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Agrobiodiversity in Middle Casamance (South Senegal): collection and agro-morphological assessment of traditional rice landraces.

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Abstract: Casamance is the main centre of cultivated traditional rainfed rice in Senegal including *Oryza sativa* and *O. glaberrima* species. Traditional landraces are an important source of genetic variability essential for the improvement of elite varieties adapted to local conditions. The study aimed at collecting and characterizing rice accessions cultivated in Middle Casamance. A total of 11 valleys and 29 villages were visited. Based on farmers' perception on agronomic traits, 171 rice accessions were collected and 136 characterized in an open-field trial. Accessions presented a high variability for all the traits assessed (p-value < 0.001) and were grouped into three main clusters based on seven quantitative variables with grain yield and panicle number as the most discriminative traits. Most of the accessions was semi dwarf to intermediate plant type, early to medium flowering, early maturing and with a low to moderate tillering ability. An average overall mean diversity (0.52 ± 0.21) was observed with the highest in Sindina, Balmadou and Bambali valleys. This study revealed an interesting diversity in the collection, representing a pre-breeding material for future rice improvement programs targeted to Casamance local conditions. It highlights also the significant role played by local farmers in maintaining and shaping local germplasm.

Keywords: Rainfed rice, Adaptation, Germplasm evaluation, Diversity Index

Introduction

Rice is considered as a staple food in many countries of the world (Khush, 1997). In West Africa, rice farming is one of the principal sources of income and food energy for smallholder farmers (Seck et al., 2013). In Senegal, rice represent 50% of annual cereal crop consumption (Gergely and Baris, 2009). Rainfed rice system, which now covers 20% of Senegalese rice production, is supposed to cover 40% according to the objectives of the National Program for Rice Self-Sufficiency (PNAR – Programme National d'Autosuffisance en Riz). It is practiced specifically in the Southern, South-eastern and South-central zones of the country

(Kanfany *et al*., 2016). Casamance region remains however the principal rainfed rice area of Senegal due to its potentiality in terms of rainfall and its Costal South Sudanese climate (Sambou *et al*., 2014). Furthermore, a vast hydrographic scheme represented by Casamance river and its tributaries provides a huge number of valleys and land areas suitable for rice cultivation (Sambou *et al*., 2014). In this region, rice is cultivated all along the topographic sequence from upland to swamp areas with the traditional inland valleys remaining the most exploited. Casamance region has a long tradition in rice cultivation, representing the main part of Senegal where both species *O. sativa* and *O. glaberrima* are cultivated (Linares, 2002) allowing therefore the existence of diverse rice landraces adapted to different ecologies. Further, this region is considered as one of the secondary centers of diversification of *O. glaberrima* (Porteres, 1950). For these reasons, a huge rice variability is expected to exist with local farmers acting as main force in preserving and shaping these genetic resources. In Casamance rice cultivation practices remains non-intensive with a low use of input (Gergely & Baris, 2009), related to a strong tradition of self-sufficiency, with the consequence that, often, yields remain weak, around 1 t ha^{-1} (Kanfany, 2014).

To increase rice production and yields in Casamance region, recent technologies were introduced and proposed to farmers for their adoption. More recently, it should be reminded the introduction and promotion of NERICA varieties (New Rice for Africa). These technologies include high yielding improved varieties which were promoted for years by the Senegalese Government, international donors and non-governmental organizations with the aim of increasing domestic production to ensure food security. However, Casamance traditional rice resources that include both cultivated rice species are neglected and thus threatened to disappear. Even though they have generally limited performances in terms of yields, traditional varieties are considered to be more adapted to local conditions and constitute therefore an important reservoir of genetic variability for crop improvement (Sanni et al., 2016). In fact, their greater genetic variability could make them better adapted to local ecologies in terms of soil types, stress tolerance, ripening period and yield stability (Oka, 1988; Guei and Traore, 2001). These genetic resources, grown for years, are certainly unique for their adaptability to local conditions and adopted by smallholder farmers for a set of characteristics such as plant height and harvesting facilities, grain quality and maturing period. Moreover, in a such typical subsistence agriculture where seasons are unpredictable, some local varieties can outperform modern varieties (Sall et al., 1998).

