

Genomic studies of Myanmar rice (*Oryza sativa* L.) varieties using DArT and SNP markers

Aye Aye Thant¹, Hein Zaw², Marie Kalousová¹, Rakesh Kumar Singh³, Bohdan Lojka¹

¹ Czech University of Life Sciences Prague, Fac. Tropical AgriSciences, Czech Republic

² Plant Biotechnology Center, Myanmar

³ Crop Diversity and Genetics section, The International Center for BioSaline Agriculture, Dubai



Introduction

- Rice (*Oryza sativa* L.) is the main cereal grain crop of Myanmar and occupies two-thirds of the total area under cultivation.
- Local varieties of rice have evolved from their wild progenitors under both natural and human selection, resulting in a high level of genetic diversity (Huang et al. 2010).
- The identification of genetic diversity in local varieties of rice is important in breeding elite varieties for sustainable agriculture.

Objective

- to characterize genetic diversity, and population structure of 117 diverse Myanmar rice accessions collected from Ayeyarwady delta region using DArT platforms

Results

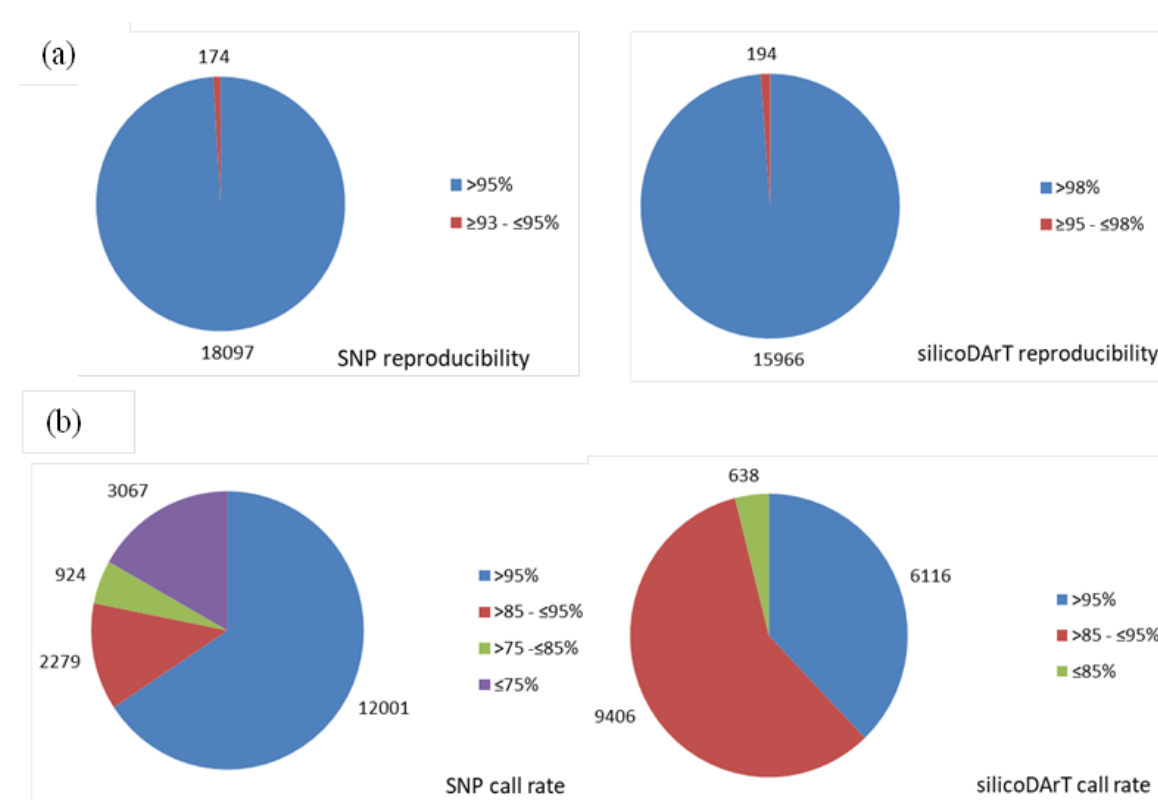


Figure 2. Distribution of SNP and silicoDArT marker data for several quality parameters. (a) Reproducibility. (b) Call Rate.

