

CZECH UNIVERSITY OF LIFE SCIENCES PRAGUE

FACULTY OF TROPICAL AGRISCIENCES

**Morphological and Genetic Diversity, Agronomic Traits of Indigenous
Landraces of Rice (*Oryza sativa* L.) in Myanmar**



Dissertation thesis

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DECLARATION OF AUTHORSHIP

I, Aye Aye Thant, hereby declare that this thesis entitled “**Morphological and Genetic Diversity, Agronomic Traits of Indigenous Landraces of Rice in Myanmar**”, submitted in partial fulfillment of the requirements for the degree of Ph.D. at the Faculty of Tropical AgriSciences, Czech University of Life Sciences Prague, and the work presented in it is entirely my own work. Information derived from the published or unpublished work has been acknowledged in the text and a list of references is given. Furthermore, I declare that no part of this work is being submitted for any other degree to this or any other university.

Prague, April 2022

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LIST OF ABBREVIATIONS

AC1	Auricle Color
AC2	Amylose Content
AD	Awns Distribution
AFLP	Amplified Fragment Length Polymorphism
AMOVA	Analysis of Molecular Variance
BVP	Basic Vegetative Phase
CA	Correspondence Analysis
CAPS	Cleaved Amplified Polymorphic Sequence
CL	Culm Length
CoA	Lemma: Color of Apiculus
CTAB	Cetrimonium bromide/ Cetyltrimethylammonium bromide
CULS	Czech University of Life Sciences Prague
CURE	Consortium for Unfavorable Rice Environments
CV	Coefficient of Variation
DAMD	Directed Amplification of Minisatellite-region DNA
DAR	Department of Agricultural Research
DArT	Diversity Array Technology
DARwin	Dissimilarity Analysis and Representation for windows
DNA	DeoxyriboNucleic Acid
DTH	Days to Heading
ECQ	Eating and Cooking Qualities
ET	Endosperm Type
FG%	Filled Grain %
FLA_E	Flag Leaf Attitude (Early observation)
FLA_L	Flag Leaf Attitude (Late observation)
FTA	Faculty of Tropical AgriSciences
GL	Grain Length
GRET	Group For Research and Technology Exchanges
GW	Grain Width
GWAS	Genome Wide Association Studies
HI	Harvest Index
HYV	High Yielding Variety
ICBA	International Center for Biosaline Agriculture
IRRI	International Rice Research Institute
ISSR	Inter Simple Sequence Repeats
LPC	Lemma and Palea: Color
LPP	Lemma and Palea: Pubescence
LSL	Long Sterile Lemma
LWR	Length-Width Ratio
MoALI	Ministry of Agriculture, Livestock and Irrigation
NGS	Next Generation Sequencing

NPP	No. of Panicles per Plant
NSPP	No. of Spikelets Per Panicle
NTPP	No. of Tillers Per Plant
PA_B	Panicle Attitude of Branches
PA_M	Panicle Attitude of Main axis
PCA	Principal Component Analysis
PGS	Participatory Guarantee System
PIC	Polymorphism Information Content
PL	Panicle Length
PSP	Photoperiod Sensitive Phase
Q	Membership coefficient
QTL	Quantitative Trait Loci
RAD	Restriction site-Associated DNA
RAPD	Random Amplified Polymorphic DNA
RCBD	Randomized Complete Block Design
RFLP	Restriction Fragment Length Polymorphism
SC	Stigma Color
SFP	Single Feature Polymorphism
SLC	Sterile Lemma Color
SNP	Single Nucleotide Polymorphism
SSL	Short Sterile Lemma
SSR	Simple-Sequence Repeats/ Microsatellites
TGW	Thousand Grain Weight
USDA	United States Department of Agriculture
Y	Yield (5 hills) kg

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ABSTRACT – ENGLISH

Myanmar falls within the center of the genetic diversity of many economically important crops, including wild and cultivated rice (*Oryza sativa* L.). The majority of the genetic resources of Myanmar, particularly traditional varieties, and their distribution still remain poorly understood. The present study was conducted to delineate the on-farm varietal diversity and the determinants of farmers' variety choices in the Ayeyarwady delta in southern Myanmar, specifically for rainfed rice cultivation and investigate the morphological and genetic diversity of Myanmar rice genotypes based on important agro-morphological traits and DArTseq markers. For on-farm rice diversity survey, data, such as basic household characteristics, rice varieties grown, farmers' preferences for varietal traits, etc. from 150 randomly selected households, distributed across five townships, were collected through a semi structured questionnaire. The on-farm diversity appeared to be relatively high, particularly rich in local landraces according to the number of varieties grown by the farmers. In total, 39 varieties were identified within the survey area. Only 34% of the interviewed farmers grew high yielding varieties (HYVs) at least on a fraction of their land. Except for high yields, farmers preferred varieties that were resistant to environmental constraints such as resistance to salinity, flooding, and pest infestation. For morphological characterization, a total of 117 rice genotypes involving a collection of currently cultivated rice varieties from farmers' survey in addition to the collections from local gene banks were used. Significant phenotypic variations among rice genotypes were found for 13 quantitative traits and high heritability (H^2) (>70%) was observed for almost all the traits. The standardized Shannon-Weaver diversity index ranged from 0.41 (least polymorphic) to 0.84 (highly polymorphic) with an average of 0.7 among qualitative traits. Cluster analysis separated the different varieties into two major clusters. Two pairs of genotypes (TLNKYAUK and MSEIK, MSWE and KYTUN) clustered together in the genetic distances (0.00) due to their similar qualitative traits. To assess the genetic diversity and population structure of the same rice panel, 7,643 SNPs and 4,064 silicoDArT derived from DArT platform were employed. DArTseq markers revealed genetic variance among the genotypes ranged from 0 to 0.753 in SNPs, and from 0.001 to 0.954 in silicoDArT. Two distinct population groups were identified from SNP data analysis. Cluster analysis with both markers clearly separated traditional Pawsan varieties and modern high yielding varieties. A significant divergence was found between populations according to the F_{st} values (0.737) obtained from the analysis of molecular variance, which revealed 74% genetic variation at the population level. Overall, this study provided evidence of on-farm rice varietal diversity in the study area and the determinants of varietal selection. Moreover, the establishment of a product profile for developing new rice varieties should give priority to farmers' desired attributes and the vital features, such as resistance to environmental stresses or/and preferred qualitative rice properties, rather than solely considering increased yield. Characterization of landraces, particularly farmers' currently cultivated rice varieties in the study area based on agro-morphological traits is important for preliminary evaluation as well as determining the regional landrace distribution and diversity under consideration of adaptability of local environment and farmers' preferences. The findings through DArTseq markers support rice researchers in identifying useful DNA polymorphisms in genes and pinpointing specific genes conferring desirable phenotypic traits

for further genome-wide association studies and parental selection for recombination breeding to enhance rice varietal development and release.

Key Words: Agro-morphological traits, DArTseq markers, genetic diversity, Myanmar, on-farm diversity, traditional rice varieties

ABSTRACT – CZECH

ABSTRAKT – ČESKY

Myanmar patří mezi centra genetické diverzity mnoha hospodářsky důležitých plodin, včetně divoké a kultivované rýže (*Oryza sativa* L.). Většina genetických zdrojů Myanmaru, především tradiční odrůdy, a jejich distribuce jsou stále málo prozkoumány. Tento výzkum byl proveden s cílem popsat diverzitu pěstovaných odrůd nezavlažované rýže a faktory určující výběr odrůd farmáři v deltě Ayeyarwady v jižním Myanmaru a prozkoumat morfologickou a genetickou diverzitu genotypů myanmarské rýže pomocí agromorfologických znaků a DArTseq markerů. Pro výzkum diverzity rýže na farmách byla sesbírána data pomocí polostrukturovaných rozhovorů (základní údaje o domácnosti, pěstované odrůdy rýže, preference zemědělců pro vlastnosti odrůd, atd.) ze 150 náhodně vybraných domácností z pěti správních oblastí. Diverzita na farmách byla relativně vysoká a obzvláště se objevila bohatost místních odrůd v závislosti na počtu pěstovaných odrůd. Celkem bylo ve zkoumané oblasti objeveno 39 odrůd. Pouze 34% farmářů pěstovalo vysoce výnosné odrůdy alespoň na části svých pozemků. Kromě vysokých výnosů farmáři upřednostňovali odrůdy rezistentní vůči enviromentálním vlivům jako zasolení, záplavám a škůdcům. Pro morfologickou analýzu bylo použito 117 genotypů rýže získaných z průzkumu mezi farmáři a doplněných o zástupce ze sbírek místních genových bank. Mezi genotypy byla zjištěna významná fenotypická rozmanitost pro 13 kvantitativních znaků a u téměř všech znaků byl pozorován vysoký index heritability (H^2) (>70%). Standardizovaný Shannon-Weaverův index diverzity se pro kvantitativní znaky pohyboval mezi 0,41 (nejméně polymorfni) a 0,84 (nejvíce polymorfni) s průměrem 0,7. Shluková analýza rozdělila odrůdy do dvou hlavních skupin. Dva páry genotypů (TLNKYAUK a MSEIK, MSWE a KYTUN) se seskupily kvůli velmi podobným hodnotám kvalitativních znaků. Pro stanovení genetické diverzity a populační struktury stejných genotypů rýže bylo použito 7643 SNP a 4064 silicoDArT markerů vygenerovaných platformou DArT. Markery DArTseq odhalily genetické vzdálenosti mezi jednotlivými genotypy v rozmezí 0 až 0,753 pro SNP a 0,001 až 0,954 pro silicoDArT. Analýza SNP dat odhalila dvě odlišné skupiny. Shluková analýza s použitím obou typů markerů jasně rozdělila tradiční Pawsan odrůdy od moderních vysoce výnosných odrůd. Mezi populacemi byl nalezen významný rozdíl vyjádřený hodnotou F_{st} (0,737), získanou z analýzy molekulární variance, která určila 74% genetické variace na úrovni populací. Výsledky této studie poskytly důkazy o diverzitě odrůd rýže na farmách ve zkoumané oblasti a určující faktory pro výběr odrůd. Kromě toho by se při ustanovení produktového profilu pro vývoj nových odrůd rýže měly brát v úvahu priority zemědělců co se týče zásadních atributů, jako odolnost vůči enviromentálním stresorům a upřednostňované vlastnosti, spíše než se pouze soustředit na zvyšování výnosů. Charakterizace místních odrůd na základě agromorfologických znaků, především odrůd, které farmáři v současnosti ve zkoumané oblasti využívají, je důležitá pro předběžné vyhodnocení i pro určení distribuce a diverzity místních odrůd z pohledu adaptability k místnímu prostředí a preferencí zemědělců. Výsledky získané z analýzy DArT markerů slouží vědcům k identifikaci užitečného polymorfismu v genech a k určení specifických genů podmiňujících fenotypické znaky pro

budoucí genomových asociačních studií a výběru rodičovských genotypů pro šlechtění a podporu vývoje nových odrůd rýže.

Klíčová slova: Agromorfologické znaky, DArTseq markery, genetická diverzita, Myanmar, diverzita na farmě, tradiční odrůdy rýže

ABSTRACT – MYANMAR

အကျဉ်းချုပ် - မြန်မာ

မြန်မာနိုင်ငံတွင် စီးပွားရေးအရအရေးပါသော သီးနှံများစွာရှိသည့်အနက် စပါးသည် တစ်ခုအပါအဝင် ဖြစ်ပါသည်။ ဗီဇကွဲစပါးမျိုးပေါင်းများစွာ ပေါ်ကြွယ်လာသည့် နိုင်ငံတစ်နိုင်ငံလည်း ဖြစ်ပါသည်။ မြန်မာနိုင်ငံရှိ စပါးမျိုးအများစု အထူးသဖြင့် ဒေသမျိုးများ၊ ၎င်းတို့၏ ပြန့်နှံ့ပုံ နှင့် မျိုးရိုးဗီဇအရင်းအမြစ်များအား လေ့လာမှတ်တမ်းပြုမှုများမှာ အားနည်းနေဆဲဖြစ်ပါသည်။ ထို့ကြောင့် တောင်သူလယ်သမားများ မိုးစပါးရာသီချိန်၌ လက်ရှိစိုက်ပျိုးနေသည့် စပါးမျိုးပြားများ နှင့် မျိုးရွေးချယ်စိုက်ပျိုးရာတွင် လယ်သမားများ ထည့်သွင်းစဉ်းစားလေ့ရှိသည့် စံသတ်မှတ်ချက်များအား ဆန်းစစ်လေ့လာရန်၊ ပြင်ပရုပ်သွင်လက္ခဏာများ (Agro-morphological traits) နှင့် မော်လီကျူလာမတ်ကာနည်းပညာ (DArTseq markers) များအသုံးပြု၍ ၎င်းစပါးမျိုးများ၏ မျိုးရိုးဗီဇဆိုင်ရာ ကွဲပြားမှုများအား ဖော်ထုတ်ရန် ဤသုတေသနကို လုပ်ဆောင်ခဲ့ပါသည်။ မြန်မာနိုင်ငံ၏အဓိက စပါးစိုက်ပျိုးရာဒေသဖြစ်သော ဧရာဝတီမြစ်ဝကျွန်းပေါ်ဒေသကို သုသေသနဧရိယာ အဖြစ်ရွေးချယ်ခဲ့ပါသည်။ ဧရာဝတီတိုင်းဒေသကြီးအတွင်းရှိ လယ်သမားများစိုက်ပျိုးသော စပါးမျိုးပြားများ အား စစ်တမ်းကောက်ယူရန် မြို့နယ် ၅ ခုမှ အိမ်ထောင်စုပေါင်း ၁၅၀ အားကျပန်းရွေးချယ်ခဲ့ပါသည်။ ယင်းသို့ကောက်ယူရာတွင် သုတေသနအချက်အလက်များဖြစ်သော အခြေခံအိမ်ထောင်စုလက္ခဏာများ၊ စပါးမျိုးအမည်များ၊ လယ်သမားအများစုစိုက်ပျိုးရန် ကြိုက်နှစ်သက်သည့် စပါးမျိုးလက္ခဏာများ စသည့် အချက်အလက်များ ပါဝင်ခဲ့ပါသည်။ ကောက်ယူရရှိသည့် စစ်တမ်းအဖြေများအရ စပါးမျိုးပြားများစွာ စိုက်ပျိုးနေကြသည်ကို တွေ့ရှိရပါသည်။ မျိုးစုစုပေါင်း ၃၉ မျိုးအထိ ကောက်ယူရရှိခဲ့ပြီး အများစုမှာ ဒေသမျိုးများဖြစ်ကြပါသည်။ မျိုးကွဲစုစုပေါင်း၏ ၃၄ ရာခိုင်နှုန်းသာလျှင် အထွက်ကောင်းမျိုးများ (HYVs) ဖြစ်ကြပြီး စိုက်ပျိုးမြေ၏အစိတ်အပိုင်းအနည်းငယ်မျှသာ စိုက်ပျိုးကြကြောင်း သိရှိရပါသည်။ တောင်သူများ သည် အထွက်နှုန်းမြင့်မားသည့် စပါးမျိုးများအပြင် သဘာဝပတ်ဝန်းကျင်ဆိုင်ရာထိခိုက်မှုများ အထူးအားဖြင့် ဆားငန်ရေဝင်ရောက်ခြင်း၊ ရေကြီး/ရေမြုပ်ခြင်း နှင့် ပိုးမွှားကျရောက်မှုဒဏ်များကို ခံနိုင်ရည်ရှိသော စပါးမျိုးများကို စိုက်ပျိုးရန်နှစ်သက်ကြသည်။ စပါးမျိုးများ၏ ရုပ်သွင်လက္ခဏာများအား လေ့လာရန် ဧရာဝတီတိုင်းအတွင်းမှ စုဆောင်းရရှိခဲ့သော စပါးမျိုးပေါင်း ၁၁၇ မျိုးအားအသုံးပြုခဲ့ပါသည်။ ယင်း ၁၁၇ မျိုး တွင် လယ်သမားများလက်ရှိစိုက်ပျိုးနေသည့် စပါးမျိုးများ၊ ရေဆင်းစိုက်ပျိုးရေးသုတေသနဌာနရှိ မျိုးစေ့ သို့လှောင်ဘဏ်မှ စပါးမျိုးများပါဝင်ခဲ့ပါသည်။ စပါးမျိုးများကြားတွင် လေ့လာခဲ့သော ပြင်ပရုပ်သွင်လက္ခဏာ ၁၅ ခုအနက် ၁၃ ခုမှာ သိသာထင်ရှားစွာ ကွဲပြားခဲ့သည်။ လက္ခဏာအားလုံးနီးပါးတွင် မျိုးရိုးအမွေဆက်ခံနိုင်မှု (H^2) ၇၀ ရာခိုင်နှုန်းထိမြင့်မားကြောင်း တွေ့ရှိခဲ့ရသည်။ အရည်အသွေးဆိုင်ရာရုပ်သွင်လက္ခဏာများကြား တွင် စံသတ်မှတ်ထားသော ကွဲပြားမှုအညွှန်းကိန်း (Shannon-Weaver index) သည် ၀.၄၁ (ကွဲပြားမှု အနိမ့်ဆုံး) မှ ၀.၈၄ (ကွဲပြားမှု အမြင့်ဆုံး) အထိရှိပြီး ပျမ်းမျှအားဖြင့် ၀.၇၀ ကွာဟသည်။ ရုပ်သွင်ပြင် လက္ခဏာများပေါ်အခြေခံ၍ မျိုးအုပ်စုခွဲရာတွင် အဓိကအုပ်စုကြီး ၂ ခုခွဲခြားပေးခဲ့ပါသည်။ ဒေသမျိုး များဖြစ်ကြသော သက်လတ်ကြောက် နှင့် ၊ မုတ်ဆိတ်၊ မွေးဆွေ နှင့် ခေါင်ရိုးထွန်း တို့သည် အများစုသော

ရုပ်သွင်လက္ခဏာများတူညီခြင်းကြောင့် မျိုးရိုးဗီဇအကွာအဝေးမရှိဘဲ (၀.၀၀) တွင်တပေါင်းတည်း တွေ့ရှိခဲ့ရပါသည်။ မော်လီကျူးလာမတ်ကာဗီဇအမှတ်အသားလက္ခဏာနည်းပညာအသုံးပြု၍ မျိုးရိုးဗီဇကွဲပြားမှုကို လေ့လာရန် DArTseq ပလပ်ဖောင်းမှရရှိသည့် ၇,၆၄၃ SNPs နှင့် ၄,၀၆၄ silicoDArT မတ်ကာတို့ကို အသုံးပြုခဲ့ပါသည်။ SNPs မတ်ကာအားဖြင့် စပါးမျိုးများကြားမျိုးရိုးဗီဇကွဲလွဲမှု ၀ မှ ၀.၇၅၃ နှင့် silicoDArT အားဖြင့် ၀.၀၀၁ မှ ၀.၉၅၄ ကြား ရှိသည်ကိုတွေ့ရှိခဲ့ရပါသည်။ SNPs မတ်ကာအသုံးပြု၍ မျိုးအုပ်စု ခွဲခြားရာတွင် အုပ်စု ၁ - ပေါ်ဆန်းမျိုးများ နှင့် ဒေသမျိုးများ၊ အုပ်စု ၂ - အထွက်ကောင်းမျိုးများ နှင့် ဒေသမျိုးများ ဟူ၍တွေ့ရပါသည်။ SNPs နှင့် silicoDArT မတ်ကာ ၂ မျိုးလုံးသည် ပေါ်ဆန်းမျိုးများ နှင့် အထွက်ကောင်းမျိုးများကို မျိုးရိုးဗီဇအားဖြင့် မတူညီကြောင်း အုပ်စု ၂ ခုအဖြစ် ရှင်းလင်းစွာခွဲခြားပေးခဲ့သည်။ မော်လီကျူးကွဲလွဲမှုကို ခွဲခြမ်းစိတ်ဖြာသည့်တန်ဖိုးဖြစ်သော Fst တန်ဖိုး (၀.၇၃၇) အရ အုပ်စု ၂ ခုကြား သိသာထင်ရှားသော ကွဲပြားမှုရှိသည်ကို တွေ့ရှိရပြီး ၎င်းတန်ဖိုးသည် အုပ်စု ၂ ခုကြားမျိုးရိုးဗီဇကွဲလွဲမှု ၇၄ ရာခိုင်နှုန်း ရှိကြောင်းကိုယ်စားပြုပါသည်။ ခြုံငုံသုံးသပ်ကြည့်မည်ဆိုလျှင် ဤလေ့လာမှုသည် သုသေသန ဧရိယာအတွင်း စပါးမျိုးပြားများကွဲပြားမှုနှင့် လယ်သမားများ၏ စပါးမျိုးရွေးချယ်စိုက်ပျိုးမှုကို အဆုံးအဖြတ် ပေးသော သွင်ပြင်လက္ခဏာများကို ဖော်ထုတ်ပေးနိုင်ခဲ့ပါသည်။ ထို့အပြင် စပါးမျိုးအသစ်များထုတ်လုပ် ရာတွင် အထွက်နှုန်းတိုးရန်တစ်ခုတည်းကိုသာ ဦးစားပေးစဉ်းစားခြင်းထက် လယ်သမားများကြိုက်နှစ်သက် သော စပါးမျိုးအရည်အသွေးလက္ခဏာများ၊ အရေးပါသောအင်္ဂါရပ်များဖြစ်သည့် ပတ်ဝန်းကျင်နှင့်လိုက်လျော ညီထွေမှုရှိသည့်၊ ရာသီဥတုဒဏ်ခံနိုင်ရည်ရှိသည့် (ဆားငံခံ၊ ရေမြုပ်ခံ ကဲ့သို့သော) အချက်များကိုလည်း ထည့်သွင်းစဉ်းစားသင့်ကြောင်း ဤသုတေသနအဖြေများက ထောက်ပြလျက်ရှိပါသည်။ DArTseq မတ်ကာ များမှတစ်ဆင့် တွေ့ရှိချက်များသည် စပါးသုတေသနပညာရှင်များအတွက် စပါးမျိုးစပ်ရာတွင် အရေးပါသည့် မျိုးရိုးဗီဇကွဲပြားမှု (DNA polymorphisms) ကိုလေ့လာခြင်း၊ ပြင်ပရုပ်သွင်လက္ခဏာများနှင့် ၎င်းတို့ကို ဖြစ်စေသော မျိုးဗီဇများအားချိတ်ဆက်လေ့လာခြင်း (Genome-Wide Association Studies) များတွင်လည်း အထောက်အကူဖြစ်စေမည့်အပြင် မိဘမျိုးများရွေးချယ်ခြင်းအတွက်လည်း အသုံးဝင်စေမည့် သုတေသန အဖြေတစ်ခုပင် ဖြစ်ပါသည်။

အဓိကအချက်များ။ ။ ရုပ်သွင်ပြင်လက္ခဏာများ၊ DArTseq မတ်ကာဗီဇအမှတ်အသားလက္ခဏာနည်းပညာ၊ မျိုးဗီဇကွဲပြားမှု၊ မြန်မာ၊ မျိုးပြားများ၊ ဒေသစပါးမျိုးများ

1. INTRODUCTION

Rice is one of the most widely cultivated cereal crops distributed across the world and serves as a major food source for more than half of the global human population (Xu et al. 2016). Due to the rapid growth of the world's population as well as increasing urbanization and climatic changes, higher or at least sustainable rice yield is urgently required to meet world food demands (Ta et al. 2018). Myanmar is the seventh-largest rice-producing country in the world, with a total rice production of 25.9 million tons in 2018 (FAOSTAT 2020). Rice is grown extensively in the country, and it covers an area of 7.26 million ha and its production reaches 28.1 million t with an average yield of 3.92 t/ha in the growing season 2017-2018 (MoALI 2019). Myanmar has diverse landscapes and geographic variation from the delta area of the Ayeyarwady River in the southern region, to the mountainous areas in the northern region. This landscape diversity resulted in the diverse agricultural system for examples, deep water fields in the delta areas, irrigated and rain-fed paddy fields in plain areas and slash and burn fields in the mountainous areas. Geographic and crop diversity, coupled with diverse traditional agricultural systems, contributes to the diversity of crop genetic resources in Myanmar (Garcia et al. 2003; Yi et al. 2008).

Rice is mostly grown in the Ayeyarwady region, where it spreads through the deltaic watershed of the Ayeyarwady River, the longest river in Myanmar, which flows from the northernmost snowcap of the Himalayan Mountain range to the southern Ayeyarwady delta, ultimately draining into the Bay of Bengal. The richness of fertile alluvial soil and abundant monsoon rainfall in this delta provide vast fertile farmland. These attributes have defined the Ayeyarwady region as the largest "rice bowl" in Myanmar (Oo et al. 2012). The total paddy area in the Ayeyarwady region covers approximately 2 million ha, which is 28% of the total cultivated rice area in Myanmar, and it produces 7.8 million t (MoALI 2019). However, this region is highly vulnerable to the impacts of climatic aberrations evident from frequent saltwater intrusion and flooding. To tolerate such stresses, some unique rice landraces have been nurtured and cultivated by the farmers in this region for decades. However, there is little formal documentation of variety diversity on-farm and farmers' preferences, which are needed to guide rice varietal breeding and development of the Ayeyarwady region. Information about the factors that affect farmers' variety choice and the impact of their choice on varietal diversity is important for programs that are aiming to introduce new modern varieties.

While in the upper Ayeyarwady region, the HYVs occupy 98% of rice fields because of favorable conditions (Subedi et al. 2017); the proportion of such varieties in the lower delta was only 59%, indicating the importance of the traditional varieties to counter the harsh environmental conditions in the lower delta region. Among the most commonly grown traditional varieties, the Pawsan group, which has aroma, grain quality, and eating quality similar to the reputable aromatic rice varieties of the world, namely Basmati of India and Pakistan and Jasmine of Thailand, is of special economic importance on the local market (Myint & Napsintuwong 2016).

Local genetic resources (approximately 7,000 genotypes including Pawsan varieties) are conserved in the seed bank at the Department of Agricultural Research, Ministry of Agriculture, Livestock and Irrigation, Yezin (Wunna et al. 2016). They represent the main sources with which to undertake genetic improvement for the toleration of biotic and abiotic stress. Moreover, identifying the genetic diversity of local varieties compared with improved or introduced varieties will assist the breeding of elite varieties for use in sustainable agriculture. However, in Myanmar, unfortunately, only a small percentage of the available rice genetic resources have been used in past breeding programs. To overcome this bottleneck due to poor or scant information on the genetic characterization of available resources, this study could be a game changer for the development of Myanmar's future rice breeding strategy that involves local diverse resources of paramount importance for climate resilience.

The use of agro-morphological characterization is the traditional method of measuring diversity because farmers identify or distinguish varieties by using some standards such as growth duration, water region, seed size, etc. Farmers select among the plants in the crop population to maintain these desirable characteristics and to increase the prevalence of other valued traits in the population over time. Therefore, evaluation of agronomic traits variation among farmers' handling landrace pools, which was shaped by cultural practices, ecosystems and diverse ethnic groups, may serve as a source for study leading to effective utilization and conservation for region specific (Wunna 2015). A number of studies on genetic diversity using agro-morphological characterization have been conducted in Myanmar and it led to the identification of the phenotypic variability in rice (Aung 2007; Yamanaka et al. 2011; Wunna 2015). Nevertheless, phenotypic characterization subject to the inaccuracy involved in the selection of elite accessions due to the effects of environment and genotype-environment interactions. To reduce inaccuracies in selection procedure and to accelerate breeding efficiency, genomic techniques can be used as a tool of rice breeding program.

Molecular markers to analyze genetic relationships in crops have become increasingly popular since they are more reliable than other phenotypic or biochemical markers (Winter & Kahl 1995; Adhikari et al. 2017). Most studies up to now were conducted on limited sets of resources, using an older generation of markers such as restriction fragment length polymorphism (RFLP), amplified fragment length polymorphism (AFLP) and cleaved amplified polymorphic sequence (CAPS) (Saw et al. 2006), which are seldom used now because of poor marker-trait association information and, only in the recent past, have simple sequence repeat (SSR) markers been used (Aung 2007; Oo et al. 2008; Yamanaka et al. 2011; Thein et al. 2012; Thein et al. 2014; Wunna et al. 2016). SSR markers have been used most extensively to study the genetic diversity of more than 600 Myanmar rice landraces (Thant et al. 2018).

DArT (diversity array technology) markers developed by Jaccoud et al. (2001) are useful for whole-genome profiling of crops without the need for prior sequence knowledge. This unique genotyping tool is characterized by hybridization rather than electrophoretic gel resolution and this helps to improve both throughput and accuracy. It can produce thousands of polymorphic loci in a single assay. DArT has generated two types of markers, SNP and

silicoDArT, over the past decade. DArTseq-based SNPs are codominant markers. SilicoDArT markers are microarray markers that are dominant and scored for the presence or absence of a single allele. Compared to other marker technology, DArT markers have merits in terms of cost effectiveness and time (Kilian et al. 2003). There are only a few studies about genetic structure and diversity in rice using DArTseq markers (Mogga et al. 2018; Adeboye et al. 2020), although many studies have been made on other crops such as barley (*Hordeum vulgare*), rye (*Secale cereale*), bean (*Phaseolus vulgaris*), macadamia (*Macadamia integrifolia*), etc. (Alam et al. 2018).

The present study was designed to assess the on-farm rice varietal diversity in the Ayeyarwady region for monsoon rice growing season and the determinants of farmers' preferences on rice varieties and collect rice germplasm then; to investigate genetic diversity using agro-morphological traits and DArTseq markers involving a collection of currently cultivated rice germplasm from farmers' fields in addition to the collection from local gene bank using the most robust set of markers to obtain repeatable inferences.

2. LITERATURE REVIEW

2.1 Rice production in Myanmar

2.1.1 Myanmar

Republic of the Union of Myanmar is situated in Southeast Asia, sharing borders with China on the north and northeast, India and Bangladesh on the west and northwest, Lao PDR and Thailand on the east and southeast. The country covers an area of 678,500 square kilometers spanning 2,361 km from North to South and 1,078 km from East to West. It has a long coastline of 2,832 km along the Bay of Bengal and Andaman Sea. Myanmar is ethnically diverse; it is a union of eight major ethnic groups and 135 minor sub-groups with their own languages and dialects. In 2021, Myanmar's population is 54.9 million with a population density of 83 people per square kilometer and 31.4 % of the population is urban (Myanmar Population [n.d.](#)).

There are two major climatic regions: tropical and sub-tropical or temperate, generally with three seasons; a cool winter from November to February, a hot summer season in March and April and a rainy season from May to October, dominated by the southwest monsoon (MoNREC [2017](#)). Three distinct topographical areas represent the coastal zone as western ranges, the central dry zone as central plains and the hilly zone as eastern hills (Figure 1) (Lwin [2015](#)). Annual climate patterns, as well as seasonal temperatures and precipitation vary across the country (MoNREC [2016](#)). The regions in the coastal zone receive the highest mean annual rainfall (2,500–5,500 mm) and are prone to flooding. Mean annual rainfall is the lowest in the central dry zone (500–1,000 mm per year) that is prone to extreme heat events and drought. Temperatures in this zone could reach 40–43°C during the hot dry season. The northern hilly region has the lowest mean and maximum annual temperatures. The Eastern and Northern Hilly areas receive the lowest wet season precipitation, and both regions are exposed to heat waves, droughts and floods, as well as landslides.

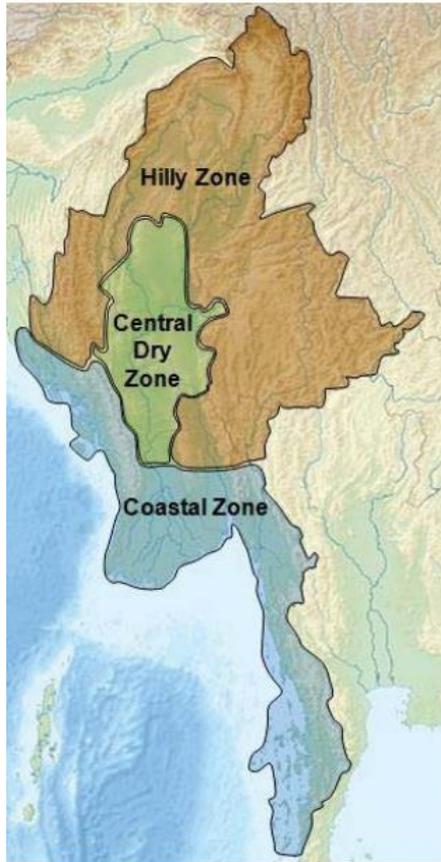


Figure 1. Topographical zones in Myanmar (MoNREC 2016)

2.1.2 Agriculture sector

Myanmar's economy and food security totally rely on agriculture sector including crops, livestock and fishery sub-sectors, accounting for 25.6% of GDP, 24.4% of total export earnings in 2017-2018, and 61.2% of the labor force (MoALI 2019). Current net sown area of agricultural lands occupies 12.1 million ha, accounting for 17.8% of total cultivable land resources. For the area expansion of new agricultural land, the remaining 0.47 million ha of fallow land and 5.54 million ha of cultivable waste land could be developed (MoALI 2018).

Rice, which covers about two-thirds of the total area under cultivation, is the most important crop in Myanmar, requiring large investments, large manpower, and earning the large return from the land. Myanmar has reached self-sufficiency in rice production for domestic consumption and surplus is being exported. The second principal crop are beans and pulses, which have recently become major export crops and are grown on about 4.44 million ha. Oilseed crops (especially groundnut and sesame) are third, grown on 2.67 million ha; production is insufficient to meet national demand and about 200,000 tons of palm oil is imported annually. Currently, oil palm plantations are established at southern coral Thaninthayi division to catch the domestic need. Fruits and vegetables are grown on about 0.5 million ha, principally in highland areas. Other crops, including maize, cotton, rubber,

sugarcane, and tropical fruit crops are also important crop for agriculture sector (MoALI 2019).

2.1.3 Rice production

Myanmar is the world's seven-largest rice producing country and rice is the country's most important crop because of its crucial role in food security, as well as its attribute to the economic viability, political stability, and social prosperity of the country. The major rice-producing regions of Myanmar are Ayeyarwady, Bago and Yangon regions, which make up almost half of the country's harvested rice area. In the growing season 2017-2018, paddy-sown area occupied 7.26 million ha, and its production was 28.1 million t with an average yield of 3.92 t/ha (MoALI 2019). Between 1900 and 1940, Myanmar exported 2 to 3 million t rice annually, up to 70% of national production (Win 1991). In the early 1960s, annual exports were in the range 1.3 to 1.7 million t (United States Department of Agriculture (USDA) data from World Rice Statistics). In recent years, exports have dropped below 1 million t per annum (USDA), as population growth has outpaced productivity improvement (Denning et al. 2013).

According to the cropping calendar in the country, the majority of rice is sown in wet season (monsoon crop from the end of May through November) and followed by growing rice during dry season as the summer crop under irrigation (from December to May) that was introduced in late 1970s to fulfill the need for the increasing population and to increase the peasant income (Oo et al. 2012). Presently, high yielding varieties (HYVs) are cultivated extensively, especially as a summer crop, but farmers are not achieving the yield potential of these varieties as they cannot afford high amounts of inputs (e.g., fertilizer and herbicide). Rice yield rose to 4.1 t/ha in 2010 from about 3 t/ha in 1995 and yield growth was 1.9% per year in 2005-10. Rice yield reached 4 t/ha in 2008 and the stagnation since then could be due to the lower amount of fertilizer applied by rice farmers. Domestic rice consumption is rising gradually with population rate. With the politic transformation during 2011 to 2015, country's recent re-engagement with the international community has set the stage for renewed and reinvigorated attention to develop the agricultural export market and regain the world rice market share (Wunna 2015).