Rice genetic resources existing in Casamance have not yet been assessed, despite their key role in the achievement of household food security. Indeed, very few studies, mainly from sixties and seventies, were conducted on rice traditional varieties from this southern part of Senegal (ISRA-Djibelor, 1978; 1980). Conversely, the collection, the preservation and the valorisation of this genetic material are crucial in terms of contribution to local food security, reliability to climate change and use in plant breeding programs targeted towards the diversification of varieties' genetic base and the adaptation to local edaphic conditions and needs of local communities. For an effective use of these genetic resources, an accurate assessment of phenotypic and genetic variability is required.

In this study, we aimed at conducting a germplasm collection and assessing the phenotypic variability in rice landraces from Middle Casamance. Knowledge and conservation of local landraces will provide a broad base of genetic variability from which improved rice varieties can be developed, thus aiding in the stabilization of a secure and sustainable food supply for households of Casamance region.

Materials and Methods

Germplasm collection

The germplasm collection was carried out during the dry season of 2015 in the agro-ecological zone of Middle Casamance which corresponds to the administrative region of Sedhiou (Senegal). A total number of 29 villages polarized around 11 valleys and located in the district of Sedhiou and Goudomp, were visited (Figure 1). Visited valleys were Balmadou, Bambali, Boumouda, Djiredji, Kinthiengrou, Medina, Same, Samiron, Sindina, Badiary and Djimbana, which insist on a territory of about 600 km^2. Samples were collected on farmers' choice criteria and perceptions such as grain yield, earliness, plant height, grain quality, tolerance to biotic and abiotic stresses. A total number of 171 rice accessions were collected (Table 1). Most of these samples were collected as panicles. In the case where they were collected as grain, off-type seeds were visually separated based on grain shape and color.

