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Agro-morphological Characterization of the Wild Rice (*Oryza longistaminata*) Populations From Selected Areas of Tanzania

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Abstract

The study assessed the variations in ten agro-morphological characters among and within the wild rice (*O. longistaminata*) populations from four selected areas, namely Bagamoyo, Kibaha, Kilombero and Mbarali in Tanzania. The study was carried out through measurement of ten quantitative agro-morphological characters, based on morphological descriptors for wild and cultivated rice species developed by Bioversity International and International Rice Research Institute (IRRI). Measurement of morphological characters of *O. longistaminata* plants was done soon after heading. One-way Analysis of Variance (ANOVA) was used to compare the variations in quantitative characters between *O. longistaminata* populations from the four study areas. The results showed existence of high genetic variations in the study areas, both within populations and among populations. In addition, the results showed significant differences between populations for all quantitative agro-morphological characters, except grain length. Moreover, some agro-morphological characters showed positive correlations to each other. The dendogram generated from the agro-morphological data grouped the *O. longistaminata* from the four study areas into four groups irrespective of their geographic origins, implying that there was gene flow between populations. Generally, the four *O. longistaminata* populations assessed exhibited a wide range of variations in morphological characters, both within the population and among populations.

Keywords: Morphological character, *Oryza longistaminata*, Quantitative character, Wild rice.

Introduction

Oryza longistaminata is the only perennial wild rice species in Tanzania. It is also the most widely distributed wild rice species in Tanzania (Katayama 1990). *O. longistaminata* possesses some agronomically important genes which can be used in rice breeding to improve rice quality and productivity (Kiambi et al. 2005). Rice breeders are interested in developing cultivars with improved yields and other desirable agronomic characters (Kamara et al. 2017). Yield or productivity is a complex quantitative character controlled by several genes interacting with the environment and is the product of many factors called *yield components* (Khan et al. 2009). Literature shows that *O. longistaminata* has genes for tolerance to biotic and abiotic stresses (Kiambi et al. 2005). These genes can be introgressed into the cultivated rice for the purpose of improving rice quality and productivity. However, selection and utilization of genetic resources from this wild rice species in rice breeding requires thorough understanding of its genetic diversity (Kiambi et al. 2005), which can be determined through genetic characterization using morphological, molecular or biochemical markers. Information on the genetic diversity is important not only for selection and utilization of genetic resources in breeding but also for effective conservation and management of the species (Lin 1991).

Agro-morphological characterization is the characterization which is based on agronomically important morphological characters, such as culm length or panicle length. According to Crispo (2008), morphological variation in populations can be caused by additive genetic variation or processes of phenotypic plasticity in response to different selective pressures or environmental factors within the habitat. Among the key environmental factors influencing plant's morphological characteristics include the soil physico-chemical properties. Studies show that evaluation of genetic diversity of plant populations based on morphological characteristics does not usually provide accurate estimates of the genetic differences because morphological characters are highly influenced by environmental factors (Nguyen et al. 2004). However, morphology-based characterization of plant populations is quicker, easier and less costly than DNA-based evaluation (Aida et al. 2007). In this study, *O. longistaminata* populations from four districts of Tanzania were characterized based on agronomic or performance-related morphological characters, i.e., based on characters which have direct or indirect effects to the plant productivity or plant's physiological processes, including growth and reproduction. Before this study, there was limited information on the extent of morphological variations among and within *O. longistaminata* populations from the four study areas. This study was therefore intended to characterize the *O. longistaminata* populations from selected areas of Tanzania basing on their quantitative agro-morphological characters.