2.1.4 Rice ecosystems

Rice is grown in Myanmar during the wet (June to November) and dry (December to May) seasons. Myanmar's major rice ecosystems include rain-fed lowland rice, irrigated lowland rice, deep water rice and upland rice. There are two dominant rice production systems: rain-fed lowland and irrigated lowland. During the wet season, Myanmar's rainfall in the delta and coastal region is sufficient for growing rice without supplemental irrigation from dams, river and stream diversions or groundwater. Where available, irrigation coupled with drainage structures, improves stability of production, and reduces the risks of flooding and stagnant water (Denning et al. 2013). Nearly 60% of the delta region, including the Ayeyarwady, Bago, and Yangon region of Lower Myanmar, is cultivated with rain-fed rice (GRiSP 2013).

In the dry zone, annual rainfall of 500–1,000 mm is generally inadequate to produce a rain-fed rice crop except in low lying areas with a high water table. Rice grown in the dry zone can be productive when grown under irrigated conditions because of the increased hours of sunshine, especially during the summer season. Upland rice is grown in the hilly areas under a shifting cultivation, however the yields lag behind the paddy field grown rice. Main theme of rice cultivation in upland mountainous area is culture value and regional self-sufficiency. Upland rice is direct seeded into moist soil with the first rains. As much as 2 million ha of upland rice is planted in Myanmar, more than half of which is grown in Shan State (Denning et al. 2013).

2.1.5 Rice landraces

Myanmar has heterogeneous geographical and ecological conditions such as hills and mountains so that it has been recorded as one of the countries, which have been found high in genetic diversity of rice and a part of the crop diversity area (Saw et al. 2006). The genetic diversity of rice landraces has been brought about by natural factors such as various topographic and climatic conditions and by different social and agricultural circumstances. Their diversity is considered to result from adaptation to various natural and agro-ecological environments and to be caused by unconscious selection by farmers for several thousand years and by artificial selection applied through preference for different qualities and ingredients of rice grains (Tun et al. 2005).

Landraces are precious genetic resources, because they contain huge genetic variability which can be used to complement and broaden the gene pool of advanced genotypes and to improve modern crops as they have the ability to exchange their genetic material (Kobayashi et al. 2006). Landraces are also an integral part of cultural heritage due to their close link with the local territory as well as the community with their associated traditional uses, knowledge and habits. Myanmar traditional landraces are still under cultivation by resource poor farmers who practice subsistence farming (Yamanaka et al. 2011). Although the output is lower usually lower yield, these landraces have wide adaptation to local or harsh conditions. For example, in the Ayeyarwady delta, local varieties such as Annarwarbo and Latyonegyi are usually grown for their salinity tolerant ability; Manaw and Madama are grown for their submergence tolerance. They do not need high inputs (fertilizers, herbicides, pesticides) for their growth and are assumed as a harbor of great genetic potential for rice improvement.

Most of Myanmar farmers use their own seeds for reproduction and distribution of planting material is usually done from farmer to farmer. Temporal variability and spatial heterogeneity among landraces can be found especially in farmers' seeds and it depends on the landrace's local adaptability, and farmer selection impact and their cultural practices. For survival of landraces, market-based incentives play an important role to encourage farmers to grow landraces as economically interesting. Therefore, a complex combination of environmental, socio-cultural and economic factors influences upon farmer manageable on-farm varietal diversity (Wunna 2015).

In Myanmar, rice growing farmers in mountainous and hilly regions have better possession of diverse landraces and maintain their richness or evenness of landrace diversity due to their assets of cultivating diverse land types, dispersed plots that are located further away from market centers (Watanabe et al. 2007) and coexistence of diverse tribes. Mountainous and hilly regions farmers know the benefit of diversity richness and appreciate upon it as cultural, social and religious norm. In irrigated lowland and dry-land area, local farmer groups' link to extension service, therefore, government's regional self-sufficiency policy and incentive of irrigation facility negatively influence landrace diversity. Richness of lowland landraces directly relate to irrigation facilities, farmer's choice and market demands while richness of upland landraces is totally related to ethnic diversity, culture and religious facts. Due to overwhelming introduction of modern varieties among lowland rain-fed and irrigated area and awareness and interest of farmers upon those varieties, promising local landraces need to be conserved at both in situ and ex situ level (Wunna 2015).

2.2 The Ayeyarwady delta

The Ayeyarwady River is the longest river in Myanmar. It flows from the northern most snow cap Himalaya mountain range (the peak of Hkakabo Razi) to the southern Ayeyarwady delta and drains to the Bay of Bengal. The richness of fertile alluvial soil and abundant monsoon rainfall at this delta area is providing a wide farmland and contributing to the nation as the biggest rice pot (Oo et al. 2012). The region is vulnerable to extreme weather conditions in the form of storm surges, floods, cyclones and coastal inundation. For example, Cyclone Nargis hit the coast in May 2008 and it was the most devastating cyclone that Myanmar has ever experienced. The Ayeyarwady delta and the eastern part of Yangon were most affected experiencing wind speeds of more than 250 km/h causing loss of livelihoods and homes affecting about 3.2 million people and mortality of 138,373 and bringing catastrophic damage to the region (MOAI 2015). The average annual rainfall is 2,000–3,000 mm, however, water scarcity affects over 50% of the villages, particularly between March and May.

The total paddy area in the region covers around 2 million ha, which is 28% of the total cultivated rice area in Myanmar and produces 7.8 million t. Before 1991-92, mono cropping of rice was the dominant cropping pattern of the Ayeyarwady delta which can make farmers extremely vulnerable to climate-induced shocks. The majority of farmers grow local rice varieties, which have photoperiod-sensitive, long life span (about six to seven months from planting to maturation and harvest) and low yield (SeinnSeinn et al. 2015; MoALI 2019). A summer paddy program was introduced in 1992 by using short-duration high-yielding varieties together with proper irrigation management (MoALI 2019). At present, farmers grow rice on a double-cropping basis, using different varieties with short maturation periods. Many of these are cross-breeds and varieties from the International Rice Research Institute (IRRI).

Significantly, in recent years, climate change shocks rice production of the Ayeyarwady delta due to the alterations of rainfall pattern, intensity of rainfall and sea level rise (SeinnSeinn et al. 2015). Large areas of the delta are subject to flooding ranging in duration

from a few days to two or three months, damaging the thousand acres of rice field leading to significant risks to farmers. Therefore, to combat flooding, farmers in the delta cultivate flood-adapted landraces that can withstand and resume normal growth after flood subsides. Some areas, though declining in importance, are suitable for deep water rice (floating rice), a low yielding rice type that can escape complete submergence by enhancing the growth of internodes. . Other varieties, including a new variety carrying the submergence 1 (SUB1) gene (Bailey-Serres et al. 2010), demonstrate adaptation to periods of total submergence, a potentially valuable trait as more frequent and prolonged submergence events may be a consequence of climate change.

The Ayeyarwady region can be generally divided into the upper (northern) and lower (southern) parts. The upper delta is located beyond salt water intrusion (Van Driel & Nauta 2014); whereas there are three main ecological zones in the lower delta which are related to the distance to the sea and to the salinity level: i) a floodplain zone characterized by freshwater or a very low salinity maximum, where freshwater irrigation allows rice cultivation as a summer crop, ii) brackish-water zone, salt-water interface with freshwater region, where summer crop cultivation is not guaranteed for all areas in this zone because water salinity levels increase progressively during the dry season, and iii) saline-water zone, located near coastal areas, where only monsoon rains support paddy growth and no summer crops are able to grow. The major causes of salinity in the delta region were sea level rise and the seasonal tidal regime. In delta area, the mean tidal level as well as the spring tide level fluctuates seasonally. Spring tides are highest in summer season leading to strong seasonal salinization. Consequently, the rice growing areas in delta are not completely protected against a periodic saltwater intrusion even in rainy season (Driel & Nauta 2013). Floodwater with the electric conductivity level more than 2 dS/m leads to yield loss up to 1 t/ha in rice (Asch & Wopereis 2001). Rice, being transplanted crop, can alleviate the salinity at the seedling stage by management i.e. transplanting of aged seedlings but cannot avoid stress at the flowering stage (Reddy et al. 2017). High salt reduces pollen viability of the flowering stage, which in turn determines grain yield (Khatun & Flowers 1995; Singh et al. 2004). Growth differences among various genotypes in response to salinity are dependent on the salt concentration and the degree of salt tolerance (Eynard et al. 2005). Identifying rice germplasm that is salt tolerant and breeding rice cultivars that are salt tolerant are the most economic and effective methods for the reduction of rice yield loss caused by salinity (Sun et al. 2019).

In the lower delta, resource-poor farmers largely depend on their own farm-saved seed; whereas in the upper delta, there is more formal sector engagement, with farmers increasingly becoming engaged in commercial farming and improved variety uptake. The difference in rice agroecology between the lower and the upper Ayeyarwady delta results in different rice growing techniques and different rice varieties. The transplanting technique in the lower delta uses the fork because of deeper water levels at the time of transplanting.

2.3 Varieties used and seed system in the Ayeyarwady delta

Farmers manage their own genetic resources and local varieties in the field. The diversity of local varieties is important for farmers in relation to risk management and resilience. Most of the farmers in the Ayeyarwady delta only grow one or two rice varieties per season, with the local aromatic Pawsan varieties of high importance in the lower Ayeyarwady delta and local varieties of minor importance in the upper delta due to more accessible to improved/modern rice varieties. Many different sub-selections from the original variety Pawsan Hmwe exist based on different growing areas. Pawsan Bay Kyar and Pawsan Yin are such sub-selections made by farmers, which are only slightly photosensitive as compared to the highly photosensitive variety Pawsan Hmwe (Subedi et al. 2017).

According to the survey results of the Ayeyarwady delta (Subedi et al. 2017), in the main rice production season farmers mostly grow improved high yielding varieties (HYVs). In the lower Ayeyarwady delta, the main variety sown was an improved variety in 59% and a local variety in 41%. In the upper Ayeyarwady delta, improved varieties are sown on 98% of the fields. Farmers closely follow rice market trends which in turn impact variety selection. Appreciated varieties by consumers and varieties fetching high prices on the market are most commonly used. In the upper delta, farmers can grow two crops per year according to agro-ecological conditions (no salt water intrusion) and for that reason they use HYVs for both seasons. However, in the lower delta, summer rice cannot be guaranteed for all areas so that farmers grow more local varieties, particularly Pawsan, which can fetch a high market price.

Farmers gain access to seed from different seed sources, particularly informal seed system such as saving part of their own grain harvest for the next crop production cycle, farmers' source seed from family, friends and neighbors. Seeds in this system are generally available at low costs but there is no standard mechanism for seed quality control. The general recommendation for farmers is to recycle certified seed for three generations, which will not result in significant quality deterioration. To gain information about new rice varieties, the informal network, as well as own observations play a substantial role in terms of variety selection. Formal networks such as specialized seed producers, companies or projects play a much more limited role in contributing to variety selection. In marginal and remote areas, it is often the only seed system or seed source available to farmers (Subedi et al. 2017).

The four generally recognized classes of seeds are: breeder's seed, foundation seed, registered seed and certified seed (Rice Knowledge Bank n.d). Breeder seed is the seed of a new variety that has the highest purity and is produced, developed, controlled, and provided directly by breeders or their institutions for further multiplication. Foundation seed is the progeny of the breeder seed produced by trained officers of an agricultural station in conformity with regulated national standards and handled to maintain genetic purity and identity of the variety. Registered seed is the progeny of the foundation seed grown by selected farmers, handled to maintain genetic purity and identity, and has undergone field and seed inspections to ensure conformity with standards. Certified seed is the progeny of foundation, registered, or certified seeds, handled to maintain sufficient varietal identity and

purity, grown by selected farmers under prescribed conditions of culture and isolation, and subjected to field and seed inspections prior to approval by the certifying agency. Harvest from this class is used for commercial planting. In the Ayeyarwady delta, there are five government seed farms that produce foundation seed, registered seed and certified seed only. The farms can get the breeder seed from the DAR (Department of Agricultural Research) headquarters in Yezin. In the public-private system, government actors interact with and support contact farmers who produce certified seed; sometimes rice millers become involved as intermediaries. This system works with the local Pawsan varieties as well as with improved varieties. Seeds in this system are subject to external quality assurance and certified. Field inspections are done by staff of the regional and district agriculture office to keep seed quality (Subedi et al. 2017).

Other seed sources are private companies like Gold Delta Company and several development organizations including GRET (Group for Research and Technology Exchanges), Mercy Corps, and IRRI. Gold Delta produce certified seeds with selected contract growers and then sold to their grain contract growers as a service. GRET used a system called Participatory Guarantee System (PGS) where individual farmers are members of small groups for a shared mechanism of seed quality control i.e. farmers check the quality of each other's seed. Mercy Corps involves millers to facilitate the linkage of seed producers with seed users. Farmers are trained in seed production of popular local and improved varieties. International Rice Research Institute (IRRI) also involves with key farmers in participatory selection of new improved varieties, especially climate resilient varieties (Subedi et al. 2017).

2.4 The rice botanical description

2.4.1 Taxonomy and origin

The cultivated rice plant (*Oryza sativa* L.) belongs to the tribe Oryzeae under the sub family Oryzoideae in the grass family Poaceae. The genus *Oryza* contains 24 species (2 cultivated and 22 wild). The two cultivated species and six wild relatives belonged to *O. sativa* complex are classified as AA genome diploid species with 24 chromosomes, including Asian cultivated rice (*O. sativa*), African cultivated rice (*O. glaberrima*), common wild rice (*O. rufipogon*), annual wild rice (*O. nivara*), Barth's rice (*O. barthii*), longstamen rice (*O. longistaminata*), Australian wild rice (*O. meridionalis*), and South American wild rice (*O. glumaepatula*). The geographical distributions of these eight species are totally different: *O. sativa* is grown all around the world with a concentration in Asia, *O. rufipogon* and *O. nivara* mainly distribute in Asia, *O. glaberrima* is concentrated in West Africa, *O. barthii* and *O. longistaminata* are found in tropical Africa, *O. meridionalis* is in Australia, and *O. glumaepatula* is in South America. Generally, the *O. sativa*, *O. glaberrima*, *O. nivara*, *O. barthii*, and *O. meridionalis* are annual species whereas the others are perennial species (Wei & Huang 2019). Asian cultivated rice, *O. sativa*, is divided into the indica and japonica subspecies. These two subspecies are commonly associated with differences in growth habitat, with indica rice usually found in the lowlands of tropical Asia and japonica rice typically found in the upland hills of southern China, southeast Asia, and Indonesia, as well

as outside of Asia (Africa, North America, and South America) (Khush 1997). Generally, indica is long grain, relatively less sticky, with less amylopectin, while japonica is short grain and more sticky. Besides seed shape and texture, indica and japonica show significant differences in plant height, leaf shape, leaf color, plant type, awn length, density of glume pubescence, germinating rate, cold tolerance, lodging resistance, disease resistance, seed shattering, tiller number, and many other agronomic traits. Compared to indica, the japonica varieties have shorter plant height, shaper leaf shape, light leaf color, strong cold tolerance, strong lodging resistance, are nonshattering, but have lower tiller number, slower germinating rate, are sensitive to rice blast, have long and dense glume pubescence and long awn in some varieties (Wei & Huang 2019). *O. sativa* japonica can be further differentiated into tropical (javanica) and temperate (japonica) forms, with the temperate form appearing to be a derivative of tropical japonica (Garris et al. 2005). The cluster analysis by Lu et al. (2002) revealed that the indica varieties were randomly scattered among the wild and weedy rice accessions, but the japonica varieties were grouped together independently from all other accessions.

The rice plant may be characterized as an annual grass, with round, hollow, jointed culms, rather flat, sessile leaf blades, and a terminal panicle.

2.4.2 Rice plant morphology

Rice grain

The rice grain, commonly called a seed, consists of the true fruit or brown rice (caryopsis) and the hull, which encloses the brown rice. Brown rice is largely composed of embryo and endosperm. The surface contains several thin layers of botanically differentiated tissues that enclose the embryo and endosperm. The embryo lies on the ventral side of the spikelet next to the lemma. The embryo contains the embryonic leaves (plumule) and the embryonic primary root (radicle). The plumule is enclosed by the coleoptile and the radicle ensheathed by the coleorhiza. The coleoptile is surrounded by the scutellum and the epiblast, the vascular trace which is fused with the lateral parts of the scutellum (cotyledon) (Figure 2) (Chang & Bardenas 1965).

The endosperm is enclosed by the aleurone layer which lies beneath the tegmen (Figure 2). The white starchy endosperm consists of starch granules embedded in a proteinaceous matrix. In the waxy (glutinous) varieties, the starch fraction is composed almost entirely of amylopectin and stains reddish-brown with weak potassium iodide-iodine solution. In the common, non-waxy (non-glutinous) types, the starch fraction contains amylose in addition to amylopectin and stains dark blue with potassium iodide-iodine solution. Chalky white spots often appear in the starchy endosperm. Soft textured, white spots occurring in the middle part on the ventral side (side on which the embryo lies) are called white bellies (Chang & Bardenas 1965).

The palea, lemma, sterile lemmas, and rachilla constitute the hull of indica rice (Figure 2). In japonica rice, however, because of the non-shattering characteristic, the hull usually

includes rudimentary glumes and perhaps a portion of the pedicel. A single grain weighs 10–45 milligrams at 0% moisture content. Grain length, width, and thickness vary widely among varieties (Yoshida 1981).

There are five rice varietal groups in Myanmar according to length/width ratio; (A) Emata (>3.30), (B) Latywezin (2.8–3.3), (C) Ngasein (2.4–2.8), (D) Meedon (2.0–2.4) and (E) Byat (2.25–3) (Irie et al. 2004). Emata long-grain rice has a long, slender kernel, three to four times longer than its width. Due to their starch composition, cooked grains are lighter, fluffier and more separated than medium or short grain rice. They are very exportable in quality as the kernel is hard and translucent and more attractive rice for overseas buyer especially European market because of shining and good appearance after polishing and sorting. Medium-grain rice (Latywezin) has a shorter, wider kernel than long grain rice, two to three times longer than its width. Cooked grains are moist and tender, with a tendency to cling together. Short-grain rice (Ngasein), named for its size, is only a tiny bit longer than it is wide. It's not uncommon for medium- and short-grain rice to get combined into the same category, which can make for some confusion. The kernels are hard, fairly translucent, often with abdominal white (Rice Myanmar Gold Delta n. d). Meedon rice varieties, particularly Pawsan are important for local adaptability, grain quality, premium price and market availability. They are bold, round grain and its length can increase three to four times the original size after cooking. Strong aroma, good taste and its elongation during cooking are the key characteristics of Pawsan rice (Thein et al. 2012). Byat grain type is more or less the same as Meedon. This classification is still used widely among Myanmar farmers for milling processes and marketing.

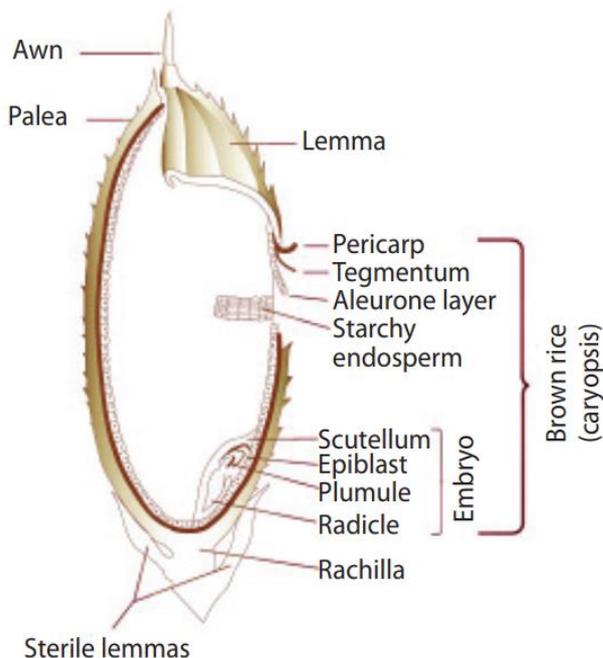


Figure 2. Cross-section of the rice grain (GRiSP 2013)

Germination

Germination and seedling development start when seed dormancy has been broken and the seed absorbs adequate water and is exposed to a temperature ranging from 10 to 40 °C. The physiological definition of germination is usually the time when the radicle or coleoptile (embryonic shoot) emerges from the ruptured seed coat. Under aerated conditions, the seminal root is the first to emerge through the coleorhizae from the embryo, and this is followed by the coleoptile. Under anaerobic conditions, however, the coleoptile is the first to emerge, with the roots developing when the coleoptile has reached the aerated regions of the environment. If the seed develops in the dark as when seeds are sown beneath the soil surface, a short stem (mesocotyl) develops, which lifts the crown of the plant to just below the soil surface (Figure 3). After the coleoptile emerges, it splits and the primary leaf develops (GRiSP 2013).

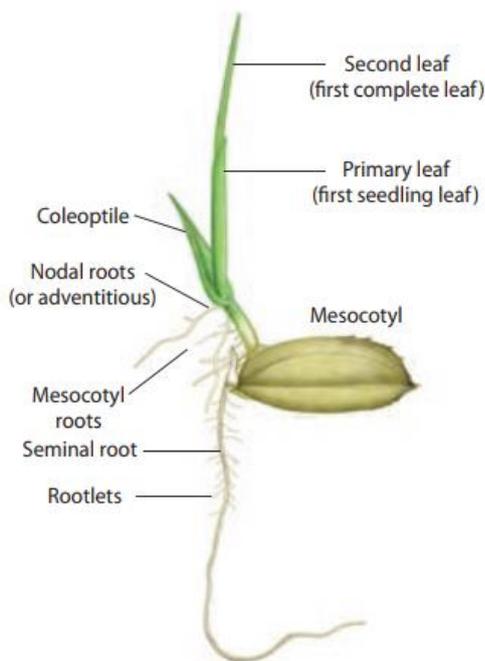


Figure 3. Parts of a young seedling germinated in the dark (GRiSP 2013)

Tillers

The tillering stage starts as soon as the seedling is self-supporting and generally finishes at panicle initiation. Tillering usually begins with the emergence of the first tiller when seedlings have five leaves. Tillers growing from the main stem are called primary tillers. These may generate secondary tillers, which may in turn generate tertiary tillers. These are produced in a synchronous manner. Although the tillers remain attached to the plant, at later stages they are independent because they produce their own roots. Varieties and races of rice

differ in tillering ability. Numerous environmental factors also affect tillering such as spacing, light, nutrient supply, and cultural practices (GRiSP 2013).

Leaf

A typical rice leaf is composed of the sheath, the blade, the ligule, and the auricles (Figure 4).

The leaf sheath is continuous with the blade. It envelops the culm above the node in varying length, form, and tightness. A swelling at the base of the leaf sheath just above the point of its insertion on the culm is the sheath pulvinus. The sheath pulvinus is usually above the nodal septum and is frequently misnamed the node (Chang & Bardenas 1965).

The leaf blade is attached at the node by the leaf sheath, which encircles the stem. The blades are generally flat and sessile. Varieties differ in blade length, width, area, shape, color, angle, and pubescence. The uppermost leaf below the panicle is the flag leaf. The flag leaf generally differs from the others in shape, size, and angle. Varieties also differ in leaf number (Chang & Bardenas 1965).

Leaf angle especially flag leaf angle (FLA) makes a large contribution to grain yield in rice. Erect leaves maximize carbon gain by optimizing the interception of photosynthetically active radiation for canopy photosynthesis and by mitigating heat stress induced by excess infrared radiation (Zhu et al. 2008; Van et al. 2010; Song et al. 2013). In addition, the more upright leaves also improve the accumulation of leaf nitrogen for grain filling in rice (Sinclair & Sheehy 1999).

Where the leaf blade and the leaf sheath meet is a pair of claw like appendages, called the auricles. Coarse hairs cover the surface of the auricles. The presence of well-developed auricles is often used as a convenient guide for differentiating rice from barnyard grasses (*Echinochloa crus-galli*), which lack auricles. The auricles may not persist on older leaves. Immediately above the auricles is a thin, upright membrane called the ligule, which is a small, white, triangular scale that looks like a continuation of the sheath (Yoshida 1981; GRiSP 2013). The ligule varies in length, color, and shape from variety to variety. Some rice varieties, however, lack the ligule and auricles (liguleless rice). The junction of the sheath and blade is the collar or junctura. The collar often appears as a raised region on the back of the leaf. The sheath pulvinus, auricles, ligule and collar on the same plant may be differentially pigmented. When pigmented, the dorsal, ventral and lateral parts of the collar may slightly differ in color (Chang & Bardenas 1965).

Culm/stem

The culm is one of the most important agronomic traits and plays a critical role in determining the lodging resistance and final architecture of crop plants (Wang et al. 2017). The culm/stem is composed of a series of nodes and internodes (Figure 4). It is enclosed within the sheath before heading, and a small portion of the culm right below the panicle becomes exposed after heading. The internodes vary in length depending on variety and

environmental conditions, but generally increase from the lower to the upper part of the stem. Each upper node bears a leaf and a bud, which can grow into a tiller. The number of nodes varies from 13 to 16, with only the upper 4 or 5 separated by long internodes. Under rapid increases in water level, some deep-water rice varieties can also increase the lower internode lengths by more than 30 centimeters (cm) each (Yoshida 1981; GRiSP 2013).

Roots

The rice root system consists of two major types: crown roots (including mat roots) and nodal roots (Figure 4). In fact, both these roots develop from nodes, but crown roots develop from nodes below the soil surface. Roots that develop from nodes above the soil surface usually are referred to as nodal roots. Nodal roots are often found in rice cultivars growing at water depths above 80 cm. Most rice varieties reach a maximum depth of 1 m or more in soft upland soils. In flooded soils, however, rice roots seldom exceed a depth of 40 cm. That is largely a consequence of limited oxygen diffusion through the gas spaces of roots (aerenchyma) to supply the growing root tips (GRiSP 2013).

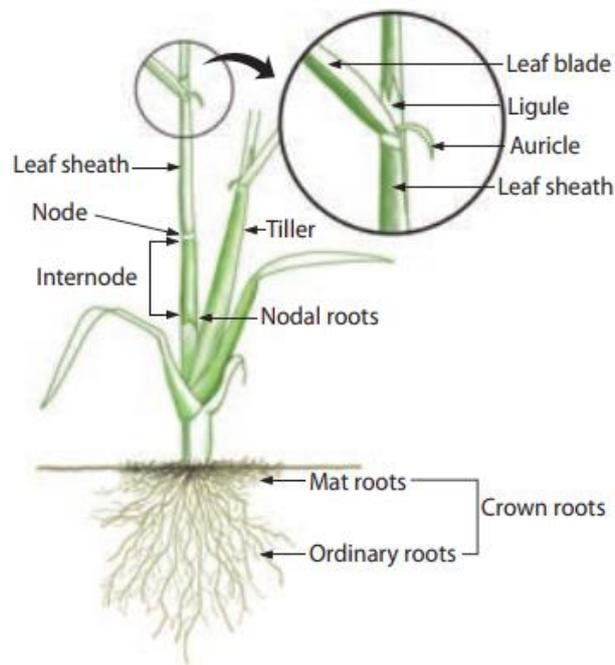


Figure 4. Parts of the rice stem and tillers (GRiSP 2013)

Inflorescence

The major structures of the panicle are the base, axis, primary and secondary branches, pedicel, rudimentary glumes, and spikelets (Figure 5). The panicle base often appears as a ciliate ring and is used as a dividing point in measuring culm length and panicle length. The region about the panicle base is often called the neck (Chang & Bardenas 1965). The panicle axis (rachis) extends from the panicle base to the apex; it has 8–10 nodes at 2 to 4 cm

intervals, from which primary branches develop. Secondary branches develop from the primary branches. Pedicels develop from the nodes of the primary and secondary branches. The spikelet is borne on the pedicel which is morphologically a peduncle. Varieties differ greatly in the length, shape, and angle of the primary branches, and in the weight and density (number of spikelets per unit of length) of the panicle (Chang & Bardenas 1965).

A spikelet consists of a minute axis (rachilla) on which a single floret is borne in the axils of 2-ranked bracts. The bracts of the lower pair on the rachilla, being always sterile, are the sterile lemmas. The upper bracts or the flowering glumes consist of the lemma (fertile lemma) and palea. The lemma, palea, and the included flower form the floret (Chang & Bardenas 1965). The flower consists of the pistil and stamens, and the components of the pistil are the stigmas, styles, and ovary (GRiSP 2013). The sterile lemmas are generally shorter than the lemma and palea, seldom exceeding one-third the length of the latter. The sterile lemmas may be equal or unequal in size, the upper one generally being larger. The lemma is the larger, indurate (hardened), 5-nerved bract which partly envelops the smaller, 3-nerved palea. The middle nerve or keel may be ciliate or smooth. The extended tips of the lemma and the palea are the apiculi. The apiculi may be separated into lemmal apiculus and paleal apiculus. The awn is a filiform extension of the keel of the lemma. The surface of the lemma and the palea may be pubescent or glabrous. In some varieties, a pair of lateral nerves on each side of the central nerve of the lemma may fuse to form a knob-like mucro on either side of the lemmal apiculus (Chang & Bardenas 1965).

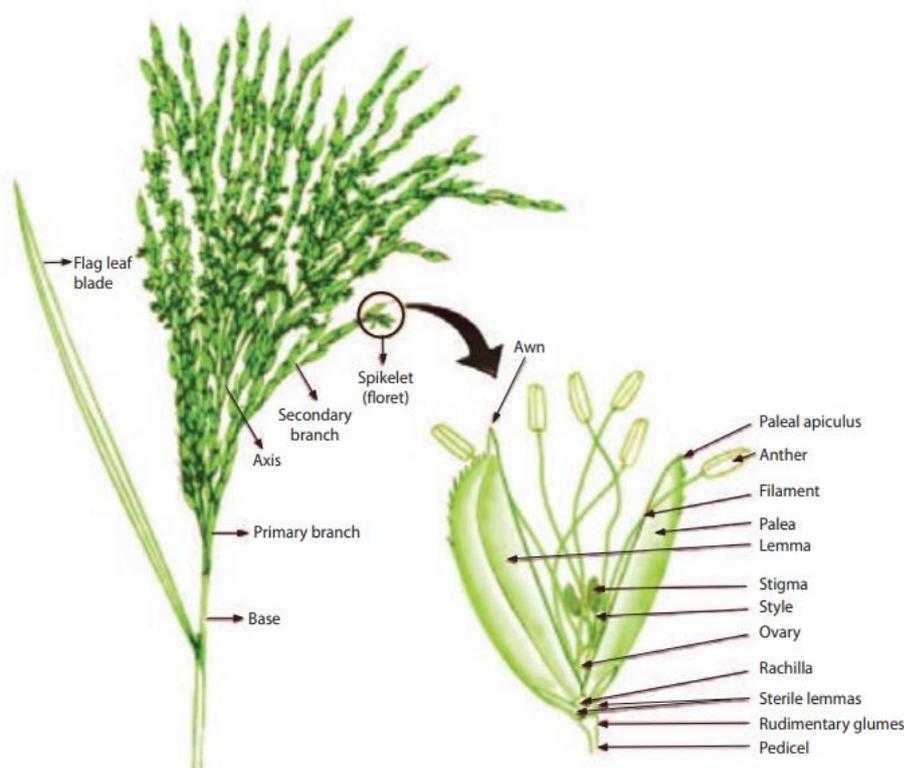


Figure 5. Rice panicle and spikelets (GRiSP 2013)

2.4.3 Rice growth stages

The growth duration of the rice plant is 3–6 months, depending on the variety and the environment under which it is grown. The growth stages of rice are divided into the vegetative phase, the reproductive phase and ripening phase. The vegetative stage refers to a period from germination to the initiation of panicle primordia; the reproductive stage, from panicle primordial initiation to heading; and the ripening period, from heading to maturity. A typical 120-day variety, when planted in a tropical environment, spends about 60 days in the vegetative stage, 30 days in the reproductive stage, and 30 days in the ripening period (Figure 6) (Yoshida 1981).

The vegetative phase is characterized by active tillering, a gradual increase in plant height, and leaf emergence at regular intervals. Tillering may start when the main culm develops the 5th or 6th leaf. Active tillering refers to a stage when the increase in tiller number per unit of time is high. The maximum tiller number stage follows active tillering. It is a stage when tiller number per plant or per square meter is maximum before or after the initiation of panicle primordia, depending on a variety's growth duration. Because tiller number declines after the maximum tiller number stage, there is a period before that stage (often called the end stage of effective tillering) when the tiller number becomes numerically equal to panicle number at maturity. That does not necessarily mean that tillers developed after the end stage will not bear panicles. But tillers developed at early growth stages normally produce panicles, while those developed later may or may not. Tillers that do not bear panicles are called ineffective tillers (Yoshida 1981). The vegetative growth phase can be further divided into the basic vegetative phase (BVP) and the photoperiod-sensitive phase (PSP). The BVP refers to the juvenile growth stage of the plant, which is not affected by photoperiod. It is only after the BVP has been completed that the plant is able to show its response to the photoperiodic stimulus for flowering, which is during the PSP of the plant (Vergara & Chang 1985).

The reproductive growth phase is characterized by culm elongation (which increases plant height), a decline in tiller number, emergence of the flag leaf (the last leaf), booting, heading, and flowering of the spikelets. Panicle initiation is the stage about 25 days before heading when the panicle has grown to about 1 mm long and can be recognized visually or under magnification following stem dissection. Spikelet anthesis (or flowering) begins with panicle exertion (heading) or on the following day. Consequently, heading is considered a synonym for anthesis in rice. It takes 10–14 days for a rice crop to complete heading because there is variation in panicle exertion among tillers of the same plant and among plants in the same field. Agronomically, heading is usually defined as the time when 50% of the panicles have exerted. Anthesis normally occurs from 1,000 h to 1,300 h in tropical environments and fertilization is completed within 6 hours. Very few spikelets have anthesis in the afternoon, usually when the temperature is low. Within the same plant, it takes 7–10 days for all the panicles to complete anthesis; the spikelets themselves complete anthesis within 5 days (Yoshida 1981).

Ripening follows fertilization and can be subdivided into milky, dough, yellow ripe, and maturity stages. These terms are primarily based on the texture and color of the growing grains. Ripening is characterized by leaf senescence and grain growth (increases in grain size and weight and changes in grain color). The length of ripening varies among varieties from about 15 to 40 days. Ripening is also affected by temperature, with a range from about 30 days in the tropics to 65 days in cool temperate regions, such as Hokkaido, Japan; and Yanco, Australia (Yoshida 1981).

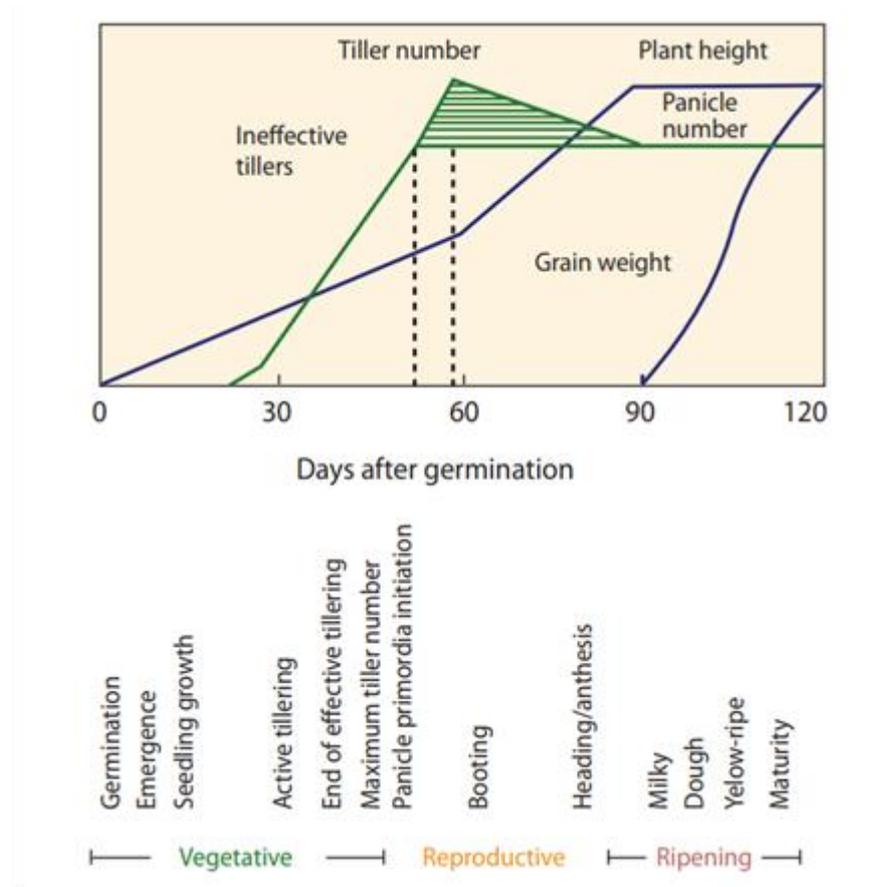


Figure 6. Schematic growth of a 120-day rice variety in the tropics (GRiSP 2013)

2.4.4 Yield and yield components

Yield

Rice is harvested as paddy with the hull comprising of about 20%, the bran and embryo 8–12% and the endosperm part 70–72% (Gujral et al. 2012). Paddy after removal of husk gives brown rice that is further polished to remove the bran and germ (embryo) resulting in white rice. Rice yield is usually reported as rough rice (paddy) at 14% moisture content, except in a few countries, such as Japan and Korea, where yield is expressed in terms of brown rice or sometimes in terms of polished rice. Conversion factors of 0.8 and 1.25 are

usually used to obtain brown-rice weight from grain weight, and grain weight from brown-rice weight, respectively (Yoshida 1981).

