	0.3	1.8	1.5	1.6	1.1	1.2	18.5	19.0	12.5	15.5	12.5	16.5	15.8
39	TZA5463	TARI	0.4	0.4	1.1	1.1	1.2	0.9	0.8	15.5	14.5	14.5	16.0	15.0	16.0	15.3
40	MWPLR 5	GENEBANK	0.8	0.4	1.9	1.8	1.5	0.9	1.2	18.5	14.0	12.5	18.0	12.5	13.5	14.8
41	MWPLR 15	GENEBANK	0.8	0.3	1.1	1.0	1.5	1.2	1.0	10.5	14.0	12.5	17.5	5.0	14.0	12.3
42	ICEAP 87105	ICRISAT	0.6	0.4	0.9	0.5	0.4	0.4	0.5	12.0	12.5	14.0	19.0	20.0	14.5	15.3
43	MWPLR 16	GENEBANK	1.7	1.7	1.6	1.8	1.6	1.9	1.7	17.0	14.5	14.0	17.0	22.5	13.0	16.3
44	TZA 2496	TARI	1 9	0.8	0.9	0.9	1.8	1.9	1.3	15.5	13.0	18.0	16.5	7.5	9.0	13.3
45	TZA 5582	TARI	1 5	0.9	1.4	1.5	2.3	1.9	1.6	18.4	19.0	15.5	16.0	16.5	18.0	17.2
46	TZA 5596	TARI	1 2	0.8	2.2	0.5	1.5	1.4	1.3	15.0	14.0	10.0	17.5	10.0	14.0	13.4
47	Chitedze Pigeonpea 2	DARS	1 2	0.6	1.0	1.0	1.5	1.5	1.1	20.5	10.0	9.5	19.5	7.5	13.5	13.4
48	MWPLR 7	GENEBANK	0.6	0.4	1.6	1.6	1.5	1.4	1.2	10.0	15.0	15.0	17.0	17.5	13.0	14.6
49	Babati	TARI	1.0	0.7	1.1	0.5	0.8	1.4	0.9	15.0	12.5	12.5	17.0	18.5	12.5	14.7
50	TZA 5557	TARI	0 9	0.5	0.4	0.5	0.4	0.7	0.6	13.0	10.5	17.5	21.0	19.0	14.5	15.9
51	MWPLR 14	ICRISAT	1.8	1.0	2.1	2.1	1.7	1.7	1.7	16.5	17.5	14.5	18.5	21.5	13.5	17.0
52	ICEAP 01101/2	ICRISAT	0.8	0.5	1.2	1.1	1.7	1.0	1.0	14.0	12.0	17.0	19.0	5.0	15.5	13.8
53	TZA 2456	TARI	1 2	0.7	1.1	1.1	1.6	1.5	1.2	16.0	19.5	13.5	16.5	12.5	14.0	15.3
54	TZA 5464	TARI	1 5	0.9	1.0	1.0	1.5	1.4	1.2	16.0	13.5	13.5	16.5	12.5	14.5	14.4
55	TZA 5463	TARI	1.4	0.5	1.0	1.0	1.3	1.5	1.1	20.0	12.5	12.5	21.5	2.5	14.5	13.9
56	ICEAP 01285	ICRISAT	1.0	0.5	1.5	1.5	1.1	1.1	1.1	13.0	14.5	7.5	23.5	15.0	5.5	13.2
57	MWPLR 25	GENEBANK	0.7	0.5	2.1	2.1	1.4	1.3	1.3	15.5	15.5	15.0	19.5	18.0	14.5	16.3