Materials and Methods

The study areas

The study was conducted in four districts of Tanzania, namely Bagamoyo, Kibaha, Kilombero and Mbarali. In each of these districts, wild rice (*O. longistaminata)* population was sampled in swampy areas within the valleys or flood plains. In some cases, the wild rice and the cultivated rice species seemed to grow sympatrically. Moreover, the four districts (study sites) showed more or less variations in several ecological factors, including altitude, climatic conditions and soil characteristics. In the four study sites, the sampling of *O. longistaminata* was carried out around the following grid points: S 62°8'35.7" and E 38°50'19" (in Bagamoyo), S 60°44'35.1" and E 38°53'49.5" (in Kibaha), S 80°7'29" and E 36° 38'38" (in Kilombero) and S 84°05'0.3" and E 34°17'24.6" (in Mbarali). The locations of the study sites in the four districts are shown in Figure 1.

Sampling method

Agro-morphological characterization of *O. longistaminata* populations involved assessment of variations in morphological characters among and within *O. longistaminata* populations from different sites in the study areas. Morphological data were collected based on descriptors for wild and cultivated rice species developed by the Bioversity International and International Rice Research Institute (IRRI). The study involved assessment of ten selected quantitative morphological characters, namely culm length, flag leaf length, flag leaf width, penultimate leaf length, penultimate leaf width, panicle length, number of primary branches per panicle, number of grains per panicle, grain length and grain width. The study involved measurement of a total of 140 randomly selected *O. longistaminata* samples from the four study areas. Each sample was recorded as an average of 10 replicates (Kioko et al. 2015). All measurements and assessments were done soon after heading. The culm length was measured (using a tape measure) from the ground level (base of the plant) to the base of the panicle and panicle length was measured from the base of the panicle to its tip. Flag leaf was measured from the ligule to the tip of the blade. Penultimate leaf, which is the highest leaf below the flag leaf, was also measured from the ligule to the tip of the blade. The leaf width was measured at the widest portion of the leaf. Grain length was measured as the distance from the base of the lower most glume to the tip of the lemma or palea. The grain width was measured as the distance across the lemma and palea at the widest point. All the measurements were done using either a tape measure or a ruler. Grain number per panicle and number of primary branches per panicle were determined by counting the respective parts.

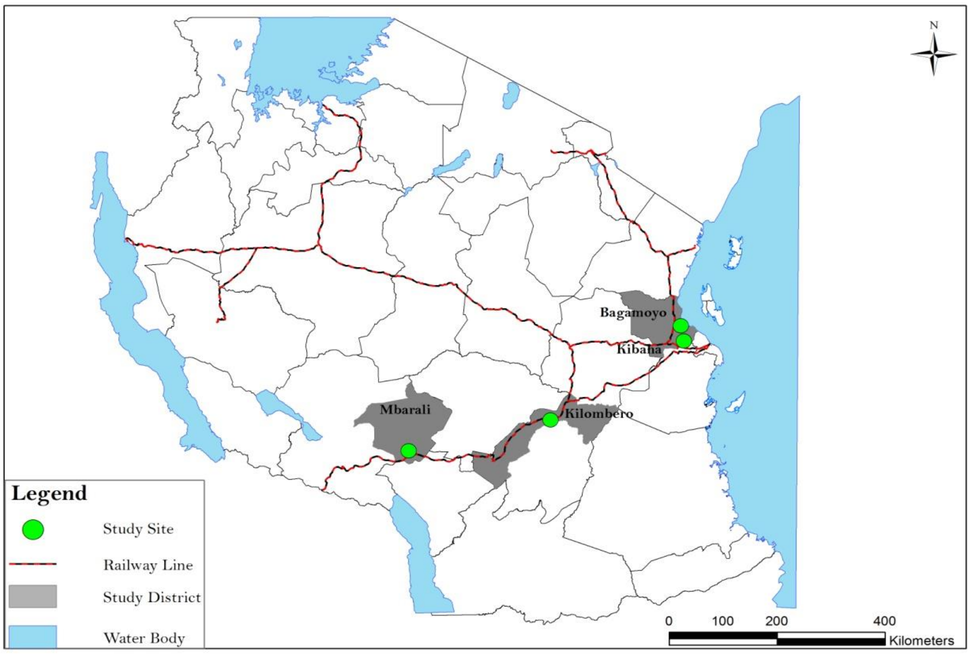

Figure 1: The map of Tanzania showing the location of the study sites/areas.