Harvest index

The harvest index is a measure of the economically useful fraction of the biological yield. It is a common way of examining rice grain yield.

$$\begin{aligned}\text{Harvest index (HI)} &= \text{economic yield/biological yield} \\ &= \text{dry grain yield/total dry weight}\end{aligned}$$

$$\text{Dry grain yield} = \text{HI} \times \text{total dry weight}$$

The last equation indicates that grain yield can be increased by either increasing total dry matter production or by increasing the harvest index. The total dry weight is a measure of a crop's photosynthetic performance and the harvest index is a measure of the economically useful fraction of the biological yield. The total dry weight of a good rice crop is around 10–20 t/ha, depending on variety, management and environment. The harvest index is about 0.3 for traditional tall varieties and 0.5 for improved, short varieties (Yoshida 1981).

Yield components

A second method for examining yield performance is to break the yield into its components:

$$\begin{aligned}\text{Grain yield (t/ha)} &= \text{panicle number/m}^2 \times \text{spikelet number/panicle} \times \% \text{ filled spikelets} \times \\ &1,000\text{-grain weight (g)} \times 10^{-5} \\ &= \text{spikelet number/m}^2 \times \% \text{ filled spikelets} \times 1,000\text{-grain weight (g)} \times 10^{-5}\end{aligned}$$

The spikelet number includes filled, partially filled, and unfertilized spikelets. The filled spikelet is called grain. The percentage of filled spikelets is a ratio of the number of grains to the total number of spikelets. The 1000-grain weight is the average weight, in grams, of 1000 grains (Yoshida 1981).

2.4.5 Flowering response to photoperiod

The length of a day, defined as the interval between sunrise and sunset, is known as photoperiod. Rice is basically a short-day plant. It initiates panicle primordia in response to short photoperiods. Panicle primordia may be initiated late or they may not develop when the plant is subjected to long photoperiods. Most of the wild *Oryza* species and many of the primitive cultivated rice (*O. sativa* L.) are photoperiod sensitive. The present tendency is to select photoperiod-insensitive cultivars so that most of the cultivated varieties may eventually become photoperiod-insensitive ones. These improved, early maturing cultivars may fit into the multiple cropping system characteristic of progressive agriculture (Vergara & Chang 1985).

Optimum photoperiod and critical photoperiod are terms commonly used to characterize the PSP. Optimum photoperiod is the day length at which the duration from sowing to

flowering is at a minimum. The optimum photoperiod of most varieties is about 9–10 hours. A longer or shorter photoperiod delays flowering, the delay depending upon the sensitivity of the variety. Critical photoperiod is the longest photoperiod at which the plant will flower or the photoperiod beyond which it cannot flower. The critical photoperiod of most varieties ranges from 12 to 14 hours (Yoshida 1981).

In photoperiod-sensitive cultivars, the PSP determines the rice plant's sensitivity. The PSP of photoperiod-insensitive cultivars ranges from 0 to 30 days while that of sensitive cultivars lasts 31 days or longer. Under continually long photoperiods, some cultivars have been reported to remain vegetative even after 12 year of growth (Kondo et al. 1942). The PSP is usually determined by subtracting the minimum growth duration (growth duration at the optimum photoperiod) from the maximum growth duration (growth duration at the critical photoperiod) of a cultivar (Vergara et al. 1965). Because many cultivars remain vegetative for a long period if grown under long-day conditions, experiments are usually terminated after 200 d and the PSP of the cultivar is given the value of 200+.

2.5 Variations of rice germplasm

A wide range of vernacular names for each variety does not always represent genetically diversified cultivars because of linguistic differences among different ethnic groups (Watanabe et al. 2007). If the farmer-named varieties are not genetically distinct, farmer taxonomies and nomenclature which are typically localized and culturally determined would not affect in identification and qualification of diversity in agricultural ecosystem (Sadiki et al. 2007). Asian cultivated rice (*Oryza sativa* L.) is normally classified into two subspecies; indica and japonica. Indica-type landraces predominate in Myanmar as 81% of total landraces (Khush et al. 2003) and japonica-type landraces specially prevail in Eastern plateau region (Saw 2007). However, genetic variations and dissemination of Myanmar rice landraces depends on the adaptation to local agro-ecology, socio-economic and environmental harsh conditions; biotic and abiotic stress.

Wide range of photoperiod sensitivity among landraces is important for crop adaptability at different ecological environments. Less sensitive photoperiod cultivars take a popular role in irrigated dry-zone areas because they are widely adaptable and suitable for cultivation year around. Tun et al. (2005) reported that highly photoperiod sensitive landraces predominate countrywide in Myanmar but low sensitive landraces are exclusively distributed in the mountainous regions. In the low to intermediate rain-fed lowland areas, medium to medium-late maturing-type, photoperiod sensitive or insensitive-type varieties are usually overwhelming, while the traditional medium-late to late maturing varieties are dominant in large areas of the intermediate to semi-deep rain-fed lowland. In deep water areas, the traditional tall pant-type with photoperiod sensitive are predominant. Mainly, traditional rice are monsoon rice that are grown only once in a year. Early maturing landraces are more adaptable to high altitude mountainous regions (Tun 2006).

Amylose content of grain starch and type of endosperm are one of the attributes for selection. High ethnical complexity, diverse dietary habits and arts of rice cooking are the

main factors that create a great diversity of rice landraces in Myanmar. Aung et al. (2003) reported that Myanmar local rice cultivars have a wealth of genetic diversity for seed storage protein and endosperm starch characters. Very low amylose landraces are frequently found in the North-East mountainous zone. Opaque and waxy endosperm types are dispersed in the mountainous region related to diverse cooking and eating habits and processing techniques traditionally transmitted by various ethnic groups settled in mountainous regions and affected by cultural influences of surrounding areas, especially the so-called waxy rice zone expanding around Thailand, Laos and Cambodia.

2.6 Agro-morphological traits

Agro-morphological traits (passport data) play a very important role in varietal identification which ultimately helps rice breeders (Laxuman et al. 2011). They are important also in farmer communities because farmers identify or distinguish varieties by using some standards such as growth duration, water regime, seed size, etc. Agro-morphological properties of rice cultivars determine their yield potential, local agronomic suitability and ability to adapt or tolerate biotic and abiotic stresses (Xu & MacKill 1996).

These marker traits are often susceptible to phenotypic plasticity; conversely, this allows assessment of diversity in the presence of environmental variation which cannot be neglected from the genotypic variation. They do not require expensive technology but large tracts of land area are often required for these field experiments, making it possibly more expensive than molecular assessment in western (developed) countries and equally expensive in Asian and Middle East (developing) countries considering the labor cost and availability. These types of markers are still having advantage and they are mandatory for distinguishing the adult plants from their genetic contamination in the field, for example, spiny seeds, bristled panicle, and flower/leaf color variants (Govindaraj et al. 2015).

In Myanmar, a number of works have been reported using different morphological and agronomic traits in rice and it led to the identification of the phenotypic variability in rice. Aung (2007) studied on the diversity of 124 Myanmar rice accessions revealed that the mean Shannon Weaver diversity index H' for both quantitative and qualitative traits is 0.57 while for quantitative traits, H' is 0.71, and for qualitative traits, H' is 0.52. It indicated that a moderate level of diversity exists for morphological characters in Myanmar rice varieties. Wunna (2015) also studied the diversity of 175 rice accessions from upland and lowland ecosystems in Myanmar. The result explained that significant variation of agronomic traits, namely, seed size, cooking and eating quality and heading date, was investigated among upland and lowland populations, and regional attributed agronomic characters. Variation of upland population was more variable than lowland group except heading date trait with effect of harsh environment and diverse human tribes. Yamanaka et al. (2011) conducted an assessment of phenotypic variation to characterize rice genetic resource in Myanmar. Two populations of rice landraces, a seed bank population maintained by seed-propagation in a gene bank for several generations and an “on farm” population collected from agricultural lands were evaluated. Phenotypic characteristics of representative agronomic traits in rice, such as culm length, panicle length, number of tillers and days to heading, were

measured in both populations. Principal Component Analysis (PCA) analyses of agronomic traits showed that a variation in the seed-bank population had narrower genetic bases than the on-farm population. Genetic bias caused by ‘unconscious selection’ during the gene bank management processes may have occurred in the landraces. The importance of the conservation on on-farm landraces of *Oryza sativa* and its wild relatives was proposed in order to ensure the genetic resources for further breeding and conserve biological diversity. All the above studies indicated that morphological traits were useful for preliminary evaluation for crop improvement programs and can be used for assessing genetic diversity among morphologically distinguishable rice landraces. However, the use of morphological traits to evaluate the genetic relationships among rice varieties has encountered difficulties in obtaining reproducible results in different environments, times and management strategies (Yue et al. 2008; Tanaka & Shiraiwa 2009). In contrast, since molecular markers to evaluate genetic relationships is not affected by the environments and/or management practices, molecular markers to analyze genetic relationships in crops has become increasingly popular.

2.7 Molecular markers for genetic studies

Molecular markers are the most widely used type of marker predominantly due to their abundance. These markers can detect the variation that arises from deletion, duplication, inversion, and/or insertion in the chromosomes and they are selectively neutral because they are usually located in noncoding regions of DNA in a chromosome. Unlike other markers, DNA markers are unlimited in number and represent the variation in DNA sequences among the individuals of a species. These markers are stable and not confounded by the environment, pleiotropic and epistatic effects. Thus, among molecular markers, DNA based markers are considered as potential molecular markers. (Winter & Kahl 1995; Adhikari et al. 2017). They offer numerous advantages over conventional, phenotype-based alternatives as they are detectable in all tissues regardless of growth, differentiation, development, or defense status of the cell.

These markers are extensively utilized for assessment of genetic diversity, inferring systematics and molecular phylogeny, identification of close relatives, varieties/cultivars, management and improvement of crops, conservation of endangered species, etc. Molecular markers may be dominant (unable to distinguish allelic difference of a gene in heterozygous conditions), or codominant (allelic difference can be detected). Protein markers (Allozymes, Isozymes) and DNA-based single locus markers (RFLP, SSR, SNPs) are codominant, whereas multilocus (e.g., RAPD, DAMD, ISSR, AFLP) are dominant markers (Adhikari et al. 2017). Limitations associated with these marker systems include low marker density, poor genome coverage, and less cost-effectiveness. Some of these methods (e.g., SSR) require pre-identification of a polymorphism (or a potential site for it) before analysis of other individuals is possible. Furthermore, all methods based on size separation of multiple DNA fragments suffer from difficulties in precisely correlating bands on gels with allelic variants (Jaccoud et al. 2001).

Most of the studies on Myanmar landraces until now were conducted on limited sets of resources, using old generation of markers like restriction fragment length polymorphism (RFLP), amplified fragment length polymorphism (AFLP) and cleaved amplified polymorphic sequence (CAPS) (Saw et al. 2006), which are seldom used now due to poor marker-trait association information and, only in recent past, simple sequence repeat (SSR) (Aung 2007; Oo et al. 2008; Yamanaka et al. 2011; Thein et al. 2012; Thein et al. 2014; Wunna et al. 2016). According to a review article of microsatellite markers (SSRs) and their application on *Oryza sativa* L., SSR markers have been utilized most extensively in the genetic diversity studies of over 600 Myanmar rice landraces, while there is no report of using SNP markers up to now (Thant et al. 2018).

Sequence-based single nucleotide polymorphism (SNP), biallelic markers, are excellent for genomic approaches particularly, for those studies that require a high number of markers such as marker-trait association, genomic selection, and determining population structure (Kumar et al. 2012). SNP markers developed through automated sequencers can constitute an important choice for molecular studies due to their wide and uniform genome coverage with high-throughput and cost-effectiveness (Gupta et al. 2008). Many studies have investigated the genomic structure and genetic diversity of rice, and have identified millions of SNPs (Huang et al. 2010; Huang et al. 2011; Huang et al. 2012; Xu et al. 2011). The advantage of SNPs as markers is that they have a very high density in the genome, approximately 1.6 to 1.7 SNPs/kb in rice (Feltus et al. 2004; McNally et al. 2009). In recent times, advanced Genotyping-by-Sequencing (GBS) platforms, such as Diversity Array Technology (DArT) (Sansaloni et al. 2011), have enabled timely mining of high-density SNP information at a significantly reduced cost (Poland & Rife 2012). Consequently, diversity studies of several crop species have shown relatively better coverage and reduced missing data with DArT compared to other GBS platforms (Chen et al. 2016; Nazzicari et al. 2016; Yang et al. 2016).

2.8 DArTseq markers

Diversity Array Technology Pty Ltd. (DArT, Canberra, ACT, Australia) developed cost-effective sequence-independent ultra-high-throughput marker systems in 2001 (Jaccoud et al. 2001). The technology was based on hybridization and solid-state surfaces, rather than relying on resolving DNA polymorphisms through electrophoretic gel separation, and thus helped to improve both throughput and accuracy (Melville et al. 2017). Additionally, by combining the complexity reduction of the DArT method with high-throughput next-generation sequencing (NGS) technologies, the DArTseq platform was developed signifying a new implementation of sequencing of complexity-reduced representations (Qiu et al. 2012). Over the last decade, DArTseq has generated two types of markers: i) silicoDArT and ii) SNP markers. SilicoDArT markers are microarray markers that are dominant and scored for the presence or absence of a single allele. SilicoDArT markers have been rarely utilized in rice (Jaccoud et al. 2001; Xie et al. 2006). DArTseq-based SNPs are co-dominant markers. The first step of marker development involves the creation of a library of genomic fragments using restriction enzymes to digest DNA and reduce genome complexity. Fragments selected from the library are spotted on a glass slide using a microarray platform. The target DNA is

treated in the same way as the DNA used to constitute the library. It is digested with the same enzymes, and the fragments are hybridized on a chip to reveal the presence/absence of certain sequences. The sequences are aligned to the reference species genome and SNPs are identified in the sequences (Phung et al. 2014). Both types of markers have been successfully applied in several crop species for genetic diversity, genetic mapping, and population structure studies (Alam et al. 2018). The many advantages of DArTseq include no prior knowledge about sequencing of the plant genome and the capacity to produce high-density results, scoring thousands of unique genomic-wide DNA fragments in one single experiment with low-cost genotype information (Jaccoud et al. 2001; Kilian et al. 2016). Additional advantages of DArTseq technology are its suitability for polyploid species as well as the possibility of developing rapidly for virtually any genome (Barilli et al. 2018). Hence, DArTseq has been widely applied (Kilian et al. 2012; Courtois et al. 2013; Von et al. 2013) and is rapidly gaining popularity as a preferred method of genotyping by sequencing (Sánchez-Sevilla et al. 2015).

Mogga et al. (2018) used DArTseq markers to investigate genetic diversity in rice (*Oryza sativa* L.). Their study was performed using 59 rice genotypes with 525 SNPs derived from DArTseq platform. Phung et al. (2014) also characterized the panel of 182 rice genotypes with 25,971 markers using DArT and SNP markers. To date, there are only few studies about genetic diversity on rice by using DArTseq markers though many studies have been applied on other crops such as barley, rye, bean, macadamia etc. Therefore, silicoDArT and SNP markers may better suit genetic diversity studies, association/linkage mapping, and/or sequence based physical mapping in rice. Additionally, the co-dominant inheritance pattern of SNP markers may increase the utility of DArT platforms for genetic identity and parentage analysis (Alam et al. 2018).

3. OBJECTIVES AND HYPOTHESIS

The main objective of the study was to assess the variability within the Myanmar traditional rice varieties and classify them based on agro-morphological traits and genomic content, which would provide useful information to facilitate the choice of genitors for rice breeding programs.

The specific objectives were:

- (i) to delineate the on-farm varietal diversity and the determinants of farmers' variety choices in the Ayeyarwady delta in southern Myanmar
- (ii) to characterize the genetic resources of Myanmar rice (*Oryza sativa* L.) on the basis of agro-morphological traits for genetic diversity analysis
- (iii) to investigate the genetic variation, relatedness and population structure of rice genotypes using DArTseq markers.

Hypothesis

1. On-farm rice varietal diversity is expected to be high because of different agro-ecological regions in the study area and the variety choice by farmers mainly depends on the adaptation to local agro-ecology, socio-economic and environmental harsh conditions; biotic and abiotic stress.
2. Wide range of agro-morphological variations of rice genotypes in the Ayeyarwady region is expected to be high due to different rice ecosystem, especially water regime. Farmers usually name rice varieties based on farmers' taxonomic system such as tillering pattern, leaf type, grain shape, etc. Therefore, there are many different local landraces even though they are given to the same name and vice versa.
3. Investigating genetic diversity and population structure of local rice genotypes at molecular level will help rice varieties identifying based on the extent of genetic variability and relatedness among them. There may be different genetic background rather than agronomic characterization among groups. Further, DArTseq marker analysis will pinpoint specific genes conferring desirable phenotypic traits for further study.

4. METHODOLOGY

4.1 On-farm rice diversity and farmers' preferences for varietal attributes in Ayeyarwady Delta, Myanmar

4.1.1 Sites selection and farmers survey

The study area covered five townships: Bogale, Mawlamyinegyun, Labutta, Myaungmya, and Pathein in the lower Ayeyarwady region (Figure 7). We visited key informants, including researchers, township agricultural staff, local leaders, and experienced farmers, to identify the survey areas and select respondents in the five townships in the Ayeyarwady region. In total, 150 respondents were chosen from 15 villages (three villages in each township), representing three agro-ecological zones: i) freshwater zone, where freshwater irrigation allows rice cultivation as a summer crop; ii) saline-water zone, located near coastal areas, where only monsoon rains support paddy growth and iii) brackish-water zone, salt-water interface with freshwater region, where summer crop cultivation is not guaranteed because water salinity levels increase progressively during the dry season.

A structured questionnaire was developed after a thorough literature review and consultation with experts and extension officers (Appendix 3). It consisted of several parts to obtain the following information: a) basic household characteristics, b) land-tenure status and soil type, c) rice varieties grown during the monsoon season, d) awareness of the benefits and disadvantages of traditional rice varieties, e) farmers' preferences for rice variety traits, and f) major constraints in rice production.

Farmers were further asked to select the three most important problems related to rice cultivation from a list of 25 potential constraints (3 = the most serious cultivation constraint, 1 = the least important constraint, and 0 if the problem was not considered by the farmer). The problems were classified into three categories: agro-ecological, technical, and socio-economic constraints and the mean value was calculated for each particular problem, for a category as well as for all study sites. Furthermore, the mean values of each problem were calculated for different agro-ecological zones, different soil fertility types, terrain, and soil texture to determine the possible effects of these variables on rice cultivation constraints. The mean scores were calculated as a mean of scores given by farmers within a particular agro-ecological zone, soil-fertility type, terrain, and soil texture.

In addition, farmers were asked to score the most appreciated rice properties, with scores ranging from 3 (the most appreciated) to 1 (the least appreciated). The mean score was calculated for each characteristic as well as for a group of characteristics (cultivation characteristics, resistance to stresses and grain characteristics), similar to the rice cultivation constraint scores.

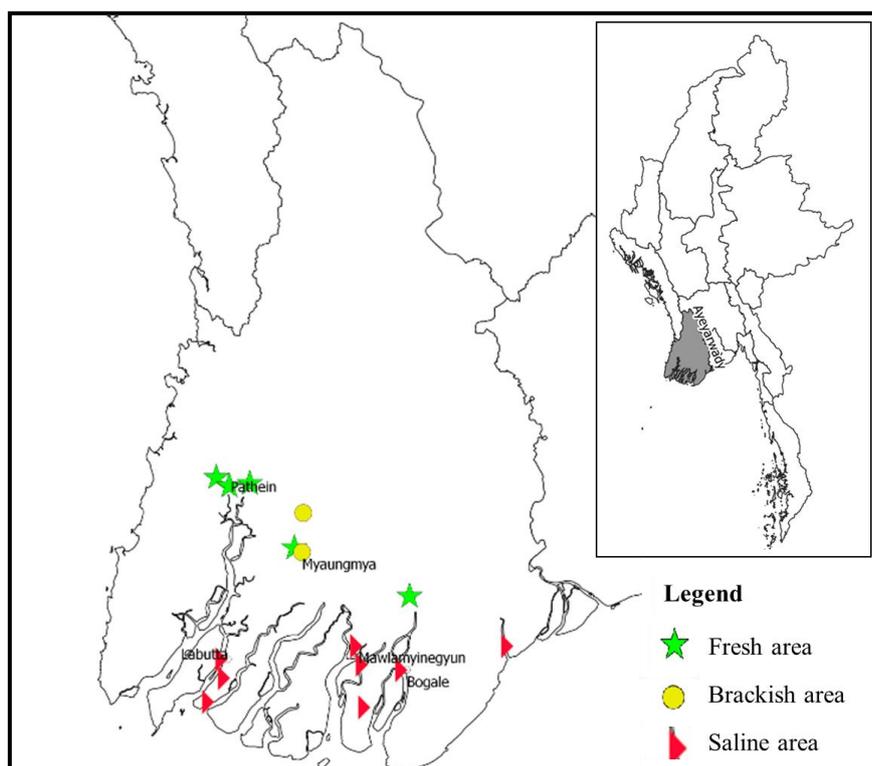


Figure 7. Study area-showing location of surveyed villages in five townships

4.1.2 Data analysis

The survey data were processed using SPSS 22 software (IBM SPSS, Inc., Chicago, USA). The effect of each agro-ecological zone, soil fertility, type of terrain, and soil texture on the number of rice varieties grown by the farmers, yield, and area sown was tested using the non-parametric Kruskal-Wallis test ($p < 0.05$), as the parametric test assumptions were not fulfilled. Similarly, Kruskal-Wallis test was used to determine the significance of the effects of different regions (agro-ecological zone, soil fertility, terrain, soil texture) on the scores given to particular cultivation constraints and preferred rice traits because data were not normally distributed.

To assess the impact of the data collected on the use of HYVs, Pawsan or other traditional varieties (scale: 1 = farmers adopting HYVs, 0 = farmers not adopting HYVs, only using traditional varieties), separate binominal logistic regressions were used and the significant models ($p < 0.05$) are presented. We included socio-economic characteristics of households (farming experience, education, land-tenure status, age, total sown area) as well as region and agronomic parameters (agro-ecological zone, soil fertility, type of terrain, soil texture) in the models. Similar binomial logistic regressions were performed to evaluate the impact of the same parameters on the use of more than two rice varieties on the farm (1 = more than two varieties, 0 = less than two varieties, corresponding to farmers growing only one rice variety on their land) to reveal the drivers of crop diversification by the farmers. Binomial logistic regression was also used to analyze the effect of the scores given by the farmers to the

different rice traits and cultivation constraints on the adoption of traditional varieties, Pawsan varieties, and HYVs. The power of the model was determined by Nagelkerke R^2 , which evaluates the goodness of fit of the logistic regression model.

The correspondence ordination results and the corresponding relationships were projected in the two-dimensional biplot diagrams. Same procedure was used for farmers' preferred rice traits to detect the possible correlation between farmers' preferences and actual varietal choices.

4.2 Phenotypic characterization of rice varieties

4.2.1 Plant material

A total of 117 rice genotypes were used in this study and all genotypes except the HYVs were originally derived from the Ayeyarwady region (Appendix Table A1). Seventy-two genotypes were provided by the Department of Agricultural Research (DAR) seed bank section in Yezin and 40 were directly collected from farmers' fields in different parts of the Ayeyarwady region (Figure 8). Five popular HYVs, which can be grown across the country, were obtained from the International Rice Research Institute (IRRI-DAR), Yezin, and they were used as controls (check varieties). Rice genotypes in this study are grouped into traditional and high yielding varieties (HYVs). However, traditional varieties were further divided into Pawsan varieties and traditional varieties (other than Pawsan) since Pawsan rice varieties are of high importance in the Ayeyarwady delta region. Thus, in this study, 17 Pawsan, 95 traditional (other than Pawsan), and 5 HYVs were included.

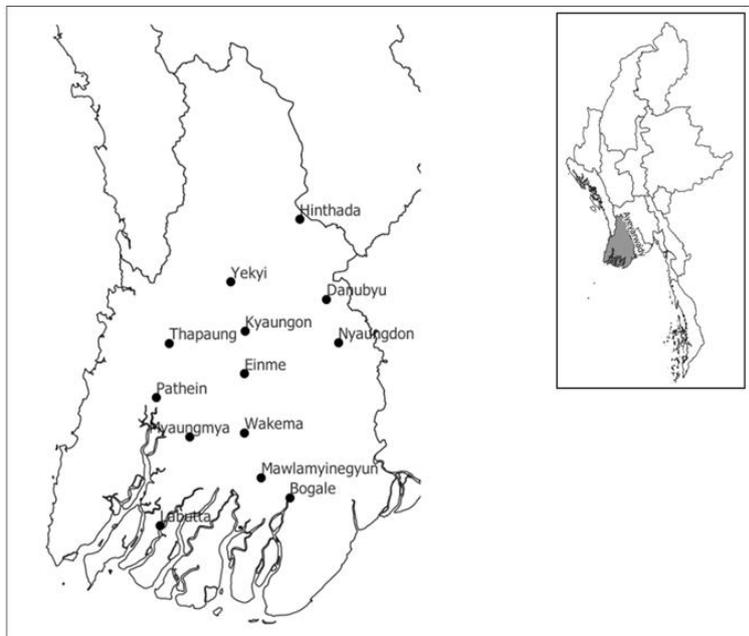


Figure 8. Area-showing collection sites of 112 rice genotypes in the Ayeyarwady delta

4.2.2 Experimental design and data collection

The field experiment was conducted in 2017 monsoon cropping season (June to November) at DAR/IRRI research farm, which is located in Yezin, at latitude 19°84'N, longitude 96°26'E at 107 m above sea level, Nay Pyi Taw, Myanmar. Monthly weather data such as minimum and maximum temperature, rainfall, and relative humidity for 2017 were collected from Yezin Agrometeorological station, Nay Pyi Taw, Myanmar (Appendix Table A2). Ten grams of seeds for each variety were sown on seedbed and let to germinate. The statistical experimental design was Augmented Randomized Complete Block Design (RCBD), contained both replicated and un-replicated treatments (Federer & Raghavarao 1975). Replicated treatments “c” checks (control) are tested in each block as in a RCBD. Un-replicated treatments “n” new treatments or genotypes occur in only one block - so each block has a different set of un-replicated treatments. When the seedlings were 17 days old, 2–3 vigorous seedlings for one hill of each genotype were transplanted to the experimental field. Each block consisted of 33 rows of 50 hills spaced 15 cm apart, with the rows spaced 30 cm apart. Layout for Augmented RCBD and field experiment at DAR-IRRI farm is shown in Appendix Figure A1 and Appendix Figure A2. Recommended dosage of fertilizer by IRRI (60-30-25 kg/ha of N-P-K and 16 kg/ha of gypsum) were applied at 14 days after transplanting, tillering stage (24–30 days after transplanting), and panicle initiation stage (60 days before harvest). In addition, 30 kg/ha of P and 16 kg/ha of gypsum were applied at land preparation. Carbofuran 3G was applied to prevent nematodes at 14 days after transplanting and Cyclone 505 EC (Active ingredients: Chlorpyrifos and Cypermethrin) was used to control stem borer incidence at 30 days before harvesting. Irrigation and weeding were done whenever necessary throughout the crop growth period.

All genotypes were characterized according to descriptors established by the International Rice Research Institute and the International Board for Plant Genetic Resources (Bioversity International 2007). Five representative plants in each genotype were tagged randomly for recording the observations. A total of 15 quantitative and 13 qualitative traits were measured (Appendix Figure A3). Methods of measurement and evaluation phase are described in Appendix Table A3. The assessment of amylose content was done in the Laboratory of Biotechnology Centre, Ministry of Agriculture, Livestock, and Irrigation, Yangon, Myanmar. The iodine colorimetric method based on the absorbance spectra using a UV spectrophotometer was used to determine the value of AC in rice genotypes (Avaro et al. 2011).

4.2.3 Data analysis

For quantitative traits, all the basic statistical parameters and analysis of variance were generated using PBTools v1.4 (PBTools 2014). Correlations and ordination (principal components analysis) analyses were performed using SPSS v22 statistical software (IBM SPSS, Inc., Chicago, USA), R package, FactoMineR (Husson et al. 2020), and factextra (Kassambara & Mundt 2017).

Shannon-Weiner information index was computed by using the phenotypic frequencies of qualitative characters in order to estimate the diversity in different clusters. An arbitrary scale was adapted to categorize the computed indices into maximum ($H' = 1.00$), high ($H' = 0.76–0.99$), moderate ($H' = 0.46–0.75$) and low diversity ($H' = 0.01–0.45$) (Jamago & Cortes 2016). The index (H') was calculated by:

$$H' = - \frac{\sum p_i * \log_2 p_i}{\log_2 k}$$

where p_i is frequency proportion of the descriptor state and k = number of states. The H' for each trait was normalized by its maximum value ($\log_2 k$) that ensured that the diversity index ranged from 0 to 1. Cluster analysis and principal component analysis were carried out on qualitative morphological traits using R package factextra (Kassambara & Mundt 2017).

4.3 Genotypic characterization of rice varieties

4.3.1 Plant materials and DNA extraction

The same set of 117 rice genotypes from phenotypic characterization was used for genomic studies. Young healthy leaf tissue was collected from 3-week-old rice plants and stored on silica gel in plastic tubes for desiccation. DNA extraction was performed in the Laboratory of Molecular Biology of Faculty of Tropical AgriSciences, Czech University of Life Sciences Prague (CZU). DNA was isolated from dried leaves using the CTAB method developed by Doyle and Doyle (1987) and modified by Faleiro et al. (2002). The content and purity of DNA were measured on a Nanodrop (Thermo Scientific, Waltham, MA, USA) spectrophotometer. DNA quality of the samples was controlled by incubating 1 μ L of DNA in restriction enzyme buffer at 37 °C for 2 hrs and resolving the DNA on a 0.8% agarose gel in 1x TAE buffer. The DNA concentration was adjusted within the range of 50–100 η g/ μ L.

4.3.2 Genotyping of individual DNA samples using DArTseq technology

A total of 20 μ L of each sample with a concentration of 100 η g/ μ L DNA were sent to Diversity Arrays Technology (DArT) Pty. Ltd., Bruce, Australia (<https://www.diversityarrays.com>, accessed November 2021) for whole-genome scans using a combination of DArT complexity reduction method and next-generation sequencing platform. Whole-genome genotyping for the 117 rice genotypes was carried out using DArTseq technology as described by Barilli et al. (2018) using 18,271 SNPs and 16,160 silicoDArT markers (Appendix Table A4 and Appendix Table A5). This method involved the digestion of DNA samples with a rare cutting enzyme, PstI, paired with a set of secondary

frequently cutting restriction endonucleases, ligation with site-specific adapters, and amplification of adapter-ligated fragments. The DNA samples were processed in digestion/ligation reactions according to the prescribed standard procedures of Kilian et al. (2012) but replacing a single PstI-compatible adapter with two different adapters corresponding to two different restriction enzymes overhangs. The genomic representations were generated following the procedures described by Kilian et al. (2012) and Barilli et al. (2018). PstI-MseI was selected as the appropriate complexity reduction method for rice and consequently next-generation sequencing technology using HiSeq2500 (Illumina, San Diego, CA, USA) was employed to detect SNPs and silicoDArT markers. The sequence data were analyzed using DArTsoft14 and silicoDArT (presence/absence of markers in genomic representations) were scored '1' for presence, and '0' for absence.

4.3.3 Data analysis

Quality analysis of marker data

The following parameters for the DArT marker assaying pipeline for quality control were used for marker screening: reproducibility (%), call rate (%), polymorphism information content (PIC), and one ratio (Kilian et al. 2012). A total of 18,271 SNP and 16,160 silicoDArT markers were reported, of which 7,643 SNP and 4,064 silicoDArT markers were considered for analyses after filtering with quality control parameters including >95% reproducibility, >95% call rate, and >0.1 one ratio. Scoring of reproducibility involved the proportion of technical replicate assay pairs for which the marker score exhibited consistency. The call rate determined the success of reading the marker sequence across the samples and was estimated from the percentage of samples for which the score was either 0 or 1. PIC is the degree of diversity of the marker in the population and it showed the usefulness of the marker for linkage analysis. One ratio constitutes the proportion of the samples for which genotype scores equaled 1 (Alam et al. 2018).

Genetic relationship among genotypes and population structure

Genetic dissimilarity matrices were constructed in DARwin v. 6.0.21 to identify the genetic relationships among the genotypes (Perrier et al. 2003). Weighted neighbor-joining dendrograms were constructed in both marker (7,643 SNP and 4,064 silicoDArT) platforms. Clade strength in the dendrograms was tested by 10,000 bootstrap analyses. Principal component analysis for 7,643 SNP and 4,064 silicoDArT markers was conducted by R package factoextra (Kassambara & Mundt 2017). Genetic structure using STRUCTURE v.2.3.4 among 117 rice genotypes using 7,643 DArTseq-derived SNP markers was investigated (Pritchard et al. 2010). Bayesian clustering method was applied to identify clusters of genetically similar individuals using STRUCTURE. The parameters used were burn-in period of 50,000 steps followed by 100,000 Monte Carlo Markov Chain iterations, admixture model with correlated frequencies, K varying from 1 to 5, and three runs per K value in order to obtain consistent results. The log-likelihood of the observed data for each K value was calculated and compared across the range of K values. The best K value was estimated based on the membership coefficient (Q) for each individual in each cluster. The Q

values indicate the level of relatedness of each genotype to various subgroups. The STRUCTURE results were subsequently analyzed by the STRUCTURE HARVESTER application to identify the best value of K (Earl & vonHoldt 2012).

Analysis of molecular variance (AMOVA) and genetic diversity indices

The number of clusters determined with STRUCTURE using 7,643 SNPs was used for AMOVA and the calculation of Nei's genetic distance using GenAlEx v6.503 (Peakall & Smouse 2012). From AMOVA, the F_{st} (fixation index) and N_m (haploid number of migrants) within the population were obtained. In addition, genetic indices such as number of loci with private allele, number of different alleles (N_a), number of effective alleles (N_e), Shannon's information index (I), observed heterozygosity (H_o), and expected heterozygosity (H_e) were also calculated using GenAlEx v6.503 (Peakall & Smouse 2012).