58	ICEAP 87091	ICRISAT	1.0	0.5	0.9	0.9	1.1	0.9	0.9	15.0	17.5	10.0	19.0	12.5	18.5	15.4

Appendix 2. Continued

59	TZA 2692	TARI	1 5	0.7	1.7	1.6	1.1	1.2	1.3	15.5	14.0	14.5	16.0	15.0	12.5	14.6
60	TZA 2807	TARI	1.6	0.8	0.9	0.9	2.2	1.8	1.4	17.0	16.0	10.0	20.0	15.5	19.0	16.3
61	ICEAP 00068	ICRISAT	0 9	0.7	1.0	1.2	1.4	1.5	1.1	15.5	12.5	10.0	17.0	10.0	14.5	13.3
62	TZA 2785	TARI	1.4	0.7	1.8	1.7	1.4	1.4	1.4	15.5	18.0	13.0	17.5	2.5	12.5	13.2
63	MWPLR 10	GENEBANK	0.8	0.4	1.3	1.5	2.2	1.2	1.2	19.5	16.0	12.5	18.0	13.0	17.5	16.1
64	ICEAP 00612	ICRISAT	0.6	0.4	1.5	1.7	1.1	0.7	1.0	17.5	14.5	12.5	15.0	20.0	15.5	15.8
65	MWPLR 21	GENEBANK	1.7	0.4	1.8	1.7	1.5	1.0	1.3	19.5	17.0	14.5	18.0	17.5	15.0	16.9
66	TZA 2514	TARI	2.4	1.5	1.2	1.2	1.4	1.5	1.5	15.5	15.5	15.0	17.5	17.5	13.5	15.8
67	TZA 2466	TARI	1 5	0.9	1.5	1.4	0.8	1.9	1.3	16.5	7.5	13.0	21.0	15.0	17.5	15.1
68	ICEAP 01179	ICRISAT	0.8	0.5	0.8	0.9	1.7	1.1	0.9	14.0	6.0	16.0	17.0	2.5	14.0	11.6
69	MWPLR 13	GENEBANK	0.6	0.4	1.7	1.7	2.1	1.4	1.3	15.0	15.5	18.5	20.0	14.5	14.0	16.3
70	MWPLR 2	GENEBANK	0.6	0.3	1.5	1.4	1.9	1.4	1.2	15.0	10.5	12.5	18.0	15.5	17.5	14.8
71	TZA 250	DARS	1.4	0.9	1.6	1.5	0.7	0.8	1.1	17.5	17.0	16.5	17.0	9.5	16.0	15.6
72	MWPLR 3	GENEBANK	1 1	0.8	1.8	1.5	1.0	1.2	1.2	11.0	12.0	7.5	18.0	5.0	12.5	11.0
73	TZA 5541	TARI	1 1	0.6	1.0	1.1	1.5	1.4	1.1	14.5	13.5	12.5	16.5	10.0	17.0	14.0
74	MWPLR 23	GENEBANK	2 2	1.6	1.1	1.0	1.1	1.8	1.5	14.5	14.5	15.5	16.9	20.0	13.5	15.8
75	ICEAP 00979/1	ICRISAT	0 9	0.7	1.1	1.2	1.3	1.2	1.0	16.0	15.0	12.5	19.0	10.0	13.5	14.3
76	TZA 197	TARI	1 3	0.7	1.0	1.0	1.5	1.5	1.1	13.0	13.5	11.0	16.0	17.5	14.5	14.3
77	MWPLR 20	GENEBANK	1 2	0.8	1.9	1.8	1.7	1.1	1.4	15.5	17.0	15.5	18.0	7.5	17.0	15.1
78	HOMBOLO	TARI	1 1	0.7	1.5	1.4	1.1	1.3	1.2	15.5	15.0	13.5	15.5	16.0	10.5	14.3
79	ICEAP 86012	ICRISAT	0.8	0.3	0.4	0.5	0.3	0.5	0.5	13.0	16.5	14.5	17.5	17.5	14.0	15.5
80	ICEAP 01106/1	ICRISAT	0.7	0.6	1.1	0.8	1.2	0.7	0.9	18.5	0.0	13.0	18.0	14.0	15.0	13.1
81	Sauma	DARS	1 3	0.5	1.5	1.6	2.3	2.3	1.6	19.5	16.0	15.5	19.0	15.0	11.0	16.0
Mean			1 1	0.6	1.3	1.3	1.5	1.3	1.2	15.9	14.0	13.5	17.5	13.0	14.2	14.7
STD			0.4	0.3	0.4	0.4	0.5	0.4	0.3	2.4	3.2	2.4	2.4	4.6	2.5	1.3
SED±			0.0	0.0	0.0	0.0	0.1	0.0	0.0	0.3	0.4	0.3	0.3	0.5	0.3	0.1
CV (%)			38.0	45.8	33.3	33.1	31.0	33.3	21.5	14.9	22.7	18.1	13.9	35.3	17.4	8.9

ICRISAT=International Crops Research Institute for the Semi-Arid Tropics, DARS= Department of Agricultural Research Services, TARI= Tanzania Agricultural Research Institute, STD= standard deviation, SED= standard error of difference, CV= coefficient of variation, S1= site 1 (Bvumbwe), S2= site 2 (Chitedze), S3= site 3 (Makoka), Y1= year 1 (2017/18), Y11= year 2 (2018/19), DTF= days

to flowering, DTM= days to 75% maturity, PH= plant height (cm), NPB= number of primary branches, NRP= number of racemes per plant, NPP= number of pods per plant, GYD= grain yield (t ha^{-1}), HSWT= 100 seed weight (g), See genotype codes (G1-G81) in Table 3 1