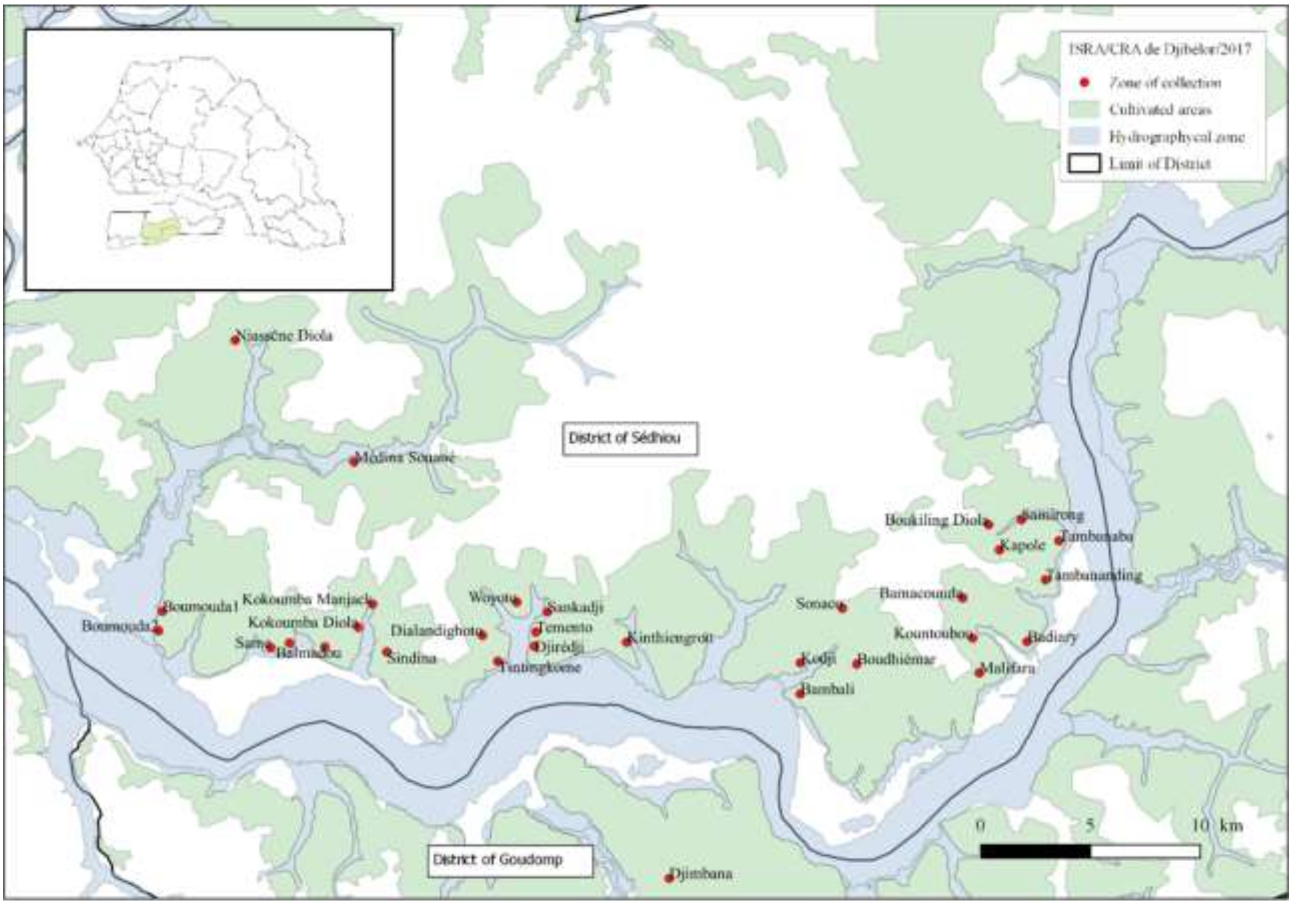

Figure 1 - Map of the valleys and villages surveyed

Table 1 - Number of accessions per valley and village of collection

COLLECTION DATE	VALLEY	VILLAGE	N OF ACCESSIONS
29/01/2015	Samiron	Tambanaba	11
29/01/2015	Samiron	Boukiling Diola	6
30/01/2015	Samiron	Kapole	7
31/01/2015	Djimbana	Djimbana	6
02/02/2015	Djiredji	Sankadji	5
02/02/2015	Djiredji	Tingtingkom	9
02/02/2015	Djiredji	Dialandighoto	4
02/02/2015	Djiredji	Temento	5
03/02/2015	Boumouda	Boumouda 1	2
03/02/2015	Boumouda	Boumouda 2	9
03/02/2015	Sindina	Koukoumba Diola	4
04/02/2015	Balmadou	Balmadou	8
04/02/2015	Samet	Samet	8
04/02/2015	Sindina	Koukoumba Manjaque	4
05/02/2015	Balmadou	Balmadou Manjaque	7
05/02/2015	Djiredji	Woyoto	4
05/02/2015	Kinthiengrou	Kinthiengrou	6
05/02/2015	Sindina	Sindina	7
06/02/2015	Bambali	Kodji	6
06/02/2015	Bambali	Sonaco	2
06/02/2015	Bambali	Bambali	6
06/02/2015	Bambali	Boudhiemar	3
18/02/2015	Badiary	Badiary	6
18/02/2015	Badiary	Bamacounda	4
18/02/2015	Badiary	Kountoubou	6
18/02/2015	Badiary	Malifara	6
18/02/2015	Badiary	Tambananding	4
19/02/2015	Medina Souané	Medina Souané	10
19/02/2015	Medina Souané	Niassène Diola	6
Gran total	11	29	171

Agro-morphological characterization

Based on seeds purity and quantity, a total of 136 accessions were selected and characterized (Table 2). Accessions with the same local names but collected in different villages were maintained separated. Four elite varieties grown in Casamance region, DJ 11-509 (upland rice variety), BG 90-2 (lowland and irrigated perimeters), TOX 728-1 (hydromorphic soil, intermediate between upland and lowland) and WAR 77 (mangrove zone), were used as control.