Data analysis

The morphological data were analysed using the SPSS software package (Saramajeewa et al. 2004). A one-sample Kolmogorov-Smirnov test was used to determine the variations in the quantitative morphological characters among the *O. longistaminata* samples from the four populations. One-way Analysis of Variance (ANOVA) was used to compare the variations in morphological characters among the four populations for all the quantitative data. A correlation analysis was performed to determine the interrelationships among the assessed morphological characters. The UPGMA (Unweighted Pair Group Method with Arithmetic mean) dendogram was also generated to assess clustering of *O. longistaminata* individuals from the four populations based on their genetic distance.

Results

Variation in quantitative morphological characters among and within *Oryza longistaminata* populations

The *O. longistaminata* from the four populations showed a wide range of variations in the quantitative morphological characters. Generally, *O. longistaminata* individuals from Mbarali and Kilombero had relatively high mean values for most of the characters assessed while the individuals from Kibaha had relatively low mean values. Moreover, statistical analysis using one way ANOVA showed significant differences among populations (p ≤ 0.02) in all the assessed quantitative morphological characters except the grain length. The summary results on the variation in quantitative morphological characters between and within the *O. longistaminata* four populations are presented in Table 1.

Correlation among the agro-morphological characters in *Oryza longistaminata*

Most of the agro-morphological characters assessed in the present study showed significant positive correlations ($r \geq 0.01 \leq 0.05$) to each other as shown in Table 2.

Table 1: Variations in quantitative morphological characters within and between *Oryza longistaminata* populations (Mean ± SD) n = 140

Character	Bagamoyo	Kibaha	Kilombero	Mbarali	*F*-value	p-value	Conclusion
Culm length (CL)	146.47 ± 5.61	132.56 ± 4.20	178.11 ± 6.17	155.83 ± 5.5	12.36	0.000	Significant
Penultment leaf length (pLL)	58.59 ± 2.89	46.84 ± 1.55	60.04 ± 2.90	55.25 ± 2.20	5.68	0.001	Significant
Penultment leaf width (pLW)	1.44 ± 0.04	1.25 ± 0.05	1.53 ± 0.06	1.62 ± 0.07	7.66	0.000	Significant
Flag leaf length (fLL)	35.22 ± 1.84	25.65 ± 1.41	31.28 ± 1.24	33.69 ± 1.92	6.52	0.000	Significant
Flag leaf width (fLW)	1.34 ± 0.04	1.21 ± 0.06	1.43 ± 0.06	1.54 ± 0.08	5.79	0.001	Significant
Panicle length (PL)	34.44 ± 0.92	34.8 ± 1.12	38.17 ± 1.12	39.89 ± 1.26	5.85	0.001	Significant
Number of primary branches per panicle (PB)	11.06 ± 0.35	10.71 ± 0.46	13.00 ± 0.56	15.08 ± 0.80	12.40	0.000	Significant
Number of grains per panicle (GN)	115.40 ± 5.69	110.65 ± 6.03	138.06 ± 6.14	160.03 ± 7.40	12.73	0.000	Significant
Grain length (GL)	0.85 ± 0.01	0.85 ± 0.01	0.84 ± 0.01	0.82 ± 0.01	1.73	0.164	Not significant
Grain width (GW)	0.20 ± 0.00	0.20 ± 0.00	0.22 ± 0.00	0.20 ± 0.00	6.95	0.000	Significant

Table 2: Correlations among some agro-morphological characters in *O. longistaminata* accessions from the study areas

