### Full Length Research Paper

# Assessing phenotypic diversity of interspecific rice varieties using agro-morphological characterization

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Accepted 20 April, 2011

The aim of this study is to identify the phenotypic variability of 60 lowland NERICA varieties and 18 promising lines for better management of their genetic inheritance. The experimental design used was an augmented randomized complete block. Data on 23 agro-morphological traits were collected and analyzed. The analysis of variance showed highly significant differences (P<0.0001) between the variables and revealed the structure of the different genotypes from 11 quantitative discriminate traits. Cluster analysis and principal component analysis were carried out. The entries were grouped into three clusters irrespective of the level of backcrossing of genotypes. Eighty percent of lowland NERICA containing the desired characters adapted to lowland rice cultivation were grouped in cluster 1. The second cluster includes lowland NERICA varieties that can be grown in both upland and lowland ecologies, while the last group comprised only varieties close to their African parent and mainly adapted to the upland. Tillering at 60 days after sowing (DAS), plant height at maturity, maturity and spikelet fertility were the most discriminate and could therefore supplement a wide range of variables for genetic diversity study.

Key words: Lowland NERICA, promising lines, agro-morphological descriptors, cluster.

#### INTRODUCTION

Rice is a major food crop, ranking second to wheat among the most cultivated cereals in the world (Abodolereza and Racionzer, 2009). In 2009, world rice production was evaluated at about 680 million tons with a projected record harvest of 710 million tons in 2010 (FAO, 2010), alongside an increase in consumption of about 8 million tons. If the rains are regular in 2011, rice production in Africa should grow by 3.6% to reach 23.2 million tons (FAO, 2010). In West and Central Africa, the increasing demand from urban and rural populations was estimated at 4% per year (WARDA, 2008). Moreover, rice farming in Africa is mainly done by poor small stakeholders limited by use of input constraints. The development of high performance varieties that combine

By definition, genetic diversity is an inherited variation among and between populations, created, activated and maintained by evolution (Demol, 2001). It is a fundamental characteristic without which breeders are very limited and powerless in plants breeding. The study of genetic diversity reposes on adapted and appropriate techniques. Techniques, such as plant characterization

good adaptation to farmers' cultural practices with good organoleptic characteristics is a crucial step towards food security. Lowlands are also the main ecologies for rice cultivation in Africa accounting for 30% of rice cultivated area (WARDA, 2005). Sixty varieties of New Rice for Africa (NERICA) adapted to lowland ecology and 18 promising lines were therefore developed by the AfricaRice scientists to meet the demand of production and deal with abiotic and biotic stresses related to lowland ecologies. The understanding of the genetic variability of these varieties will provide farmers the opportunities to choose the varieties of their preference.

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have been successfully used in recent years to help in identifying elite individuals. It is an indispensable tool for selecting varieties or lines based on agronomical, morphological, genetic or physiological characters (Ndour, 1998). Characterization is the technique used to evaluate the phenotypic diversity through agromorphological traits (Bajracharya et al., 2006).

Many studies on genetic diversity using agromorphological characterization have been conducted and it led to the identification of the phenotypic variability in rice (Ogunbayo et al., 2005; Bairacharva et al., 2006; Barry et al., 2007). A previous study reported by Semagn et al. (2006) on the genetic diversity of 18 upland NERICA from agro-morphological characterization revealed that the shortest Euclidian distances were obtained between NERICA 4 and 5 (3.61) and NERICA 1 and 2 (2.01), respectively. The farthest remotest distances were recorded with, NERICA 1 and 5 (6.23) using agronomic descriptors, and NERICA 5 and 6 (7.87) using microsatellite markers. For both molecular and agronomic data, NERICA 6 is the least genetically linked to all other varieties belonging to the principal group shown by the cluster analysis (Semagn et al., 2006).

The objective of the present study is to evaluate the agro-morphological traits of 60 lowland NERICA and 18 promising lines in order to contribute to a greater understanding of the diversity between varieties and lines, in contribution to future plant breeding.

#### **MATERIALS AND METHODS**

The experiments were conducted during two consecutive rainy seasons (2008 and 2009) at the AfricaRice Ouedeme site in Lokossa, located at the southern part of Benin (6º42'46"N, 1º41'07"E, altitude 21 m). The average rainfall during the wet seasons (2008 and 2009) were 1044 and 851 mm, respectively (Chinese cooperation, 2009). The soils were black, very clayed and thick, and turned out to be very fertile with a pH of 5.5.

The plant material consisted of 60 lowland NERICA varieties at different backcross levels: 4 BC1, 22 BC2, 19 BC3 and 15 BC4, and 18 promising lines (WAB1 to WAB18). The promising lines were varieties originating from interspecific crosses in the process of nomination at AfricaRice, and they were all BC2 (Table 1). Six control varieties originating from the Asian species O. sativa and 14 parents (African and Asian species) were also used. The 'Augmented Experimental Design' (Federer, 1956) was laid in three blocks. Each block contained 14 parents, 6 control varieties and 26 varieties (lowland NERICA and promising lines). The parents and control varieties were replicated in each block. Direct sowing was carried out, in July 14, 2008 and July 27, 2009, on 5 rows of 5 m length with a spacing of 20 cm × 20 cm and within rows. Recommended cultural practices for the characterization included fertilizer NPK (15-15-15) as basal application at a rate of 200 kg ha-1 during land preparation and urea was applied at the rate of 50 kg ha-1 as top dressing at 14 DAS (first weeding) and at panicle initiation. Agro-morphological data were collected for twenty-three (23) characters at appropriate growth stage of rice plant following the Standard Evaluation System (IRRI, 1996) and descriptors for rice, Bioversity International-IRRI-AfricaRice (2007) (Table 2). Descriptive analysis and ANOVA (using the mixed model) were performed. Cluster analysis, using Euclidean distances within

and between lowland NERICA and promising lines, was calculated and principal component analysis was carried out. A dendrogram was generated using Ward's (1963) minimum variance through XLSTAT (2010) and SAS (2003) software.