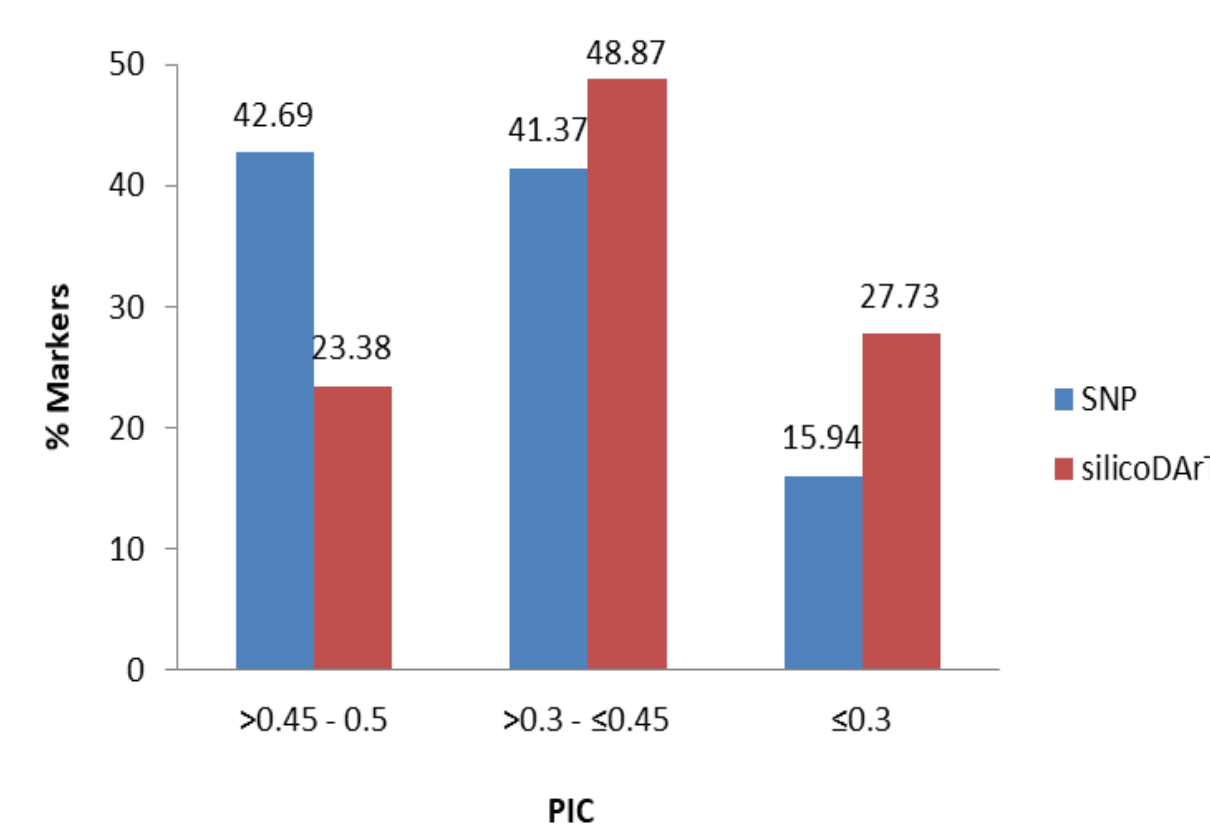


Figure 3. Distribution of PIC values of SNP and silicoDArT markers used for genomic studies in rice