5. RESULTS

5.1 On-farm rice diversity and farmers' preferences for varietal attributes in Ayeyarwady Delta, Myanmar

5.1.1 Demographic characteristics of the selected farmers

The average respondents' age was 48 years, with 24 years of experience in rice farming (Table 1). The average number of years of schooling was six, in reference to an education of secondary level. Farmers owned various sizes of farms (0.4 to 81 ha), with the average and median size of landholding being 7.9 ha and 5.3 ha, respectively. Some farmers cultivated rice during both the monsoon and summer seasons, whereas most farmers cultivated only monsoon rice, depending on the agro-ecological region.

5.1.2 The on-farm rice varietal diversity

On average, farmers grew two varieties on their farms during the monsoon season. As shown in Appendix Table A6, 83% percent of the interviewed farmers cultivated more than one variety, with the majority of the farmers growing two to four varieties. Thirty-nine different varieties, i.e., nine HYVs and 30 traditional varieties, were identified by the farmer-given names in this survey. A total number of 21, 12, and 27 varieties were observed in fresh, brackish, and saline agro-ecological regions, respectively. The average yields of HYVs, Pawsan varieties and other traditional varieties were 2.8 t/ha, 2.6 t/ha, and 2.7 t/ha, respectively (Table 2). The Pawsan varieties group was the most popular among the farmers and those varieties were grown by all farmers at least on a portion of their land (61% of the survey area was devoted to Pawsan group). The yield of these varieties was influenced by the agroecological region and was higher ($p < 0.05$) in the brackish-water zone compared with the fresh-water zone. Farmers were well aware of the advantages of local varieties, namely, good market value, adaptability to harsh weather conditions, good eating quality, resistance to lodging, low production costs, and stable yield (Appendix Table A7).

A larger number of rice varieties per farm were used by the farmers in the saline and brackish-water regions compared with the fresh-water region (Table 2) and on soil described as low to medium fertile. Similarly, farmers in the undulated type of terrain usually used more varieties simultaneously compared to farmers in the flat terrain.

The regression model (Table 3), used to explain the use of more rice varieties (more than two) by the socio-economic factors ($\chi^2 = 41.058$, $p < 0.001$), accounted for 32.8% of variability (Nagelkerke R^2) and classified correctly 68% of the cases. Farmers' experience and education, agroecological zone, soil fertility and terrain were the main predictors of the use of more than two rice varieties on farmers' fields.

Farmers with longer farming experience and higher education (in number of years) grew more than two rice varieties on their fields when compared to less experienced and less educated farmers. Furthermore, more farmers growing at least two varieties belonged to the saline-water agro-ecological region compared to the fresh and brackish-water regions (Table

3). The use of at least two rice varieties was also more common in the undulated type of terrain and lower fertility soils compared to flatland type and fertile soils, where growing only one rice variety was more common. The average number of varieties per farmer was 2.55, 2.25, and 2.06 in low, medium, and high soil-fertility areas, respectively (Table 2).

Table 1. Physical and socio-economic characteristics of the survey farmers in five selected townships

Characteristics	Study sites				
	Bogale	Mawlamyinegyun	Labutta	Myaungmya	Pathein
No. of respondents	30	31	30	29	30
Average age (years)	45	47	52	45	51
Education (years)	6	6	6	6	7
Farming experience (years)	20	23	28	23	27
Average sown area (ha)	8.1	5.2	14.5	5.1	6.4
No. of growing cycles (%)					
Two rice cultivations	-	77	-	100	80
One rice cultivation	100	23	100	-	20
Agro-ecological region (%)					
Fresh	-	35	-	31	100
Brackish	-	-	-	69	-
Saline	100	65	100	-	-
Soil texture (%)					
Clay	80	65	90	59	70
Loam	17	29	7	31	20
Sand	3	-	3	3	-
Sandy loam	-	6	-	7	10
Soil fertility (%)					
High	23	16	27	28	17
Average	50	77	60	59	70
Low	27	6	13	14	13

Table 2. Number of grown varieties per farmer, total sown area and yields of Pawsan varieties, other traditional varieties and high-yielding varieties

Variable	No. ¹	Total		High-yielding varieties			Pawsan varieties			Traditional varieties ³		
		Sown area ²	Yield	No.	Sown area	Yield	No.	Sown area	Yield	No.	Sown area	Yield
		(ha)	(t/ha)		(ha)	(t/ha)		(ha)	(t/ha)		(ha)	(t/ha)
Agro-ecological zones												
Fresh	2.06b ⁴	5.06b	2.60b	0.46b	1.31b	2.93	1.04a	2.83b	2.43b	0.56b	0.90b	2.72
Brackish	2.10ab	4.92b	3.22a	0.45b	2.40a	3.49	0.65b	1.71b	2.85a	1.00a	0.79b	2.94
Saline	2.41a	9.91a	2.63b	1.11a	0.01c	2.00	1.29a	6.65a	2.54ab	0.13c	3.23a	2.69
Soil fertility												
Low fertility	2.55a	7.10	2.61	0.91	0.82	3.21ab	1.14	4.03	2.36	0.50	2.22	2.50
Medium fertility	2.25ab	6.95	2.67	0.79	0.73	2.87b	1.17	4.34	2.56	0.29	1.86	2.74
High fertility	2.06b	9.91	2.85	0.79	0.79	3.78a	0.98	6.26	2.58	0.30	2.84	2.80
Terrain												
Flat	2.09b	6.97b	2.73	0.78	0.36	3.32	1.08	4.56	2.47	0.23	2.02	2.81
Undulated	2.38a	8.13a	2.67	0.82	1.06	3.04	1.15	4.84	2.59	0.40	2.21	2.65
Soil texture												
Clay	2.25	7.74	2.70	0.86	0.55	2.92	1.14	4.88	2.57	0.25	2.29	2.73
Loam	2.32	6.89	2.71	0.65	1.47	3.42	1.10	4.12	2.45	0.58	1.28	2.74
Sand	2.67	21.1	2.29	1.33	1.33	3.11	-	11.87	2.16	0.33	7.83	2.04
Sandy loam	1.86	3.34	2.80	0.43	0.63	3.67	1.00	1.82	2.55	0.43	0.86	2.77

¹ the number of varieties grown per farmer; ² the sown area per farmer; ³ Traditional varieties other than Pawsan group

⁴ Different letters indicate differences ($p < 0.05$, Kruskal-Wallis) between subcategories within the same groups of rice varieties.

Table 3. Model using the socioeconomic variables to explain the use of more varieties (more than two) was significant ($\chi^2=41.058$, $p<0.001$). The model explained 32.8% (Nagelkerke R^2) of the variance in the use of more (>2) rice varieties and classified correctly the 68% of the cases.

Variable	Coefficient	Standard error	<i>p-value</i>
Experience	0.058	0.027	0.031
Agro-ecological zone ¹			0.008
Brackish	0.831	0.694	0.232
Saline	2.103	0.683	0.002
Age	0.008	0.030	0.796
Education	0.154	0.076	0.044
Total sown area	0.016	0.019	0.414
Soil texture ²			0.244
Loam	0.848	0.504	0.117
Sand	0.264	1.346	0.845
Sandy loam	-1.502	1.315	0.253
Soil fertility ³			0.006
High	-2.323	0.752	0.002
Medium	-1.807	0.635	0.004
Terrain	1.121	0.427	0.009
No. of cultivation cycles	-0.852	0.619	0.168
Constant	-3.507	1.450	0.016

¹ Brackish and saline agro-ecological regions were compared to fresh water region; ² loam, sand and sandy loam texture was compared to clay texture; ³ high and medium fertility soils were compared to low fertility soils.

5.1.3 The adoption of high-yielding varieties

Nine HYVs were identified; however, the percentage of the farmers growing these varieties was low in the saline-water regions (0.1%), whereas 27.8% and 48.8% of the land was under HYVs in the fresh and brackish-water regions, respectively. The most commonly reported HYVs across all agro-ecological zones were Ayeyarpadaethar, Shwe War Yin, Manawthukha, and Thee Htat Yin (Appendix Table A8).

The likeliness of the farmers to adopt HYVs was evaluated using the logit model, with socio-economic characteristics used to describe the use of HYVs at least on some part of the agricultural land. The significant model ($\chi^2 = 78.572$, $p < 0.001$) classified correctly 90% of the cases and explained 62% of variability (Nagelkerke R^2), with farming experience, agro-ecological zone, education, total sown area, and soil fertility being the most pertinent variables of the model (the *p*-values are given in Table 4).

According to the model, farmers with less farming experience and less education were more likely to adopt HYVs. The probability of the farmers adopting HYVs also increased with an increase in the total rice area per farmer (Table 4). Furthermore, farmers, who had classified the fertility of the soil as low, were more likely to use HYVs at least on part of their land. The yields of HYVs were the highest on the soils classified as “high fertility soils” but did not follow a linear trend with decreasing soil fertility.

Table 4. Model using the socioeconomic variables to explain the use or not of HYV (the adoption likelihood) was significant ($\chi^2 = 78.572$, $p < 0.001$). The model explained 62.0% (Nagelkerke R^2) of the variance in the selection of HYV and classified correctly the 90% of the cases.

Variable	Coefficient	Standard error	<i>p-value</i>
Experience	-0.067	0.037	0.068
No. of cropping cycles	-1.036	1.338	0.439
Family members	0.027	0.221	0.903
Agro-ecological zone ¹			0.002
Brackish	1.529	0.725	0.035
Saline	-4.995	1.753	0.004
Age	0.058	0.042	0.167
Education	-0.198	0.110	0.072
Total sown area	0.078	0.037	0.034
Soil texture ²			0.365
Loam	0.802	0.654	0.220
Sand	0.930	2.182	0.670
Sandy loam	-1.524	1.370	0.266
Soil fertility ³			0.074
High	-2.369	1.166	0.042
Medium	-2.278	1.027	0.027
Terrain	0.133	0.597	0.824
Constant	0.901	2.131	0.672

¹ Brackish and saline agro-ecological regions were compared to fresh water region; ² loam, sand and sandy loam texture was compared to clay texture; ³ high and medium fertility soils were compared to low fertility soils.

5.1.4 The main cultivation constraints

The main rice cultivation constraints reported by the farmers were grouped into agro-ecological, technical, and socio-economic constraints (Table 5). Mean scores for the severity of the rice cultivation constraints given by farmers were calculated and nine problems were identified as the most important: flooding, rainfall during harvest, extreme water conditions, and abnormal weather (agro-ecological constraints); pest infestation, labor scarcity, rodent infestation, and poor germination rate were identified as the main technical rice cultivation constraints, whereas inadequate input was the main socio-economic constraint. The mean score given to each constraint group was 0.159 for agro-ecological constraints, 0.314 for technical problems, and 0.056 for socio-economic problems.

Farmers in the freshwater agro-ecological region perceived flooding as a more important problem limiting rice cultivation (score = 1.180), whereas farmers from the saline region reported flooding less frequently or of less importance (score = 0.550). On the other hand, pest infestation seemed to be the most important constraints in the saline region (score = 1.713) compared with the freshwater regions (score = 1.020). The labor scarcity was severe in the fresh-water region (score = 1.28); however, the significance of labor scarcity was more obvious in the brackish-water region (score = 2.1). Soil fertility did not influence the majority of the problems reported by the farmers, except for labor scarcity and rodent infestation (Table 5). Logically, only farmers in the undulated type of terrain reported poor leveling as a significant problem (score = 0.118). Farmers who cultivated on sandy loam soils did not report rainfall during harvest as a constraint (unlike farmers who cultivated other soil types) but identified the lack of access to water as a constraint limiting rice cultivation (score = 0.857).

Additionally, cultivation problems reported by the farmers were used to explain the selection of rice varieties, but no significant logistic regression model was found. On the other hand, the correspondence analysis (Figure 9a) of nine most important cultivation constraints (scores>0.1) and rice varieties revealed a relationship between both variables ($\chi^2 = 397.289$, $p = 0.002$). No correlation between problems and the adoption of HYVs or Pawsan group varieties was found, and only the use of other traditional varieties (Figure 9b) seemed to be influenced by the cultivation problems reported by the farmers ($\chi^2 = 186.127$, $p = 0.077$).

Table 5. The scores of main rice cultivation problems given by the farmers (3= the most severe, 1= the least important, 0=not considered a constraint by the farmer)

Constraints	Total (n=150)	Agro-ecological zone			Soil fertility			Terrain		Soil texture			
		Fresh (n=50)	Brackish (n=20)	Saline (n=80)	High (n=33)	Medium (n=95)	Low (n=22)	Flat (n=65)	Undulat ed (n=85)	Clay (n=109)	Loam (n=31)	Sand (n=3)	Sandy loam (n=7)
Agro-ecological	0.159	0.198	0.120	0.145	0.155	0.155	0.186	0.154	0.164	0.165	0.132	0.133	0.200
Flooding	0.773A ¹	1.180a ²	0.650ab	0.550b	0.848	0.758	0.727	0.754	0.788	0.789	0.710	0.000	1.143
Raining during harvest	0.160BC	0.160	0.200	0.150	0.121	0.168	0.182	0.062	0.235	0.110ab	0.355a	0.333a	0.000b
Separate land	0.007C	0.000	0.000	0.013	0.000	0.011	0.000	0.000	0.012	0.009	0.000	0.000	0.000
Poor levelling	0.067C	0.020	0.000	0.113	0.061	0.074	0.045	0.000b	0.118a	0.092	0.000	0.000	0.000
Sea water intrusion	0.067C	0.000	0.000	0.125	0.152	0.021	0.136	0.046	0.082	0.092	0.000	0.000	0.000
Mudding after flooding	0.013C	0.000	0.000	0.025	0.000	0.011	0.045	0.015	0.012	0.009	0.032	0.000	0.000
Extreme water conditions	0.107BC	0.060	0.000	0.163	0.030	0.137	0.091	0.154	0.071	0.147	0.000	0.000	0.000
Drought	0.033C	0.000	0.000	0.063	0.030	0.042	0.000	0.031	0.035	0.046	0.000	0.000	0.000
Lack of water access	0.040C	0.120	0.000	0.000	0.091	0.032	0.000	0.092	0.000	0.000b	0.000b	0.000b	0.857a
Abnormal weather	0.327B	0.440	0.350	0.250	0.212	0.295	0.636	0.385	0.282	0.358	0.226	1.000	0.000
Technical	0.314	0.258	0.365	0.335	0.264	0.331	0.314	0.322	0.307	0.319	0.310	0.400	0.200
Pest infestation	1.400A	1.020b	1.100ab	1.713a	1.303	1.421	1.455	1.338	1.447	1.358	1.710	2.333	0.286
Labor scarcity	1.233A	1.280ab	2.100a	0.988b	0.727b	1.453a	1.045ab	1.400	1.106	1.312	0.871	1.000	1.714
Rodent infestation	0.147B	0.000b	0.000b	0.275a	0.303ab	0.053b	0.318a	0.231	0.082	0.147	0.129	0.667	0.000
Crab incidence	0.047B	0.060	0.000	0.050	0.091	0.011	0.136	0.015	0.071	0.009	0.194	0.000	0.000
No access to quality seeds	0.047B	0.080	0.000	0.038	0.091	0.032	0.045	0.015	0.071	0.037	0.097	0.000	0.000
Slow growth	0.027B	0.000	0.100	0.025	0.061	0.021	0.000	0.031	0.024	0.037	0.000	0.000	0.000
Poor soil quality	0.080B	0.100	0.100	0.063	0.000	0.105	0.091	0.123	0.047	0.083	0.097	0.000	0.000
Weed problem	0.027B	0.000	0.000	0.050	0.000	0.042	0.000	0.000	0.047	0.037	0.000	0.000	0.000
Poor germination rate	0.120B	0.020	0.250	0.150	0.061	0.158	0.045	0.062	0.165	0.165	0.000	0.000	0.000
Impure seeds	0.007B	0.020	0.000	0.000	0.000	0.011	0.000	0.000	0.012	0.009	0.000	0.000	0.000
Socio-economic	0.056	0.048	0.020	0.070	0.115	0.036	0.054	0.058	0.054	0.055	0.065	0.000	0.057
Inaccessible machinery	0.067	0.080	0.000	0.075	0.182	0.032	0.045	0.092	0.047	0.046	0.097	0.000	0.286
Poor market	0.020	0.000	0.000	0.038	0.030	0.021	0.000	0.015	0.024	0.028	0.000	0.000	0.000
High inputs	0.020	0.000	0.000	0.038	0.091	0.000	0.000	0.046	0.000	0.028	0.000	0.000	0.000
Inadequate credit	0.067	0.040	0.100	0.075	0.061	0.063	0.091	0.062	0.071	0.037	0.194	0.000	0.000
Inadequate inputs	0.107	0.120	0.000	0.125	0.212	0.063	0.136	0.077	0.129	0.138	0.032	0.000	0.000

¹ Different capital letters indicated differences ($p < 0.05$) between given scores within the constraint category (Agro-ecological, technical and socio-economic constraints)

² Different lower-case letters indicate statistically significant differences ($p < 0.05$, Kruskal-Wallis) between scores given by the farmers in different regions within the main four categories (Agro-ecological zone, soil fertility, terrain, soil texture). No letters indicate no statistically significant differences within category.

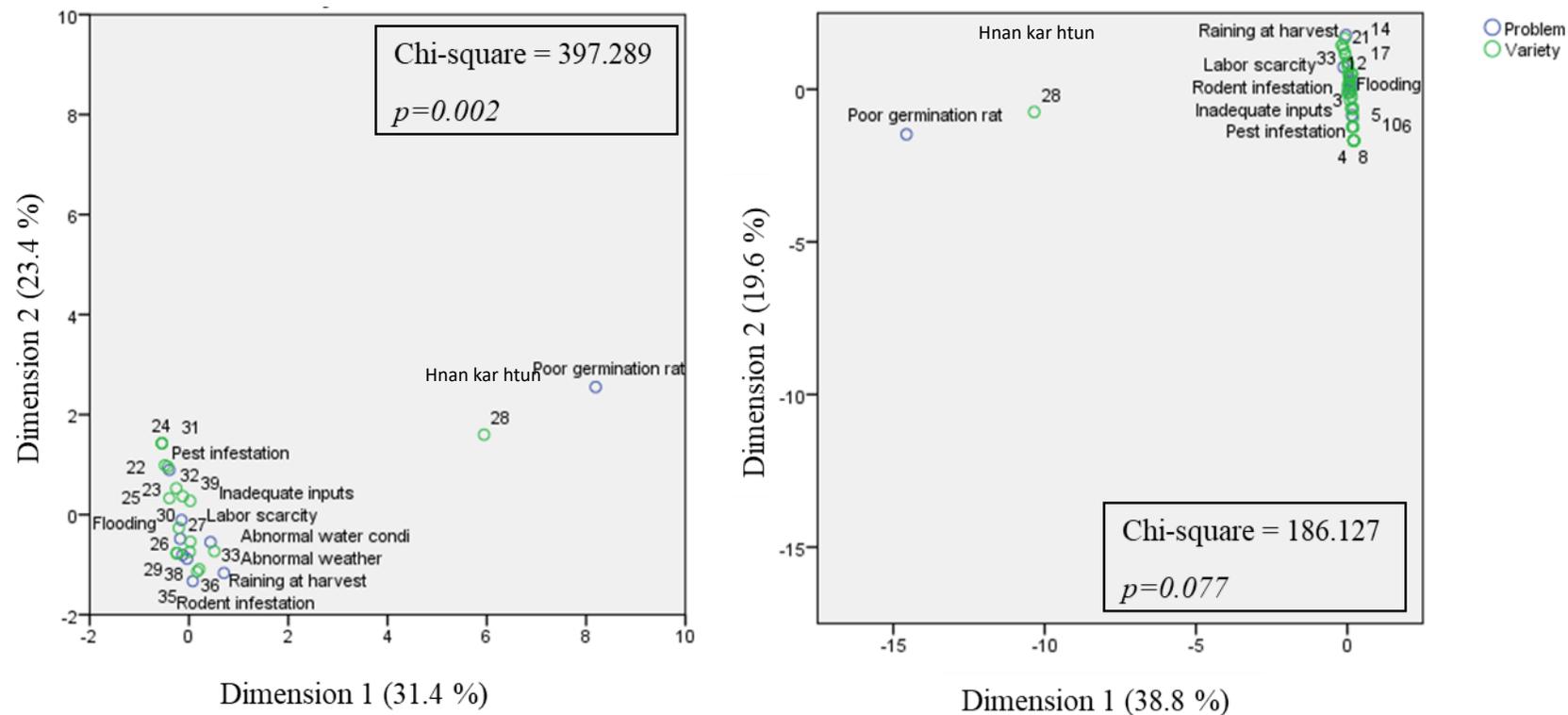


Figure 9. Correspondence analysis (CA) of nine main rice cultivation problems and all cultivated varieties (n=39) (a), and traditional varieties others than Pawsan group rice (n=20) (b)

5.1.5 Rice characteristics valued by farmers

Farmers gave a score of 2.504, 2.394, and 2.621, respectively, to production characteristics, resistance to stress and grain characteristics (Table 6). Within the production characteristics, low production costs (2.947), high yield (2.900), resistance to grain shattering (2.900) and resistance to lodging (2.860) were the most appreciated rice traits. Resistance to insects (2.893) and resistance to diseases (2.893) were the most important traits among the characteristics linked to the resistance to stress. Among grain characteristics, high market demand for specific rice grains scored the highest among the traits (3.000), followed by high milling recovery (2.933), low amount of broken grains (2.933) and taste (2.840). The importance of the resistance to insects and of the stickiness of cooked rice increased with decreasing soil fertility, and rice aroma was more valued by the farmers from the undulated areas compared to the farmers from flatlands.

Three regression models were constructed to predict the use of traditional varieties, HYVs and Pawsan group varieties. While no model for the use of HYVs and Pawsan varieties was significant, the use of traditional varieties (other than Pawsan) was positively related to the scores given to resistance to drought, high volume expansion, and the filling value (the characteristic valued by the farmers) ($\chi^2 = 41.058$, $p = 0.007$) (Table 7). On the other hand, farmers, who placed high values on the low amount of broken grains and the properties of cooked rice with high values, were more likely to adopt HYVs.

Table 6. The scores of the rice traits given by the farmers (3= the most appreciated, 1= the least appreciated)

Rice traits	Total (n=150)	Agro-ecological zone			Soil fertility			Terrain		Soil texture			
		Fresh (n=50)	Brackish (n=20)	Saline (n=80)	High (n=33)	Medium (n=95)	Low (n=22)	Flat (n=65)	Undulated (n=85)	Clay (n=109)	Loam (n=31)	Sand (n=3)	Sandy loam (n=7)
Production characteristics	2.504	2.520	2.486	2.498	2.468	2.510	2.532	2.486	2.518	2.509	2.461	2.571	2.592
High yield	2.900A ¹	2.960	2.850	2.875	2.909	2.884	2.955	2.908	2.894	2.890	2.903	3.000	3.000
Low production costs	2.947A	2.960	2.900	2.950	2.939	2.947	2.955	2.939	2.953	2.936	2.968	3.000	3.000
High straw production	1.413D	1.380	1.600	1.388	1.303	1.411	1.591	1.385	1.435	1.431	1.258	1.333	1.857
Short growing period	1.793C	1.840	1.800	1.763	1.636	1.832	1.864	1.846	1.753	1.752	1.903	2.000	1.857
Resistance to lodging	2.860AB	2.900	2.900	2.825	2.879	2.842	2.909	2.800	2.906	2.881	2.742	3.000	3.000
Resistance to shattering	2.900AB	2.860	2.850	2.938	2.849	2.905	2.955	2.892	2.906	2.917	2.807	3.000	3.000
High tillering ability	2.713B	2.740	2.500	2.750	2.758	2.747	2.500	2.631	2.777	2.752	2.645	2.667	2.429
Resistance to stress	2.394	2.313	2.442	2.433	2.237	2.430	2.477	2.403	2.388	2.401	2.376	2.389	2.381
Resistance to insects	2.893A	2.900	2.900	2.888	2.758b ²	2.926ab	2.955a	2.846	2.929	2.890	2.871	3.000	3.000
Resistance to diseases	2.893A	2.900	2.950	2.875	2.788	2.916	2.955	2.862	2.918	2.872	2.936	3.000	3.000
Resistance to drought	2.093C	1.880	2.200	2.200	1.970	2.105	2.227	2.246	1.977	2.101	2.065	2.333	2.000
Tolerance to flooding	2.540B	2.520	2.550	2.550	2.455	2.558	2.591	2.554	2.529	2.569	2.387	2.667	2.714
Tolerance to salinity	2.200C	1.980	2.300	2.313	2.091	2.168	2.500	2.169	2.224	2.211	2.226	2.000	2.000
Tolerance to cold injury	1.747D	1.700	1.750	1.775	1.364b	1.905a	1.636ab	1.739	1.753	1.762	1.774	1.333	1.571
Grain characteristics	2.621	2.578	2.595	2.653	2.565	2.633	2.653	2.589	2.645	2.625	2.592	2.879	2.571
High milling recovery	2.933AB	2.940	2.900	2.938	2.970	2.926	2.909	2.923	2.941	2.936	2.968	3.000	2.714
High demand	3.000A	3.000	3.000	3.000	3.000	3.000	3.000	3.000	3.000	3.000	3.000	3.000	3.000
Softness	2.760B	2.740	2.800	2.763	2.697	2.758	2.864	2.708	2.800	2.780	2.742	3.000	2.429
Short cooking time	1.747D	1.680	1.750	1.788	1.697	1.705	2.000	1.862	1.659	1.651b	1.871ab	2.667a	2.286ab
Taste	2.840AB	2.800	2.850	2.863	2.879	2.863	2.682	2.754b	2.906a	2.853	2.839	3.000	2.571
Aroma	2.733B	2.680	2.700	2.775	2.606	2.790	2.682	2.600b	2.835a	2.762	2.645	3.000	2.571
Stickiness of cooked rice	2.393C	2.320	2.400	2.438	2.000b	2.463a	2.682a	2.277	2.482	2.431	2.161	3.000	2.571
Longer keeping quality	2.747B	2.680	2.650	2.813	2.788	2.737	2.727	2.646	2.824	2.725	2.807	2.667	2.857
High volume expansion	2.740B	2.640	2.700	2.813	2.849	2.705	2.727	2.708	2.765	2.771a	2.742a	2.667ab	2.286b
Hard-texture white rice with high amylose	2.000D	1.920	1.850	2.088	1.788	2.074	2.000	2.092	1.929	2.037	1.807	2.667	2.000
Less broken grains	2.933AB	2.960	2.950	2.913	2.939	2.937	2.909	2.908	2.953	2.927	2.936	3.000	3.000

¹ Different capital letters indicated differences ($p < 0.05$) between given scores within the trait category (production characteristics, resistance to stress, grain characteristics)

² Different lower-case letters indicate statistically significant differences ($p < 0.05$, Kruskal-Wallis) between scores given by the farmers in different regions within the main four categories (Agro-ecological zone, soil fertility, terrain, soil texture). No letters indicate no statistically significant differences within category.

Table 7. Model using the rice traits preferred by the farmers to explain the adoption of traditional varieties (other than Pawsan group) was significant ($\chi^2=41.058$, $p=0.007$). The model explained 34.1 % (Nagelkerke R^2) of the variance in the selection of traditional varieties and classified correctly the 75.3% of the cases.

Rice traits	Coefficient	Standard error	<i>p-value</i>
Production characteristics			
High yield	-0.285	0.801	0.722
Low production costs	0.355	1.157	0.759
High straw production	0.948	0.969	0.328
Short growing period	0.182	0.590	0.758
Resistance to lodging	-0.842	0.814	0.301
Resistance to shattering	0.710	0.925	0.443
High tillering	-0.214	0.520	0.681
Resistance to stress			
Resistance to insects	-0.812	1.389	0.559
Resistance to diseases	0.073	1.395	0.958
Resistance to drought	0.824	0.486	0.090
Tolerance to flooding	0.457	0.475	0.336
Tolerance to salinity	-1.156	0.501	0.021
Tolerance to cold injury	0.487	0.604	0.420
Grain characteristics			
High milling recovery	0.152	0.891	0.865
Softness	-0.429	0.517	0.407
Short cooking time	0.806	0.591	0.173
Taste	0.123	0.731	0.867
Aroma	0.615	0.625	0.325
Stickiness of cooked rice	-0.641	0.461	0.164
Longer keeping quality	-1.320	0.620	0.033
High volume expansion	2.168	0.578	0.000
Hard-texture white rice with high amylose	1.113	0.493	0.024
Less broken grains	-1.198	1.175	0.092
Constant	1.876	1.806	0.299

5.2 Phenotypic characterization of rice varieties

5.2.1 Diversity in quantitative traits

Considering the 15 quantitative traits analyzed in this study, significant phenotypic variation among genotypes were found for 13 traits (Table 8). Days to heading (DTH) of rice genotypes varied from 66–114 days with low coefficient of variation (CV) of 9.07%; only six genotypes fell in the shortest duration group (66–80 days) (Appendix Table A9). Culm length (CL) at reproductive stage ranged from 65.80 cm to >120 cm and 56% of the studied genotypes fell within the range of 91–120 cm and 41% fell in 71–90 cm. All genotypes have medium length of the panicle (PL) (21–30cm), except the genotype SHTUN (31.07 cm) and low CV of 8.71%. Sterile lemma length (short and long) ranged from short (≤ 1.5 mm) to long (>2.5 mm but shorter than the lemma) with high CV of 39.13% for short and 38.9% for long sterile lemma; 7 genotypes has long sterile lemma. The number of tillers per plant (NTPP) varied from 10 to 36 with a mean of 21. The number of panicles per plant (NPP) ranged from 6 to 33 with a mean of 16. No significant variations were observed for NTPP and NPP. The number of spikelets per panicle (NSPP) varied from 382 to 1510 with a mean of 827. TGW (1000-grain wt.) ranged from 16.7 to 35.2 g and most of the genotypes had 26–30 g TGW. Grain length (GL) varied from 6.58 mm to 11.15 mm. Grain width (GW) varied from 2.2 mm to 4.14 mm. Length width ratio (LWR) ranged from 1.77 to 4.32. Filled grain % (FG %) ranged from 45–96 % and 46% of rice genotypes found highly fertile ($>90\%$ filled grains). Harvest index (HI) varied from 0.13–0.85 and 60% genotypes fell within the range of 0.5–0.6. Forty-nine percent of genotypes produced 0.2–0.3 kg rice grain yield (Y) for 5 hills and high CV of 38.09%. The heritability (H^2) for the traits ranged from 62 to 98% and high heritability ($>70\%$) was observed for almost all the traits (Table 8). The contribution of genotypic variance to the total phenotypic variance was significant for almost all traits.

Principal component analysis (PCA) was employed to reduce the complexity of the data set while retaining the variation within the data set as far as possible. PCA using 15 quantitative traits produced five principal components (eigenvalue >1) which cumulatively accounted for 73% of the total phenotypic variance (Table 9). The first component accounted for 22.6% of the total variation in the data set. The traits with high positive loadings are grain width (GW), short sterile lemma (SSL), 1000-grain weight (TGW), and long sterile lemma (LSL). The PC2 explained an additional 21% of the total variance. The traits such as yield (Y), harvest index (HI), days to heading (DTH), and panicle length (PL) showed high positive loadings. PC3 accounted for 10.46% of the total variance. The characters grain length (GL), panicle length (PL), 1000-grain weight (TGW), and length width ratio (LWR) contributed more to the total variance in PC3. PC4 explained 10.17% of the total variance and number of panicles per plant (NPP), number of tillers per plant (NTPP), grain length (GL), and filled grain % (FG%) had a strong effect in variation, whereas in PC5, culm length (CL) and number of spikelets per panicle (NSPP) contributed more to the total variance.

Among the 15 different quantitative agro-morphological traits studied, the highest correlation corresponded to the LSL and SSL ($r = 0.98$). Traits presenting highly significant correlation with these two characteristics were GL and LWR ($r = 0.85$), HI and Y ($r = 0.73$)

and TGW and GW ($r = 0.62$). Grain length width ratio (LWR) negatively correlated with grain width (GW) ($r = -0.87$), grain length (GL) negatively correlated with the grain width (GW) ($r = -0.51$) (Table 10). In agreement with correlation analysis, the variables PCA plot showed that positively correlated variables are grouped together and negatively correlated variables are positioned on opposite sides of the plot origin (Figure 10).

Phylogenetic relationships (the cluster analysis) for quantitative traits using the Euclidean distance showed three major clusters out of which 4 rice genotypes were grouped together (TPBGYAR, NKTPYAN, KKTPYAN, TPYIN) in cluster-I, 35 genotypes including 13 Pawsan varieties and 22 traditional varieties formed together in cluster-II, and rest of the genotypes formed a very large cluster-III comprising 2 Pawsan varieties, 5 HYVs, and 71 traditional varieties. A careful observation on the cluster-III revealed that it was divided further into seven sub-clusters (sub-cluster-III.1 to sub-cluster-III.7.) Three-check HYVs (C1, C2, and C3) formed together in sub-cluster III.1, whereas C4 and C5 in sub-cluster-III. 2 and 4 (Figure 11).