#### **RESULTS**

Most of the traits were highly significant (Table 3). A high phenotypic variability (p<0.0001) was observed for leaf length and leaf width, primary branching, maturity and grain thickness. On the other hand, plant height, secondary branching and grain width showed significant differences at the probability of  $\alpha=0.05$  significance level. The analysis of phenotypic diversity was performed using qualitative and quantitative (most discriminating) traits.

#### **Plant vigor**

It was scored between 3 and 5 for all varieties. Moreover, 95% of the lowland NERICA and 85% promising lines grew normally. The growth of control and parent varieties of Asian species varies between 60 and 70% unlike those of the African species for which only 30% of the plants grew normally.

#### Culm habit

There are 5 types of culm habit of which 3 characterize plant materials: erect (15°), semi-erect (20°) and open (40°). All parent varieties were semi-erect and the control varieties were semi-erect (30%) and erect (70%). For lowland NERICA, 20% were erect and 80% were semi-erect. With regard to promising lines, 30% were semi-erect, 60% erect and 10% open.

#### Basal leaf sheath color

Three colors (green, green with purple lines and purple) were observed. Major phenotypic variability was observed in lowland NERICA: green (65%), green color with purple lines (30%) and purple (5%). However, 90% of the promising lines showed green basal leaf sheath color, but only 10% had green color with purple lines. The parental varieties from Africa had either green basal leaf sheath color in the frequency of 50% or green to purple basal leaf sheath color. For the parental varieties from the Asia, no variability was observed; all varieties had green basal leaf sheath color.

#### Flag leaf attitude

High variability was also recorded in flag leaf attitude: erect (1), semi-erect (2), horizontal I (5) and descending

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Table 1. Plant material used including control and parental varieties, lowland NERICA and promising lines.

N°	Designation	Pedigree	Origin	N°	Designation	Pedigree	Origin	N°	Designation	Pedigree	V_BC*	Origin
1	NERICA-L-1	TOG5681/3*IR64	AfricaRice	35	NERICA-L-35	TOG5681/4*IR64	Africa Rice	9	WAB1159-4-3- 2-1-7-2-2	WAB 56- 50/SHAWHON	BC2	AfricaRice
2	NERICA-L-2	TOG5681/3*IR64	AfricaRice	36	NERICA-L-36	TOG5681/4*IR64	Africa Rice	10	WAB1159-2- 12-11-2-1-2-1	WAB 56- 50/SHAWHON	BC2	AfricaRice
3	NERICA-L-3	TOG5681/3*IR64	AfricaRice	37	NERICA-L-37	TOG5681/4*IR64	Africa Rice	11	WAB1159-2- 12-11-2-5-2-2	WAB 56- 50/SHAWHON	BC2	AfricaRice
4	NERICA-L-4	TOG5681/3*IR64	AfricaRice	38	NERICA-L-38	TOG5681/4*IR64	Africa Rice	12	WAB1159-2- 12-11-2-7-2-1	WAB 56- 50/SHAWHON	BC2	AfricaRice
5	NERICA-L-5	TOG5681/3*IR64	AfricaRice	39	NERICA-L-39	TOG5681/4*IR64	Africa Rice	13	WAB1159-2- 12-11-2-10-2-1	WAB 56- 50/SHAWHON	BC2	AfricaRice
6	NERICA-L-6	TOG5681/3*IR64	AfricaRice	40	NERICA-L-40	TOG5681/4*IR64	Africa Rice	14	WAB1159-2- 12-11-2-10-2-2	WAB 56- 50/SHAWHON	BC2	AfricaRice
7	NERICA-L-7	TOG5681/3*IR64	AfricaRice	41	NERICA-L-41	TOG5681/4*IR64	Africa Rice	15	WAB1159-2- 12-11-6-7-1-2	WAB 56- 50/SHAWHON	BC2	AfricaRice
8	NERICA-L-8	TOG5681/3*IR64	AfricaRice	42	NERICA-L-42	TOG5681/4*IR64	Africa Rice	16	WAB1031-3-3- 13-4-3-2-2	WITA 7/CG 20	BC2	AfricaRice
9	NERICA-L-9	TOG5681/3*IR64	AfricaRice	43	NERICA-L-43	TOG 5674/4*IR 31785-58-1-2-3-3	Africa Rice	17	WAB1039-1-3- 1-1-2-2-2	WITA 2/IG 10	BC2	AfricaRice
10	NERICA-L-10	TOG5681/3*IR64	AfricaRice	44	NERICA-L-44	TOG5681/5*IR64	Africa Rice	18	WAB1039-1-1- 1-1-5-2-1	WITA 2/IG 10	BC2	AfricaRice
11	NERICA-L-11	TOG5681/3*IR64	AfricaRice	45	NERICA-L-45	TOG5681/5*IR64	Africa Rice	1	FKR 19	MAHSURI/IET 1444	Oryza sativa (Indica)	IITA- Ibadan
12	NERICA-L-12	TOG5681/3*IR64	AfricaRice	46	NERICA-L-46	TOG5681/5*IR64	Africa Rice	2	FKR 54	IR 12979- 241	Oryza sativa (Indica)	-

Table 1. Cont.