Table 2 - List of selected and evaluated accessions

CODE	SPECIES	DESIGNATION/LOCAL NAME	REGION OF COLLECT	VILLAGE OF COLLECT
CP1	*O.sativa*	Ablaye mano	Sédhiou	Kountoubou
CP2	*O.sativa*	Koudang	Sédhiou	Kodji
CP3	*O.sativa*	Ablaye mano	Sédhiou	Bounkiling diola
CP4	*O.sativa*	Bandi	Sédhiou	Kodji
CP5	*O.sativa*	Kayuro	Sédhiou	Kodji
CP6	*O.sativa*	Manofing	Sédhiou	Djimbana
CP7	*O.sativa*	Awa biaye	Sédhiou	Malifara
CP8	*O. glaberrima**	Tiebouding	Sédhiou	Sindina
CP9	*O. glaberrima**	Colsa	Sédhiou	Tingtinckome
CP10	*O.sativa*	Dania	Sédhiou	Tingtinckome
CP11	*O.sativa*	Wankarang	Sédhiou	Tambanaba
CP12	*O. glaberrima**	Moussofing	Sédhiou	Bambali
CP13	*O.sativa*	Noire	Sédhiou	Balmadou Manjaque
CP14	*O.sativa*	Guinée	Sédhiou	Koucoumba Diola
CP15	*O.sativa*	Chinois	Sédhiou	Tingtinckome
CP16	*O.sativa*	Wankara	Sédhiou	Balmadou
CP17	*O.sativa*	Toure toure	Sédhiou	Balmadou
CP18	*O.sativa*	Nokono	Sédhiou	Sindina
CP19	*O. glaberrima**	Nene	Sédhiou	Kinthiengrou
CP20	*O.sativa*	Mariama	Sédhiou	Dialandighoto
CP21	*O.sativa*	kondjimi	Sédhiou	Bambali
CP22	*O.sativa*	Gabou	Sédhiou	Bamacounda
CP23	*O.sativa*	Moussoukoi	Sédhiou	Temento
CP24	*O.sativa*	Richard toll	Sédhiou	Balmadou
CP25	*O.sativa*	Nioukourouba	Sédhiou	Tingtinckome
CP26	*O.sativa*	Pekene	Sédhiou	Boumouda
CP27	*O.sativa*	Koudjimi	Sédhiou	Badiary

CP28	*O.sativa*	Kolda	Sédhiou	Balmadou Manjaque
CP29	*O.sativa*	yayang	Sédhiou	Sindina
CP30	*O. glaberrima**	Ablaye mano	Sédhiou	Dialandighoto
CP31	*O.sativa*	Awa biaye	Sédhiou	Malifara
CP32	*O. glaberrima**	Niokono	Sédhiou	Madina Souané
CP33	*O.sativa*	Chinois	Sédhiou	Boumouda
CP34	*O.sativa*	Tiebouding	Sédhiou	Kapole
CP35	*O. glaberrima**	Manowouledingo	Sédhiou	Boumouda
CP36	*O.sativa*	Cisse thiopi	Sédhiou	Sankadji
CP37	*O.sativa*	Simbandi	Sédhiou	Balmadou
CP38	*O.sativa*	Denanomano	Sédhiou	Sindina
CP39	*O. glaberrima**	Diaouro	Sédhiou	Balmadou Manjaque
CP40	*O.sativa*	Diakong	Sédhiou	Balmadou
CP41	*O.sativa*	Synkape	Sédhiou	Kinthiengrou
CP42	*O.sativa*	Diarbancounda	Sédhiou	Kountoubou
CP43	*O. glaberrima**	Emana Yemeuy	Sédhiou	Same
CP44	*O.sativa*	Ngata	Sédhiou	Bambali
CP45	*O.sativa*	Badiouré	Sédhiou	Djimbana
CP46	*O.sativa*	Nimzat	Sédhiou	Bounkiling diola
CP47	*O.sativa*	Tuntunai	Sédhiou	Bounkiling diola
CP48	*O.sativa*	Adama diallo	Sédhiou	Balmadou Manjaque
CP49	*O.sativa*	Wankaran	Sédhiou	Djimbana
CP50	*O.sativa*	Koundjini	Sédhiou	Temento
CP51	*O.sativa*	kouboni	Sédhiou	Bounkiling diola
CP52	*O.sativa*	Koudjini	Sédhiou	Temento
CP53	*O.sativa*	Guinee	Sédhiou	Kapole
CP54	*O.sativa*	Barafita	Sédhiou	Kapole
CP55	*O.sativa*	Kolda	Sédhiou	Same
CP56	*O.sativa*	foulo	Sédhiou	Same
CP57	*O.sativa*	Koussy3	Sédhiou	Boudhiemar
CP58	*O.sativa*	Awa biaye	Sédhiou	Tambananding
CP59	*O.sativa*	Badiouré	Sédhiou	Temento
CP60	*O.sativa*	Barafita	Sédhiou	Tambanaba
CP61	*O. glaberrima**	Ballefoudon	Sédhiou	Sankadji
CP62	*O.sativa*	Mandina	Sédhiou	Bambali
CP63	*O.sativa*	Kaourou	Sédhiou	Koukoumba Manjaque
CP64	*O.sativa*	Badjouré	Sédhiou	Kinthiengrou