	CL	pLL	pLW	fLL	fLW	PL	GN	GL	PB	GW
CL	1									
pLL	0.429**	1								
PLW	0.360**	0.480**	1							
fLL	0.269**	0.695**	0.590**	1						
fLW	0.357**	0.548**	0.772**	0.712**	1					
PL	0.394**	0.394**	0.400**	0.361**	0.327**	1				
GN	0.278**	0.214**	0.299**	0.262**	0.364**	0.354**	1			
GL	-0.044	0.042	-0.027	0.036	-0.031	-0.011	-0.163	1		
PB	0.311**	0.203*	0.313**	0.231**	0.315**	0.494**	0.697**	-0.036	1	
GW	0.233**	0.087	0.063	0.067	0.05	0.044	-0.022	0.082	0.066	1

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

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

Legend: CL = Culm length, pLL = Penultimate leaf length, pLW = Penultimate leaf width, fLL = Flag leaf length, fLW = Flag leaf width, PL = Panicle length, GN = Grain number, GL = Grain length, PB = Number of primary branches, GW = Grain width

Clustering of the *Oryza longistaminata* individuals from the four populations based on quantitative morphological data

The UPGMA dendogram generated from the quantitative morphological data based on genetic distance grouped the *O. longistaminata* individuals from the four populations into four major clusters or groups and several sub-clusters with cophenetic correlation of 0.72. The genetic distance between groups ranged between 0 and 0.35 (0% and 35%). Clustering of *O. longistaminata* individuals was not strictly based on their geographical origins, therefore some individuals from different populations clustered together as shown in Figure 2.

Discussion

Variation in quantitative morphological characters among *Oryza longistaminata* populations

The present study demonstrated existence of high levels of variations in the morphological characters among and within the four *O. longistaminata* populations in the study areas, implying that *O. longistaminata* individuals from the four districts exhibited a wide range of variations, both within and among populations. This seems to be consistent with what was reported in the study by Housman et al. (2002) that terrestrial plants exhibit enormous morphological diversity. According to White (1979), this diversity or variation reflects underlying diversity in total resource acquisition and allocation to root, stem and leaf tissues, as well as diversity in sizes and patterns of accretion of modular units during growth. In the present study statistical analysis showed significant differences in morphological characters between populations ($p < 0.05$) in all the assessed quantitative characters, except grain length ($p = 0.164$). The morphological variation between *O. longistaminata* individuals from different geographical areas is probably the result of their genetic make-up or spatial variations in environmental factors or resources between sites. Thus, the size and shape of an individual plant reflect both a genetically determined developmental programme, as well as effects of the environment on resource acquisition and growth rates (Housman et al. 2002). Rao and Hodgkin (2002) indicated that geographical differences in the distribution of genetic variation are extremely common. Moreover, Zhou et al. (2003) demonstrated that morphological variations among populations increase with geographic distances between the populations, implying that isolation by distance increases genetic differentiation. According to the literature, populations may differ with respect to all aspects of diversity and show variations in the number of alleles, the identity of those alleles and the effects they have on plants' morphological characteristics in a population (Rao and Hodgkin 2002).

According to the results, *O. longistaminata* individuals from Mbarali and Kilombero demonstrated higher values, hence higher performance than those from Bagamoyo and Kibaha. This could be an indication that environmental conditions in Kilombero and Mbarali were more favourable to *O. longistaminata* than the conditions in Bagamoyo and Kibaha. The implication is that there were relatively high morphological similarities (low morphological variations) between Bagamoyo and Kibaha populations, as well as between Kilombero and Mbarali populations. This pattern of variations in the morphological characters among *O. longistaminata* individuals from the four populations could be related to spatial variation in environmental factors, such as variation in climatic conditions and soil physico-chemical characteristics.