13	NERICA-L-13	TOG5681/3*IR64	AfricaRice	47	NERICA-L-47	TOG 5675/4*I	R 28	Africa Rice	3	WITA 3	11975 / IR 13146-45-2-3	Oryza sativa (Indica)	Côte d'Ivoire
14	NERICA-L-14	TOG5681/3*IR64	AfricaRice	48	NERICA-L-48	IR 6- 5681/4*IR 64		Africa Rice	4	WITA 4	11975 / IR 13146-45-2-3	Oryza sativa (Indica)	Côte d'Ivoire
15	NERICA-L-15	TOG5681/3*IR64	AfricaRice	49	NERICA-L-49	TOG5681/3*IF	R64	Africa Rice	5	WITA 9	IR 2042-178-1 / CT 19	Oryza sativa (Indica)	Côte d'Ivoire
16	NERICA-L-16	TOG5681/3*IR64	AfricaRice	50	NERICA-L-50	IR 6- 5681/4*IR 64	4//TOG	Africa Rice	6	WITA 12	ITA 235/IR 9828-91-2-3 // CT 19	Oryza sativa (Indica)	Côte d'Ivoire
17	NERICA-L-17	TOG5681/3*IR64	AfricaRice	51	NERICA-L-51	IR 6- 5681/4*IR 64	4//TOG	Africa Rice	7	CG 20	-	Oryza glaberrima	Sénegal
18	NERICA-L-18	TOG5681/3*IR64	AfricaRice	52	NERICA-L-52	IR 6- 5681/4*IR 64	4//TOG	Africa Rice	8	IG 10	-	Oryza glaberrima	Côte d'Ivoire
19	NERICA-L-19	TOG5681/3*IR64	AfricaRice	53	NERICA-L-53	IR 6- 5681/4*IR 64	4//TOG	Africa Rice	9	TOG 5674	-	Oryza glaberrima	Nigeria
20	NERICA-L-20	TOG5681/3*IR64	AfricaRice	54	NERICA-L-54	IR 6- 5681/4*IR 64	4//TOG	Africa Rice	10	TOG 5675	-	Oryza glaberrima	Nigeria
21	NERICA-L-21	TOG5681/3*IR1529 -680-3-2	AfricaRice	55	NERICA-L-55	IR 6- 5681/4*IR 64	4//TOG	Africa Rice	11	TOG 5681	-	Oryza glaberrima	Nigeria
22	NERICA-L-22	TOG 5681 / 2*IR 64 //IR 31785-58-1-2-3- 3	AfricaRice	56	NERICA-L-56	IR 6- 5681/4*IR 64	64//TOG	Africa Rice	12	SHAWHON	-	Oryza glaberrima	Liberia
23	NERICA-L-23	TOG 5681/2*IR 64//IR31851-96-2-3- 2-1	AfricaRice	57	NERICA-L-57	IR 6- 5681/4*IR 64	64//TOG	Africa Rice	13	IR 1529-680-3	IR 305-3-17-1-3 / IR 661-1-140- 3	Oryza sativa (Indica)	Burkina Faso
24	NERICA-L-24	TOG 5681/2*IR 64//IR31851-96-2-3- 2-1	AfricaRice	58	NERICA-L-58	IR 6- 5681/4*IR 64	64//TOG	Africa Rice	14	IR 28	IR 5853-162-1- 2-3/IR 7963-30- 2 // IR 9828-36- 3	Oryza sativa (Indica)	IRRI

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Table 1. Cont.

25	NERICA-L-25	TOG 5681/2*IR 64//IR31851-96-2-3- 2-1	AfricaRice	59	NERICA-L-59	IR 31785-58-1- 3/TOG 5674/IR	-2-3-	Africa Rice	15	IR 31851-96-2- 3-2-1	IR 17491-5-4-3-3/IR 2415-90-4-3 // IR 9129-209-2-2-2-1	Oryza sativa (Indica)	IRRI
26	NERICA-L-26	TOG5681/4*IR64	AfricaRice	60	NERICA-L-60	IR 64// 5681/4*IR 64	ГОG	Africa Rice	16	IR 31785-58-1- 2-3-3	-	Oryza sativa (Indica)	IRRI
27	NERICA-L-27	TOG5681/4*IR64	AfricaRice	1	WAB1159-2-12- 11-5-1-1-1	WAB 50/SHAWHON	56-	Africa Rice	17	IR 64	IR 5657-33-2-1 / IR 2061-465- 1-5-5	Oryza sativa (Indica)	IRRI
28	NERICA-L-28	TOG5681/4*IR64	AfricaRice	2	WAB1159-2-12- 11-5-1-2-2	WAB 50/SHAWHON	56-	Africa Rice	18	WAB 56-50	-	Oryza sativa (Japonica)	Côte d'Ivoire
29	NERICA-L-29	TOG5681/4*IR64	AfricaRice	3	WAB1159-4-7- 12-3-5-1-2	WAB 50/SHAWHON	56-	Africa Rice	19	WITA 2	IR 42 / SUAKOKO 8	Oryza sativa (Indica)	Côte d'Ivoire
30	NERICA-L-30	TOG5681/4*IR64	AfricaRice	4	WAB1159-2-12- 11-7-2-1-1	WAB 50/SHAWHON	56-	Africa Rice	20	WITA 7	TOX 891-212- 1-201-1-105 / TOX 3056-5-1	Oryza sativa (Indica)	Côte d'Ivoire
31	NERICA-L-31	TOG5681/4*IR64	AfricaRice	5	WAB1159-2-12- 11-7-4-1-1	WAB 50/SHAWHON	56-	Africa Rice					
32	NERICA-L-32	TOG5681/4*IR64	AfricaRice	6	WAB1159-2-14- 1-2-2-1-1	WAB 50/SHAWHON	56-	Africa Rice					
33	NERICA-L-33	TOG5681/4*IR64	AfricaRice	7	WAB1159-4-3-2- 1-1-2-2	WAB 50/SHAWHON	56-	Africa Rice					
34	NERICA-L-34	TOG5681/4*IR64	AfricaRice	8	WAB1159-4-3-2- 1-7-1-2	WAB 50/SHAWHON	56-	Africa Rice					

V\_BC\*; Variety and level of backcross.

Table 2. List of characters studied.