Table 8. Descriptive statistics for 15 quantitative traits in 117 Myanmar rice genotypes

Traits	Mean± SE	F-value	Pr(>F)	CV (%)	H ²
DTH	96.55±0.86	25.59***	0.0000	9.70	0.96
CL (cm)	94.39±1.18	7.44***	0.0000	14.97	0.80
PL (cm)	25.78±0.21	12.53***	0.0001	8.71	0.91
LSL(mm)	2.23±0.08	32.91***	0.0000	38.91	0.97
SSL (mm)	2.08±0.08	38.92***	0.0000	39.13	0.97
NTPP	20.64±0.51	0.85	0.7011	27.66	0.00
NPP	16.42±0.37	1.09	0.4520	24.56	0.12
NSPP	827.39±20.00	5.89**	0.0002	26.40	0.80
TGW (g)	26.65±0.34	60.61**	0.0130	14.57	0.98
GL (mm)	8.18±0.08	4.23***	0.0014	10.95	0.75
GW (mm)	3.03±0.04	12.81***	0.0000	13.80	0.90
LWR	2.77±0.05	91.04***	0.0000	21.40	0.98
FG%	87.92±0.71	2.36*	0.0305	8.58	0.62
HI	0.51±0.01	3.16**	0.0161	27.45	0.64
Y (kg)	0.21±0.01	6.90**	0.0003	38.09	0.86

DTH, days to heading; CL, culm length; PL, panicle length; LSL, long sterile lemma; SSL, short sterile lemma; NTPP, no. of tillers per plant; NPP, no. of panicles per plant; NSPP, no. of spikelets per panicle; TGW, thousand grain weight; GL, grain length; GW, grain width; LWR, length-width ratio; FG %, filled grain %; HI, harvest index; Y, yield (5 hills) kg; CV, co-efficient of variation (%); H², heritability

* p< 0.05, **p<0.01, ***p<0.001

Table 9. Eigenvalue of five principal components and factor loadings (eigenvectors) for different quantitative traits

	PC1	PC2	PC3	PC4	PC5
Eigenvalue	3.383	3.155	1.569	1.525	1.288
Percent variance	22.551	21.032	10.460	10.167	8.586
Cumulative percent of total variance	22.551	43.584	54.043	64.210	72.796
Eigenvectors/coefficient vectors					
DTH	0.222	0.627	0.136	-0.044	-0.172
CL (cm)	-0.174	0.265	0.213	0.196	0.818
PL (cm)	0.107	0.598	0.501	0.168	0.048
LSL(mm)	0.587	-0.558	0.304	0.190	0.049
SSL (mm)	0.612	-0.495	0.325	0.198	0.045
NTPP	-0.039	0.028	-0.410	0.744	-0.092
NPP	0.027	-0.114	-0.303	0.816	-0.040
NSPP	-0.277	0.570	-0.041	0.090	0.486
TGW (g)	0.605	0.211	0.432	0.132	-0.045
GL (mm)	-0.637	-0.157	0.587	0.223	-0.133
GW (mm)	0.916	-0.064	-0.025	-0.042	0.146
LWR	-0.869	-0.087	0.365	0.153	-0.199
FG%	0.154	0.282	0.320	0.216	-0.366
HI	0.144	0.752	-0.154	-0.049	-0.349
Y (kg)	0.284	0.845	-0.065	0.113	-0.024

DTH, days to heading; CL, culm length; PL, panicle length; LSL, long sterile lemma; SSL, short sterile lemma; NTPP, no. of tillers per plant; NPP, no. of panicles per plant; NSPP, no. of spikelets per panicle; TGW, thousand grain weight; GL, grain length; GW, grain width; LWR, length-width ratio; FG %, filled grain %; HI, harvest index; Y, yield (5 hills) kg

Table 10. Correlation coefficient matrix of the quantitative characters of 117 Myanmar rice genotypes

	DTH	CL	PL	LSL	SSL	NTPP	NPP	NSPP	TGW	GL	GW	LWR	FG	HI
CL	.07													
PL	.29**	.22*												
LSL	-.14	-.14	-.13											
SSL	-.04	-.12	-.12	.98**										
NTPP	-.04	.01	-.01	-.07	-.06									
NPP	-.12	.02	-.1	.11	.1	.51**								
NSPP	.15	.46**	.27**	-.30**	-.27**	.01	-.03							
TGW	.24**	.04	.45**	.16	.20*	-.03	.	-.25**						
GL	-.13	.13	.08	-.12	-.13	-.02	.02	-.02	-.03					
GW	.09	-.07	.04	.46**	.47**	-.03	.	-.26**	.62**	-.51**				
LWR	-.11	.07	.01	-.26**	-.27**	.01	.	.11	-.41**	.85**	-.87**			
FG	.16	-.08	.26**	.06	.07	.	.03	.11	.18*	-.02	.02	-.01		
HI	.47**	-.13	.32**	-.27**	-.22*	.03	-.07	.28**	.09	-.23*	.01	-.13	.21*	
Y	.57**	.16	.43**	-.23*	-.16	.06	.04	.41**	.28**	-.28**	.17	-.27**	.22*	.73**

DTH:Days to heading, CL:Culm length, LSL:Long sterile lemma, SSL:Short sterile lemma, NTPP:No. of tillers per plant, NPP:No. of panicles per plant, NSPP:No. of spikelets per panicle, TGW:Thousand grain weight, GL:Grain length, GW:Grain width, LWR:Length-width ratio, FG%:Filled grain %, HI:Harvest index, Y:Yield (5 hills) kg

**p<.05; *p<.01

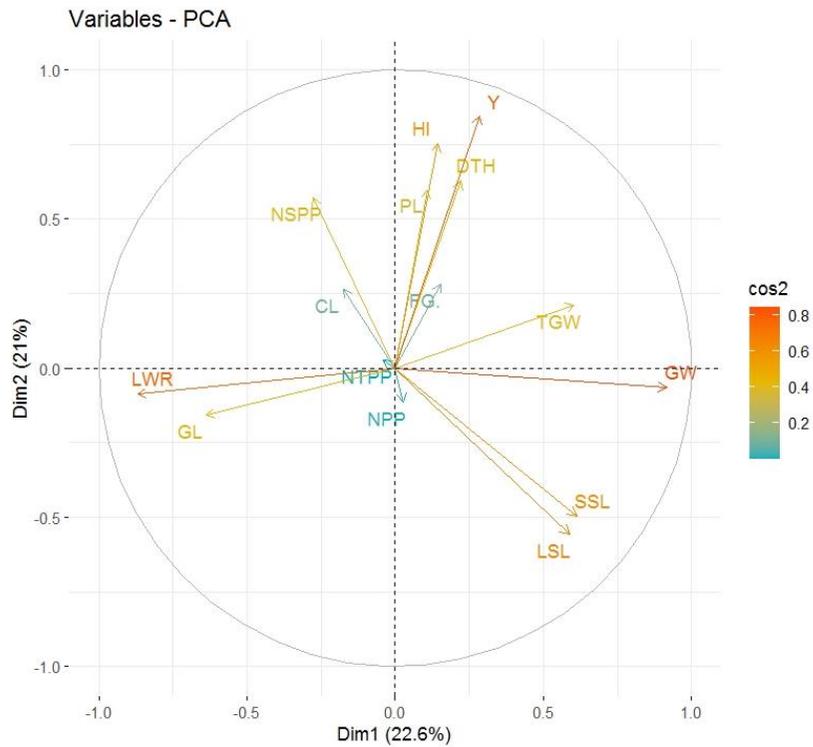


Figure 10. Correlation between variables (quantitative traits) and dimensions (Dim)/principal components (PCs); The \cos^2 values are used to estimate the quality of representation. High \cos^2 indicates a good representation on PC. Variables that are closed to the center of the plot are less important for the first component.

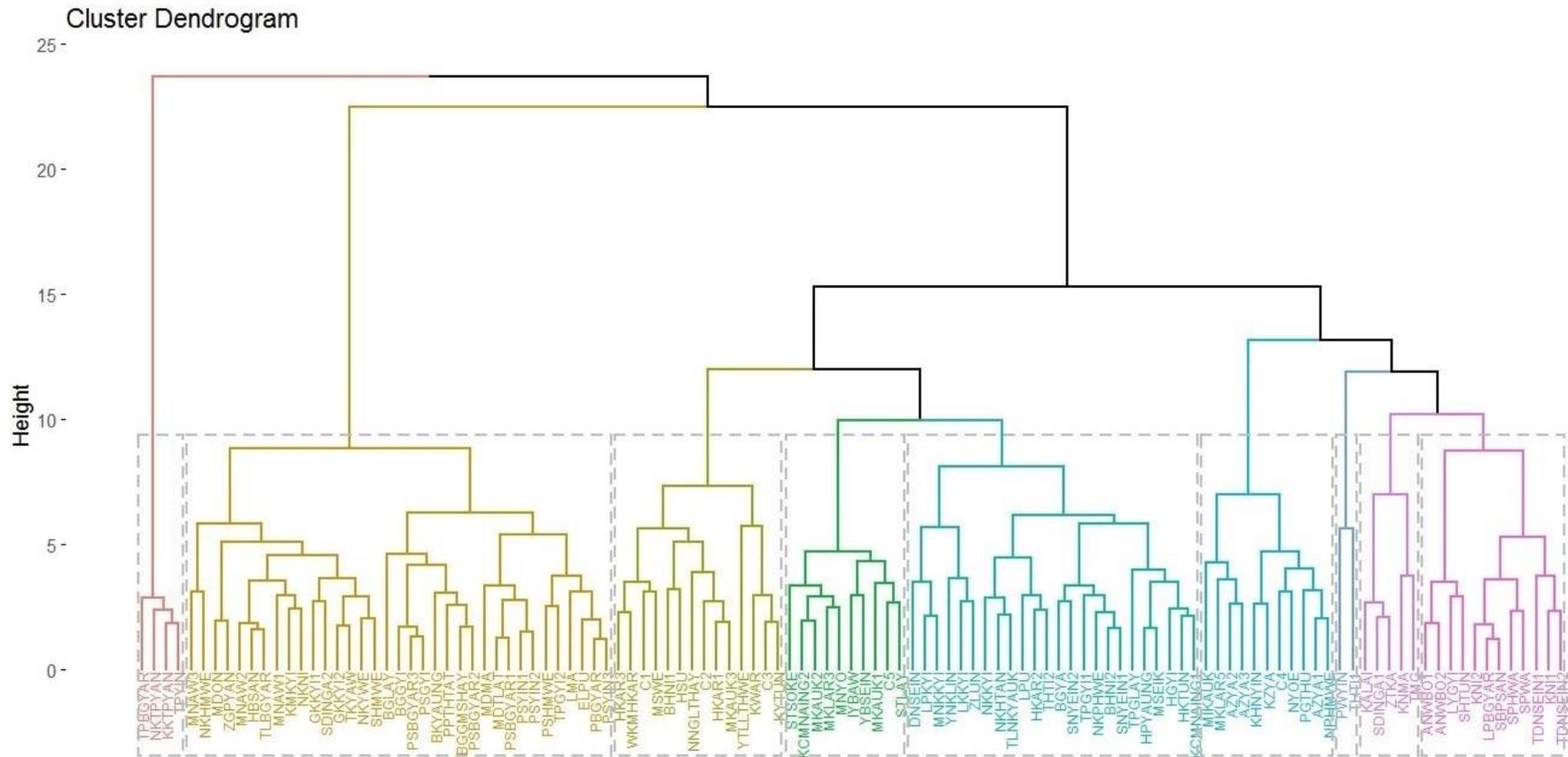


Figure 11. Cluster analysis showing the diversity and relatedness among the 117 Myanmar rice genotypes based on 15 quantitative traits; Sub-clusters are colored differently.

5.2.2 Diversity in qualitative traits

Thirteen of the qualitative traits scored were dominated by one character in each trait with a distribution ranging between 34%–86%. The Shannon diversity index (H') ranged from 0.41 (least polymorphic) for sterile lemma color (SLC) to 0.84 (highly polymorphic) for amylose content (AC2). Trait SLC 86% genotypes showed straw color, whereas 12% and 2% genotypes showed gold and purple color, respectively (Table 11 and Appendix Table A10).

Moderately diverse traits were observed for 6 traits within the range of 0.48–0.73. Moderate diversity observed for awn distribution (AD) (0.48), endosperm type (ET) (0.66), stigma color (SC) (0.67), flag leaf altitude (late observation) FLA_L (0.68), panicle altitude of main axis (PA_M) (0.72) and auricle color (AC1) (0.73). Regarding the AD, 79% of the genotype showed lack awn and 6% genotype consist of few randomly, 3% whole length, and 2% upper three-quarters only. Cloudy and translucent type of endosperm was the predominant state (68% genotype), whereas 30% genotype showed intermediate and 3% genotype had waxy white type. SC showing the highest variability where 58% of the genotype showed light green color and only 3% genotype showed purple color which was the lowest variability. The characters showing higher variability were FLA_L (50% semi-erect, 42% horizontal, 6% descending, 2% erect), PA_M (50% strong drooping, 48% slightly drooping, 3% semi upright). Light green (57%), purple (39%), and green (3%) were observed for AC1 (Table 11 and Appendix Table A10).

Six of the 13 traits (panicle altitude of branches (PA_B), color of apiculus (CoA), lemma and palea pubescence (LPP), lemma and palea color (LPC), flag leaf altitude (early observation) (FLA_E), amylose content (AC2) scored had a high diversity with an average index of 0.8. Regarding trait PA_B, 54% genotype showed spreading, 25% showed semi erect, 20% and 2% genotype shows horizontal and erect respectively. In case of CoA, absent (50%) of genotype and present (22% brown, 9% purple, 8% straw, 7% red, 4% black) were observed. The characters showing higher variability were LPP (34% short hairs, 33% hairs on upper portion, 23% long upper hairs, 4% glabrous, 3% long hairs, 2% short upper hairs), LPC (45% straw furrows, 27% straw, 16% brown furrows, 9% gold and gold furrows, 2% black furrows), and FLA_E (62% semi erect, 24% erect, 14% horizontal). Very high (>25) amylose content was the predominant state (40%), followed by intermediate (~20) (32%), high (~23) (15%), low (~17) (7%), and very low (~9) (5%). High value of Shannon diversity index is the resultant of the presence of different alleles for the characters. Overall, the diversity in qualitative traits was moderate with an average H' index of 0.7 (Table 11 and Appendix Table A10). Nearly all the traits measured showed moderate to high diversity.

The cluster analysis on the basis of the qualitative traits classified the genotypes into two major clusters (Figure 12). The two major clusters were further subdivided into nine sub-clusters (3 in cluster-I and 6 in cluster-II). Cluster-I consisted of 32 genotypes, which were mostly Pawsan rice varieties, whereas cluster-II comprised of 85 genotypes including traditional varieties (other than Pawsan) and HYVs (C1 to C5). C1, C3, and C4 clustered together in sub-cluster-II. 4, whereas C2 and C5 formed together in sub-cluster-II. 3. The genotype pairs (TLNKYAUK and MSEIK, MSWE and KYTUN) in sub-cluster-II clustered

together in the genetic distances 0.00 indicating they have similar morphological characters. The principal component analysis showed similar pattern of groupings as in the cluster analysis (Figure 13).

Table 11. Qualitative traits showing the predominant state observed, distribution (%) and the calculated Shannon diversity indices (H') for each trait scored

Traits	Predominant state	%	Evaluation phase	H' index
AC2	Very high (>25)	40.17	After harvest	0.84
FLA_E	Semi erect	62.39	Anthesis	0.83
LPC	Straw furrows	45.30	Ripening	0.81
CoA	None	50.43	After anthesis to hard dough stage	0.78
LPP	Short hairs	34.19	Ripening	0.78
PA_B	Spreading (open panicle)	53.85	Near maturity	0.77
AC1	Light green	57.26	Late vegetative	0.73
PA_M	Strongly drooping	49.57	Near maturity	0.72
FLA_L	Semi erect	50.43	Maturity	0.68
SC	Light green	58.12	Anthesis (between 09:00 and 14:00)	0.67
ET	Cloudy and translucent	67.52	After polishing	0.66
AD	Awnless	78.63	Flowering to maturity	0.48
SLC	Straw	86.32	7 days after flowering	0.41
Average diversity				0.70

AC2, amylose content; FLA_E, flag leaf altitude (early observation); LPC, lemma and palea color; CoA, color of apiculus; LPP, lemma and palea pubescence; PA_B, panicle altitude of branches; AC1, auricle color; PA_M, panicle altitude of main axis; FLA_L, flag leaf altitude (late observation); SC, stigma color; ET, endosperm type; AD, awns distribution; SLC, sterile lemma color; H', Shannon diversity indices

Cluster Dendrogram

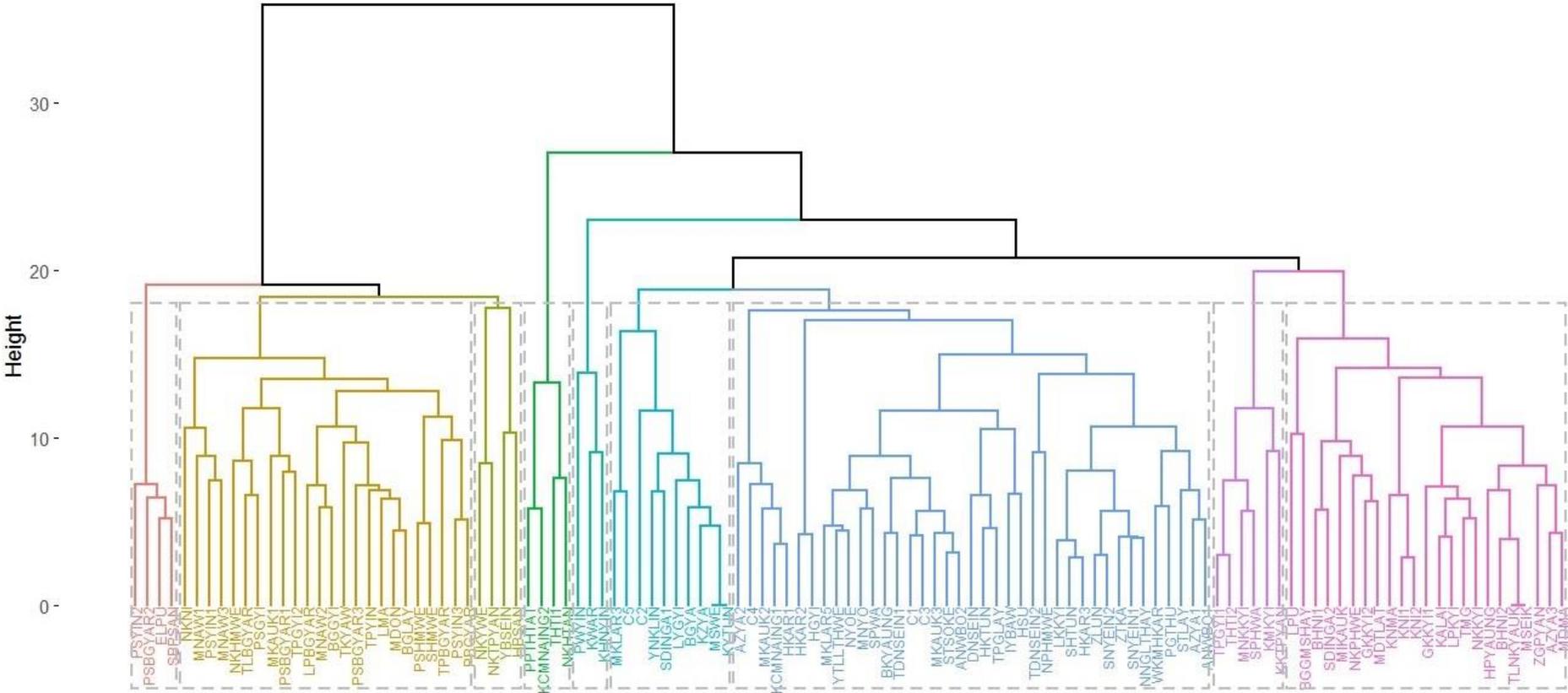


Figure 12. Cluster analysis showing the diversity and relatedness among the 117 Myanmar rice genotypes based on 13 qualitative traits; Sub-clusters are colored differently.

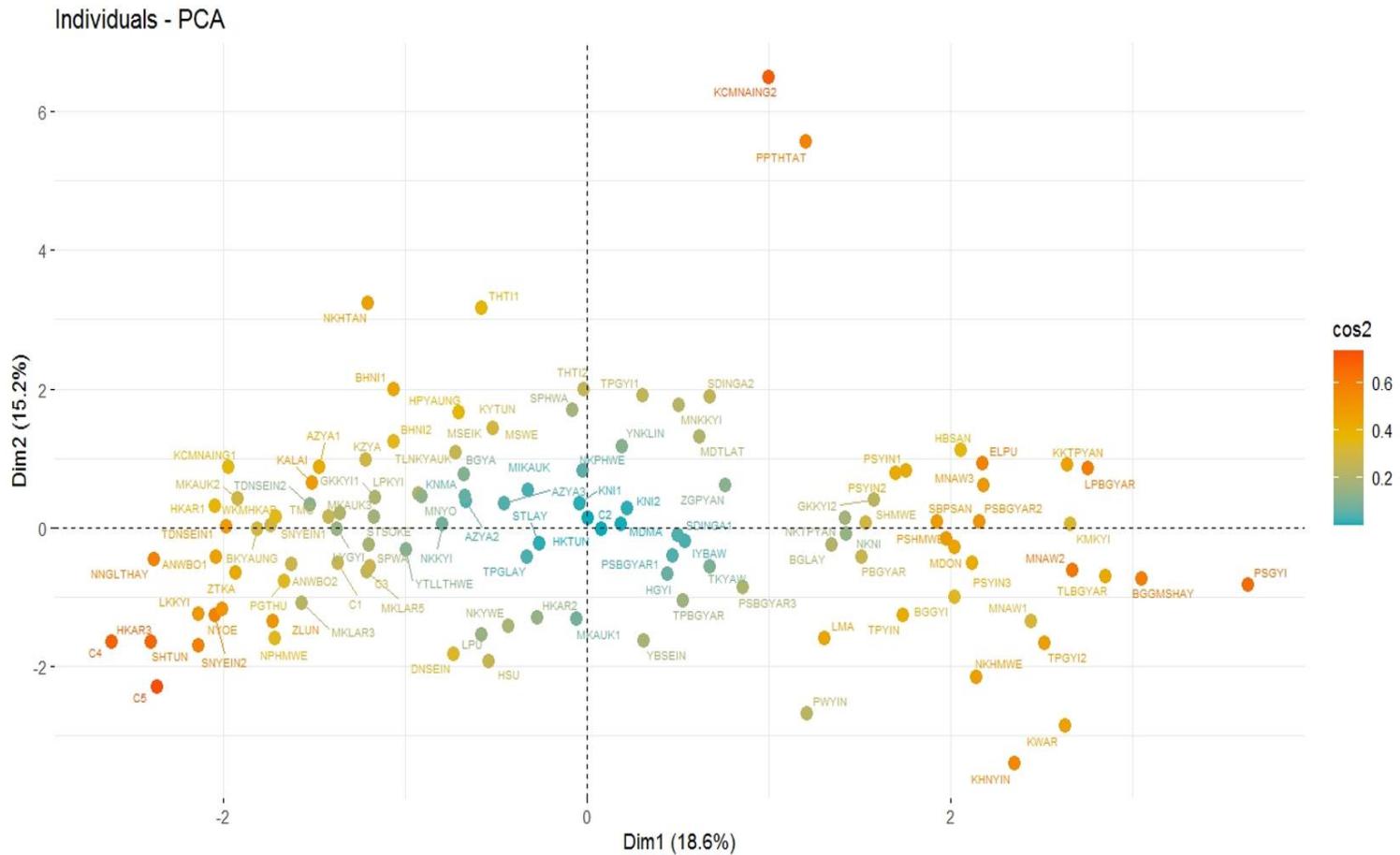


Figure 13. Principal component analysis to explain the diversity across 117 rice genotypes based on 13 qualitative traits; The \cos^2 values are used to estimate the quality of representation. High \cos^2 indicates a good representation on PC.

5.3 Genotypic characterization of rice varieties

5.3.1 Marker quality analysis

Out of 18,271 SNP markers, a total of 7,643 markers cleared and passed all the quality parameters (>95% reproducibility, >95% call rate, and >0.1 one ratio) (Appendix Figure A4). Among the 7,643 informative SNPs, 43% were observed in the PIC class >0.45 to 0.50 and 41% in the >0.30 to ≤0.45 class (Appendix Figure A5). The median (0.44) was located close to the average PIC value of 0.41 (Appendix Table A11). A total of 16,160 silicoDArT markers were generated and they had an average of 99% reproducibility and 93% call rate and 87% of all the identified markers had a >0.1 average one ratio (Appendix Table A5 and Appendix Figure A4). Considering all of the quality parameters, 4,064 silicoDArT markers were used for subsequent analysis. These markers were determined to be moderately informative, with an average PIC value of 0.37 and 0.41 median (Appendix Table A11). Approximately 28% of the markers had a PIC value of ≤0.30 and 23% were in the high PIC value range (>0.45 to 0.50) (Appendix Figure A5).

5.3.2 Genetic relationships among genotypes

The genetic dissimilarities among the genotypes estimated through the SNP markers ranged from 0 to 0.753 (Appendix Table A12). The Pawsan group of varieties revealed the least amount of genetic dissimilarity, ranging from 0 to 0.115, whereas the HYVs ranged from 0.037 to 0.217. Among the traditional varieties other than Pawsan, the dissimilarity indices ranged from 0 to 0.753. The weighted neighbor-joining phylogenetic tree obtained with SNP markers produced two major clusters (Figure 14). Cluster I consisted of 17 high-quality aromatic Pawsan rice, while the remaining 23 genotypes were traditional genotypes, which fell within the genetic dissimilarity range of 0 to 0.13 (Appendix Table A12), revealing that those varieties were closely related to the Pawsan group. Cluster II had a combination of traditional genotypes and HYVs. Among the traditional genotypes, NYOE, MKLAR3, MKLAR5, ZLUN, KZYA, AZYA3, KYTUN, and LPKYI displayed closer genetic similarity (dissimilarity indices of 0.11 to 0.233) to HYVs (Figure 14 and Appendix Table A12).

SilicoDArT markers were also useful for the identification of genetic relationships among rice genotypes. The range of genetic dissimilarities identified through silicoDArT markers was broader than that observed through SNP markers. Among the 117 rice genotypes, dissimilarity ranged from 0.001 to 0.954 (Appendix Table A13). The genetic dissimilarity index among Pawsan varieties fell within the range of 0 to 0.223, suggesting that they were closely related to each other. The HYVs ranged from 0.169 to 0.582, whereas the rest of the traditional varieties revealed a wide dissimilarity range, from 0 to 0.952. Similar to SNP markers, silicoDArT markers also formed two clusters of rice genotypes based on their relatedness (Figure 15). The proportion of membership of individual genotypes in each cluster showed consistency in grouping with the results of SNP markers.

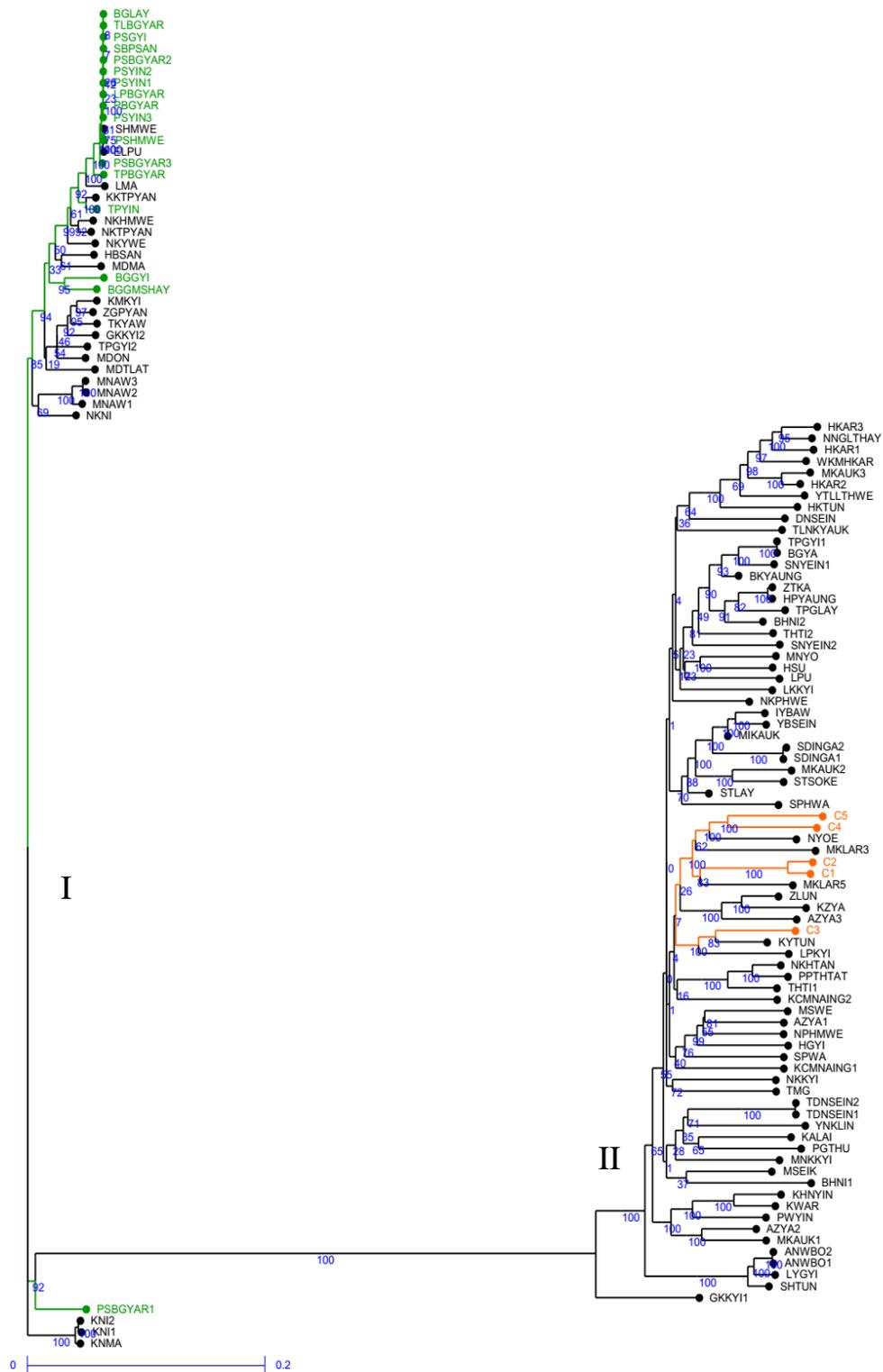


Figure 14. The weighted neighbor-joining phylogenetic tree based on 7,643 SNP markers representing the grouping of 117 rice genotypes; Traditional varieties are shown with black color; Pawsan varieties with green; HYVs with orange.

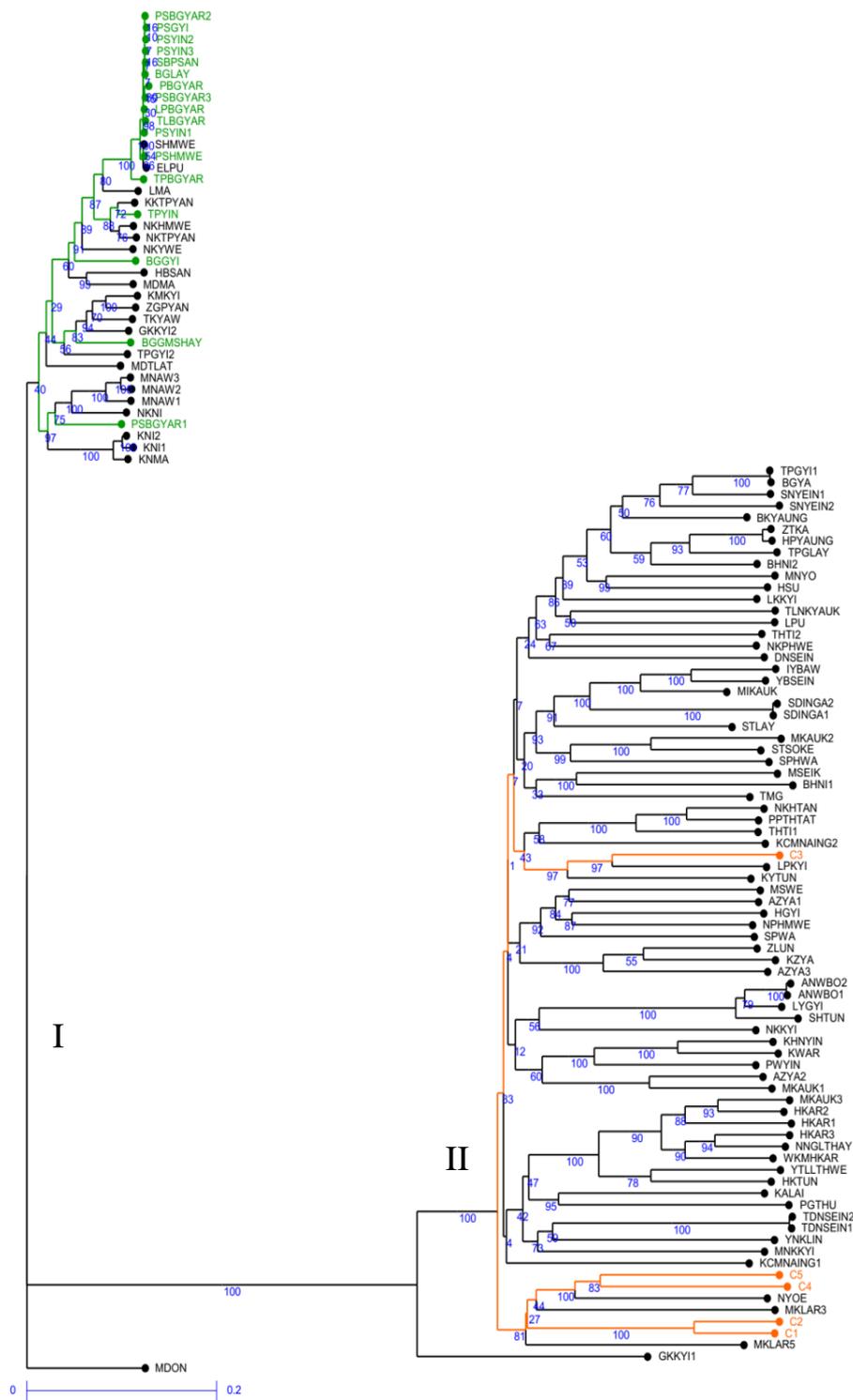


Figure 15. The weighted neighbor-joining phylogenetic tree based on 4,064 silicoDArT markers representing the grouping of 117 rice genotypes; Traditional varieties are shown with black color; Pawsan varieties with green; HYVs with orange.

5.3.3 Population structure

A total of 7,643 SNP markers were used for population structure analysis. The model-based Bayesian cluster analysis in STRUCTURE visualized the genetic structure of the population under examination. K value was used to estimate the number of clusters of the genotypes based on the genotypic data throughout the whole genome. In order to find the optimal K value, the number of clusters (K) was plotted against ΔK , which showed a sharp peak at K = 2 (Figure 16), and the membership of individual genotypes in each population is listed in Table S5. The optimal K value indicates that two populations showed the highest probability for population clustering and these two populations consisted of 40 (Pawsan plus non-Pawsan) and 77 (non-Pawsan plus HYVs), respectively (Table 12 and Appendix Table A14).

A significant divergence was found among individuals within populations according to the Fst values of 0.832 for pop1 and 0.687 for pop2 obtained from STRUCTURE (Table 12). Principal component analysis (PCA) illustrated the genetic divergence among the genotypes (Figure 17). In SNP and silicoDArT markers, the first two axes of the PCA explained 93.5% and 91.5% of the total genetic divergence, respectively. The population distribution determined by both markers is consistent with the output of population structure analysis (Figure 17).

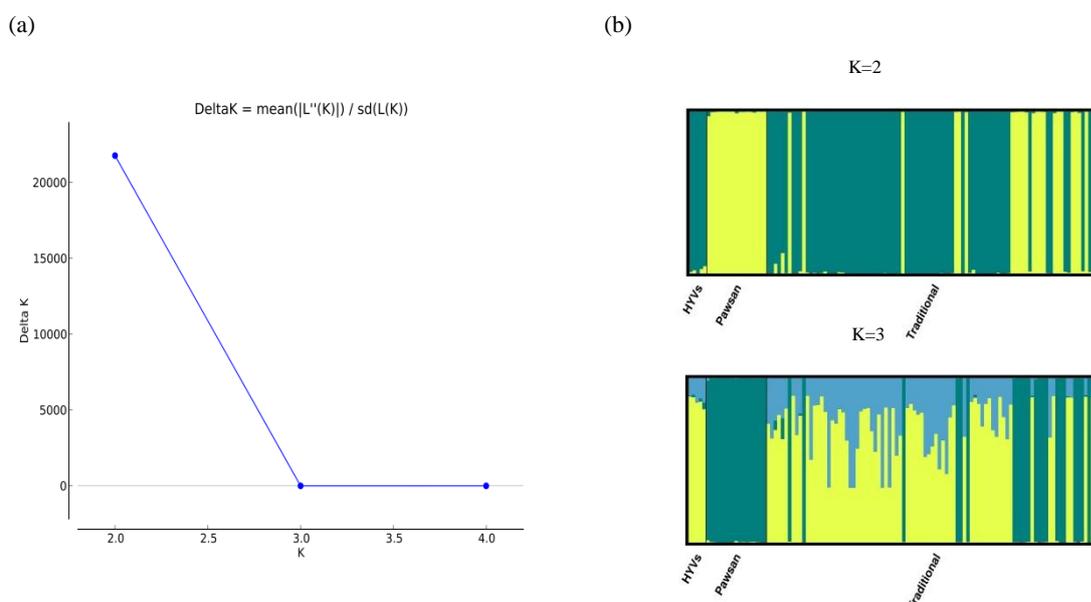


Figure 16. Population structure of 117 rice genotypes based on 7,643 SNPs: (a) ΔK values plotted as the number of populations, (b) populations (K = 2 and K = 3) inferred using STRUCTURE. The number of individuals at K = 3 was also observed as the result of farmers' survey although there is no peak at K = 3. Traditional varieties, Pawsan varieties, and HYVs are colored differently.

Table 12. STRUCTURE results of 117 rice genotypes assigned to each population

Population	Inferred Clusters	Mean Fst ¹	Exp. Het. ²	No. of Genotypes
pop1 ³	0.449	0.832	0.128	40
pop2 ⁴	0.551	0.687	0.169	77

¹ Fst, fixation index; ² Exp. het., expected heterozygosity; ³ pop1, Pawsan plus non-Pawsan traditional varieties; ⁴ pop2, non-Pawsan plus HYVs.

