Breeding for Resistance to Rice Yellow Mottle Virus and Improved Yield in Rice (*Oryza sativa* L.) in Tanzania

By

William Titus Suvi

BSc. and MSc. Sokoine University of Agriculture, Tanzania

A thesis submitted in partial fulfilment of the requirements for the degree of Doctor of Philosophy (PhD) in Plant Breeding

African Center for Crop Improvement (ACCI)

School of Agricultural, Earth and Environmental Sciences
College of Agriculture, Engineering and Science
University of KwaZulu-Natal
Republic of South Africa

November 2020

ABSTRACT

Rice [*Oryza sativa* (L.), 2n = 2x = 24] is the second most important staple food crop after wheat (*Triticum aestivum* L.) serving more than half of the world's population. In Tanzania, rice is the second most important cereal crop after maize (*Zea mays* L.). However, rice production and productivity in the country is hindered by several factors. One of the leading biotic constraints is the rice yellow mottle virus (RYMV) disease which is devastating the existing rice varieties, and causes severe yield losses of 20 to 100 % under field conditions. Both landraces and introduced varieties that are grown by farmers succumb to RYMV. Several control strategies have been recommended to reduce RYMV infection: however, the development and deployment of RYMV resistant varieties is the most effective, economical and environmentally friendly approach for subsistence farmers. Breeding for resistance to RYMV and improved yields are the main goals for rice breeders aiming to develop and release improved rice cultivars that meet the preferences of the farmers and their markets. Therefore, the objectives of this study were to: (i) assess farmers' perceptions, production constraints and variety preferences of rice in Tanzania to guide breeding; (ii) determine variation among Tanzanian rice germplasm collections based on agronomic traits and resistance to RYMV to select unique parents for breeding; (iii) assess the genetic diversity and population structure of rice genotypes using simple sequence repeat (SSR) markers to complement phenotypic profile and select parents; and (iv) determine the combining ability and gene action for resistance to RYMV disease and for key agronomic traits in rice, and thereby to develop new populations of parental germplasm for future breeding.

A participatory rural appraisal study was conducted involving 180 participants, using a structured questionnaire and focused group discussions with 90 farmers in the Mvomero, Kilombero and Kyela districts of Tanzania. The results indicated that rice was the most important food and cash crop, followed by maize, cassava (*Mannihot esculenta* Crantz), sweetpotato (*Ipomoea batatas* [L..] Lam.), sugarcane (*Saccharum officinarum* L.), pigeonpea (*Cajanus cajan* L.), cowpea (*Vigna unguiculata* [L.] Walp.), sesame (*Sesamum indicum* L.), common bean (*Phaseolus vulgaris* L.), cocoa (*Theobroma cacao* L.), banana (*Musa acuminate* L.), groundnut (*Arachis hypogaea* L.), and oil palm (*Elaeis guineensis* Jacq.). The majority of the respondents (67.2%) used farm saved seed from the previous rice harvest. The major constraints limiting rice production and productivity in all studied areas were diseases, insect pests, frequent droughts, the non-availability and high cost of fertilizers, a limited number of improved cultivars, poor soil fertility and bird damage. The farmers preferred rice varieties with high yield, disease resistance, drought tolerance, high market value, early maturity, attractive aroma, and local adaptation. A systematic rice-breeding program aimed

at improving RYMV resistance and incorporating farmers' preferred traits should be designed and implemented as a means to increase the productivity and adoption of new cultivars by the farmers across the rice-growing areas of Tanzania.

Fifty-four rice genotypes were field evaluated at two important rice production sites (Ifakara and Mkindo), which are recognized as RYMV hotspots in Tanzania, using a 6 × 9 alpha lattice design with two replications. There were significant ($p<0.05$) genotypic variations for agronomic traits and RYMV susceptibility in the tested germplasm. Seven genotypes with moderate to high RYMV resistance identified, namely Salama M-57, SSD1, IRAT 256, Salama M-55, Mwangaza, Lunyuki, and Salama M-19 were identified as new sources of resistance genes. Positive and significant correlations were detected between grain yield and number of panicles per plant (NPP), panicle length (PL), number of grains per panicle (NGP), percentage-filled grains (PFG), and thousand-grain weight (TGW), which are useful traits for simultaneous selection for rice yield improvement. A principal component analysis resulted in five principal components accounting for 79.88% of the total variation present in the assessed germplasm collection. Traits that contributed most to the total genotypic variability included NPP, number of tillers per plant (NT), PL, grain yield (GY), and days to 50% flowering (DFL). Genotypes, Rangimbili, Gigante, and SARO have complementary agronomic traits and RYMV resistance, and can be recommended for further evaluation, genetic analysis and breeding.

The genetic relationship and divergence of the 54 rice selected genotypes mentioned above were examined using 14 polymorphic simple sequence repeats (SSR) markers to select unique parents for breeding. Data analysis was based on marker and population genetic parameters. The mean polymorphic information content (PIC) was 0.61, suggesting a high level of polymorphism for the selected SSR markers among the rice accessions. The population structure revealed a narrow genetic base, with only two major sub-populations. Analysis of molecular variance revealed that only 30% of the variation was attributed to population differences, while 47% and 23% were due to variation among individuals within populations and within individual variation, respectively. The genetic distance and identity among genotypes varied from 0.083 to 1.834 and 0.159 to 0.921, respectively. A dendrogram grouped the genotypes into three clusters with wide variation. The selected genetic resources, namely IR56, Mwanza, Salama M-55, Sindano nyeupe, SARO, Gigante, Lunyuki, Rangimbili, IRAT 256, Zambia and Salama M-19, will be useful resources for rice breeding in Tanzania and other African countries because they are genetically diverse.

The final study involved combining ability analysis of the above selected genotypes and derived families to assess gene action conditioning RYMV resistance and agronomic traits.

Ten parental lines and their 45 F_2 progenies were field evaluated at three selected locations using a 5 × 11 alpha lattice design with two replications. The genotype × site interaction effects were significant ($p<0.05$) for the NT, NPP, NGP, percentage of filled grains (PFG), TGW, rice yellow mottle virus disease (RYMVD) resistance and GY. The variance due to the general combining ability (GCA) and the specific combining ability (SCA) effects were both significant for all assessed traits, indicating that both additive and non-additive gene actions were involved in governing trait inheritance. The high GCA to SCA ratios calculated for all the studied traits indicate that additive genetic effect was predominant. Parental lines, Mwangaza, Lunyuki, Salama M-57, Salama M-19, IRAT 256 and Salama M-55, which had negative GCA effects for RYMVD, and families such as SARO × Salama M-55, IRAT 245 × Rangimbili, Rangimbili × Gigante and Rangimbili × Mwangaza, which had negative SCA effects for RYMVD were selected for RYMV resistance breeding. The crosses such as Rangimbili × Gigante, Gigante × Salama M-19 and Rangimbili × Salama M-55 were selected due to their desirable SCA effects for GY. The predominance of additive gene effects for agronomic traits and RYMVD resistance in the present breeding populations suggest that rice improvement could be achieved through gene introgression using a recurrent selection method.

Overall, the present study resulted in selection of agronomically superior and RYMV resistant breeding parents and new rice families for further evaluation and variety release in Tanzania.

Declaration

I, William Titus Suvi, declare that

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
    b. Where their exact words have been used, then their writing has been placed in italics and inside quotation marks, and referenced.
5. This thesis does not contain text, graphics or tables copied and pasted from the Internet, unless specifically acknowledged, and the source being detailed in the thesis and in the references sections.

Signed:

……

William Titus Suvi

As the candidate's supervisors, we agree to the submission of this thesis:

……

Prof. Hussein Shimelis (Supervisor)

……..

Prof. Mark Laing (Co-Supervisor)

Acknowledgements

Thanks to the Almighty God for his abundant blessings and protection, guidance and continuous support during the whole period of my studies.

I express special thanks and appreciation to my main supervisor, Prof. Hussein Shimelis, for his patience, excellent supervisory and guidance roles, genuine and constructive criticisms, suggestions, contributions and co-operation, which in no doubt helped to make this work a success. My heartfelt appreciations are extended to my co-supervisor, Prof. Mark Laing, for his advice, encouragement, support, and valuable contribution for the completion of this thesis. Dr Learnmore Mwadzingeni, Dr Admire Shayanowako and Dr Isack Mathew are acknowledged for providing insights and comments to the proposal and literature write-up and draft chapters.

Special thanks go to the Alliance for a Green Revolution in Africa (AGRA) for granting a scholarship opportunity through the African Centre for Crop Improvement (ACCI) and the University of KwaZulu-Natal for enrolling me as a PhD Scholar.

I am grateful to the entire members of the academic and administrative staff at the ACCI in general, and especially Mrs. Rowelda Donnelly for her facilitation and logistical support during my study.

Thanks are due to the Permanent Secretary of the Ministry of Agriculture, Food Security and Cooperatives of the United Republic of Tanzania for granting me study leave.

My heartfelt thanks are extended to the centre director and staff of the Tanzania Agricultural Research Institute (TARI) - Ilonga for their tremendous contribution in making my study successful through administrative support and facilitation during my research.

I would also like to thank the District Agriculture, Irrigation and Cooperative Officers (DAICO) of Kilombero, Mvomero and Kyela for their help during the survey and data collection. Along with this, I would like to extend sincere gratitude to all interviewees who willingly accepted to be interviewed during the survey. I also acknowledge Dr Ashura Luzi-Kihupi, Sokoine University of Agriculture (SUA) for lending me their rice vacuum emasculator to assist in the making of crosses.

I am thankful to my beloved wife, Yustina Henry Lyandala, my daughter, Lackisha, my son Livingstone, and all family members for their tireless prayers, patience, understanding, inspiration and support throughout my study.

I am also indebted to Fred Martin (late), Agrey Kyando, Philemon Maweya, Sungwa Mbuga, Fadhil Paramana, Tarish Mustapha, Dorice said Shemkuya and Fabian Makuwa for field assistance during experiment establishment and data collection.

Dedication

I dedicate this thesis to:

1) To the Almighty God in Jesus Christ's name for giving me blessings and protection, guidance and continuous support during the whole period of my studies.
2) To my beloved wife Yustina Lyandala; my children, Lackisha Suvi and Livingstone Suvi;
3) To my house assistant Edda ng'hambi
4) To my parents, my late father, Titus William Suvi, and my mother, Celestina Conrad Mbuswa; and
5) To my brothers, sisters, nephews, and friends for their overall support during my studies.

TABLE OF CONTENTS

LIST OF FIGURES

LIST OF TABLES

LIST OF ABBREVIATIONS AND ACRONYMS

ACCI, African Centre for Crop Improvement

AFLP, Amplified fragments length polymorphism

AGRA, Alliance for a Green Revolution in Africa

AMOVA, Analysis of molecular variance

ANOVA, Analysis of variance

CV, Coefficient of variation

DAICO, District Agriculture, Irrigation and Cooperative Officers

DF, Degree of freedom

DFL, Days to 50% flowering

F_1, First filial generation

F_2, Second filial generation

FAO, Food and Agriculture Organisation of the United Nations

FGDs, Focus group discussions

GCA, General combining ability

GDP, Gross domestic product

GY, Grain yield

IRRI, International Rice Research Institute

LSD, Least significance difference

MAS, Marker assisted selection

NERICA, New Rice for Africa

NGP, Number of grains per panicle

NPP, Number of panicles per plant

NT, Number of tillers per plant

ORF, Open reading frame

PCA, Principal component analysis

PCR, Polymerase chain reaction

PFG, Percent filled grains

PH, Plant height

PIC, Polymorphic information content

PL, Panicle length

PRA, Participatory rural appraisal

RAPD, Random amplified polymorphism DNA

RFLP, Restricted fragment length polymorphism

RNA, Ribonucleic acid

RYMV, Rice yellow mottle virus

SCA, Specific combining ability

SNP, Single nucleotide polymorphisms

SSA, Sub-Sahara Africa

SSR, Simple sequence repeats

SUA, Sokoine University of Agriculture

TARI, Tanzania Agricultural Research institute

TE, Tris-EDTA

TGW, Thousand grain weight

Publication pertaining to this thesis

Chapter One

Suvi, W.T., H. Shimelis, M. Laing. 2018. Breeding rice for rice yellow mottle virus resistance in Sub-Saharan Africa: a review, Acta Agriculturae Scandinavica, Section B - Soil & Plant Science 69(2):1-8. doi: 10.1080/09064710.2018.1523454.

Chapter Two

Suvi, W.T., H. Shimelis, M. Laing. 2020. Farmers' perceptions, production constraints and variety preferences of rice in Tanzania. Journal of Crop Improvement. 1-18. doi:10.1080/15427528.2020.1795771.

Chapter Three

Suvi, W.T., H. Shimelis, M. Laing, I. Mathew, and A.I.T. Shayanowako. 2021. Variation among Tanzania rice germplasm collections based on agronomic traits and resistance to rice yellow mottle virus. Agronomy11, 391. https://dx.doi. org/10.3390/agronomy11020391.

Chapter Four

Suvi, W.T., H. Shimelis, M. Laing, I. Mathew, and A.I.T. Shayanowako. 2019. Assessment of the genetic diversity and population structure of rice genotypes using SSR markers. Acta Agriculturae Scandinavica, Section B- Soil & Plant Science 70: 76–86. doi: 10.1080/09064710.2019.1670859.

Chapter Five

Suvi, W.T., H. Shimelis, M. Laing, I. Mathew, and A.I.T. Shayanowako. 2021. Determining the Combining Ability and Gene Action for Rice Yellow Mottle Virus Disease Resistance and Agronomic Traits in Rice (*Oryza sativa* L.). Agronomy11, 12. https://dx.doi. org/10.3390/agronomy11010

THESIS INTRODUCTION

Background

Rice (*Oryza sativa* L., 2n = 2x = 24) is the second most important staple food crop in the world after wheat (*Triticum aestivum* L.) (FAO, 2015; Yelome et al., 2018). The global production of rice is 744.4 million tons per annum from an estimated area of 158.4 million hectares (FAO, 2017). Asia accounts for more than 90% of world rice production (Srujana et al., 2017). The bulk of rice produced (85%) is used for human consumption, compared with only 72% for wheat and 19% for maize (*Zea mays* L.) (FAO, 2013). Rice provides up to 50% of dietary calories and a substantial part of Asia's protein intake (Muthayya et al., 2014). In sub-Saharan Africa (SSA), rice consumption among urban dwellers has tripled from 9.2 to 31.5 million tons (Muthayya et al., 2014; USDA, 2018; Tsujimoto et al., 2019). This makes it the second-largest source of calories after maize in the region (van Oort et al., 2015). In response to the growing demand, total annual rice production in SSA increased from 11.58 to 14.5 million tons, contributing to 15% of the total cereal production (FAOSTAT, 2015). In addition, rice production, processing and marketing play important roles in providing employment opportunities and income for many households in Africa and Asia (Mghase et al., 2010). West Africa accounts for 70.4% of the rice produced in SSA, followed by East Africa (16.1%) and Central and Southern Africa (7.5%) (Del Villar and Lancon, 2015).

Tanzania produces about 1.1 million tons of rice per year, making it the second-largest rice producer after Madagascar in the East, Central, and South African region (FAOSTAT, 2010; Match maker, 2010). In Tanzania, rice is produced predominantly by small-scale farmers under both dryland and irrigated systems. The main rice production regions in Tanzania are Shinyanga, Mbeya, Morogoro, Mwanza, Tabora, and Rukwa. Most of the rice production in Tanzania is in the lowlands with 72% as rain-fed and 8% as irrigated production, with 20% produced by upland or dry-land rice systems (Kitilu et al., 2019).

Population growth, improved household incomes, urbanization, and changes in consumer preferences have significantly increased the demand for rice in Tanzania. Rice contributes about 37% of the gross domestic product (GDP) in the country (Hubert et al., 2017). Despite the high demand and potential of rice in Tanzania, the productivity of the crop is hindered by many factors including biotic and abiotic stresses, and numerous socio-economic constraints (Lamo et al., 2015; Suvi et al., 2018; Suvi et al., 2020). About 91% of the rice is produced by small-scale farmers who still rely on local varieties (Hubert et al., 2016), many of which are low yielding and susceptible to diseases. The average yield of rice in Tanzania is very low

(1.5 t ha^{-1}) compared to the potential yield in the region of 4.6 t ha^{-1}, and a mean yield of 8.48 t ha^{-1} reported in Asia and USA, respectively (Kilimo-Trust, 2012; FAO, 2015).

Constraints to rice production

Drought and heat stress, poor soil fertility, salinity, and iron toxicity are the key abiotic constraints affecting rice production in Tanzania (Mghase et al., 2010; Reynolds et al., 2015). Socio-economic factors such as a shortage of labour, lack of production inputs such as fertilizers and pesticides due to poorly developed distribution systems, frequent droughts and poorly developed irrigation systems, and obsolete production technologies are among the key constraints affecting rice productivity (Mghase, et al., 2010). Persistent insect pests and diseases, and high levels of weed infestation are important biotic constraints frequently encountered in the rice production systems in Tanzania. The most important diseases of rice in Tanzania include rice yellow mottle virus (RYMV), bacterial leaf spot (*Xanthomonas* oryzae pv. *oryzae*), rice blast (*Pyricularia grisea*) and brown leaf spot (*Cochliobolus miyabeanus*) (Chuwa et al., 2015; Duku et al., 2016; Suvi et al., 2018).

The RYMV disease causes yield losses ranging from 20% to 100% in the susceptible rice varieties that are currently grown by small-scale farmers in Tanzania (Luzi-Kihupi et al., 2009; Longué et al., 2016). RYMV is a single-stranded, positive-sense RNA virus (ssRNA) belonging to the genus Sobemovirus (Hull and Fargette, 2005). The disease is widespread in almost all the rice-growing regions in both rain-fed lowland and irrigated ecosystems (Zouzou et al., 2008; Ndikumana et al., 2011; Pinel-Galzi et al., 2016). The disease affects all susceptible local and introduced varieties (Kouassi et al., 2005). RYMV infected rice plants show mottling and yellowing symptoms. The symptoms may resemble iron or nitrogen deficiency, or iron toxicity. The disease also causes stunted growth, reduced tillering ability, non-synchronous flowering, poor panicle exertion, and brown to dark brown discolouration of grains (Kouassi et al., 2005; Sereme et al., 2016). The disease interferes with the accumulation of carbohydrates necessary for spikelets development. It also triggers pollen degeneration and drying up of the stigma, resulting in spikelet sterility (Onwughalu et al., 2011). Diverse RYMV strains are distributed in Tanzania (Banwo et al., 2004; Kanyeka et al., 2007; Hubert et al., 2017). Strain S5 is found in the Morogoro region, while S4 and S6 are found throughout the country (Abubakar et al., 2003; Kanyeka et al., 2007). The disease is prevalent in almost all rice growing areas, causing major yield losses, which may be exacerbated by the susceptibility of the rice genotype, the earliness of infection and the viral strain, and their interactions (Longué et al., 2016). Therefore, the development of novel cultivars of rice that carry RYMV resistance and farmer-preferred agronomic traits is the major goal of the regional rice breeding efforts in order to bolster yields and ensure food security.

RYMV control methods

Various control strategies (e.g. cultural practices, crop protection chemicals and resistant cultivars) have been recommended for RYMV disease management (Traore et al., 2015; Suvi et al., 2020). A number of cultural control practices have been proposed including manipulating planting dates, rogueing of diseased plants, crop rotation, removal of rice residues and ratoons, periodically disinfecting farm tools, minimizing inter-plot infection, and field inspection and isolation. The aim of these cultural practices is to reduce disease incidence and spread. However, their adoption among smallholder farmers remains low. Cultural control requires substantial labour inputs, yet it is relatively ineffective in reducing the spread of the virus, making it an unattractive option for the majority of farmers in developing countries (Suvi et al., 2018). Chemical control is effective when applied during the early crop growth stage to reduce the population of RYMV insect vectors (Traore et al., 2015). However, the presence of diverse alternate host plants harbouring a number of insect vectors limits the efficacy of chemical pesticides under field conditions. Also, the repeated use of pesticides can lead to a build-up of pesticide resistance by the insect pests. The continuous application of pesticides is also associated with severe health risks and environmental pollution. In addition, pesticides and their application equipment are too expensive for most small-scale farmers (Suvi et al., 2018). Consequently, the deployment of varietal resistance is considered as the most economical and environmentally sustainable method to control RYMV disease (Thiemélé et al., 2010; Sow, 2012; Kam et al., 2013). The deployment of RYMV resistant cultivars would be suitable for communal and subsistence rice production systems because of the low cost, ease of implementation and compatibility with other integrated disease management systems.

Breeding for RYMV resistance in Rice

The development and deployment of RYMV resistant rice cultivars have the potential to reduce the impact of the disease on rice production in RYMV endemic areas. The presence of RYMV strains with a diversity of virulence genes requires an understanding of the genetic basis conditioning RYMV resistance and associated agronomic traits. Successful breeding for RYMV resistance depends on the availability of sources of resistance genes as well as effective phenotyping and pathotyping methods, an understanding of the pattern of inheritance of resistance, and the adoption of the most suitable breeding methods for exploiting available genetic variations to create durable resistance. Most of the introduced rice varieties available in Tanzania have not been widely adopted by farmers because they lack farmer-preferred agronomic and quality traits. Landraces or farmers' varieties of rice that express farmer-preferred traits have not previously been used as the primary source of genetic variation to initiate pre-breeding in Tanzania.

Breeding for high-yielding and RYMV resistant varieties could have a significant economic effect if it were to be based on a demand-led approach. Rice breeding should consider the demands of the end-users for quality traits such as cooking and eating quality, grain shape, and aroma (Mogga et al., 2018, Suvi et al., 2020). Farmer-preferred traits are considered to be the major drivers for the widespread adoption of a new variety. Hence, RYMV resistance breeding programs should integrate farmer preferred traits and tolerance to other production constraints. This requires adequate genetic variation to select breeding parents through genetic diversity analyses. Understanding genetic diversity and relatedness is an important component of crop improvement because it allows for an informed selection of diverse parents that are required to generate recombinants and transgressive segregants with superior performance. It is also imperative to understand combining ability effects, gene action and inheritance of RYMV resistance and agronomic traits because these are major determinants of the selection procedure to maximize genetic gain (Acquaah, 2012). Therefore, this study aims to develop high yielding rice varieties with farmer-preferred traits coupled with durable resistance to the dominant RYMV strains in Tanzania.

Overall objective

The overall objective of this study was to develop high-yielding rice genotypes with resistance to the rice yellow mottle virus disease in Tanzania. To attain this objective, the specific objectives included the following.

Specific objectives

The study had the following specific objectives:

i. To assess farmers' perceptions, production constraints, and variety preferences of rice in Tanzania to guide breeding.
ii. To determine variation among Tanzania rice germplasm collections, based on agronomic traits and resistance to rice yellow mottle virus, aiming to select unique parents for breeding.
iii. To assess the genetic diversity and population structure of a selected population of rice genotypes using simple sequence repeat (SSR) markers to complement phenotypic data.
iv. To determine the combining ability and gene action for rice yellow mottle virus disease resistance and agronomic traits in rice (O*ryza sativa* L.) and to develop new populations of rice progenies for future breeding.

Research hypotheses

The main hypotheses of the study were:

i. Farmers' have different perceptions, production constraints, and variety preferences for rice in Tanzania.
ii. There will be differential expressions for RYMV resistance and agronomic traits among the selected rice germplasm.
iii. There exists genetic variability among selected rice genotypes that can be exploited in breeding for RYMV resistance, agronomic traits and increased grain yield.
iv. The selected parents and their progenies have adequate general combining ability and specific combining ability as a basis to breed RYMV resistant cultivars that include good agronomic and farmer-preferred traits.

Outline of the thesis

This thesis consists of six distinct chapters (Table 0.1) following a number of activities related to the above objectives. Chapter 1 is written as a separate review paper, while Chapters 2 to 5 are written in the form of research chapters. Chapter 6 gives a general discussion of the results of the respective chapters and conclusions, and identifies future research directions. Each of these chapters follows the format of a publishable paper. The format of a published chapter follows the formatting protocols of the journal in which it was published. This format follows the dominant thesis format adopted by the University of KwaZulu-Natal. Consequently, there is inevitable repetition of references and introductory information between some chapters. Chapter 1 and 4 have been published in the Journal of Acta Agriculturae Scandinavica, Section B-Soil & Plant Science. Chapter 2 has been published in the Journal of Crop Improvement, and Chapter 3 and 5 have been published in the Journal of Agronomy.

Table 0.1. Thesis outline

Chapter	Title
-	Thesis introduction
1	A review: Breeding rice for rice yellow mottle virus resistance
2	Farmers' perceptions, production constraints and variety preferences of rice in Tanzania to guide breeding
3	Variation among Tanzania rice germplasm collections based on agronomic traits and resistance to rice yellow mottle virus to select unique parents for breeding
4	Assessment of the genetic diversity and population structure of rice genotypes using SSR markers to complement phenotypic data and select parents
5	Combining ability and gene action for rice yellow mottle virus disease resistance and agronomic traits in rice (O*ryza sativa* L.) and develop new populations for future breeding
6	An overview of the research findings

References

Abubakar, Z., F. Ali, A. Pinel-Galzi, O. Traoré, P. N'Guessan, J. Notteghem, F. Kimmins, G. Konaté, and D. Fargette. 2003. Phylogeography of rice yellow mottle virus in Africa. Journal General Virology 84: 733-743. doi:10.1099/vir.0.18759-0.

Acquaah, G. 2012. Principles of Plant Genetics and Breeding, 2nd edition, Wiley-Blackwell, Oxford, UK

Banwo, O.O., M.D. Alegbejo, and M.E. Abo. 2004. Rice yellow mottle virus genus Sobemovirus: A continental problem in Africa. Journal of Plant Protection Science 40: 26-36.

Chuwa, C.J., R.B. Mabagala, and S.O.W.M. Reuben. 2015. Pathogenic variation and molecular characterization of *Pyricularia oryzae*, causal agent of rice blast disease in Tanzania. International Journal of Science and Research 4(11): 1131-1139.

Del Villar, P.M., and F. Lançon. 2015. West African rice development: Beyond protectionism versus liberalization? Global Food Security 5: 56-61.

Duku, C., A.H. Sparks, and S.J. Zwart. 2016. Spatial modelling of rice yield losses in Tanzania due to bacterial leaf blight and leaf blast in a changing climate. Climatic Change 135 (3): 569 -583.

FAO. 2013. http://faostat.fao.org/default.aspx. (Accessed 3 January 2018).

FAO. 2017. Rice market monitor. http://faostat.fao.org/default.aspx. (Accessed 28 September 2020).

FAOSTAT. 2010. http://faostat.fao.org/default.aspx. (Accessed 17 March 2020).

FAOSTAT. 2015. The Rice Value Chain in Tanzania. A Report from the Southern Highlands Food Systems Programme. http://faostat.fao.org/default.aspx. (Accessed 5 August 2018).

Hubert, J., A. Luzi-Kihupi, E. Hébrard, and H.J.F Lyimo. 2017. Pathogenic variation and occurrence of multiple resistance-breaking rice yellow mottle virus strains in Tanzania. African Journal of Biotechnology 16 (24): 1331-1337.

Hubert, J., A. Luzi-Kihupi, E. Hébrard, and H.J.F. Lyimo. 2016. Farmers' knowledge and perceptions of rice yellow mottle virus in selected rice growing areas in Tanzania. International Journal of Science and Research 5: 549-559.

Hull, R., and D. Fargette. 2005. *Sobemovirus*. In: Fauquet C.M., M.A. Mayo, J. Maniloff, U. Desselberger, and L.A. Ball (Eds). Virus Taxonomy: Eighth Report of the International Committee on Taxonomy of Viruses. Elsevier, London, pp. 885 - 890.

Kam, H., M.D. Laing, Y. Séré, D.Thiémélé, A. Ghesquière, N. Ahmadi, and M.N Ndjiondjop. 2013. Evaluation of a collection of rice landraces from Burkina Faso for resistance or tolerance to rice yellow mottle virus. Journal of Plant Pathology 95: 485-492.

Kanyeka, Z.L., E. Sangu, D. Fargette, A. Pinel-Galzi, and E. Hébrard. 2007. Distribution and diversity of local strains of rice yellow mottle virus in Tanzania. African Journal of Crop Science 15: 201-209.

Kilimo-Trust (2012). Expanding Rice Markets in the EAC: An Opportunity for Actors in the Value Chain. East African Community, 45 p.

Kitilu, M.J.F., A.M.S. Nyamora, and J. Charles. 2019. Growth and yield performances of selected upland and lowland rain-fed rice varieties grown in farmers and researchers' managed fields at Ifakara, Tanzania. African Journal of Agriculture Research 14 (4):197-208.

Kouassi, N.K., P. N'Guessan, L. Albar, C.M. Fauquet, and C. Brugidou. 2005. Distribution and characterization of rice yellow mottle virus: A threat to African farmers. Plant Disease 89: 124-133.

Lamo, L., G. Cho, I. Jane, P.K.A. Dartey, E. James, M. Ekobu, S. Alibu, S. Okanya, B. Oloka, M. Otim, G. Asea, and K. Kang. 2015. Developing lowland rice germplasm with resistance to multiple biotic stresses through anther culture in Uganda. The Korean Society Journal of International Agriculture 27: 415-420.

Longué, R.D.S., I. Zinga, S. Semballa, N. Barro, and O. Traoré. 2016. Detection and serological characterization of rice yellow mottle virus in Central African Republic. Agricultural Sciences 7: 911-919.

Luzi-Kihupi, A., J.A. Zakayo, H. Tusekelege, M. Mkuya, N.J.M. Kibanda, K.J. Khatib, and A. Maerere. 2009. Mutation breeding for rice improvement in Tanzania. In: Induced Plant Mutations in the Genomics Era. FAO, Rome, pp. 385-387.

Match Maker Associates. 2010. Value Chain Analysis of Rice and Maize in Selected Districts in Tanzania, Tap Rice and Maize Context Report. Dar es Salaam, Tanzania.

Mghase, J.J., H. Shiwachi, K. Nakasone, and H. Takahashi. 2010. Agronomic and socio-economic constraints to high yield of upland rice in Tanzania. African Journal of Agriculture Research 5: 150 -158.

Mogga, M., J. Sibiya, H. Shimelis, J. Lamo, and N. Ochanda. 2018. Appraisal of major determinants of rice production and farmers choice of rice ideotypes in South Sudan: Implications for breeding and policy interventions. Experimental Agriculture 55(1):143-156. doi: 10.1017/s0014479718000017.

Muthayya, S., J.D. Sugimoto, S. Montgomery, and G.F. Maberly. 2014. An overview of global rice production, supply, trade, and consumption. Annals of the New York Academy of Science 1324: 7-14. doi: 10.1111/nyas.12540.

Ndikumana, I., R. Gasoré, S. Issaka, A. Pinel-Galzi, A. Onasanya, A. Hassani-Mehraban, D. Fargette, D. Peters, and Y. Séré. 2011. Rice yellow mottle virus in rice in Rwanda: first report and evidence of strain circulation. New Disease Reports 23: 18. doi:10.5197/j.2044-0588.2011.023.01.

Onwughalu, J.T., M.E. Abo, J. Okoro, K.A. Onasanya, and Y. Sere. 2011. Rice yellow mottle virus infection and reproductive losses in rice (*Oryza sativa* L.). Trends in Applied Sciences Research 6: 182-189.

Pinel-Galzi, A., C. Dubreuil-Tranchant, E. Hébrard, C. Mariac, A. Ghesquière, and L. Albar. 2016. Mutations in rice yellow mottle virus polyprotein P2a involved in RYMV2 gene resistance breakdown. Frontiers in Plant Science 7: 1779. doi: 10.3389/fpls.2016.01779.

Reynolds, T.W., S.R. Waddington, C.L. Anderson, A. Chew, Z. True, and A. Cullen. 2015. Environmental impacts and constraints associated with the production of major food crops in Sub-Saharan Africa and South Asia. Food Security 7: 795-822.

Sereme, D., I. Ouedraogo, B.J. Neya, P.E. Zida, N. Yao, and M. Sie. 2016. Screening improved rice varieties (*Oryza spp*) for their resistance /tolerance to rice yellow mottle virus in West Africa. International Journal Agriculture Innovation Research 5: 481-486.

Sow, M.E. 2012. Genetic Diversity of *Oryza species* in Niger; Screening and Breeding for Resistance to Rice Yellow Mottle Virus (RYMV). PhD Thesis, University of KwaZulu-Natal, 217 p.

Srujana, G., B.G. Suresh, G.R. Lavanya, J.B. Ram, and V. Sumanth. 2017. Studies on genetic variability, heritability and genetic advance for yield and quality components in rice (*Oryza sativa* L.). Journal Pharmacognosy and Phytochemistry 6:564-566.

Suvi, W.T., H. Shimelis, and M. Laing. 2018. Breeding rice for rice yellow mottle virus resistance in sub-Saharan Africa: a review, Acta Agriculturae Scandinavica, Section B-Soil & Plant Science 69 (2): 1-8. doi:10.1080/09064710.2018.1523454.

Suvi, W.T., H. Shimelis, and M. Laing. 2020. Farmers' perceptions, production constraints and variety preferences of rice in Tanzania. Journal of Crop Improvement. 1-18. doi:10.1080/15427528.2020.1795771.

Thiémélé, D., A. Boisnard, M.N. Ndjiondjop, S. Chéron, Y. Séré, S. Aké, A. Ghesquière, and L. Albar. 2010. Identification of a second major resistance gene to rice yellow mottle virus, RYMV2, in the African cultivated rice species, *O. glaberrima*. Theoretical and Applied Genetics 121: 169 -179.

Traore, V.S.E., M.D. Asante, V.E. Gracen, S.K. Offei, and O. Traore. 2015. Screening of rice accessions for resistance to rice yellow mottle virus. American Journal of Experimental Agriculture 9(4): 1-12.

Tsujimoto, Y., T. Rakotoson, A. Tanaka, and K. Saito. 2019. Challenges and opportunities for improving N use efficiency for rice production in sub-Saharan Africa. Plant Production Science 22(4): 413-427. doi: 10.1080/1343943X.2019.1617638.

USDA. 2018. Production, supply and distribution online. Retrieved from https://apps.fas.usda.gov/psdonline/app/index.

Van Oort, P. A. J., K. Saito, A. Tanaka, E. Amovin-Assagba, L.G.J. Van Bussel, J. van Wart, and M.C.S. Wopereis. 2015. Assessment of rice self-sufficiency in eight African countries in 2025. Global Food Security 5: 39-49.

Yelome, O. I., K. Audenaert, S. Landschoot, A.D.W. Vanhove, D. Silue, P. Van Damme, and G. Haesaert. 2018. Combining high yields and blast resistance in rice (*Oryza spp*.): A screening under upland and lowland conditions in Benin. Sustainability. 10: 2500. doi: 10.3390/su10072500.

Zouzou, M., T.H. Kouakou, M. Kone, and S. Issaka. 2008. Screening rice (*Oryza sativa* L.) varieties for resistance to rice yellow mottle virus. Scientific Research and Essay 3: 416-424.

CHAPTER ONE: A REVIEW OF LITERATURE

Abstract

Rice (*Oryza sativa* L.) is the world's second most produced staple cereal crop after wheat (*Triticum aestivum* L.). Currently, rice production and consumption have steadily increased in sub-Saharan Africa (SSA). To date, rice is the largest imported commodity crop in the region. The low productivity is due to a number of biotic and abiotic stresses, and socio-economic constraints. Among the biotic constraints, rice yellow mottle virus (RYMV) is the most important constraint in SSA, causing yield losses ranging from 20% to 100%. Various control strategies (host resistance, cultural practices and chemicals) have been recommended to manage RYMV disease. The management of this disease through generic crop protection chemicals is not economic nor is it successful due to the presence of a large number of vector species spreading the virus. In addition, cultural practices are ineffective against RYMV because the virus is spread by several agents including insect vectors. The use of RYMV resistant cultivars remains the most effective, economic and environmentally friendly method for resource poor farmers. However, RYMV resistant varieties have not yet been developed and deployed in SSA including Tanzania. The aim of this review was to present the main components in the development of rice cultivars with RYMV disease resistance. The paper provides a comprehensive review on the genetic variability of the RYMV, its epidemiology and control measures, and the gene action responsible for RYMV resistance. The review also summarises complementary genomic tools useful in RYMV disease resistance breeding. Successful breeding of rice for RYMV resistance depends on the availability of genes for stable resistance, knowledge of the genetics of the host, and the availability of efficient phenotyping and pathotyping methods, and an understanding of the genes involved and their pattern of inheritance. Information presented in the review can serve as a reference guide for rice breeding emphasising RYMV resistance, high yields and farmers-preferred traits.

Keywords: Resistance breeding, rice, rice yellow mottle virus, sub-Saharan Africa

This chapter was published in the Journal of Acta Agriculturae Scandinavica, Section B - Soil & Plant Science Suvi, W.T., H. Shimelis, M. Laing. 2018. Breeding rice for rice yellow mottle virus resistance in Sub-Saharan Africa: a review, Acta Agriculturae Scandinavica, Section B - Soil & Plant Science 69(2):1-8. doi: 10.1080/09064710.2018.1523454.

1.1 Introduction

Rice [*Oryza sativa* (L.), 2n= 2x = 24] is a staple food crop supporting more than half of the world's population. It is the second most important cereal crop after wheat (*Triticum aestivum* L) in terms of total production (Bagati et al., 2016). Global annual production of rice is 744.4 million tons from an estimated area of 158.4 million hectares of agricultural lands (FAO, 2017). Rice is a major source of calories and protein for humans (Lussewa et al., 2016). Nearly 85% of the total world rice production is destined to human consumption compared with wheat and maize of which 72% and 19% are used for food, in that order (FAOSTAT, 2012). Asia supports 59% of the world's population through rice production and market place, while it produces and consumes more than 90% of global rice output (Muthayya et al., 2014; Srujana et al., 2017).

In sub-Saharan Africa (SSA), rice production and consumption have steadily increased over the past 20 years (Ogunbayo et al., 2014). In the region, rice has become a staple food for millions of people and constitutes a major part of the diet (Atera et al., 2011; Maclean et al., 2013). Rice production and marketing is reported to be the source of income and employment opportunity along the value chains for millions of households in SSA. In the region, annual rice production is estimated to vary from 11.58 to 14.5 million tons, contributing 15% of SSA's total cereal production (FAOSTAT, 2015). The major rice producing regions in SSA is West Africa contributing to 70.4% of total production, followed by East Africa (16.1%) and Central and Southern Africa (7.5%) (Del Villar and Lancon, 2015). In SSA, rice is cultivated mainly under dryland farming systems, contributing to 38% of the total cultivated area, followed by rain-fed wetland (33%), irrigated wetland (20%) and deep water and mangrove swamps (9%) production systems (Balamurugan and Balasubramanian, 2017).

Tanzania is the second largest rice producer and consumer after Madagascar in East, Central and Southern Africa (ECSA) region with total annual production of more than 1.1 million tons (Match Maker Associates 2010). In Tanzania, rice is the second most important staple food next to maize and is grown by more than 18% of the farming households (Bucheyeki et al., 2011). The rice is mainly grown in the regions of Morogoro, Shinyanga, Mbeya, Mwanza, Tabora, Kilimanjaro and Rukwa. The crop is grown mainly by small-scale farmers as food and cash crop. About 74% of rice production is under rain-fed condition, 20% in upland and 6% under irrigation (EUCORD, 2012). About 42% of the rice produced in Tanzania is marketed, compared to 28% and 18% for maize and sorghum, respectively (MAFAP, 2013). Population growth, increased household incomes, urbanisation, diverse consumer preferences, changes in the dietary habit in favour of rice have significantly increased the demand for rice in SSA, more than elsewhere globally. Despite increased production and

growing demands for rice, the productivity of the crop is relatively low in SSA including Tanzania, with mean yields of 1.5 to 2.5 t ha^{-1} compared with mean yields of 4.6 t ha^{-1} and 8.48 t ha^{-1} reported in Asia and USA, respectively (FAOSTAT, 2015; Atera et al., 2018). The low productivity of rice in SSA including Tanzania is attributed to a number of biotic and abiotic stresses and socio-economic constraints. Rice diseases, including rice yellow mottle virus (RYMV), rice blast (*Pyricularia grisea*), bacterial leaf blight (*Xanthomonas campestris pv. oryzae*) and brown leaf spot (*Cochliobolus miyabeanus*) and insect pests (e.g. African rice gall midge (*Orseolia oryziovora*; Diptera: Cecidomyiidae) are the main yield limiting biotic constraints (Sie et al., 2012; Drame et al., 2013). Moreover, abiotic stresses such as poor soil fertility, salinity and drought are other key factors affecting rice yields.

RYMV is the most important viral disease in most rice growing regions. Most severe RYMV infections are reported under rain-fed and irrigated lowland rice production agro-ecologies. RYMV causes pronounced crop damage from seedling to booting growth stages. RYMV causes yield losses ranging from 20% to 100% in susceptible rice varieties that are currently grown by small-scale farmers in SSA (Luzi-Kihupi et al., 2001; Longué et al., 2016). Yield losses depend on plant growth stage at the onset of disease infection, level of resistance/ susceptibility of rice variety and environmental conditions and their interaction (Kouassi et al., 2005; Kam et al., 2013).

Several RYMV control strategies are internationally recommended, including the use of resistant varieties, application of the cultural practices and spraying crop with chemicals (Kouassi et al., 2005; Zouzou et al., 2008). The use of crop protection chemicals has contributed to improved rice production by controlling RYMV transmitting vectors (Traore et al., 2015). However, chemical control of RYMV is not economic and it may be unsuccessful due to the presence of many vectors of RYMV (Traore et al., 2009). In addition, the high cost of implementing chemical control measure hinders its adoption by smallholder farmers. Chemical control has led to the development of resistant insect populations due to mutation events. Furthermore, chemicals may not be safe for farmers and may cause environmental pollution (Hashmi and Khan, 2011).

Cultural control practices (e.g. removal of crop residues, synchronous planting, shifting nursery sites, early transplanting, rogueing of infected plants and reduced use of fertilisers on infected plots) are widely used by rice farmers. However, RYMV can easily spread by irrigation water or rainfall and other vectors such as Chrysomelid beetle and grasshoppers, limiting the value of these strategies in controlling the disease.

Use of RYMV resistant rice varieties is considered to be the most effective, economic and environmentally friendly method for RYMV disease management, especially for smallholder farmers. All current rice varieties grown in SSA have succumbed to RYMV infection (Kouassi et al., 2005). There is a need to develop and deploy RYMV resistant varieties in SSA. Successful breeding for RYMV resistance depends on the availability of effective sources of resistance, understanding of the genetics of the host and the causative agent, availability of effective phenotyping and pathotyping methods, knowledge of the genes conditioning resistance and the pattern of inheritance, and the choice of a suitable breeding method. Therefore, the aim of this review was to present the main components in the development of rice cultivars with RYMV disease resistance. The review highlights the genetic variability of the RYMV, its epidemiology and control measures, and the gene action responsible for RYMV resistance. The review also summarises complementary genomic tools intentionally useful in RYMV disease resistance breeding.