N°	Characters	Abbreviation
1	Plant vigor	Pv
2	Basal leaf sheath coloration	Bsc
3	Culm habit	Ch
4	Flag leaf attitude	Fla
5	Panicle exsertion	P_ex
6	Number of tillers at 15DAS	T15
7	Number of tillers at 60DAS	T60
8	Plant height at maturity	Hmat
9	50 percent of heading	Hd
10	Maturity	M
11	Leaf length (mm)	LL
12	Leaf width (mm)	LW
13	Number of panicles per plant	Npl
14	Panicle length (cm)	PL
15	Grain length (mm)	GL
16	Grain width (mm)	Gw
17	Grain thickness (mm)	Gth
18	Panicle primary branching	Ppb
19	Panicle secondary branching	Psb
20	Spikelet fertility (%)	S_fert
21	Spikelet sterility (%)	S_ster
22	1000 grain weight (g)	Gw
23	Grain yield	Gyld

Table 3. Significance of 18 quantitative traits.

Traits	Probability level
Tillering at 15 days after sowing	0.4465 <sup>ns</sup>
Tillering at 60 days after sowing	0.0674 <sup>ns</sup>
Leaf length	<0.0001***
Leaf width	<0.0001***
Plant height at maturity	0.0129 <sup>*</sup>
Panicle primary branching	<0.0001***
Panicle secondary branching	0.0037*
Number of panicles per plant	0.6735 <sup>ns</sup>
Grain length	0.135 <sup>ns</sup>
50 percent of heading	<0.0001***
Maturity	<0.0001***
Grain yield	0.1832 <sup>ns</sup>
Panicle length	0.0655 <sup>ns</sup>
Grain width	0.0341*
Grain thickness	<0.0001***
1000 grain weight	0.6094 <sup>ns</sup>
Spikelet sterility	0.3568 <sup>ns</sup>
Spikelet fertility	0.3568 <sup>ns</sup>

<sup>\*\*\*:</sup> significance (P<0.0001); \*\* and \*: significance (0.001< P< 0.05) and ns: not significance P> 0.05.

(7). Lowland NERICA were erect (40%), inter-mediate (45%) and horizontal (15%). Distribution among promising lines was different, in that 65% were erect and 20% were horizontal. For all parent (*O. sativa* and *O. glaberrima*) and control varieties, the percent obtained variedbetween erect (50%) and horizontal (50%).

#### Tillering at 60 DAS (T60)

The number of tillers ranged from 5 to 16 for lowland NERICA and 4 to 14 for the promising lines. The means recorded with the parents of *O. glaberrima* and *O. sativa* varied between 9 and 11 tillers. There was a poor variability of tillering ability at 60 DAS for all groups, and the standard deviation was between 0.32 and 0.49 for lowland NERICA and promising lines. More than 85% of lowland NERICA showed high tillering ability, except for N-L1, N-L15, N-L17, N-L22 and N-L55. For the promising lines, 44.44% varieties showed high tillering with the exception of WAB16, WAB18, WAB17, WAB15, WAB4, WAB11, WAB14, WAB6 and WAB3.

#### Plant height at maturity (Hmat)

The recorded mean for lowland NERICA was 91.8 cm, with a minimum height of 65.24 cm and a maximum of 127.23 cm. For the promising lines, the mean height was 119.64 cm. In a descending order, the means recorded for the parents were: 131.68 (*O. glaberrima*) and 95.7 cm(*O. sativa*), whereas the mean recorded for the controlvarieties was 104.5 cm. Most of the lowland NERICA (87.27%) were semi-dwarf (110 cm) except for N-L15, N-L16, N-L17, N-L26, N-L39, N-L44 and N-L57. However, 83.33% of the promising lines' plants were tall (130 cm), except for WAB13, WAB14 and WAB15 (semi-dwarf).

#### Leaf length and width (LL and LW)

Leaf width varied between 9 and 19 mm with a means of 11 mm for lowland NERICA, while 11.9, 20.34 and 15.4 mm means were recorded with promising lines. The minimum, maximum and means for *O. glaberrima*, *O. sativa* and control varieties were 15.15, 12 and 11.8 mm, respectively. The minimum and maximum recorded for LL were 25.7 and 55.2 mm for lowland NERICA and were 31.5 and 65.8 mm for promising lines. The means LW of 37.2, 50, 50.87, 32.58 and 40.86 mm were obtained from lowland NERICA, promising lines, *O. glaberrima* and *O. sativa* parents, and control varieties, respectively.

#### Grain length (LG)

The minimum and maximum obtained with lowland

NERICA were 8.9 and 10.6 mm, while the values of 8.6 and 10.1 mm were recorded for promising lines. The means recorded with *O. glaberrima*, *O. sativa* species and the control varieties were 8.49, 9.43 and 9.21 mm, respectively.

## Panicle primary and secondary branching (Ppb and Psb)

For lowland NERICA, 6.5 and 11.46 were recorded as minimum values and 12.58 and 35.72 as maximum values. Panicles of promising lines were, for the majority, dense and compact, whereas those of lowland NERICA were dense, rarely open and compact. The means obtained were 11.2, 23.2 and 27.8 for *O. glaberrima, O. sativa* and the control varieties, respectively. Regarding Ppb, 23.63% of the lowland NERICA were below the average. These included N-L2, N-L8, N-L9, N-L10, N-L11, N-L13, N-L18, N-L32, N-L38, N-L45, N-L48 and N-L58.

#### Spikelet fertility (S\_fert)

The percentage of fertility ranged from 70.58 to 98.02%, with a mean of 88.36% for all lowland NERICA. For promising lines, the means recorded was 81.98% with minimal and maximal values varying between 65.72 and 91.80%. Mean obtained from the parents were 82.8 (*O. glaberrima*) and 88% (*O. sativa*). The control varieties had a mean of 87.5%. A total of 90.9% of all plant materials recorded a percentage of fertility above 80% with the exception of N-L7, N-L14, N-L16, N-L52 and N-L53.

#### Number of panicles per plant (Npl)

The means for panicles per plant was 9.7 for lowland NERICA, 6 for promising lines and 11.3, 11.8 and 8 for *O. glaberrima*, *O. sativa* and control varieties, respectively.