CP65	*O. glaberrima**	Gambie1	Sédhiou	Koukoumba Manjaque
CP66	*O.sativa*	Gambie2	Sédhiou	Koukoumba Manjaque
CP67	*O.sativa*	Diakone	Sédhiou	Tingtinckome
CP68	*O.sativa*	Mansara	Sédhiou	Kapole
CP69	*O.sativa*	Kuboni	Sédhiou	Sindina
CP70	*O.sativa*	Manowek	Sédhiou	Bambali
CP71	*O.sativa*	Koundjimi	Sédhiou	Malifara
CP72	*O.sativa*	Barafita	Sédhiou	Boumouda
CP73	*O.sativa*	Glory	Sédhiou	Same
CP74	*O.sativa*	Lekoutey	Sédhiou	Same
CP75	*O.sativa*	Karfamano	Sédhiou	Boudhiemar
CP76	*O.sativa*	Soranse	Sédhiou	Kapole
CP77	*O.sativa*	Diourimi	Sédhiou	Boumouda
CP78	*O.sativa*	Guinee	Sédhiou	Tambanaba
CP79	*O. glaberrima**	Parfumé	Sédhiou	Tambanaba
CP80	*O.sativa*	OUBOME	Sédhiou	Same
CP81	*O.sativa*	guinee	Sédhiou	Balmadou Manjaque
CP82	*O.sativa*	Mbomano	Sédhiou	Madina Souané
CP83	*O.sativa*	Kayoura	Sédhiou	Tingtinckome
CP84	*O. glaberrima**	Rolifin	Sédhiou	Sindina
CP85	*O.sativa*	Nimzat	Sédhiou	Kapole
CP86	*O.sativa*	Badjouré	Sédhiou	Sankadji
CP87	*O.sativa*	Awa biaye	Sédhiou	Tambanaba
CP88	*O.sativa*	Barafita	Sédhiou	Tambananding
CP89	*O.sativa*	Konene	Sédhiou	Tambanaba
CP90	*O. glaberrima**	Adama diallo	Sédhiou	Sankadji
CP91	*O. glaberrima**	Adama diallo	Sédhiou	Kountoubou
CP92	*O. glaberrima**	Manodiouno	Sédhiou	Woyoto
CP93	*O.sativa*	Abdoulaye mano	Sédhiou	Kodji
CP94	*O.sativa*	Massa mano	Sédhiou	Boumouda
CP95	*O. glaberrima**	Adama diallo	Sédhiou	Tingtinckome
CP96	*O.sativa*	Diafar	Sédhiou	Koucoumba Diola
CP97	*O.sativa*	Bonti	Sédhiou	Balmadou Manjaque
CP98	*O.sativa*	Awa biaye	Sédhiou	Kapole
CP99	*O.sativa*	Barafita	Sédhiou	Badiary
CP100	*O.sativa*	Poti	Sédhiou	Boumouda
CP101	*O.sativa*	Falkinto	Sédhiou	Djimbana