Appendix 3. Correlations of qualitative and quantitative traits of 81 Pigeonpea genotypes evaluated in six environments

	GH	FP	FMC	FSC	FSP	LS	LH	PF	PC	PL	DTF	TTM	NPB	NSB	NRP	NPP	NSP	SC	PH	GYD	HSWT	SMC	SEC	SCP	SSH
GH	1																								
FP	-0.047	1																							
FMC	0.112**	-0.003	1																						
FSC	0.150**	.134**	0.051	1																					
FSP	0.055	.082*	0.054	.417**	1																				
LS	0.041	-.190**	-.205**	-0.057	0.045	1																			
LH	0.054	.097**	.129**	-0.023	-.429**	-.121**	1																		
PF	0.022	-.127**	-.234**	0.041	0.026	.241**	-0.041	1																	
PC	-0.011	-.167**	.149**	-.275**	-0.026	.121**	0.000	.105**	1																
PH	0.038	0.004	-0.015	0.025	0.052	-0.043	-0.057	-.127**	-0.039	1															
DTF	0.033	.084**	-0.027	.173**	.121**	-.092**	-.118**	-0.025	-.109**	.107**	1														
DTM	-0.003	.106**	-0.032	.135**	.124**	-.105**	-.124**	.070*	-0.059	.073*	.780**	1													
NPB	.067*	-.121**	-.070*	-0.018	.096**	.096**	-.341**	-0.024	0.032	-0.020	0.047	0.004	1												

NSB	.123**	-.174**	-.119**	-0.001	.113**	.086**	-.215**	.083**	-0.040	-0.022	0.050	.073*	.334**	1											
NRP	.272**	-.218**	-0.037	0.030	-.076*	0.063	.202**	0.055	-0.025	.093**	-0.047	-0.015	0.062	.285**	1										

Appendix 3. Continued

NPP	.150**	-.092**	-.141**	0.014	-0.054	0.082*	.116**	.315**	-.069*	.114**	-0.008	.065*	-0.005	.287**	.458**	1									
NSP	0.037	-0.002	-0.035	0.047	0.016	-.087**	-0.014	.078*	-0.026	.283**	.084**	.075*	-0.012	0.016	0.030	.231**	1								
SC	0.025	-0.009	.090**	-.086**	.066*	0.055	-0.002	-0.015	0.051	0.037	-0.055	-0.061	-0.030	-0.041	-0.021	-0.014	-0.010	1							
PH	-0.041	.205**	-.158**	0.061	-0.004	0.011	0.021	.251**	-.068*	-0.017	.091**	.145**	-0.018	-0.009	-0.001	.124**	0.023	0.025	1						
GYD	0.059	0.006	-.132**	0.005	-0.021	.074*	-0.041	.286**	-0.006	.087**	.088**	.108**	0.008	0.049	0.053	.319**	.271**	-.068*	.141**	1					
HSWT	.066*	-0.037	.103**	0.039	-.077*	-0.053	.124**	-.181**	0.044	.221**	0.041	-0.061	0.009	-0.050	.134**	.088**	.195**	-0.028	-0.039	.471**	1				
SMC	-0.012	0.060	0.035	-0.047	-0.011	-.094**	0.006	0.043	0.000	-.071*	-0.011	0.029	0.024	-0.013	-0.004	0.010	0.035	0.045	0.054	.099**	0.061	1			
SEC	.084**	-0.051	.114**	-.110**	-0.016	-0.013	-.072*	0.059	.149**	0.027	-.067*	-.080*	0.019	0.038	.075*	0.037	0.029	.088**	-0.028	0.025	0.038	.141**	1		
SCP	0.052	-.094**	0.033	-0.050	0.023	.093**	-.092**	0.059	0.084**	0.037	-0.030	-.063*	0.021	0.048	0.043	0.056	.069*	0.023	-0.013	.122**	.107**	.143**	.242**	1	
SSH	-0.035	0.040	0.029	.069*	.068*	-0.021	-0.023	.073*	-.070*	.076*	.073*	.111**	-0.022	0.017	.082*	.071*	.073*	-0.081*	0.033	.075*	-0.006	0.055	-0.003	-0.051	1

**. Correlation is significant at the 0.01 level (2-tailed).

*. Correlation is significant at the 0.05 level (2-tailed).