#### Maturity (M)

(M) means was 116 days. Minimum and maximum values for lowland NERICA were 96 and 127 days, and were 101 and 136 days for promising lines. Means recorded for *O. sativa*, *O. glaberrima* parents and control varieties were 110, 110 and 116 days, respectively. There was a poor variability (standard deviation<5) for this trait. However, two distinctive groups were found in lowland NERICA: 20% of varieties had a short cycle (M<100 days) and 80% had an intermediate cycle (110  $\leq$  M  $\leq$  130 days). On the other hand, promising lines had an intermediate cycle.

**Table 4.** Pearson correlation coefficients (n) of 11 quantitative traits.

Traits	М	GW	Gth	LL	LW	T60	S_fert	Hmat	Ppb	Psb	Npl
M	1										
GW	0.000	1									
Gth	-0.096	-0.187	1								
LL	0.517	0.188	-0.071	1							
LW	0.357	0.319	0.042	0.682	1						
T60	-0.210	-0.178	0.163	-0.458	-0.425	1					
S_fert	-0.418	0.019	0.043	-0.545	-0.373	0.253	1				
Hmat	0.272	0.425	-0.032	0.767	0.629	-0.410	-0.388	1			
Ppb	0.447	0.205	0.032	0.646	0.650	-0.485	-0.464	0.677	1		
Psb	0.264	-0.214	0.174	0.445	0.326	-0.302	-0.292	0.261	0.608	1	
Npl	-0.175	-0.060	0.026	-0.311	-0.426	0.581	0.192	-0.284	-0.593	-0.464	1

Bold values are different from 0 at a significance level of alpha = 0.05.

**Table 5.** Principal component analysis showing the contribution of each trait among the characteristics of 98 genotypes.

Traits	Prin 1	Prin 2	Prin 3	
M	0.548	-0.112	0.506	
GW	0.263	0.805	-0.148	
Gth	-0.047	-0.515	-0.018	
LL	0.860	0.073	0.267	
LW	0.794	0.147	-0.007	
T60	-0.644	-0.080	0.445	
S_fert	-0.595	0.134	-0.479	
Hmat	0.784	0.325	0.082	
Ppb	0.885	-0.110	-0.087	
Psb	0.584	-0.605	-0.142	
Npl	-0.614	0.210	0.597	
Eigen value	4.626	1.505	1.169	
% variance	42.056	13.686	10.623	
Cumulative % variance	42.056	55.742	66.365	

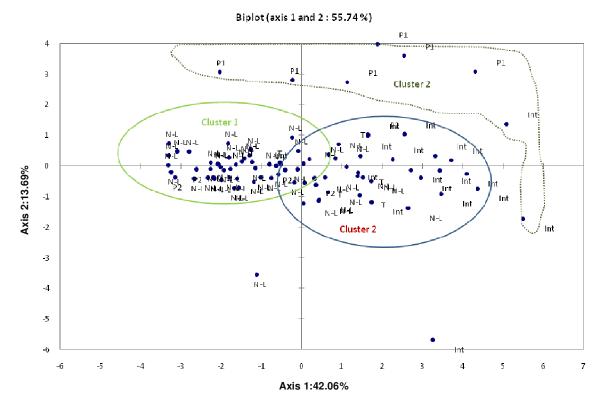
#### Grain yield (Gyld)

Potential yields vary between 3008.31 and 7756.74 kg/ha for lowland NERICA and 2702.38 and 6428.20 kg/ha for the promising lines. More than 50% of lowland NERICA and promising lines had potentially high yields (above the mean of control varieties ≈ 5415 kg/ha).

Varietal structuring was conducted on 11 significant quantitative variables selected during the first season. Table 4 shows a positive and strong correlation (0.77) between the height at maturity and leaf length. Furthermore, another high correlation (0.63) was obtained between the height at maturity and leaf width. Each of these traits was negatively correlated with spikelet fertility (-0.55). Positive correlations between T60 and Npl (0.58) and between primary and secondary

panicle branching (0.61) were recorded. These correlations showed that tall varieties had long and wide leaves. These varieties had an average number of tillers at 60 DAS and the number of panicles per plant was high. They had an intermediate but hardly late Maturity, with profuse primary and secondary branching.

The first 2 axes were retained for principal component analysis (PCA) because they express better total variability of the plant material (55.74%). The contributions of each discriminating trait are listed in Table 5. Axis 1 (42.06%) is defined by Hmat, S\_fert, Ppb, LL and LW, T60, M and Npl traits. These variables express the vegetative and reproductive characters of the varieties. This axis contrasts varieties with long, wide leaves, abundant primary branching with dense panicles to those having short leaves and very dense and compact panicles. It also contrasts varieties with high



**Figure 1.** Agro-morphological diversity from the principal component analysis of the 11 quantitative traits and 98 genotypes.

plant height at maturity and poor tillering at 60 DAS compared to semi-dwarf varieties at maturity with good tillering ability and small number of panicles per plant. Axis 2 (13.69%) explains the yield components: Psb, GL and GW. It opposes varieties with very dense panicles bearing long and round grains to those with less dense panicles bearing round and intermediate-size grains. Figure 1 shows the distribution of the 98 genotypes on both axes of the PCA.

Cluster analysis based on Euclidian distances between the 98 genotypes enabled the identification of three major phenotypic clusters (Figure 2). Cluster 1 was composed of 83.63% lowland NERICA and 26.66% promising lines. Genotypes in this cluster were morphologically similar to their Asian parents (IR64 and IR31785-1-2-3-3). They had the characteristics of the *Indica* group of the Asian species. A strong variability was observed within Cluster 1 which was divided into three sub-clusters when truncated between 0 and 1 250 of distance. Cluster 2 comprised 33.33% of promising lines and 16.36% of lowland NERICA. Lowland NERICA in this cluster were morphologically similar to their African parents (TOG5681 and TOG5674), while promising lines were similar to their Asian parents of Japonica group WAB56-50. As in cluster 1, phenotypic variability at a low level was observed in cluster 2. Three sub-clusters were also identified according to their level of affinity.