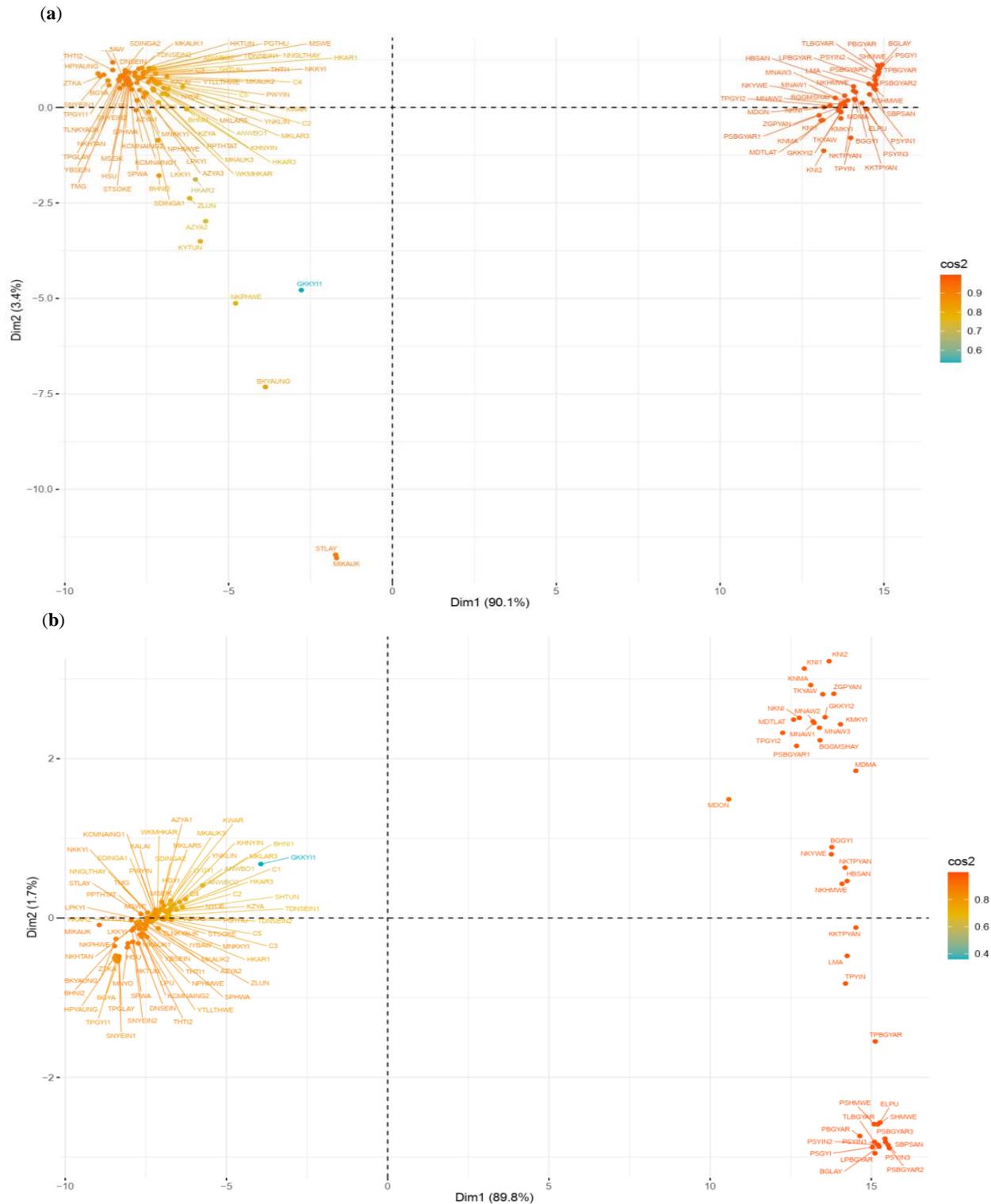


Figure 17. Principal component analysis (PCA) to explain the genetic diversity across 117 rice genotypes: (a) PCA based on 7,643 SNP markers and (b) PCA based on 4,064 silicoDArT markers; The \cos^2 values are used to estimate the quality of representation. High \cos^2 indicates a good representation on PC.

5.3.4 Genetic differentiation of populations

Results from the analysis of molecular variance (AMOVA) revealed that 4% of the total variance was found within individuals, whereas maximum diversity was partitioned between the two populations (74%) and among individuals within populations (22%) (Table 13). In addition, a high F_{st} (0.737) from the AMOVA results was found between populations, indicating a high genetic differentiation between these two populations, and a low N_m value (0.089) was obtained according to Nei's genetic distance analysis (Table 13).

Table 13. Analysis of molecular variance (AMOVA) using 7,643 SNPs of the genetic variation among and within two populations of 117 rice genotypes

Source	df	SS	MS	Est. Var.	%
Among Populations	1	209102.877	209102.877	1973.534	74
Among Individuals	115	148509.820	1291.390	585.915	22
Within Individuals	117	13988.500	119.560	119.560	4
Total	233	371601.197		2679.009	100
F_{st} ¹		0.737 (P = 0.001)			
N_m ²		0.089			

¹ F_{st} , Fixation index; ² N_m , haploid no. of migrants

5.3.5 Allelic pattern across populations

The average value for the number of different alleles (N_a) and effective alleles (N_e) across the populations was 1.604 and 1.252, respectively (Table 14), and the mean value for the overall population in Shannon's index (I), expected heterozygosity (H_e), and unbiased expected heterozygosity (uH_e) was 0.232, 0.150, and 0.152, respectively. Of the two populations, pop2 was more diverse than pop1 and the percentage of polymorphic loci per population (PPL) ranged from 41.07% (pop1) to 79.73% (pop2), with an average of 60.40%.

Table 14. Genetic diversity indices for the two population structures of 117 rice genotypes based on 7,643 SNPs

Pop	N_a ¹	N_e ²	I ³	H_o ⁴	H_e ⁵	uH_e ⁶	F ⁷	PPL (%) ⁸
pop1	1.411	1.146	0.137	0.025	0.087	0.088	0.515	41.07
pop2	1.797	1.358	0.327	0.035	0.214	0.215	0.776	79.73
Mean	1.604	1.252	0.232	0.030	0.150	0.152	0.688	60.40

¹ N_a , number of different alleles; ² N_e , number of effective alleles; ³ I , Shannon's index; ⁴ H_o , observed heterozygosity; ⁵ H_e , expected heterozygosity; ⁶ uH_e , unbiased expected heterozygosity; ⁷ F , fixation index; ⁸ PPL, percentage of polymorphic loci.

6. DISCUSSION

6.1 On-farm rice diversity and farmers' preferences for varietal attributes in Ayeyarwady Delta, Myanmar

6.1.1 Rice varietal diversity

Eighty-three percent of the interviewed farmers cultivated more than one variety during the monsoon season. Growing several varieties per season could be a strategy to address diverse weather conditions (Subedi et al. 2017). In agreement with the hypothesis 1, the on-farm diversity was relatively high as 39 different rice varieties were cultivated: nine of them were HYVs, 10 were from the Pawsan group and 20 were traditional varieties (other than Pawsan). Only 17 rice varieties out of 39 were included for further studies i.e., phenotypic and genotypic characterization because of lack of seed availability for the rest varieties when the survey was conducted.

The number of farmers simultaneously growing more rice varieties was positively affected by farmers' experience and education, which suggests that farmers' decision to increase rice varietal diversity was a factor of cumulative outcome of farmers' choices during the past years. Similarly, farmers located in areas less suitable for rice cultivation, such as saline-water agro-ecological region or undulated type of terrain, were more likely to grow more varieties (especially traditional ones) compared to farmers in the brackish or freshwater regions and flatlands. Similar trend was observed for the farmers who reported their soil as non-fertile, as they tended to adopt more rice varieties on their farms. These findings indicated that growing more than one variety was farmers' strategy to secure rice production and decrease the risk of crop failure in areas less suitable for rice cultivation and in areas more prone to risks related to harsh environmental conditions (i.e., saline-water intrusion, soil erosion in hilly areas). Furthermore, as the terrain and soil fertility of such areas are likely more diverse compared with flatlands, farmers adapt by dividing their fields into smaller portions and by sowing different varieties in particular areas on their farms.

Farmer's reliance on local traditional varieties because of their adaptability to local conditions and to environmental stresses is a well-accepted fact. In agreement with previous results (Cleveland et al. 1994), these traits in our study were among the most valued rice characteristics reported by the farmers in the survey area and partly explained (34%) the use of traditional varieties in the regression model. In particular, farmers, who considered resistance to drought, tolerance to salinity, and several rice grain traits to be important, were the ones that preferentially used traditional varieties (other than Pawsan group); traditional varieties generally show little improvement when high amounts of fertilizers are applied (Yi et al. 2005). Saito et al. (2007) reported that when comparisons were made between traditional and modern improved varieties, there were significant differences in response only to N, whereas there was no significant difference between the NP treatments and the control. This suggests that cultivar differences in response to N fertilizer depend on P availability. The lack of response of the traditional varieties to only N coupled with their response to NP

suggest that either P only or both N and P are responsible for the yield increase of the traditional cultivars when NP fertilizers are applied.

Local varieties, particularly the aromatic Pawsans, were considered to be of high importance in the Ayeyarwady deltaic region (occupying 61% of total cultivated land) and were valued mainly for their high market price and demand, grain texture, aroma and flavor and relatively good yield (2.62 t/ha). Many different sub-selections from the original landrace “Pawsan Hmwe,” a strongly photosensitive variety, which cannot flower beyond a critical photoperiod, were identified in the delta. Pawsan Bay Kyar and Pawsan Yin, which are only slightly photosensitive, were farmers’ selections from Pawsan Hmwe, as they could flower under any length of photoperiod (Subedi et al. 2017). Manaw, Hnan Kar, and Madama were photoperiod sensitive varieties, with higher yield (3 t/ha) compared to the Pawsan group (Appendix Table A8). Farmers, whose fields were located in the lowland areas, usually grew these varieties because of their tolerance to submergence. Moreover, the landrace Hnan Kar was resistant to nematode (*Ditylenchus angustus*) attacks, rice blast disease (*Magnaporthe oryzae*) and rice stem borer (*Scirpophaga incertulas*). Nevertheless, farmers, who reported pest and disease attacks, did not seem to select these varieties specifically, either because they did not have access to the seeds or because of the lack of knowledge of the advantages of particular rice varieties. Similarly, no conclusions could be drawn from the farmers’ preferences for rice characteristics because of the adoption of Pawsan group varieties by almost all farmers in the study area; therefore, their adoption could not be described by the selected logit model.

6.1.2 Socio-economic and agronomic factors to determine the adoption of HYVs

Farmers predominantly preferred traditional varieties to HYVs in the study area, with only 34% of respondents growing HYVs. Despite the higher yields, farmers in the survey area adopted fewer HYVs compared to Pawsan and traditional variety groups, which indicated that traits other than yield were important in their decision-making. While Khanal et al. (2017) reported that rice yield was the most important factor influencing farmers’ varietal selection, the farmers in the Ayeyarwady region were generally aware that HYVs were less resistant to stresses, unless specially bred varieties were included, which, however, was not the case for the HYVs used by the farmers in the survey area. This clearly indicates that while high yields are desired by the farmers, growing varieties capable of coping with harsh local conditions remains essential and highlights the usefulness of such traditional varieties in breeding programs.

According to the regression model, socio-economic variables and agronomic factors (farming experience, education, total sown area, soil fertility, and water region) had a significant impact on the likeliness of cultivating HYVs. While Islam et al. (2012) identified farmers’ age, experience, irrigation coverage, off-farm income, access to microfinance, and membership in village local groups as the key factors driving the HYVs adoption; we found the likeliness of HYVs adoption to be positively influenced by total area sown to rice, but negatively influenced by farmers’ experience and education. The negative impact of farming experience can be explained by a higher awareness of the benefits of the traditional varieties.

Furthermore, while HYVs may offer immediate benefit with respect to increased yields, farmers with longer experience could have faced long-term disadvantages of HYVs in increased risks of incidents, including pest or disease attacks or lower tolerance to stresses resulting in production losses (Wale 2012).

The negative relationship between HYVs adoption likeliness and farmers' education is surprising as education is generally related to a more efficient use of inputs and facilitated perception and interpretation of new technologies and new improved varieties. It is well accepted that HYVs are relatively more labor-intensive and have higher input costs when compared to traditional varieties, which may result in reduced adoption of HYVs if a large proportion of family members lives off-farm. Higher education and longer experience also likely influence farmers' capacity to determine the most suitable rice variety based on available resources and may rely more on stable, low-input traditional varieties in the case of the limited access to inputs, such as labor, irrigation, fertilizers, or pesticides, which are often the key determinants of HYVs adoption (Samal et al. 2011). Moreover, since HYVs adoption likeliness decreased with education but increased with total area sown to rice, it could be speculated that more educated farmers had other incomes besides rice cultivation and diversified their portfolio of cash crops.

In addition, new unknown varieties could be considered risky because of the lack of an assured market. On the other hand, Wale (2012) concluded that technology and market, particularly farmers' access to new HYVs, were among the most important drivers related to the abandonment of traditional varieties. As the majority of the farmers in the study area were using their own seeds and did not generally purchase seed, farmers' lack of access to the seeds of modern HYVs could also be the reason for the relatively low adoption of HYVs.

The farmers' perception of the fertility of their fields seemed to influence the likeliness of the adoption of HYVs, as these varieties were more commonly used by those farmers who described their soil fertility as low or medium. As higher subsistence pressure has been indicated as an important driver of HYVs adoption (Hollaway et al. 2002), farmers with non-fertile soils likely had, in general, very low rice yields. As the yields of traditionally grown varieties are generally lower compared to HYVs, these farmers may be more eager to experiment with varieties that can guarantee higher yields (HYVs) and thus, higher income. Nevertheless, HYVs perform well only under optimal or near-optimal growing conditions (Bardsley & Thomas 2005) and on fertile soil. Thus, subsistence pressure and low yields can hardly explain the adoption of HYVs, as farmers with knowledge of disadvantages of HYVs, under specific growing conditions, likely corresponding to more educated farmers, are apparently less prone to experiment with HYVs.

The distribution of HYVs was uneven. These varieties were almost absent in the saline-water region, since traditional varieties were more resistant to salinity. Thus, developing new rice varieties, which are tolerant to the major abiotic stresses, such as drought, flooding, salinity, and high temperature, will affect the adoption of HYVs by the farmers located in unfavorable growing environments, especially in saline areas. Therefore, the identified

traditional varieties used by local farmers are a valuable source of genetic material required for successful breeding of new improved varieties.

6.1.3 Rice cultivation constraints and the preferred characteristics

From all the 25 rice cultivation constraints identified by the farmers, pest infestation received the highest score. Farmers revealed that the control of pests and diseases incurred high costs while considerably reducing rice productivity and quality. Lack of available labor was identified as a second constraint because young people often migrate to other areas or neighboring countries, where labor wages had risen in the recent years. As a result of these shortages, farmers were unable to plant their paddy crop in a timely manner, ultimately delaying harvest and risking damage to the crops by heavy rains. Consequently, such paddies fail to fetch a satisfactory price on the market. The third constraint, flooding, seriously impacted rice productivity. Large areas of the delta are subject to flooding, ranging in duration from a few days to 2 or 3 months, presenting significant risks to farmers. Some areas are suitable for deep water rice, a low-yielding rice type that elongates to stay above the rising water. The native deep water rice is of low palatability and is therefore grown as animal feed in Myanmar (Nguyen & Pittock 2016). Thus, there is a need to develop flood-tolerant rice varieties that also produce high yields and good quality. Without the benefit of submergence tolerance, excessive flooding severely limits the scope of using improved HYVs and crop management (Denning et al. 2013).

Nowadays, because of abnormally heavy rain at the rice-ripening stage, farmers encounter crop damage from both plant lodging and shattering of rice grains. Hence, farmers continue to grow local rice varieties, as resistances to shattering and lodging are prominent traits, which were highly scored by the farmers. Tillering ability is a yield-determining characteristic of rice plants. The growth and development of tillers depend partially on environmental factors, such as radiation, temperature, and nutritional conditions, and partially on varietal characteristics (Hanada 1993). Generally, varieties with more tillers have a higher number of panicles and their contribution to yield is higher than that of the low-tillering varieties (Nuruzzaman et al. 2012). Thus, it is not surprising that farmers highly rated tillering ability. On the other hand, especially in areas with harsh environmental conditions or with unpredictable climate, varieties with shortened growing period could be preferred, as the risk of harvest loss is reduced. Surprisingly, a short-growing period was among the least important traits mentioned by the farmers. High straw production was scored as the least important trait because straw is commonly used for animal fodder and bedding; however, cattle or buffalo were not often employed in farming activities.

Because of high production losses attributable to pests and diseases, farmers tend to grow local varieties, which can be tolerant to some extent, while modifying agronomic practices (spacing, adjusting planting time, water management) to reduce pest and disease attacks. Farmers wanted new resistant varieties and new methods of control, including biological control methods. Besides biotic stresses (diseases/insects), the rice crop frequently faces abiotic stresses, such as submergence, salinity, drought, and cold stresses. Excess water is the main constraint to rice productivity in large areas of rainfed lowland ecosystems. This

regularly affects some 15–20 million ha of rice land in Asia. Farmers in flood-prone areas highly value rice that can withstand submergence for prolonged periods, such as Swarna-Sub1 (Arora et al. 2019). Dar et al. (2013) also confirmed higher yields when submergence-tolerant rice variety Swarna-Sub1 was grown on fields submerged for as long as 7–14 days. Farmers indicated that salt-tolerant rice varieties were required, as salt-water intrusion had become a problem during the last several years. Clearly, the rising sea levels and problems linked with reduced availability of freshwater will lead to even higher demand for varieties that can cope with such environmental stresses without jeopardizing rice yields.

The International Rice Research Institute (IRRI), through the Consortium for Unfavorable Rice Environments (CURE), has continued to work with Myanmar's Department of Agricultural Research (DAR) to develop suitable rice varieties for challenging areas. This cooperation brings the prospect of additional promising varieties in the future. According to a former Deputy Director General of Department of Agricultural Research (Myint 2017), a number of stress-tolerant rice varieties have been developed in Myanmar. Some of these varieties include Yemyokekhan 1 (Swarna-Sub1), Yemyokekhan 2 (BR11-Sub 1) for submergence tolerance, and Sangankhan Sinthwelatt (Salttol Sin Thwe Latt), Pyi Myanmar Sein (IR10T107), Shwe Asean (CSR 36) for salinity tolerance. Farmers can receive information on or seeds of these improved varieties from regional agricultural offices and seed farms. However, only a limited number of varieties are commonly grown by farmers in the Ayeyarwady delta. Farmers are only interested in those rice varieties that have a good market. As it takes at least 4–5 years to develop a new market, farmers tend to stick to their old local varieties. Farmers preferred varieties with quality characteristics of milled rice, such as fewer broken grains and high milling recovery. Both traits are mutually related and depend on variety type, environmental factors, and postharvest handling, especially moisture content of rice grains at harvest (Fan et al. 2000).

6.2 Phenotypic characterization of rice varieties

6.2.1 Diversity in quantitative traits

Among the quantitative traits evaluated in this study, the high coefficient of variation was observed for a majority of the agronomic traits (yield, number of tillers per plant, number of panicles per plant, number of spikelets per panicle, and 1000-grain weight). As we expected as in hypothesis 2, their variations found quite high for 13 quantitative traits. The presence of high variability suggests that these characters might be of interest for selection and can be improved either through selection or hybridization. Aung (2007) also reported that there was remarkable diversity in traits such as grain size and shape, 1000-grain weight, sterile lemma length, and culm length. Moreover, PCA analysis indicated that grain width, sterile lemma length, yield, days to heading, harvest index, panicle length, 1000-grain weight, and numbers of spikelets per panicle were the major factors contributing to the variation in rice genotypes.

Myanmar traditional rice genotypes are usually sensitive to photoperiod so that flowering (DTH) begins in November regardless of sowing time. In this study, DTH of rice genotypes ranged from 66–114 days showing significant variation among genotypes (Appendix Figure

A6). Traditional genotypes such as PWYIN, SHTUN, ANWBO2, ANWBO1, LYGVI, MSWE were found as the early growth duration genotypes (average DTH = 96 days). Among them, ANWBO1, ANWBO2, and LYGVI are typically late matured and photoperiod sensitive. However, they were unexpectedly found as the early matured genotypes in this study, probably due to late sowing (~60 days later than usual sowing time), which means late sowing might have the vegetative stage shortened and early flowering begun at 2nd week of November. DTH for Pawsan group was in the range of 84–109 days with a mean of 101 days, whereas DTH for HYVs was a mean of 97 days, which is a standard DTH for them and in addition, they are not sensitive to photoperiod. For this reason, they have gotten popular in irrigated areas throughout Myanmar.

Grain shape and size has long been used as a convenient criterion among Myanmar farmers (Irie et al. 2004) to recognize different varieties. Size, shape and weight of rice grains of Myanmar landraces were diverse countrywide as well as locally. Myanmar rice genotypes in this study greatly varied in grain length, grain width, and length/width ratio. Among the 117 genotypes, bold grain type (Meedon) was observed as the predominant grain type (all Pawsan and 21 non-Pawsan) followed by Ngasein (32 non-Pawsan), Letywezin (2 HYVs and 23 non-Pawsan), and Emata (3 HYVs and 19 non-Pawsan (Appendix Figure A7).

Trait correlations can be used by breeders either to simultaneously improve correlated traits or reduce undesirable side effects when trying to improve only one of the correlated traits (Chen & Lubberstedt 2010). A significant correlation of grain yield with days to heading, panicle length, number of spikelets per panicle, 1000-grain weight, filled grain %, and harvest index showed that these integrated traits can be used to improve grain yield in rice. According to on-farm survey result, farmers preferred varieties with high tillering ability, shorter growing duration, and high yield. The genotypes such as MNAW3, KNMA, TMG, YTLLTHWE, ZTKA, KALAI, and PSGVI produced more tillers and BGLAY, PSHMWE, LPKYI, MNAW1, BGGMSHAY, and PSBGYAR3 produced more grain yield than other genotypes (Appendix Table A9). In this study, there was no correlation between number of tillers per plant and yield, however, the correlation between the panicle number per plant and the tiller number ($r = 0.51$) was significant correlation suggesting that all tillers were productive tillers and able to bear inflorescence.

The cluster analysis for quantitative traits showed three major clusters; the genotypes with same name grouped together. The genotypes found in cluster-I own the significantly long length of sterile lemma (Appendix Figure A8). Traditional varieties were scattered in both cluster-II (with Pawsan) and cluster-III (with Pawsan and HYVs) based on quantitative traits. HYVs formed separately from Pawsan group except 2 Pawsans (LPBGYAR and SBPSAN) suggesting that HYVs shared similar quantitative traits of those 2 Pawsans more than the rest Pawsans. However, clustering result using DArT seq-based SNPs and silicoDArT markers showed all 17 Pawsans and HYVs found in separate group (Figure 14 and Figure 15). Nevertheless, to validate this clustering result, we would need to implement more research for different seasons under different environments since morphological characters especially quantitative traits are more dependent to environmental responses.

Heritability close to zero indicates that almost all of the variability in a trait among genotypes is due to environmental factors with very little influence from genetic differences (Wray & Visscher 2008). In this study, number of tillers per plant and number of panicles per plant showed 0% heritability meaning those traits are not under genetic control. The heritability values were high ($H^2 >70\%$) for most of the traits except for filled grain % and harvest index. The highest heritability 0.98 for 1000-grain weight and length/width ratio means that 98% of the variability in the traits is due to genetic differences among genotypes. These findings agree with Descalsota et al. (2018) who reported that the heritability values were high for grain length, grain width, 1000-grain weight, days to heading, and culm length in the MAGIC Plus lines developed at IRRI. Fahlani et al. (2010) discussed that the better prediction of the environmental component of phenotypic variance helps the more reliable estimation of both broad and narrow sense heritability, and so the better planning program for cross and self-pollinated plants improvement.

6.2.2 Diversity in qualitative traits

Qualitative traits are considered as the most important characters to identify a particular plant variety and they are less independent to the environmental response since they are mostly genetically controlled (Sinha & Mishra 2013). Analyzing 13 qualitative traits in our study, the least polymorphism was observed in the trait sterile lemma color while the rest qualitative traits showed moderate to high polymorphism.

Amylose content (AC) was the trait that showed the highest H' index and among 117 genotypes, 40% genotypes were very high AC type (>25) and both traditional non-Pawsans and HYVs were included in it. High amylose type show high volume expansion and a high degree of flakiness after cooling. Intermediate type is moist and tender, and do not become hard after cooling. Pawsan varieties were founded in intermediate AC class. In major rice productive lowland area, intermediate AC type (~ 20) was predominant due to market and customer preference (Wunna 2015). Amylose is almost absent from the waxy (glutinous) rice, and low amylose cooked rice is moist and sticky (Wunna 2015). A few samples were observed as very low amylose content (~ 9) such as PWYIN, IYBAW, KHNYIN, etc. AC can be determined by difference in the endosperm types, and amylose and amylopectin ratio (Farias & DeLaCruz 1995). Endosperm type of Myanmar rice genotypes varied from non-glutinous (translucent endosperm) to glutinous (opaque endosperm). Most of the genotypes in this study were non-glutinous, which is in line with the results of Aung (2007).

The trait flag leaf attitude was taken on two stages early and late. It was observed that semi erect type flag leaf attitude was the predominant type among genotypes at both stages. Twenty-eight genotypes found erect flag leaf attitude at early stage but only 2 genotypes (C5 and MKLAR3) found erect at late observation (Appendix Table A15). Sinclair and Sheehy (1999) reported that leaf angle has been a target in breeding programs because erect flag leaves can capture more sunlight. Further, flag leaves are important in grain filling, as 80% of the total carbohydrate stored in the grains is produced by the top two leaves in rice (Gladun & Karpov 1993). Traits in relation with panicle in which first trait is panicle attitude of main axis found drooping type in 114 genotypes, and only 3 had semi upright. The attitude of

panicle branches found spreading (open panicle) in 63 genotypes. Bonow et al. (2007) stated that panicle type is generally affected by water deficiency before flowering because this condition changes the angle of the panicle branches and the density of the grains.

In case of color pigmentation traits, they all showed the predominant color in their respective states, for instance, straw furrows for lemma and palea color, colorless for apiculus, light green for auricle and stigma, and straw for sterile lemma. However, some genotypes clearly differed from one another, for their pigmentation showing unique color in each trait such as the genotypes PPTHTAT and KCMNAING2 observed purple color of sterile lemma, NKHTAN, THTI1, PPTHTAT and KCMNAING2 observed purple stigma and green auricle. Regarding awn distribution, majority of Myanmar rice were awnless. In our study, 79% genotypes were awnless and the rest 21% were awned varieties with different length. Among the awned varieties, LPU possessed significant long lengthy awn compared with others (Appendix Figure A9). Fonseca et al. (2002) discussed that the soil fertilization and plant density influence the awn length. The presence of awn is not a desirable character and farmers and rice traders do not prefer awned varieties since the awns reduced the seed weight, number of seeds and milling recovery (Aung 2007). The awned genotypes are primitive and well adapted to adverse environment factors viz., drought, salinity and low temperature as reported by Chandraratna (1964).

The clustering based on qualitative traits data revealed two main clusters out of which 32 rice genotypes were grouped together in cluster-I and half of them were Pawsans. The rest of the genotypes formed a very large cluster-II comprising traditional non-Pawsans and HYVs. It can be assumed that although other traditional genotypes might have similar characters in some qualitative traits with HYVs, Pawsan traditional group markedly differed from HYVs based on 13 qualitative traits, particularly, amylose content, endosperm type, awn distribution, color and pubescence of lemma and palea (Appendix Figure A10 and Appendix Figure A11). Clustering analysis using DArTseq markers was in agreement with this result forming Pawsans and HYVs separately (Figure 14 and Figure 15). Two pairs of genotypes (TLNKYAUK and MSEIK, MSWE and KYTUN) found in cluster-IIb in the genetic distances (0.00) due to their identical qualitative traits, which we may conclude that those two genotypes altogether in each pair should not be used for further rice improvement because of no differences between them. This clustering analysis confirmed our hypothesis 2, which is that those genotypes shared same qualitative traits although they have different names. The genetic dissimilarities using DArTseq-based SNP markers identified that they were closely related having less amount of dissimilarity index (0.169 between TLNKYAUK and MSEIK and 0.189 between MSWE and KYTUN).

6.3 Genotypic characterization of rice varieties

6.3.1 Marker quality analysis

Our study highlights the suitability of DArT platforms that can be applied for genomic studies of rice genotypes. A total of 18,271 DArTseq SNPs were developed, of which 7,643

markers provided robust information from the rice genome in the absence of sequence information, while silicoDArT markers provided 4,064 informative markers.

The average PIC values of both types of markers in rice were similar to the values identified in DArT markers in sorghum (0.41) (Mace et al. 2008), cassava (0.42) (Xia et al. 2005), and wheat (0.44) (Akbari et al. 2006). The PIC values are a good indication of informative markers that can be used for genotyping plant populations and studying genetic diversity (Salem & Sallam 2016). According to a previous study, (1) markers with a PIC value of ≥ 0.50 were considered to be highly informative, (2) markers with a PIC value from 0.25 to 0.50 were moderately informative, and (3) markers with a PIC value of less than 0.25 were slightly informative (Botstein et al. 1980). The average PIC values of both SNP and silicoDArT markers in our results suggested that those markers were moderately informative. Marker density has a high correlation with gene density; therefore, the abundance of SNP and silicoDArT markers may achieve better genome coverage through the sampling of a greater number of points in the whole genome (Dierig & Ray 2009; Kilian et al. 2012).

Of the different types of molecular markers, microsatellite markers (SSRs) have been used most extensively in Myanmar rice genotypes (Aung 2007; Oo et al. 2008; Wunna et al. 2016). Mogga et al. (2018) used DArTseq markers to investigate genetic diversity in rice (*Oryza sativa* L.). Their study was performed using 59 rice genotypes with 525 SNPs derived from a DArTseq platform. Phung et al. (2014) also characterized a panel of 182 rice genotypes with 25,971 markers using DArT and SNP markers. Therefore, SNP and silicoDArT markers may be better suited for genetic diversity studies, association/linkage mapping, and sequence-based physical mapping in rice (Alam et al. 2018). Furthermore, this study will be useful for international trade to avoid adulteration of Myanmar Pawsan varieties, which are priced varieties. Markers specific for identifying the sub-sub group of Pawsan could be helpful in identifying true Pawsan. This is similar to Basmati trade, in which international trading checks actual Basmati varieties through Basmati-specific markers.

6.3.2 Population structure and relationships

Population structure analysis is informative in understanding genetic diversity and facilitates subsequent association mapping studies (Eltaher et al. 2018). The presence of structure in this population was expected to have three groups based on the findings of an on-farm rice diversity survey in the study area. However, the population structure results did not support the expectation. In fact, 117 rice genotypes were divided into only two groups with STRUCTURE ($K = 2$) (Table 12). The dendrogram analysis (neighbor-joining tree) and the PCA results were in agreement with STRUCTURE results (Figure 14, Figure 15, Figure 16, and Figure 17).

In total, 23 non-Pawsan traditional genotypes and all 17 Pawsans were clustered together into one genetically related population (pop1), which indicated that those non-Pawsans were genetically close to Pawsans. With regard to genetic dissimilarity indices, they revealed a smaller range (0 to 0.133) for SNP markers (Appendix Table A12) and a larger range (0 to 0.287) for silicoDArT markers (Appendix Table A13). All 40 genotypes clustered in pop1

had bold grain shape, which is called Meedon type in Myanmar. Most of the genotypes in the Meedon group are local rice with good eating quality (Thein et al. 2012). It can be assumed that those Pawsan and non-Pawsan rice genotypes possessed the same genes that controlled rice grain shape. In addition, in the studies of Wunna et al. (2016) and Thein et al. (2012) using SSR markers, four genotypes (KMKYI, NKYWE, NKTPYAN, and ZGPYAN) out of 23 non-Pawsans clustered together with Pawsans. Thein et al. (2014) studied the variation in genetic structure of 38 Pawsan rices and reported that the Pawsan group was separate from two controls (IR36 for indica and Koshihikari for japonica). However, the genetic similarity of the Pawsan group was closer to japonica type (Koshihikari) suggesting that they were tropical japonica or javanica-type landraces. A former deputy director general of the Department of Agricultural Research also confirmed that the Pawsan group belongs to tropical japonica type varieties (Myint 2021). Therefore, it can be concluded that those 23 non-Pawsans might also be tropical japonica types. In addition, morphological characterization on this panel of rice genotypes pointed out that some traits, particularly culm length (91–120 cm), panicle length (~25 cm), and grain type (2–2.4 mm) were observed as common traits among genotypes within pop1.

Further, 72 non-Pawsan traditional genotypes and all 5 HYVs were clustered together in the second genetically related population (pop2), probably reflecting the fact that breeding activities led to genetic similarities since new varieties were usually selected from local landraces. Most of the local landraces have unique taste and shape, which breeders want to keep intact, whereas breeding for traits such as short duration, short stem, low canopy type, etc. However, local varieties have some undesirable traits such as long duration, tall or plant architecture etc. Although these genotypes fell into the same group, they had a broad range of dissimilarity: 0–0.281 and 0.001–0.688 for SNP and silicoDArT markers, respectively (Appendix Table A12 and Appendix Table A13). Moreover, Khush et al. (2003) reported that indica-type landraces predominate in Myanmar (81% of the total landraces). Some genotypes in pop2 such as NPHMWE, KHNYIN, HKAR, C1, and C3 were confirmed as indica type (Wunna 2015). It might be important that those 77 genotypes (non-Pawsan and HYVs) were found in the same population although further studies such as adding more control indica-type genotypes would be helpful to clarify this.

The assignment of individuals to each population at $K = 3$ was also observed as the farmers' survey result (Figure 16b). The membership and number of genotypes (40) comprising pop1 at $K = 3$ were identical to the results at $K = 2$, whereas 77 genotypes in pop2 at $K = 2$ were divided into two groups (Appendix Table A16). This clearly indicates that there is no genetic similarity between Pawsans and HYVs. Pawsan is known as a highly photoperiod-sensitive variety and its grain quality (a prized trait) depends upon photosensitivity and requires ecology (special growing season, etc.). For this reason, rice breeders/researchers failed to develop HYVs from the Pawsan group and consumers also refuse to accept any change from the original Pawsan. Seven traditional genotypes (other than Pawsan) out of 77 genotypes (72 non-Pawsans and 5 HYVs) found separation as a new group (pop3), suggesting that the genetic relatedness of those 7 non-Pawsans with HYVs was less than with other traditional genotypes grouped together with HYVs in pop2 (Appendix Table

A16). Moreover, in the work of phenotypically characterization, some traits (e.g., grain type, culm length, and number of spikelets per panicle) distinguished seven non-Pawsan traditional genotypes from HYVs.

In addition, some duplicate genotypes found in this study, by their name (farmer-named varieties based on farmers' taxonomies and nomenclature), were consistent with the naming and distinguishing through very close genetic relationships (e.g., in pop1: MNAW1, MNAW2, and MNAW3) (Figure 14). On the other hand, different genotypes accounted for by the same name also existed in this germplasm collection since the genotypes under the same name were classified into different groups, for instance, GKKYI1 (pop2) and GKKYI2 (pop1) (Figure 14). Unlikely, GKKYI1 and GKKYI2 found together in the same cluster as their phenotypic variations in both quantitative and qualitative traits (Figure 11 and Figure 12). In this case, the combined use of molecular markers and morphological characters may allow further correct discrimination, which confirmed the hypothesis 3. Therefore, testing the underlying population structure is crucial for rice improvement strategies involving marker-trait association studies, such as genome-wide association scanning to identify a true association between markers and traits and the underlying genes controlling the traits (Eltaher et al. 2018). The information obtained from such testing will build confidence in the outcome of the potential association that may be detected.