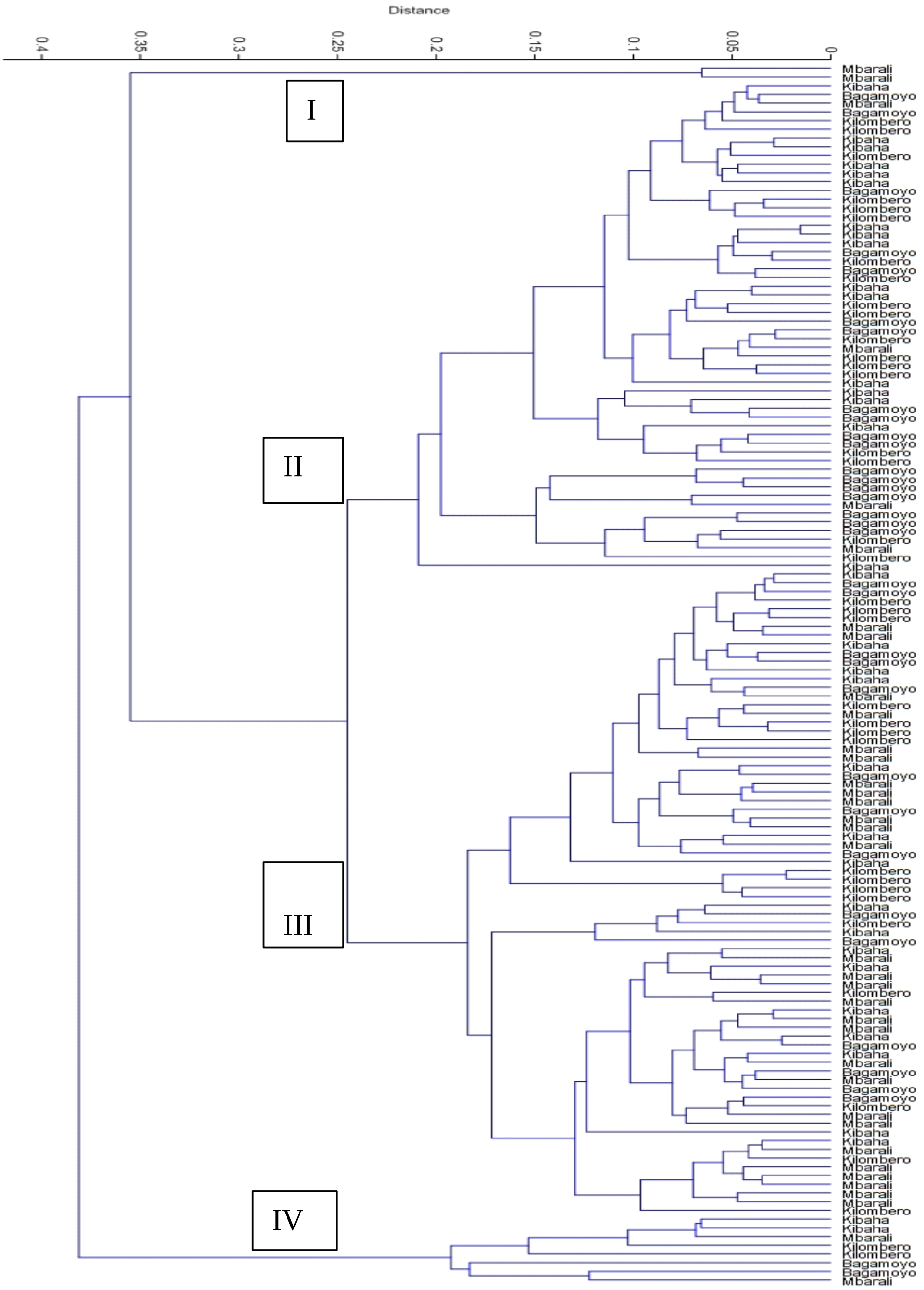

Figure 2: Clustering of *O. longistaminata* individuals from the four populations based on their quantitative morphological characters.