There was no lowland NERICA in cluster 3, but 45.45% of the promising lines were found here. These varieties were agronomically and morphologically similar to the African species IG10 and SHAWHON (parents of promising lines) and TOG 5675 (NL-47). However, there was no Asian species in cluster 3.

Eleven significant quantitative traits were chosen based on multivariate analyses carried out during the first season. The variance analyses carried out on these traits during rainy seasons 1 and 2 showed significant differences among them. Variables T60, Hmat, Fert\_ep, Npl and SMC did not show variation during both seasons. On the other hand, significant variation was recorded for LL and LW, Ppb and Psb and G\_L and GW. The variance analysis performed on G × E interaction confirmed the absence of phenotypic variability of these traits (Table 6).

#### DISCUSSION

Qualitative and quantitative agronomic and morphological traits were examined as recommended by Arraudeau (1975) and Jacquot and Arnaud (1979). The most discriminating quantitative traits were T60, Hmat, LL, LW, GW, Ppb and Psb. Similar results were reported by Sié (1991) for LL, LW, GL and GW as discriminating traits through a study based on genetic evaluation of traditional

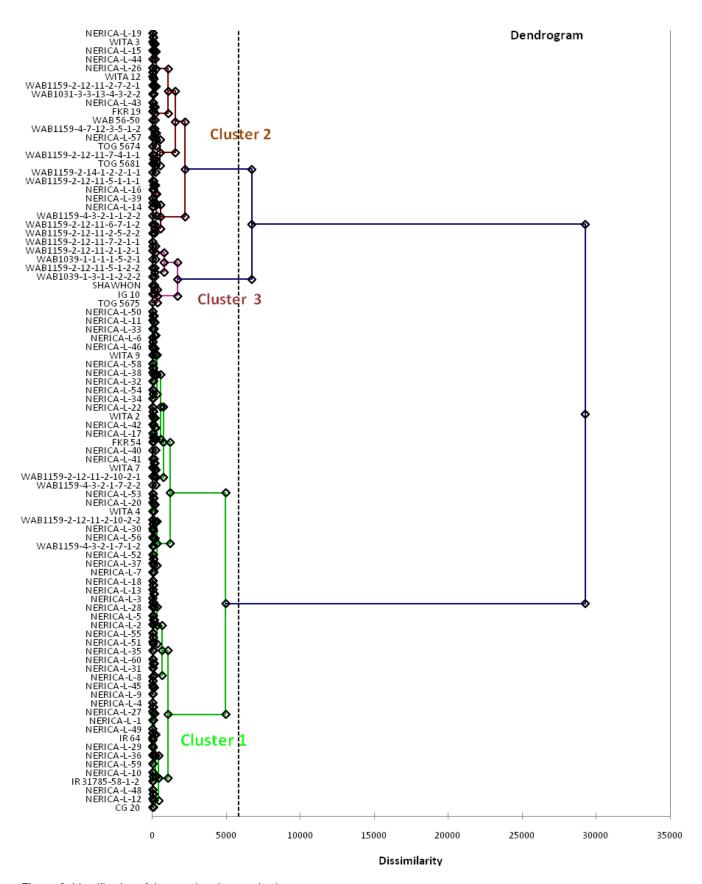


Figure 2. Identification of three major phenotypic clusters.

Traits	Season 1(WS1)	Season2(WS2)	InteractionWS1 x WS2
	Probability(& = 0.05)	Probability(& = 0.05)	Probability(& = 0.05)
Tillering 60 DAS	0.0674 <sup>ns</sup>	0.1354ns	0.8459ns
Leaf length	< 0.0001****	0.0007**	0.0498*
Leaf width	< 0.0001****	< 0.0001***	< 0.0001***
Plant height at maturity	0.0129ns	0.2796ns	0.9999ns
Spikelet fertility	0.3568ns	0.4105ns	0.9743ns
Panicle primary branching	< 0.0001**	0.1209ns	0.9997ns
Panicle secondary branching	0.0037*	0.0421*	0.2808ns
Number of panicles per plant	0.6735ns	0.9857ns	0.9999ns
Length of grain	0.135ns	< 0.0001***	0.0175*
Thickness of grain	< 0.0001**	0.1126ns	0.0029*
Maturity	0.0341*	0.0976ns	0.2027ns

**Table 6.** Variability of phenotypic expression of agro-morphological traits.

rice varieties in Burkina Faso. A strong phenotypic variability was observed within lowland NERICA and within promising lines according to the qualitative traits P ex, Fla and Bsc. We hypothesize that the qualitative traits such as anthocyanine content for Bsc could be used as a marker enabling the selection desired lines. Multivariate analysis of T60, Hmat, LL, LW, Ppb and Psb traits explain the maximal variability between genotypes. Genotype-environment interaction (G x E) effect was evaluated in order to describe the morphological variability between varieties used. G x E effect also showed high variability regarding only the characters related to panicle (Ppb, Psb, GL and G W). Although Jacquot and Arnaud (1979) and Sié (1991) demonstrated that variables related to panicle were relatively stable. some explanation can be given to help the understanding of these differences. Some traits such as "color of flowers" were very stable and they differ from one genotype to another depending on their genes and gene associations. However, other traits such as the sensitivity to a disease do not show any morphological variability (INRA, 2006). In this study, morphological modifications observed on some genotypes during the second season might have resulted from the action of environmental effects on plant growth and development. In a similar study, Cissé et al. (2006) reported that phenotypic variability can be observed even between genotypes belonging to the same group with the same parents. In addition, morphological variability observed in the lowland NERICA and promising lines could be explained by the combined action of rainfall and soil preparation during the two seasons on the vegetative plant cycle. The latter could have a negative incidence on the supply of ground water by running and seepage water and consequently on the level of the water. The results of this study are similar to those of Steel (1972) who showed that morphological variations can be observed with the same genotype due to environmental conditions such as fertility

of soil, water regime, light and temperature. Moreover, Morkinyo and Ajibade (1998) obtained similar results by using morphological characters. Nevertheless, intravarietal variations were not observed agronomically in lowland NERICA, or in promising lines.