1.2 Description of rice yellow mottle virus

RYMV belongs in the genus Sobemovirus (Hull and Fargette, 2005). It is an icosahedral particle of 30 nm in diameter that contains a single strand, positive-sense genomic RNA (ssRNA), with attributes peculiar to the members of this genus (Tamm and Truve, 2000; Kouassi et al., 2005). Through extensive sequencing of various isolates (Fargette et al., 2004), the genome organisation of RYMV has been found to be 4452 nucleotides (nt) with the following coding sequences from 5′ to 3′: open reading frame (ORF1), ORFx, ORF2a, ORF2b and ORF3 (Ling et al. 2013). ORF1 is 17.8 Da region that codes for protein movement (P1) and suppresses gene silencing. The ORFx has an unknown function but is needed to establish infection. ORF2b is translated via frameshifting and ORF3 via sub-genomic ribonucleic acid (RNA). The ORF2a and ORF2b, encode for a polyprotein and putative proteins. The ORF3 encodes for a coat protein (CP) of 239 aa (26 kDa) (Ling et al., 2013).

1.3 Genetic variation and distribution of RYMV

Knowledge of the genetic variability present among RYMV strains is important to designing resistance breeding and gene deployment programmes. Molecular variants and serological differences between RYMV strains have been reported (Fargette et al. 2002). RYMV has a high level of genetic diversity. Several serotypes and strains of RYMV have been identified at various geographical locations in Africa. Based on the genomic analysis and serological differences, five serotypes have been identified in Africa (Pinel et al., 2000). These include Serotype 1, Serotype 2 and Serotype 3 that are predominantly found in West and Central Africa, and Serotype 4, Serotype 5 and Serotype 6 found in East Africa (Kouassi et al., 2005; Kanyeka at el., 2007). In SSA, RYMV was first detected in 1966 in the Otonglo area, Kenya,

along with the shores of Lake Victoria (Bakker 1974). Later, it spread to all the rice-growing countries of West Africa, East African, Madagascar and Mozambique. Presently RYMV is found in most SSA countries, as shown in Figure 1.1.

Figure 1.1. Map showing the distribution of rice yellow mottle virus in various countries in Africa.

Note: countries with RYMV epidemics are labelled (adapted from Banwo, 2002).

1.4 Epidemiology and transmission of RYMV

The RYMV pathogen has been found in the two cultivated rice species (*O. glaberrima and O. sativa*) and two wild rice species (*O. longistaminata and O. barthii*). Several wild grasses such as jungle rice (*Echinochloa colona*), creeping grass (*Panicum repens*), elastic grass (*Erasgrostis tenuifolia*) and viper grass (*Dinebra retroflexa*) are also hosts of the virus. RYMV can be transmitted by either insect vectors or mechanical agents (Bakker, 1971; Konate et al., 1997). Beetles belonging to the Chrysomelidea family such as *Sesselia pusilla, Chaetocnema pulla, Trichispa sericea* and *Dicladispa viridicyanea*, as well as the grasshopper *Conocephalus merumontanus* are known vectors of the virus (Abo et al., 1998). RYMV can be transmitted by wind mediated leaf contact, contaminated hands of field workers, and contact-transmission by domestic or wild animals. Transplanting of rice into a field in which infected rice seed from a previous crop has germinated can be another source of RYMV transmission to a healthy crop (Woin et al., (2007). In addition, the virus can be

transmitted through irrigation water or by humans during field activities such as weeding or fertiliser application (Abo et al., 2000; Pinel-Galzi et al., 2016).

1.5 Symptoms of RYMV

The leaves of RYMV infected rice plants show mottling and yellowing symptoms depending on disease severity and the reactions of various genotypes. These symptoms may resemble iron or nitrogen deficiency, or iron toxicity (Abo et al., 2005; Onasanya et al., 2009). Once infected with RYMV, rice plants show stunted growth, reduced tillering ability, non-synchronous flowering, poor panicle exertion and brown to dark brown discolouration of grains (Sereme et al., 2016). Under severe infection and disease development, plants may develop conspicuous bronze or orange pigmentation, followed by leaf rolling and leaf desiccation, leading to complete crop failure (Hubert et al., 2016). Disease development after inoculation is manifested by the appearance of yellow-green spots on the youngest leaves (Munganyinka et al., 2016; Sereme et al., 2016). Resistant rice genotypes may not show distinctive symptoms when compared to susceptible controls (Sereme *et al*., 2016). RYMV incidence and severity is dependent on rice genotype, the growing environment, the virulence of viral strains and the stage of infection.

1.6 Control strategies of RYMV disease

RYMV is a difficult plant disease to control, especially under the complex farming systems prevalent in SSA (Nwilene et al., 2009). RYMV survives under harsh weather conditions compared with other common viruses such as the African cassava mosaic virus, maize streak virus, groundnut rosette virus, or tomato yellow leaf curl virus (Abo et al., 2000). Various control measures are internationally recommended for the management of RYMV disease. These include cultural, chemical and biological approaches, which are briefly described below.

1.6.1 Cultural control

Cultural control depends on managing the rice agroecosystem to create a growing environment unfavourable for insect vectors, and to make it ideal for crop growth and development (Abo et al., 2004). This approach aims to minimise disease incidence and damage. The following are the common cultural control methods of RYMV: optimal planting date, rogueing of diseased plants, crop rotation, removal of rice residues and ratoons, disinfection of farm tools, minimising inter-plot infection and field inspection and isolation (Salaudeen, 2014). Reduced level of fertiliser application, growing diverse varieties or multi-lines on a single plot, changing nursery sites and phytosanitary measures can prevent the introduction of virulent viral strains into another rice production region (Traore et al., 2015).

However, successful implementation and adoption of cultural practices by smallholder farmers remain low due to several practical and socio-economic hindrances. The method is less effective in reducing the spread of the virus making it a less practical option for the majority of farmers in developing countries. Furthermore, cultural practices may alter crop value or gross income e.g. delayed planting date and market supply.

1.6.2 Chemical control

Crop protection chemicals are widely used to control RYMV vectors such as *Sesselia pusilla, Chaetocnema pulla and Trichispa sericea*, as well as the grasshopper, *Conocephalus merumontanus*. The most commonly used insecticides include Decis, Karate, Super Gro, lambda-cyhalothrin, abamectin and diazinon. Chemical control of vectors is effective when applied during the early crop growth stage to reduce the insect population. However, the presence of diverse alternative host plants harbouring insect vectors limits the value of this technique under field conditions. Repeated use of crop protection chemicals can lead to a build-up of chemical resistance by the insect pests, requiring a search for a new generation and effective chemicals. Moreover, chemical control measures are expensive and not environmentally friendly (Shelepchikov et al., 2008), limiting their application under resource poor and smallholder production systems such as in Africa and Asia.

1.6.3 Biocontrol control

Biocontrol involves the use of living organisms to control the population of pests. This approach has not been widely used to control vectors for RYMV in rice production. Woin et al. (2007) reported the potential of biological control method to control RYMV vectors. The authors indicated that bio-agents such as *Eurytoma spp* and *Pediobius spp* decreased the population of the RYMV vectors such as *Chaetocnema pulla* and *Oxya hyla*, respectively. Further research is needed to explore on the use of predators and parasitoids as biocontrol agents against RYMV vectors in rice.

1.6.4 Host resistance

Breeding for resistance to RYMV disease is based on the identification and incorporation of resistance genes into economically important and susceptible varieties. Genes conditioning RYMV resistance have been reported by several workers (Ndjiondjop et al., 1999; Traore et al., 2015; Sereme et al., 2016). To date, two RYMV resistance genes including RYMV1 and RYMV2 are reported globally (Ndjiondjop et al., 1999; Thiemele et al., 2010). Expression of RYMV resistance genes is subject to the rice genotype, environment and their interaction. In most African countries including Tanzania, RYMV resistant cultivars are yet to be developed and deployed to farmers. Key aspects in breeding for RYMV resistance is an understanding

of the mode of gene action and the number of genes conditioning resistance for the prevailing RYMV strains. This will guide selection and development of parental of populations for use in breeding programmes. There is also a need for continuous disease surveillance and pathotyping of the current virus strains to ensure the usefulness of RYMV resistance screening. Figure 1.1 is a map showing the distribution of RYMV in various countries in Africa. Most rice cultivars grown globally are derivatives of *O. sativa*. The majority of these cultivars are highly susceptible to RYMV. Breeding rice for RYMV tolerance is a breeding strategy to create cultivars that yield well despite being RYMV susceptible. Measuring virus load present in xylem parenchyma cells and sieve elements is one method to assess tolerance to RYMV in rice (Opalka et al., 1997).

Complete (high) and incomplete (partial) resistance to RYMV has been reported to depend on the rice genotype, virulence of viral strains, the environment and their interaction (Salaudeen, 2014). Partial resistance is conditioned by minor genes. It is characterised by low virus titres (virus accumulation) at early stages of infection and delayed symptom development. Partial resistance is quantitative and is polygenic. Markers targeting eight regions of the rice genome have been used to map quantitative trait loci influencing partial RYMV resistance. Complete resistance is associated with a lack of symptom development, undetectable virus content and blockage of virus movement. Completely resistant cultivars that are genetically monogenic, with recessive inheritance have been identified (Ndjiondjop et al., 1999; Thiemele et al., 2010). RYMV1 was the first resistance gene described in rice (Ndjiondjop et al., 1999; Pinel-Galzi et al., 2016), and it has been mapped onto chromosome 4. It controls resistance in a recessive way and encodes eIF (iso) 4G1, a translation initiation factor. This gene is responsible for the resistance present in *O. glaberrima* accessions Tog5681, Tog5672 and Tog5674, whose alleles Rymv1-3, Rymv1-4 and Rymv1- 5, respectively, are distinct from each other and from that of another resistant cultivar, Gigante. Most of the resistance genes to RYMV come from *O. glaberrima*. In addition, RYMV2 and RYMV3 resistance genes have been identified on *O. glaberrima* (Pinel Galzi et al., 2016). Therefore, the above candidate genes are important in breeding for RYMV resistance, high yielding and farmer preferred rice varieties in SSA.

The existence of high genetic variation of RYMV may be associated with the emergence of pathogen virulence and new strains. New strains emerge through genetic mutation and recombination. Such strains are capable of overcoming the resistance of commercial rice varieties. Pathogen variability and adaptability has seriously affected efforts of breeding RYMV disease resistant rice varieties. The highly resistant rice cultivar Gigante has been reported to be effective against a range of different RYMV strains from Central and West

Africa (Ndjiondjop et al., 1999). In Tanzania, RYMV strain S6 has been reported to break the resistance of rice cultivar Gigante (Pinel-Galzi et al., 2007). Furthermore, the rice cultivar Tog12387 reported to be as resistant to the West African RYMV strains was susceptible to all Tanzanian RYMV strains suggesting the emergence of virulent and new RYMV pathotypes (Jaw, 2010).

1.7 Genetic variability and genetic analysis of rice for RYMV resistance

Genetic diversity is fundamental in any crop breeding programmes. The use of genetically variable and complementary parents for breeding provides plant species the ability to adapt to the prevailing biotic (pests and diseases) and abiotic (poor soil fertility and drought) stresses (Parmesan and Yohe, 2003). Genetic diversity analysis in rice breeding is essential for identifying complementary and unrelated parents for hybridisation, and subsequent selection. Genetic diversity will limit genetic vulnerability and ensure enhanced genetic variation through recombination. The availability of a broad genetic base is key in rice breeding, enabling to maximise genetic gains through selection. Knowledge of genetic variation among germplasm accessions and genetic relationships between genotypes is important considerations in variety design and development. There is a need to intensively characterise modern and obsolete varieties, breeding lines and landraces for resistance to RYMV. Table 1.1 presents some of the key RYMV resistant rice genetic resources and genes reported globally. These genetic resources can further be characterised to identify the genetic basis of their resistance to guide future gene introgression and gene pyramiding. Landraces may serve as important sources of genes that can be transferred through hybridisation, while wild rice species that are not cross compatible with cultivated rice can be exploited through bridge crossing or transgenics.

Table 1.1. Resistant rice genotypes to RYMV and corresponding resistance genes reported globally

Genotype	Species	RYMV resistance genes	References
Gigante	*Oryza sativa*	RYMV1	Coulibaly et al. (1999)
Bekarosaka	*O. sativa*	RYMV1	Coulibaly et al. (1999)
Tog12387	*O. glaberrima*	RYMV1	Jaw (2010)
Tog5672	*O. glaberrima*	RYMV1 and RYMV2	Thiemélé et al. (2010)
Tog5674	*O. glaberrima*	RYMV1	Thiemélé et al. (2010)
Tog5438	*O. glaberrima*	RYMV1	Thiemélé et al. (2010)
Tog7291	*O. glaberrima*	RYMV2	Ndjiondjop et al. (1999)

RYMV1 and RYMV2 are resistance gene to RYMV 1 and 2, respectively.

1.8 Gene action and heritability for RYMV resistance, yield and yield components

Plant traits are broadly classified as quantitative or qualitative depending on phenotypic expression. This is usually linked to the number of genes conditioning their inheritance.

Quantitative traits are controlled by numerous genes, each contributing to a small effect on the overall phenotype expression. According to Acquaah (2007), these genes function individually contributing to the phenotypic expression. Gene action is further partitioned into additive, dominance and epistatic effects. Additive gene action results in progeny that are intermediate in phenotype between contrasting parents for the alternative genes. Additive gene action will make some parents, in a population, combine favourably with most parents. Additive genes are fixable, and genetic improvement of a desired trait can successfully be achieved through selection (Acquaah, 2007). Conversely, dominance gene action results in a heterozygote whose phenotype may not be midway between two parents but with a tendency be like one of the best parents. The magnitude of association towards one of the parents is related to the degree of dominance, which might be complete dominance, partial dominance, or over-dominance. Unlike additive gene action, dominance gene action is not fully inherited by the progeny generation through continuous selection. However, homozygous dominance genes can be fixed in a self-fertilising crop such as rice.

Knowledge of the nature and magnitude of gene action governing RYMV resistance and other complementary traits is essential in order to design efficient rice breeding programmes. This determines breeding methodologies to develop RYMV resistant cultivars with yield gains. Resistance to RYMV is controlled by both additive and dominance gene action. Various genetic studies have indicated the importance of both additive and non-additive gene action for yield and yield related components in rice (Kumar et al., 2010; Hassan, 2012). Kumar et al. (2010) reported dominance gene action for days to 50% flowering, plant height, number of productive tillers per plant, number of grains per panicle, 1000-grain weight and grain yield per plant. Hassan (2012) reported that additive gene action was significant for the panicle length, number of panicles/plant, number of filled grains/ panicle, 1000-grain weight and grain yield/plant. Pedigree selection methods would be effective for the improvement of traits that are largely controlled by additive gene action.

Understanding the mode of inheritance of characters is an essential component in plant breeding programmes. Success of breeders in changing the characteristics of a population depends on the degree of correspondence between phenotypic and genotypic values. The degree of correspondence is provided by quantitative measures such as heritability, which is estimated for a particular trait and population in a given environment. Two types of heritability estimates are distinguished, broad-sense and narrow sense, depending whether it refers to the genotypic value or breeding value, respectively. Heritability estimates indicate the extent to which a given character would be transmitted to the next generation. The knowledge of heritability of a character helps plant breeders to predict genetic advance that should result

from selection. The higher the heritability, the greater the response to selection. Mirarab and Ahmadikhah (2010) observed high narrow-sense heritability estimates for days to heading, whereas Ghara et al. (2014) reported a low narrow sense heritability estimate (3.35%) for days to 50% flowering. The type of gene action plays a significant role in determining heritability. According to Hefena et al. (2016), traits controlled by additive gene effects tend to have higher heritability values than traits controlled by non-additive gene effects. For instance, Bagati et al. (2016) reported high heritability (98%) for spikelet fertility in rice, indicating that this trait was simply inherited controlled by additive gene effect, therefore selection for traits with high heritability values would be more effective for improvement.

1.9 Genomic approaches towards RYMV resistance improvement

Molecular markers are useful tools for marker-assisted selection (MAS) that complements phenotypic selection, aiming to accelerate the overall breeding progress. A wide range of molecular breeding methods has been described including MAS for selection of major genes and large-scale genomic selection for quantitative traits. MAS appears to be most useful for the introgression of a few genes, and which allows for earlier selection, and reduces the plant population size used during selection programmes. Recently, the successful introgession of the RYMV1 resistance gene from the cultivar Gigante into the background of locally adapted cultivars using microsatellite markers (SSR) was reported (Taylor and Jalloh, 2017). Polymerase chain reaction (PCR) based single nucleotide polymorphism (SNP) markers have been also used to tag RYMV1 resistance alleles (Thiemele et al., 2010). RYMV1 has been mapped on the long arm of chromosome 4 using SSR markers to provide tools for MAS. The sequencing of the CP gene is able to distinguish RYMV strains (Fargette et al., 2002). The transfer of resistance alleles through MAS and their functionality in new genetic backgrounds can be confirmed through phenotypic evaluation under artificial inoculation or in hotspot areas.

1.10 Identifying the needs and preferences of farmers in improved rice varieties

Participatory plant breeding enables adoption of newly developed RYMV resistant cultivars, particularly by smallholder farmers in marginal agro-ecological and socio-economic groups (Ceccarelli et al., 2007). Plant breeders have often focused on developing high yielding and improved crop cultivars in favourable environments and under controlled experiments. Most of the breeding programmes did not consider farmers' preferences and attributes, available landraces and the real conditions of small-scale farmers (Ceccarelli *et al*., 2000). Failure to engage with the realities faced by local farmers has been identified as the primary cause of the consistently low adoption of improved cultivars and their production packages.

Information on farmers' knowledge and perceptions about RYMV disease in Africa is limited and farmers' management of the disease is not well documented. Such knowledge requires proper and recent information for rice improvement purposes. Collaboration of farmers with rice researchers and stakeholders may ensure that the newly developed cultivars are relevant to all value chain actors for local and regional markets (Joshi and Witcombe, 1996). Demand led plant breeding is an effective way to select locally adapted RYMV resistant landraces and to improve farmers' access to useful crop genetic diversity (Ashby and Lilja, 2004). Participatory rural appraisal (PRA) is a rapid and cost-effective technique for identifying farmers-preferred cultivars and market demand. The technique helps to reveal a number of important traits that would not have been considered by breeders in developing new cultivars (Danial et al., 2007). Therefore, crop-breeding technologies developed through participatory research have a greater chance of adoption by farmers because they are developed in response to local constraints, and meet end-users needs and preferences.

1.11 Conclusion

RYMV disease is widespread and severe under lowland rain-fed and irrigated rice faming systems globally. There is a need for developing genotypes that can yield better under existing constraints in order to bridge the existing yield gap. Several control strategies are recommended for RYMV, but the use of resistant cultivars remains to be the most efficient and economical option, particularly for subsistence farmers. Landraces might be excellent sources for resistance breeding against RYMV. These are readily available, adapted to local environments and have been kept by farmers because of their variable-desired traits that evolved over long agricultural history. Rice breeders were unsuccessful in developing stable RYMV resistant cultivars. This was due to the emergence of new and virulent RYMV strains through genetic mutation. Therefore, there is continued need to develop RYMV resistant cultivars using stably expressing genes. This is contingent up on the availability of novel genetic and genomic resources, knowledge of the genetics of the host and the causative agent, the availability of efficient phenotyping and pathotyping methods, and understanding of the genes involved and their pattern of inheritance.

References

Abo, M.E., A. Sy, and M.D. Alegbejo. 1998. Rice yellow mottle virus (RYMV) in Africa. Evolution, distribution and economic significance on sustainable rice production and management strategies. Journal of Sustainable Agriculture 11: 85-111.

Abo, M.E., A.S. Gana, A.T. Maji, M.N. Ukwungwu, and E.D. Imolehin. 2005. The resistance of farmers rice varieties to rice yellow mottle virus (RYMV) at Badeggi. Nigeria, Journal of Tropical Agriculture 21:100-104.

Abo, M.E., M. Alegbejo, A, Sy, and S. Misari. 2000. An overview of the mode of transmission, host plants and methods of detection of rice yellow mottle virus. Journal of Sustainable Agriculture 17: 19-36.

Abo, M.E., M.D. Alegbejo, A. Sy, and Y. Sere. 2004. Retention and transmission of rice yellow mottle virus (RYMV) by beetle vectors in Cote d'Ivoire. African Journal of Agronomy 16: 71-75.

Acquaah, G. 2007. Principles of plant genetics and breeding. Blackwell Publishing Ltd, Malden, UK. 569 p.

Ashby, J.A., and N. Lilja. 2004. Participatory research does it work? Evidence from participatory plant breeding. Crop Science Congress, Brisbane, Australia.

Atera, E., J.C. Onyango, T. Azuma, S. Asanuma, and K. Itoh. 2011. Field evaluation of selected NERICA rice cultivars in Western Kenya. African Journal of Agriculture Research 6: 60-66.

Atera, E.A., F.N. Onyancha, and E.B.O. Majiwa. 2018. Production and marketing of rice in Kenya: Challenges and opportunities. Journal of Development and Agricultural Economics 10: 64-70.

Bagati, S., A.K. Singh, R.K. Salgotra, R. Bhardwaj, M. Sharma, S.K Rai, and Bhat, A. 2016. Genetic variability, heritability and correlation coefficients of yield and its component traits in basmati rice (*Oryza sativa* L.). Journal of Breeding and Genetics 48: 445-452.

Bakker, W. 1971. Rice yellow mottle virus, a mechanically transmissible virus disease of rice in Kenya. Netherlands Journal of Plant Pathology 76: 53-63.

Bakker, W. 1974. Characterization and ecological aspects of rice yellow mottle virus in Kenya. Agriculture research reports. Wageningen: Centre for Agricultural Publishing and Documentation, 152 p.

Balamurugan, P., and Balasubramanian, V. 2017. Challenges and opportunities for increasing rice production in sub Saharan Africa. Journal International of Agriculture 4:1–10.

Banwo, O.O. 2002. Vector Identity, Bionomics and Molecular Characterization of Rice Yellow Mottle Virus in Tanzania [PhD Thesis]. Sokoine University of Agriculture, Morogoro, Tanzania.

Bucheyeki, L.K., E. Shennkawa, D. Kadadi, and J. Lobulu. 2011. Assessment of rice production constraints and farmers preferences in Nzega and Igunga district, Tanzania. Journal of Advance in Development Research 2: 30 - 37.

Ceccarelli, S., and G. Stefania. 2007. Decentralized participatory plant breeding: an example of demand driven research. Euphytica 155: 349-360.

Ceccarelli, S., S. Grando, R. Tutwiler, J. Baha, A.M. Martini. H. Salahieh, A. Goodchild, and M. Michael. 2000. A methodological study on participatory barley breeding: Selection phase. Euphytica 111: 91-104.

Coulibaly, M.M., G. Konate, and J.D. Zongo. 1999. Criblage varietal du riz pour la resistance au RYMV au Sahel. Revue du Cames. Science Medecine 1:7–13.

Danial, D., J. Parlevliet, C. Almekinders, and G. Thiele. 2007. Farmer's participation and breeding for durable disease resistance in the Andean region. Euphytica 153: 385-396.

Del Villar. P.M., and F. Lancon. 2015. West African rice development: beyond protectionism versus liberalization? Global Food Security 5:56–61.

Drame, N.K., and B. Manneh. 2013. Rice Genetic improvement for abiotic stress tolerance in Africa. In: Wopereis, M.C.S., D.E. Johnson, N. Ahmadi, E. Tollens, and A. Jalloh, (Eds) Realizing Africa's Rice Promise. CABI, Boston, pp.144-160.

EUCORD. 2012. Rice Sector Development in East Africa. A Desk Study Prepared for Common Fund for Commodities. European Cooperative for Rural Development, Brussels, 73 p.

Fargette, D., A. Pinel, H. Halimi, C. Brugidou, C. Fauquet, and M Van Regenmortel. 2002. Comparison of molecular and immunological typing of isolates of rice yellow mottle virus. Archives of Virology 147:583–596.

Fargette, D., A. Pinel, Z. Abubakar, O. Traoré, C. Brugidou, S. Fatogoma, E. Hébard, M. Choisy, Y. Séré, C. Fauquet, and G. Konaté. 2004. Inferring the evolutionary history of rice yellow mottle virus from genomic, phylogenetic and phylogeographic studies. Journal of Virology 78: 3252-3261.

Food and Agriculture Organization of the United Nations (FAO). 2017. Rice market monitor. Available at http://faostat.fao.org/default.aspx. (Accessed 16 January 2018).

Food and Agriculture Organization of the United Nations (FAOSTAT). 2012. Database available from: http://faostat.fao.org/default.aspx. (Accessed 17 December 2017).

Food and Agriculture Organization of the United Nations (FAOSTAT). 2015. Database available from: http://faostat.fao.org/default.aspx. (Accessed 13 February 2018).

Ghara, A.G., G. Nematzadey, N. Bagheri, M. Oladi, and A. Bagheri. 2014. Heritability and heterosis of agronomic traits in rice lines. International Journal of Farming and Allied Sciences 3:66–70.

Hashmi, I., and A.D. Khan. 2011. Adverse health effects of pesticides exposure in agriculture and industrial workers of developing countries. In: Stoycheva M, editor.

Pesticides -the Impacts of Pesticide Exposure. 1st ed. Slavka Krautzeka, pp.156-178.

Hassan, H.M. 2012. Genetic studies on grain dimension yield and its related traits in rice under saline soil conditions (*Oryza sativa* L.). Journal of Plant Production 3:3149–3164.

Hefena, A.G., M.S. Sultan, M.A. Abdel-Moneam, S.A Hammoud, C. Barutçular, and A. EL-Sabagh. 2016. Assessment of genetic variability and correlation coefficient to improve some agronomic traits in rice. Journal of Experimental Agriculture Internationa14:1–8.

Hubert, J., A. Luzi-Kihupi, E. Hébrard, H. John, and J. Lyimo. 2016. Farmers' knowledge and perceptions of rice yellow mottle virus in Selected rice growing areas in Tanzania. International Journal of Science and Research 5: 549-559.

Hull, R., and D. Fargette. 2005. Sobemovirus. In: Fauquet, C.M., M.A. Mayo, J. Maniloff, U. Desselberger, and L.A. Ball, (eds). Virus taxonomy. Classification and nomenclature of viruses. Eighth Rport of the International Committee on Taxonomy of Viruses. Elsevier/Academic Press, London, pp. 885-890.

Jaw, A. 2010. Screening and Molecular Characterisation Of near-Isogenic Lines for Resistance to Rice Yellow Mottle Virus. MSc Thesis, Kwame Nkrumah University of Science and Technology, Kumasi, Ghana, 61 p.

Joshi, A., and J.R. Witcombe. 1996. "Farmer participatory crop improvement II," Participatory varietal selection, a case study in India. Journal of Expansion Agriculture 32: 461-477.

Kam, H., M.D. Laing, J. Ouoba, and M.N. Ndjiondjop. 2013. Rice traits preferred by farmers and their perceptions of rice yellow mottle virus (RYMV) disease in Cascades Region of Burkina Faso. African Journal of Agricultural Research 8: 2703-2712.

Kanyeka, Z.L., E. Sangu, D. Fargette, A. Pinel-Galziand, and E. Hérbrard. 2007. Distribution and diversity of local strains of rice yellow mottle virus in Tanzania. African Crop Science Journal 15: 201-209.

Konaté, G., O. Traoré, and M.M. Coulibaly. 1997. Characterization of rice yellow mottle virus isolates in Sudano-Sahelian areas. Archives of Virology 142: 117-124.

Kouassi, N.K., P. N'Guessan, L. Albar, C.M. Fauquet, and C Brugidou. 2005. Distribution and characterization of rice yellow mottle virus: A threat to African farmers. Plant Disease 89:124-133.

Kumar, S.P., K. Saravanan, and T. Sabesan. 2010. Combining ability for yield and yield contributing characters in rice (*Oryza sativa* L.). Electronic Journal of Plant Breeding 1:1290-1293.

Ling, R., A.E Pate, J.P Carr, and A.E Firth. 2013. An essential fifth coding ORF in the sobemoviruses. Journal of Virology 446:397-408.

Longué, R.D.S., I. Zinga, S. Semballa, N. Barro, and O. Traoré. 2016. Detection and serological characterization of rice yellow mottle virus in Central African Republic. Agricultural Sciences 7: 911-919.

Lussewa, R.K., R. Edema, P. Gibson, and J. Lamo. 2016. Inheritance of bacterial leaf blight resistance in crosses involving interspecific and intraspecific rice genotypes. Indian Journal of Science 23: 698-717.

Luzi-Kihupi, A. 2001. Rice yellow mottle virus in Tanzania. In: Rice Research workshop for rice researchers in Kenya, Malawi, Tanzania and Zambia, Kilimanjaro Agricultural and Training Centre, Moshi, Tanzania, March 12 -16, 2001, pp. 41-174.

Maclean, J.L., B. Hardy, and G. Hettel. 2013. Rice Almanac: Source Book for the Most Important Economic Activities on Earth, 4th Edition. IRRI, Los Baños, Philippines.

MAFAP. 2013. Review of Food and Agricultural Policies in the United Republic of Tanzania. Monitoring African Food and Agricultural Policies, Food and Agriculture Organization, Rome, 222 p.

Match Maker Associates. 2010. Value Chain Analysis of Rice and Maize in Selected Districts in Tanzania, Rice and Maize Context Report: Agricultural Council Publishing House, Dar es Salaam. Tanzania.

Mirarab, M. and A. Ahmadikhah. 2010. Study on genetics of some important phenological traits in rice using line x tester analysis. Annals of Biological Research 1:119-125.

Munganyinka, E., R. Edema, J. Lamo, and P. Gibson. 2016. The reaction of intraspecific and interspecific rice cultivars for resistance to rice yellow mottle virus disease. European Journal of Experimental Biology 6:13-18.

Muthayya, S., J.D. Sugimoto, S. Montgomery, and G.F. Maberly. 2014. An overview of global rice production, supply, trade, and consumption. Annals of the New York, Academy of Sciences 1324: 7-14.

Ndjiondjop, M.N., L. Albar, D. Fargette, C. Fauquet, and A. Ghesquiere. 1999. The genetic basis of high resistance to rice yellow mottle virus (RYMV) in cultivars of two cultivated rice species. Plant Disease 83:931–935.

Nwilene, F.E., A.K. Traore, A.N. Asid, Y. Séré, A. Osanya, and M.E Abo. 2009. New records of insect vectors of rice yellow mottle virus (RYMV) in Cote d'Ivoire, West African Journal of Entomology 6: 198-206.

Ogunbayo, S.A., M. Sie, D.K. Ojo, K.A. Sanni, M.G. Akinwale, B. Toulou, A. Shittu, E.O. Idehen, A.R. Popoola, I.O. Daniel, and G.B. Gregorio. 2014. Genetic variation and heritability of yield and related traits in promising rice genotypes (*Oryza sativa* L.) Journal of Plant Breeding and Crop Science 6 (11): 153-159.

Onasanya, A., M.M. Ekperigin, F.E. Nwilene, Y. Sere, and R.O. Onasanya. 2009. Two pathotypes of *Xanthomonas oryzae* pv. oryzae virulence identified in West Africa. Current Research in Bacteriology 2: 22-35.

Opalka, N., C. Brugidou, C. Bonneau, M. Nicole, and Beachy R. 1997. Movement of rice yellow mottle virus between xylem cells through pit membranes. Protocol National Academic Science 95:3323–3328.

Parmesan, C., and G. Yohe. 2003. A globally coherent fingerprint of climate impacts across natural systems. Nature 421: 37-42.

Pinel, A., P. N'Guessan, Bousalem, M. and D. Fargette. 2000. Molecular variability of geographically distinct isolates of rice yellow mottle virus in Africa. Archives of Virology 145: 1621-1638.

Pinel-Galzi, A., C. Dubreuil-Tranchant, E. Hébrard, C. Mariac, A. Ghesquière, and Albar, L. 2016. Mutations in rice yellow mottle virus polyprotein P2a involved in RYMV2 gene resistance breakdown. Frontier of Plant Science 7:1–11.

Pinel-Galzi, A., M. Rakotomalala, E. Sangu, F. Sorho, Z. Kanyeka, O. Traoré, D. Sérémé, N. Poulicard, Y. Rabenantoandro, and Y. Séré. 2007. Theme and variations in the evolutionary pathways to virulence of an RNA plant virus species. PLoS pathogens 3:1761-1770.

Salaudeen, M.T. 2014. Relative resistance to rice yellow mottle virus in rice. Plant Protection Science 50:1-7.

Sereme, D., I. Ouedraogo, B.J. Neya, P.E. Zida, N. Yao, and M. Sie. 2016. Screening improved rice varieties (*Oryza spp*) for their resistance /tolerance to rice yellow mottle virus in West Africa. International Journal of Agriculture Innovations and Research 5:481-486.

Shelepchikov, A.A., V.V. Shenderyuk, E.S. Brodsky, D. Feshin, L.P. Baholdina, and S.K. Gorogankin. 2008. Contamination of Russian Baltic fish by polychlorinated dibenzo-p-dioxins, dibenzofurans and dioxin-like biphenyls. Environmental Toxicology and Pharmacology 25: 136-143.

Sié, M., K. Sanni, K. Futakuchi, B. Manneh, S. Mandé, R. Vodouhé, S. Dogbe, K. Dramé, A. Ogunbayo, M.N. Ndjiondjop, and K. Traoré. 2012. Towards a Rational Use of African Rice (*Oryza glaberrima* Steud.) for Breeding in Sub-Saharan Africa. Genes, Genomes and Genomics, 6: 1-7.

Srujana, G., B.G. Suresh, G.R. Lavanya, J.B. Ram, and V. Sumanth. 2017. Studies on genetic variability, heritability and genetic advance for yield and quality components in rice (*Oryza sativa* L.). *Journal of Pharmacognosy and Phytochemistry* 6: 564-566.

Tamm, T., and E. Truve. 2000. Sobemovirus (mini-review). Journal of Virology 74 (14): 6231 - 6231.

Taylor, D.R., and Jalloh, A.B. 2017. Evaluation of a set of near isogenic lines (NILS) for rice yellow mottle virus (RYMV) resistance and farmers' participatory varietal evaluation in Sierra Leone. African Journal of Agricultural Research 12:1149–1157.

Thiemele, D., A. Boisnard, M.N. Ndjiondjop, S. Cheron, Y. Sere, S. Ake, A. Ghesquiere, and L. Albar. 2010. Identification of a second major resistance gene to rice yellow mottle virus, RYMV2, in the African cultivated rice species, O*. glaberrima*. Theoretical Applied Genetics 121:169–179.

Traore, O., A. Pinel-Galzi, F. Sorho, S. Sarra, and M. Rakotomalala. 2009. A reassessment of the epidemiology of rice yellow mottle virus following recent advances in field and molecular studies. Virus Research 141: 258 - 267.

Traore, V.S.E., M.D Asante, V.E. Gracen, S.K. Offei, and O. Traore. 2015. Screening of rice accessions for resistance to rice yellow mottle virus. American Journal of Experimental Agriculture 9:1–12.

Woin, N., D. Djonmaila, S. Ismael, S. Bourou, and T. Bebom. 2007. Potential for biological control of rice yellow mottle virus vectors. African Crop Science Journal 15:211–222

Zouzou, M., T.H. Kouakou, M. Koné, and S. Issaka. 2008. Screening rice (*Oryza sativa* L.) varieties for resistance to rice yellow mottle virus. Scientific Research and Essay, 9:416-424.

CHAPTER TWO: FARMERS' PERCEPTIONS, PRODUCTION CONSTRAINTS AND VARIETY PREFERENCES OF RICE IN TANZANIA

Abstract

Rice (*Oryza sativa* L.) is a major food and cash crop cultivated under diverse farming systems in Tanzania. The objective of this study was to assess farmers' perceptions, production constraints, variety preferences and breeding priorities of rice in selected agro-ecologies in Tanzania to guide variety development and release. A participatory rural appraisal study was conducted involving 180 participants using a structured survey. Focus group discussions were held with 90 discussants in the Mvomero, Kilombero and Kyela districts of Tanzania. The survey results indicated that rice was the most important food and cash crop followed by maize (*Zea mays* L.), cassava (*Mannihot esculenta* Crantz), sweetpotato (*Ipomoea batatas* [L.] Lam.), sugarcane (*Saccharum officinarum* L.), pigeonpea (*Cajanus cajan* L.), cowpea (*Vigna unguiculata* [L.] Walp), simsim (*Sesamum indicum* L.), beans (*Phaseolus vulgaris* L.), cocoa (*Theobroma cacao* L.), banana (*Musa acuminata* L.), groundnuts (*Arachis hypogaea* L.), and palm (*Elaeis guineensis* Jacq.). Most of the respondents used saved seed from a previous harvest. Major constraints limiting rice production and productivity in all studied areas were disease, insect pests, recurrent drought, the non-availability and high cost of fertilizers, a lack of improved cultivars, poor soil fertility and bird damage. The farmers preferred rice varieties with high yield, disease resistance, drought tolerance, high market value, early maturity, aroma, and local adaptation. A systematic rice breeding program aimed at improving rice yellow mottle virus resistance and incorporating farmers' preferred traits should be designed and implemented to increase productivity and adoption of new cultivars by the farmers across the ricegrowing areas of Tanzania.

Key words: Farmers' preferred traits; participatory rural appraisal; production constraints; rice; Tanzania

---

This chapter was published in the Journal of Crop Improvement. Suvi, W.T., H. Shimelis, M. Laing. 2020. Farmers' perceptions, production constraints and variety preferences of rice in Tanzania. Journal of Crop Improvement 1-18. doi: 10.1080/15427528.2020.1795771.

2.1 Introduction

Rice (*Oryza sativa* L.) is an important global commodity crop. It is the third most preferred cereal in the world after maize (*Zea mays* L.) and wheat (*Triticum aestivum* L.) (Bagati et al., 2016). It supports the livelihoods of more than two thirds of the global human population (Krupa et al., 2017). Human consumption of rice accounts for 85% of total production, compared with 72% for wheat and 19% for maize (FAO, 2012). Rice provides approximately 23% of daily caloric intake for the human population (Chemutai et al., 2016). Globally, rice is cultivated on approximately 159 million hectares of land, with an annual total production of 744.4 million tons, with Asia producing more than 90% of global production (Srujana et al., 2017). Although rice production is still low in sub-Saharan Africa (SSA), it is steadily increasing and becoming an important component in the national economies and food security of several countries in this region. The production of rice in SSA is categorized into rain-fed lowland, rain-fed upland, irrigated lowland, deep water or flooding and mangrove swamps.

Tanzania is the second largest rice producer after Madagascar in the east, central and southern African region, with an annual production of 1.1 million tons of milled rice (Matchmaker, 2010). The rice industry is a major source of employment, income and food security. In Tanzania, rice is the second most important food crop after maize (Bucheyeki et al., 2011). The crop is mostly grown in the regions of Morogoro, Shinyanga, Mbeya, Mwanza, Tabora, Kilimanjaro and Rukwa. Nearly 25% of the national rice production comes from the Kyela and Mbarali districts, situated in Mbeya region and Kilombero, Kilosa and Mvomero districts in the Morogoro region.

Despite the increasing importance of rice in Tanzania, the mean yield of the crop is 1.5 tons ha^{-1}, which is far below the yield averages reported in SSA (4.4 tons ha^{-1}), Asia (4.6 tons ha^{-1}) and South America (5.2 tons ha^{-1}) (Atera et al., 2018). The low rice productivity in Tanzania is attributed to biotic and abiotic stresses and diverse socio-economic constraints.

Among the biotic constraints to rice production, the rice yellow mottle virus (RYMV) is the most important (Hubert et al., 2016). RYMV disease is endemic to Africa and is considered the most damaging pathogen of rice (Pinel-Galzi et al., 2016). The disease is prevalent in almost all SSA countries, causing major yield losses in susceptible rice varieties cultivated in the lowland and irrigated agro-ecologies. The RYMV disease causes yield losses ranging from 20 to 100%, depending on the rice genotype, time of infection and viral strain, and the interaction of these factors (Longué et al., 2016). Control strategies, such as the use of resistant cultivars, cultural practices and crop protection chemicals, have been recommended for RYMV disease. Developing RYMV-resistant rice cultivars is considered to be an

economic, environmentally friendly and effective control strategy against the disease, especially for smallholder farming systems (Sereme et al., 2016).

The development of new high-yielding and resistant varieties cannot have an appreciable impact unless the selection takes into account end-user qualities (Mogga et al., 2018). End-user qualities, such as cooking and eating quality, grain shape and aroma, are considered major drivers for widespread adoption of a new variety. Improved varieties that do not meet or surpass physical, cooking and eating qualities of landraces would not be competitive on the market. Thus, researchers have become increasingly aware that incorporating end-user preferred qualities in technology development may substantially enhance chances of adoption of the technology. Developing cultivars with improved grain yield and quality will also boost production and aid in penetrating lucrative international markets, which would increase income generation for the farmers.

Currently, there is only limited information on constraints affecting rice production, trait preferences and disease-management strategies for sustainable rice production among smallholder farmers in Tanzania. A strategy for improving rice productivity through breeding requires current information regarding farmers' perceived constraints, and their needs and preferences. In turn, this strategy requires documenting current circumstances and constraints of the farmers through farmers' participatory methods across rice-farming systems. Rice-improvement programs should focus on the needs of smallholder farmers, value chains, and other stakeholders to ensure the successful release and adoption of newly developed cultivars and production technologies.

The participatory rural appraisal (PRA) approach has been widely used to identify farmers' production constraints, preferred crop varieties and traits for deployment of production packages and suitable varieties (Mrema et al., 2017; Mogga et al., 2018). This approach considers the value of farmers' knowledge, their trait preferences, experiences and production constraints, abilities and innovation. Collaboration of farmers with the formal research sector may offer researchers a mechanism to ensure that their work is relevant to farmers' needs and conditions. According to Shelton and Tracy (2016), involvement of farmers in the research process has increased the chances of success in the generation of appropriate agricultural technology and adoption of varieties. This information will be valuable for participatory plant breeding that has been shown to be an effective way to develop demand-led, locally adapted rice genotypes and to improve farmers' access to useful crop genetic diversity (Shelton and Tracy, 2016). Therefore, the objective of this study was to assess farmers' perception, production constraints, preferences and breeding priorities

regarding rice in selected agro-ecologies in Tanzania to guide variety development and release.

2.2 Materials and methods

2.2.1 Description of study areas

The study was carried out in the Kilombero and Mvomero districts in the Morogoro region and in the Kyela district in the Mbeya region in Tanzania (Figure 2.1). Morogoro and Mbeya regions are located in the eastern and southern highland agro-ecological zones, respectively. Morogoro region experiences maximum temperatures varying from 26^0C to 32^0C and has a bimodal rainfall pattern with short rains that begin towards the end of November and end in early February, and long rains that usually start in March and end in May. This region receives a mean total annual rainfall of 935 mm. The Mbeya region has maximum temperatures ranging from 16^0C in July to 32^0C in October. Mbeya receives a total mean annual rainfall of 944 mm, and has a long dry season of about four months. The Kilombero District is characterized by alluvial lowlands covered mostly by heavy clay soils. The Mvomero District is characterized by sandy clay loam textured soils. The dominant soil texture in the Kyela district is clay loam.

Figure 2.1. Map of Tanzania showing the study areas highlighted in yellow.