6.3.3 Genetic differentiation of populations

Fst (fixation index) is a measure of population differentiation due to genetic structure. A Fst value of 0.25 can be considered as significant in differentiating populations. The range 0.15–0.25 indicates moderate differentiation, whereas differentiation is negligible if the Fst value is 0.05 or less (Wright 1987). A significant divergence was found among individuals within populations according to the Fst values obtained from STRUCTURE (Table 12). The AMOVA results showed that high genetic variation existed between populations, which may be due to low genetic exchange and gene flow (Nm value) (Arora et al. 2014). An Nm value of less than 1 indicates limited gene exchange among populations (Wright 1965). In this study, the Nm value (0.089) was quite low; therefore, the low genetic exchange between the two populations led to their high genetic differentiation (Eltaher et al. 2018). Various cultural and agro-ecological factors influence the mechanisms of gene flow in rice fields (Nuijten & Richards 2013). In this study, the main factor influencing gene flow may be agro-ecological conditions. For instance, farmers whose fields were located near the sea (saline-water region) usually grew local landraces such as LYGVI, ANWBO1, and ANWBO2 because of their tolerance of salinity, whereas there was no possibility to grow the Pawsan group in such areas. This might have influenced the high genetic variation observed.

The allelic pattern and genetic diversity indices provided useful information on the genetic diversity in each population. The higher value for diversity indices is an indication of a higher level of genetic diversity (Eltaher et al. 2018; Luo et al. 2019). Previous studies in Myanmar rice using SSR markers observed a higher level of polymorphism with respect to heterozygosity (Thein et al. 2012; Wunna 2015) because of their multi-allelic nature and their rapid mutation rates (Tsykun et al. 2017). SNPs are mostly bi-allelic; however, a higher

number of loci sufficiently polymorphic can potentially give a similar genetic resolution as randomly chosen and multi-allelic SSRs (Guichoux et al. 2011). In this study, comparing genetic diversity indices revealed that pop2 appeared to be more diverse with higher values for private alleles, Shannon's index, expected heterozygosity, unbiased expected heterozygosity, and parental percentage of polymorphic loci. The level of diversity represents a valuable resource for future rice improvement programs. In the survey results of farmers' preferences for rice varieties, except for high yield, farmers were interested in rice varieties with a good response to stress conditions and suitability in particular agro-ecological regions, specifically those with salinity, submergence, and pests/diseases. Among the genotypes in pop2, some of them have those properties; for example, HKAR is resistant to nematode (*Ditylenchus angustus*) attacks, rice blast disease (*Magnaporthe oryzae*), and rice stem borer (*Scirpophaga incertulas*); MKAUK, HKAR, and MKLAR are well known for their submergence tolerance and elongation ability; and LYGVI and ANWBO are known for their salinity tolerance. Thus, these findings based on whole-genome genotyping could be a pillar for region-specific rice improvement programs that could meet local farmers' demand.

7. SUMMARY DISCUSSION

Myanmar has prosperous plant genetic diversity source with a wide range of extensive ecological background. It constitutes part of the center of genetic diversity of cultivated rice. Rice production is a prominent contribution to national food security and economy. In the Ayeyarwady region, which is major rice production area in Myanmar, farmers mainly grow local landraces in the rainy season because of harsh environmental conditions, specifically salinity and flooding. There are several vernacular names for each variety, which does not always mean to represent genetically diversified cultivars. For efficient conservation and utilization, the genetic diversity and structure of the existing germplasm must be known and therefore, this current study attempted to examine the rice varieties farmers grow in monsoon rice cultivation and their preferred traits on rice varieties, and to evaluate genetic diversity as well as agronomic variations between and among rice genotypes as no such attempt regarding farmer handling landrace pools in the Ayeyarwady region has not yet been conducted.

Farmers' survey provided evidence of on-farm rice varietal diversity in the Ayeyarwady delta and investigated the determinants of varietal selection on-farm. The assessment of the diversity status is the initial but essential step for the implementation of germplasm conservation strategies. Moreover, on-farm seed source is important and popular for indigenous selection for regional adaptation. These assessments are required both at the national level and in regional contexts. In this study, 39 different varieties, i.e., 9 high yielding varieties (HYVs), 10 Pawsans, and 20 traditional non-Pawsans were identified by the farmer-given names in the survey. We planned to include all those rice varieties for genetic diversity study at both phenotypic and genotypic level. However, only 17 varieties were involved because of the lack of seed availability when the survey was conducted. The main drivers of varietal diversification were farming experience and education, which both positively increased the likeliness of growing more varieties at the same time. More varieties were grown by farmers in conditions less suitable for rice cultivation (saline water regions, undulated type of terrain, low-fertility soils), as a strategy to secure at least part of rice production under harsh conditions. These findings may be useful to form the basis for the formulation of farmer-oriented extension and research programs by helping to focus on a particular group of farmers. This study pointed out reduced likeliness of HYVs adoption with higher education, which could be linked to the farmers' awareness of considerable input (labor and costs) requirements for HYVs cultivation and high risk of failure in case of limited access to irrigation, fertilizers, and pesticides.

After the identification of on-farm diversity and farmers' preferences on rice varieties, the study continued to evaluate phenotypic variations of 117 Myanmar rice genotypes involving traditional rice genotypes from farmers' survey and also from local gene bank. Phenotyping of germplasm materials is an important undertaking in genetic resource conservation. Eighty-seven percent of the quantitative traits showed significant variations among rice genotypes, particularly days to heading, culm length, number of tillers per plant, number of spikelets per panicles, and grain length width ratio. According to on-farm survey, farmers preferred the varieties with short culm length (plant height) because of resistance to lodging. Number of

tillers per plant and number of spikelets per panicle are essential traits for high yield, thus, farmers also mentioned them as their preferred traits. In terms of growth duration (DTH), Myanmar traditional rice genotypes are mostly sensitive to photoperiod and grow only in the rainy season. Therefore, DTH depends on the sowing time i.e., late cultivation will shorten the duration and vice versa, however, the optimum growth duration to achieve maximum yield is about 120 days. Diversity analyses for qualitative traits showed that 46% of the traits had high diversity indices with an average index of 0.7. The eating and cooking qualities (ECQ) of rice grains are the major determinants of consumer preferences and, consequently, the economic value of a specific rice variety. ECQ is mainly determined by the level of amylose content (AC), where high AC in the endosperm is usually associated with dry, fluffy, and separated cooked rice grains, and represents the key determinant of poor cooking and eating quality. AC had the highest diversity index and a very high AC was observed as the predominant type followed by medium type AC (~20) that is dominant in local market because of consumer preference. Flag leaf altitude, which is a yield related trait showed the second highest diversity index. Although semi erect type found a predominant character in the rice panel, some genotypes such as MKLAR3, MKLAR5, and KWAR observed as erect type flag leaf that capture efficient light and support to grain filling. Overall, this phenotypic characterization can be utilized as baseline information for any breeding program, particularly for selecting potential parents in consideration of collaborating with farmers' desired attributes observed in on-farm survey, for example, MNAW, TMG, ZTKA for high tillering ability; NYOE, TDNSEIN, HSU for short culm length, etc.

The same panel of rice genotypes was used to investigate genetic variations and relatedness at molecular level employing DArTseq technology (SNPs and silicoDArT). SNPs and silicoDArT markers developed a large number of highly polymorphic markers. To date, this study was the first to use DArTseq technology in genetic diversity analysis on Myanmar rice genotypes. Based on the findings, the rice panel was genetically diverse and two populations that could be explained by regional adaptation and natural selection were identified. The presence of structure in this rice panel was not in accordance with the type of variety observed from farmer's survey, which were to have three populations such as Pawsans, traditional non-Pawsans, and high yielding varieties. HYVs were not found as a separate group. Instead, they were clustered together with several non-Pawsans in pop2. This might be due to breeding activities, suggesting that some breeding activities led to genetic similarities since new varieties were selected from local/traditional varieties. Some non-Pawsans were closely related to Pawsans, and they were grouped together in pop1. The pop2 has a larger number of genotypes and exhibited higher values for genetic diversity indices and was thus more diverse than pop1. According to phenotypic clustering, HYVs also did not group together with Pawsans, except 2 (LPBGYAR and SBPSAN) suggesting that these two Pawsans were phenotypically related to HYVs to some extent. The level of genetic diversity investigated in this study could be the basis for developing new rice varieties with farmers' desirable characteristics such as high yield potential and high tillering capacity. In addition, the traits good salt and submergence tolerance must be considered as the major traits when developing new rice since serious waterlogging and prolonged stagnant flood and salinity are the most common abiotic stresses limiting crop production in the lower Ayeyarwady delta.

The findings using DArTseq markers will be important for future genetic analyses, such as allele/gene identification using genome-wide association studies, which is an approach that can identify the most important alleles for grain quality. Furthermore, this study is advantageous for international trade to avoid adulteration, which means that markers specific for identifying the subgroup of Pawsans could be helpful in identifying true Pawsan, which are highly priced varieties.

8. CONCLUSION AND RECOMMENDATIONS

This study aimed at investigating the on-farm varietal diversity and the determinants of farmers' variety choices in the Ayeyarwady delta in southern Myanmar, specifically for rainfed/monsoon rice cultivation, characterizing rice genotypes by means of agro-morphological traits, and examining the genetic diversity and population structure using DArTseq technology.

The on-farm diversity in the Ayeyarwady region appeared to be relatively high according to the number of varieties grown by the farmers, particularly rich in local landraces. Farmers grew predominantly local varieties over modern HYVs. Diversity and survival of local landraces is totally relying on farmers and their on-farm management. The elite landrace markets need to be promoted for supporting the stability and sustainability of landrace diversity among farmer level since farmers generally appreciate their current varieties for which there is market demand. Information such as farmers' preferences on rice varieties and their awareness of traditional rice varieties should be considered as the preliminary factors for innovation and motivation of systematic on-farm conservation procedures among farmers.

Assessment of genetic diversity using agro-morphological characters is the traditional method of measuring diversity. We suggest that characterizing phenotypic traits of the studied rice panel should be repeated under different environments to check whether they equally express in all environments. An easy and quick discriminated character between phenotypes; in this study e.g., days to heading, culm length, stigma color, panicle and spikelet numbers, etc. may be a pick-up varietal attribute from local landraces for rice improvement plan. High variations of rice genotypes at phenotypic level were found in the studied rice panel suggesting that they can be the potential pool for breeding purpose. Moreover, farmers' preferred traits observed in on-farm survey should also be put together under consideration of varietal improvement. Other than high yield, farmers preferred rice varieties that are tolerant to abiotic and biotic stress conditions i.e., saline environment and flooding, and pest infestation. Though breeding cannot incorporate all desired attributes, the vital features, such as tolerance to those stresses, should be introgressed in particular varieties, so that they can meet the demands of farmers.

Two populations clearly identified at molecular level using DArTseq technology and diversity level between populations can also be a promising gene pool for quality improved breeding purposes. Cluster analysis clearly separated traditional Pawsan varieties and modern high-yielding varieties. More information will be gained through genome-wide association studies (GWAS) of rice landraces as combining SNPs based sequencing and phenotypic data, however, a larger number of broadly representative landraces (at least 500) should be sampled. Larger sample size and higher genetic diversity provide sufficient power for association analysis. The smaller population size and low genetic diversity would limit the power of GWAS. Several follow-up steps could be taken to pinpoint candidate genes via application of rice functional-genomics approaches.

For effective utilization in breeding program, grain quality traits such as grain shape (determined by three dimensions including grain length, grain width and grain length to width ratio), amylose content (volume expansion, texture tenderness, and gloss scores of cooked rice regardless of water/rice ratio used during cooking), gelatinization temperature (GT) (the time required to cook milled rice), and gel consistency (GC) (measures the tendency of the cooked rice to harden on cooling) should be evaluated. This data can be used for grouping the Myanmar rice varieties. However, in this study, only grain shape and amylose content were evaluated and therefore, we suggest that evaluations on GT and GC of the same rice panel should be conducted in order to cluster rice varieties based on grain quality traits since they could also be useful genomic resources for genome-wide association studies of rice grain quality traits to accelerate varietal development and release.

All in all, this whole study could be a complete set of research for region specific rice improvement program by providing information about farmers' current grown rice varieties and their preferences on rice varietal traits, phenotypic characters for each variety, and identifying the genetic basis of these diverse varieties will provide important insights for breeding elite varieties.

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Appendix 4. Author's curriculum vitae

Appendix 1. List of publications

Thant AA, Zaw H, Kalousova M, Singh RK, Lojka B. 2021. Genetic diversity and population structure of Myanmar rice (*Oryza sativa* L.) varieties using DArTseq-based SNP and SilicoDArT markers. *Plants* **10** (12): 2564. DOI: 10.3390/plants10122564.

Thant AA, Teutscherova N, Vazquez E, Kalousova M, Phyto A, Singh RK, Lojka B. 2020. On-farm rice diversity and farmers' preferences for varietal attributes in Ayeyarwady Delta, Myanmar, *Journal of Crop Improvement* **34** (4): 549–570. DOI: 10.1080/15427528.2020.1746457.

Thant AA, Htun TM, Kalousova M, Singh RK, Lojka B. 2018. Microsatellite markers and their application on genetic diversity studies of rice landraces (*Oryza sativa* L.) in Myanmar - A Review. *International Journal of Environmental and Rural Development* **9** (2): 8–13. DOI: 10.32115/ijerd.9.2_8.

Appendix 2. List of conference contributions

Thant AA, Zaw H, Kalousova M, Singh RK, Lojka B. 2020. Genomic studies of Myanmar rice (*Oryza sativa* L.) varieties using DArT and SNP markers. Poster presentation at TROPENTAG on 9–11 Sep 2020, Virtual Conference, ATSAF e.V., Germany

Thant AA, Zaw H, Kalousova M, Aung HH, Singh RK, Lojka B. 2019. Phenotypic diversity of rice landraces collected from Ayeyarwady region, Myanmar using agro-morphological characterization. Poster presentation at TROPENTAG on 18–20 Sep 2019, University of Kassel, Kassel, Germany.

Thant AA, Kalousova M, Singh RK, Lojka B. 2018. On-farm rice diversity and farmers' preferences to varietal attributes in Ayeyarwady delta, Myanmar. Poster presentation at TROPENTAG on 17–19 Sep 2018, Ghent University, Ghent, Belgium.

Thant AA, Htun TM, Kalousova M, Singh RK, Lojka B. 2018. Microsatellite markers and their application on genetic diversity studies of rice landraces (*Oryza sativa* L.) in Myanmar - A review. Poster presentation at The 9th International Conference on Environmental and Rural Development on 24–25 Feb 2018, Yezin Agricultural University, Nay Pyi Taw, Myanmar.

Thant AA, Kalousova M, Than H, Lojka B. 2017. Effects of seed sizes and varieties on growth, yield, and oil and protein contents of groundnut (*Arachis hypogaea* L.). Poster presentation at TROPENTAG on 20–22 Sep 2017, University of Bonn, Bonn, Germany.

Appendix 3. Questionnaires for diversity of rice varieties in the Ayeyarwady region, Myanmar

Questionnaires for diversity of rice varieties in the Ayeyarwaddy region, Myanmar

CONSENT STATEMENT

Hello! You were randomly selected to participate in the research study “**Agro-morphological traits and genetic diversity of rice landraces in the Ayeyarwaddy Region, Myanmar**” which is conducted to understand “**Diversity of rice varieties**”. I would like to ask you some questions related to your rice production, varietal use, and general characteristics of your household and farm. The information you provide will be used to document the rice varieties which are grown in this region, and identify factors why farmers are growing those varieties.

The interview will take approximately 20 minutes. Your participation is voluntary. You may opt to refuse to participate or withdraw at any time. All information you provide will be kept confidential and your privacy will be protected.

You may ask any questions you have about the study. If you have questions later, they can be directed to **Ms. Aye Aye Thant**, phone number **+95 9420318612**.

Date of Interview: / /

***Continue survey if farmers grow traditional variety on any of their fields.**

A. Basic information

Q1. Name of Farmer

Q2. Village/Village Tract

Q3. Township

Q4. District/Division

Q5. No. of Years Spent on Farming

Q6. Cropping Pattern Rice-Rice Rice-Pulse Other

Q7. Latitude

Q8. Longitude

Q9. Elevation (m)

[Water region Fresh Brackish Saline]

Note: Interview the farmer who manages the farm and not any other household member.

B. Household characteristics

(Include only persons staying in the house for more than 2 days per week in 2016)

Q1. Name of HH member	Q2. Relation to farmer respondent (<i>See codes</i>)	Q3. Sex Male (M) /Female (F)	Q4. Age	Q5. Marital Status (<i>See codes</i>)	Q6. Education (years completed)	Q7. Occupation specific (<i>See codes</i>)	Q8. Years in farming	Q9. Notes

Q2.

- 1=Spouse
- 2=Son
- 3=Daughter
- 4=Brother
- 5 =Sister
- 6= Father
- 7=Mother
- 8=Son-in-law
- 9=Daughter-in-law
- 10=Father-in-law
- 11=Mother-in-law

- 12=Brother-in-law
- 13=Sister-in-law
- 14=Nephew
- 15=Niece
- 16=Permanent servant
- 17=Friend
- 18= Others (specify)

Q5.

- 1= Single
- 2= Widow/Widower
- 3= Married
- 4= Separated
- 5= Live-in partner
- 6= Others (specify)

Q7.

- 1=Jobless
- 2=Farmer
- 3=Agricultural labor
- 4=Housewife
- 5=Student
- 6=Retired
- 7=Small shop owner
- 8=Plumber
- 9=Others (specify)

C. Landholding

(List all types of land and water bodies owned or operated by the farmer during wet season 2016.)

Q1. Parcel type (<i>See codes</i>)	Q2. Area	Q3. Unit (Area)	Q4. Tenure status (<i>See codes</i>)	Q5. Rental value (kyat/year)	Q6. Current value of parcel if owned (kyat)	Q7. Field type (<i>See codes</i>)	Q8. Soil type (<i>See codes</i>)	Q9. Soil fertility (<i>See codes</i>)	Q10. Most serious constraints (<i>See codes</i>)	Q11. Planted to rice in at least 1 season (Yes/No)	Q12. Notes

- Q1.**
 1=Cultivable/arable land
 2=Pasture
 3=Bush/forest
 4=Waster/non-arable land
 5=Land in riverbed
 6=Cultivable pond
 7=Derelict pond

- 8=Orchard
 9=Others (specify)
Q4.
 1= Owner-cultivator
 2= Share-tenant
 3= Leasehold (fixed rent)
 4= Others (specify)

- Q7.**
 1= Flat
 2= Undulated
Q8.
 1= Clay
 2= Loam
 3= Sandy
 4= Sandy Loam
 5= Others (specify)

- Q9.**
 1=High fertility
 2=Average fertility
 3=Low fertility
Q10.
 1=Drought
 2=Submergence
 3=Salinity
 4=Acidity
 5= Others (specify)

D. Crops grown and rice varieties

(List all rice and non-rice crops grown by the farmer in wet season 2016.)

Q1. Crop	Q2. Area planted	Q3. Unit (Area)	Q4. Variety name (for rice only)	Q5. Farmers classification of rice variety (TV/MV/Hybrid)	Q6. Production	Q7. Unit (Grains)	Q8. Kg equivalent per unit	Q9. Farm gate price per unit	Q10. Unit of price	Q11. Notes

E. Rice varieties and seeds

Rice varieties (from module D)	Q1. Method of establishment (See codes)	Q2. Date of TPR/sow ing (month)	Q3. Date of TPR/sow ing (week)	Q4. Date of harvestin g (month)	Q5. Quantity of seeds used	Q6. Unit (Grains)	Q7. Kg- equivalent/u nit	Q8. Price of seed if purchased (kyat/unit)	Q9. Unit (Price)	Q10. Source of seed (See codes)	Q11. Seed type/class (See codes)	Q12. Notes

Q1.

- 1=Transplanting
- 2=Wet seeding
- 3=Dry seeding
- 4=Line sowing
- 5=Others (specify)

Q10.

- 1=Own harvest
- 2= Exchange/co-farmer
- 3= Seed grower
- 4= Input dealer
- 5= DOA
- 6= Private companies (specify)
- 7= Landowner
- 8= Others (specify)

Q11.

- 1= Hybrid Seeds
- 2= Registered Seeds
- 3= Certified Seeds
- 4= Good Seeds
- 5= Own or Farmer's Seeds
- 6= Farmer Exchange
- 7= Others (specify)

F. Farmer's preferences for rice variety traits

1 = extremely important 2 = important 3 = not important

	Rice variety traits	Farmer's preference	Notes
F1. Production characteristics			
1	High yield		
2	Give high yield with less inputs/low production costs		
3	High straw production		
4	Shorter growing duration		
5	Resistant to lodging		
6	Resistant to shattering		
7	High tillering		
F2. Stress tolerance characteristics			
1	Resistant to insect		
2	Resistant to diseases		
3	Tolerant to drought		
4	Tolerant to flood/submergence		
5	Tolerant to salinity		
6	Tolerant to cold injury		
F3. Grain quality characteristics			
1	Size (long/medium/short)		
2	Shape (short/bold/slender/medium)		
5	High milling recovery		
6	High consumer/trader/market demand (marketability)		
7	Softness		
8	Short cooking time		
9	Good taste/palatability/eating quality		
10	Good aroma/smell		
11	Stickiness of cooked rice		
12	Longer keeping quality of cooked rice		
13	High volume expansion of cooked rice		
14	Hard-texture white rice with high amylose content (slower starch digestion)		
15	Less broken grains		
F4. Other preferred traits/include soil problems			

G. Major problems in rice production

What are the top 3 major problems/constraints in rice cultivation? Please rank the problems according to importance on a scale of 1-3 with 1 = most important and 3 = least important.

Q1. Problems	Q2. Rank
1.	
2.	
3.	

H. Awareness of traditional rice varieties

(Put a tick mark on the appropriate response)

Q1. Have you noticed the advantages of traditional rice varieties?

- Yes - No

If yes, what are those? *(See codes IN F)*

Q2. Have you noticed the disadvantages of traditional rice varieties?

- Yes - No

If yes, what are those?

Q3. Do you have any plan to keep growing traditional rice varieties?

Yes No

If yes, please provide information on the following:

Q4. Traditional rice varieties	Q5. Source of seed <i>(See codes)</i>	Q6. Why you want to keep growing that particular traditional variety

Q5.

1=Own harvest

3= Seed grower

5= DOA

7= Landowner

2= Exchange/co-farmer

4= Input dealer

6= Private companies
(specify)

8= Others (specify)

If No, please provide information on the following:

Q7. Which varieties want to replace current varieties	Q8. Source of seed (See codes)	Q9. Why you want to grow that particular variety

Q8.

1=Own harvest

3= Seed grower

5= DOA

7= Landowner

2= Exchange/co-farmer

4= Input dealer

6= Private companies
(specify)

8= Others (specify)

Appendix Table A1. 117 Myanmar rice genotypes and their collection locations

No.	Genotypes	Short name	Place of collection	Year released	Prominent traits
1	Tu_Maung	TMG	Myaungmya		
2	Nga_Kywe_Phyu	NKPHWE	Mawtin		
3	Bahat_Ni_1	BHNI1	Mawtin		
4	Pauk_War_Yin	PWYIN	Thayaungchaung		
5	Gay_Ku_Kama_Kyi_1	GKKYI1	Myaungmya		
6	Phya_Pon_Thee_Htat	PPTHTAT	Myaungmya		
7	Emata_Lone_Pu	ELPU	Myaungmya		
8	Mae_Khalar_5	MKLAR5	Thayaungchaung	1980	Deep water rice, high yield, medium duration, flooding and elongation ability
9	Wa_Khe_Ma_Hnan_Kar	WKMHKAR	Thapaung		
10	Sein-Ta_Lay	STLAY	Thayaungchaung	1975	High yield, rainfed lowland rice with low amylose
11	Zaw_Gyi_Pyan_(Mee_Don)	ZGPYAN	Thapaung		
12	Khun_War	KWAR	Thayaungchaung		
13	Bay_Kyaung	BKYAUNG	Yekyi		
14	Aung_Ze_Ya_1	AZYA1	Thapaung		
15	Shwe-Ta_Soke	STSOKE	Pathein	1979-80	High yield, rainfed lowland rice with high amylose
16	Kyaw_Ze_Ya	KZYA	Pathein	1980	
17	Pyi_Gyi_Thu	PGTHU	Thayaungchaung		
18	Shan_Nyein_1	SNYEIN1	Thayaungchaung	1924	Late, photosensitivity, local glutinous rice
19	Ye_Baw_Sein	YBSEIN	Thayaungchaung	1952	Late, local quality rice, long grain, low amylose
20	Inma_Ye_Baw	IYBAW	Thayaungchaung		Late, local var, photosensitivity, long grain, low amylose, soft eating quality
21	Ye_Ngan_Ka_Lin	YNKLIN	Myaungmya		
22	Kalai	KALAI	Myaungmya		
23	Lone_Pu	LPU	Myaungmya		
24	Bala_Gya	BGYA	Myaungmya		High millig outcomes, resistant to rice blast
25	Hteik_Pyaung	HPYAUNG	Myaungmya		
26	Hnan_Su	HSU	Myaungmya		
27	Taung_Hti_1	THTI1	Myaungmya		
28	Thee_Dat_Nga_Sein_1	TDNSEIN1	Myaungmya		

29	Thee_Dat_Nga_Sein_2	TDNSEIN2	Einme		
30	Longyi_Kauk_Kyi	LKKYI	Myaungmya		
31	Maung_Nyo	MNYO	Myaungmya		
32	Za_Lun	ZLUN	Myaungmya		
33	Zaw_Ti_Ka	ZTKA	Myaungmya		
34	Mya_Naung_Kauk_Kyi	MNKKYI	Myaungmya		
35	Toe_Pwa_Gyi_1	TPGYI1	Myaungmya		
36	Shwe_Dinga_1	SDINGA1	Myaungmya	1956	Late, photosensitivity, local quality rice, long grain
37	Hnan_Kar_Tun	HKTUN	Myaungmya		
38	Daung_Nga_Sein	DNSEIN	Kyaungon		
39	Nga_Kywe	NKYWE	Myaungmya	1925	Late, photosensitivity, roundish grain, aromatic, good elongation ability
40	Shwe_Dinga_2	SDINGA2	Myaungmya	1956	Late, photosensitivity, local quality rice, long grain
41	Nga_Kyauk_Kyi	NKKYI	Myaungmya		
42	Nga_Kyauk_Htan	NKHTAN	Myaungmya		
43	Kywe_Chae_Manaing_1	KCMNAING1	Myaungmya		
44	Sit_Pwa	SPWA	Myaungmya		
45	Shan_Nyein_2	SNYEIN2	Myaungmya	1924	Late, photosensitivity, local glutinous rice
46	Taung_Hti_2	THTI2	Myaungmya		
47	Ya_Tha_Lay_Lone_Thwe	YTLLTHWE	Myaungmya		
48	Thet_Lat_Nga_Kyauk	TLNKYAUK	Myaungmya		
49	Bahat_Ni_2	BHNI2	Myaungmya		
50*	Paw_San_Bay_Gyar_1	PSBGYAR1	Myaungmya	1960	Late, weakly photosensitive, aromatic chalky
51	Kywe_Chae_Manaing_2	KCMNAING2	Myaungmya		
52	Toe_Pwa_Ga_Lay	TPGLAY	Myaungmya	1952	High milling outcomes, good eating quality
53	Mee_Kauk_1	MKAUK1	Myaungmya	1969	Late, medium long, tolerance to submergence, local variety
54	Aung_Ze_Ya_2	AZYA2	Myaungmya		
55	Toe_Pwa_Gyi_2	TPGYI2	Myaungmya		
56	Gay_Ku_Kama_Kyi_2	GKKYI2	Myaungmya		
57	Moke_Seik	MSEIK	Myaungmya		
58	Mee_Don	MDON	Wakema		
59	Aung_Ze_Ya_3	AZYA3	Thapaung		

60	Mae_Khalar_3	MKLAR3	Thayaungchaung	1980	Deep water rice, high yield, medium duration, flooding and elongation ability
61	Nat_Pyi_Hmwe	NPHMWE	Einme		
62	Soke_Phwa	SPHWA	Nyaungdon		
63	Lu_Pyo_Kyi	LPKYI	Danubyu		
64	Mwe_Swe_(Yekyi)	MSWE	Yekyi		
65	Kyaung_Yoe_Tun	KYTUN	Hinthada		
66	Nga_Ni_Gyi_Lone_Thay	NNGLTHAY	Labutta		
67*	Paw_San_Yin_1	PSYIN1	Labutta		
68	Naung_Yoe	NYOE	Hinthada		
69	Mine_Kauk	MIKAUK	Hinthada		
70	Hnan_Kar_1	HKAR1	Einme	1957	Late, tolerance to submergence, elongation ability, photosensitivity
71	Mee_Kauk_2	MKAUK2	Pathein	1969	Late, medium long, tolerance to submergence, local variety
72*	Paw_San_Hmwe	PSHMWE	Pathein	1944	Late, photosensitivity, chalky, roundish grain
73	Ka_Mar_Kyi	KMKYI	Bogale		
74	Nga_Kywe-Taung_Pyan	NKTPYAN	Pathein		Late, photosensitivity, RG, chalky, long sterile lemma
75	Nga_Kywe_Hmwe	NKHMWE	Pathein		
76	Thaung_Kyaw	TKYAW	Mawlamyinegyun		
77	Khun_Ni_1	KNI1	Labutta		
78	Annarwarbo_1	ANWBO1	Labutta		
79	Mee_Don_Thet_Lat	MDTLAT	Labutta		
80	Late_Ma	LMA	Bogale		
81*	Thet_Lat_Bay_Gyar	TLBGYAR	Bogale		
82*	Bay_Gyar_Lay	BGLAY	Bogale		
83	Manaw_1	MNAW1	Bogale		
84*	Paw_San_Gyi	PSGYI	Bogale		
85	Manaw_2	MNAW2	Bogale		
86	Hnan_Kar_2	HKAR2	Bogale	1957	Late, tolerance to submergence, elongation ability, photosensitivity
87	Annarwarbo_2/Ye_Ngan_Bo	ANWBO2	Mawlamyinegyun		
88*	Paw_San_Yin_2	PSYIN2	Bogale		
89	Khun_Ni_2	KNI2	Bogale		
90	Madama	MDMA	Mawlamyinegyun		

91*	Bay_Gyar_Gyi_Mee_Shay	BGGMSHAY	Bogale		
92*	Taung_Pyan_Yin	TPYIN	Bogale		
93*	Lat_Pan_Bay_Gyar	LPBGYAR	Bogale		
94	Sakarwar_Hmwe	SHMWE	Bogale		
95	Hai_Gyi	HGYI	Bogale		
96	Si_Htun	SHTUN	Bogale		
97	Nga_Kyi_Ni	NKNI	Bogale		
98	Manaw_3	MNAW3	Bogale		
99	Khun_Ni_Ma	KNMA	Bogale		
100	Kauk_Hnyin	KHNYIN	Bogale		
101	Hmaw_Bi_San	HBSAN	Bogale		
102	Latyonegyi	LYGYI	Bogale		
103*	Paw_San_Bay_Gyar_2	PSBGYAR2	Myaungmya	1960	Late, weakly photosensitive, aromatic chalky
104*	Paw_San_Bay_Gyar_3	PSBGYAR3	Myaungmya	1960	Late, weakly photosensitive, aromatic chalky
105	Hnan_Kar_3	HKAR3	Myaungmya	1957	Late, tolerance to submergence, elongation ability, photosensitivity
106*	Shwe_Bo_Paw_San	SBPSAN	Myaungmya		
107	Kauk_Kyi-Taung_Pyan	KKTPYAN	Myaungmya		
108	Mee_Kauk_3	MKAUK3	Myaungmya	1969	Late, medium long, tolerance to submergence, local variety
109*	Paw_San_Yin_3	PSYIN3	Myaungmya		
110*	Taung_Pyan_Bay_Gyar	TPBGYAR	Myaungmya		
111*	Pathein_Bay_Gyar	PBGYAR	Myaungmya		
112*	Bay_Gyar_Gyi	BGGYI	Myaungmya		
113↓	Manawthukha	C1	Yezin (IRRI)	1978	Medium, rainfed lowland and irrigated, medium grain, high milling out turn, popular variety in Myanmar
114↓	Sinthukha	C2	Yezin (IRRI)	2007	Medium, resistance to BB, small grain size, good eating quality
115↓	Ayeyarmin	C3	Yezin (IRRI)	1977	Medium, rainfed lowland rice, intermediate amylose
116↓	Pyimyanmarsein	C4	Yezin (IRRI)	2014	Early, tolerance to salinity and drought tolerance
117↓	Pyitawyin	C5	Yezin (IRRI)	2013	Early, irrigated, tolerance to inland salinity

Genotype no. 1-72 were provided by the Department of Agricultural Research (DAR) seed bank section in Yezin. Pawsan varieties are marked as (*); HYVs are marked as (↓).

Appendix Table A2. Monthly weather data for 2017, Yezin Agrometeorological Station, Nay Pyi Taw, Myanmar

Month	Temperature (°C) ¹		Rainfall (mm) ²	Relative Humidity (%) ³
	Max.	Min.		
January	33.00	17.01	17.00	65.65
February	35.04	17.97	9.00	53.30
March	37.51	20.54	18.00	53.39
April	37.40	24.37	58.00	61.00
May	38.53	26.25	93.00	65.87
Jun	32.96	25.27	293.00	85.97
July	32.07	24.78	182.00	86.10
August	32.10	24.90	392.00	87.45
September	33.42	25.21	213.00	86.20
October	32.62	24.35	493.00	87.58
November	33.24	21.95	46.00	81.70
December	31.30	17.86	0.00	79.16

¹ Monthly means maximum and minimum temperature; ² Monthly total rainfall; ³ Monthly mean relative humidity

Appendix Table A3. Quantitative and qualitative traits, the methods of measurement and evaluation phase

Traits	Methods of measurement	Evaluation phase
Quantitative		
Days to heading (DTH)	Arithmetic means of the five random sample	80% heading date
Culm length (cm) (CL)	Arithmetic means of the five random sample	After flowering to maturity
Panicle length (cm) (PL)	Arithmetic means of the five random sample	7 days after anthesis or upon full panicle exertion
Long sterile lemma length (mm) (LSL)	Arithmetic means of the five random sample	Ripening
Short sterile lemma length (mm) (SSL)	Arithmetic means of the five random sample	Ripening
No. of tillers per plant (NTPP)	Arithmetic means of the five random sample	Ripening
No. of panicles per plant (NPP)	Arithmetic means of the five random sample	Ripening
No. of spikelets per panicle (NSPP)	Arithmetic means of the five random sample	Maturity
1000-grain weight (TGW)	Random sample of 1000 well-developed, whole grains, 13% moisture content	Maturity
Grain length (mm) (GL)	Arithmetic means of 10 representative grains	Post-harvest
Grain width (mm) (GW)	Arithmetic means of 10 representative grains	Post-harvest
Length/width ratio (LWR)	Arithmetic means of 10 representative grains	Post-harvest
Filled grain % (FG %)	Arithmetic means of 5 representative panicles	Post-harvest
Harvest index (HI)	Arithmetic means of the five random sample	Post-harvest
Yield (Y) 5hills (kg)	Arithmetic means of the five random sample	Post-harvest
Qualitative		
Auricle color (AC1)	1, light green; 2, purple; 3, green	Late vegetative
Flag leaf attitude (early observation) (FLA_E)	1, erect; 3, semi erect; 5, horizontal; 7, descending	Anthesis
Stigma color (SC)	1, white; 2, light green; 3, yellow; 4, light purple; 5, purple	Anthesis (between 09:00 and 14:00)
Sterile lemma color (SLC)	1, straw; 2, gold; 3, red; 4, purple	7 days after flowering
Lemma color of apiculus (CoA)	1, white; 2, straw; 3, brown; 4, green; 5, red; 6, red apex; 7, purple; 8, purple apex; 9, black; 10, none	After anthesis to hard dough stage
Awns distribution (AD)	0, none; 1, tip only; 2, upper quarter only; 3, upper half only; 4, upper three-quarters only; 5, whole length; 6, few randomly	Flowering to maturity
Lemma and palea color (LPC)	1, white; 2, straw; 3, gold and gold furrows; 4, brown; 5, brown spots; 6, brown furrows; 7, purple; 8, reddish to light purple; 9, purple spots; 10, purple furrows; 11, black; 12, straw furrows; 13, black furrows	Ripening
Lemma and palea pubescence (LPP)	1, glabrous; 2, hairs on lemma keel; 3, hairs on upper portion; 4, short hairs; 5, long hairs (velvety); 6, long upper hairs; 7, short upper hairs	Ripening

Panicle attitude of main axis (PA_M)	1, upright; 2, semi upright; 3, slightly drooping; 4, strongly drooping	Near maturity
Panicle attitude of branches (PA_B)	1, erect (compact panicle); 3, semi erect (semi compact panicle); 5, spreading (open panicle); 7, horizontal; 9, drooping	Near maturity
Flag leaf attitude (late observation) (FLA_L)	1, erect; 3, semi erect; 5, horizontal; 7, descending	Maturity
Amylose content (AC2)	0, waxy-glutinous (<3); 1, very low (~9); 3, low (~17); 5, intermediate (~20); 7, high (~23); 9, very high (>25)	After harvest
Endosperm type (ET)	1, non-glutinous (non-waxy); 2, intermediate; 3, glutinous (waxy)	After polishing

Appendix Table A4 List of SNP markers is available online at https://osf.io/vy736/?view_only=c07f9565c7de472c82cd976e9054ac5e .