Environmentally-induced phenotypic variation or phenotypic plasticity in plants is often considered to be functional response that maximizes fitness in variable environments (Coleman et al. 1994). Variation in morphological characters exhibited in response to varying resource availability have been used to support the notion that plasticity may be an adaptation to patterns of resources availability in the habitat in which a particular species evolved (Coleman et al. 1994).

Correlation among the agro-morphological characters in *Oryza longistaminata*

The results of this study further indicated that some of the quantitative agro-morphological characters assessed significantly correlated to each other. This finding is similar to the findings of several other studies on cultivated rice (*Oryza sativa*), such as Kole et al. (2008), Jayasudha and Sharma (2010), Basavaraja et al. (2011), Shanthi et al. (2011) and Kohnaki et al. (2013). Knowledge on correlation between morphological characters in agriculturally important plants, such as rice is highly important in selection of phenotypes for breeding purpose. Correlation analysis results can also provide information on the cause and effects relationships between pairs of characters or variables which in turn help to bring improvement of yields (Tandekar et al. 2008). In the light of these results, it is obvious that the characters can be selected simultaneously from *O. longistaminata* populations for improved yield. This is because selection and use of *O. longistaminata* phenotypes or populations with a combination of several performance or yield-related characters such as panicle length, number of grains per panicle in rice breeding programmes would ensure improved rice productivity.

Clustering of the *Oryza longistaminata* individuals from the four populations based on quantitative morphological data

A dendogram generated from the morphological data grouped *O. longistaminata* individuals from the four study sites into four main groups with the cophenetic correlation of 0.72, implying that there was low distortion of clustering of the individuals. The grouping pattern of the *O. longistaminata* individuals from the study area signifies existence of high genetic diversity, among and within the clusters or populations. Genetic variation among individuals from different geographical areas is likely to be caused by variations in environmental factors between sites (Nevo et al. 1991). Literature shows that genetic variation resulting from adaptation to habitats is expected in plant individuals from different habitats or ecological conditions. According to Rao and Hodgkin (2002), adaptive genetic variation, which is usually quantitative, is responsive to even small habitat differences.

The results further indicated that, grouping of *O. longistaminata* individuals was not based on their geographical origins, as individuals from different geographical origins clustered together. Cluster I, which was the smallest of all clusters or groups, consisted of only few individuals all from Mbarali, implying that these individuals were either outliers or individuals with unique genetic characteristics. Cluster II was large and highly diverse, consisting of majority of individuals from Kibaha and Bagamoyo, few individuals from Kilombero and very few from Mbarali. Cluster III, which was the largest and most diverse group consisted of majority of individuals from Kilombero and Mbarali and few individuals from Bagamoyo and Kibaha. Cluster IV, which was relatively small in size consisting of individuals from all the four study sites in almost equal proportions. Grouping together of individuals from different geographical areas is probably the result of gene flow among the populations. On the other hand, the association between individuals from different populations as demonstrated by clustering together of individuals from different geographical locations (study sites) may also be caused by similarity in environmental conditions (Nevo et al. 1991), such as similarity in soil physico-chemical characteristics among the study sites (Nevo et al. 1983). Therefore, phenotypic similarity among individuals from different geographic locations or from different populations may be caused by occurrence of gene flow among the populations or similarity in environmental conditions. If the observed levels of agro-morphological variations among *O. longistaminata* populations from the study area were resulting from gene flow, then it could be concluded that the variations are more genetically controlled than environmentally controlled.

Conclusion

The study revealed that the four *O. longistaminata* populations in the study areas exhibited a wide range of agro-morphological variations, both within and among populations. Moreover, most of the agro-morphological characters assessed showed significant positive correlations between each other, implying that these characters can be selected together. The study further revealed clustering together of individuals from different study sites or populations, which can probably be regarded as an evidence for the occurrence of gene flow between *O. longistaminata* populations. Due to long distances between the four study sites or populations, the said gene flow is probably accomplished through seed transfer by floods, animals or birds or through pollen transfer by agents of pollination such as winds and insects.

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