2.2.2 Sampling procedures

Purposive sampling was employed to identify regions, districts, villages, and farmers for the survey. The districts were purposely selected on the basis of their high potential for rice production. Further, more than 80% of the district residents' income comes from paddy rice production and trade. The following nine villages were chosen: Mkindo (S06º15.344', E037º32.387'), Kigugu (S06º20.674', E037º59.176') and Lukenge (S06º24.263', E037º67.511') (from the Mvomero district); Lusungo (S09°30.000, E033°05.860'), Tenende (S09°33.050', E033°05.326´) and Kilasilo (S09º05.858', E033º82.841') (from the Kyela District); Mkula (S07º84.895', E036º91.903'), Ichonde (S07º90.812', E036º81.897') and Mgudeni (S07º88.679', E036º08.318') (from the Kilombero District); on the basis of prior information on the importance of rice in these areas, and their accessibility. Planning meetings were conducted in each district and village, and the breeder explained the objectives of the study and selection criteria to farmers. Following consultative discussions with the extension officers, the survey routes were mapped, farms selected and the questionnaire pretested. The target survey group was smallholder rice farmers. In each village, 20 farmers were sampled for household interviews. This provided 180 farmers to be

interviewed using a semi-structured questionnaire. Village leaders, with the help of agricultural extension officers, identified farmers for household interviews. The team that carried out the survey consisted of a breeder, two socio-economists, and one agricultural extension officer in each district.

Focus group discussions (FGDs) were conducted in each village to understand farmers' varietal preferences and the specific traits that influence a farmer's decision in selecting a rice variety for production, and the major constraints affecting rice production. It involved nine focus groups comprising farmers, local leaders, and key informants with broad knowledge on diverse social issues and rice cultivation in the village. Each focus group was composed of 10 representative farmers, who were sampled on the basis of gender balance and their experience in rice farming. A total of 90 farmers participated in the FGDs across the three districts. Checklists were developed and used to guide focused group discussions with farmer groups and individual key informants. The farmers were encouraged to use their local Swahili language. In both individual interviews and FGDs, both male and female farmers were selected.

2.2.3 Data collection

A number of participatory methods were used for data collection. Both informal and semi-formal methods were employed to obtain information. Primary data were collected using semi-structured questionnaire, FGDs and field visits. A semi-structured questionnaire was used to collect household information regarding rice variety grown, preferred variety attributes, production constraints and cropping systems used, and the traits of their preferred varieties that they used in selection. Other data included seed sources, preferred rice traits, and farmers' awareness of rice diseases, such as RYMV, and their control methods.

During data collection, farmers expressed their opinions through group discussions regarding food and cash crops grown, the commonly grown varieties, constraints to rice production, preferred rice variety attributes, and their needs and preferences. Participants were given a flip chart to list names and types of rice varieties grown, preferred traits and problems facing rice production. Group observations on selected rice fields were made during transect walks in the selected districts to provide complementary data.

2.2.4 Data analysis

Quantitative and qualitative data collected through the questionnaire were coded and subjected to statistical analyses using the Statistical Package for Social Sciences software version 24 (SPSS, 2017). Cross-tabulations were constructed and descriptive statistics were calculated to summarize data collected during the survey and the FGDs. To make statistical

inferences, contingency chi-square tests were computed to analyze relationships between variables. This allowed empirical analyses and description of associations between the collected parameters across the three study districts.

2.3 Results

2.3.1 Demographic characterization of households

Detailed descriptions regarding the socioeconomic characteristics of the households in the study area are presented in Table 2.1. The number of male farmers (62.8%) was significantly (χ^2 = 13.885; p = 0.001) higher than female farmers (37.2%) in all the study districts. The majority of respondents (63.3%) were aged between 26 and 50 years, whereas 26.7% of respondents were above 50 years of age and 10% of respondents were below 25 years of age (Table 2.1). The proportion of respondents, who were married, was 75.6%, whereas 12.2, 7.2 and 5% were divorced, single and widowed, respectively (Table 2.1).

The differences in level of education attained by the farmers across the districts were not significant (χ^2 = 9.66; p = 0.140). Most respondents (81.6%) had attended primary school and were able to read and write in the local Kiswahili language only, whereas 10.6% and 2.2% had attended college and secondary education, respectively, and were able to read and write in both English and Kiswahili. The remainder, i.e., 5.6% of respondents had not attended school (Table 2.1). The size of land owned by the farmers was consistent across the different districts (χ^2 = 12.444; p = 0.053). The land size allocated for rice cultivation is summarized in Table 2.1. Across districts, 41.7% of rice farmers had production fields ranging from 1.6 to 3.4 ha. About 27.8% of the respondents owned between 0.5 and 1.5 ha, 20.6% of respondents owned rice fields of between 3.5 and 5 ha, and 10% had farm sizes greater than 5 ha.

Table 2.1. Description of household characteristics in surveyed areas in Tanzania

Variable	Class	Mvomero	Kilombero	Kyela	Mean	Chi-square	DF	*p*-value
Gender	Male	51.7	55.0	81.7	62.8	13.885	2	0.001
	Female	48.3	45.0	18.3	37.2			
Age (years)	<25	13.3	10.0	6.7	10.0			
	25-50	56.7	63.3	70.0	63.3	2.675	4	0.614
	>50	30.0	26.7	23.3	26.7			
Marital status	Married	73.3	75.0	78.3	75.6			
	Single	13.3	5.0	3.3	7.2	7.630	6	0.267
	Divorced	11.7	11.7	13.3	12.2			
	Widowed	1.7	8.3	5.0	5.0			
Education level	Illiterate	5.0	6.7	5.0	5.6			
	Primary	85.0	85.0	75.0	81.6			
	Secondary	5.0	0.0	1.7	2.2	9.663	6	0.140
	College	5.0	8.3	18.3	10.6			
Farm size (ha)	0.5-1.5	36.7	33.3	13.3	27.8			
	1.6-3.4	35.0	45.0	45.0	41.7	12.444	6	0.053
	3.5-5.0	18.3	13.3	30.0	20.6			
	> 5.0	10.0	8.3	11.7	10.0			

DF = degrees of freedom; ha = hectare

2.3.2 Crop production

All participants were actively involved in crop production as their major source of food, feed and cash income. The results from surveyed districts (Table 2.2) showed that most of the farmers cultivated different types of crops. Overall, rice and maize were the major crops across the districts. Rice and maize were cultivated by 53.6 and 16.6% of the respondents, respectively. The other crops, such as cassava and sugarcane, were regarded as minor, with less than 5% of the respondents affirming their cultivation. Across the districts, rice was predominantly cultivated as a sole crop for food and income generation, unlike other crops, such as maize and common bean, which were cultivated as intercrops. In Kilombero, sugarcane was considered to be an essential crop by 11% of the farmers. Cocoa, oil palm and groundnut were regarded as important in Kyela by 4% of the respondents. These crops were primarily grown for household consumption and, occasionally, for income generation.

Table 2.2. List of crops grown and proportion of participants (%) cultvating these in three surveyed districts in Tanzania

Crop	Mvomero	Kilombero	Kyela	Mean
Rice	49.9	53.0	57.8	53.6
Maize	15.7	20.2	14.0	16.6
Cassava	3.7	5.1	4.7	4.5
Sweetpotato	2.0	5.0	5.7	4.2
Sugarcane	0.0	11.0	0.0	3.7
Pigeonpea	8.7	0.0	0.0	2.9
Cowpea	8.3	0.0	0.0	2.8
Simsim	8.3	0.0	0.0	2.8
Horticultural crops	1.7	3.7	1.7	2.4
Common beans	1.7	0.0	2.7	1.5
Banana	0.0	2.0	2.0	1.3
Cocoa	0.0	0.0	4.0	1.3
Oil palm	0.0	0.0	3.7	1.2
Groundnut	0.0	0.0	3.7	1.2
Total (%)	**100**	**100**	**100**	**100**

2.3.3 Sources of rice planting material

The different sources of rice planting materials are summarized in Table 2.3. The majority of the farmers (67.2%) used farm-saved seed from previous crops, followed by seed purchased from agro-dealers (15%), and sourced from neighbours (8.9%). In addition, research centres and local government organizations were considered to be minor sources of improved seed, representing only 6.7% and 1.7%, respectively. Very few farmers (0.6%) sourced planting materials from local non-government organizations (NGO), such as Nafaka.

Table 2.3. Sources of rice seed (%) reported by farmers across the three surveyed districts in Tanzania

Sources of seed	Mvomero	Kilombero	Kyela	Mean
Farm saved	58.3	61.7	81.7	67.2
Neighbours	16.7	3.3	6.7	8.9
Non-government organizations (NGOs)	1.7	0.0	0.0	0.6
Research centre	8.3	3.3	8.3	6.7
Local government	3.3	1.7	0.0	1.7
Agro- dealers	11.6	30.0	3.4	15.0
Total (%)	**100**	**100**	**100**	**100**

2.3.4 The types of rice cultivars grown and aromatic traits

Highly significant differences regarding choice of rice varieties grown by the farmers (χ^2 = 53.32; p = 0.000) were detected among the respondents (Table 2.4). About 51.4% of the respondents cultivated landraces, whereas 25.7% of the respondents cultivated both landraces and improved cultivars. The rest of the respondents (22.9%) were growing improved varieties.

There were non-significant statistical differences for scented or unscented aroma in rice (χ^2 = 3.103; p = 0.201) among farmers across the districts. The non-significant difference was attributable to the higher proportion (97%) of farmers needing a scented rice variety compared to only 3% of the farmers that preferred non-aromatic type. All the interviewed farmers (100%) in the Kilombero district preferred aromatic rice cultivars (Table 2.4).

Table 2.4. Types of rice cultivars grown and preferred aromatic attributes by farmers in the three districts

Input	Type/use	Mvomero	Kilombero	Kyela	Mean	Chi- square	DF	p-value
Variety	Landrace	31.7	35.6	86.7	51.4			
	Improved	25.0	40.7	3.3	22.9	53.32	4	0.000
	Both	43.3	23.7	10.0	25.7			
Aroma	Scented	95.0	100.0	95.0	96.7	3.103	2	0.201
	Unscented	5.0	0.0	5.0	3.3			

DF = degrees of freedom

2.3.5 Major constraints to rice production

The chief constraints of rice production included both biotic and abiotic stresses (Table 2.5). The most prominent constraint to rice production, according to 89.4% of the respondent farmers, was the rice diseases including RYMV. The ranking of diseases did not show significant differences (χ^2 = 8.594; p = 0.198) across the districts, indicating that it was equally important across the districts.

Insect pests were also identified as a major problem in rice production and their ranking in importance showed significant variation (χ^2 = 23.92; p = 0.001) among the districts. The least

insect pest problem was recorded in Kyela district (Table 2.5). A higher proportion of the respondents (73.4%) considered insect pests as a high-priority constraint in rice production across all the districts. However, 11.7% and 15% of the respondents considered insects as moderate and low constraints to rice production, respectively. Drought was considered an important constraint in rice production by 69.4% of the respondents, followed by the high cost of fertilizers (64.4%), limited access to improved varieties (59.4%), poor soil fertility (46.7%), bird damage (46.1%) and the limited of access to fertilizers (42.2%), across the districts.

Table 2.5. Main constraints to rice production in the surveyed districts

Constraints	Class	Mvomero	Kilombero	Kyela	Mean	Chi- square	DF	*p*-value
Bird damage	High	70.0	26.7	41.7	46.1			
	Moderate	23.3	26.7	26.7	25.6	32.94	4	0.000
	Low	6.7	46.7	31.7	28.3			
Poor soil fertility	High	51.7	55.0	33.3	46.7			
	Moderate	26.7	25.0	25.0	25.6	11.19	4	0.083
	Low	21.7	20.0	41.7	27.8			
Limited access to improved variety	High	68.4	50.0	60.0	59.4			
	Moderate	10.0	13.3	28.3	17.2	23.51	4	0.001
	Low	21.7	36.7	11.7	23.3			
Limited use of fertilizers	High	50.0	35.0	41.7	42.2			
	Moderate	13.3	26.7	16.7	18.9	12.86	4	0.045
	Low	36.7	38.3	41.7	38.9			
High cost of fertilizers	High	83.3	56.7	53.3	64.4			
	Moderate	8.3	26.7	15.0	16.7	35.35	4	0.000
	Low	8.3	16.7	31.7	18.9			
Drought	High	76.7	73.3	58.3	69.4			
	Moderate	15.0	15.0	18.3	16.1	9.61	4	0.142
	Low	8.3	11.7	23.3	14.4			
Insect pests	High	80.0	88.3	51.6	73.4			
	Moderate	6.7	6.7	21.7	11.7	23.92	4	0.001
	Low	13.3	5.0	26.7	15.0			
Diseases	High	88.4	91.7	88.4	89.4			
	Moderate	3.3	1.7	3.3	2.8	8.59	4	0.198
	Low	8.3	6.7	8.3	7.8			

DF = degrees of freedom

2.3.6 Field observation

The interviewers, extension officers, village officials and farmers conducted field visits to assess the main cropping systems, and the incidence and severity of RYMV disease reported above. The visited fields were severely affected by RYMV (Figure 2.2). Moreover, the co-occurrence of RYMV and prolonged drought were observed to have seriously retarded plant growth.

Figure 2.2. Photo showing infection of RYMV in one of the farmer field's in the Kyela district in Tanzania

2.3.7 Farmers' preferred trait improvements for rice varieties

The farmer-preferred traits included high grain yield, drought tolerance, disease resistance, marketability and early maturity (Table 2.6). High grain yield was ranked as the most preferred trait and the ranking was not significantly different ($\chi^2 = 8.299$; $p = 0.081$) among the districts. About 93% of the respondents preferred rice varieties with high grain yields. On the other hand, 10% farmers, across the study districts, preferred rice varieties with moderate grain yield. The second most preferred trait in rice was disease resistance, with 48.3% to 55% of the respondents across all districts being affirmative. The preference for disease resistance was not significantly different across the districts ($\chi^2 = 3.568$; $p = 0.468$). Mean ranks in all districts showed that drought tolerance (52.1%), marketability (51.1%) and early maturity (47.2%) were the third, fourth, and fifth most preferred traits by farmers, respectively. The severity of these constraints varied from district to district and within districts (Table 2.6).

Table 2.6. Farmers' preferred trait improvements in rice variety in three districts in Tanzania

Trait	Class	Mvomero	Kilombero	Kyela	Mean	Chi - square	DF	*p*-value
	High	90.0	90.0	98.3	92.8			
Grain yield	Moderate	10.0	10.0	0.0	6.7	8.299	4	0.081
	Low	0.0	0.0	1.4	0.5			
Drought tolerance	High	54.3	56.0	46.0	52.1			
	Moderate	25.3	27.0	23.7	25.3	3.568	4	0.468
	Low	20.3	19.0	28.3	22.6			
	High	55.0	56.7	48.3	53.3			
Disease resistance	Moderate	33.4	28.3	23.5	28.4	11.821	4	0.019
	Low	21.7	15.0	18.3	18.3			
	High	56.7	40.0	56.7	51.1			
Marketability	Moderate	35.0	53.3	33.3	40.6	6.218	4	0.183
	Low	8.3	6.7	10.0	8.3			
	High	50.0	48.3	43.3	47.2			
Earliness	Moderate	38.3	41.7	36.7	38.9	2.986	4	0.560
	Low	11.7	10.0	20.0	13.9			

DF = degrees of freedom

2.3.8 RYMV disease and yield loss

The severity of RYMV disease in rice was reported to have been increasing across time in all the surveyed areas (Table 2.7). RYMV infection and yield loss showed significant differences (χ^2 = 47.475; p = 0.000) across the districts, with Kyela suffering the most severe RYMV infection. Across the districts, 92.3% of the respondents reported that RYMV infection was severe, whereas 7% and 0.7% reported mild and no infection, respectively (Table 2.7).

Table 2.7. Farmers' assessment of the levels of rice yellow mottle virus infection

Infection level	Mvomero	Kilombero	Kyela	Mean	Chi-square	DF	p-value
None	0.4	1.7	0.0	0.7			
Mild	11.0	8.3	1.7	7.0	47.475	4	0.000
severe	88.6	90.0	98.3	92.3			

DF = Degrees of freedom

2.4 Discussion

The present study assessed farmers' perceptions of production constraints, variety preferences and breeding priorities for rice in selected agro-ecologies in Tanzania to guide

future cultivar development and release. The proportion of males engaged in rice production was expectedly higher than that of females (Table 2.1) because men were usually the household heads, who took the lead in farm planning and decision-making regarding crops to grow. Males were custodians of common household wealth in the districts in which the study was conducted. This result is in agreement with Mogga et al. (2018), who conducted a study of similar nature in South Sudan and reported that males were heads of the families and took the lead in farm planning and decision-making in most African agricultural systems. The present findings, however, were in contradiction to Kam (2011), who found that there were more females than males involved in rice production in Burkina Faso and the females were more influential in variety adoption. Most respondents (Table 2.1) were between 25 and 50 years of age, which is considered an active group for rice production. Aldosari et al. (2017) also found that rice production was practiced by farmers of a similar age group. Most of the respondents had a level of literacy and numeracy skills, which would facilitate adoption of agricultural technologies required for increased rice productivity. This is in agreement with Farid et al. (2015), who reported that farmers in developing countries, including Tanzania, required at least five years of schooling to follow good farming practices and to enhance their choice and adoption of production technologies. The results agreed with Mrema et al. (2017), who found that educated farmers could easily access information from various sources. The segmentation of rice production by demographic groups is important, especially for raising awareness about new technologies or cultivars to achieve maximum possible impact. Thus, in this study, it would be more effective to target information or newly developed cultivar dissemination toward males, who are the decision makers regarding farming. Some of the innovations can be introduced through posters, leaflets, and brochures that require individuals with reading and numeracy capabilities.

Size of the land holding plays an important role in crop productivity, dissemination and adoption of agricultural practices among farming communities (Aldosari et al., 2017). With respect to land size, most of respondents owned a small-size farm because of limited capital. The farm sizes reported in this study were similar to those reported by Tsinigo (2014), who found that the average farm size of farmers in Ghana was 2.21 ha. Furthermore, Aldosari et al. (2017) described that small land holdings had lower productivity potential and were less efficient in adopting modern technologies. The small farm sizes found in this study could be an impediment to efforts to improve rice productivity in Tanzania even when improved cultivars with high yield potential are availed. With smaller sized farms, the respondents would likely continue to employ obsolete farming methods, which are inefficient and counterproductive, or fail to secure credit for investing on their farm, as financial institutions were likely to regard smallholder farming as too risky to invest in.

The majority of farmers allocated most of their agricultural land to rice, but they also planted other crops as a strategy to diversify their livelihoods and improve resilience to biotic and biotic stresses and socio-economic challenges. The wide range of crops, which included maize, cassava, sweet potato, sugarcane, pigeonpea, cowpea, simsim, horticultural crops, beans, cocoa, banana, groundnut and palm, indicated that the districts in this study fell under an agro-ecological zone with high agricultural potential. Mogga et al. (2018) also found various staple crops across lowland and upland rice ecologies in South Sudan. There was evident gender segmentation regarding choice of crops to cultivate, with females being more associated with predominantly family-subsistence crops, such as maize, sweet potato, pigeonpea, simsim and groundnut, whereas the male counterparts were more actively cultivating crops, such as sugarcane, rice, cocoa, and palm, which had more economic value in income generation.

The majority of the farmers retained seed from preceding seasons, which was a common practice in self-pollinating crops, such as rice. Retention of rice seed for future planting has been widely reported in major rice-producing countries in Africa (Hubert et al., 2016). However, the exchange of rice planting materials was not only restricted within farming communities but also across districts. This can be an advantage, resulting in the diffusion of varieties, especially when farmers utilize improved cultivars. The saving of seed can also act as a selection method, allowing suitably adapted varieties to perpetuate. However, the widespread retention of seed presents a huge challenge to seed companies because they may not be able to realize viable returns on their newly developed cultivars. Once a cultivar of a self-pollinating crop is released on the market, farmers can use farm-retained seeds for a number of seasons before the seed loses vigour or viability. This means that the farmers will not buy new seed for a number of seasons, which represents a loss of potential loss to seed companies. The presence of different agents in seed dissemination, such as government and NGOs, must be used to leverage the sustainability of developing improved cultivars of rice. The government could play a pivotal role by introducing tariffs on rice products that will be channelled to research and development or subsidizing rice-breeding organizations to promote continuous improvement of rice genetic resources.

The majority of respondents cultivated obsolete rice varieties or landraces in all the surveyed districts, which could be a contributing factor for the low yields obtained by the farmers. Although landraces may harbour important attributes preferred by farmers, such as aroma, good cooking and eating quality, they consistently exhibit low yield. Asante et al. (2013) also found that landraces cultivated in the Ashanti region of Ghana had local farmers' preferred traits. The widespread cultivation of landraces by farmers in this study provides opportunities for introducing new cultivars with higher yield potential. However, the landraces must be

collected and characterized to identify the important attributes that confer advantage over introduced varieties. Landraces are known to carry important genetic attributes and genetic variation that can be useful in rice breeding programs (Mogga et al., 2018). Besides, the development of new high-yielding varieties cannot have an appreciable impact unless the selection takes into account end-user qualities. Thus, researchers have become increasingly aware that incorporating end-user-preferred qualities, including appearance, milling quality, cooking, processing and nutritional quality in technology development, may substantially increase chances of adoption of the technology. The observed differences between males and females in preference for aroma showed that males had no regard for cooking quality but were more concerned with economic returns. In most Africa cultures, males are not involved in food preparation and would thus not be concerned with aroma or cooking quality of rice.

There were only 22.9% of farmers cultivating improved cultivars in this study, highlighting that there was a very low adoption rate of improved cultivars in the study districts. The new improved cultivars could have had higher yield potential but lacked other traits preferred by the farmers. Although high yield is a priority trait in any crop, farmers are also cognizant of preferred traits other than the yield; and any variety lacking the additional traits may be adopted to a lesser extent. This is common when varieties are developed without consulting the farmers or consumers. For instance, Nzomoi and Anderson (2013) found that newly released varieties in East Africa were not widely adopted because they lacked the taste and aroma expected by the farmers and consumers in that region. Hence, the developed improved cultivars cannot have an appreciable impact unless the selection takes into account end-user-preferred qualities, including appearance, milling quality, cooking and eating quality, processing and nutritional quality (Mogga et al., 2018). Therefore, this study suggests that to increase adoption of improved rice varieties, farmers' and consumers' preferred traits and specific end user product profiles should be taken into account through participatory breeding approaches.

Farmers in all the three districts recognized the constraints affecting rice production. Major rice production constraints included diseases, insect pests, drought, the high cost of and inaccessibility to fertilizers, a lack of improved varieties, poor soil fertility and bird attack (Table 5). These factors have been reported in many parts of Africa, especially SSA (Huberth et al., 2016; Atera, et al., 2018; Alibu et al., 2016; de Mey et al., 2012; Mogga et al., 2018). The presence of multiple constraints presents challenges for breeding a single variety incorporating tolerance to multiple stresses. There are concerted efforts to breed for drought tolerance in rice but there has been relatively low success rate because of the complexity of drought tolerance and the inherently high water requirements for rice production. Similarly, research on developing pests and disease resistance in rice has been on-going but success

has been limited because of a lack of stable and horizontal resistance. Most of the pests-resistant and disease-resistant cultivars exhibit vertical resistance and become susceptible to different strains of the pathogen or under different environments. For instance, high yield losses attributable to RYMV are common in Africa because currently there are no known varieties with resistance to the disease, which opens an opportunity to intensify breeding programs aimed at developing resistant varieties. Birds are very difficult to control and there are currently only a limited number of breeding programs aimed at developing cultivars that deter bird attack. Further, high cost of fertilizers was also identified as a major constraint in rice production, contributing to low rice yields across surveyed areas. In addition, lack of improved varieties was a major cause of crop failure and, therefore, was viewed as a major challenge in rice production across all rice ecologies surveyed.

Poor soil fertility was identified as a major constraint to rice production and farmers rarely used fertilizers because of high cost. Low fertilizer use or application of sub-optimal rates occurs in SSA. The consensus among the farmers that poor soil fertility is a production constraint informs breeders to consider developing low nitrogen-tolerant or nutrient-use efficient varieties for dissemination among these farmers. There have been efforts to improve nitrogen use in cereals and it has been shown that nitrogen use can also be improved in rice.

Majority of the respondents preferred breeders to improve agronomic and market traits (Table 2.6). The preferences were related to grain yield, drought tolerance, resistance to disease and earliness. The differences in the ranks between the districts could be attributed to variations in soil type, levels of annual rainfall, rice varieties grown, and the duration of dry spells. Farmers indicated that they would prefer early-maturing varieties and this could help in drought escape as most of the cultivated landraces were late maturing and suffered drought spells and susceptibility to RYMV disease. Developing high-yielding rice varieties that performed better in harsh and unpredictable environments and possessed farmers' preferred traits should maximize the adoption of such varieties and improve productivity in the study areas. The impact of yield improvement in new varieties that lack most of the traits preferred by farmers will be very minimal in most parts of Africa because of low adoption. Incorporation of farmers' knowledge, preferences, and use of the local landraces as a basis for breeding programs will be expected to maximize the adoption of newly developed varieties. In previous surveys conducted by Mehar et al. (2017), farmers considered agronomic traits and marketability as their main criteria for selecting rice varieties.

Most of the female farmers preferred early-maturing cultivars and high marketability as the most important characteristics. The female farmers considered late-maturing varieties more time- and labour-consuming because such varieties would require more weeding, which

would reduce the amount of time available for other household duties. The criteria for selecting rice varieties were influenced by gender differences. Both women and men played prominent roles in rice farming across the districts. However, results indicated that more males than females participated in rice production across the studied districts and this was strongly attributed to the traditional set-up and cultures, where men took the lead in farm planning and decision-making.

2.5 Conclusions

Rice is an important food security crop and source of income for households in rice growing regions in Tanzania. Diseases, such as RYMV, insect pests, drought, the high cost of and inaccessibility to fertilizers, a limited of improved varieties, poor soil fertility and bird damage were the main production constraints. The study also highlighted that rice farming across the study areas was largely dominated by the use of landraces and farmer-saved seed. Rice attributes preferred by farmers were high yield, disease resistance (e.g., resistance to RYMV), drought tolerance, good cooking and eating grain quality, aroma, earliness to maturity and high market value. The information obtained from this study should assist breeders to use farmer-preferred traits as their selection criteria in future cultivar development. A systematic rice-breeding program aimed at improving RYMV resistance and incorporating farmers' preferred traits should be designed and implemented to increase productivity and adoption of new cultivars by the farmers in eastern and southern highland Tanzania.

References

Aldosari, F., M.S.A. Shunaifi, M.A. Ullah, M. Muddassir, and M.A. Noor. 2017. Farmers' perceptions regarding the use of information and communication technology (ICT) in Khyber Pakhtunkhwa, Northern Pakistan. Journal of the Saudia Society of Agricultural Sciences. 18(2): 211-217. doi: 10.1016/ j.jssas.2017.05.004.

Alibu, S., M.H. Otim, S E.A. Okello, J. Lamo, M. Ekobu, and G. Asea. 2016. Farmer's knowledge and perceptions on rice insect pests and their management in Uganda. Agriculture 6: 1-10.

Asante, M.D., B.O. Asante, G.K. Acheampong, S.K. Offei, V. Gracen, H. Adu-Dapaah, and E.Y. Danquah. 2013. Farmer and consumer preferences for rice in the Ashanti region of Ghana: Implications for rice breeding in West Africa. Journal of Plant Breeding and Crop Science *5:* 229-238.

Atera, E.A., F.N. Onyancha, and E.B.O. Majiwa. 2018. Production and marketing of rice in Kenya: Challenges and opportunities. Journal of Development and Agricultural Economics 10: 64-70.

Bagati, S., A.K. Singh, R.K. Salgotra, R. Bhardwaj, M. Sharma, S.K. Rai, and A. Bhat. 2016. Genetic variability, heritability and correlation coefficients of yield and its component traits in basmati rice (*Oryza sativa* L.). SABRAO Journal of Breeding and Genetics 48: 445-452.

Bucheyeki, L K., E. Shennkawa, D. Kadadi, and J. Lobulu. 2011. Assessment of rice production constraints and farmers preferences in Nzega and Igunga district, Tanzania. Journal of Advance in Development Research 2: 30-37.

Chemutai, L.R., M.A. Musyoki, W.F. Kioko, N.S. Mwenda, K.G. Muriira, and N.M. Piero. 2016. Genetic diversity studies on selected rice (*Oryza sativa* L.) genotypes based on gel consistency and alkali digestion. Journal of Rice Research 4:172. doi: 10.4172/2375-4338.1000172.

dey Mey, Y., M. Demont, and M. Diagne. 2012. Estimating bird damage to rice in Africa: Evidence from the Senegal River valley. Journal of Agricultural Economics 63: 175-200.

Food and Agriculture Organization of the United Nations (FAO. 2012). Database available from: http://faostat.fao.org/default.aspx. (Accessed 3 June 2018).

Farid, K.S., N.Z. Tanny, and P.K. Sarma. 2015. Factors affecting adoption of improved farm practices by the farmers of Northern Bangladesh. Journal of Bangladesh Agriculture 13: 291-298.

Hubert, J., A. Luzi-Kihupi, E. Hébrard, and H.J.F. Lyimo. 2016. Farmers' knowledge and perceptions of Rice yellow mottle virus in selected rice growing areas in Tanzania. International Journal of Science and Research 5: 549-559.

Kam, H. 2011. A study of the diversity of Burkina Faso rice landraces and identification of source of resistance to rice yellow mottle virus (RYMV). PhD Dissertation, University of KwaZulu-Natal, Pietermaritzburg, South Africa.

Krupa, K.N., H.E. Shashidhar, N. Dalawai, M. Reddy, and V.H. Swamy. 2017. Molecular marker based genetic diversity analysis in rice genotypes (*Oryza sativa* L.) using SSR markers. International Journal of Pure and Applied Bioscience 5: 668-674.

Longué, R.D.S., I. Zinga, S. Semballa, N. Barro, and O. Traoré. 2016. Detection and serological characterization of rice yellow mottle virus in Central African Republic. Agricultural Sciences 7: 911-919.

Match Maker Associates. 2010. Value Chain Analysis of Rice and Maize in Selected Districts in Tanzania, Rice and Maize Context Report: Dar es Salaam, Tanzania.

Mehar, M., T. Yamano, and A. Panda. 2017. The role of gender, risk, and time preferences in farmers' rice variety selection in eastern India. Asian Journal of Agriculture and Development 1(14): 1-20.

Mogga, M., J. Sibiya, H. Shimelis, J. Lamo, and N. Ochanda. 2018. Appraisal of major determinants of rice production and farmers choice of rice ideotypes in South Sudan: implications for breeding and policy interventions. Experimental Agriculture 1-14. doi: 10.1017/S0014479718000017.

Mrema, E., H. Shimelis, M. Laing, and T. Bucheyeki. 2017. Farmers' perceptions of sorghum production constraints and Striga control practices in semi-arid areas of Tanzania. International Journal of Pest Management 63: 146-156.

Nzomoi, J., and I. Anderson. 2013. The rice market in East Africa. Paper presented at the 4th International Conference of the African Association of Agricultural Economists, Hammamet, Tunisia, September 22-25. http://ageconsearch. umn.edu/bitstream/161272/2/JOSEPH%20NZOMOI%20%20%26%20IAN%20ANDERSON.

Pinel-Galzi, A., C. Dubreuil-Tranchant, E. Hébrard, C. Mariac, A. Ghesquière, and L. Albar. 2016. Mutations in rice yellow mottle virus Polyprotein P2a involved in RYMV2 gene resistance breakdown. *Frontier of Plant Science* 7: 1-11.

Sereme, D., I. Ouedraogo, B.J. Neya, P.E. Zida, N. Yao, and M. Sié. 2016. Screening improved rice varieties (*Oryza spp*) for their resistance/tolerance to rice yellow mottle virus in West Africa. International Journal of Agriculture Innovations and Research 5: 481-486.

Shelton, A.C., and W.F. Tracy. 2016. Participatory plant breeding and organic agriculture: A synergistic model for organic variety development in the United States. Elementa Science of the Anthropocene 4: 1-12.

SPSS. Statistical Package for Social Sciences. 2017. SPSS version 24, Chicago (IL): SPSS Inc.

Srujana, G., B.G, Suresh, G.R, Lavanya, J.B. Ram, and V. Sumanth. 2017. Studies on genetic variability, heritability and genetic advance for yield and quality components in rice (*Oryza sativa* L.). Journal of Pharmacognosy and Phytochemistry 6: 564-566.

Tsinigo, E. 2014. Economics of Technological Change in Rice Production in the Ejura-Sekyedumase and Atebubu-Amintin districts. Master Dissertation, College of Agriculture and Renewable Natural Resources, Ghana.

CHAPTER THREE: VARIATION AMONG TANZANIA RICE GERMPLASM COLLECTIONS BASED ON AGRONOMIC TRAITS AND RESISTANCE TO RICE YELLOW MOTTLE VIRUS

Abstract

Rice (*Oryza* species) is a commercial crop worldwide. Across Africa, the potential yield and quality of rice is diminished by lack of high performance, locally adapted varieties, and the impact of rice yellow mottle virus (RYMV). The objective of this study was to assess the performance of diverse collections of rice germplasm for RYMV resistance and agronomic traits, and to select promising lines for breeding under Tanzanian conditions. Fifty-four rice genotypes were field evaluated in two important rice production sites (Ifakara and Mkindo) in Tanzania, which are recognized as RYMV hotspots, using a 6 × 9 alpha lattice design with two replications. There was significant ($p<0.05$) genotypic variation for agronomic traits and RYMV susceptibility in the tested germplasm. Seven genotypes with moderate to high RYMV resistance were identified, namely, Salama M-57, SSD1, IRAT 256, Salama M-55, Mwangaza, Lunyuki, and Salama M-19, which were identified as new sources of resistance genes. Positive and significant correlations were detected between grain yield and number of panicles per plant (NPP), panicle length (PL), number of grains per panicle (NGP), percentage-filled grains (PFG), and thousand-grain weight (TGW). These would be useful traits for simultaneous selection for rice yield improvement. A principal component analysis resulted in five principal components accounting for 79.88% of the total variation present in the assessed germplasm collection. The traits that contributed most to the gross variability included NPP, NT, PL, GY, and DFL. The genotypes Rangimbili, Gigante, and SARO possess complementary agronomic traits and RYMV resistance, and can be recommended for further evaluation, genetic analysis and breeding.

Keywords: Agronomic traits, cultivar development, principal components, RYMV resistance

---

Suvi, W.T., H. Shimelis, M. Laing, I. Mathew, and A.I.T. Shayanowako. 2021. Variation among Tanzania rice germplasm collections based on agronomic traits and resistance to rice yellow mottle virus. Agronomy11, 391. doi. org/10.3390/agronomy11020391.

3.1 Introduction

Rice (*Oryza sativa* L., 2n = 2x = 24) is an economically important crop in East, Central and West Africa (Mwalyego et al., 2018) and globally (Zhang et al., 2014). Rice is a source of 80% of the caloric intake for nearly one billion people in Africa (Tao and Li, 2018). Africa accounts for nearly 3% of the global rice production. About 25% of Africa's rice requirements have to be met with imports due to the low levels of local production, high levels of population growth, quality preferences, urbanisation and changes in life style (Balasubramanian et al., 2007).

Rice is widely cultivated and commercialized in Tanzania for food security and as a cash crop, ranking second after maize in total production and consumption (Hubert et al., 2017a). The crop is largely cultivated by small-scale farmers on less than one-hectare per household. Rice yields in Tanzania remain low, with yields of 1.0 and 1.5 t ha^{-1} compared to the yield potential of the crop of 5.0 t ha^{-1} (Kilimo-Trust, 2012; FAO, 2015). The low yields are caused by biotic, abiotic and socio-economic constraints prevalent in sub-Sahara Africa (Mghase et al., 2010; Hubert et al., 2016; Suvi et al., 2018).

Rice yellow mottle virus (RYMV) disease is the major biotic constraint under both the rain-fed and irrigated rice production agro-ecologies in Tanzania (Lamo et al., 2015; Suvi et al., 2018). Due to RYMV infection, yield losses between 20 and 100% have been recorded in susceptible rice varieties (Kouassi et al., 2005; Luzi-Kihupi et al., 2009; Longué et al., 2016). RYMV emerged in 1966 in sub-Sahara Africa (SSA) (Bakker, 1974). The RYMV is found in all rice production systems in Africa affecting 23 countries including Tanzania (Rossel et al., 1982; Hubert et al., 2013; Longué et al., 2014). RYMV transmission and distribution is mainly facilitated by insect vectors, irrigation water, wind, field workers and farm animals (Ochola et al., 2015). Infected volunteer rice plants from a previous crop are ideal sources of RYMV infection to newly planted and healthy crops (Suvi et al., 2018). Several chewing insect species, notably the Chrysomelid beetles (*Sesselia pusilla, Chaetocnema pulla, Trichispa sericea, Dicladispa viridicyanea*) and grasshoppers (*Conocephalus merumontanus*) are among the key vectors that transmit RYMV from cultivated rice, wild hosts and weeds to healthy rice crop stands (Kanyeka et al., 2007).

The RYMV is an icosahedral virus belonging to the genus *Sobemovirus* (Fauquet and Mayo, 1999). The pathogen is extremely stable and remains highly infectious under favourable environmental conditions (Bakker, 1974). Under controlled environment conditions, the RYMV remains infectious for 33 days but looses its pathogenicity after about 51 days (Sy and Sere, 1996). Bakker (1974) reported that with high ambient temperatures (>30^{o}C), RYMV induces systemic symptoms 4 to 5 days after infection, while prolonged periods of temperatures below 20 ^{o}C delay symptom appearance up to 10 to 12 days. There are various RYMV strains, based

on their geographical and ecological origins (Traoré et al., 2010). The diversity among RYMV strains in Africa was first assessed using polyclonal and monoclonal antibodies (N'guessan et al., 2000; Kanyeka et al., 2007). The RYMV diversity was subsequently characterized using the reverse transcriptase polymerase chain reaction for two primers, Prymv1 and Prymvy2 in Tanzania. This indicated the presence of three RYMV strains in the country, each belonging to a specific and restricted geographical range (Mpunami et al., 2012; Longue et al., 2017). RYMV strains exhibit differences in virulence and pathogenicity, resulting in differential reactions by rice genotypes. Some RYMV-resistant rice cultivars have become susceptible when cultivated in new agro-ecologies due to the emergence of virulent strains (Kam et al., 2013).

RYMV infection and disease development is characterized by the appearance of mottling and yellowing spots (Kouassi et al., 2005), which coalesce and become parallel to the leaf veins about two weeks after infection (Koudamilor et al., 2014). Infected plants show stunted growth, reduced tillering ability, non-synchronous flowering, poor panicle exertion, reduced number of spikelets, grain sterility and brown to dark brown discoloration of grains. Under severe infection RYMV leads to the death of infected plants and crop failure (Abo et al., 2000; Sereme et al., 2016). RYMV infection and disease development is affected by the virulence of the virus strain, the rice genotype, the growth stage of the plants, the environment (e.g. light intensity, day length, humidity and temperature) and their interactions. Field incidence, severity assessment and serological analysis are the most widely used methods for RYMV diagnosis, rating and cultivar evaluation. Visual rating of RYMV infection is relatively easy, and is more efficient when evaluating a set of genotypes that include both resistant and susceptible controls (Abo et al., 2000).

Various control measures have been recommended for the management of RYMV (Suvi et al., 2018). These include the use of various crop protection chemicals, cultural practices, biological control agents and host plant resistance. Chemical insecticides are widely used for suppressing the population of the RYMV transmitting vectors (Traore et al., 2015). However, there are many vectors of RYMV, which are present at different crop growth stages, thereby necessitating repeated applications of pesticides. Consequently, this practice is expensive and increases the cost of rice production. Small-scale farmers in Tanzania cannot afford to purchase chemical pesticides, and consequently they use sub-optimal rates, leading to poor efficacy and pesticide resistance. The prolonged use of chemicals with similar modes of action or from the same group such as belonging to organochlorines, organophosphates, and carbamates has led to the development of pesticide-resistant pest populations due to mutation events (Suvi et al., 2018). Pesticide resistance leads to the application of increasingly higher volumes of chemicals driving the cost of production even higher. Furthermore, these insecticides pose health hazards to the

farmers, and create environmental pollution issues in the long term. RYMV can be partially managed using cultural practices such as residue burning, destroying volunteer plants, and using scheduled crop rotations to deprive the pathogen of any alternate hosts. However, these practices are time-consuming and have limited efficiency in controlling RYMV disease (Pidon et al., 2017). In addition, crop rotation is not implemented by smallholder farmers because their farms are too small to apply this effectively. Furthermore, the availability of labour is limited, impeding the practice of field sanitation (Hubert et al., 2016; Nkuba et al., 2016). Furthermore, the spread of RYMV by multiple agents renders these management practices relatively ineffective (Suvi et al., 2018), and hence alternative and effective integrated options are required.

Host plant resistance is a cost-effective and sustainable strategy to control RYMV. High levels of RYMV resistance have the potential to increase rice productivity in RYMV endemic regions, while reducing the cost of production. Cultivars with RYMV resistance require reduced levels of crop protection chemicals and should attain significantly higher yields. Successful deployment of RYMV-resistant cultivars depends on the identification of sources of RYMV resistance genes among divergent and complementary parental lines. The *RYMV1* (allele *rymv1-2*) and *RYMV2* genes have been identified as two RYMV resistance genes in *Oryza* species (Ndjiondjop et al., 1999; Thiemele et al. 2010; Pinel-Galzi et al., 2016). Furthermore, resistance conferred by the RYMV3 gene has been identified in an *O. glaberrima* accession, Tog5307 (Pinel-Galzi et al., 2016).

Currently, there are no rice varieties with known RYMV resistance grown in Tanzania. The majority of introduced rice varieties and landraces that have been grown in Tanzania have succumbed to RYMV infection (Kouassi et al., 2005). Most introduced cultivars and landraces that are currently in production or have been preserved in gene banks have not been systematically evaluated in RYMV resistance breeding programs in Tanzania. There is a need to evaluate the locally available genotypes and introductions with known RYMV resistance to develop agronomically superior and resistant cultivars. Sources of RYMV resistance have been identified in *O. sativa* varieties such as Gigante and Bekarosaka, and *O. glaberrima* varieties such as Tog5681, Tog5672, Tog5674 and Tog7291 (Munganyinka et al., 2016; Pidon et al., 2017). These genetic resources could be valuable for breeding RYMV-resistant rice varieties for Tanzania.

Understanding the extent of genetic variation present in a germplasm collection and selection of complementary lines with economic traits and RYMV resistance is a prerequisite for rice improvement (Xiao et al., 2016). Wide phenotypic variations exist among Tanzanian rice

landraces and introduced genotypes (Mausa, 2014). The genetic diversity present in the Tanzanian rice collections could be explored using morphological, biochemical and molecular (DNA) markers. Bakar (2010) used SSR markers to analyse 70 rice landraces in Tanzania. Mausa (2014) characterized the genetic diversity present in 79 Tanzanian rice landraces using SSR markers. Similarly, Suvi et al. (2019) assessed the genetic diversity and population structure of 54 rice genotypes using SSR markers. Morphology-based characterization has been widely used in rice as a quick, easy, and less costly approach than DNA-based marker systems (Aida et al., 2007).

There are few recent phenotypic diversity studies on rice for agronomic traits and RYMV resistance in Tanzania. Mangosongo et al. (2019) characterized wild rice populations from some selected areas of Tanzania using morphological traits. Furthermore, Musyoki et al. (2015) undertook diversity analysis based on selected Tanzanian and Kenyan rice genotypes. However, comprehensive and up-to-date data is lacking on agro-morphological descriptions and assessment of RYMV resistance in Tanzanian rice genetic resources using diverse populations, landraces and introduced varieties. This will ensure the selection of parental genotypes with resistance to RYMV and desirable agronomic traits for genetic enhancement and effective breeding. Therefore, the objective of this study was to assess the performance of diverse rice germplasm collections for RYMV resistance and agronomic traits, and to select promising lines for breeding.