No difference between varieties with regard to the level of backcrossing and the origin of the pedigree was observed within lowland NERICA and within promising lines. The first two clusters obtained after the cluster analysis are the mixture of lowland NERICA from backcross 2, 3 and 4. Cluster 1 contains lowland NERICA varieties with agro-morphological characteristics recommended in lowland rice-growing. These NERICA were derived from the crossing TOG5681 and IR64, and a low proportion of promising lines resulting from crossing WAB56-50 and SHAWHON. However, a minority of lowland NERICA varieties resulting from crossing TOG5681 and IR64 and the majority of promising lines resulting from crossing WAB56-50 and SHAWHON belong to cluster 2.

The varieties in cluster 1 have the best quality due to the level of introgression of the inheritance of their African parent which provide these varieties the capacity to resist many biotic stresses such as weeds competitiveness (Rodenburg et al., 2009), resistance to nematodes (Bimpong et al., 2006) and resistance to many abiotic stresses such as absence of water control and lack or toxicity of certain minerals (Gregorio et al., 2006; Kambou, 2008). Referring to Mendelian genetics, the recurrent parent during the process of backcrossing provide to its progeny, 25% (BC1), 12.5% (BC2) and 6.25% (BC3) of its genetic background. The differences observed between lowland NERICA and promising lines could possibly result from the level of introgression of the genetic inheritance of their African parent. A study on lowland NERICA BC2, BC3 and BC4 obtained from crossing TOG5681 and IR64 carried out by Ndjiondjop et al. (2008) revealed that the proportions of the donor

Traits	Cluster 1	Cluster 2	Cluster 3
Leaf length	Short	Intermediate	Long
Leaf width	Close	Large	Narrow
Plant height at maturity	Semi-dwarf	Intermediate	Tall
Spikelet fertility	Fertile	Fertile	Fertile
Panicle primary branching	Low	Middle	Middle
Panicle secondary branching	Dense	Dense	Very dense
Number of panicles per plant	High	Little	Intermediate
Length of grain	Long	Long	Long
Maturity	Intermediate	Intermediate	Intermediate
Tillering 60 DAS	Good	Medium	Medium

**Table 7.** Agro-morphological characteristics of 3 different clusters obtained.

parent (TOG5681) were 7.2, 8.5 and 8.1%, respectively. On the other hand, the high tillering capacity recorded in the varieties of cluster 1 confirmed the results obtained by Johnson et al. (1998) that showed the tendency of varieties resulting from O. glaberrima × O. sativa. However, differences obtained for this trait between lowland NERICA of clusters 1 and 2 could explain the expression of a transgressive variation. Although hybridization produces heterosis or hybrid vigor in the progeny, Rieseberg et al. (1999) and Semon et al. (2006) showed that interspecific hybridization between O. glaberrima x O. sativa can be an efficient means to generate superior rice cultivars adapted to environmental conditions of rice-growing in Africa. The 10 lowland NERICA released in West and Central Africa were mainly found in cluster 1: NL-18 (Benin), NL-19 (Benin, Burkina Faso, Cameroon, Liberia, Sierra-Leone and Togo), NL-20 (Mali, Burkina Faso and Sierra-Leone), NL-26 (Benin), NL-34 (Togo), NL-39 (Niger), NL-41 (Burkina Faso), NL-42 (Mali), NL-49 (Niger) and NL-60 (Burkina Faso), released in West and Central Africa were mainly found in cluster 1.

It should be noted that there are two types of lowland NERICA: lowland NERICA from cluster 1 exclusively adapted to lowland rice-growing and those of cluster 2 that can be grown in lowland conditions as well as in upland conditions (Table 7).

Varieties from cluster 3 were morphologically very similar to their African parents: TOG5675, TOG5674, IG10 and SHAWHON. Similar results were obtained by Cissé et al. (2006) by considering other parameters such as early good vigor, red caryopsis, absence of secondary branching on the panicle and susceptibility to shelling.

#### Conclusion

Agro-morphological characterization of 60 lowland NERICA and 18 promising lines showed phenotypic variability between varieties. The genetic diversity of 78

genotypes can be studied using the following quantitative traits: T60, Hmat, LL, LW, Ppb and Psb. Lowland NERICA and promising lines belonging to cluster 1 are the best varieties for rice-growing in lowland conditions. Those belonging to cluster 2 can be considered as intermediate varieties, that can be grown in the two major ecologies: lowland and upland. The level of backcross does not show any significant difference between lowland NERICA. Molecular characterization of all lowland NERICA and promising lines could be used to further study each genotype in order to better exploit their diversity according to the genetic inheritance of their African parent.

#### **ACKNOWLEDGEMENTS**

The authors are thankful to **L'**OREAL - UNESCO program for funding this PhD work. We also thank AfricaRice for the facilities and experimental sites used during the study. The first author would also like to thank Dr. Drame Khady Nani and M. Kouadio Yao Nasser and M. Awe Vincent for their helpful assistance on the manuscript.

#### **REFERENCES**

Abodolereza A, Racionzer P (2009). Food Outlook: Global Market Analysis (December 2009). www.fao.org/docrep/012/ak341f/ak341f00, pp. 23-27.

Africa Rice Center (WARDA) (2008). 2007 Africa Rice Trends. Cotonou, Benin: Africa Rice Center (WARDA), p. 84.

Arraudeau M (1975). About the choice of parents and some ways of obtaining new varieties in rice (*Oryza sativa* L.). Agron. Trop., 1: 8-17.