Appendix Table A5 List of silicoDArT markers is available online at https://osf.io/vy736/?view_only=c07f9565c7de472c82cd976e9054ac5e .

Appendix Table A6. Percentage of farmers growing one or more varieties of rice in the study area

No. of varieties	Percentage distribution					
	Bogale	Mawlamyinegyun	Labutta	Myaungmya	Pathein	Total
1	23.3	16.1	6.67	24.1	16.7	17.3
2	30.0	38.7	56.7	44.8	63.3	46.7
3	40.0	29.0	36.7	31	16.7	30.7
4	6.67	16.1	0.00	0.00	3.33	5.33

Appendix Table A7. Awareness of the most important benefits of traditional rice varieties

Benefit	Frequency	%
Good market value	74	49
Adaptability to weather condition	28	19
Good taste/palatability	20	13
Resistance to lodging	10	7
Low production costs	6	4
Yield stability	6	4
Others	6	4
Total	150	100

Appendix Table A8. Percentage of farmers' used and yield of each variety

Traditional varieties			Pawsan varieties			High yielding varieties		
Name	% of farmer used	Yield (t/ha)	Name	% of farmer used	Yield (t/ha)	Name	% of farmer used	Yield (t/ha)
Manaw	13.33	2.98	Pawsan yin	35.33	2.43	Ayeyar padathar	9.33	1.88
Hnan kar	10.00	2.90	Baygyar (thet nge) 4.5 months	23.33	3.16	Thee htet yin	7.33	5.06
Ma da ma	9.33	2.79	Baygyar (thet gyi) 5.5 months	20.00	2.43	Manaw thukha	6.00	3.16
Kyauk kyi	9.33	2.36	Baygyar (thet latt) 5 months	13.33	2.45	Shwe war yin	3.33	3.16
Kauk nyin	6.00	2.47	Mee shay	9.33	2.76	Yet koe sae (90-days)	2.00	3.41
Nga kyauk	6.00	2.44	Pawsan sheal (6months)	6.00	2.85	Sin thwe latt	1.33	3.29
Nga sein	4.67	3.19	Pathein pawsan	1.33	2.37	Thai baykyar	1.33	3.89
Khun ni ma	3.33	2.82	Hmawbi pawsan	1.33	2.98	Ma gyan daw	1.33	2.59
Chaung thar	3.33	2.86	Ayeyarwady pawsan	1.33	2.82	Sin shwe lee	0.67	6.75
Nga kyi ni	3.33	2.49	Taung pyan	0.67	3.11			
Pakhan	2.00	2.72						
Hnan kar htun	2.00	2.82						
Phoe shin	2.00	3.00						
Sar gyin	1.33	1.78						
Late ma	1.33	2.21						
Khun war	0.67	2.13						
Yoe sein	0.67	2.34						
Sagarwar hmawe	0.67	2.08						
Nga kyawe	0.67	2.79						
Mee done	0.67	3.63						

Appendix Table A9 Complete data set of quantitative traits is available online at https://osf.io/vy736/?view_only=c07f9565c7de472c82cd976e9054ac5e .

Appendix Table A10. Variability in qualitative traits of 117 Myanmar rice genotypes

No.	Traits	States	Frequency	Percent
1	Auricle color (AC1)	light green	67	57.26
		purple	46	39.32
		green	4	3.42
2	Flag leaf altitude (early observation) FLA_E	erect	28	23.93
		semi erect	73	62.39
		horizontal	16	13.68
3	Stigma color (SC)	white	30	25.64
		light green	68	58.12
		yellow	15	12.82
		purple	4	3.42
4	Sterile lemma color (SLC)	straw	101	86.32
		gold	14	11.97
		purple	2	1.71
5	Color of apiculus (CoA)	straw	9	7.69
		brown	26	22.22
		red	8	6.84
		purple	10	8.55
		black	5	4.27
		none	59	50.43
6	Awns distribution (AD)	none	92	78.63
		tip only	13	11.11
		upper three-quarters only	2	1.71
		whole length	3	2.56
		few randomly	7	5.98
		none	59	50.43
7	Lemma and palea color (LPC)	straw	32	27.35
		gold and gold furrows	11	9.40
		brown furrows	19	16.24
		straw furrows	53	45.30
		black furrows	2	1.71
8	Lemma and palea pubescence (LPC)	glabrous	5	4.27
		hairs on upper portion	39	33.33
		short hairs	40	34.19
		long hairs (velvety)	4	3.42
		long upper hairs	27	23.08
		short upper hairs	2	1.71
9	Panicle altitude of main axis (PA_M)	semi upright	3	2.56
		slightly drooping	56	47.86
		strongly drooping	58	49.57
10	Panicle altitude of branches (PA_B)	erect (compact panicle)	2	1.71
		semi erect (semi compact panicle)	29	24.79
		spreading (open panicle)	63	53.85
		horizontal	23	19.66
11	Flag leaf altitude (late observation) (FLA_L)	erect	2	1.71
		semi erect	59	50.43

		horizontal	49	41.88
		descending	7	5.98
12	Amylose content (AC2)	very low (~9)	6	5.13
		low (~17)	8	6.84
		intermediate (~20)	38	32.48
		high (~23)	18	15.38
		very high (>25)	47	40.17
13	Endosperm type (ET)	cloudy and translucent	79	67.52
		intermediate	35	29.91
		waxy white	3	2.56

Appendix Table A11. Summary statistics of informative markers of both markers of DArT platform

Component	SNP	silicoDArT
Mean	0.40497508	0.371728278
Standard Error	0.001521129	0.00152924
Median	0.449996	0.405983
Mode	0.449996	0.429688
Standard Deviation	0.096971251	0.097488291
Sample Variance	0.009403423	0.009503967
Kurtosis	1.500348356	-0.91676601
Skewness	-1.404134877	-0.551033185
Range	0.5	0.48261
Minimum	0	0.01739
Maximum	0.5	0.5

Appendix Table A12 Dissimilarity indices among 117 Myanmar rice genotypes estimated by neighbor-joining analysis of 7,643 DArTseq based SNP markers in DARwin software is available online at https://osf.io/vy736/?view_only=c07f9565c7de472c82cd976e9054ac5e.

Appendix Table A13 Dissimilarity indices among 117 Myanmar rice genotypes estimated by neighbor-joining analysis of 4,064 silicoDArT markers in DARwin software is available online at https://osf.io/vy736/?view_only=c07f9565c7de472c82cd976e9054ac5e.

Appendix Table A14 A list of membership in two population groups is available online at https://osf.io/vy736/?view_only=c07f9565c7de472c82cd976e9054ac5e.

Appendix Table A15 Complete data set of qualitative traits is available online at https://osf.io/vy736/?view_only=c07f9565c7de472c82cd976e9054ac5e.

Appendix Table A16 A list of membership in three population groups is available online at https://osf.io/vy736/?view_only=c07f9565c7de472c82cd976e9054ac5e.

DESIGN PARAMETERS for Augmented RCBD:

Number of Trials = 1

Number of Replicated Treatments = 5

Levels of Replicated Treatments = check1, check2, check3, check4, check5

Number of Blocks = 4

Number of Unreplicated Treatments = 112

Levels of UnReplicated Treatments = new1, new2, new3..., new112

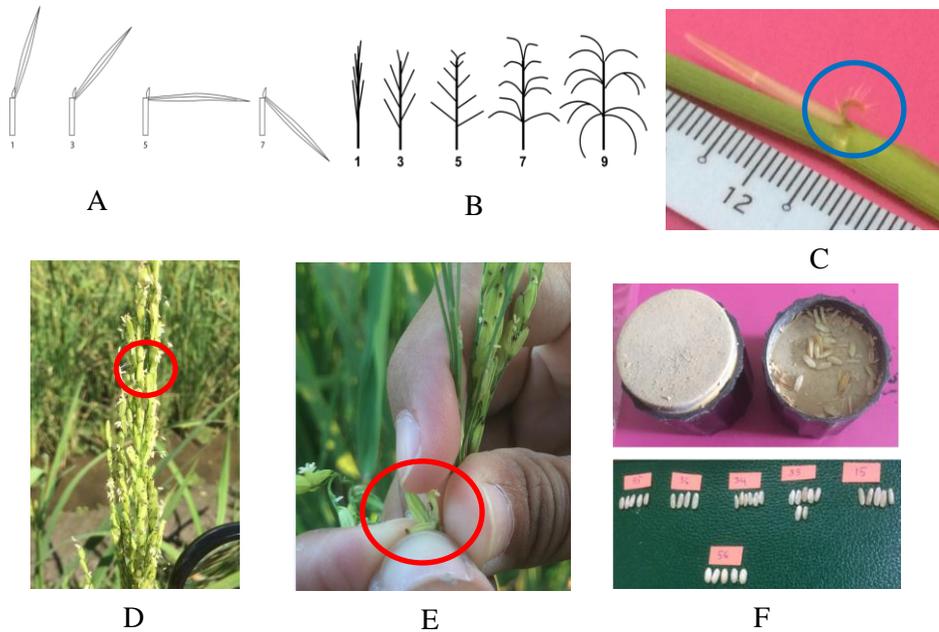
Number of Field Rows = 4

FieldRow1	101	102	103	104	105	106	107	108	109	110	111	112	113	114	115	116	117	118	119	120	121	122	123	124	125	126	127	128	129	130	131	132	133
	new82	check2	new32	new8	new54	new103	new30	new102	new69	new36	new16	new45	new104	check4	new55	new2	new33	check3	new21	new91	check1	check5	new4	new20	new112	new92	new84	new107	new19	new24	new34	new41	new110
FieldRow2	201	202	203	204	205	206	207	208	209	210	211	212	213	214	215	216	217	218	219	220	221	222	223	224	225	226	227	228	229	230	231	232	233
	new83	check1	new93	check3	new106	new62	new85	new13	new12	new100	new40	new87	new46	new78	new59	new76	new70	new47	new53	new68	new35	new23	new94	check5	new42	new95	new22	check4	new10	new72	new15	check2	new63
FieldRow3	301	302	303	304	305	306	307	308	309	310	311	312	313	314	315	316	317	318	319	320	321	322	323	324	325	326	327	328	329	330	331	332	333
	new89	new80	new67	new52	new101	new49	new60	new81	new14	new56	new6	check2	new58	new97	check1	new50	new73	check4	new5	new74	new64	check5	new39	new66	check3	new11	new48	new86	new18	new79	new96	new61	new99
FieldRow4	401	402	403	404	405	406	407	408	409	410	411	412	413	414	415	416	417	418	419	420	421	422	423	424	425	426	427	428	429	430	431	432	433
	new31	new51	new108	new88	check4	new37	new75	new109	new26	check3	check5	new105	new44	check2	new65	new57	new25	new77	new28	new7	new98	new38	new111	new3	new9	new1	new29	new17	new90	new27	new43	new71	check1

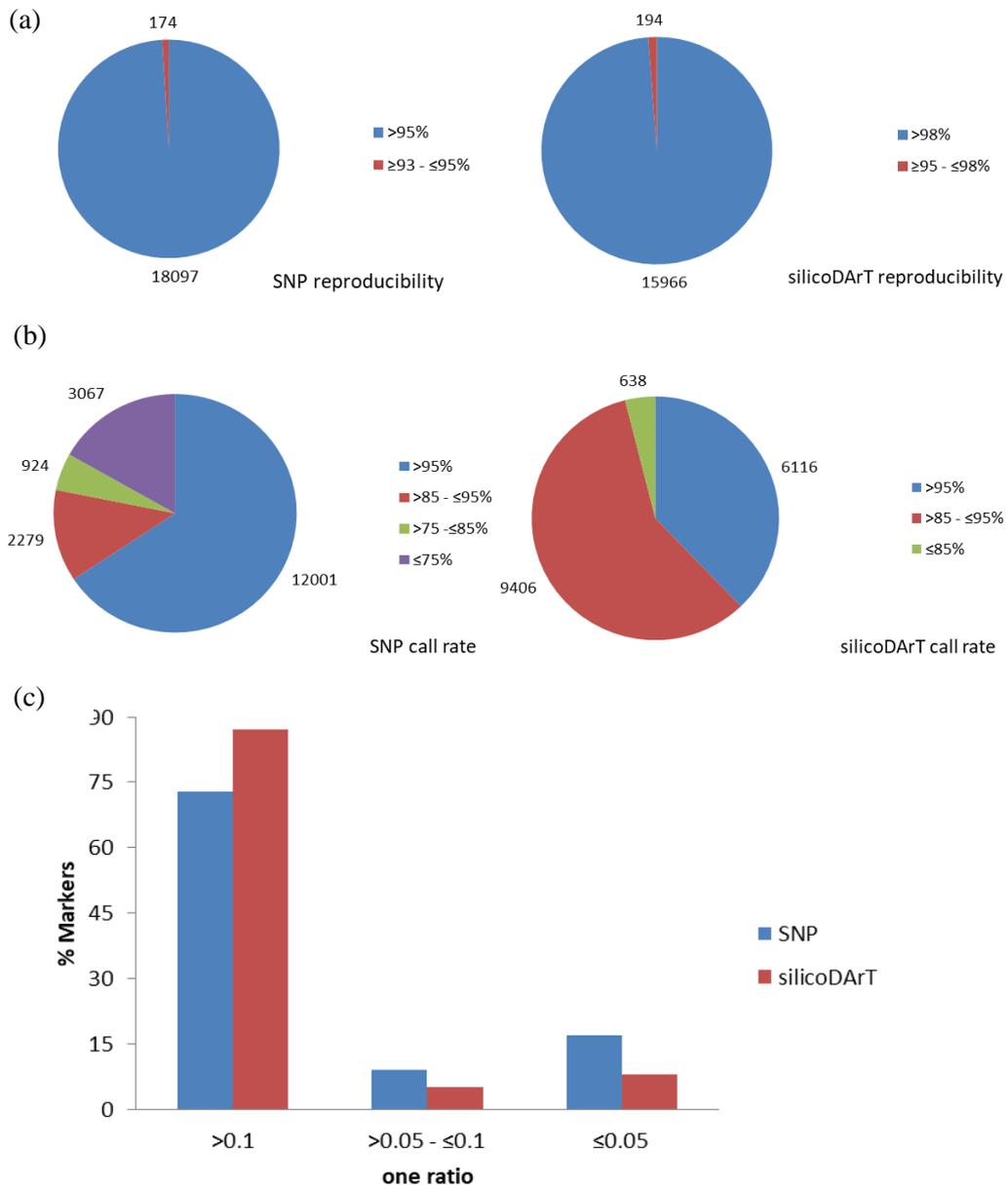
Appendix Figure A1. Layout for Augmented Randomized Complete Block Design



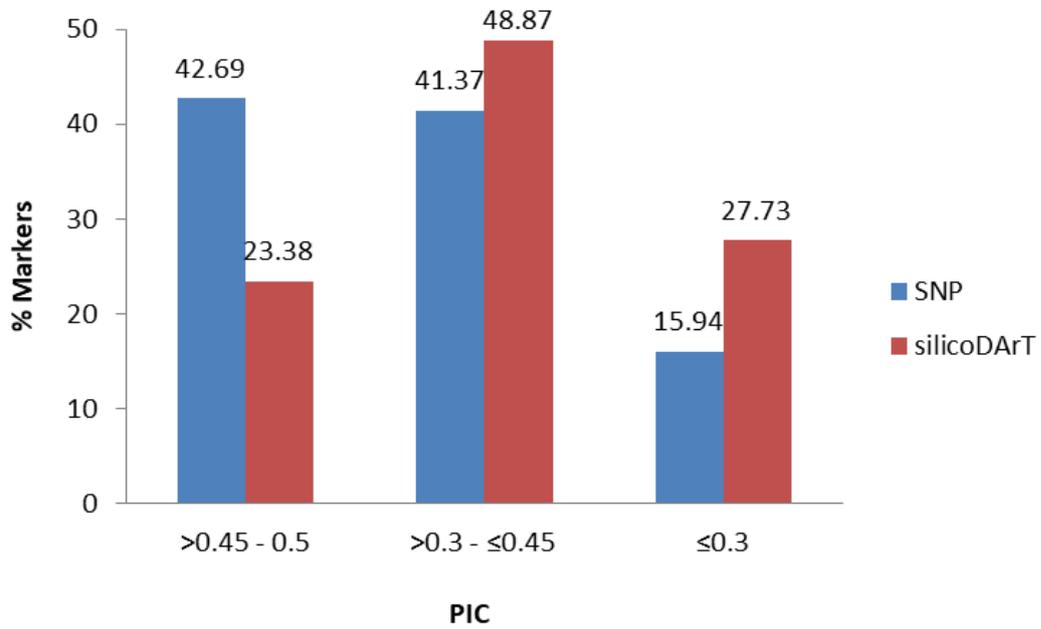
Appendix Figure A2. Field experiment for phenotypic characterization of 117 Myanmar rice genotypes at DAR/IRRI research farm in Myanmar



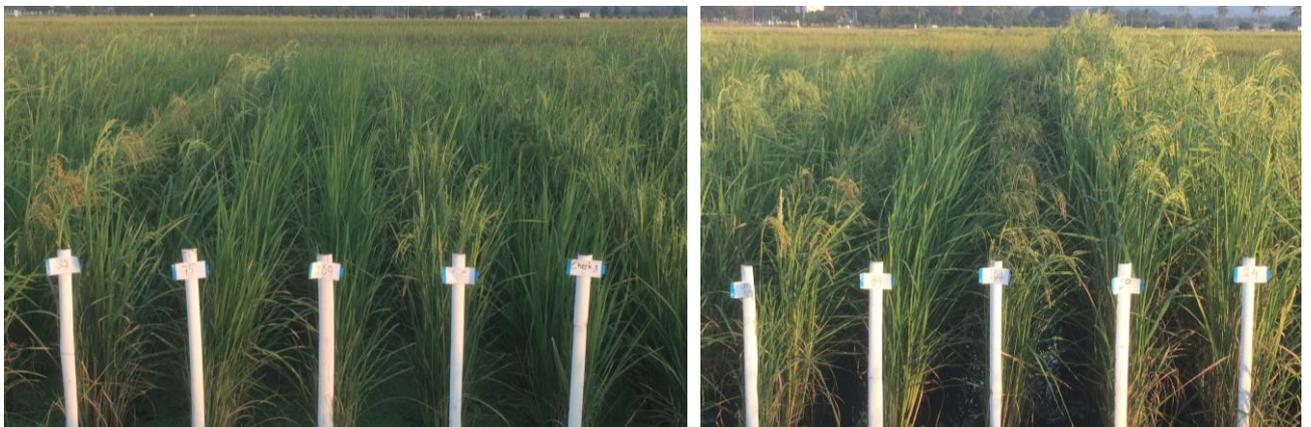
Appendix Figure A3. Qualitative traits: A, flag leaf attitude; B, attitude of panicle branches; C, auricle color; D, color of apiculus; E, stigma color; F, endosperm type



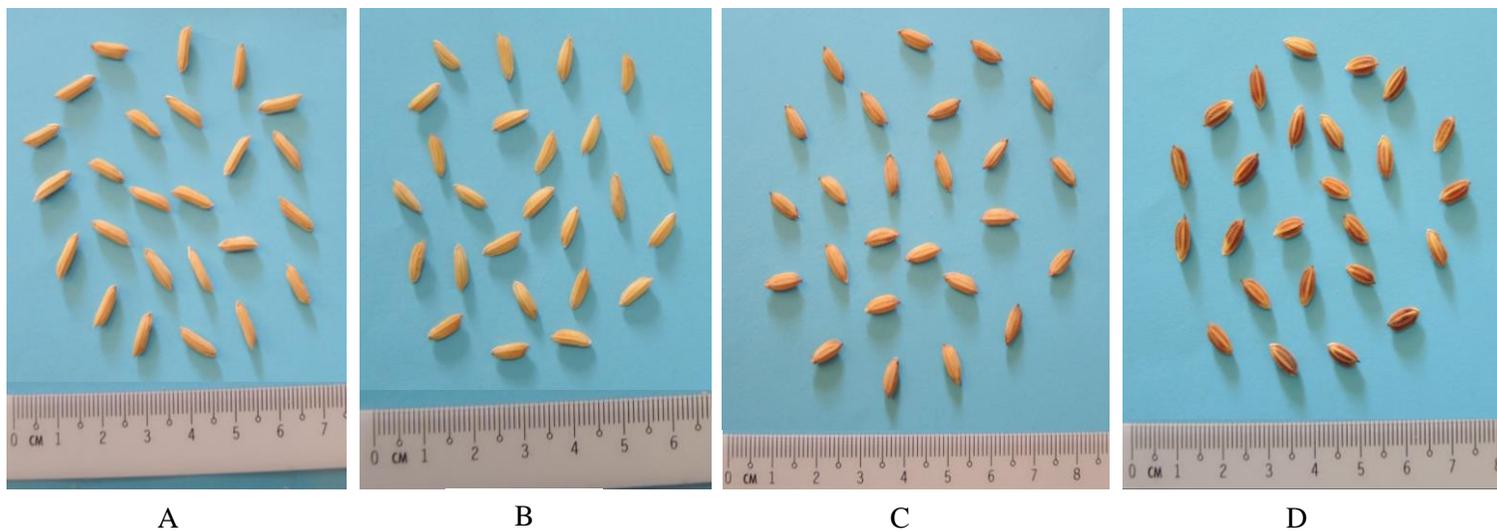
Appendix Figure A4. Distribution of SNP and silicoDArT marker data for several quality parameters: (a) reproducibility, (b) call rate, and (c) one ratio



Appendix Figure A5. Distribution of PIC values of SNP and silicoDArT markers used for genomic studies in rice



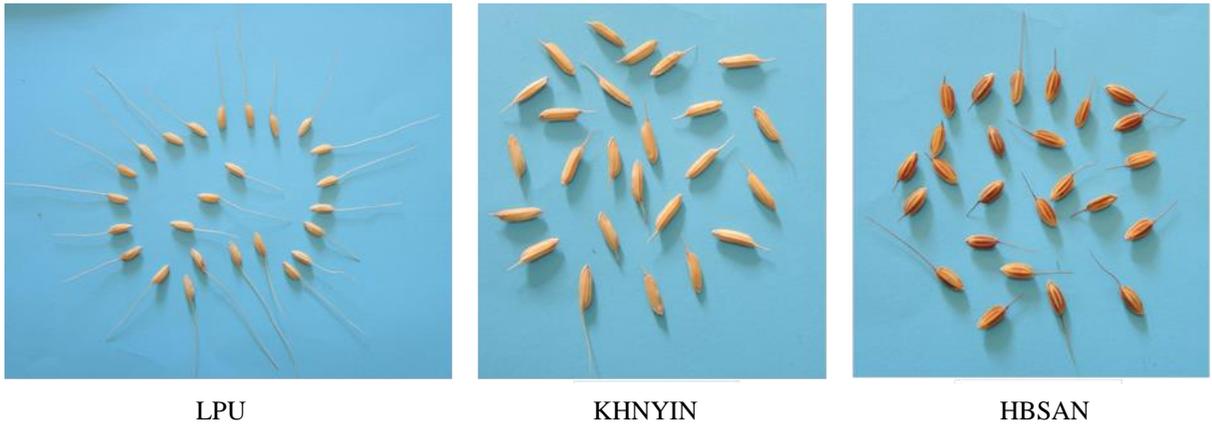
Appendix Figure A6. Diversity in days to heading (DTH) and culm length (CL)



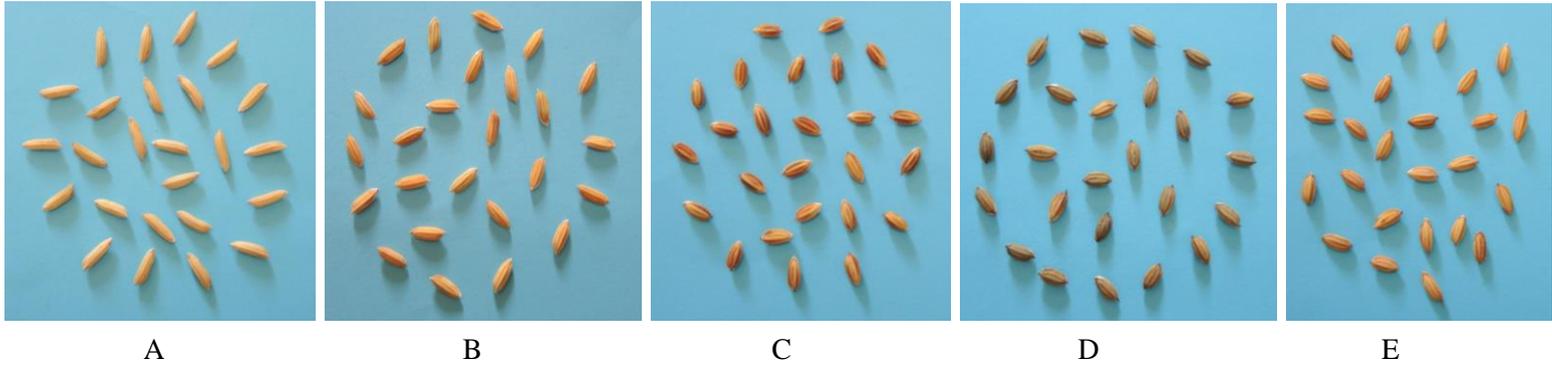
Appendix Figure A7. Type of grains: A, Emata (GL/GW = >3.3mm); B, Latywezin (GL/GW = 2.8–3.3mm); C, Ngasein (GL/GW = 2.4–2.8mm); D, Meedon (GL/GW = 2.0–2.4mm)



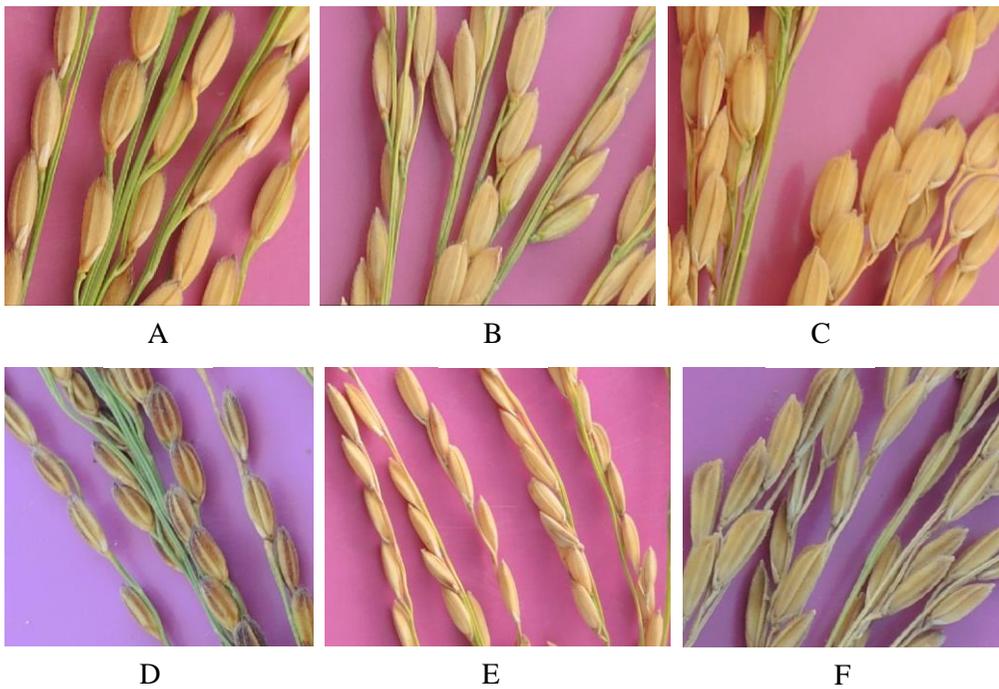
Appendix Figure A8. Genotypes showing pronounced sterile lemma (>2.5 mm but shorter than the lemma)



Appendix Figure A9. Awned varieties with different length



Appendix Figure A10. Grain color (Lemma and palea color): A, straw; B, straw furrows; C, brown furrows; D, black furrows; E, gold and gold furrows



Appendix Figure A11. Lemma and palea pubescence: A, long upper hairs; B, hairs on upper; C, short hairs; D, long hairs (velvety); E, glabrous; F, short upper hairs

Appendix 4. Author's curriculum vitae

CURRICULUM VITAE

Name: Aye Aye Thant, M.Agr.Sc.

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Address: Lathar st., Kyaukpadaung tsp., Nyaung U dist.,
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EDUCATION

Ph.D. in Agriculture in Tropics and Subtropics at Czech University of Life Sciences Prague (2016–2022*anticipated)

M.Agr.Sc. in Field Crop Production (Agronomy) at Yezin Agricultural University, Myanmar (2008–2011)

B.Agr.Sc. in Field Crop Production (Agronomy) at Yezin Agricultural University, Myanmar (2004–2008)

WORK EXPERIENCE

Assistant Scientist – Agronomist at International Rice Research Institute (2012–2015) under Crop and Environmental Sciences Division (CESD)

- Assist in designing experiment/survey based on the research outline provided, in the area of Agriculture
- Manage the implementation of experiments/surveys and other research studies
Consolidate, evaluate and analyze research data
- Contribute to the preparation of reports, publications, and presentations
- Coordinate and prepare training
- Supervise the work of Researchers, Research Technicians and/or contract workers
- Carry out performance appraisals of Researchers/Research Technicians
- Contribute to the professional development of Researchers/Research Technicians

RESEARCH ARTICLES

Wehmeyer H, Malabayabas A, San SS, Thu AM, Tun MS, Thant AA, Connor M. 2022. Rural development and transformation of the rice sector in Myanmar: Introduction of best management practices for sustainable rice agriculture. Outlook on Agriculture 1–15. DOI: 10.1177/00307270221086008.

Thant AA, Zaw H, Kalousova M, Singh RK, Lojka B. 2021. Genetic diversity and population structure of Myanmar rice (*Oryza sativa* L.) varieties using DArTseq-based SNP and SilicoDArT markers. *Plants* **10** (12): 2564. DOI: 10.3390/plants10122564.

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Thant AA, Htun TM, Kalousova M, Singh RK, Lojka B. 2018. Microsatellite markers and their application on genetic diversity studies of rice landraces (*Oryza sativa* L.) in Myanmar - A Review. *International Journal of Environmental and Rural Development* **9** (2): 8–13. DOI: 10.32115/ijerd.9.2_8.

Rahman MA, Thant AA, Win M, Tun MS, Moet Moet P, Thu AM, Win KT, Myint T, Myint O, Tun YT, Labios RV, Casimero MC, Gregorio GB, Johnson DE, Singleton GR and Singh RK. 2015. Participatory Varietal Selection (PVS): A “bottom-up” breeding approach helps rice farmers in the Ayeyarwady delta, Myanmar. *SABRAO Journal of Breeding and Genetics* **47** (3): 299–314.

CONFERENCE CONTRIBUTIONS

Thant AA, Zaw H, Kalousova M, Singh RK, Lojka B. 2020. Genomic studies of Myanmar rice (*Oryza sativa* L.) varieties using DArT and SNP markers. Poster presentation at TROPENTAG on 9–11 Sep 2020, Virtual Conference, ATSAF e.V., Germany

Thant AA, Zaw H, Kalousova M, Aung HH, Singh RK, Lojka B. 2019. Phenotypic diversity of rice landraces collected from Ayeyarwady region, Myanmar using agro-morphological characterization. Poster presentation at TROPENTAG on 18–20 Sep 2019, University of Kassel, Kassel, Germany.

Thant AA, Kalousova M, Singh RK, Lojka B. 2018. On-farm rice diversity and farmers' preferences to varietal attributes in Ayeyarwady delta, Myanmar. Poster presentation at TROPENTAG on 17–19 Sep 2018, Ghent University, Ghent, Belgium.

Thant AA, Htun TM, Kalousova M, Singh RK, Lojka B. 2018. Microsatellite markers and their application on genetic diversity studies of rice landraces (*Oryza sativa* L.) in Myanmar - A review. Poster presentation at The 9th International Conference on Environmental and Rural Development on 24–25 Feb 2018, Yezin Agricultural University, Nay Pyi Taw, Myanmar.

Thant AA, Kalousova M, Than H, Lojka B. 2017. Effects of seed sizes and varieties on growth, yield, and oil and protein contents of groundnut (*Arachis hypogaea* L.). Poster presentation at TROPENTAG on 20–22 Sep 2017, University of Bonn, Bonn, Germany.

Thant AA, Soe MN, MoetMoet P, Thu AM, Casimero MC, Labios RV, Myint TT, Myint O, Rahman MA, Singleton GR, Johnson DE, Singh RK. 2014. Evaluation of four rice varieties

for higher yield in the Ayeyarwaddy delta region of Myanmar. The 4th International Rice Congress (IRC 2014), 27 Oct–1 Nov 2014, Bangkok, Thailand.

EDUCATIONAL TRAININGS

Hands-on training on data management, analysis, and interpretation using different software and weather outputs at International Rice Research Institute on 19-22 Jan 2015 in Yangon, Myanmar.

2nd GIS training course using Quantum GIS at International Rice Research Institute on 7-10 Apr 2014 in Yangon, Myanmar.

Ecological management of rodents, weeds, insects and birds in rice agro-ecosystems - biological and social dimensions in Philippines at International Rice Research Institute on 4-15 Nov 2013 in Los Baños, Philippines.

Hands – on training on quality rice seed production at International Rice Research Institute on 10-12 Oct 2013 in Yangon, Myanmar.

Training on Geospatial techniques using Quantum GIS at International Rice Research Institute on 23-27 Sep 2013 in Yangon, Myanmar.

Field and data management of on-farm participatory varietal selection trials at International Rice Research Institute on 3-4 Apr 2013 in Yangon, Myanmar.

Training on postharvest management for improved quality of rice grain and seed at International Rice Research Institute on 3-5 Nov 2012 in Yangon, Myanmar.

Participatory adaptive research on stress tolerant rice in Ayeyarwaddy Delta, Myanmar at International Rice Research Institute on 18-20 Sep 2012 in Yangon, Myanmar.

COMPUTER SKILLS AND COMPETENCES

Programming: CROPSTAT, SPSS, PBtools, R programming, Genomic data analysis tools (DARwin, Mega, TASSEL, Structure)

Packages: Microsoft Office Word, Excel, Power point, Access (all excellent)

Browsers: Internet Explorer, Mozilla Firefox, Google Chrome (all excellent)