3.2 Materials and methods

3.2.1 Plant materials

The study used a panel of 54 rice genotypes, which comprised of farmers' landraces and introduced collections from the Tanzania Agricultural Research Institute (TARI), Sokoine University of Agriculture (SUA) in Tanzania, AfricaRice in Benin and Côte d'Ivoire, and from smallholder farmer fields in Tanzania. The details of the germplasm used in the study are summarised on Table 3.1. The panel included 29 landraces that are adapted to Tanzania agro-ecologies and grown widely by small-scale farmers. The landraces are predominantly aromatic and are preferred by farmers and local markets. Six genotypes belonging to the New Rice for Africa (NERICA) types were included. The NERICA types were developed by the AfricaRice Consortium from interspecific crosses between *O. glaberrima* (African rice) and *O. sativa* (Asian rice) species. Genotype, Gigante, a rice cultivar widely cultivated in West Africa, was included. The NERICA and Gigante genotypes were introduced in Tanzania in 2008 by AfricaRice, and are usually grown under upland and lowland agro-ecologies, respectively. There were five genotypes that were introduced from the International Rice Research Institute (IRRI) in the Philippines. These genotypes are adapted and cultivated under paddy production systems. The

paddy types were included in the study for their high yield potential, although these genotypes are susceptible to drought stress. Furthermore, the test genotypes included six irrigated and seven lowland rain-fed genotypes, which had been developed by TARI and SUA, respectively.

Table 3.1. List of the rice genotypes used in the study and their sources

Sr. No	Genotypes	Origin/source	Sr. No.	Genotypes	Origin/source
1	Salama M-57	SUA/Tanzania	28	Kalubangala	Landrace/Tanzania
2	SSD 1	SUA/Tanzania	29	Mpaka wa bibi	Landrace/Tanzania
3	Nerica 7	AfricaRice/Benin	30	Mbawambili nyekundu	Landrace/Tanzania
4	Kalalu	SUA/Tanzania	31	Limota	Landrace/Tanzania
5	IRAT 256	AfricaRice/Benin	32	Moshi	Landrace/Tanzania
6	SARO	TARI/Tanzania	33	Shingo ya mwali	Landrace/Tanzania
7	Nerica 1	AfricaRice/Benin	34	Kalundi	Landrace/Tanzania
8	Serena	Landrace/Tanzania	35	IR54	IRRI/Philippines
9	Nerica 4	AfricaRice/Benin	36	TXD 88	TARI/Tanzania
10	WAB450	AfricaRice/Benin	37	IR 56	IRRI/Philippines
11	Mbega	Landrace/Tanzania	38	IR64	IRRI/Philippines
12	Salama M-55	SUA/Tanzania	39	Mzinga	Landrace/Tanzania
13	Mwangaza	SUA/Tanzania	40	Afaa mwanza	Landrace/Tanzania
14	Nerica 2	AfricaRice/Benin	41	TXD 85	TARI/Tanzania
15	Lunyuki	TARI/Tanzania	42	TXD 307	TARI/Tanzania
16	Turiani	Landrace/Tanzania	43	Sumbawanga	Landrace/Tanzania
17	Mbawa ya njiwa	Landrace/Tanzania	44	Supa	Landrace/Tanzania
18	Chamota	Landrace/Tanzania	45	Rangi mbili nyekundu	Landrace/Tanzania
19	IR72	IRRI/Philippines	46	Faya mzinga	Landrace/Tanzania
20	Salama M-19	SUA/Tanzania	47	TAI	TARI/Tanzania
21	Masantula	Landrace/Tanzania	48	Gombe	Landrace/Tanzania
22	IR 68	IRRI/Philippines	49	Kisegese	Landrace/Tanzania
23	Kalamata	Landrace /Tanzania	50	Gigante	AfricaRice
24	Zambia	Landrace/Tanzania	51	Sindano nyeupe	Landrace/Tanzania
25	Ringa	Landrace/Tanzania	52	Kihogo red	Landrace/Tanzania
26	Wahiwahi	Landrace/Tanzania	53	Cherehani	Landrace/Tanzania
27	Mwanza	Landrace/Tanzania	54	ITA 303	TARI/Tanzania

SUA = Sokoine University of Agriculture; IRRI = International Rice Research Institute; TARI = Tanzania Agricultural Research Institute; Sr. No = serial number.

3.2.2 Description of experimental sites

The field trials were conducted at two selected sites in Tanzania; namely: Mkindo situated in the Mvomero district; and Ifakara in the Kilombero district. The sites were purposefully selected for being the major rice production agro-ecologies (Wilson, 2018) with high levels of RYMV infection (Hubert *et al*., 2016). The Ifakara site (08°03'693''S; 036°40'005'' E, 286 masl) is characterized by two cropping seasons based on the amount of rainfall received. The short crop season commences in November and ends in February, while the long rainy season starts in March and ends in May or June. The total annual rainfall received at this site is 935 mm. The monthly temperatures range between 26 ^{0}C and 32 ^{0}C. Heavy clay soils with a pH of 6.0 are dominant at the Ifakara site. The site at Mkindo is located at latitude of 06º15.344' S and longitude of 037º32.387' E, with an altitude of 345 to 365 meters above sea level (masl). The site has a bimodal rainfall. The short rainy season extends from October to December, while the long rainy season occurs between March and May. The average annual temperature is 24 ^{0}C with a minimum of 15 ^{0}C in June and a maximum of 32 ^{0}C in February. The dominant soil texture at the Mkindo site is clay loam with a pH of 6.2. The Mvomero and Kilombero districts are

recognized hotspots for RYMV. The disease can cause yield losses of 100% under epidemic conditions (Kanyeka et al., 2007).

3.2.3 Experimental design and management

The experiments at both sites were laid out in a 6 × 9 alpha lattice design with two replications. The plot size was 2.4 m x 2.4 m in which plants were spaced 20 cm between rows. Seeds were directly sown at the Ifakara site at the beginning of February in 2018. Experimental units at the Mkindo site were established using seedling transplants. Seedlings were transplanted in April, 21 days after sowing, with one seedling per hill. Gap filling was done as necessary within two weeks after direct sowing or transplanting to ensure uniform crop stands. Nitrogen fertilizer was applied at a rate of 80 kg N ha^{-1} in the form of urea (46% N) in two installments as a top dressing. The first and second applications were done at the tillering and booting stages, respectively. Hand weeding was carried out three times at each site to prevent weed infestation. After direct seeding or transplanting, sufficient soil moisture was maintained in each plot using supplemental irrigation at both sites.

3.2.4 Data collection

Quantitative agronomic traits and RYMV resistance were recorded according to the descriptors of IRRI (2002). RYMV severity was scored on a scale of 1 to 9; where: 1 represented no symptoms; 3 represented plants with sparse dots or streaks on green leaves and less than 5% reduction in plant height;;5 represented plants with mottling on green or pale green leavesand 6 to 25% reduction in plant height and slightly delayed flowering; 7 represented plants with yellow or pale yellow leaves with a 26 to 75% reduction in plant height and delayed flowering; and 9 was assigned to plants with yellow or orange leaves with more than 75% reduction in plant height and no flowering.

Data on the following agronomic traits were collected: days to 50% flowering (DFL) counted from sowing to the date when half of the plants in a particular plot had flowered; number of tillers per plant (NT) counted at physiological maturity and recorded as the average of 10 selected and tagged plants in a row; number of panicles per plant (NPP) counted from ten plants at harvest and recorded as the number of fully exerted panicles bearing grains and recorded as an average per plant; plant height (PH in cm) measured from the soil surface to the tip of the longest panicle on ten tagged plants in each plot; panicle length (PL) measured in centimetre from the tip of the panicle to the ciliate ring at the base on the 10 selected plants per plot; number of grains per panicle (NGP) counted using a seed counter and recorded as a mean of 10 panicles per plot; percent filled grains (PFG) calculated as the proportion of unfilled grains to the total number of grains from 10 sampled panicles per plot; 1000-grain weight (TWG expressed in grams) for each

genotype using an Elmor seed counter (model, source) and weighed on an electric balance in grams; and grain yield (GY) weighed per plot after adjusting to 14% moisture content and converted to tons per hectare.

3.2.5 Data analysis

The data were subjected to analysis of variance (ANOVA) using the restricted maximum likelihood model (REML) procedure for alpha lattice designs in GenStat 24th edition (Payne et al., 2017). Genotype was set as a fixed factor, while location and genotype by location interaction, replication and block were treated as random factors using the following model:

$$Y_{ijkl} = \mu + G_i + L_j + GL_{ij} + R_{k(j)} + B_{l(j,r)} + \varepsilon_{ijkl}$$

Where: μ is the overall mean, and Gi, Lj, GLij, Rk(j), and Bl(j,r) represent the effects of genotype, location, the genotype × location interaction, replication in location, and the incomplete block in replication, in that order. Ɛ is the random error term. Traits means were separated by the Fischer's unprotected least significant difference at the 5% probability level. The correlations among traits were computed using the Pearson correlation procedure with the SPSS version 24 (SPSS, 2017). A correlation matrix based principal component analysis (PCA) was performed to elucidate the genotype-trait relationships with a biplot generated in Genstat 24th edition (Payne et al., 2017).

3.3 Results

3.3.1 Analysis of variance (ANOVA) for grain yield and yield-related traits, and the RYMV disease score

Table 3.2 summarizes the results from the combined ANOVA for all the measured agronomic traits and the RYMV disease score. The genotype × site interaction effects were highly significant ($p<0.001$) for PH, PL, NGP, PFG, TGW, RYMV, and GY. Highly significant differences were detected among the genotypes and sites for all the measured agronomic and RYMVD parameters, except for DFL.

3.3.2 Mean performance of genotypes for agronomic traits and the RYMVD resistance

The genotypes exhibited variable agronomic performance and RYMVD reactions across the two sites (Table 3.3). The mean DFL among the test genotypes was 85 days. Genotypes such as Cherehani, SSD1,WAB450,Mwangaza, Ringa and Mbawambili were the earliest to reach 50% flowering, after 57, 62, 64, 69, 71 and 72 days at the Ifakara and Mkindo sites. Genotype, Mpaka wa bibi was the slowest to flower after 104 days at the Ifakara and Mkindo sites. In terms of tillering capacity, genotypes, Gigante, Rangimbili nyekundu, IR64, IR72 and Shingo ya mwali produced the most tillers per plant at both sites, while Sumbawanga had a mean of five tillers at

each site. The PH ranged between 77 and 156.7 cm, with a mean of 108.7 cm. Genotypes Mwanza, TXD85 and TXD307 were the shortest genotypes with PHs of 77.8 cm, 82.6cm and 84.2 cm, respectively, at both sites. Genotype, IRAT 256 was the tallest at 157.7cm. NPP ranged from 4 to 10 with a mean of 7. Genotypes, Gigante and Sumbawanga recorded the highest and lowest NPP values, respectively. The trait PL ranged from 18.7 to 25.3cm, with a mean value of 22 cm. Genotypes, IRAT 256, Serena and Mpaka wa bibi had the longest PL values, while the shortest PLs were recorded for the genotypes, Nerica 1 (19.8 cm) and Nerica 7 (20 cm). NGP ranged from 85 to184 with a mean of 143. Genotypes, Serena, Kisegese, Gigante and Zambia had the highest NGPs of 184, 182, 179 and 178, respectively. The lowest NGPs were recorded for genotypes, Mwangaza (88) and IR64 (100). PFG varied from 83.7 to 97.4% with a mean of 92.3%. Genotypes, Nerica 7, IRAT 256 and Salama M-55 had the highest PFG values of 96.8, 95.9 and 95.8%, respectively. The TGW ranged from 23 to 37.2 g with a mean of 30.2 g. Heavier TGW values of 37.2, 36.5 and 35 g were recorded for the genotypes, Mwangaza, Mbega and Salama M-55, respectively. The genotype, Mpaka wa bibi followed by Limota, Kalalu, and IR56 had the lowest TGWs.

The RYMVD ratings ranged from 1 to 7 with a mean of 5. Genotypes, Salama M-57, SSD1, IRAT 256, Lunyuki, Salama M-19, Salama M-55, and the resistant check Mwangaza exhibited highly resistant reactions to RYMVD with scores of 1. Genotypes with a RYMVD score of 3 included Nerica 1, Nerica 2, Nerica 7, IR56, IR64, IR68, Kalalu, TXD307, and TAI. Moderately resistant genotypes with RYMVD ratings of 5 included Turiani, Moshi and Shingo ya mwali. The other genotypes, including the susceptible check SARO were susceptible with RYMVD ratings between 5 and 7. The mean GY of the test genotypes was 2.5 t ha^{-1}. The genotype,s with the highest GY values were SARO (4.1 t ha^{-1}), and Rangimbili nyekudu and Mbega (>3.7 t ha^{-1}), while Nerica 4 (1.0 t ha^{-1}) had the lowest GY.

Table 3.2. Mean squares and F-tests for agronomic traits and RYMVD reaction among 54 rice genotypes evaluated at two locations in Tanzania

Source of variation	DF	DFL	NT	NPP	PH	PL	NGP	PFG	TGW	RYMVD	GY
Site	1	0.00^{ns}	6.97*	21.41***	797.72***	228.93***	1533.33***	1561.80***	97.34***	1.85***	52.25***
Rep (Site)	1	0.93^{ns}	0.47^{ns}	1.16ns	22.89^{ns}	5.55*	22.26^{ns}	772.20***	2.08	0.00	0.17*
Block(Rep)	32	167.83***	2.82***	2.20**	371.84***	3.25***	623.91***	26.53***	10.19***	7.73***	0.42***
Genotype	53	450.42***	3.95***	4.13***	945.34***	3.25***	2539.17***	10.57***	52.20***	7.62***	1.65***
Genotype x Site	53	0.00^{ns}	1.27^{ns}	1.34^{ns}	120.77***	4.11***	146.09***	11.67***	4.98**	1.73***	0.59***
Residual	106	1.40	1.22	1.15	31.37	1.28	36.83	4.78	2.39	0.08	0.04

DF= degrees of freedom; DFL= Days to 50% flowering; NT= number of tillers/plant; NPP=number of panicles/plant; PH= plant height; PL= panicle length; NGP = number of grains/panicle; PFG = percentage filled grains/panicle; TGW = 1000 grain weight; RYMVD = rice yellow mottle virus disease reaction; GY = grain yield; * = $P < 0.05$; ** = $P < 0.01$; *** = $P < 0.001$; ns, non-significant.

Table 3.3. Mean values for agronomic traits and RYMVD reaction of 54 rice genotypes evaluated at two locations in Tanzania

Entry	Genotype	DFL		NT		NPP		PH		PL		NGP		PFG		TGW		RYMVD		GY	
		Ifa	Mk	Ifa	Mk	Ifa	Mk	Ifa	Mk	Ifa	Mk	Ifa	Mk	Ifa	Mk	Ifa	Mk	Ifa	Mk	Ifa	Mk
1	Salama M-57	86	84	8	6	7	6	133.2	130.9	22.8	22.4	141	143	96.3	92.9	30.5	34.5	1	1	2.0	2.3
2	SSD1	60	63	7	7	6	5	123.9	124.0	20.0	20.9	100	99	95.1	92.7	33.0	35.5	1	1	2.7	2.8
3	Nerica 7	74	76	8	7	7	7	108.7	102.3	19.2	20.8	142	135	96.1	97.4	26.5	30.5	3	3	2.2	2.0
4	Kalalu	77	74	7	6	7	6	100.1	95.0	19.3	22.3	142	137	94.5	87.1	23.0	25.5	3	3	2.3	2.4
5	IRAT 256	73	76	7	5	7	4	156.7	146.2	24.0	25.3	106	105	96.3	95.5	29.5	33.0	1	1	1.4	1.7
6	Gigante	95	93	10	11	10	10	97.1	98.5	19.7	22.0	176	181	96.4	91.4	30.0	30.0	5	5	3.7	3.5
7	Nerica 1	74	72	8	7	7	6	85.1	89.9	19.4	20.2	127	164	95.7	89.5	29.5	32.5	3	3	2.5	2.6
8	Serena	91	94	8	9	7	8	110.2	112.9	23.6	23.9	183	184	84.1	93.3	31.0	31.5	5	3	3.6	3.3
9	Nerica 4	78	76	7	5	6	5	101.5	96.9	20.1	22.3	116	119	95.1	91.6	29.0	29.5	5	3	1.0	1.1
10	WAB450	65	63	8	8	6	7	103.1	99.4	20.2	21.1	118	98	94.8	95.9	28.0	30.5	5	5	1.8	1.7
11	Mbega	85	82	7	6	6	6	127.5	126.8	23.2	22.2	160	156	95.4	87.2	36.4	36.5	5	5	3.9	3.6
12	Salama M-55	86	89	9	6	9	7	133.2	117.6	21.1	21.3	149	163	96.0	95.6	35.0	35.0	1	1	1.5	3.2

Table 3.3. Continued

Entry	Genotype	DFL		NT		NPP		PH		PL		NGP		PFG		TGW		RYMVD		GY	
		Ifa	Mk	Ifa	Mk	Ifa	Mk	Ifa	Mk	Ifa	Mk	Ifa	Mk	Ifa	Mk	Ifa	Mk	Ifa	Mk	Ifa	Mk
13	Mwangaza	67	70	8	7	7	6	116.5	99.2	19.6	21.0	91	85	94.7	94.8	37.0	37.3	1	1	1.2	1.3
14	Nerica 2	77	79	7	7	6	6	85.6	86.1	19.2	21.1	137	140	93.4	90.0	25.5	32.0	3	3	1.9	2.1
15	Lunyuki	78	76	8	7	8	7	124.0	122.1	18.9	21.5	146	139	95.5	89.5	29.5	32.5	1	1	3.2	3.3
16	SARO	90	93	7	7	6	6	89.0	87.4	23.0	22.9	154	153	91.0	91.8	34.0	34.0	5	5	3.9	4.0
17	Mbawa ya njiwa	76	80	8	7	8	7	112.8	109.7	20.2	22.9	142	156	95.1	88.7	28.0	27.4	7	5	1.9	2.1
18	Chamota	91	89	8	7	8	6	118.0	127.6	19.1	22.6	164	168	95.5	88.4	25.5	23.5	7	5	2.7	2.2
19	IR72	92	90	9	9	8	8	89.5	84.5	20.8	23.8	155	152	93.6	89.4	29.5	30.5	5	3	2.0	2.5
20	Salama M-19	79	81	9	6	8	5	115.1	114.4	20.5	22.4	129	115	96.9	91.4	30.5	32.5	1	1	1.7	1.8
21	Masantula	102	101	8	9	8	8	124.0	126.7	20.1	22.8	109	123	96.9	88.7	23.0	26.5	7	5	2.1	2.5
22	IR68	94	90	7	9	6	8	87.6	87.9	19.2	22.9	147	143	96.2	89.9	25.0	26	3	3	1.9	2.1
23	Kalamata	91	96	7	6	6	6	126.4	118.4	18.7	19.1	168	174	95.6	89.6	34.0	35.0	5	5	2.7	2.7
24	Zambia	90	91	6	7	6	6	115.8	125.2	21.5	20.5	177	179	90.9	92.3	30.0	27.5	5	5	2.5	2.8
25	Ringa	73	69	9	7	9	7	116.2	113.3	20.8	21.5	163	161	95.4	90.9	31.0	33.0	6	7	1.5	2.0
26	Rangimbili nyekundu	73	75	10	10	8	9	105.6	112.9	21.3	23.4	97	139	93.6	89.9	32.5	34.0	7	5	3.7	3.8
27	Mwanza	88	87	8	7	8	6	78.5	77.0	19.6	23.1	143	154	94.5	88.3	26.5	32.0	7	5	1.5	1.7
28	Kalubangala	88	89	8	6	7	6	84.5	108.3	19.0	24.7	115	160	96.0	85.4	29.5	35.5	7	3	2.8	2.2
29	Mpaka wa bibi	103	104	9	8	9	7	104.3	113.3	22.2	23.6	114	145	96.1	89.1	23.0	23.5	5	7	1.7	2.3
30	Mbawambili	71	72	7	8	7	7	116.6	116.4	21.2	22.3	123	134	93.9	89.7	28.5	27.5	7	5	2.3	2.0
31	Limota	79	80	7	7	7	6	116.5	109.9	19.8	21.4	143	152	95.0	85.1	24.5	24.0	5	7	1.5	2.0
32	Moshi	92	93	7	7	7	6	129.0	126.6	21.6	23.7	169	174	96.5	87.3	28.0	29.2	5	5	2.8	3.1
33	Shingo ya mwali	73	74	9	9	9	8	110.7	104.1	21.9	25.0	102	103	96.5	91.7	33.5	36.0	5	5	3.1	3.3
34	Kalundi	99	101	7	6	6	6	127.2	105.5	21.5	22.6	163	166	96.5	88.9	30.5	29.0	5	7	1.9	2.0
35	IR54	90	94	8	6	8	5	95.2	91.1	19.6	21.5	148	176	94.8	84.9	27.0	27.5	5	3	2.0	2.0
36	TXD88	92	95	7	9	7	7	90.0	86.0	19.8	21.4	126	149	95.2	86.0	29.5	32.0	5	3	3.1	2.9
37	IR 56	77	74	6	7	6	7	96.4	97.8	19.8	21.7	163	141	94.4	86.1	22.5	26.0	3	3	2.3	2.4
38	IR 64	75	79	9	10	8	9	86.9	85.8	20.7	21.7	98	102	96.6	88.8	27.0	28.5	3	3	2.8	3.2
39	Mzinga	92	95	8	9	7	8	98.3	87.3	20.4	21.8	117	129	95.8	88.8	26.5	27.5	5	5	2.0	2.3
40	Afaa Mwanza	89	92	6	7	6	6	117.3	117.6	22.3	22.1	168	166	93.5	87.3	31.5	35.5	7	5	1.8	1.8

Table 3.3. Continued

Entry	Genotype	DFL		NT		NPP		PH		PL		NGP		PFG		TGW		RYMVD		GY	
		Ifa	Mk	Ifa	Mk	Ifa	Mk	Ifa	Mk	Ifa	Mk	Ifa	Mk	Ifa	Mk	Ifa	Mk	Ifa	Mk	Ifa	Mk
41	TXD 85	97	95	9	7	8	7	83.1	82.1	19.8	22.3	119	117	96.1	85.4	29.0	30.5	3	3	2.2	3.1
42	TXD 307	98	100	8	10	8	8	89.4	78.9	19.1	23.8	110	113	93.2	85.1	29.0	30.5	3	3	1.8	2.5
43	Sumbawanga	80	81	5	5	4	5	123.3	123.6	22.7	20.4	179	173	96.2	93.1	34.0	35.0	5	5	2.6	2.8
44	Supa	84	87	7	7	7	7	130.0	115.9	20.9	23.2	153	169	95.9	89.8	34.0	33.0	5	7	1.9	2.5
45	Wahiwahi	80	83	6	6	6	5	121.3	116.3	22.4	22.5	159	168	83.7	84.5	25.0	26.0	5	7	1.5	1.4
46	Faya mzinga	87	88	8	7	6	6	128.0	119.3	20.9	21.0	156	172	96.4	91.3	34.5	35.0	5	5	3.2	3.4
47	TAI	79	80	7	9	7	8	95.0	87.6	20.5	22.0	116	112	96.1	85.5	26.0	28.0	3	3	3.5	3.7
48	Gombe	88	89	6	6	6	6	132.4	126.0	22.1	23.7	166	165	96.3	90.6	29.5	29.0	5	7	1.9	2.4
49	Kisegese	95	96	7	6	7	6	106.9	102.7	19.5	23.2	181	183	93.0	89.1	36.5	34.5	5	7	1.3	2.4
50	Turiani	88	89	8	7	8	6	94.4	93.2	20.9	21.0	145	157	96.2	85.5	32.5	34.5	5	5	2.6	3.1
51	Sindano nyeupe	97	98	7	8	7	7	127.7	136.2	22.2	23.0	160	169	93.8	90.7	26.5	27.0	5	7	2.1	2.7
52	Kihogo red	95	96	6	6	6	6	124.0	114.5	20.5	22.4	164	174	93.8	89.6	32.0	35.0	7	5	2.3	2.0
53	Cherehani	57	56	7	8	7	7	93.8	109.3	21.1	24.3	106	105	91.0	87.3	29.0	33.5	3	5	2.2	2.8
54	ITA 303	85	81	8	9	8	7	131.3	126.1	21.0	22.8	150	147	96.2	86.4	33.0	27.0	5	5	2.3	2.6
Mean		84.0	85.0	7.6	7.3	7.1	6.6	110.0	107.4	20.3	22.3	140.6	146.0	95.0	89.6	29.5	30.9	4.9	4.1	2.3	2.6
CV (%)		1.42	1.43	13.31	17.05	14.24	17.4	6.99	1.8	4.26	6.03	1.68	5.66	1.73	2.92	4.13	5.87	16.71	16.39	1.35	11.34
LSD (5%)		2.38	2.86	1.99	2.43	2.04	2.28	15.48	3.85	1.74	2.71	4.74	16.62	3.32	5.27	2.46	3.65	0.60	0.55	0.04	0.60

DFL= Days to 50% flowering; NT= number of tillers/plant; NPP=number of panicles/plant; PH= plant height; PL= panicle length; NGP = number of grains/panicle; PFG = percentage filled grains/panicle; TGW = 1000 grain weight; RYMVD = rice yellow mottle virus disease reaction; GY = grain yield; t ha^{-1}= tons per hectare; LSD= least significance difference; CV = coefficient of variation; Ifa = Ifakara and Mk = Mkindo.

3.3.3 Correlations among agronomic traits and RYMVD reaction

The magnitude of trait correlations revealed variable pairwise associations within and between sites (Tables 3.4 and 3.5). GY exhibited moderate and positive correlations with NPP (0.29≤ r ≤0.44, p≤0.05), PL (0.28≤ r ≤0.34, p≤0.05), NGP (0.28 ≤ r ≤ 0.54, p≤0.05), PFG (0.34 ≤ r ≤ 0.38, p≤0.05) and TGW (r= 0.43≤ r ≤ 0.48, p≤0.05) within and across sites. The associations between GY and RYMVD were also moderate but negative (-0.33 ≤ r ≤ -0.40, p≤0.05) within and between sites. There were also variable and significant associations among the secondary traits. For instance, TGW had significant and positive associations with PH (0.28 ≤ r ≤ 0.36, p≤0.05), NGP (0.29 ≤ r ≤ 0.48, p≤0.05) and PFG (0.31 ≤ r ≤ 0.41, p≤0.05) at the two sites. RYMVD exhibited negative correlations with most traits and significantly correlated to DFL (r=-0.27, p≤0.05) at Ifakara and NGP (r=-0.34, p≤0.05) across the two sites.

Table 3.4. Pearson correlation coefficients of phenotypic traits and RYMV reaction of 54 rice genotypes screened at Ifakara (upper diagonal) and Mkindo (lower diagonal) sites in Tanzania

Traits	DFL	NT	RYMVD	NPP	PH	PL	NGP	PFG	TGW	GY
DFL	1	-0.01	-0.27*	0.05	-0.02	0.22	0.41**	0.13	-0.07	0.13
NT	0.19	1	-0.05	0.83***	-0.26	0.03	0.39**	0.25	0.04	0.12
RYMVD	-0.29	0.24	1	0.04	0.05	0.23	-0.24	-0.14	0.01	-0.40**
NPP	0.20	0.85***	0.21	1	-0.19	0.16	-0.32*	0.18	-0.10	0.44**
PH	0.03	-0.21	0.21	-.0.27	1	0.07	0.29*	0.22	0.36*	0.05
PL	0.23	0.29*	-0.26	0.31*	0.09	1	-0.06	-0.15	0.07	0.34*
NGP	0.47**	0.31*	-0.42	-029*	0.33*	-0.02	1	-0.16	0.29*	0.28*
PFG	-0.23	-0.14	-0.27	-0.15	0.40**	-0.22	-0.17	1	0.31*	0.36*
TGW	-0.09	-0.09	-0.14	-0.3	0.28*	0.06	0.46**	0.41**	1	0.48**
GY	0.12	0.25	-0.33*	0.29*	0.01	0.28*	0.54***	0.34*	0.43**	1

DFL= Days to 50% flowering; NT= number of tillers/plant; NPP=number of panicles/plant; PH = plant height; PL= panicle length; NGP = number of grains/panicle; PFG = percentage filled grains/panicle; TGW = 1000 grain weight; RYMVD = rice yellow mottle virus disease reaction; GY = grain yield.

Table 3.5. Pearson correlation coefficients of phenotypic traits and RYMVD reaction of 54 rice genotypes evaluated acros two sites

Traits	DFL	NT	RYMVD	NPP	PH	PL	NGP	PFG	TGW	GY
DFL	1	-0.01	-0.27	0.05	-0.02	0.22	0.43**	0.12	-0.07	0.13
NT		1	-0.05	0.83***	-0.31*	0.03	0.36*	0.25	0.04	0.12
RYMVD			1	0.04	0.05	0.23	-0.34*	-0.14	-0.01	-0.37*
NPP				1	-0.29	0.16	-0.32*	0.28*	-0.10	0.32*
PH					1	0.07	0.33*	0.22	0.34*	0.05
PL						1	-0.06	-0.15	0.07	0.33*
NGP							1	-0.16	0.32*	0.45**
PFG								1	0.37*	0.38*
TGW									1	0.47**
GY										1

DFL= Days to 50% flowering; NT= number of tillers/plant; NPP = number of panicles/plant; PH= plant height; PL= panicle length; NGP = number of grains/panicle; PFG = percentage filled grains/panicle; TGW = 1000 grain weight; RYMVD = rice yellow mottle virus disease reaction; GY = grain yield.

3.3.4 Principal component analysis (PCA)

The rotated component matrix revealed the proportion of total variance described by each principal component (PC) and their correlations with the traits (Table 3.6). The first five and four PCs with Eigenvalues greater than 1 explained 78.32% of the genotype variation at the Ifakara and Mkindo sites, in that order. The first PC accounted for 23.44% and was positively associated with NT (with a loading score of 0.87) and NPP (0.82), while NGP (-0.71) had a negative contribution. The traits with major contribution on PC2 were DFL (0.71), RYMVD (0.62) and PL (0.58).The key traits allocated on PC3 were TGW and GY. The variation on the fourth and fifth PCs was contributed by DFL, PFG, PH and PL. At the Mkindo site, the first four PCs accounted for 70.78% of the total variation. PC1 accounted for 27.40% of the variation, which was mostly due to the positive contributions by NPP (0.81), NT (0.77), and RYMVD (0.54), whereas PFG (-0.61) was negative contributor. In comparison, trait variation linked with PC2 was accounted for by differences in NGP (0.82), RYMVD (0.53), DFL (0.52) and PH (0.50). The variation on PC3 was largely due to TGW (0.75) and GY (0.60), while the PC4 was negatively correlated with GY (-0.52). The combined results showed that 79.88% of the total variation across sites was elucidated by the first five PCs. The PC1, PC2, PC3, PC4, and PC5 accounted for 24.30%, 20.15%, 14.16%, 11.19%, and 10.08% of the variation, respectively. The PC1 was mostly correlated with NPP, NT, PL, GY, and DFL. Much of the variation on PC2 was contributed by NGP, RYMVD, and DFL. The traits most strongly correlated with PC3 were TGW, GY, and PH. The fourth PC accounted for much of the variation in PFG, PH, and DFL, while PC5 was correlated to PL, NGP, and GY.

Table 3.6. Rotated component matrix of phenotypic traits and RYMVD reaction on 54 rice genotypes evaluated at Ifakara and Mkindo sites, and across sites

Trait	Ifakara					Mkindo				Across locations				
	PC1	PC2	PC3	PC4	PC5	PC1	PC2	PC3	PC4	PC1	PC2	PC3	PC4	PC5
Eigen-values	2.34	1.81	1.49	1.18	1.01	2.74	1.82	1.43	1.09	2.43	2.02	1.42	1.12	1.01
Proportion variance (%)	23.44	18.06	14.89	11.84	10.09	27.4	18.2	14.33	10.86	24.3	20.15	14.16	11.19	10.08
Cumulative variance (%)	23.44	41.5	56.39	68.22	78.32	27.4	45.5	59.92	70.78	24.3	44.45	58.61	69.8	79.88
DFL	-0.17	**0.71**	-0.2	**0.41**	-0.17	**0.49**	**0.52**	0.01	-0.04	**0.32**	**0.62**	-0.08	**0.36**	0.26
NT	**0.87**	0.26	0.21	0.14	-0.13	**0.77**	-0.4	0.21	0.17	**0.86**	-0.3	0.17	0.17	0.13
RYMVD	-0.24	**0.62**	-0.38	0.02	0.1	**0.54**	**0.53**	-0.14	0.16	0.29	**0.66**	-0.26	0.07	-0.14
NPP	**0.82**	**0.38**	0.12	0.2	0.02	**0.81**	-0.4	0.12	0.19	**0.88**	-0.23	0.08	0.27	0.08
PH	-0.44	-0.21	**0.41**	**0.56**	**0.32**	-0.37	**0.5**	**0.43**	**0.49**	-0.51	0.24	**0.43**	**0.52**	-0.18
PL	0.04	**0.53**	0.24	-0.09	**0.74**	**0.41**	0.03	**0.41**	**0.47**	**0.39**	0.25	0.3	0.14	**-0.76**
NGP	**-0.71**	**0.4**	-0.02	0.19	-0.3	0.25	**0.82**	0.06	-0.18	-0.12	**0.83**	0.01	0.08	**0.34**
PFG	0.28	-0.36	-0.22	**0.75**	-0.07	-0.61	-0.17	0.29	**0.38**	-0.34	-0.47	0.08	**0.68**	0.24
TGW	-0.24	-0.04	**0.77**	0.12	-0.01	-0.31	0.04	**0.75**	-0.3	-0.25	0.06	**0.79**	-0.15	0.05
GY	-0.02	0.31	**0.61**	-0.16	-0.47	**0.35**	-0.08	**0.6**	-0.52	**0.34**	0.15	**0.63**	-0.32	**0.32**

DFL= Days to 50% flowering; NT= number of tillers/plant; NPP =number of panicles/plant; PH= plant height; PL= panicle length; NGP = number of grains/panicle; PFG = percentage filled grains/panicle; TGW = 1000 grain weight; RYMVD = rice yellow mottle virus disease reaction; GY = grain yield; PC = principal component.

3.3.5 Principal component biplot analysis

Figures 3.1, 3.2, and 3.3 depict the associations among the various traits and genotypes with respective principal components within and across locations. The two PCs of the PCA biplot explained only 41.5% of the total variation at Ifakara (Figure 3.1). The biplot revealed strong and positive correlations among NPP, NT, RYMVD, DFL, PL, and GY. Furthermore, the biplot showed that there were pairwise correlations between TGW and PH, and NT and NPP. Genotypes E8 (Serena) was in close proximity to the vectors for DFL, PL, GY, and RYMVD. The vectors for TGW and PH were associated with genotype E43 (Sumbawanga), while genotypes E38 (IR64) and E19 (IR72) were in close proximity with the NPP and NT vectors. The PFG vectors correlated with genotypes E13 (Mwangaza) and E2 (SSD1), though they exhibited a negative association with the vector for GY.

The biplot dimension vectors at the Mkindo site explained 45.59 % of the variation (Figure 3.2). The biplot showed positive correlations between NT and NPP, DFL and RYMV, PL, and GY. The vectors for PH and NGP; TGW, and PFG were also close, suggesting their positive correlation. Genotype E19 (IR72) was plotted next to the vectors for NT, and NPP, indicating higher values for these traits than most other genotypes. The vectors for DFL and RYMVD were associated with genotypes E49 (Kisegese) and E51 (Sindano nyeupe), although these were not vertex genotypes. For PL, and GY, the associated genotype was E32 (Moshi) and E27 (Mwanza). Also, E34 (Kalundi) and E40 (Afaa Mwanza) were correlated with NGP, though these genotypes were not on the polygon vertices. The traits TGW and PFG were associated with genotypes E5 (IRAT 256) and E9 (Nerica 4).

The PCA biplot based on combined data showed that 44.45% of the variation could be explained by PC1 and PC2 (Figure 3.3). There were positive associations among NGP, DFL, and RYMVD. Similarly, there were positive pairwise associations between PL and GY, NPP and NT, and PH and TGW, while PFG was not positively correlated with any particular trait. The vertex genotypes included genotype E8 (Serena), which was associated with NGP, DFL, RYMVD, PL and GY, genotype E19 (IR72) that correlated to NPP and NT and genotype E43 (Sumbawanga) with correlation to PH and TGW. The last vertex genotype was E10 (WAB450), which had a correlation with PFG. Vertex genotypes attained higher values for the associated traits.

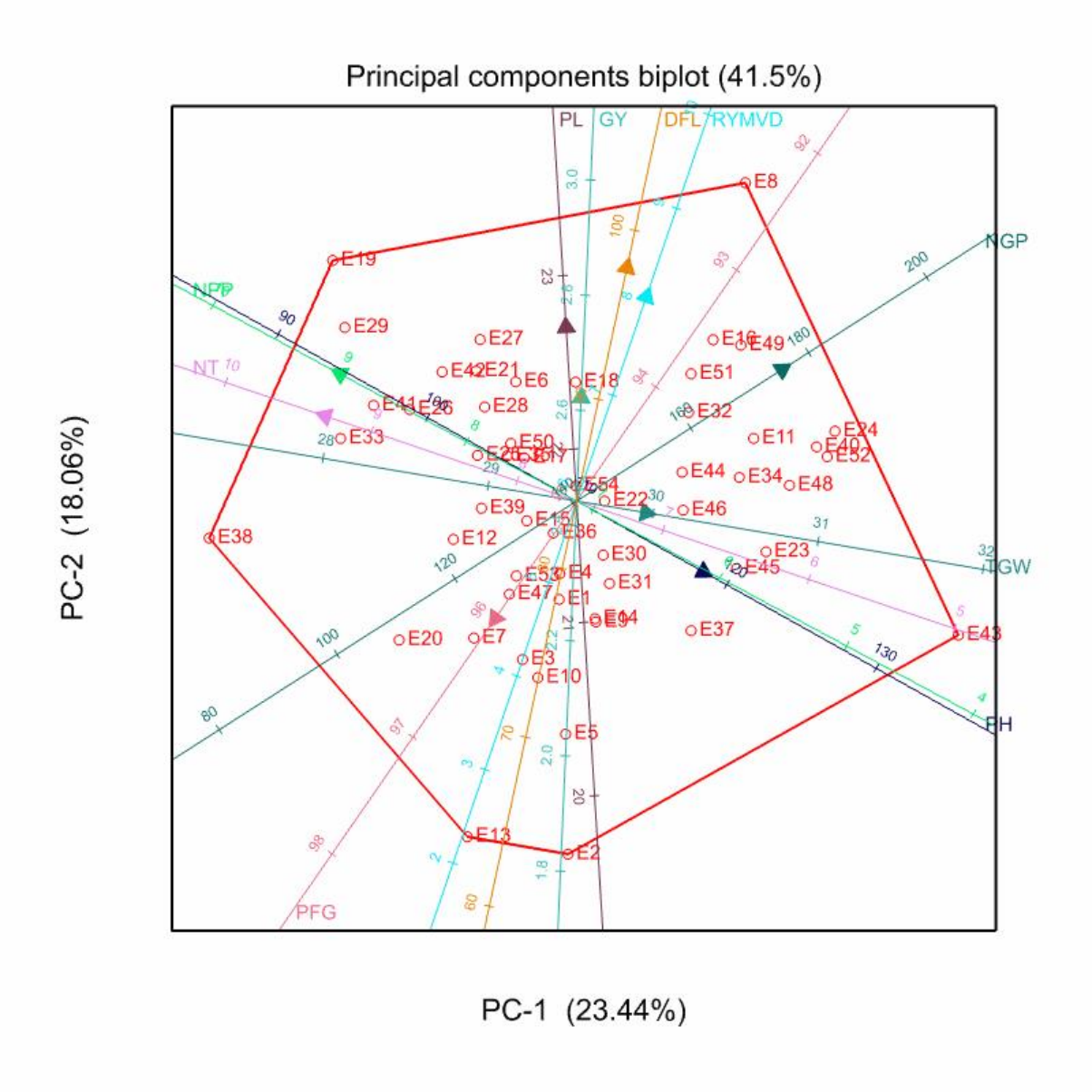

Figure 3.1. Genotype-trait biplot showing the relationship of agronomic traits in 54 rice genotypes evaluated at the Ifakara site

Notes: DFL= Days to 50% flowering; NT= number of tillers/plant; NPP=number of panicles/plant; PH= plant height; PL= panicle length; NGP = number of grains/panicle; PFG = percentage filled grains/panicle; TGW = 1000 grain weight; RYMVD = rice yellow mottle virus disease reaction; GY = grain yield; E = entry number designated on Table 3.3.

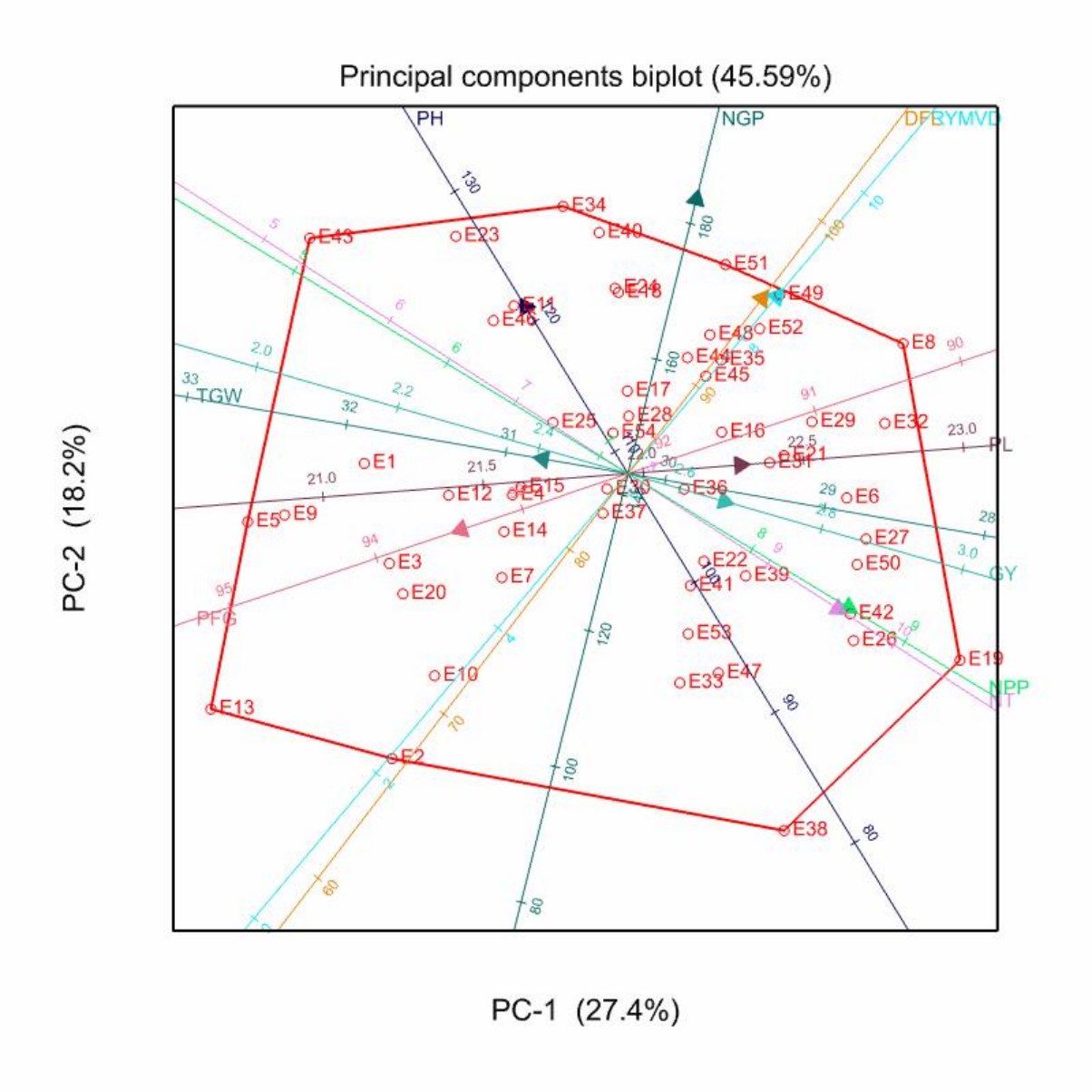

Figure 3.2. Genotype-trait biplot showing the relationship of various traits in 54 rice genotypes evaluated at the Mkindo site

DFL= Days to 50% flowering; NT= number of tillers/plant; NPP = number of panicles/plant; PH= plant height; PL= panicle length; NGP = number of grains/panicle; PFG = percentage filled grains/panicle; TGW = 1000 grain weight; RYMVD = rice yellow mottle virus disease reaction; GY = grain yield. E = entry number designated on Table 3.3.