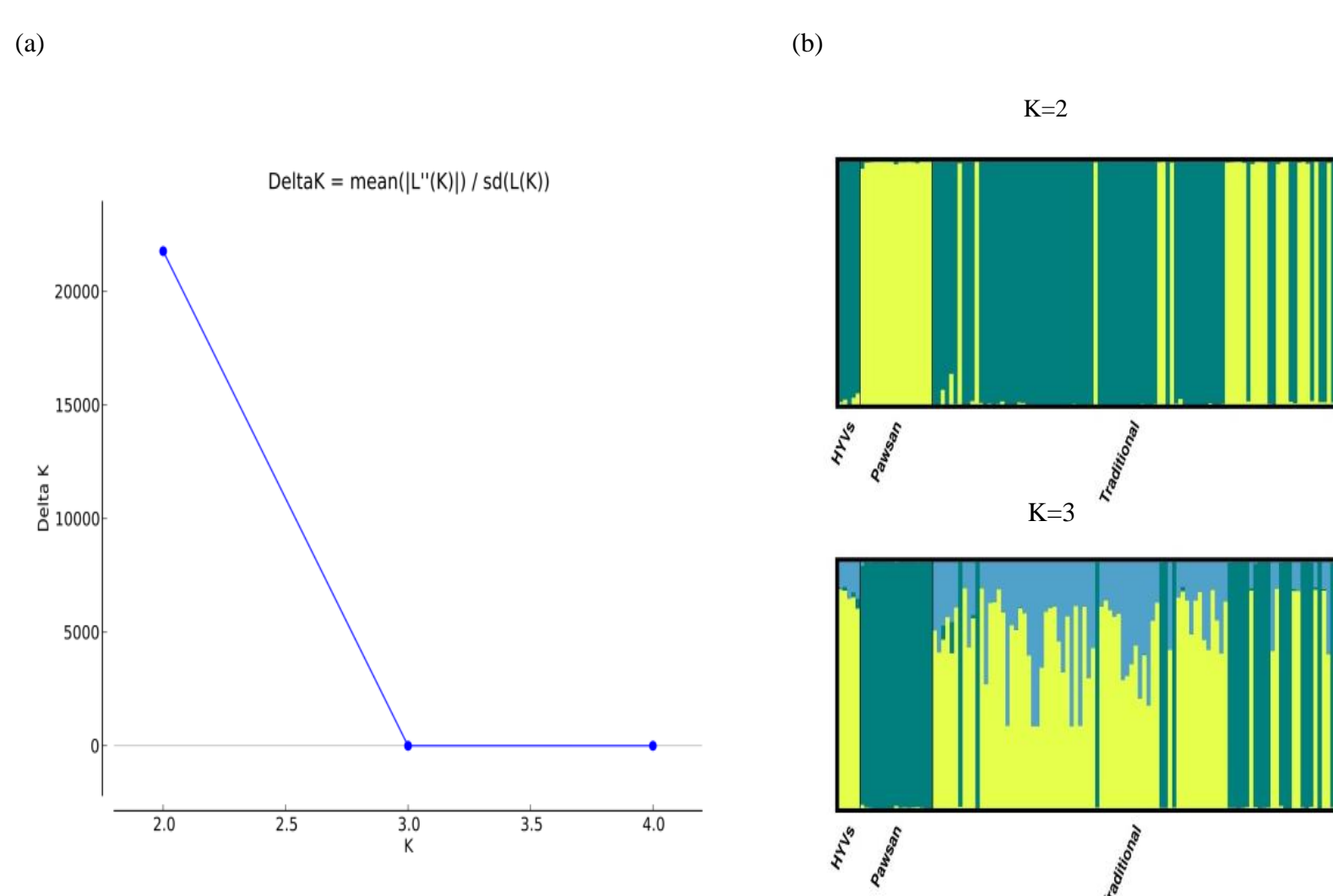


Figure 4. Population structure of 117 Myanmar rice accessions based on 7643 SNPs. a. ΔK values plotted as the number of subpopulations. b. Subpopulations (K=2, and K=3) inferred using STRUCTURE. Traditional, Pawsan, and HYVs are colored differently

Table 1. Analysis of molecular variance (AMOVA) using 7,643 SNPs of the genetic variation among and within two subpopulations of 117 rice accessions

Source	df	SS	MS	Est. Var.	%
Among Pops	1	209102.877	209102.877	1973.534	74%
Among Indiv	115	148509.820	1291.390	585.915	22%
Within Indiv	117	13988.500	119.560	119.560	4%
Total	233	371601.197		2679.009	100%
Fst (Fixation index)		0.737			
Nm (haploid no. of migrants)		0.089			

Table 2. Mean of different genetic parameters including number of samples (N), number of different alleles (Na), number of effective alleles (Ne), Shannon's index (I), observed heterozygosity (Ho), expected heterozygosity (He), unbiased expected heterozygosity (uHe), fixation index (Fst), and percentage of polymorphic loci (PPL) in each of the two subpopulations

Pop	N	Na	Ne	I	Ho	He	uHe	F	PPL
POP1	39.765	1.411	1.146	0.137	0.025	0.087	0.088	0.515	41.07 %
POP2	76.353	1.797	1.358	0.327	0.035	0.214	0.215	0.776	79.73 %
Mean	58.059	1.604	1.252	0.232	0.030	0.150	0.152	0.688	60.40%

References

Huang, Xuehui, Xinghua Wei, Tao Sang, Qiang Zhao, Qi Feng, Yan Zhao, Canyang Li, et al. 2010. "Genome-Wide Association Studies of 14 Agronomic Traits in Rice Landraces." *Nature Genetics* 42 (11): 961–67. doi: 10.1038/ng.695.