Bajracharya J, Steele KA, Jarvis DI, Sthapit BR, Witcombe JR (2006). Rice landrace diversity in Nepal: Variability of agro-morphological traits and SSR markers in landraces from a high-altitude site. Field Crops Res., 95: 327-335.

Barry MB, Pham JL, Noyer JL, Billot C, Courtois B, Ahmadi N (2007). Genetic diversity of the two cultivated rice species (*O. sativa* and *O. glaberrima*) in Maritime Guinea: Evidence for interspecific recombination. Euphytica, 154: 127-137.

Bimpong IK, Carpena AL, Gregorio GB, Reversat G, Brar DS (2006).

- Evaluation of progeny of  $Oryza\ sativa\ \times\ O.\ glaberrima$  for resistance to nematodes and molecular characterization of introgression in summary of proceedings beyond the first generation NERICA in Africa: paradigms and partnerships for the next decade. AfricaRice: Dar es Salaam, p. 9.
- Bioversity International-IRRI-AfricaRice (2007). Descriptors for wild and cultivated rice (*Oryza* spp.), p. 63.
- Cissé F, Yalcouye N, Doumbia Y, Hamadoun A (2006). Development of interspecific populations *O. sativa* L. and *O. glaberrima* Steud and identification of strains resistant to drought conditions in lowland rice cultivation and controlled flooding in summary of proceedings: Beyond the first generation NERICA in Africa: paradigms and partnerships for the next decade. Africa Pica E or Salaam, p. 23.
- Chinese cooperation (2009). Meteorological data collection area in the province of Deve Mono Couffo. Internal Report.
- Demol J, Baudoin JP, Louant BP, Maréchal R, Mergeai G, Otoul E (2001). Plant breeding: application to the main species grown in tropical regions. Ed presses of Gembloux, p. 583
- FAO (2010). Trends of rice paddy production: monitoring the market. http://www.fao.org\esc\fr\15\70\highlight\_71html.
- Federer WT (1956). Augmented designs. Hawaiian Planter's Rec., 55: 191-208
- Gregorio GB, Sié M, Ndjiondjop MN, Tsunematsu H, Wissuwa M, Atlin GN, Brar DS, Singh RK, Bennett J, Ismail AM, Mackill DJ (2006). Design of rice varieties tolerant to abiotic stress for rainfed lowlands in sub-Saharan Africa. In summary of proceedings: Beyond the first generation NERICA in Africa: paradigms and partnerships for the next decade. AfricaRice: Dar es Salaam, p. 8.
- INGER IRRI (1996). Standard Evaluation System for rice. 4<sup>th</sup>ed. IRRI, p. 52.
- INRA (2006). Analysis and management of genetic diversity. http://www.inra.fr/gap/departement/activites/diversite.htm.
- Jacquot M, Arnaud M (1979). Numerical classification of rice varieties. Agro. Trop., 34: 157-173.
- Johnson DE, Dingkhun M, Jones MP, Mahamane MC (1998). The influence of rice plant type on the effect of weed competition on *Oryza sativa* and *Oryzaglaberrima*. Weed Res., 38: 207-216.
- Kambou Kambou KA (2008). Evaluation of water stress in lowland rice based on varieties and planting dates. Memory Engineering, Polytechnic University of Bobo-Dioulasso, pp. 1-84.

- Morkinyo JA, Ajibade SR (1998). Characterization of the segregants of an improved cowpea line IT84K-124-6. Nig. J. Sci., 32: 27-32.
- Ndjiondjop MN, Semagn K, Sie M, Cissoko M, Fatondji B, Jones M (2008). Molecular profiling of interspecific lowland rice populations derived from IR64 (*Oryza sativa*) and Tog5681 (*Oryza glaberrima*). Afri. J. Biotechnol., 23: 4219-4229.
- Ndour D (1998). Tests of Agro-morphological characterization and genetics of salt tolerance in rice (*Oryza sativa* L.) in the Senegal River Delta. Memory Master II, University Cheikh Anta Diop in Dakar, pp. 1-27
- Ogunbayo SA, Ojo DK, Guei RG, Oyelakin OO, Sanni KA (2005). Phylogenetic diversity and relationships among 40 rice accessions using morphological and RAPDs techniques. Afr. J. Biotechnol., 11: 1234-1244.
- Rodenburg J, Saito K, Glele KR, Toure A, Mariko M, Kiepe P (2009). Weed competitiveness of the lowland rice varieties of NERICA in the southern Guinea Savanna. Field Crops Res., 114: 411-418.
- Rieseberg LH, Margaret AA, Wayne RK (1999). Transgressive segregation, adaptation and speciation. Heredity, 83: 363-372.
- Semagn K, Ndjiondjop MN, Cissoko M (2006). Microsatellites and agronomic traits for assessing genetic relationships among 18 New Rice for Africa (NERICA) varieties. Afr. J. Biotechnol.. 10: 800-810.
- Semon M, Kobi KO, Gridley H, Ndjiondjop MN, McCouch S (2006). Identification de la variation transgressive associée aux QTL pour la résistance à la sécheresse chez les descendances interspécifiques (*O. sativa* L. × *O. glaberrima* Steud.) in summary of proceedings: Beyond the first generation NERICA in Africa: paradigms and partnerships for the next decade. AfricaRice: Dar es Salaam. p. 20.
- Sié M (1991). Exploration for and Evaluation of genetic varieties of traditional rice (*Oryza sativa* L. and *O. glaberrima* Steud.) Burkina Faso. PhD thesis Engineer, National University of Côte d'Ivoire, Abidjan, p. 118.
- Steel WM (1972). Cowpeas in Nigeria Doctoral thesis, University of Reading, United Kingdom, p. 241.
- Ward JH (1963). Hierarchical Grouping to optimize an objective function. J. Am. Stat. Assoc., 58: 236-244.
- WARDA. (2005). Vers de nouveaux horizons. Rapport annuel 2003 2004.