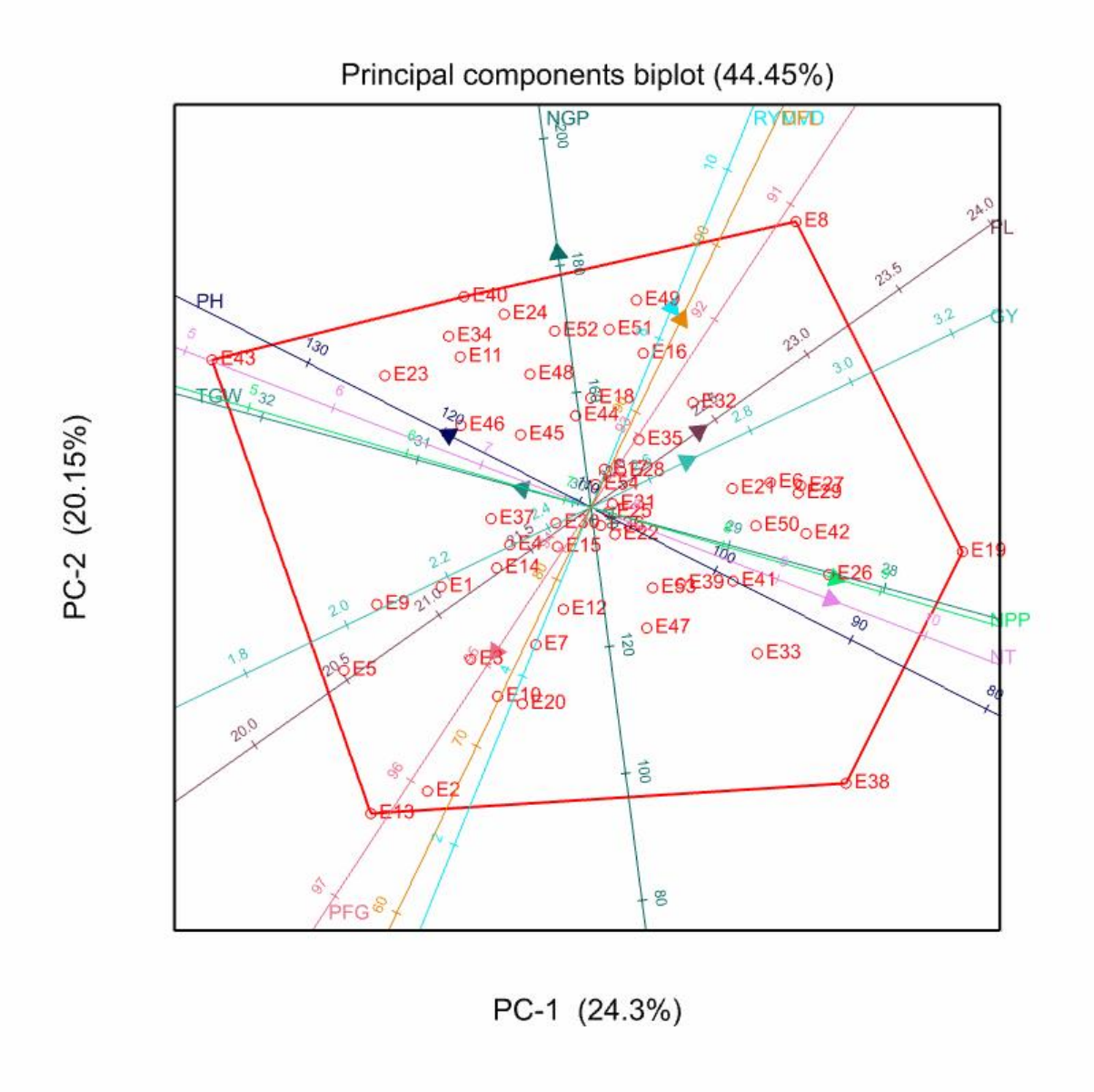

Figure 3.3. Genotype-trait biplot showing the relationship of various traits in 54 rice genotypes evaluated across two locations.

DFL= Days to 50% flowering; NT= number of tillers/plant; NPP=number of panicles/plant; PH= plant height; PL= panicle length; NGP = number of grains/panicle; PFG = percentage filled grains/panicle; TGW = 1000 grain weight; RYMVD = rice yellow mottle virus disease reaction; GY = grain yield; E = entry number designated on Table 3.3.

3.4 Discussion

3.4.1 Genotypic variation and mean performance

The study assessed variation present among 54 rice genotypes grown in Tanzania using agronomic traits and RYMV parameter to identify suitable parental lines for RYMV resistance breeding. The test genotypes exhibited significant variation for yield and yield components and RYMV resistance (Table 3.2). This suggests that the genotypes harbour adequate genetic

variation for improving agronomic performance and RYMV resistance in rice. The variation among genotypes emanates from differences in their genetic constitution and the environment (Adhikari et al., 2018; Gyawali et al., 2018). Genetic variability among rice genotypes for yield and yield-related traits was also reported by Summanth et al. (2017) and Bandi et al. (2018) in India.

The rice genotypes used in this study were collected from different sources where they were developed with different pedigrees and breeding objectives, which gave rise to significant variation. For instance, the NERICA genotypes were specifically developed for upland and drier ecologies and are derivatives of *O. glaberrima* and *O. sativa* interspecific-crosses. Other genotypes such as Supa, SARO 5, Gigante, TAI, Salama M-19, Salama M-55, Lunyuki, and Salama M-57 are Asian genotypes developed for lowland and wet ecologies. The genetic differences conferred variable performances and adaptation in genotypes of diverse genetic backgrounds. The landraces such as Rangimbili nyekundu and Mbega performed well (Table 3.3), probably due to their adaptation to the growing conditions in Tanzania. Adaptation among landraces reflects successful adaptation due to selection pressure applied by farmers, and to suitable climatic factors (Mercer and Perales, 2010).

The tested rice genotypes had significant genotype x site interactions (Table 3.2), showing differential performances over the two test locations. The results are in agreement with reports by Sandhu et al. (2019), who found that the test environment was influential in genotype performance. Genotype × environment interaction effects become significant when genotype performance is not consistent over different locations. The observed phenotypic expression that is quantified during germplasm evaluation is partially conditioned by genetic and environmental factors that influence trait expression. The differential response over locations can provide opportunities to identify genotypes with stable and broad adaptation to different ecologies. The genotypes exhibited significant variation and differential RYMV scores in different sites, which provides an opportunity to identify the genotypes with the most stable RYMV resistance and to identify the best site for RYMV screening. According to Joseph et al. (2011) and Hebert et al. (2017b), RYMV reactions depend on the test environment. Genotype × environment effects confound selection efforts by masking genotypic potential due to significant interaction with environments. Significant genotype × environment interaction effects can reduce the correlation between genotype and phenotypic expression, limiting selection response during breeding or cultivar recommendation (Bustos-Korts et al., 2018).

Genotypes such as Salama M-55, IRAT 256, Lunyuki, Salama M-19, Salama M-57, SSD1, and Mwangaza had low values for RYMV scores (Table 3.3) and are potential sources of new RYMV

resistance genes, and were therefore selected for subsequent breeding activities. However, breeding for high performance in stress-prone environments and in diverse rice-producing ecologies must target selection for multiple traits to increase adaptability to the erratic and harsh growing conditions. It is imperative to consider other agronomic traits to complement RYMV resistance. The genotypes with RYMV resistance did not exhibit a comparative advantage in agronomic performance or grain yield probably due to poor yield potential. Such genotypes must be crossed with high potential and complementary genotypes with suitable genetic backgrounds. Genotypes such as Salama M-55, IRAT 256, Lunyuki, Salama M-19. Salama M-57 and Mwangaza can provide new genes for RYMV resistance while Gigante, Rangimbili nyekundu, Zambia, and SARO 5 can provide suitable agronomic traits such as high grain yield.

3.4.2 Traits associations

Grain yield is a complex trait that is influenced by several inter-dependent secondary traits. Understanding the relationships among the secondary traits and grain yield is vital to devise appropriate selection strategies. Due to environmental variance that reduces selection efficiency (Romagosa and Fox, 1993), direct selection for grain yield may not be effective. Thus, knowledge of its relationship with secondary traits is important to guide indirect selection. The variable correlations exhibited by secondary traits with grain yield present both opportunities and challenges for indirect selection. Selection for traits such as the number of panicles per plant, panicle length, number of grains per plant, percentage filled grains, and thousand-grain weight that exhibited positive correlations with grain yield will simultaneously improve grain yield potential. The positive relationship between these traits and grain yield was previously reported in other studies. For instance, Bhuvaneswari et al. (2015) and Getachew and Burhan (2017) found that grain yield was positively correlated with productive tillers per plant, a number of grains per panicle, and 1000 grains weight in rice in their independent studies. However, indirect selection becomes complicated when at least one of the traits positively linked to other secondary traits exhibiting unfavourable associations with grain yield. There were un-favourable negative associations among the number of grains and the number of panicles per plant, and RYMVD resistance, with the number of grains per panicle, which would complicate indirect selection for grain yield. The selection of the number of panicles and grains per plant would indirectly increase grain yield but reduce negatively associated traits such as grains per panicle. Such un-favourable correlations have been identified in some traits due to linkage drag. Li et al. (2018) found that grain number per panicle and panicle number had a negative association that compromised grain yield. They subsequently conducted a genetic association study that revealed linkage drag among desirable traits in rice. Similarly, linkage drag attributed to a negative correlation between root capability and tillering capacity was found to limit breeding progress for drought-tolerant rice (Luo et al., 2015). Negative correlations caused by genetic

linkage drag would be difficult to break unless alternative breeding techniques are used such as mutation breeding. A significant negative correlation between RYMVD and grain yield indicates that the RYMV is the main cause of yield losses, as reported by others (Hubert et al., 2017b). Moreover, RYMV disease causes spikelet sterility and reduced grain weight, both leading to yield losses (Onwughalu et al., 2011).

Assessing genetic variability using principal component analysis allows the breeder to quantify the relative importance of each trait in discriminating a set of genotypes. The high proportion of variation accounted by the first two PCs in this study (Table 3.6) shows that traits that are associated with these PCs will explain much of the variation in the test genotypes and offer an opportunity to select for the best genotypes. The high and positive loadings by NT, NPP, PL, and GY on PC1 and PC2 at the Ifakara and Mkindo sites indicate that these traits exhibit wide variation that enabled for discrimination between the test genotypes. These traits can be simultaneously selected for rice improvement. The findings of this study are corroborated with those reported by Sahu et al. (2017), Yugandha et al. (2018), and Ranjith et al. (2019). Similarly, Nachimuthu et al. (2014) found that the number of grains per panicle, plant height, and days to 50% flowering contributed the most to the total variation in rice. In addition, Gana et al. (2013) reported that NPP contributed highly to the total variation in rice evaluated in rain-fed lowland ecologies. Therefore, the selection of these traits should achieve rapid improvement of grain yield.

The genotype-trait biplot depicts relationships between genotypes and traits, which assists in the selection of genotypes with multiple desirable traits. This is unlike univariate analysis methods that can only compare one trait at a time (Flores et al., 1998). Genotypes IR72, Rangimbili nyekundu, and TXD 307 were positively associated with traits NT and NPP. On the other hand, genotype Moshi associated most with PL and GY, while genotypes Serena and Afaa mwanza were highly correlated with DFL and NGP. These correlations indicated that the genotypes performed well for these traits. Conversely, genotypes Nerica 4, IRAT 256, and IR64 were not associated with a specific trait vector, showing that they performed below average for most traits. The close association of genotypes Kisegese and Sindano nyeupe with the RYMV vector and their plotting in the direction of the RYMV vector indicates that they had high RYMV scores that are linked to susceptibility. The depiction in the biplots corroborated with the analysis of variance, which showed that there was significant variation, and the genotypic means, which identified the genotypes with superior performance for particular traits. Genotypes SARO, Rangimbili and Gigante were selected for grain yield. On the other hand, genotypes Salama M-57, SSD1, IRAT 256, Salama M-55, Mwangaza, Lunyuki, and Salama M-19 were identified as possible sources of RYMV resistance genes due to their consistently low RYMVD scores.

3.5 Conclusion

The study evaluated a diverse rice collection at two locations in Tanzania where RYMV is prevalent in rice crops. It provided a basis to select the best genotypes and to understand genotype and environmental influences on agronomic performances and RYMV reactions. Significant variation was detected among the assessed genotypes for selection for grain yield and RYMV resistance improvement in Tanzania. The PCA identified number of tillers, number of panicle per plant, panicle length and grain yield as the most important traits for discriminating between the test genotypes.

Genotypes Salama M-57, SSD1, IRAT 256, Salama M-55, Mwangaza, Lunyuki, and Salama M-19 were selected as new sources of RYMV resistance genes under Tanzania condition. Genotypes such as Rangimbili, Zambia, SARO, and Gigante were selected with desirable agronomic traits, high yield potential, and RYMV resistance. Further studies to assess grain quality will be required to incorporate market-preferred traits, while combining ability tests will identify breeding populations with good combining ability effects for RYMV resistance and high grain yield potential.

References

Abo, M.E., M.D. Alegbejo, A.A. Sy, and S.M. Misari. 2000. An overview of the mode of transmission, host plants and methods of detection of rice yellow mottle virus. Journal of Sustainable Agriculture 17 (1):19-36.

Adhikari, B.N., B.P. Joshi, J. Shrestha, and N.R. Bhatta. 2018. Genetic variability, heritability, genetic advance and correlation among yield and yield components of rice (*Oryza sativa* L.). Journal of Agriculture and Natural Resources 1(1): 149-160.

Aida, A., M. Huseain, A.S. Mohamad, S. Hani, and M. Sameer. 2007. Assessment of genetic variation among Jordanian barely landraces (*Hordeum vulgare*) as revealed by molecular markers. American-Eurasian Journal of Agricultural and Environmental Science 2: 68-74.

Bakari, M.M. 2010. Genetic diversity of some rice (*Oryza sativa* L.) landraces grown in Tanzania using simple sequence repeats (SSR). MSc thesis, Sokoine University of Agriculture, Morogoro, Tanzania, 72 p.

Bakker, W. 1974. Characterization and ecological aspects of Rice yellow mottle virus in Kenya. PhD thesis, Wageningen Agricultural University, the Netherlands, 128 p.

Balasubramanian, V., M. Sie, R.J. Hijmans, and K. Otsuka. 2007. Increasing rice production in sub-Saharan Africa: Challenges and opportunities. Advanced Agronomy 94: 55-133.

Bandi, H.R.K., P.V. Satyanarayana, D.R. Babu, N. Chamundeswari, V.S. Rao, and S.K. Raju. 2018. Genetic variability estimates for yield and yield components traits and quality traits in rice (*Oryza sativa* L.). International Journal of Current Microbiology and Applied Sciences 7(5): 551-559. doi:10.20546/ijcmas.2018.705.069.

Bhuvaneswari, S., S. Kumar, I.M. Singh, S. Takhellambam, K.S. Shasidhar, R.Y. Singh, and N. Prakash. 2015. Genetic variability and association studies on grain yield components in F2 populations of black rice (*Oryza sativa* L.) of Manipur. *Indian* Journal of Hill Farming 28: 85-89.

Bustos-Korts, D., I. Romagosa, G. Borràs-Gelonch, A.M. Casas, G.A. Slafer, and F.A. Van Eeuwijk. 2018. Genotype by Environment Interaction and Adaptation. Encyclopaedia of Sustainability Science and Technology. Springer Science and Business Media, LLC: 29-71.

Fauquet, M.C., and M.A. Mayo. 1999. Abbreviations for plant virus name. *Archives of Virology* 144 (6): 1249-1273.

Flores, F., M.T. Moreno, and J.I. Cubbero. 1998. A comparison of univariate and multivariate methods to analyze G x E interaction. *Field Crop Research* 56: 271-286.

Food and Agriculture Organization. 2015. The Rice Value Chain in Tanzania. A report from the Southern Highlands food systems programme. (Wilson, R.T. and I. Lewis, Eds). FAO, Tanzania, 9 p.

Gana, A.S., S.Z. Shaba, and E.K. Tsado. 2013. Principal component analysis of morphological traits in thirty-nine accessions of rice (*Oryza sativa* L.) grown in a rainfed lowland ecology of Nigeria. Journal of Plant Breeding and Crop Science 5: 120-126.

Getachew, M., and T. Burhan. 2017. Growth and yield of rice (*Oryza sativa* L.) as affected by time and ratio of nitrogen application at Jimma, South-West Ethiopia. International Journal of Agriculture Innovations and Research 4(1): 175-182.

Gyawali, S., A. Poudel, and S. Poudel. 2018. Genetic variability and association analysis in different rice genotypes in mid hill of western Nepal. Acta Scientific Agriculture 2(9): 69-76.

Hubert, J.G., A. Pinel-Galzi, D. Dibwe, E. Cinyabuguma, A. Kabore, D. Fargette, D. Silue, E. Herbrard, and Y. Sere. 2013. First report of rice yellow mottle virus on rice in the Democratic Republic of Congo. Plant Disease 97(12):1664. doi: 10.1094PDIS-06-13-0650PDN.

Hubert, J., A. Luzi-Kihupi, E. Hébrard, and H.J.F. Lyimo. 2016. Farmers' knowledge and perceptions of Rice yellow mottle virus in selected rice growing areas in Tanzania. International Journal of Science and Research 5 (2): 549- 559.

Hubert, J., H.J.F. Lyimo, and A. Luzi-Kihupi. 2017a. Geographical variation, distribution and diversity of rice yellow mottle virus phylotypes in Tanzania. American Journal of Plant Sciences 8(6): 1264-1284.

Hubert, J., H.J.F. Lyimo, and A. Luzi-Kihupi. 2017b. Pathogenic variation and occurrence of multiple resistance-breaking rice yellow mottle virus strains in Tanzania. African Journal of Biotechnology (16) 24: 1331-1337.

International Rice Research Institute (Eds.) .2002. Standard Evaluation System for Rice, 5th edition. Genetic Resources Center, IRRI, Manila, Philippines, 27 p.

Joseph, A., D.B. Olufolaji, F.E. Mwilene, A. Onasanya, M.M. Omale, R.O. Onasanya, and Y. Sere. 2011. Effect of leaf age on rice yellow mottle virus severity chlorophyll content with mechanical inoculation and vector transmission method. Trends Applied Sciences Research 6(12): 1345 -1351.

Kam, H., M.D. Laing, Y. Sere, D. Thiemele, A. Ghesquiere, N. Ahmadi, and M.N. Ndjiondjop. 2013. Evaluation of a collection of rice landraces from Burkina Faso for resistance or tolerance to rice yellow mottle virus. Journal of Plant Pathology 95 (3): 485-492.

Kanyeka, Z.L., E. Sangu, D. Fargette, A. Pinel-Galzi, and E. Hébrard. 2007. Distribution and diversity of local strains of Rice yellow mottle virus in Tanzania. African Journal of Crop Science 15(4): 201-209.

Kilimo-Trust. 2012. Expanding Rice Markets in the EAC: An Opportunity for Actors in the Value Chain. Kilimo Trust, Dar es Salaam, Tanzania, 45p.

Kouassi, N.K., P. N'guessan, L. Albar, C. Fauquet, and A. Ghesquiere. 2005. Distribution and characterization of rice yellow mottle virus: A threat to African farmers. Plant Disease 89: 124-133.

Koudamilor, A., F.E. Nwilene, D. Silue, A. Togola, O. Oyetunji, Y. Sere, and M. Akogbeto. 2014. Identification insect vectors of rice yellow mottle virus (RYMV) in Benin. Journal of Entomology 11: 153-162. doi:10.1155/2015/721751.

Lamo, L., G. Cho, I. Jane, P.K.A. Dartey, E. James, M. Ekobu, S. Alibu, S. Okanya, B. Oloka, M. Otim, G. Asea, and K. Kang. 2015. Developing lowland rice germplasm with resistance to multiple biotic stresses through anther culture in Uganda. The Korean Society Journal of International Agriculture 27: 415-420.

Li, F., J. Xie, X. Zhu, X. Wang, Y. Zhao, X. Ma, Z. Zhang, M.A.R. Rashid, Z. Zhang, L. Zhi, S. Zhang, J. Li, Z. Li, and H. Zhang. 2018. Genetic basis underlying correlations among growth duration and yield traits revealed by GWAS in rice (*Oryza sativa* L.). Frontiers in Plant Science 9: 650. doi: 10.3389/fpls.2018.00650.

Longué, D.R.S., A. Galzi-Pinel, S. Semballa, I. Zinga, D. Fargette, and O. Traoré. 2014. First report of Rice yellow mottle virus in rice in the Central African Republic. Plant Disease 98 (1): 162-162.

Longué, D.R.S., A. Galzi-Pinel, I. Zinga, S. Semballa, D. Fargette, N. Baro, and O. Traoré. 2017. Genetic diversity and molecular epidemiology of rice yellow mottle virus in Central African Republic. International Journal of Virology 1-11. doi: 10.3923/ijv.2017.

Longué, R.D.S., I. Zinga, S. Semballa, N. Barro, and O. Traoré. 2016. Detection and serological characterization of rice yellow mottle virus in Central African Republic. Agricultural Sciences 7: 911-919.

Lou, Q., L. Chen, H. Mei, H. Wei, F. Feng, P. Wang, H. Xia, T. Li, and L. Luo. 2015. Quantitative trait locus mapping of deep rooting by linkage and association analysis in rice. Journal of Experimental Botany 66(15): 4749-4757. doi: 10.1093/jxb/erv246.

Luzi-Kihupi, A., J.A. Zakayo, H. Tusekelege, M. Mkuya, N.J.M. Kibanda, K.J. Khatib, and A. Maerere. 2009. Mutation breeding for rice improvement in Tanzania. In: Shu, G.Y. (Ed.). Induced Plant Mutations in the Genomics Era.. FAO, Rome, pp. 385-387.

Mangosongo, H.M., H.V. Lyaruu, and E.E. Mneney. 2019. Agro-morphological characterization of the wild rice (*Oryza longistaminata*) populations from selected areas of Tanzania. Tanzania Journal of Science 45(1): 9-17.

Mausa, E.A. 2014. Genetic diversity of rice (*Oryza sativa* L.) landraces conserved at the national genebank as revealed by simple sequence repeat (SSR) DNA markers. MSc thesis, Sokoine University of Agriculture, Morogoro, Tanzania, 83 p.

Mercer, K.L., and H.G. Perales. 2010. Evolutionary response of landraces to climate change in centres of crop diversity. Evolutionary Applications, 15 p. doi:10.1111/J.1752-4571.2010.00173.

Mghase, J.J., H. Shiwachi, K. Nakasone, and H. Takahashi. 2010. Agronomic and socio-economic constraints to high yield of upland rice in Tanzania. African Journal of Agriculture Research 5: 150-158.

Mpunami, A., I. Ndikumana, J. Hubert, A., Pinel-Galzi, N.J.M. Kibanda, F. Mwalyego, P. Tembo, B. Kola, M. Mkuya, Z. Kanyeka, R. Mutegi, S. N'chimbi-Msolla, P. Njau, Y. Séré, D. Fargette, and E. Hébrard. 2012. Tanzania, biodiversity hotspot of Rice yellow mottle

virus. In: Proceedings of the 12th International Plant Virus Epidemiology Symposium, 29 January -1 February 2013, Arusha, Tanzania, 70 p.

Munganyinka, E., R. Edema, J. Lamo, and P. Gibson. 2016. The reaction of intraspecific and interspecific rice cultivars for resistance to rice yellow mottle virus disease. European Journal of Experimental Biology 6: 13 -18.

Musyoki, M.A., W.F. Kioko, N.P. Mathew, A. Daniel, K.G. Muriira, N.D. Wavinya, M. Felix, L.R. Chemutai, N.S. Mwenda, M.J. Kiambi, and N.L. Ngithi. 2015. Genetic Diversity Studies on Selected Rice (*Oryza Sativa L.*) Genotypes Based on Amylose Content and Gelatinization Temperature. Advances in Crop Science and Technology 3:193. doi:10.4172/2329-8863.1000193.

Mwalyego, F.S., R.M., Maganga, A. Luzi- Kihupi, A.A. Ndunguru, and D.A. Kabungo. 2017. Evaluation and deployment of rice genotypes resistant to rice yellow mottle virus disease in the southern highlands of Tanzania. Huria Journal 2(24): 33-45.

N'Guessan, P., A. Pinel, M.L. Caruana, R. Frutos, A. Sy, A. Ghesquiere, and D. Fargette. 2000. Evidence of the presence of two serotypes of rice yellow mottle Sobemovirus in Côte d'Ivoire. European Journal of Plant Pathology 106: 167- 178.

Nachimuthu, V.V., S. Robin, D. Sudhakar, M. Raveendran, S. Rajeswari, and S. Manonmani. 2014. Evaluation of rice genetic diversity and variability in a population panel by principal component analysis. Indian Journal of Science and Technology 7(10): 1555-1562.

Ndjiondjop, M.N., L. Albar, D. Fargette, C. Fauquet, and A. Ghesquiere.1999. The genetic basis of high resistance to rice yellow mottle virus (RYMV) in cultivars of two cultivated rice species. Plant Disease 83: 931-935.

Nkuba, J., A. Ndunguru, R. Madulu, D. Lwezaura, G. Kajiru, A. Babu, B. Chalamila, and G. Ley. 2016. Rice value chain analysis in Tanzania: Identification of constraints, opportunities, and upgrading strategies. African Crop Science Journal 24: 73-87.

Ochola, D., S. Issaka, M. Rakotomalala, A. Pinel-Galzi, I. Ndikumana, J. Hubert, E. Hébrard, Y. Séré, G. Tusiime, and D. Fargette. 2015. Emergence of rice yellow mottle virus in eastern Uganda: Recent and singular interplay between strains in East Africa and in Madagascar. Journal of Virus Research 195: 64-72.

Onwughalu, J.T., M.E. Abo, J.K. Okoro, A. Onasanya, and Y. Sere. 2011. Rice yellow mottle virus infection and reproductive losses in rice (*Oryza sativa* Linn.). Trends in Applied Sciences Research 6(2): 182-189.

Payne, R.W., D.A. Murray, and S.A. Harding. 2017. An introduction to the GenStat Command Language, 18th edition. Hemel Hempstead, UK. VSN International.

Pidon, H., A. Ghesquière, S. Chéron, S. Issaka, E. Hébrard, F. Sabot, O. Kolade, D. Silué, and L. Albar. 2017. Fine mapping of RYMV3: A new resistance gene to rice yellow mottle virus from *Oryza glaberrima.* Theoretical and Applied Genetics 130: 807-818.

Pinel-Galzi, A., C. Dubreuil-Tranchant, E. Hébrard, C. Mariac, A. Ghesquière, and L. Albar. 2016. Mutations in rice yellow mottle virus polyprotein P2a involved in RYMV2 gene resistance breakdown. Frontier in Plant Science 7:1-11.

Ranjith, P., S. Sahu, S.K. Dash, D.N. Bastia, and B.D. Pradhan. 2019. Principal component analysis of rice varieties under bacterial leaf blight incidence. International Journal of Chemical Studies 7(3): 4184-4187.

Romagosa, I., and P.N. Fox.1993. Genotype × environment interaction and adaptation. In: M. D., Hayward, N.O. Bosemark, I. Romagosa, and M. Cerezo. (Eds). Plant Breeding. Plant Breeding Series, Springer, Dordrecht.

Rossel, H.W., Thottapilly, G. and I.W. Buddenhagen. 1982. Occurrence of rice yellow mottle virus in two important rice-growing areas of Nigeria, FAO Plant Protection Bulletin 30 (3/4): 137-139.

Sahu, H., R.R. Saxena, S.B. Verulkar, and V. Kumar. 2017. Principal component analysis of agro-morphological traits in RIL population of rice (*Oryza sativa* L.) grown in an irrigated lowland water regime. Green Farming 8(1): 11-15.

Sandhu, N., R.B. Yadaw, B. Chaudhary, H. Prasai, K. Iftekharuddaula, C. Venkateshwarlu, A. Annamalai, P. Xangsayasane, K.R. Battan, M. Ram, M.T.S. Cruz, P. Pablico, P.C. Maturan, K.A. Raman, M. Catolos, and A. Kumar. 2019. Evaluating the performance of rice genotypes for improving yield and adaptability under direct seeded aerobic cultivation conditions. Frontiers in Plant Science 10: 159. doi: 10.3389/fpls.2019. 00159.

Sereme, D., I. Ouedraogo, B.J. Neya, P.E. Zida, N. Yao, and M. Sie. 2016. Screening improved rice varieties (*Oryza* spp) for their resistance /tolerance to rice yellow mottle virus in West Africa. International Journal Agriculture Innovation Research 5: 481-486.

SPSS. 2017. Statistical Package for Social Scientists. SPSS for Windows Release 24.0, Chicago, IL.

Sumanth, V., B.G. Suresh, B.J. Ram, .0and G. Srujana. 2017. Estimation of genetic variability, heritability and genetic advance for grain yield components in rice (*Oryza sativa* L.). Journal of Pharmacognosy and Phytochemistry 6: 1437-1439.

Suvi, W.T., H. Shimelis, and M. Laing. 2018. Breeding rice for rice yellow mottle virus resistance in Sub-Saharan Africa: a review, *Acta Agriculturae Scandinavica, Section B-Soil & Plant Science* 69 (2): 1-8. doi:10.1080/09064710.2018.1523454.

Suvi, W.T., H. Shimelis, M. Laing, I. Mathew, and A.I.T. Shayanowako. 2019. Assessment of the genetic diversity and population structure of rice genotypes using SSR markers. *Acta Agriculturae Scandinavica, Section B - Soil & Plant Science* 70:76-86 doi:10.1080/09064710.2019.1670859.

Sy, A. A., and Y. Sere. 1996. Manuel de Formation en Pathologie du Riz. Le publication de l'Association pour le developpment de la riziculture en Afrique de l'Ouest (ADRAO), Bouake, Cote d'Ivoire.

Tao, J., and Y.O. Li. 2018. Edible insects as a means to address global malnutrition and food insecurity issues. Food Quality and Safety 2:17-26.

Thiémélé, D., A. Boisnard, M.N. Ndjiondjop, S. Chéron, Y. Séré, S. Aké, A. Ghesquière, and L. Albar. 2010. Identification of a second major resistance gene to *Rice yellow mottle virus*, *RYMV2*, in the African cultivated rice species, *O. glaberrima*. Journal of Theoretical and Applied Genetics 121: 169 -179.

Traoré, O., A. Pinel-Galzi, S. Issaka, N. Poulicard, J. Aribi, S. Aké, A. Ghesquiéré, Y. Séré, G. Konaté, E.Hébrard, and D. Fargette. 2010. The adaptation of *Rice yellow mottle virus* to the eIF(iso)4G-mediated rice resistance. Journal of Virology 408: 103 -108.

Traoré, V.S.E. B.J. Néya, M. Camara, V. Gracen, S.K. Offei, and O. Traoré. 2015. Farmers' perception and impact of rice yellow mottle disease on rice yields in Burkina Faso. Agricultural Sciences 6: 943-952.

Wilson, R.T. 2018. The rice value chain in Tanzania. International Journal of Agriculture Innovations and Research 7(2): 237-244.

Xiao, X.Y., Y.P. Wang, J.Y. Zhang, S.G. Li, and T.Z., Rong. 2016. SSR marker based genetic diversity fingerprinting of hybrid rice in Sichuan, China. China Journal of Rice Science 20:1-7.

Yugandhar P.R., S. Kota, B.U. Kiran, and Sridhar, M. 2018. Principal component analysis for agro-morphological and quality characters in germplasm of rice (*Oryza sativa* L.). International Journal of Advanced Biological Research 8 (2): 268-273.

Zhang, Q.J., T. Zhu, E.H. Xia, C. Shi, Y.L. Liu, Y. Zhang, Y. Liu, W.K. Jiang, Y.J. Zhao, and S.Y. Mao. 2014. Rapid diversification of five *Oryza* AA genomes associated with rice adaptation. Proceedings of the National Academy of Sciences of the United States of America 111(46):E4954-E462.

CHAPTER FOUR: ASSESSMENT OF THE GENETIC DIVERSITY AND POPULATION STRUCTURE OF RICE GENOTYPES USING SSR MARKERS

Abstract

Genetic diversity is a pre-requisite for rice (*Oryza sativa* L.) breeding and population development. Hence, the objective of this study was to assess the genetic diversity and population structure of 54 rice accessions using 14 polymorphic simple sequence repeat (SSR) markers to select unique parents for breeding. Data analysis was based on marker and population genetic parameters. The mean polymorphic information content (PIC) was 0.61 suggesting high polymorphism for the selected SSR markers among the rice accessions. The population structure revealed a narrow genetic base with only two major sub-populations. Analysis of molecular variance revealed that only 30% of the variation was attributed to population differences while 47% and 23% were due to variation among individuals within populations and within individual variation, respectively. The genetic distance and identity among genotypes varied from 0.083 to 1.834 and 0.159 to 0.921, respectively. A dendrogram grouped the genotypes into three clusters with wide variation among the accessions. The study established the existence of considerable genetic diversity among the tested 54 accessions. The selected genetic resources will be useful for rice breeding in Tanzania or other African countries.

Key words: Genetic diversity, Polymorphisms, Population structure, Rice, SSR markers, Tanzania

---

This chapter was published in the journal of Acta Agriculturae Scandinavica, Section B- Soil & Plant Science. Suvi, W.T., H. Shimelis, M. Laing, I. Mathew, and A.I.T. Shayanowako. 2019. Assessment of the genetic diversity and population structure of rice genotypes using SSR markers. Acta Agriculturae Scandinavica, Section B- Soil & Plant Science 70: 76–86. doi: 10.1080/09064710.2019.1670859.

4.1 Introduction

In sub-Sahara Africa (SSA), rice (*Oryza sativa* L.; 2× = 24) has become a pivotal crop in ensuring food security and in sustaining the livelihoods of millions of people. In Tanzania, rice is the second most important food and cash crop after maize (*Zea mays* L) (Bucheyeki et al., 2011). Tanzania is the second largest rice producer in East and Central Africa after Madagascar, with an annual production of 1.2 to 1.5 million tons (Nkuba et al., 2016; FAO, 2017). The majority of rice production in Tanzania is carried out by small-scale farmers using landrace varieties, which have low yield potential (Mogga et al., 2018). There is need to develop modern and improved varieties to serve the diverse needs of the rice value chains. There is an evident lack of adoption of improved rice cultivars because they lack the taste or aroma preferred by farmers and consumers (Mogga et al., 2018). Hence, most farmers opted to grow landraces, which have important attributes such as aroma and good cooking qualities that are absent in the introduced cultivars. There is an urgent need to develop cultivars that incorporate farmer and consumer-preferred traits.

Previous studies on rice focused on evaluations for agronomic performance and value for cultivation (Mligo and Msuya, 2015; Ansah et al., 2017) with less emphasis on breeding for improved yield and related traits. Progress in rice breeding is strongly related to the genetic variation within the germplasm resources (Yan et al., 2016). Therefore, understanding the population structure and genetic variation in germplasm is a prerequisite for crop genetic improvement (Xiao et al., 2016). Genetic diversity in rice has been investigated using morphological, biochemical and DNA markers (Palanga et al., 2016; Luther et al., 2017). However, both morphological and biochemical traits are highly influenced by environments, genotype × environment interaction effects, and may not provide accurate genetic classification of the crop (Aljumaili et al., 2018; Mulualem et al., 2018). Moreover, morphological traits cannot define the exact level of genetic diversity among germplasm, because of the presence of polygenic control on the expression of traits. Therefore, rice genetic resources should be effectively characterised using genomic tools for efficient utilisation and conservation.

A range of DNA techniques, including amplified fragment length polymorphism (AFLP) (Sorkheh et al., 2016), restriction fragment length polymorphism (RFLP) (Sun et al., 2000), random amplified polymorphic DNA (RAPD) (Ali et al., 2014), microsatellites (Liu et al., 2015; Chen et al., 2017), single nucleotide polymorphisms (SNP) (Sun et al., 2013) markers have been applied in rice genetic diversity studies. However, the choice of markers depends on the availability of genetic information on the genome sequence, cost of marker development, ease of documentation and level of polymorphism (Mittal and Dubey, 2009). The SSR markers are widely used because of their high degree of polymorphism, multi-allelic variation, codominance,

high reproducibility, and ease of detection by polymerase chain reaction (PCR), and relatively abundance with a uniform coverage. Moreover, SSR markers have remarkable potential to discriminate rice genotypes due to their high polymorphic nature and transferability (Islam et al., 2012; Mousavi et al., 2017). Further, SSRs markers that are linked to major genes could increase the efficiency of classical breeding by significantly reducing the number of selection generations required to identify superior and stable progenies. Recently, Yelome et al., (2018) used SSR markers to assess the extent of genetic divergence among *O. sativa* and West African rice *O. glaberrima* accessions. To develop breeding populations, a panel of 54 genetically diverse rice genotypes including landraces were collected from farmers' and different research organisations in Tanzania. Based on agro-morphological classification, these accessions were found to be phenotypically distinct. However, the extent of genetic diversity and genetic relationships present in these collections has not been rigorously studied using molecular markers. Knowledge of genetic diversity and relationships among the rice germplasm will play a significant role in local and regional breeding programmes. Therefore, the objective of the present study was to determine the genetic relationship and population structure present among 54 rice collections using SSR markers to identify genetically divergent genotypes for breeding.

4.2 Materials and methods

4.2.1 Plant materials

The study used 54 rice genotypes acquired from Tanzania Agricultural Research Institute (TARI), International Rice Research Institute (IRRI)/Philippines, Africa Rice/ Benin, Sokoine University of Agriculture (SUA)/Tanzania and farmer fields in Tanzania. The details of the germplasm are described in Chapter 3.

4.2.2 DNA extraction

Prior to DNA extraction, seeds of 54 rice genotypes were planted at University of KwaZulu-Natal (latitude 29° 37'51.75″ S; longitude 30°23'59.10″ E), South Africa. All genotypes were established under glasshouse conditions. Four seeds of each rice genotype were sown in a plastic pot, and from each pot, three healthy and vigorous plants were randomly selected and fresh young leaves collected for DNA extraction. The DNA was extracted following the Cetyl-tetramethyl ammonium bromide (CTAB) method. Approximately 200 mg of ground plant tissue combined with 500 µL of CTAB buffer, was incubated for one hour at 65°C, and subjected to centrifugation at 3500 rpm for 10 min. The supernatant was then transferred into new micro-tubes, and 400 µl chloroform: iso-amyl alcohol (24:1) was added into the tubes and mixed gently. After a second centrifugation (centrifuged at 3500 rpm for 30 min), the DNA was precipitated from the aqueous layer by addition of salt and ethanol. The upper aqueous phase containing

DNA was transferred to a clean microfuge tube. The resulting pellet was dried and re-suspended in Tris-EDTA (TE) buffer.

The PCR amplification reaction contained a total volume of 12 µL of PCR mix. The PCR mix contained 0.72 µL magnesium chloride (50 mM MgCl2), 1.2 µL dNTPs (25 µM), 0.12 µL Taq (5U/µL), 0.06 µL forward primer (10 µM), 0.3 µL reverse primer (10 µM), 1.2 µL of 1× reaction buffer, 6.16 µL PCR grade water and 0.24 µL dye. A PCR profile of initial denaturation for 2 min at 94°C, and 33 cycles of denaturation for 1 min at 55–60°C, an annealing temperature of 63°C for 2 min, and an extension for 2 min at 72°C was used. The PCR products (DNA samples) were fluorescently analysed using a Genetic Analyzer 3130xl labelled and separated by capillary electrophoresis on an ABI 3013 automatic sequencer. Analysis of the electropherograms was performed using Gene Mapper 4.0 and the marker data was presented as fragment sizes in an Excel spreadsheet.

4.2.3 Microsatellite analysis

Fourteen simple sequence repeats (SSRs) distributed on the 12 chromosomes of rice were used in this study and chosen based on their use in published rice diversity analysis reports (Chen et al., 1997; Ashfaqa and Khan, 2012; Ashraf et al., 2016). Forward and reverse primers of the SSR markers are presented in Table 4.1.

Table 4.1. Sequence of SSR markers used for rice genetic diversity analysis

Name	Sequence	AT	Reference
RM 206	F: CCCATGCGTTTAACTATTCT R: CGTTCCATCGATCCGTATGG	55	Ashfaqa and Khan (2012)
RM 235	F: AGAAGCTAGGGCTAACGAAC R: TCACCTGGTCAGCCTCTTTC	55	Ashfaqa and Khan (2012)
RM 202	F: CAGATTGGAGATGAAGTCCTCC R: CCAGCAAGCATGTCAATGTA	55	Pervaiz et al. (2010)
RM 215	F: CAAAATGGAGCAGCAAGAGC R: TGAGCACCTCCTTCTCTGTAG	55	Chen et al. (1997)
RM1261	F: GTCCATGCCCAAGACACAAC R: GTTACATCATGGGTGACCCC	55	Dixit et al. (2012)
RM319	F: ATCAAGGTACCTAGACCACCAC R: TCCTGGTGCAGCTATGTCTG	55	Wang et al. (2005)
RM11	F: TCTCCTCTTCCCCCGATC R: ATAGCGGGCGAGGCTTAG	55	Panaud et al. (1996)
RM19	F: CAAAAACAGAGCAGATGAC R: CTCAAGATGGACGCCAAGA	55	Panaud et al. (1996)
RM125	F: ATCAGCAGCCATGGCAGCGACC R: AGGGGATCATGTGCCGAAGGCC	55	Temnykh et al. (2000)

Table 4.1. continued

Name	Sequence	AT	Reference
RM161	F: GCAGATGAGAAGCGGCGCCTC R: TGTGTCATCAGACGGCGCTCCG	61	Temnykh et al. (2000)
RM 338	F: GCCAGCAAAACCAGGGATCCGG R: CAAGGTCTTGTGCGGCTTGCGG	61	Ashraf et al. (2016)
RM252	F: TTCGCTGACGTGATAGGTTG R: ATGACTTGATCCCGAGAACG	55	Lee et al. (2011)
RM421	F: AGCTCAGGTGAAACATCCAC R: ATCCAGAATCCATTGACCCC	55	Lee et al. (2011)
RM433	F: TGCGCTGAACTAAACACAGC R: AGACAAACCTGGCCATTCAC	53	Islam et al. (2008)

F = forward primers; R = reverse primers; AT = annealing temperature (°C)

4.2.4 Data analysis

Genetic diversity was assessed using GenAlex version 6.5 (Peakall and Smouse, 2007). The following parameters were computed: total number of alleles per locus (Na), number of effective alleles per locus (Ne), Shannon's information index (I), observed heterozygosity (Ho), gene diversity (He), and inbreeding coefficient (FIS) were determined using the protocol of Nei and Li (1979). The Polymorphic information content (PIC) values were calculated for each SSR locus as PIC = 1–Σ (pi2), where pi is the frequency of i^{th} allele. Analysis of molecular variance (AMOVA) was performed to test the degree of differentiation among and within the sources of collection of the rice genotypes. The population structure of the 54 rice accessions was established using the Bayesian clustering method in STRUCTURE version 2.3.4 (Pritchard et al., 2000). The length of the burn-in period and Markov Chain Monte Carlo (MCMC) were set at 10,000 iterations (Evanno et al., 2005). To obtain an accurate estimation of the number of populations, 20 runs were performed for each K-value (assumed number of subpopulations), ranging from 1 to 10. Further, Delta K values were calculated and the appropriate K value was estimated by implementing Evanno et al. (2005) method using STRUCTURE Harvester program (Earl and von Holdt, 2012). The genetic relationships or relatedness (cluster analysis) of the 54 genotypes were estimated using the genetic dissimilarity coefficients and the dendrogram were drawn using the unweighted pair group method (UPGMA) in DARwin 6.0 (Perrier and Jacquemoud- Collet, 2006).