Jaccoud, Damian, Kaiman Peng, David Feinstein, and Andrzej Kilian. 2001. Diversity Arrays : A Solid State Technology for Sequence Information Independent Genotyping. *Nucleic Acids Research* 29 (4): 1–7.

Materials and methods

Germplasm and DNA extraction

- 117 rice genotypes from different parts of Ayeyarwady delta region
- 2-3 leaves of three-week old rice plants taken for DNA extraction

Genotyping with DArT (Diversity Array Technology) markers

- 20 μ l of a 100 ng/ μ l DNA of each sample was sent to DArT Pty Ltd, Australia.
- DArT markers were developed by (Jaccoud et al. 2001) to enable whole genome profiling of crops without the need for sequence information.
- DArT has generated two types of markers: i) SNP (co-dominant markers) and ii) silicoDArT markers (dominant and scored for the presence or absence of a single allele).



Figure 1. Rice field growing 117 genotypes

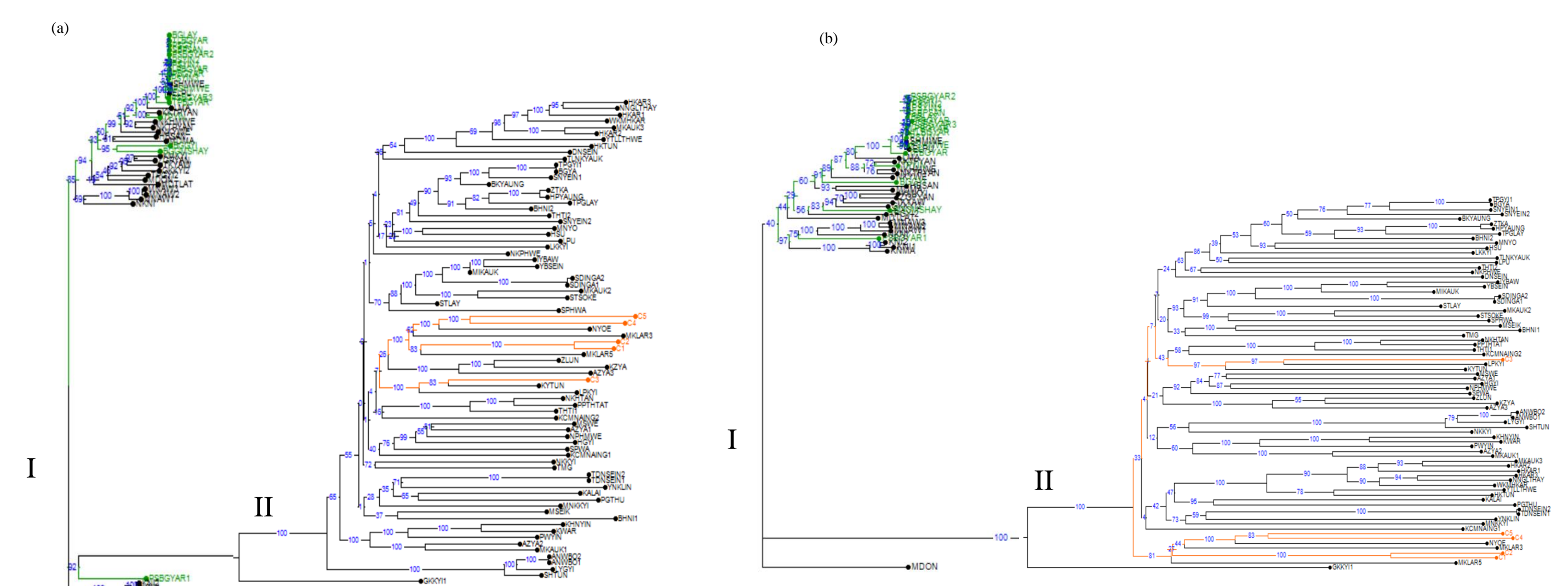


Figure 5. Genetic relationships among 117 rice genotypes. (a) Weighted neighbour-joining dendrogram based on SNP markers. (b) Weighted neighbour-joining dendrogram based on silicoDArT markers.

Conclusions

- 7,643 SNP and 4,064 silicoDArT markers provided robust information of the rice genome in the absence of sequence information.
- Based on our findings, the rice panel was genetically diverse. This level of genetic diversity could be the basis for developing new rice varieties with desirable characteristics while being adapted to diverse environments.
- These findings support researchers to identify useful DNA polymorphisms in genes and germplasm of interest and apply for future allele/gene identification.