4.3 Results

4.3.1 Genetic variability of 54 rice accessions based on SSR markers

The number of alleles scored per locus ranged from 2 for the markers RM319 and RM338, to 20 for marker RM206 with a mean of 7.43 per locus (Table 4.2). The number of effective alleles (Ne) per locus varied from 1.43 to 9.57 with a mean of 3.97 and markers RM319 and RM206 had the lowest and highest numbers of effective alleles, respectively. Expected heterozygosity

(He) ranged from 0.30 (M319) to 0.90 (RM206 and RM235) with a mean of 0.62 (Table 4.2). The observed heterozygosity (Ho) values had a mean of 0.18 and a range of 0.00 (RM319) to 0.80 (RM125 and RM235). The inbreeding coefficient (FIS) ranged from 0.10 to 0.93 with a mean of 0.74 (Table 4.2). The PIC values of the 14 SSR markers ranged from 0.30 (RM319) to 0.90 (RM206 and RM235) with a mean of 0.61.

Table 4.2. Genetic parameters generated by 14 SSR markers on 54 rice genotypes

	Genetic parameters					
Marker	Na	Ne	Ho	He	F_{is}	PIC
RM11	9	5.98	0.23	0.84	0.72	0.83
RM19	6	3.08	0.12	0.68	0.83	0.67
RM125	4	2.68	0.80	0.63	-0.27	0.63
RM1261	7	3.63	0.11	0.73	0.84	0.72
RM202	8	3.01	0.09	0.67	0.86	0.67
RM215	4	1.96	0.04	0.50	0.92	0.49
RM252	15	7.32	0.06	0.87	0.93	0.86
RM319	2	1.43	0.00	0.30	1.00	0.30
RM206	20	9.57	0.15	0.90	0.83	0.90
RM161	3	1.53	0.02	0.35	0.95	0.35
RM235	14	9.53	0.80	0.90	0.10	0.90
RM338	2	1.44	0.04	0.31	0.88	0.31
RM421	3	1.50	0.04	0.33	0.89	0.33
RM433	7	2.86	0.06	0.66	0.91	0.65
Mean	7.43	3.97	0.18	0.62	0.74	0.61
SE	1.46	0.78	0.07	0.06	0.10	0.06

Na = total number of alleles per locus, Ne = Number of effective alleles per locus, Ho = Observed gene diversity within landraces, He = Average gene diversity within landraces, F_{IS} = Inbreeding coefficient, PIC = Polymorphic information content and SE = Standard error.

4.3.2 Genetic relationship among 54 rice accessions based on source of collection

The genetic variability among rice genotypes based on source of collection is presented in Table 4.3. The mean values of observed (Na) and effective (Ne) number of detected alleles were 3.47 and 2.36, respectively. IRRI and Africa Rice recorded the lowest Na (2.53) and Ne (1.87), respectively. Similarly, the highest Na and Ne values of 5.67 and 3.26 were recorded for landrace collections. The mean observed Ho and He across rice genotypes were 0.17 and 0.47, respectively. The lowest values of Ho (0.12) and He (0.36) were observed from rice genotypes collected from Africa Rice and IRRI, respectively. The highest value of Ho = 0.22 and He = 0.53 was recorded from TARI and SUA, genotypes, respectively (Table 4.3). Shannon's information index ranged from 0.65 to 1.05 with a mean of 0.82. High heterozygosity values recorded were associated with F values ranging from 0.38 (TARI) to 0.77 (SUA), with a mean of 0.63 at population level (Table 4.3).

Table 4.3. Genetic diversity of 54 rice genotypes classified by areas of collection

Source	Genetic parameters					
	Na	Ne	I	Ho	He	F
SUA	3.27	2.66	0.95	0.16	0.53	0.77
AfricaRice	2.60	1.87	0.68	0.12	0.40	0.76
TARI	3.27	1.93	0.75	0.22	0.40	0.38
Landrace	5.67	3.26	1.05	0.18	0.48	0.61
IRRI	2.53	2.07	0.65	0.17	0.36	0.62
Mean	3.47	2.36	0.82	0.17	0.43	0.63
S.E.	0.57	0.26	0.08	0.02	0.03	0.07

Na = total number of alleles per locus; Ne = Number of effective alleles per locus; I = Shannon's information index; Ho = Observed gene diversity within landraces; He = Average gene diversity within landraces; F= Fixation index; SUA = Sokoine University of Agriculture; IRRI = International Rice Research Institute; SE= Standard error

The genetic differentiation (Fst) ranged from low (0) between IRRI and TARI accessions, while a large Fst (0.49) was observed between IRRI and Africa Rice collections (Table 4.4). Gene flow ranged between 0.05 and 1.06. The average Nei's unbiased genetic distance showed that the greatest genetic distance (1.84) was between genotypes collected from Africa Rice and landraces followed by Africa Rice and IRRI (1.74), SUA and IRRI (1.46), Africa Rice and TARI (1.41), SUA and TARI (1.32). The lowest genetic distance (0.08) was observed between TARI and IRRI rice genotypes. The genetic identity varied from 0.16 to 0.92 (Table 4.4). The highest genetic identity (0.92) was between TARI and Africa rice, followed by IRRI and SUA (0.88), TARI and SUA (0.81) genotypes and the lowest (0.16) observed between landraces and Africa rice.

Table 4.4. Pair-wise estimates of gene flow (above diagonal off brackets), genetic indenty (above diagonal within brackets) and genetic differentiation (lower diagonal offbrackets), genetic distance (lower diagonal within brackets)

Source	Gene flow (Nm)				
	SUA	AfricaRice	TARI	Landrace	IRRI
SUA		1.04 (0.79)	0.80 (0.81)	0.07 (0.24)	0.61 (0.88)
AfricaRice	0.00 (0.13)		1.06 (0.92)	0.12 (0.16)	0.07 (0.27)
TARI	0.38 (1.32)	0.46 (1.41)		0.05 (0,18)	0.16 (0.31)
Landrace	0.36 (1.19)	0.45 (1.84)	0.12 (0.21)		0.06 (0.23)
IRRI	0.39 (1.46)	0.49 (1.74)	0.00 (0.08)	0.13 (0.24)	
	Genetic differentiation (F_{ST})				

SUA = Sokoine University of Agriculture; TARI = Tanzania Agricultural Research Institute; IRRI= International Rice Research Institute

4.3.3 Analysis of molecular variance (AMOVA)

The results from AMOVA displayed highly significant genetic differences (P ≤ 0.001) among populations, among individuals and within individuals (Table 4.5). Thirty percent of the variance was due to genetic differentiation among the populations, while 47% of the variance was accounted for by individuals within populations. The remaining 23% of the variance was due to the differences within individuals.

Table 4.5. Analysis of molecular variance among and within the 54 rice genotypes

Source of variation	DF	SS	MS	Estimated variance	Percent variation	F-statistics
Among populations	4	139.11	34.78	1.65	30%	0.001
Among Individuals	48	305.36	6.36	2.57	47%	0.001
Within Individuals	53	65.00	1.23	1.23	23%	0.001
Total	105	509.47		5.45	100%	

DF= degrees of freedom; SS = sum of squares, MS = mean square

4.3.4 Population structure of 54 rice accessions

The population structure analysis of the 54 genotypes grouped the population into two sub-populations (Figure 4.1). Forty-one rice genotypes, representing 76% of the population, were assigned into sub-population 1 (Pop 1), and the remaining 13 were grouped into subpopulation 2 (Pop 2). Results showed that sub-population 1 comprised genotypes from SUA, landraces, TARI and Africa Rice, while population 2 consisted of landraces and IRRI genotypes.

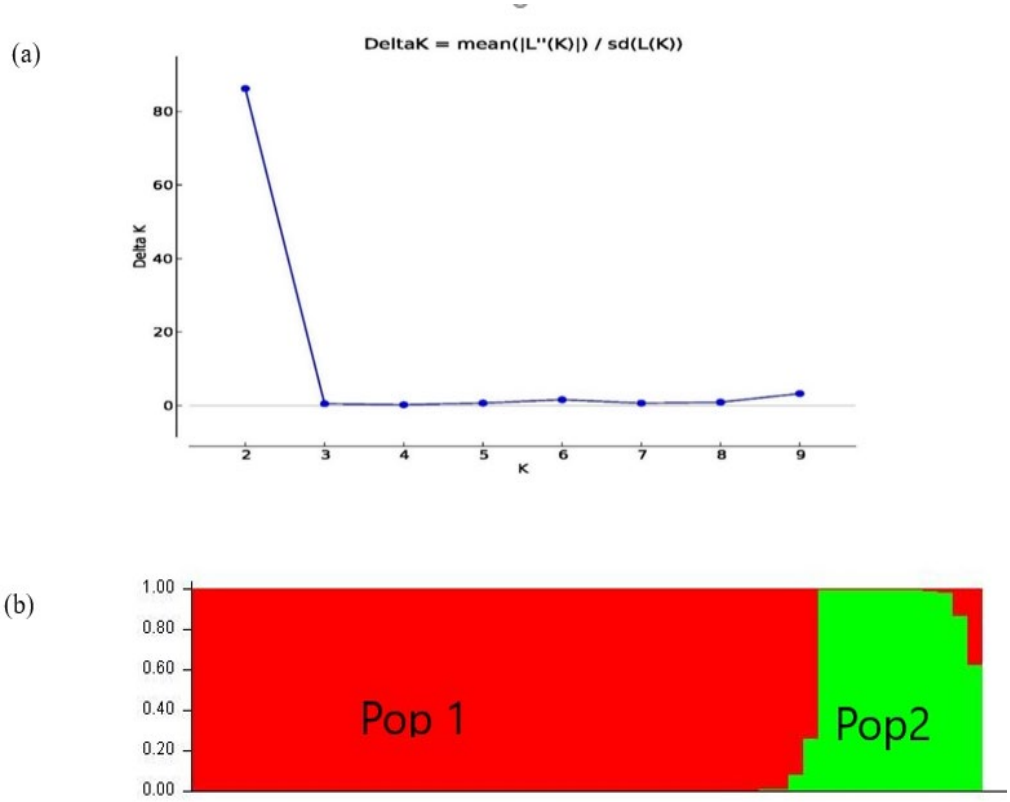

Figure 4.1. Population structure analysis of 54 rice accessions; (a) Delta K showing the number of populations, (b) = bar plot of populations sorted by kinship matrix (Pop 1 = population 1, Pop 2= population 2.

4.3.5 Genetic cluster analysis of 54 rice accessions

The UPGMA cluster analysis based on genetic dissimilarity using the neighbour-joining method grouped the 54 genotypes into three major clusters (Figure 4.2). The distribution of the genotypes into the three main clusters was not homogeneous. Cluster I consisted of one genotype. Cluster II composed of 25 (46.30%) of the rice genotypes studied (Figure 4.2). Cluster III comprised 28 (51.85%) genotypes (Figure 4.2). Genotypes, IR56 and Mwanza, Salama M-55 and Sindano nyeupe, SARO and Gigante, Mwanza and SARO, Lunyuki and Zambia, Rangimbili and IRAT 256, Zambia and Salama M-19 were highly distinct based on genetic makeup. The lowest genetic dissimilarity among rice genotypes was between Cherehani and Supa, Afaa Mwanza and Serena, Nerica 1 and Nerica 2, ITA 303 and SARO and IR 54 and 64 (Figure 4.2). These landraces may have the same genetic background but collected under different names in different locations.

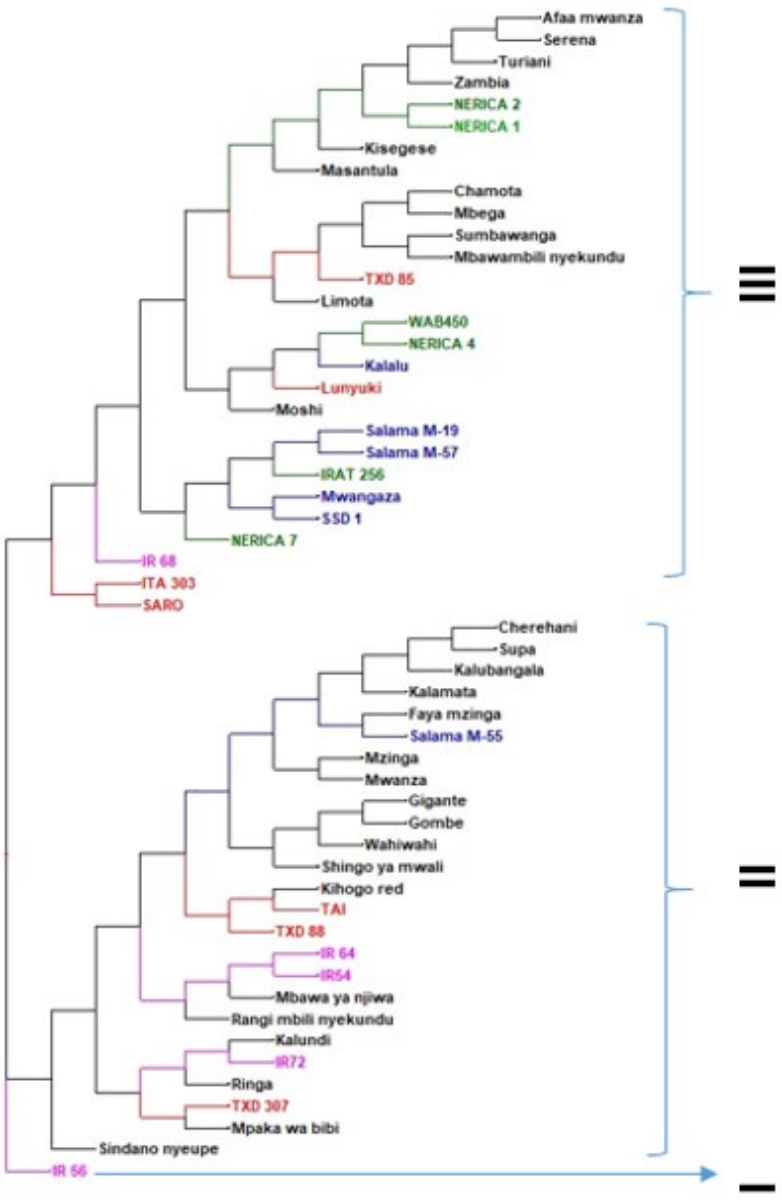

Figure 4.2. Dendrogram showing genetic relationship among 54 rice genotypes tested using 14 SSR markers. Accessions with the same colour share the same source of collection.

4.4 Discussion

Identification of genetic relationship and divergence of genetic resources is a useful step for parental choice for breeding. This will assist in minimising the use of closely related parents in breeding programs, which would otherwise lead to genetic depression and reduced genetic variation. The current study was therefore carried out to establish genetic diversity, relationship and population structure among selected rice genotypes to identify appropriate parents for hybridisation. The present study utilised 14 microsatellite markers to reveal genetic polymorphism of 54 rice accessions collected from four different sources. The genetic improvement of yield and other economically important traits in crop depends upon the genetic

diversity available within the crop species and the rice genotypes with high levels of genetic variation found in this study are beneficial resources for broadening the genetic base and for achieving rapid gains during rice breeding in Tanzania. A wide genetic diversity translates into a potentially high variation in morphological traits.

The number of alleles investigated ranged from 2 to 20, with a mean value of 7.48 per locus similar to 7.8 and 7.7 alleles per locus reported by Jain et al. (2004) and Zeng et al. (2007), respectively. This suggests that there is favourable allelic diversity, which is essential for assessment of genetic diversity. The mean number of alleles (7.48) obtained in the study was significantly higher than 6.4 alleles per locus reported by Chemutai et al. (2016). Rahman et al. (2012) detected even lower number of alleles of 4.18 using 34 SSR markers. In contrast, the number of alleles detected in the present study was lower than the average number of alleles (11.85) reported by Prathepha (2012). The variability in the number of alleles detected per locus might be due to the use of diverse genotypes.

The number of effective alleles per locus ranged from 1.43 to 9.57 with a mean of 3.97 close to 3.77 previously reported by Chen et al. (2017). Greater number of alleles generated by SSR markers suggests the usefulness of this marker system for detecting genetic polymorphism. In contrast, Aljumaili et al. (2018) detected 1.48 effective number of allele per SSR locus among 53 rice cultivars. On the contrary, effective number of alleles detected in the present study was lower than the average number of effective alleles (5.51) reported by Yelome et al. (2018), among West African rice accessions. The mean expected gene diversity was 0.62 (Table 4.2), which was similar to value reported by Wang et al. (2014). This was comparable to the findings of Aljumaili et al. (2018) who reported a gene diversity of 0.60 in a microsatellite-based study that involved 53 rice accessions. However, the mean gene diversity recorded in the present study was higher than that reported by Anh et al. (2018) and Islam et al. (2018). Further, the gene diversity obtained in the present study was higher than the findings of Chemutai et al. (2016), Choudhary et al. (2013), and Nachimuthu et al. (2015) who reported values of 0.54, 0.52 and 0.42 respectively, in rice. This could be attributed to high rate of exchange of genetic materials among the sources of germplasm collection.

The mean observed heterozygosity (Table 4.2) of the genotypes of 0.18 was similar to low heterozygosity reported by Yelome et al. (2018) among 42 rice accessions from six West African countries using 20 polymorphic SSR markers. The low level of heterozygosity has also been reported in other studies on rice (Choudhury et al., 2014; Nachimuthu et al., 2015) and this could be attributed to its autogamous mode of reproduction. Over 60% of the tested primers in the present study were highly polymorphic with mean PIC value of 61 implying the high

discriminating ability of the SSR markers. This indicates that the selected microsatellites were highly informative in distinguishing the test genotypes. The PIC value of a marker is the probability of the marker to be detected in the progeny and is a good measure of a marker's usefulness for linkage analysis. It is also a reflection of allelic diversity among varieties (Meti et al., 2013). The PIC and inbreeding coefficient (FIS) are the functions of how heterozygosity is partitioned within and among genotypes, based on differences in allele frequencies (Mulualem et al., 2018). Furthermore, high PIC value implies that the SSR markers were informative. A similar PIC value of 0.61 among rice genotypes was reported by Jain et al. (2004). In addition, the PIC values observed in this study were comparable to 0.60 and 0.62 reported by Meti et al. (2013) and Ashraf et al. (2016) using 12 and 24 SSR markers, respectively. On the contrary, the present study reported higher mean PIC value compared to 0.48 and 0.37 reported by Ashfaqa and Khan (2012) and Chemutai et al. (2016), respectively. The differences in PIC values maybe linked to the selection of different markers and the diversity of test genotypes.

Seven percent of the markers in the present study had negative inbreeding coefficient values (Table 4.2). FIS represents the average deviation of the population's genotypic proportions from Hardy-Weinberg equilibrium for a locus. The FIS values revealed that, one of the 14 markers (RM125) showed higher heterozygotes (−0.27). Populations differ with respect to richness of allelic diversity, distribution and frequency (Rao and Hodgkin, 2002). Variation in population may be attributed to the breeding system of the species and the ecological factors such as latitude, altitude, temperature, and moisture availability and other soil-related factors. Shannon's information index (I) ranged from 1.05 to 6.65 with an average of 0.82 (Table 4.3). This agrees with the findings of Aljumaili et al. (2018), who reported an index of 0.88. The high value of Shannon's information index in the present study was another indication of the presence of genetic diversity of the rice germplasm used in the study.

Population structure analysis revealed two sub-populations (Pop 1 and Pop 2) (Figure 4.1) indicating that a narrow genetic base exists among the studied rice genotypes. This result is consistent with the population structure of West African rice accessions reported by Yelome et al. (2018). Further, the population structure analysis confirmed the clustering of the sampled genotypes in a similar group, suggesting the need for crosses using genetically unrelated parents to develop breeding populations. However, AMOVA revealed highly significant genetic differences ($P \leq 0.001$) among the populations, among individuals and within individuals (Table 4.6). Of the total genetic variation in the 54 genotypes, 47% of the variation was contributed by the genetic differentiation among individuals within populations indicating that there is adequate variation among the studied genotypes useful for breeding. Variation of similar pattern has been reported in previous studies on rice germplasm (Aljumaili et al., 2018; Yelome et al., 2018).

AMOVA results suggest that a small collection within a given source will capture the genetic diversity present in the test genotypes. The presence of variability within and between the populations represents the possibility of making wide crosses for population development and to enhance genetic divergence in rice.

Genetic distance is a measure of the genetic divergence between pairs of genotypes or populations. The present study revealed average genetic distance estimate of 1.84, which is higher than previous reports. Becerra et al. (2015) reported a genetic distance of 0.87 in elite rice genotypes from Chile. Similarly, a mean genetic distance of 0.86 was reported in Ugandan rice genotypes (Mogga et al., 2017). In addition, Ndjiondjop et al. (2018) reported a genetic distance of 0.01 to 0.76. The high genetic distance (1.84) for the genotypes studied could be attributed to the uniqueness of Tanzania rice germplasm collections, which seems to be different from other regions. According to Nei (1972), genetic distance is linearly related to geographical distance. However, the genetic distance values of rice germplasm (1.74 and 1.84) require further confirmation using additional SSR primers.

According to standard interpretation of genetic differentiation, 0.0 to 0.005 shows little, 0.05 to 0.15 moderate, 0.15 to 0.25 large, and above 0.25 very large genetic differentiations (Wright 1978). The lower genetic variance among sources of collection in this study can also be associated with the observed low gene differentiation and high gene flow. According to Morjan and Rieseberg (2004), gene flow <1 is considered to be low (Nm), while Nm = 1 is considered to be moderate and Nm > 1 is considered to be high. The occurrence of high gene flow in the germplasm studied could be attributed to the evolutionary history of these populations, out-crossing between rice genotypes or effects of spontaneous mutations (Nuijten et al., 2009). Further, exchange of rice genotypes among farmers and traders may have enhanced gene flow across rice growing regions of Tanzania.

The UPGMA cluster analysis based on genetic dissimilarity using the neighbour-joining method grouped the 54 genotypes into three major distinct clusters. The clustering pattern in the present study indicated the existence of variability among rice genotypes. Chemutai et al. (2016) also grouped 50 rice genotypes into three clusters using SSR markers. However, the cluster patterns did not correspond to the predefined population structure based on the area of collection. According to Mulualem et al. (2018), this may be due to the fact that genotypes collected from similar areas belong to the same gene pool or they may have similar ancestral relationships. Conversely, genetic dissimilarity among the rice genotypes studied could arise due to the diverse ancestral origin, high gene flow caused by cross-pollination and possible gene/chromosomal mutation. In the present study, rice genotypes collected from similar regions

were grouped together in the same cluster such as Gigante and Gombe, and Salama M-55 and Faya mzinga. These results agree with earlier studies, which reported that geographical separation did not affect genetic distance among genotypes (Zhang et al., 2012). According to Ganesamurthy et al. (2010), geographic location should not be used as a measure of genetic diversity during genotype selection. This could be a consequence of exchange of genetic materials among the neighbouring farmers as well as traders in the region. Besides, farmers' selections and management practice affect the patterns of genetic diversity (Barnaud et al., 2008). Tanzanian rice farmers' recycled seed as a source of planting material, which in turn increases the genetic similarity among landraces. Mekbib (2007) reported that farmers selected and preserved genotypes are based on the phenotypic and agronomic traits. The study suggests that parents used in breeding should be chosen following assessment of genetic diversity based on molecular markers.

4.5 Conclusions

In conclusion, the current study found the existence of reasonable variability among rice genotypes, which could be exploited for future breeding. The results revealed that nine of the 14 selected SSR markers were highly polymorphic and sufficiently distinguished the tested rice genotypes. The cluster analysis classified the 54 rice genotypes into three major distinct genetic groups irrespective of the source of collection. Genotypes IR56, Mwanza, Salama M-55, Sindano Nyeupe, Gigante, SARO, Lunyuki, Rangimbili, IRAT 256, Zambia and Salama M-19 showed unique genetic pattern and relationship suggesting that they may have different genetic makeup. These can be used as sources of novel genes in rice breeding programs. Hence, the information generated will contribute significantly to rice improvement in Tanzania and other related environments in East Africa.

References

Ali, M.A., M.S. Islam, S.K. Mandal, Z. Nasrin, M.M. Rahman, R.H. Kuddus, and S.H. Prodhan. 2014. Genetic diversity among salt-tolerant rice (*Oryza sativa* L.) landraces cultivated in the coastal districts of Bangladesh. Journal of Bioscience and Biotechnology 3: 15-22.

Aljumaili, S.J., M.Y. Rafii, M.A. Latif, S.Z. Sakimin, I.W. Arolu, and G. Miah, 2018. Genetic diversity of aromatic rice germplasm revealed by SSR markers. BioMed Research International 1-11.

Anh, T.T.T., T.D. Khanh, T.D. Dat, and T.D. Xuan. 2018. Identification of phenotypic variation and genetic diversity in rice (*Oryza sativa* L.) mutants. Agriculture 8(2):30. doi: 10.3390/agriculture8020030.

Ansah, T., W. Dogbe, S. Cudjoe, A.A.B. Iddrisu, and A.S Eseoghene. 2017. Agronomic performance of five rice varieties and nutritive value of the straw from these varieties. West African Journal of Applied Ecology 25(1): 1-10.

Ashfaqa, M., and A.S. Khan. 2012. Genetic diversity in basmati rice (*Oryza sativa* L.) germplasm as revealed by microsatellite (SSR) markers. Russian Journal of Genetics 48 (1): 53-62.

Ashraf, H., A.H. Husaini, M.A. Bhat, G.A. Parray, S. Khan, and N.A. Ganai. 2016. SSR based genetic diversity of pigmented and aromatic rice (*Oryza sativa* L.) genotypes of the western Himalayan region of India. Physiology Molecular Biology Plants 22(4): 547-555.

Barnaud, A., G. Trigueros, D. McKey, and H.I. Joly. 2008. High outcrossing rates in fields with mixed sorghum landraces: how are landraces maintained? Heredity 101: 445-452.

Becerra, V., M. Paredes, E. Gutierrez, and C. Rojo. 2015. Genetic diversity, identification, and certification of Chilean rice varieties using molecular markers. Chilean Journal of Agricultural Research 75(3): 267-274.

Bucheyeki, L.K., E. Shennkawa, D. Kadadi, and J. Lobulu. 2011. Assessment of rice production constraints and farmers preferences in Nzega and Igunga district, Tanzania. Journal of Advance in Development Research 2: 30-37.

Chemutai, L.R., M.A. Musyoki, W.F. Kioko, N.S. Mwenda, K.G. Muriira. 2016. Genetic diversity studies on selected rice (*Oryza sativa* L.) genotypes based on gel consistency and alkali digestion. Journal of Rice Research 4:172. doi: 10.4172/2375-4338.1000172.

Chen, C., W. He, T. Y. Nassirou, A. Nsabiyumva, X. Dong, Y.M.N, Adedze, and D. Jin. 2017. Molecular characterization and genetic diversity of different genotypes of *Oryza sativa* and *Oryza glaberrima*. Electronic Journal of Biotechnology *30*: 48-57.

Chen, X., S. Temnykh, Y. Xu, Y.G. Cho, and S.R. McCouch. 1997. Development of a microsatellite framework map providing genome-wide coverage in rice (*Oryza sativa* L.). Theoretical and Applied Genetics 95: 553-567.

Choudhary, G., N. Ranjitkumar, M. Surapaneni, D.A. Deborah, A. Vipparla, and G. Anuradha. 2013. Molecular genetic diversity of major Indian rice cultivars over decadal period. PLoS One 8(6): e66197. doi: 10.1371/journal.pone.0066197.

Choudhury, D.R., N. Singh, A.K. Singh, S. Kumar, K. Srinivasan, and R.K. Tyagi. 2014. Analysis of genetic diversity and population structure of rice germplasm from North-eastern region of India and development of a core germplasm set. PLoS ONE 9(11): e113094. doi:10.1371/journal.pone.0113094.

Dixit, S., B.P.M. Swamy, P. Vikram, H.U. Ahmed, M.T.S. Cruz, M. Amante, D. Atri, H. Leung, and A. Kumar. 2012. Fine mapping of QTLs for rice grain yield under drought reveals

sub-QTLs conferring a response to variable drought severities. Theoretical and Applied Genetics 125(1): 155 -169.

Earl, D.A., and B.M. von Holdt. 2012. STRUCTURE HARVESTER: a website and program for visualizing STRUCTURE output and implementing the Evanno method. Conservation Genetic Resource 4:359-361. doi: 10.1007/s12686-011-9548-7.

Evanno, G., S. Regnaut, and J. Goudet, 2005. Detecting the number of clusters of individuals using the software STRUCTURE: a simulation study. Molecular Ecology 14:2611-2620.

FAO. 2017. Global paddy production. http://faostat.fao.org/default.aspx. Date visited 18th June, 2019.

Ganesamurthy, K., D. Punitha, and M. Elangovan, 2010. Genetic diversity among the land races of sorghum collected in Tamil Nadu. Electronic Journal of Plant Breeding 1: 1375-1379.

Islam, M.R., G.B. Gregorio, M.A. Salam, B.C.Y. Collard, R.K. Singh, and L. Hassan. 2012. Validation of SalTol linked markers and haplotype diversity on chromosome 1 of rice. Molecular Plant Breeding 3: 103 -114.

Islam, M.Z., M. Khalequzzaman, M.F.R.K. Prince, M.A. Siddique, M.S.M.H. Rashid, M.S.U. Ahmed, B.R. Pittendrigh, and M.P. Ali. 2018. Diversity and population structure of red rice germplasm in Bangladesh. PLoS ONE 13(5): e0196096. doi:10.1371/journal.pone.0196096.

Islam, M.R., R.K. Singh, M.A. Salam, L. Hassan, and G.B. Gregorio. 2008. Molecular diversity of stress tolerant rice genotypes using SSR markers. SABRAO Journal of Breeding and Genetics 40(2): 127-139.

Jain, S., R.K. Jain, and S.R. McCouch. 2004. Genetic analysis of Indian aromatic and quality rice (*Oryza sativa* L.) germplasm using panels of fluorescently labelled microsatellite markers. Theory of Applied Genetic 109 (5): 965-977.

Lee, H.H., P.P.N. Neoh, W.S.T. Bong, J. Puvaneswaran, S.C. Wong, P.H. Yiu, and A. Rajan. 2011. Genotyping of Sarawak rice cultivars using microsatellite markers. Pertanika Journal of Tropical Agricultural Science 34 (1): 123-136.

Liu, D., J.Y., X.X. Wang, X.L. Yang, J. Sun, and W.F. Chen. 2015. Genetic diversity and elite gene introgression reveal the japonica rice breeding in northern China. Journal of Integrative Agriculture 14: 811-822.

Luther, Z., R. Akromah, D. Nyadanu, D.P. Tokpah, Z. Page, V.M. Voor, and A.D. Kwaloe. 2017. Evaluation of genetic diversity in rice (*Oryza sativa* and *Oryza glaberrima*) germplasm from Liberia and Ghana using simple sequence repeat (SSR) markers. African Journal of Biotechnology 16:1990-1996.

Mekbib, F. 2007. Infra-specific folk taxonomy in sorghum (*Sorghum bicolor* L.) Moench] in Ethiopia: folk nomenclature, classification, and criteria. Journal of Ethnobiology and Ethnomedicine **3**:38. doi: 10.1186/1746-4269-3-38.

Meti, N., K.C. Samal, D.N. Bastia, and G. R., Rout. 2013. Genetic diversity analysis in aromatic rice genotypes using microsatellite based simple sequence repeats (SSR) marker. African Journal of Biotechnology 12(27): 4238-4250.

Mittal, N., and A.K. Dubey, 2009. Microsatellite markers- A new practice of DNA based markers in molecular genetics. A review. Plant Breeding 3:235-246.

Mligo, F.E.I., and C.P. Msuya. 2015. Farmer's adoption of recommended rice varieties: A case of Kilombero district of Morogoro region of Tanzania. Southern African Journal of Agriculture Extension 43 (1): 41-56.

Mogga, M., J. Sibiya, H. Shimelis, J. Lamo, and N. Ochanda. 2018. Appraisal of major determinants of rice production and farmers choice of rice ideotypes in South Sudan: implications for breeding and policy interventions. Experimental Agriculture 1-14. doi: 10.1017/S0014479718000017.

Mogga, M., J. Sibiya, H. Shimelis, J. Lamo, and N. Yao. 2017. Diversity analysis and genome-wide association studies of grain shape and eating quality traits in rice (*Oryza sativa* L.) using DArT markers. PLoS ONE 13(6): e0198012. doi: 10.1371/journal.pone.0198012.

Morjan, C.L. and L.H. Rieseberg. 2004. How species evolve collectively: Implications of gene flow and selection for the spread of advantageous alleles. Molecular Ecology 13: 1341-1356.

Mousavi, S., R. Mariotti, L. Regni, L. Nasini, M. Bufacchi, S. Pandofi, L. Baldoni, and P. Proietti. 2017. The first molecular identification of an olive collection applying standard simple sequence repeats and novel expressed sequence tag markers. Frontiers in Plant Science 8:1283. doi:10.3389/fpls.2017.01283.

Mulualem, T., F. Mekbib, H. Shimelis, E. Gebre, and Amelework, B. 2018. Genetic diversity of yam (*Dioscorea spp*.) landrace collections from Ethiopia using simple sequence repeat markers. Australian Journal of Crop Science 12(8):1223-1230.

Nachimuthu, V.V., R. Muthurajan, S. Duraialaguraja, R. Sivakami, B.A. Pandian, and G. Ponniah. 2015. Analysis of population structure and genetic diversity in rice germplasm using SSR Markers: An initiative towards association mapping of agronomic traits in *Oryza sativa*. Rice 8: 30-54.

Ndjiondjop, M.N., K. Semagin, and M.L. Warburton. 2018. Assessment of genetic variation and population structure of diverse rice genotypes adapted to lowland and upland ecologies in Africa using SNPs. Frontiers in Plant Science 9: 446. doi: 10.3389/fpls.2018.00446.

Nei, M. 1972. Genetic distance between populations. American Naturalist 106:283-292.

Nei, M., and W. Li. 1979. Mathematical method for studying genetic variation in terms of restriction endonucleases. Proceedings of the National Academy of Sciences 76: 5256 -5273.

Nkuba, J., A. Ndunguru, R. Madulu, D., Lwezaura, G. Kajiru, A. Babu, B. Chalamila, and G. Ley. 2016. Rice value chain in Tanzania: Identification of constraints, opportunities and upgrading strategies. African Crop Science Journal 26:73-87.

Nuijten, E., R. Struik, P.C. Van Treuren, A. Mokuwa, F. Okry, B.Teeken, and P. Richards, 2009. Evidence for the emergence of new rice types of interspecific hybrid origin in West African farmers' fields. PLoS ONE 4(10): e7335. doi: 10.1371/journal.pone.0007335.

Palanga, K.K., K. Traore, K. Bimpong, M. Jamshed, and M A.P. Mkulama. 2016. Genetic diversity studies on selected rice varieties grown in Africa based on aroma, cooking and eating quality. African Journal of Biotechnology 15: 1136-1146.

Panaud, O., X. Chenayd, and S.R. McCouch. 1996. Development of microsatellite markers and characterization of simple sequence length polymorphism (SSLP) in rice (*Oryza sativa* L.) Molecular and General Genetics 252(5): 597-607.

Peakall, R., and P.E. Smouse. 2007. GENALEX6: genetic analysis in excel. Population genetic software for teaching and research. Molecular Ecology 6: 288-295.

Perrier, X., and J.P. Jacquemoud-Collet. 2006. DARwin software http://darwin.cirad.fr/darwin

Pervaiz, Z.H. M.A. Rabbani, I. Khaliq, S.R. Pearce, and S.A. Malik. 2010. Genetic diversity associated with agronomic traits using microsatellite markers in Pakistani rice landraces. Electronic Journal of Biotechnology 13 (3): 1-12.

Prathepha, P.2012. Genetic diversity and population structure of wild rice (*Oryza rufipogon*) from North-eastern Thailand and Laos. Australian Journal Crop Science 6: 717-723.

Pritchard, J., M. Stephens, N. Rosenberg, and P. Donnelly. 2000. Association mapping in structured populations. American Journal of Human Genetics 67:170-180.

Rahman, M.M., M.G. Rasaul, M.A. Hossain, K.M. Iftekharuddaula, and H. Hasegawa. 2012. Molecular characterization and genetic diversity analysis of rice (*Oryza sativa* L.) using SSR markers. Journal of Crop Improvement 26(2): 244-257.

Rao, V.R., and Hodgkin, T. 2002 Genetic diversity and conservation and utilization of plant genetic resources. Plant Cell Tissue Organ Culture 68: 1-19.

Sorkheh, K., M. Masaeli, M.H. Chaleshtori, A. Adugna, and S. Ercisli. 2016. AFLP-based analysis of genetic diversity, population structure, and relationships with agronomic traits in rice germplasm from North region of Iran and world core germplasm set. Biochemical Genetics 54(2): 177-193. doi: 10.1007/ s10528-016-9711-7.

Sun, C.Q., X.K. Wang, A. Yoshimura, and N. Iwata. 2000. A study of the genetic diversity of common wild rice (*Oryza rufipogon* Griff.) and cultivated rice (*Oryza sativa* L.) by RFLP analysis. Acta Genetica Sinica 27: 227-234.

Sun, X.Y., S. Kang, Y.J. Zhang, X.Q. Tan, Y F. Yu, H.Y. He, X.Y. Zhang, Y.F. Liu, S. Wang, W.X. Sun, L. Cai, and S.J. Li. 2013. Genetic diversity and population structure of rice pathogen *Ustilaginoidea virens* in China. PLOS ONE 8(12). doi: 10.1371/annotation/13aec20c-1d9d-4c18-a9a3-a8767987b9b8.

Temnykh, S., W.D. Park, N. Ayes, S. Cartinhour, N. Hauck, L. Lipovich, Y.G. Cho, T. and Ishii, S.R. McCouch. 2000. Mapping and genome organization of microsatellite sequences in rice (*Oryza sativa* L.). Theoretical and Applied Genetics 100(5): 697-712.

Wang, J., T. Jiang, D. Zou, H. Zhao, Q. Li, H. Liu, and C. Zhou. 2014. Genetic diversity and genetic relationships of japonica rice varieties in Northeast Asia based on SSR markers. Biotechnology and Biotechnological Equipment 28(2): 230-237.

Wang, X., J. Zhu, L. Mansueto, and R. Bruskiewich. 2005. Identification of candidate genes for drought stress tolerance in rice by the integration of a genetic (QTL) map with the rice genome physical map. Journal of Zhejiang University Science 6B (5): 382-388.

Wright, S. 1978. Evolution and the genetics of populations: Variability within and among natural populations. University of Chicago Press, Chicago, USA.

Xiao, X.Y., Y.P. Wang, J.Y. Zhang, S.G. Li, and T.Z. Rong. 2016. SSR marker-based genetic diversity fingerprinting of hybrid rice in Sichuan, China. China Journal of Rice Science 20: 1-7.

Yan, A.O., X. Yong, C. Xiao-fen, W. An, T. Fei, S. Li-qun, and L. Qiao-quan. 2016. A genetic diversity assessment of starch quality traits in rice landraces from the Taihu basin, China. Journal of Integrative Agriculture15 (3): 493-501.

Yelome, O.I., K. Audenaert, S. Landschoot, A.D.W. Vanhove, D. Silue, P. Van Damme, and G. Haesaert. 2018. Analysis of population structure and genetic diversity reveals gene flow and geographic patterns in cultivated rice (*O. sativa and O. glaberrima*) in West Africa. Euphytica 214-215. doi: 10.1007/s10681-018-2285-1.

Zeng, Y., H. Zhang, Z. Li, S. Shen, J. Sun, M. Wang, D. Liao, X. Liu, X. Wang, F. Xiao, and G. Wen. 2007. Evaluation of genetic diversity of rice landraces (*Oryza sativa* L.) in Yunnan, China. Breeding Science 57: 91-99.

Zhang, H., L. Wei, H. Miao, T. Zhang, and C. Wang. 2012. Development and validation of genetic SSR markers in sesame by RNA-seq. Bio Medical Central Genomics 13: 316-317.

CHAPTER FIVE: COMBINING ABILITY AND GENE ACTION FOR RICE YELLOW MOTTLE VIRUS DISEASE RESISTANCE AND AGRONOMIC TRAITS IN RICE (*Oryza sativa* L.)

Abstract

Selecting genetically diverse and complementary parental lines and superior crosses are pre-requisites in developing improved cultivars. The objectives of this study were to determine the combining ability effects and gene action conditioning rice yellow mottle virus (RYMV) resistance and agronomic traits in selected parental lines and derived families in rice (*Oryza sativa* L.) for future breeding. Ten parental lines and their 45 F_2 progenies were field evaluated in three selected locations using a 5 × 11 alpha lattice design with two replications. The genotype × site interaction effects were significant ($p<0.05$) for the number of tillers per plant (NT), number of panicles per plant (NPP), number of grains per panicle (NGP), percentage of filled grains (PFG), thousand grain weight (TGW), rice yellow mottle virus disease (RYMVD) resistance and grain yield (GY). The variance due to the general combining ability (GCA) and the specific combining ability (SCA) effects were both significant for all assessed traits, indicating that both additive and non-additive gene action were involved in governing trait inheritance. The high GCA to SCA ratios calculated for all the studied traits indicated that additive genetic effect was predominant. Parental lines, Mwangaza, Lunyuki, Salama M-57, Salama M-19, IRAT 256 and Salama M-55, which had negative GCA effects for RYMVD, and families such as SARO × Salama M-55, IRAT 245 × Rangimbili, Rangimbili × Gigante and Rangimbili × Mwangaza, which had negative SCA effects for RYMVD, were selected for RYMV resistance breeding. The crosses such as Rangimbili × Gigante, Gigante × Salama M-19 and Rangimbili × Salama M-55 were selected due to their desirable SCA effects for GY. The predominance of additive gene effects for agronomic traits and RYMVD resistance in the present breeding populations suggest that rice improvement could be achieved through gene introgression using recurrent selection, but this can be challenging in a strongly self-pollinating crop such as rice.

Keywords: Cultivar development; diallel, gene action, RYMV resistance, rice breeding, yield components

---

Suvi, W.T., H. Shimelis, M. Laing, I. Mathew, and A.I.T. Shayanowako. 2021. Determining the Combining Ability and Gene Action for Rice Yellow Mottle Virus Disease Resistance and Agronomic Traits in Rice (*Oryza sativa* L.). Agronomy11, 12. https://dx.doi. org/10.3390/agronomy11010.

5.1 Introduction

Rice (*Oryza sativa* L., 2n = 2x = 24) is the second most important global crop after wheat in terms of total production (Srujana et al., 2017; FAO, 2018). Globally, rice is cultivated on about 167 million hectares with an annual production of 744.4 million tonnes of grain (FAOSTAT, 2017). More than 90% of rice is grown and consumed in Asia (IRRI, 2013; Akanksha and Jaiswal, 2019), while the sub-Saharan Africa (SSA) region accounts for about 15% of the global rice production (FAOSTAT, 2015). In East and Southern Africa region, Tanzania is the second largest producer of rice after Madagascar. Rice is ranked as the second most important staple crop after maize (*Zea mays*) in Tanzania (Hubert et al., 2017; Suvi et al., 2018).

Despite the contribution of rice to food and nutrition security, and enhanced livelihoods of millions of people in Tanzania, the average yield in the country is 1.5 t ha^{-1}, which is significantly lower than the yield potential of 4.6 t ha^{-1} reported in Asia (FAOSTAT, 2015). The low productivity of rice in Tanzania is caused by a combination of biotic and abiotic stresses, and socio-economic constraints (Chuwa et al., 2015; Duku et al., 2016; Atera et al., 2018). The rice yellow mottle virus (RYMV) disease has been identified as a major biotic constraint limiting rice productivity in SSA countries including Tanzania (Duku et al., 2016; Hubert et al., 2016; Suvi et al., 2018). The first incidence of RYMV disease in Tanzania was reported in 1980s in subsistence farming systems in the Morogoro region (Kanyekai et al., 1996). The disease has since become endemic in all the rice-growing regions under both rain-fed and irrigated production systems. The RYMV causes yield losses ranging from 20% to 100%, depending on cultivar susceptibility, and stage of growth at the onset and development of infection (Kouassi et al., 2005; Luzi-Kihupi et al., 2009; Hubert et al., 2017). The RYMV disease is characterized by mottling and yellowing symptoms, resulting in reduced photosynthetic area, stunted growth, reduced tiller formation and increased grain sterility (Koussi et al., 2005; Hubert et al., 2016). Locally grown, farmer-preferred rice varieties with good culinary properties have been reported to be susceptible to RYMV (Ochola and Tusiime, 2011). Hence, there is a need to developing improved rice varieties with RYMV resistance and farmer-preferred traits.

Host plant resistance is the most economical and environmentally friendly approach to control the RYMV. The development of RYMV resistant and agronomically superior genotypes requires genetically diverse and complementary parental lines and superior crosses for selection (Zouzou et al., 2008). Genes conditioning RYMV resistance have been reported previously (Ndjiondjop et al., 1999; Traore et al., 2015; Sereme et al., 2016). The *RYMV1* and *RYMV2* are the most widely reported genes (Ndjiondjop et al., 1999; Thiemele et al., 2010; Pinel-Galzi et al., 2016). Hence, RYMV resistant and agronomically suitable donor parents can be used in

local pre-breeding programs to develop new populations for variety development. Gene introgression requires an understanding of the nature of gene action and trait heritability.

Assessing combining ability and gene action for RYMV resistance and agronomic traits in rice would provide a basis for the development and selection of a breeding population. Combining ability analysis can facilitate the selection of suitable parents for hybridization, and identification of promising recombinants (Sprague and Tatum, 1942; Falconer et al., 1996; Acquaah, 2012). Broadly, combining ability is divided into the general combining ability (GCA) effects of the parents, and the specific combining ability (SCA) effects of the crosses. The GCA is the average performance of a line in a series of hybrid combinations and relates to additive gene action. The SCA refers to the deviation of the performance of a cross-based on the average performance of its parents. The SCA effects are associated with non-additive gene actions such as dominance and epistasis (Schlegel, 2010).

The diallel mating design has been used widely in determining the combining ability effects in self-pollinating species such as rice. Several studies have been carried out on combining ability effects in rice with varied results. Munganyinka et al. (2016) and Mogga et al. (2010) reported a preponderance of additive gene action in conditioning RYMV resistance. Conversely, Paul et al. (2003) reported that dominance gene action was responsible for conditioning resistance to RYMV. Therefore, combining ability analyses and genetic predictions are dependent on the test populations and environment. With a goal of developing rice varieties adapted to Tanzanian conditions, with high RYMV resistance, genetically diverse rice genotypes were assembled and evaluated using agronomic traits and simple sequence repeat (SSR) markers (Suvi et al., 2019). This allowed for the selection of promising and complementary parents to be included in a rice pre-breeding program in Tanzania. Therefore, the objective of this study was to determine the combining ability effects and gene action conditioning RYMV resistance and agronomic traits in selected parental lines and derived families for subsequent breeding activities.

5.2 Materials and methods

5.2.1 Plant materials

Ten selected rice genotypes were used to generate new populations. A description of the parental genotypes is presented in Table 5.1. The parents were selected from previous phenotypic and genotypic evaluations for their diversity in terms of RYMV resistance and agronomic traits. The selections included genotypes sourced from Sokoine University of Agriculture (SUA), landraces from local farmers, a variety from the Tanzania Agricultural Research institute (TARI) and one genotype from Africa Rice.

5.2.2 Population development

Crossing blocks were initiated under a screen-house condition at the Tanzania Agricultural Research Institute (TARI), Ilonga (6°50'3.39"S and 36°59'30.17"E), with an altitude of 491 metres above sea level. The temperature range and mean relative humidity during the growing period were 20 to 32°C and 87.4%, respectively. The parents were planted in 10 L capacity plastic pots. Crosses were undertaken using a 10 × 10 half-diallel mating design to produce 45 F_1 families, between May and August in 2018. Parents were stagger-planted at weekly intervals to synchronize flowering. A vacuum emasculation method was used (Lamo, 2010). Pollination was carried out between 10.00 am to 12:00 noon. After emasculation, panicles were immediately covered with a pollination bag and sealed with paper clips to avoid unintended cross-pollinations. The pollinating bag was removed from the emasculated female parent and a fertile panicle from the male parent was gently dusted onto the female panicle. The flowers were bagged immediately after hand pollination. The pollinating bag covered the female parent (Figure 5.1) to prevent cross-pollination and to maintain high relative humidity for better fertilization. Mature seeds from each successful cross were harvested 25 to 30 days after pollination and kept separately with proper records. The seed of the F_1 crosses (Figures 5.2 and 5.3) were planted for seed bulking and genetic analysis at the F_2 generation.

Table 5.1. Description of rice parental genotypes used for population development

No	Genotype	Status	Origin	RYMV resistance	Agronomic and grain quality traits
1	Salama M-57	Accession	Tanzania	Resistance	Unscented; long grain
2	IRAT 256	Accession	Tanzania	Resistance	Unscented
3	Rangimbili	Landrace	Tanzania	Susceptible	Scented; high grain quality
4	Zambia	Landrace	Tanzania	Susceptible	Scented; high grain quality,
5	Lunyuki	Accession	Tanzania	Resistance	Unscented, long grain
6	SARO	Variety	Tanzania	Susceptible	Scented, high yielding
7	Mwangaza	Variety	Tanzania	Resistance	Unscented; long grain
8	Salama M-55	Accession	Tanzania	Resistance	Unscented
9	Gigante	Accession	AfricaRice	Susceptible	Large panicle
10	Salama M-19	Accession	Tanzania	Resistance	Unscented; long grain

Figure 5.1. Emasculated and pollinated rice genotypes covered with pollination bags.

Figure 5.2. Germinating F_1 seed in a Petri-dish being advanced to F_2.

Figure 5.3. F_1 plants established in plastic pots to produce F_2 seeds.

5.2.3 Field evaluations

5.2.3.1 Descriptions of the study areas

The parental lines and their F_2 crosses were field evaluated in three sites namely, Ifakara, Ilonga and Mkindo sites in Tanzania (Table 5.2). Evaluations were conducted during the main cropping season (December 2019 to June 2020). The experimental sites are hotspot areas for RYMV disease. The sites are known for the high disease pressure that develops during the growing season. The climatic conditions of the study sites are summarized in Table 5.2. All the three sites experience a sub-humid tropical climate with a bimodal rainfall distribution. The short rainy season usually starts in October and ends in December, while the long rainy season lasts between March and May.

Table 5.2. Descriptions of the three sites used for evaluation of 45 crosses and 10 parents

Site	District	Test condition	Latitude (S)	Longitude (E)	Altitude (masl)	Total annual rainfall (mm)	TMax (^{0}C)	TMin (^{0}C)
Ifakara	Kilombero	Rain-fed	08°03'	36°40'	271	980	27.34	17.71
Ilonga	Kilosa	Irrigated	6°74'	37°05'	607	1194	28.43	19. 24
Mkindo	Mvomero	Irrigated	6°14'	38°41'	430	975	25.67	16.94

S = south; E = east; m = metre above sea level; mm = millimetre; TMax =average maximum temperature; TMin = average minimum temperature.

5.2.3.2 Experimental design and management

The 10 parents and the 45 F_2 crosses were established using a 5 × 11 alpha lattice design with two replications at each site. Each genotype was planted in a plot measuring 2.4 m × 2.4 m. The seeds were planted directly at Ifakara while transplanting of 21-day old seedlings was carried out at the Ilonga and Mkindo sites. The seeds or seedlings were sown or transplanted, respectively, in each plot at a spacing of 20 cm x 20 cm, with one plant per hill. Potassium and phosphorous fertilizers were used for basal application at all sites prior to planting at a rate of 65 kg P ha^{-1} and 54 kg K ha^{-1}, respectively. Urea fertilizer (46% nitrogen content) was broadcasted in two equal splits (the first at tillering and the second at panicle initiation) as top dressings to deliver a total level of 60 kg N ha^{-1}. The rest of the cultural practices, including thinning and hand weeding, were applied as recommended, to ensure uniform and healthy crop growth.

5.2.3.3 Data collection

Data collected included the RYMV reaction and agronomic traits, based on the IRRI Standard Evaluation System (SES) for rice (IRRI, 2013). The severity of the RYMV disease reaction was scored using a scale of 1 to 9 (IRRI, 2002) (Table 5.3).

Table 5.3. Rice yellow mottle virus disease severity rating scale and description (IRRI, 2002)

Rating scale	Description
1	Highly resistant (no symptoms observed)
3	Resistant (leaves green, but with sparse dots or streaks, less than 5% reduction of height)
5	Moderate resistant (Leaves green or pale green with mottling, 6%-25% height reduction)
7	Susceptible (Leaves pale yellow or yellow, 26-75% height reduction, flowering slightly delayed)
9	Highly susceptible (Leaves yellow or orange, more than 75% height reduction, no flowering)

Data on days to 50% flowering (DFL), number of tillers per plant (NT), plant height (PH), number of panicles per plant (NPP), panicle length (PL), number of grains per panicle (NGP), percentage-filled grains per panicle (PFG), 1000-grain weight (TGW) and grain yield (GY) were collected. The DFL were recorded by counting the number of days from sowing to when 50% of all the plants in each plot had flowered. The NT from 10 randomly selected plants in a plot were recorded at physiological maturity. PH was measured in centimetres (cm) using a ruler from the soil surface to the tip of the longest panicle at physiological maturity. The NPP was recorded by counting the number of fully exerted panicles bearing grains from a sample of selected 10 plants and their sum averaged to obtain the NPP. The PL was measured in cm using a ruler from the panicle base node to the tip (end) at the base on 10 selected plants per plot. NGP was counted using a seed counter and recorded as an average of samples from 10 panicles per plot. The PFG was calculated as the proportion of unfilled grains to the total number of grains from 10 sampled panicles per plot. TGWT in grams was obtained by counting 1000 grains from each plot using a seed counter (Elmor C1, Biotronic Bharat, India) and weighing on an electronic balance (Ohaus Scout Pro Model 502 AC, China). The GY was determined by harvesting all panicles in each plot. The panicles were threshed and winnowed to remove chaff. The weight of the grains was adjusted to 14% moisture content and was expressed in tonnes per hectare (t ha^{-1}).

5.2.3.4 Data Analysis

The performance of each cross and parent was determined through the analysis of variance using the REML procedure of GenStat 24th edition (Payne et al., 2017). Means separation was performed using the Fishers' unprotected least significant difference (LSD) procedure at a 5%

probability level. Separate ANOVAs were conducted for each location and later combined data analysis were calculated across locations after a test for homogeneity of variance was conducted. Parents and crosses were considered as fixed effects, while replication was considered as a random effect in computing the ANOVA for combining ability effects. Griffing's (1956) diallel method 2, model 1, was used to estimate the GCA and SCA effects as:

$$Yij = \mu + gi + gj + sij + \frac{1}{bc}\Sigma k\ eijkl.$$

Where Yij = observed value of the cross between parent i and j; μ = the population (general) mean; gi and gj = GCA effects of i^{th} and j^{th} parents, respectively; sij = SCA of the cross between parents i and j; eijkl = environmental effect associated with $ijkl^{h}$ individual observation in k^{th} replication. The GCA for each parent was calculated as described by Acquaah (2012):

$$GCA_P = X_P - \mu.$$

The SCA effects of the crosses were computed from the formula:

$$SCA_X = X_X – E\ (X_X) = X_X – [GCA_P + \mu];$$

Where GCA_P = general combining ability effect of the parent; X_P = Mean of the parent; μ = Overall mean of all crosses; SCA_X = specific combining ability of the parent in the cross; X_X = observed mean value of the cross; E (X_X) = expected values of the cross basing on the GCA of the parent.

Baker's ratios were also computed to estimate the relative importance of additive and non-additive gene action in the expression of traits using Baker's general predicted ratio (GPR) as follows:

$$GCA/SCA = 2MSGCA / (2MSGCA+MSSCA)$$

Where MSGCA = mean square for GCA and MSSCA = mean square for SCA.

A ratio of $>$ 0.5 implies that GCA is more important than SCA in the inheritance of the character and a ratio of $<$ 0.5 implies that SCA is more important than GCA in the inheritance of the character (Baker, 1978).

5.3 Results

5.3.1 Analysis of variance and mean performance

The mean squares and significant tests among genotypes revealed that NT, NPP, NGP, PFG, TGWT and GY were significantly ($P \leq 0.05$) affected by genotype × site interaction effects (Table 5.4). The genotypes exhibited significant differences for all assessed traits. Genotype performance also varied across the sites for all the traits except for the RYMV reaction.

Table 5.4. Mean squares and signfcant tests for 10 traits among 10 parents and 45 F2 crosses of rice evaluated in three locations in Tanzania

Sources of variation	DF	DFL	NT	PH	NPP	PL	NGP	PFG	TGW	RYMVD	GY
Site	2	1694.03***	208.37***	863.30***	90.87***	64.95***	3493.17***	416.9***	3.65*	0.18ns	6.61***
Rep (Site)	2	85.92***	33.27***	1760.60***	3.68ns	7.71*	17.55ns	19.82ns	2.37*	0.1ns	0.1ns
Block (Rep)	20	8.00ns	4.81**	129.9ns	4.23**	4.14***	257.45***	25.91ns	1.99***	0.24ns	0.2*
Genotype	54	441 33***	6.43***	342.2***	4.99***	5.98***	1649.19***	477.17***	6.87***	26.81***	0.97***
Genotype × Site	108	6.949ns	3.29*	76.9ns	3.47**	2.19ns	236.45***	45.23***	1.47***	0.45ns	0.32***
Error	102	9.33	1.98	104	1.95	1.67	81.4	25.93	0.82	0.83	0.11

DF = degrees of freedom; DFL= Days to 50% flowering; NT= number of tillers/plant; NPP = number of panicles/plant; PH= plant height; PL= panicle length; NGP = number of grains/panicle; PFG = percentage filled grains/panicle; TGW= thousand grain weight; RYMVD = rice yellow mottle virus disease reaction; GY = grain yield; * = $P < 0.05$; ** = $P < 0.01$; *** = $P < 0.001$; ns, non-significant.

Mean values, coefficients of variation (CVs) and list significant differences (LSDs) of the genotypes evaluated at three locations are presented in Table 5.5.The shortest DFL of 77 days was recorded at Ifakara. The earliest flowering crosses across all the test sites were Lunyuki × Mwangaza, Mwangaza × Salama M-19 and IRAT 256 × Mwangaza. The genotypes attained the highest average NT and NPP of 12.3 and 10.9 at Ifakara, respectively. Crosses, IRAT 256 × Zambia and IRAT 256 × Lunyuki exhibited the highest tillering capacity and NPP. Crosses, Salama M-57 × Salama M-55, Zambia × SARO and IRAT 256 × SARO were the tallest genotypes, while SARO × Gigante was the shortest with a mean PH of 97.4 cm. The means for PL were 23, 23.4 and 24.6 cm at Ifakara, Ilonga and Mkindo sites, respectively. Crosses, Zambia × SARO and Rangimbili x Salama M-55 had the longest panicles across sites. The parental genotypes, such as Zambia, Salama M-55 and Rangimbili had higher PL value, while the shortest was recorded for the parent SARO. Mean NGP values of 122.3, 125.8 and 125.4 were recorded at the Ifakara, Ilonga and Mkindo sites, in that order. Across the three sites, the best crosses for NGP were Rangimbili x Salama M-55, Rangimbili × Gigante, Gigante × Salama M-19 and Salama M-55 × Gigante. The parents, Salama M-55 and Gigante had the highest and lowest NGP, respectively. Crosses, Gigante × Salama M-19, Rangimbili × Mwangaza, Salama M-57 × Lunyuki and IRAT 256 × Mwangaza had the highest PFG across sites. Parents, Mwangaza, Salama M-55, Salama M-19 and Salama M-57 were the best combiners for PFG across sites. Mean TGW of 34.3 and 34.0 g were achieved for crosses, Rangimbili × Mwangaza and Lunyuki × Mwangaza, respectively. The panel included resistant and susceptible genotypes with RYMVD scores ranging between 1 and 7 with an overall mean score of 3.5. Parental lines, Salama M-57, IRAT 256, Lunyuki, Mwangaza, Salama M-55, and Salama M-19 were highly resistant to RYMVD. There were nine crosses that exhibited RYMV resistance with severity

ratings of 1. Highly resistant crosses, included Salama M-57 × IRAT 256, Salama M-57 × Lunyuki, Salama M-57 x Mwangaza, Salama M-57 × Salama M-19, IRAT 256 × Rangimbili, IRAT 256 × Mwangaza, Rangimbili × Mwangaza, Lunyuki × Mwangaza and SARO × Salama M-55. The genotypes exhibited wide variation in GY productivity ranging between 2.2 and 5.6 t ha^{-1}. The overall mean grain yield was 3.7 t ha^{-1} with the Ilonga site having the highest mean value of 3.8 t ha^{-1}. Across the three sites, crosses, Salama M-57 × IRAT 256, Rangimbili × Salama M-55, Rangimbili × Gigante, Gigante × Salama M-19, Salama M-55 × Gigante, and IRAT 256 × Rangimbili had mean grain yield of > 4.0 t ha^{-1}. The parental lines, Salama M-19, Salama M-57, Salama M-55 and IRAT 256 had the highest means for grain yield, producing 4.0 t ha^{-1} each.

Table 5.5. Means for agronomic traits and RYMVD reaction of 10 parental genotypes and 45 F_2 crosses of rice evaluated in three sites in Tanzania

Genotypes	DFL			NT			PH			NPP			PL		
	Ifa	Ilo	Mk	Ifa	Ilo	Mk	Ifa	Ilo	Mk	Ifa	Ilo	Mk	If	Ilo	Mk
							Crosses								
Salama M-57 × IRAT 256	76	78	78	11	12	9	113.6	114.5	113.6	9	11	8	22.3	21.5	24.8
Salama M-57 × Rangimbili	73	76	79	11	9	12	129.1	115.4	120.6	10	7	11	26.2	23.4	24.9
Salama M-57 × Zambia	69	77	81	13	10	10	125.2	118.3	109.6	11	8	9	24.9	24.0	25.0
Salama M-57 × Lunyuki	72	79	78	10	9	9	111.8	120.0	109.6	9	9	8	22.3	22.3	22.6
Salama M-57 × SARO	71	74	76	15	13	9	115.9	118.8	116.8	15	10	9	22.8	22.4	24.4
Salama M-57 × Mwangaza	66	69	73	12	8	11	117.1	120.3	120.0	11	8	10	23.2	24.6	24.9
Salama M-57 × Salama M-55	69	77	80	11	11	12	134.1	109.0	114.6	10	9	11	23.8	22.6	24.9
Salama M-57 × Gigante	74	81	81	14	11	12	110.6	115.9	116.4	13	9	8	23.0	23.1	24.9
Salama M-57 × Salama M-19	75	81	78	10	10	9	122.7	113.6	120.1	10	9	9	22.9	21.1	24.5
IRAT 256 × Rangimbili	62	69	71	13	10	9	109.8	106.6	107.8	12	10	9	21.5	21.0	21.4
IRAT 256 × Zambia	73	76	76	14	16	11	130.6	127.0	127.9	12	14	10	23.4	23.5	24.1
IRAT 256 × Lunyuki	76	78	78	13	15	12	114.4	110.3	113.4	12	14	12	23.7	20.1	22.2
IRAT 256 × SARO	74	82	79	14	12	11	119.9	108.5	167.6	11	10	10	21.8	22.5	23.6
IRAT 256 × Mwangaza	63	68	72	10	11	8	107.2	111.8	108.5	10	10	8	20.4	23.2	21.9
IRAT 256 × Salama M-55	65	70	73	11	11	10	119.0	118.0	118.1	10	10	10	22.8	22.6	24.4
IRAT 256 × Gigante	62	71	74	15	12	10	111.2	108.8	113.9	13	11	10	21.5	21.9	23.9
IRAT 256 × Salama M-19	75	79	81	14	10	10	120.6	113.1	116.7	13	11	9	21.0	21.1	21.9
Rangimbili × Zambia	83	88	88	13	10	10	131.5	120.0	123.1	12	10	10	23.6	24.5	26.0
Rangimbili × Lunyuki	72	76	80	12	10	8	116.5	116.4	119.8	10	9	8	24.1	23.2	24.0
Rangimbili × SARO	65	72	76	12	13	12	104.5	103.5	114.2	9	12	11	21.3	21.1	24.2
Rangimbili × Mwangaza	64	68	73	10	9	8	106.9	98.7	99.3	8	9	6	21.5	20.9	21.6
Rangimbili × Salama M-55	82	88	92	10	10	11	131.1	126.1	113.0	8	9	10	25.8	24.7	25.2
Rangimbili × Gigante	81	87	86	12	11	10	104.3	113.0	115.5	10	10	9	22.7	23.5	24.6

Table 5.5. Continued

Genotypes	DFL			NT			PH			NPP			PL		
	Ifa	Ilo	Mk	Ifa	Ilo	Mk	Ifa	Ilo	Mk	Ifa	Ilo	Mk	If	Ilo	Mk
							Crosses								
Rangimbili × Salama M-19	76	83	87	12	9	9	126.6	114.4	120.6	11	7	8	24.8	21.7	24.7
Zambia × Lunyuki	73	74	77	12	10	11	127.2	121.1	130.9	11	8	11	24.7	22.9	24.5
Zambia × SARO	76	83	86	14	12	8	132.9	126.2	125.9	12	11	8	23.7	25.1	27.2
Zambia × Mwangaza	62	73	73	12	8	9	125.3	109.5	119.8	11	7	8	22.0	23.9	25.2
Zambia × Salama M-55	114	117	116	10	8	9	112.9	110.4	123.0	8	7	8	23.0	24.5	26.9
Zambia × Gigante	77	81	81	14	12	9	124.8	112.4	118.0	13	9	9	23.0	26.0	24.9
Zambia × Salama M-19	62	73	76	15	11	10	126.6	118.1	116.7	10	9	9	25.2	22.7	22.8
Lunyuki × SARO	76	83	87	13	12	11	119.7	111.8	114.8	12	11	10	24.8	21.0	24.1
Lunyuki × Mwangaza	63	67	69	10	11	9	111.2	107.3	116.4	8	9	9	21.8	22.7	23.9
Lunyuki × Salama M-55	70	78	82	11	11	12	127.3	121.6	120.9	10	9	11	23.4	22.0	24.0
Lunyuki × Gigante	72	80	80	12	11	8	122.2	113.2	116.3	11	10	8	23.0	23.2	24.2
Lunyuki × Salama M-19	71	78	81	12	11	9	114.3	108.1	115.5	8	10	9	22.2	20.2	22.9
SARO × Mwangaza	75	81	81	14	11	10	109.7	113.9	115.0	12	9	10	22.7	23.9	23.8
SARO × Salama M-55	64	68	71	11	13	9	106.2	94.7	103.4	9	12	8	22.6	21.8	25.5
SARO × Gigante	85	90	93	12	12	13	99.8	94.8	97.5	11	10	11	22.4	21.6	23.5
SARO × Salama M-19	72	82	82	13	11	10	103.0	114.6	116.4	12	9	10	21.6	22.9	25.4
Mwangaza × Salama M-55	66	72	75	12	10	9	130.5	114.6	112.7	10	8	9	23.3	23.4	24.5
Mwangaza × Gigante	65	75	77	13	12	8	120.1	116.8	109.8	11	10	8	23.2	25.4	24.9
Mwangaza × Salama M-19	64	67	70	11	8	8	111.7	115.1	114.3	10	7	8	21.6	24.0	25.6
Salama M-55 × Gigante	74	86	89	11	12	9	119.8	117.6	116.4	10	9	8	22.1	24.7	24.9
Salama M-55 × Salama M-19	68	72	75	13	11	10	126.1	114.3	122.0	10	10	9	23.8	23.9	24.5
Gigante × Salama M-19	81	86	87	12	9	9	109.3	109.2	108.9	10	8	9	21.6	24.6	24.9
							Parents								
Salama M-57	76	81	84	12	8	8	124.4	118.3	118.5	11	7	8	21.8	23.1	23.9
IRAT 256	73	74	78	14	11	9	116.9	107.6	101.3	13	10	8	25.8	19.7	20.7

Table 5.5. Continued

Genotypes	DFL			NT			PH			NPP			PL		
	Ifa	Ilo	Mk	Ifa	Ilo	Mk	Ifa	Ilo	Mk	Ifa	Ilo	Mk	If	Ilo	Mk
							Parents								
Rangimbili	95	94	97	12	9	10	138.2	132.3	127.7	10	7	10	23.9	25.6	26.1
Zambia	92	97	100	10	8	10	117.3	123.1	127.7	8	6	9	23.3	25.2	27.7
Lunyuki	69	75	77	14	11	9	147.1	117.2	123.2	13	10	9	25.1	22.2	23.6
SARO	87	94	97	11	12	10	107.1	99.0	94.5	10	10	9	20.4	21.6	22.5
Mwangaza	62	66	69	15	10	11	107.7	102.4	96.9	14	9	5	21.9	20.9	25.2
Salama M-55	80	90	92	10	13	9	133.1	120.7	125.1	9	11	8	25.1	25.8	25.1
Gigante	84	94	90	14	11	10	105.7	93.3	92.3	14	10	10	22.8	22.9	24.1
Salama M-19	106	103	106	12	14	8	119.9	102.3	112.6	11	12	8	23.6	21.6	24.3
Mean	77.1	82.7	84.8	12.3	10.5	8.9	119.9	112.8	113.0	10.9	9.1	8.4	23.0	23.4	24.6
LSD	1.56	7.23	7.8	2.04	3.15	3.23	18.59	13.29	27.68	2.07	2.7	3.54	3.2	2.02	2.53
CV (%)	1.04	4.48	4.72	8.2	14.41	16.78	7.7	5.78	11.75	9.45	14.08	19.36	6.84	4.33	5.12

Table 5.5. Continued

Genotypes	NGP			PFG			TGWT			RYMVD			GY		
	Ifa	Ilo	Mk	Ifa	Ilo	Mk	Ifa	Ilo	Mk	Ifa	Ilo	Mk	Ifa	Ilo	Mk
							Crosses								
Salama M-57 × IRAT 256	141	159	140	91.2	98.7	91.2	32.5	30.5	32.5	1	1	1	4.3	5.3	4.5
Salama M-57 × Rangimbili	128	135	130	94.6	91.1	94.6	30.5	30.5	30.5	3	3	3	3.7	3.8	3.8
Salama M-57 × Zambia	128	131	104	79.1	65.1	68.6	31.0	32.5	32.0	5	5	5	3.3	3.0	2.5
Salama M-57 × Lunyuki	69	116	76	94.0	98.0	94.0	33.0	32.0	31.0	1	1	1	3.6	4.7	3.8
Salama M-57 × SARO	117	112	95	89.7	93.2	92.2	32.0	31.0	32.0	3	3	3	3.9	4.4	3.9
Salama M-57 × Mwangaza	86	151	123	85.2	97.2	91.7	34.0	32.5	32.0	1	1	1	2.7	5.2	3.7
Salama M-57 × Salama M-55	87	111	95	80.2	71.7	78.7	33.0	31.5	32.5	5	5	5	3.0	3.7	3.4
Salama M-57 × Gigante	110	119	112	73.9	83.2	78.4	31.0	32.5	32.0	5	5	5	3.5	4.1	3.7

Table 5.5. Continued

Genotypes	NGP			PFG			TGWT			RYMVD			GY		
	Ifa	Ilo	Mk	Ifa	Ilo	Mk	Ifa	Ilo	Mk	Ifa	Ilo	Mk	Ifa	Ilo	Mk
							Crosses								
Salama M-57 × Salama M-19	75	110	97	84.5	98.0	86.5	32.5	32.5	32.5	1	1	1	3.8	4.2	3.5
IRAT 256 × Rangimbili	139	150	138	93.2	98.2	93.2	33.0	32.0	32.0	1	1	1	3.9	4.7	3.9
IRAT 256 × Zambia	122	123	121	82.3	78.8	82.3	32.5	31.0	32.0	5	5	5	3.1	3.0	3.0
IRAT 256 × Lunyuki	122	105	121	86.5	73.0	77.0	32.0	30.5	31.5	5	5	5	3.8	3.3	3.6
IRAT 256 × SARO	101	108	115	75.2	78.2	84.7	31.0	30.0	31.0	5	5	5	3.2	3.5	3.7
IRAT 256 × Mwangaza	96	140	128	92.8	98.3	94.3	35.0	33.5	33.0	1	1	1	3.6	4.5	3.9
IRAT 256 × Salama M-55	80	92	94	53.6	71.1	78.1	32.0	32.0	32.0	5	5	5	2.5	3.3	3.3
IRAT 256 × Gigante	103	140	139	74.3	84.3	77.8	33.0	32.0	32.0	5	3	3	3.3	4.2	3.9
IRAT 256 × Salama M-19	95	94	109	74.0	68.9	70.8	31.5	30.0	33.0	7	7	7	2.8	2.8	2.9
Rangimbili × Zambia	126	126	127	61.7	70.2	76.7	32.5	30.0	30.0	5	5	5	3.4	3.3	3.7
Rangimbili × Lunyuki	126	133	128	84.4	90.9	84.4	31.5	30.0	30.5	3	3	3	2.9	3.8	2.7
Rangimbili × SARO	89	109	106	74.7	79.7	84.7	31.0	30.5	31.0	5	5	5	3.4	3.8	3.8
Rangimbili × Mwangaza	99	133	107	95.2	97.8	93.2	34.0	34.0	35.0	1	1	1	2.9	4.5	3.1

Rangimbili × Salama M-55	146	156	142	74.1	96.1	83.6	32.5	31.0	30.5	3	3	3	4.2	5.6	4.0
Rangimbili × Gigante	143	148	146	87.9	97.9	94.4	31.5	34.0	32.5	3	3	3	4.0	5.1	4.7
Rangimbili × Salama M-19	83	92	80	64.0	76.0	76.5	32.5	31.5	31.0	5	5	5	2.2	3.6	3.5
Zambia × Lunyuki	70	97	109	59.8	77.8	72.8	31.0	29.0	30.0	5	5	5	2.4	3.6	3.4
Zambia × SARO	100	104	110	68.6	68.6	78.6	29.0	33.0	32.0	7	7	7	3.1	3.0	3.4
Zambia × Mwangaza	85	113	92	82.4	95.9	84.9	33.5	31.5	33.0	3	3	3	3.3	3.6	3.5
Zambia × Salama M-55	135	136	136	71.2	78.2	86.2	31.0	33.5	32.5	5	5	5	3.8	3.8	4.0
Zambia × Gigante	111	114	102	69.4	69.4	71.9	32.5	33.0	33.5	7	5	7	3.3	3.4	3.3
Zambia × Salama M-19	86	95	89	70.0	78.5	71.5	32.0	30.5	30.5	5	5	5	2.5	3.3	2.4
Lunyuki × SARO	126	113	134	78.0	74.0	79.0	32.0	31.5	32.0	5	5	5	3.8	3.3	3.8
Lunyuki × Mwangaza	127	123	121	91.5	97.2	90.5	35.0	33.0	34.0	1	1	1	3.8	4.4	3.6
Lunyuki × Salama M-55	83	142	139	84.7	92.7	85.7	32.5	31.5	32.5	3	3	3	2.9	4.0	4.1

Table 5.5. Continued

Genotypes	NGP			PFG			TGWT			RYMVD			GY		
	Ifa	Ilo	Mk	Ifa	Ilo	Mk	Ifa	Ilo	Mk	Ifa	Ilo	Mk	Ifa	Ilo	Mk
							Crosses								
Lunyuki × Gigante	122	137	111	94.4	93.9	88.9	33.0	32.5	32.0	3	3	3	3.6	4.4	3.4
Lunyuki × Salama M-19	83	148	91	89.1	97.3	89.1	32.5	30.0	33.0	3	1	3	3.3	4.9	3.4
SARO 5 × Mwangaza	100	86	93	81.0	88.0	86.0	30.0	31.0	31.5	3	3	3	3.2	3.8	3.8
SARO 5 × Salama M-55	135	139	124	88.6	98.3	87.8	31.0	29.0	31.5	1	1	1	3.8	4.5	3.7
SARO 5 × Gigante	137	116	138	83.2	75.7	85.7	30.0	33.0	31.5	5	5	5	3.8	3.4	3.8
SARO 5 × Salama M-19	101	98	116	76.6	94.6	81.6	30.5	30.0	30.0	5	3	3	3.2	3.2	3.9
Mwangaza × Salama M-55	97	108	96	68.8	68.3	66.3	33.5	33.5	32.5	6	5	5	3.2	3.4	2.5
Mwangaza × Gigante	110	107	93	77.3	65.3	65.8	34.0	33.0	34.0	5	7	7	3.4	3.2	2.3
Mwangaza × Salama M-19	78	127	126	89.8	92.8	89.3	35.5	34.0	32.0	3	1	1	2.5	3.8	3.6
Salama M-55 × Gigante	144	144	144	89.8	85.8	85.8	33.5	33.5	33.5	3	3	1	4.4	4.3	4.3
Salama M-55 × Salama M-19	130	136	132	84.7	84.2	84.7	32.0	32.5	32.0	1	1	1	2.9	3.4	3.0
Gigante × Salama M-19	142	147	146	95.4	96.9	97.4	33.0	32.5	33.5	3	3	3	4.5	4.7	4.4
							Parents								

Salama M-57	111	111	112	96.2	98.3	93.7	31.5	31.0	32.5	1	1	1	4.5	4..3	4.6
IRAT 256	113	117	115	94.5	98.0	92.5	31.0	31.0	32.0	1	1	1	3.8	4.5	4.2
Rangimbili	118	129	126	67.3	70.8	67.3	30.5	29.0	29.5	7	7	7	3.3	3.0	3.3
Zambia	142	134	131	78.0	71.0	76.5	34.5	33.5	32.0	7	7	7	3.9	3.0	3.5
Lunyuki	120	125	122	95.8	98.3	88.3	29.5	29.0	30.0	1	1	1	3.5	3.8	3.6
SARO	126	105	113	54.1	72.1	74.1	29.0	31.5	31.5	7	5	5	3.3	3.0	3.7
Mwangaza	132	133	136	96.5	98.4	96.5	36.0	36.0	32.5	1	1	1	3.5	3.6	3.7
Salama M-55	165	175	172	96.1	98.6	96.1	32.0	35.0	32.5	1	1	1	3.7	5.7	4.4
Gigante	93	91	99	65.9	60.9	65.9	34.0	35.0	34.5	5	7	7	2.7	2.8	3.1
Salama M-19	161	154	157	95.1	98.1	95.1	31.0	31.8	32.0	1	1	1	5.4	4.4	4.1
Mean	122.3	125.8	125.4	83.6	85.4	83.3	32.4	32.5	32.1	3.6	3.4	3.5	3.6	3.8	3.7
LSD (5%)	16.88	18.40	19.62	9.58	11.12	10.29	2.06	1.62	1.80	0.98	1.53	1.64	0.61	0.74	0.70
CV (%)	7.39	7.32	8.19	5.79	6.41	6.05	3.14	2.51	2.77	13.56	21.43	23.03	8.84	9.28	9.6

DFL= days to 50% flowering; NT= number of tillers/plant; PH = plant height; NPP = number of panicles/plant; PL= panicle length; NGP = number of grains/panicle; PFG = percentage filled grains/panicle; TGW = thousand grain weight; RYMVD = rice yellow mottle virus disease reaction; GY = grain yield; LSD = least significance difference; CV = coefficient of variation; Ifa = Ifakara, Ilo = Ilonga; Mk = Mkindo.

5.3.2 Combining ability effects of parents and their crosses

The GCA and SCA variances for RYMVD reaction and assessed agronomic traits were significant (P≤0.001) (Table 5.6). The GCA and SCA effects for most agronomic traits exhibited marked variability across the test sites exhibited by their significant interaction with the site variance. In contrast, the GCA and SCA effects for RYMVD were not affected by site variance.

5.3.3 General combining ability effects of the parents

The GCA effects for agronomic traits and RYMVD reaction varied widely among the parental lines (Table 5.7). Lines with negative GCA effects for RYMVD reaction scores and DFL are ideal for developing RYMVD resistant and early flowering cultivars, in that order. Parents, Mwangaza, IRAT 256, Lunyuki and Salama M-57 had low negative GCA effects for DFL. SARO, IRAT 256, Gigante and Salama M-57 had high positive GCA for NT and NPP. Gigante and SARO recorded with low negative GCA effects for PH. Parents, Zambia and Salama M-55 had high positive GCA effects for PL. Further, Salama M-55, Rangimbili and Salama M-57 had large positive GCAs for NGP. Positive GCAs were also observed for the PFG on Salama M-57, Mwangaza, and Lunyuki. The parents, IRAT 256, Mwangaza and Gigante had the highest GCA effects for TGW. Negative GCAs for RYMVD reaction were observed for parents, Mwangaza, Lunyuki, Salama M-57, Salama M-19, IRAT 256 and Salama M-55. Parental lines, Salama M-57, IRAT 256, and Salama M-19 exhibited the highest GCA effects for GY, making them suitable candidates for GY improvement.

Table 5.6. Mean squares and significant tests of general and specific combining ability effects for agronomic traits and RYMVD reaction across three sites in Tanzania

Sources of variation	DF	DFL	NT	PH	NPP	PL	NGP	PFG	TGW	RYMVD	GY
Site	2	1694.03***	208.37***	863.32***	90.87***	64.95***	3493.17***	416.90***	3.65**	0.05ns	6.61***
Rep(site)	3	192.59***	66.94***	1180.31***	29.39***	6.25***	14.13***	13.38***	1.99ns	0.10ns	0.15ns
GCA	9	1258.56***	17.28***	1013.74***	14.79***	24.49***	2072.20***	1172.52***	36.37***	57.31***	1.80***
SCA	45	438.71***	4.77***	283.51***	4.14**	3.47**	2253.02***	514.30***	3.60***	14.19***	1.62***
GCA × site	18	11.24ns	6.22***	65.34ns	7.14***	5.19***	399.08***	61.65***	3.54***	0.50ns	0.54***
SCA × site	90	7.37ns	3.7*	92.76ns	3.55**	2.12ns	260.04***	46.48***	1.52***	0.30ns	0.32***
Error	162	9.43	2.63	103.57	2.31	1.96	75.2	25.05	0.90	0.44	0.11
Baker's Ratio		0.7	0.8	0.8	0.8	0.9	0.5	0.7	0.9	0.8	0.5

DF= degrees of freedom; DFL= Days to 50% flowering; NT= number of tillers/plant; PH = plant height; NPP=number of panicles/plant; PL= panicle length; NGP = number of grains/panicle; PFG = percentage filled grains/panicle; TGW = thousand grain weight; RYMVD = rice yellow mottle virus disease; GY = grain yield; * = $P < 0.05$; ** = $P < 0.01$; *** = $P < 0.001$; ns, non-significant.

Table 5.7. General combining ability effects for yield and yield components and RYMVD reaction of 10 parental genotypes across three sites

Genotype	DFL	NT	PH	NPP	PL	NGP	PFG	TGW	RYMVD	GY
Salama M-57	-1.92***	0.34**	1.76*	0.26*	0.08ns	4.88***	4.60***	-0.12ns	-0.73***	0.33***
IRAT 256	-4.46***	0.60***	-0.61ns	0.74***	-1.00***	0.14ns	1.01**	2.15**	-0.34***	0.25***
Rangimbili	2.21***	-0.42***	2.18**	-0.39***	0.30ns	5.74***	-1.01**	-0.64***	0.27***	0.01ns
Zambia	4.93***	-0.12ns	5.69***	-0.24*	1.03***	-2.63***	-7.88***	-0.05***	1.66***	-0.34***
Lunyuki	-2.71***	0.16ns	2.80***	0.26*	0.29**	2.17***	3.55***	-0.57***	-0.76***	-0.01ns
SARO	2.00***	0.81***	-4.31***	0.60***	-0.43***	-5.38***	-3.56***	-0.96***	0.86***	-0.06*
Mwangaza	-8.37***	-0.75***	4.13***	-0.58***	-0.23***	-3.70***	4.29***	1.39***	-1.09***	-0.10***
Salama M-55	2.55***	-0.27*	3.03***	-0.35**	0.70***	11.82***	0.16ns	0.31***	-0.23***	0.17ns
Gigante	2.88***	0.42***	-5.75***	0.36**	0.12ns	2.49***	-3.49***	0.91***	0.91***	0.04ns
Salama M-19	2.89***	-0.10ns	-0.67ns	-0.15***	-0.26**	-1.43***	2.33***	-0.12ns	-0.56***	0.22**

DFL= Days to 50% flowering; NT= number of tillers/plant; PH= plant height; NPP=number of panicles/plant; PL= panicle length; NGP = number of grains/panicle; PFG = percentage filled grains/panicle; TGW = thousand grain weight; RYMVD = rice yellow mottle virus disease; GY = grain yield; * = $P < 0.05$; ** = $P < 0.01$; *** = $P < 0.001$; ns, non-significant.

5.3.4 Specific combining ability effects of crosses

The crosses exhibited marked variation based on the SCA effects for the assessed traits (Table 5.8). The families, including Zambia × Salama M-19, SARO × Salama M-55, Salama M-55 × Salama M-19 and Rangimbili × SARO, had low negative SCA effects for DFL in a desirable direction. Salama M-57 × SARO, IRAT 256 × Zambia, IRAT 256 × Lunyuki and Zambia × Salama M-19 had large positive SCA effects values for NT. Crosses, SARO × Salama M-55, Rangimbili × Mwangaza, IRAT 256 × Rangimbili and SARO × Gigante recorded low SCA effects for PH. The highest positive SCA effects for NPP were obtained from the crosses, IRAT 256 × Zambia, IRAT 256 × Lunyuki, Salama M-57 × SARO and Rangimbili × Zambia. Crosses, Zambia × SARO, Mwangaza × Gigante and Salama M-57 × Rangimbili had high positive SCA effects for PL. The crosses, Salama M-57 × IRAT 256, Gigante × Salama M-19, Rangimbili × Gigante, IRAT 256 × Rangimbili and Lunyuki × Mwangaza had large positive SCA effects for NGP. Likewise, SARO × Gigante, Lunyuki × SARO, Rangimbili × Salama M-55, Salama M-57 × Rangimbili and Salama M-55 × Gigante had high positive SCA estimates for NGP. Rangimbili × Gigante, Gigante × Salama M-19, SARO × Salama M-55, IRAT 256 × Rangimbili recorded with high positive SCA effects for PFG. Crosses, Rangimbili × Mwangaza, Lunyuki × Mwangaza, Lunyuki × SARO and IRAT 256 × Rangimbili had positive SCA effects for TGW. Crosses, SARO × Salama M-55, IRAT 245 × Rangimbili, Rangimbili × Gigante, Rangimbili × Mwangaza and Salama M-57 × IRAT 256 had low negative SCA effects for RYMVD. Rangimbili × Gigante, Gigante × Salama M-19, Rangimbili × Salama M-55, Salama M-57 × IRAT 256, IRAT 256 × Rangimbili and Mwangaza × Salama M-19 had high positive SCA effects value for GY. Crosses, IRAT 256 × Mwangaza, Lunyuki × Mwangaza, Zambia × Gigante and Zambia × Salama M-55 also had high SCA effects for GY.

Table 5.8. Specific combining ability effects for agronomic traits and RYMVD reaction among 45 crosses assesed in three sites in Tanzania

Crosses	DFL	NT	PH	NPP	PL	NGP	PFG	TGW	RYMVD	GY
Salama M-57 × IRAT 256	5.17***	-0.57ns	-3.19ns	-1.04*	0.39ns	33.23***	4.56**	0.09ns	-1.45***	0.56***
Salama M-57 × Rangimbili	-2.85**	0.56ns	1.80ns	0.35ns	1.04*	11.79***	6.31***	-0.75*	-0.07ns	-0.15ns
Salama M-57 × Zambia	-5.89***	0.36ns	-5.68ns	0.25ns	0.11ns	10.17**	-9.24***	-0.00ns	0.55ns	-0.60***
Salama M-57 × Lunyuki	2.58*	-1.38*	-6.69ns	-1.08*	-0.77ns	-24.08***	3.65*	0.68ns	-1.04***	0.12ns
Salama M-57 × SARO	-4.80***	1.94***	3.78ns	1.45**	0.15ns	0.08ns	7.16***	0.73*	-0.65***	0.21ns
Salama M-57 × Mwangaza	1.07ns	0.38ns	5.52ns	0.67ns	1.00ns	10.35**	-1,00ns	-0.45ns	0.30ns	0.07ns
Salama M-57 × Salama M-55	-3.68***	0.91ns	-1.50ns	0.83ns	-0.40ns	-27.40***	-11.37***	0.14ns	2.43***	-0.70***
Salama M-57 × Gigante	-1.01ns	-0.14ns	2.36ns	-0.10ns	0.07ns	-1.95ns	-6.09***	-0.96**	0.63*	-0.17***
Salama M-57 × Salama M-19	-1.51ns	-0.75ns	1.75ns	-0.21ns	-0.39ns	-18.02***	-0.74**	0.73*	-0.57*	-0.36***
IRAT 256 × Rangimbili	-8.97***	-0.27ns	-9.43**	0.23ns	-1.40**	18.11***	11.35***	1.11***	-2.45***	0.47***
IRAT 256 × Zambia	-4.01***	1.93***	7.48*	1.75***	0.24ns	6.32*	4.50**	0.02ns	-0.84***	-0.33***
IRAT 256 × Lunyuki	5.96***	1.72**	-5.42ns	1.56**	-0.11ns	-0.32ns	-9.19***	0.03ns	2.57***	-0.12ns
IRAT 256 × SARO	2.40*	-0.15ns	20.97***	-0.43ns	0.68ns	-4.94ns	-1.58ns	-0.23ns	0.96***	-0.16ns
IRAT 256 × Mwangaza	2.28*	-0.94ns	-2.04ns	-0.78ns	-0.34ns	7.02*	6.32***	0.58ns	-1.09***	0.42***
IRAT 256 × Salama M-55	-7.30***	-0.47ns	0.02ns	-0.39ns	0.16ns	-41.50***	-17.04***	-0.17ns	2.05***	-0.82***
IRAT 256 × Gigante	-7.97***	0.42ns	1.73ns	0.40ns	-0.10ns	6.53*	-2.21ns	-0.43ns	0.24ns	0.06ns
IRAT 256 × Salama M-19	1.36ns	0.08ns	2.12ns	0.53ns	-0.82ns	-17.54***	-15.62***	-0.24ns	3.71***	-0.85***
Rangimbili × Zambia	0.81ns	0.64ns	1.05ns	1.31*	-0.03ns	4.88ns	-5.12***	-0.49ns	-0.45ns	0.14ns
Rangimbili × Lunyuki	-2.22*	-0.47ns	-3.36ns	-0.68ns	0.35ns	7.24*	0.49ns	-0.14ns	-0.04ns	-0.55***
Rangimbili × SARO	-11.44***	0.92ns	-6.40ns	0.58ns	-1.06*	-17.38***	0.81ns	0.41ns	0.35ns	0.05ns
Rangimbili × Mwangaza	-4.07***	-1.23*	-12.37***	-1.00ns	-2.13***	-7.39*	8.59***	1.57***	-1.70***	-0.06ns
Rangimbili × Salama M-55	4.18***	-0.04ns	2.24ns	0.05ns	0.84ns	12.09***	1.94ns	-0.35ns	0.43ns	0.76***
Rangimbili × Gigante	1.02ns	-0.14ns	-1.45ns	0.01ns	-0.22ns	19.09***	14.45***	0.39ns	-2.37***	0.91***
Rangimbili × Salama M-19	-1.48ns	-0.56ns	3.08ns	-0.66ns	0.28ns	-37.42***	-12.69***	0.41ns	1.77***	-0.58***
Zambia × Lunyuki	-6.26***	0.06ns	1.95ns	0.31ns	-0.10ns	-21.31***	-8.99***	-1.39***	0.57*	-0.19ns

Table 5.8. Continued

Crosses	DFL	NT	PH	NPP	PL	NGP	PFG	TGW	RYMVD	GY

Zambia × SARO	-3.48***	-0.20ns	10.99**	0.41ns	1.34**	-5.65ns	-0.07ns	0.33ns	0.96***	-0.08ns
Zambia × Mwangaza	-5.78***	-0.21ns	0.67ns	0.01ns	-0.51ns	-15.11***	7.88***	-0.68*	-1.09***	0.26*
Zambia × Salama M-55	3.31***	-1.49**	-9.25*	-1.50**	-0.34ns	8.30**	2.74ns	0.07ns	0.05ns	0.34***
Zambia × Gigante	-6.36***	0.65ns	2.51ns	0.49ns	0.08ns	-8.97***	-1.90ns	0.14ns	-0.43ns	-0.00ns
Zambia × Salama M-19	-15.86***	1.27**	-0.48ns	0.09ns	-0.60ns	-24.12***	-4.65**	-0.84*	0.38ns	-0.56***
Lunyuki × SARO	4.15***	0.32ns	1.01ns	0.37ns	0.60ns	13.53***	-6.44***	1.16***	1.38***	0.05ns
Lunyuki × Mwangaza	-0.80ns	-0.10ns	-2.97ns	-0.58ns	-0.09ns	17.69***	1.74ns	1.34***	-0.68***	0.41***
Lunyuki × Salama M-55	-1.55ns	0.58ns	1.49ns	0.61ns	-0.68ns	-6.45*	0.49ns	0.41ns	0.46ns	-0.13ns
Lunyuki × Gigante	-1.39ns	-0.79ns	4.24ns	-0.68ns	0.25ns	4.50ns	8.81***	0.15ns	-0.68**	0.13ns
Lunyuki × Salama M-19	-2.22*	-0.35ns	-5.42ns	-0.77ns	-1.08*	-7.42**	2.42ns	0.51ns	-0.54*	0.22**
SARO × Mwangaza	6.81***	0.78ns	5.39ns	0.65ns	0.72ns	-16.27***	0.79ns	-1.61***	-0.29ns	0.15ns
SARO × Salama M-55	-15.28***	-0.65ns	-13.22***	-0.38ns	-0.34ns	7.88*	11.45***	-0.86**	-3.15***	0.28*
SARO × Gigante	6.06***	0.36ns	-8.54**	0.01ns	-0.60ns	14.87***	5.07**	-0.46ns	-0.29ns	0.06ns
SARO × Salama M-19	-4.94***	-0.25ns	0.33ns	0.20ns	0.59ns	-6.38*	2.03ns	-0.77*	-0.15ns	-0.14ns
Mwangaza × Salama M-55	-1.57ns	0.47ns	4.43ns	0.28ns	-0.15ns	-25.81***	-20.16***	-0.55ns	3.13***	-0.69***
Mwangaza × Gigante	-0.57ns	0.52ns	9.53*	0.08ns	1.21*	-13.47***	-14.83***	-0.64ns	2.32***	-0.24***
Mwangaza × Salama M-19	-5.73***	-0.91ns	2.54ns	-0.66ns	0.83ns	-2.78ns	0.49ns	0.55ns	-0.20	0.46***
Salama M-55 × Gigante	-0.82ns	-0.36ns	4.70ns	-0.56ns	-0.34ns	11.27***	7.01***	0.27ns	-0.54*	-0.69***
Salama M-55 × Salama M-19	-11.98***	0.81ns	2.51ns	0.55ns	0.23ns	4.26ns	-1.41***	-0.03ns	-0.73**	-0.69***
Gigante × Salama M-19	0.35ns	-0.79ns	-0.39ns	-0.80ns	0.42ns	25.75***	14.20***	0.20ns	-0.87***	0.85***

DFL= Days to 50% flowering; NT= number of tillers/plant; PH= plant height; NPP = number of panicles/plant; PL= panicle length; NGP = number of grains/panicle; PFG = percentage filled grains/panicle; TGW = 1000 grain weight; RYMVD = rice yellow mottle virus disease reaction; GY = grain yield; * = P < 0.05; ** = P < 0.01; *** = P < 0.001; ns, non-significant.

5.4 Discussion

5.4.1 Analysis of variance and mean performance of genotypes

The significant differences for agronomic traits and RYMVD reaction among the parental lines and their progenies (Table 5.4) indicated the existence of adequate genetic variability for establishing a successful rice-breeding programme. The observed variation is underpinned by differences in genetic constitution among the parental lines, which may have evolved from different gene pools, and provided useful segregants among the F_2 progenies. The parental lines included landraces, accessions and varieties. The genetic groups have different characteristics, which gave rise to the observed variation in the F_2. For instance, landraces are known to be segregating at numerous loci, while varieties are the products of careful and deliberate selection that makes them very different from landraces (Kumbhar et al., 2015).

Genetic variability for agronomic traits such as DFL, NT, PH, NPP, NGP, TGW and GY has been also reported in previous studies. For instance, Akanksha and Jaiswal (2019) found significant genotypic variation among rice accessions evaluated in Bangladesh. Munganyinka et al. (2016) reported significant genetic variation for RYMV disease reaction and agronomic traits in rice in Uganda. Zhang et al. (2019) reported significant variation in yield and its contributing traits in newly developed rice genotypes in China. In the present study, across sites variability was not detected for the RYMVD reaction among the test genotypes (Table 5.4). This was attributed to the high disease pressure found at each of the test sites, ensuring even levels of RYMVD infection and disease development. This is contrast to Kouassi et al. (2005) and Joseph et al. (2011), who reported that RYMV disease infection and disease development were affected by the test environments. Nine crosses were highly resistant to RYMV disease, with scores of 1.0, indicating that they possessed high RYMV resistance, possessing the *RYMV1* or *RYMV2* genes, or new resistance gene(s). Ndjiondjop et al. (1999) and Thiemele et al. (2010) reported that the *RYMV1* and *RYMV2* genes were responsible for resistance to RYMV disease in most African rice varieties. Families such as Salama M-57 × IRAT 256, Salama M-57 × Lunyuki, Salama M-57 × Salama M-19, IRAT 256 × Rangimbili, IRAT 256 × Mwangaza, Rangimbili × Mwangaza, Lunyuki × Mwangaza and SARO × Salama M-55 were identified as new sources of RYMV resistance genes for breeding.

There were significant genotype × location interaction effects for agronomic traits across three test sites. For instance, crosses Rangimbili x Salama M-55 and Salama M-57 had the higher mean values for GY and NGP, respectively, at the Ilonga site than the Ifakara and Mkindo sites. This was probably because different rainfall and temperature conditions existed between the three locations, the highest rainfall being at the Ilonga site. René et al. (2016) reported that

rainfall and air temperature affected rice yield responses in the Guinea Savannah Zone. Yield components are quantitative traits and their expressions are affected by the genotype x environment interactions. Bashir et al. (2018) evaluated rice genotypes in Nigeria and reported significant environmental influence on agronomic performance. The significant genotype × environment interaction effects provide opportunities to identify genotypes with specific or broad adaptation. Some of the crosses, such as Salama M-57 × IRAT 256, Rangimbili × Salama M-55, Rangimbili × Gigante, Salama M-55 × Gigante, and Gigante × Salama M-19, performed well at all three sites with a high level of stability. Such genotypes would be ideal for developing cultivars with broader yield stability across sites. There is an opportunity to select transgressive segregants amongst the new families. The cross, Salama M-57 × IRAT 256 yielded better than the parents across test environments, indicating superior genetic combinations.

Selection of superior rice genotypes across sites should target multiple traits, including NT, PL, NGP, PFG and TGW, aiming to increase adaptability to biotic and abiotic stresses. Crosses, such as Salama M-57 x IRAT 256, Salama M-19 x Gigante, Rangimbili x Gigante, Rangimbili x Salama M-55, Salama M-55 x Gigante, IRAT 256 x Rangimbili and IRAT 256 x Mwangaza exhibited desirable grain yield and RMVD resistance, which make them suitable candidates for breeding.

5.4.2 Combining ability effects and gene action

Establishment of a successful breeding programme depends on the magnitude of the combining ability effects of parents and trait heritability to the offspring. The study found significant GCA and SCA effects for all the traits, indicating that both additive and non-additive gene actions condition the inheritance of RYMV resistance and the tested agronomic traits. This implies that crosses and recurrent selection programs can be used to exploit both additive and non-additive gene action to enhance grain yield in rice. The GCA effects indicate that selection of high performing parents would contribute to the generation of superior crosses for cultivar development. Previous studies reported the significance of both additive and non-additive gene action in the expression of agronomic and yield traits (Dar et al., 2014; Mulbah et al., 2015; Munganyinka et al., 2016; Malemba et al., 2017). The GCA and SCA effects exhibited variability across sites showing that environmental variance influenced the ability of parents to pass favourable traits to their offspring. The high GCA: SCA ratios calculated for all the traits in this study indicated that additive gene effects were preponderant over non-additive gene effects, and therefore a recurrent selection approach would be effective for trait improvement. Similarly, Yuga et al. (2018) found that additive gene action was preponderant for several agronomic traits, including DFL, NT, NGP, PFG and GY. Other studies have reported that additive gene action

has a predominant role in the inheritance of agronomic traits and RYMVD resistance in rice (Hasan et al., 2015; Munganyinka et al. 2016; Akanksha and Jaiswal, 2019; Zewdu, 2020).

5.4.3 General combining ability effects of parents for RYMV resistance and agronomic traits

The selection of parents based on *per se* performance does not always result in producing superior crosses (Falconer and Mackay, 1996; Simmonds and Smartt, 1999). The combining ability effects of parents are useful in selecting parents that can potentially improve target traits in the offspring. Selection is often based on mean performance in one or several environments but *per se* performance may not always result in the generation of superior crosses. Genotypes such as Mwangaza, IRAT 256 and Lunyuki exhibited negative GCA effects for DFL, indicating that they had the genetic potential to reduce the average number of days to flowering. Early flowering has significant benefits by escaping terminal drought stress if seasonal rains end early. Similar negative GCAs for DFL and PH were also reported in Bangladesh (Akter et al., 2010) and Uganda (Zewdu, 2020).

Gigante, SARO and Mwangaza were selected as good combiners for PH displaying negative GCA effects for shortness. Ahmadikhah and Marufinia (2016) and Shavrukov et al. (2017) reported negative GCA effects for PH. Therefore, significant negative GCA effects for DFL and PH are useful for the development of early dwarf varieties that are preferred in rainfall-constrained environments to escape potential drought stress. Parents, SARO and IRAT 256 were identified as good combiners for NT and NPP, expressing higher and positive GCA effects. Significant positive GCA effects for NT and NPP have been reported by Akter et al. (2010). The parental lines exhibited high and positive GCA effects for PL, NGP. PFG and TGW, suggesting their desirability for improvement of yield-related components because of their greater contribution of these traits to high grain yield. Previous studies reported the significance of using parents with high and positive GCA effects for the improvement of agronomic traits (Raju et al., 2014; Malemba et al., 2017).

Parental lines with negative scores for RYMVD were considered the best combiners for increasing RYMV resistance. According to Bokmeyer et al. (2009), negative GCA and SCA effects are desirable for disease resistance. The parents, Mwangaza, Lunyuki, Salama M-57, Salama M-19, IRAT 256 and Salama M-55 were selected as good combiners for RYMVD resistance due to their negative GCA effects. These genotypes may possess the *RYMV1 and RYMV2* genes or novel gene(s) responsible for RYMV resistance. These could be useful as sources of RYMV resistance in developing new cultivars suitable under Tanzanian growing

conditions. Munganyinka et al. (2016) reported significant GCA effects for resistance to RYMV. Salama M-57, IRAT 256 and Salama M-19 had high and positive GCA effects for grain yield, suggesting that the selected parental genotypes were good general combiners for grain yield by contributing favourable alleles. Previous studies also reported good general combiners for yield and yield traits in rice genotypes (Yuga et al., 2018). These genotypes could be regarded as good sources of additive genes for grain yield improvement (Zewdu, 2020).

5.4.4 Specific combining ability effects

A high value of the SCA effect of a cross for a particular trait reflects the contribution of non-additive gene action. This genetic parameter is particularly important for hybrid breeding (Acquaah 2012). Crosses with significant SCA effects in the desired direction would warrant further field evaluation to identify the best segregants. Crosses with high SCA effects are important targets for selection of transgressive segregants (Rajput and Kandalkar, 2018). The crosses, Zambia x Salama M-19, SARO x Salama M-55, Salama M-55 x Salama M-19 and Rangimbili x SARO were selected for their negative SCA effect for DFL. These are early flowering genotypes that reduce exposure to terminal drought. Salama M-57 x SARO, IRAT256 x Zambia, IRAT 256 x Lunyuki and Zambia x Salama M-19 were selected with better NT.

Negative SCA effects are required for PH to develop short stature cultivars. Thus, the crosses, SARO x Salama M-55, Rangimbili x Mwangaza, IRAT 256 x Rangimbili and SARO x Gigante were selected to develop ideotypes with medium height plants. Crosses, IRAT 256 x Zambia, IRAT 256 x Lunyuki and Salama M-57 x SARO were selected for their desirable SCA effects for NPP. Further, crosses, Zambia x SARO, Mwangaza x Gigante and Salama M-57 x Rangimbili were best specific combiners for PL. Salama M-57 x IRAT 256, Gigante x Salama M-19, Rangimbili x Gigante and IRAT 256 x Rangimbili had superior NGP values, supported by their high and positive SCA effects. Rangimbili x Gigante, Gigante x Salama M-19, SARO x Salama M-55 and IRAT 256 x Rangimbili were selected with better PFG scores. Crosses, Rangimbili x Mwangaza, Lunyuki x SARO, and IRAT 256 x Rangimbili were the best specific combiners for TGW.

The best crosses with favourable expression of RYMV resistance were SARO x Salama M-55, IRAT 245 x Rangimbili, Rangimbili x Gigante, Rangimbili x Mwangaza and Salama M-57 x IRAT 256. These crosses had significantly lower and negative SCA effect for RYMVD possessing *RYMV* resistance genes (Table 5.7). The families, Rangimbili x Gigante, Gigante x Salama M-19, Rangimbili x Salama M-55 and Salama M-57 x IRAT 256 were good specific combiners for grain yield because they expressed high SCA effects for grain yield. In a previous study

(Malemba et al., 2017) superior SCA effects were reported for grain yield in rice varieties. Crosses selected with desirable SCA effects for RYMVD, GY, PFG and NGP can be used to generate new rice varieties. The families, Rangimbili x Gigante and Salama M-57 x IRAT 256 are recommended for further breeding or production in RYMV endemic agro-ecologies in Tanzania or similar agro-ecologies.

5.5 Conclusions

The present study found marked differences in the performance of the test parents and their families. Significant GCA and SCA effects were detected for the assessed traits. The predominance of additive gene effects for RYMVD resistance and agronomic traits in the present breeding populations suggested that rice improvement could best be achieved through gene introgression via the recurrent selection method. For RYMVD resistance, the parental lines, Mwangaza, Lunyuki, Salama M-57, Salama M-19, IRAT 256 and Salama M-55 had negative GCA effects. The families, SARO × Salama M-55, IRAT 245 × Rangimbili, Rangimbili × Gigante and Rangimbili × Mwangaza had negative SCA effects for RYMVD. These parents and hybrids were therefore selected for RYMV resistance breeding. The crosses, Rangimbili × Gigante, Gigante × Salama M-19 and Rangimbili × Salama M-55 were selected due to their desirable SCA effects for enhanced GY. The selected parents and families are useful genetic resources for further breeding or production in RYMV endemic agro-ecologies.

References

Acquaah, G. 2012. Principles of Plant Genetics and Breeding, 2nd ed. Wiley-Blackwell, Oxford.

Ahmadikhah, A., and A. Marufinia. 2016. Effect of reduced plant height on drought tolerance in rice. Biotechnology 6:221. doi: 10.1007/s13205-016-0542-3.

Akanksha, B., and H.K. Jaiswal. 2019. Combining ability studies for yield and quality parameters in basmati rice (*Oryza sativa* L.) genotypes using diallel approach. Electronic Journal of Plant Breeding 10 (1): 9-17.

Akter, A., M.J. Hasan, H. Begum, M.U. Kulsum, and M.K. Hossain. 2010. Combining ability analysis in rice (*Oryza sativa* L.). Bangladesh Journal of Plant Breeding Genetics 23(2): 07-13.

Atera, E.A., F.N. Onyancha, and E.B.O. Majiwa. 2018. Production and marketing of rice in Kenya: Challenges and opportunities. Journal of Development and Agricultural Economics 10: 64-70.

Baker, R.J. 1978. Issues in diallel analysis. Crop Science 18 (4): 533-536.

Balamurugan, P., and V. Balasubramanian. 2017. Challenges and opportunities for increasing rice production in sub Saharan Africa. Journal of International Agriculture 4: 1-10.

Bashir, M., B.O. Ehirim, A.T. Maji, S.O. Bakare, A. Isong, and A. Odoba. 2018. Evaluation of yield components and genotype x environment interaction on grain yield of submergence lowland rice genotypes. International Journal of Pure and Applied Bioscience 6(3): 623-630. doi: 10.18782/2320-7051.6725.

Bokmeyer, J.M., S.A. Bonos, and W.A. Meyer. 2009. Inheritance characteristics of brown patch resistance in tall fescue. Crop Science 49: 2302-2308.

Chuwa, C.J., R.B. Mabagala, and S.O.W.M. Reuben. 2015. Pathogenic variation and molecular characterization of *Pyricularia oryzae*, causal agent of rice blast disease in Tanzania. International Journal of Science and Research 4(11): 1131-1139.

Dar, S., A.G. Rather, M.A. Ahanger, N.R. Sofi, and S. Talib. 2014. Gene action and combining ability studies for yield and component traits in rice (*Oryza sativa* L.): A review. Journal of Plant and Pest Science 1(3): 110-127.

Duku, C., A.H. Sparks, and S.J. Zwart. 2016. Spatial modeling of rice yield losses in Tanzania due to bacterial leaf blight and leaf blast in a changing climate. Climatic Change 135 (3): 569-583.

Falconer, D.S., T.F.C. Mackay, and R. Frankham. 1996. Introduction to Quantitative Genetics, Volume 7, 4th Ed. Prentice Hall, Harlow.

Food and Agriculture Organization of the United Nations (FAO). 2017. Rice market monitor. Database available from: http://faostat.fao.org/default.aspx.

Food and Agriculture Organization of the United Nations (FAO). 2018. Crop prospects and food situation. Quarterly global report. Database available from: http//www.fao.org/3/a-w732-4e.pd.

Food and Agriculture Organization of the United Nations (FAOSTAT). 2015. Database available from: http://faostat.fao.org/default.aspx.

Griffing, B. 1956. Concept of general and specific combining ability in relation to diallel crossing systems. Australian Journal of Biological Sciences 9: 463-493.

Hasan, M.J., U.K. Kulsum, L.F. Lipi, and A.K.M. Shamsuddin. 2013. Combining ability studies for developing new rice hybrids in Bangladesh. Bangladesh Journal of Botany 42(2): 215-222.

Hubert, J., A. Luzi-Kihupi, E. Hébrard, and H.J.F. Lyimo. 2016. Farmers' knowledge and perceptions of rice yellow mottle virus in selected rice growing areas in Tanzania. International Journal of Science and Research 5(2): 549-559.

Hubert, J., J.F.H. Lyimo, and A. Luzi-Kihupi. 2017. Pathogenic variation and occurrence of multiple resistance-breaking rice yellow mottle virus strains in Tanzania. African Journal of Biotechnology 16 (24): 1331-1337.

International Rice Research Institute (Eds.). 2002. Standard Evaluation System for Rice. Genetic Resources Center, IRRI, Manila, Philippines. 5th edition, 27 p.

International Rice Research Institute. 2013. Standard Evaluation system for Rice, 4th Edition, Manila, International Rice Research Institute, Philippines.

Joseph, A., D.B. Olufolaji, F.E. Mwilene, A. Onasanya, M.M. Omale, R.O. Onasanya, and Y. Sere. 2011. Effect of leaf age on rice yellow mottle virus severity chlorophyll content with mechanical inoculation and vector transmission method. Trends in Applied Sciences Research 6(12): 1345-1351.

Kanyeka, Z.L., N.J.M. Kibanda, and J. Mbapila.1996. *Rice yellow mottle virus* in Tanzania. In*: Proceedings of the Potential and Constraints for Improvement of Rice Cultivation Seminar.* 11 - 15 March 1996, Kilimanjaro Agricultural Training centre (KATC), Moshi, Tanzania, pp. 17 - 22.

Kouassi, N.K., P. N'Guessan, L. Albar, C.M. Fauquet, and C. Brugidou. 2005. Distribution and characterization of rice yellow mottle virus: A threat to African farmers. Journal of Plant Disease 89: 124-133. doi: 10.1094/PD-89-0124.

Kumbhar, S.D., P.L. Kulwal, J.V. Patil, C.D. Sarawate, A.P. Gaikwad, and A. S. Jadhav. 2015. Genetic diversity and population structure in landraces and improved rice varieties from India. Rice Science 22(3): 99-107.

Lamo, J. 2010. Genetic Studies on Drought Tolerance and Grain Shattering in Rice. PhD thesis, University of KwaZulu-Natal, Pietermaritzburg, 205 p.

Luzi-Kihupi, A., J.A. Zakayo, H. Tusekelege, M. Mkuya, N.J.M. Kibanda, K.J. Khatib, and A. Maerere. 2009. Mutation breeding for rice improvement in Tanzania in: Induced plant Mutations in the Genomics Era. Food and Agriculture Organization of the United Nations, Rome, pp. 385-387.

Malemba, G.M., F.M. Nzuve, J.M. Kimani, M.F. Olubayo, and J.W. Muthomi. 2017. Combining ability for drought tolerance in upland rice varieties at reproductive stage. Journal of Agricultural Science 9(3): 138-150.

Mogga, M.L., J. Lamo, G. Asea, and R. Edema. 2010. Inheritance of resistance to rice yellow mottle virus disease in selected rice cultivars in Uganda. Second RUFORUM biennial meeting, Entebbe, Uganda. http://mak.ac.ug/documents/ RUFORUM/ Mogga.pdf.

Mulbah. Q.S., H.A. Shimelis, and M.D. Laing. 2015. Combining ability and gene action of three components of horizontal resistance against rice blast. Euphytica 206: 805-814.

Munganyinka. E., R. Edema, J. Lamo, P. Gibson, and P. Rukundo. 2015. Combining ability for resistance to rice yellow mottle virus disease in interspecific and intraspecific rice genotypes. African Journal of Crop Science 3 (1): 102-107.

Ndjiondjop, M.N., L. Albar, D. Fargette, C. Fauquet, and A. Ghesquière. 1999. The genetic basis of high resistance to rice yellow mottle virus (RYMV) in cultivars of two cultivated rice species. Journal of Plant Disease 83: 931-935.

Ochola, D., and G. Tusiime. 2011. Pathogenicity of rice yellow mottle virus and the potential sources of resistance against the disease in Eastern Uganda. Asian Journal of Plant Pathology 5: 1-15.

Paul, C.P., N.Q. Ng and Ladeinde, Tao. 2003. Mode of gene action of inheritance for resistance to rice yellow mottle virus. African Crop Science Journal 11: 143-150.

Payne, R.W., D.A. Murray, and S.A. Harding. 2017. An Introduction to the GenStat Command Language, 18th edition. VSN International, Hemel Hempstead, UK.

Pinel-Galzi. A., C. Dubreuil-Tranchant, E. Hébrard, C. Mariac, A. Ghesquière, and L. Albar. 2016. Mutations in rice yellow mottle virus polyprotein P2a involved in RYMV2 gene resistance breakdown. Frontiers in Plant Science 7:1–11.

Rajput, S.S., and V.S. Kandalkar. 2018. Combining ability and heterosis for grain yield and its attributing traits in bread wheat (*Triticum aestivum* L.). Journal of Pharmacognosy and Phytochemistry 7(2):113-119.

Raju, C.D., S.S. Kumar, C.S. Raju, and A. Srijan. 2014. Combining ability studies in the selected parents and hybrids in rice. International Journal of Pure and Applied Bioscience 2(4): 271-279.

René, N.K., B. Koné, K.K. Firmin, Z. Florant, T.M. Joachim, Y.K. Albert, D.A. Emanuel, and K. Daouda. 2016. Variations of rainfall and air temperature affecting rain-fed rice growth and yield in a Guinea savanna zone. Journal of Agriculture and Environmental Sciences 5(1): 65-77.

Schlegel, R.H.J. 2010. Dictionary of Plant Breeding. CRC Press, Boca Raton.

Sereme, D., I. Ouedraogo, B.J. Neya, P.E. Zida, N. Yao, and M. Sie. 2016. Screening improved rice varieties (*Oryza spp*) for their resistance /tolerance to rice yellow mottle virus in West Africa. International Journal of Agricultural Innovation Research 5: 481-486.

Shavrukov, Y., A. Kurishbayev, S. Jatayev, V. Shvidchenko, L. Zotova, F. Koekemoer, S. de Groot, K. Soole, and P. Langridge. 2017. Early flowering as a drought escape mechanism in plants: How can it aid wheat production? Frontiers in Plant Science 8: 1950. doi:10.3389/fpls.2017.01950.

Simmonds, N.W., and J. Smartt. 1999. Principles of Crop Improvement, 2nd Ed.. Blackwell Science Ltd, Oxford, UK.

Sprague, G.F., and L.A. Tatum. 1942. General vs. specific combining ability in single crosses of corn. Agronomy Journal 34:923-932.

Srujana, G., B.G. Suresh, G.R. Lavanya, J.B. Ram, and V. Sumanth. 2017. Studies on genetic variability, heritability and genetic advance for yield and quality components in rice (*Oryza sativa* L.). *Journal of Pharmacognosy and Phytochemistry* 6: 564-566.

Suvi, W. T., H. Shimelis, M. Laing, I. Mathew, and A.I.T. Shayanowako. 2019. Assessment of the genetic diversity and population structure of rice genotypes using SSR markers. Acta Agriculturae Scandinavica, Section B-Soil & Plant Science 70: 76-86. doi: 10.1080/09064710.2019.1670859.

Suvi, W.T., H. Shimelis, and M. Laing. 2018. Breeding rice for rice yellow mottle virus resistance in Sub-Saharan Africa: a review, Acta Agriculturae Scandinavica, Section B - Soil & Plant Science 69(2):1-8. doi: 10.1080/09064710.2018.1523454.

Thiemele, D., A. Boisnard, M.N. Ndjiondjop, S. Cheron, Y. Sere, S. Ake, A. Ghesquiere, and L. Albar. 2010. Identification of a second major resistance gene to rice yellow mottle virus, RYMV2, in the African cultivated rice species, *O. glaberrima*. Theoretical and Applied Genetics 121: 169-179.

Traore, V.S.E., M.D. Asante, V.E. Gracen, S.K. Offei, and O. Traore. 2015. Screening of rice accessions for resistance to rice yellow mottle virus. American Journal of Experimental Agriculture 9: 1-12.

Yuga, M.E., J.M. Kimani, P.M. Kimani, M.F. Olubayo, J.W. Muthomi, and F.M. Nzuve. 2018. Combining ability and heterosis for agronomic and yield traits in *indica* and *japonica* rice crosses. Journal of Agricultural Science 10 (12): 92. doi: 10.5539/jas.v10n12p92

Zewdu, Z. 2020. Combining ability analysis of yield and yield components in selected rice (*Oryza sativa* L.) genotypes. Cogent Food and Agriculture 6 (1): 1811594. doi: 10.1080/23311932.2020.1811594.

Zhang, S.L., J.A. Hu, C.D. Yang, L. Haitao, F. Yang, J.H. Zhou, B.K. Samson, C. Boualaphanh, L.Y. Huang, G.F. Huang, J. Zhang, W.Q. Huang, D. Tao, D. Harnpichitvitaya, L.J. Wade, and F.Y. Hu. 2019. Genotype by environment interactions for grain yield of perennial rice derivatives (*Oryza sativa* L. /*Oryza longistaminata*) in southern China and Laos. Field Crops Research 207: 62-70.

Zouzou, M., T.H. Kouakou, M. Koné, I. and Souley. 2008. Screening rice (*Oryza sativa* L.) varieties for resistance to rice yellow mottle virus. Scientific Research and Essay 3 (9): 416-424.

CHAPTER 6: GENERAL OVERVIEW OF THE RESEARCH FINDINGS

6.1 Introduction and objectives of the study

Rice (*Oryza sativa* L.) is an important staple food crop for more than half of the world's population. It is the third most preferred food staple after maize (*Zea mays* L.) and wheat (*Triticum aestivum* L.) globally. In East and Central Africa, Tanzania is the second largest rice producer after Madagascar. In Tanzania, rice is the second most important food and cash crop after maize. Despite the increasing importance of rice in Tanzania, its productivity is affected by a multitude of biotic, biotic and socio-economic constraints. Rice yellow mottle virus (RYMV) is among the most important biotic constraints of rice production. RYMV disease causes yield losses ranging from 20 to 100%. Various control strategies such as the use of resistant cultivars, cultural practices and crop protection chemicals have been recommended for the control of the disease. Cultural practices are ineffective because the virus is spread by several agents. RYMV disease management through generic crop protection chemicals is not economic and presents health and environmental hazards. The deployment of varietal resistance against RYMV is economic and environmentally friendly especially for smallholder farmers.

This overview summarises the research objectives and highlights the core findings of the study. The specific objectives of the study were:

i. To assess farmers' perceptions, production constraints and variety preferences of rice production in Tanzania, as a baseline to guide breeding;
ii. To determine the genetic variation among Tanzanian rice germplasm collections based on agronomic traits and resistance to RYMV so as to select useful parents for breeding;
iii. To assess the genetic diversity and population structure of rice genotypes using Simple Sequence Repeat markers to complement phenotypic data, and thereby to select parents for subsequent breeding;
iv. To determine the combining ability effects and gene action conditioning RYMV resistance and agronomic traits in selected parental lines and derived families in rice (*Oryza sativa* L.), and to develop new populations for future rice breeding activities.

6.2 Summary of the major findings

6.2.1. Farmers' perceptions, production constraints and variety preferences of rice in Tanzania to guide breeding

A participatory rural appraisal study was conducted involving 180 participant farmers, combined with focus group discussions with 90 participants in the Mvomero, Kilombero and Kyela districts of Tanzania. Data were collected using a structured questionnaire, focus group discussions and transect walk. The main findings of this study were as follows:

- The majority (53.6%) of the interviewed farmers were rice producers while 16.6% were maize producers. Other crops grown included cassava, sweetpotato, sugarcane and pigeonpea that were produced by less than 5% of farmers each.
- The majority of the farmers (67.2%) used farm-saved seed, followed by a 15% of farmers who purchased seed from agro-dealers, or sourced seed from neighbours (8.9%), research centres (6.7%), local government (1.7%) and NGOs (0.6%).
- About 51.4% of the respondents used landraces, while 25.7% cultivated both landraces and improved cultivars and 22.9% only used improved varieties.
- Scented rice was preferred by almost all the farmers (97%) who were interviewed.
- RYMV disease was identified as the most important constraint of rice production and most famers (92.3%) noted that they experienced severe outbreaks of RYMV in their fields. Other constraints identified by the farmers included insect pests, drought stress, high cost of fertilizers, lack of access to improved varieties, poor soil fertility, bird damage and the limited access to fertilizers.
- The farmer-preferred traits included high grain yield, drought tolerance, disease resistance, marketability and early maturity.

6.2.2. Variation among Tanzania rice germplasm collections based on agronomic traits and resistance to rice yellow mottle virus to select useful parents for breeding

Fifty-four rice genotypes were field evaluated at Ifakara and Mkindo, which are recognized as RYMV hotspots, aiming to select superior genotypes for breeding high yielding and RYMV resistant rice cultivars. The experiments at each site were laid out using a 6 × 9 alpha lattice design with two replications. Phenotypic traits, including days to 50% flowering (DFL), number of tillers per plant (NT), plant height (PH), number of panicles per plant (NPP), panicle length (PL), number of grains per panicle (NGP), percent filled grain (PFG), thousand grain weight (TGW), grain yield (GY); and rice yellow mottle virus disease (RYMVD) reaction, were recorded

and subjected to analysis of variance, correlation and principal component analysis. The core findings of the study were:

- There were significant ($p<0.05$) differences among the genotypes for RYMV resistance and agronomic traits, indicating that there was marked genetic variation for selection.
- Seven genotypes, including Salama M-57, SSD1, IRAT 256, Salama M-55, Mwangaza, Lunyuki, and Salama M-19 with moderate to high RYMV resistance were selected, which will be useful as new sources of resistance gene while SARO, Rangimbili and Mbega were selected for their high GY values that averaged 3.7 t ha^{-1}.
- Positive and significant correlations were detected between GY and NPP, PL, NGP, PFG, and TGW, which would facilitate simultaneous selection for rice yield improvement.
- Principal component analysis identified that difference in NPP, NT, PL, GY, and DFL contributed much of the variation enabling discrimination between the tested genotypes.

6.2.3. Assessment of the genetic diversity and population structure of rice genotypes using SSR markers to complement phenotypic data and select parents

Fifty-four rice genotypes were genotyped using 14 polymorphic simple sequence repeat (SSR) markers to complement agronomic data and to choose parents for breeding. The genetic data based on marker and population structure were subjected to analysis to deduce the genetic parameters including polymorphic information content (PIC), total number of alleles per locus (Na), number of effective alleles per locus (Ne), Shannon's information index (I), observed heterozygosity (Ho), gene diversity (He), inbreeding coefficient (FIS) and the population structure. In addition, molecular variance was conducted to deduce variation among different populations identified in the structure. The key findings were:

- The mean PIC was 0.61 suggesting that there was high allelic diversity among the assessed rice accessions.
- The population structure revealed only two major sub-populations.
- Analysis of molecular variance revealed that only 30% of the variation was attributed to the differences between the populations while variation among individuals within population and within individuals accounted for 47 and 23% of the total variation, respectively.
- The genetic distance among genotypes varied from 0.083 to 1.834.
- Genotypes such as IR56, Mwanza, Salama M-55, Sindano Nyeupe, SARO, Gigante, Mwanza, Lunyuki, Zambia, Rangimbili, IRAT 256, Zambia and Salama M-19 were identified as genetically divergent and complementary for breeding.

6.2.4 Determining combining ability and gene action for RYMV disease resistance andagronomic traits in rice and develop new populations for future breeding

Ten selected parental lines were crossed using a half-diallel mating design without reciprocals to produce 45 first filial (F_1) generation. Forty-five F_2 families and their 10 parents were field evaluated at three locations using a 5 × 11 alpha lattice design replicated twice to select suitable parents, families and develop breeding populations. The core findings of this study were:

- The genotype × site interaction effects were significant ($p<0.05$) for NT, NPP. NGP, PFG, TGW, RYMVD reaction and GY.
- The variance due to general combing ability (GCA) and specific combing ability (SCA) effects were significant for all the traits indicating that both additive and non-additive gene action governed the inheritance of the traits.
- High GCA to SCA ratios for all the traits indicated that additive genetic effects were predominant and introgression through recurrent selection is recommended to exploit the additive gene effects for rice improvement.
- Parental lines, Mwangaza, Lunyuki, Salama M-57, Salama M-19, IRAT 256 and Salama M-55 with low negative GCA effects for RYMV disease were selected for future breeding.
- Crosses, SARO × Salama M-55, IRAT 245 × Rangimbili, Rangimbili × Gigante and Rangimbili × Mwangaza with negative SCA effects for RYMVD are suitable families for enhancing RYMV resistance in rice.
- Crosses, Rangimbili × Gigante, Gigante × Salama M-19 and Rangimbili × Salama M-55 were selected for developing breeding populations due to their desirable SCA effects for GY.

6.3 Implications of the research findings for breeding

- New rice cultivars must possess RYMV resistance, high yielding with good grain quality (e.g. aroma) to meet farmer preferences and adapt to multiple constraints prevalent under smallholder farming systems in Tanzania.
- The selected genotypes with moderate to high RYMV resistance are vital genetic resources for RYMV resistance breeding programmes to develop new varieties in irrigated and rain-fed agro-ecologies.
- The SSR markers would be useful in marker-assisted breeding and the identified genetic populations will enable breeders to design targeted crosses for hybrid development and maintain genetic diversity.
- Presence of both additive and non-additive gene effects for yield and resistance to RYMVD suggest that genetic gain can be realized through hybridization and recurrent

selection strategies in rice breeding program. The preponderance of additive genetic effects for most traits would fix the genetic homeostasis through recurrent selection in the selected families to develop and release new varieties in Tanzania.