CP102	*O.sativa*	Essaie	Sédhiou	Tingtinckome
CP103	*O.sativa*	Awa biaye	Sédhiou	Badiary
CP104	*O.sativa*	Mandran mano	Sédhiou	Malifara
CP105	*O.sativa*	Bonti	Sédhiou	Dialandighoto
CP106	*O.sativa*	Madina	Sédhiou	Temento
CP107	*O.sativa*	Mandoz	Sédhiou	Djimbana
CP108	*O.sativa*	Kayoura	Sédhiou	Same
CP109	*O.sativa*	Tiebounding	Sédhiou	Balmadou
CP110	*O.sativa*	Koudjini	Sédhiou	Tambanaba
CP111	*O.sativa*	kousobe	Sédhiou	Woyoto
CP112	*O.sativa*	Seydicounda	Sédhiou	Woyoto
CP113	*O.sativa*	Toure counda	Sédhiou	Boumouda
CP114	*O.sativa*	Cisse dadji	Sédhiou	Sindina
CP115	*O.sativa*	koundjimy	Sédhiou	Kodji
CP116	*O. glaberrima**	Niambafino	Sédhiou	Boumouda
CP117	*O.sativa*	Seydicounda	Sédhiou	Kinthiengrou
CP118	*O.sativa*	Tambana	Sédhiou	Malifara
CP119	*O.sativa*	Kolda	Sédhiou	Koukoumba Manjaque
CP120	*O. glaberrima**	Lainno	Sédhiou	Tingtinckome
CP121	*O.sativa*	Sithior	Sédhiou	Same
CP122	*O.sativa*	Chinois	Sédhiou	Sankadji
CP123	*O.sativa*	Thiabidabo	Sédhiou	Kinthiengrou
CP124	*O.sativa*	Koussithila	Sédhiou	Kodji
CP125	*O.sativa*	Adama diallo	Sédhiou	Bambali
CP126	*O.sativa*	Ametié	Sédhiou	Kinthiengrou
CP127	*O.sativa*	Mancolicounda	Sédhiou	Djimbana
CP128	*O.sativa*	Medina	Sédhiou	Koucoumba Diola
CP129	*O.sativa*	Misra	Sédhiou	Bounkiling diola
CP130	*O.sativa*	Taulé	Sédhiou	Bambali
CP131	*O.sativa*	Koundjimi	Sédhiou	Boudhiemar
CP132	*O.sativa*	Améro	Sédhiou	Balmadou
CP133	*O.sativa*	Manotice	Sédhiou	Bambali
CP134	*O.sativa*	Fansenimano	Sédhiou	Bounkiling diola
CP135	*O.sativa*	Bonti	Sédhiou	Boumouda
CP136	*O.sativa*	Kouloukouba	Sédhiou	Balmadou

*to be confirmed by molecular characterization. CP = Collection Papsen

The trial was conducted during the rainy season of 2015 in the Agricultural Research Centre of Djibelor (12°33.42'09"N; 16°18.20'94"O) located in the Lower Casamance region. The experiment was laid out in a 10 x 14 alpha lattice design with two replications in a wetland field. 30 days old seedlings were transplanted after soil plugging and flooding at a rate of two seedlings per hill in a plot of 0.60 m^2 consisting of four rows of 1 m. The spacing was 0.20 m between and within rows. The distance between plots and between blocs were respectively 0.4 and 0.8 m. One plant per hill was left after thinning at 20 days after transplanting (DAT). NPK fertilizer (15-15-15) was applied at a rate of 200 kg ha^{-1} as basal fertilizer just after transplanting. Nitrogen fertilizer (urea) was applied at a rate of 75 kg ha^{-1} at 20 and 45 DAT. Manual weeding was carried out at 20, 30 and 45 DAT.

Data collection

Agro-morphological data based were recorded on days to 50% flowering, date to 80% maturity, plant height, panicle length and on yield components such as tiller number, panicle number and grain yield. Awning and colour of pericarp were visually assessed, data on insect damages (deadhearts and whiteheads) were also recorded. *O. glaberrima* and *O. sativa* species were identified mainly based on pericarp shape and colour.

Data analysis

Descriptive statistics were performed using R package (R Development Core Team R, 2015). GenSTAT version 12 software was used to perform the analysis of variance (ANOVA) using the Linear Mixed Model (REML). Genotypes were considered as fixed factor while replications and blocks within replications as random factors. To estimate the phenotypic variability and the structuration of the different genotypes based on the traits evaluated, principal component and cluster analyses using the Agglomerative Hierarchical Clustering (ACH) procedure of XLSTTAT v7.5.2 software were performed. Dissimilarities and structuration of different accessions were respectively based on Euclidian distance and Ward methods. Adjusted mean of quantitative traits was used to perform the multivariate analysis. Prior to the Shannon-Weaver diversity index analysis based on geographical distribution, quantitative data were transformed into categorical data (Table 3) as described in rice descriptor (Biodiversity International *et al.*, 2007) and according to Rabara *et al.*, 2014 and Sanni *et al.*, 2016).

Table 3 - List of variables and their classes adopted from the rice descriptor (Biodiversity International et al.*, 2007 and Sanni* et al. *2016)*

CHARACTER	CHARACTER CLASSES	CODE	DESCRIPTION
Plant height	< 110 cm	1	Semi dwarf
	111-130 cm	5	Intermediate
	> 130 cm	9	Tall
50% Flowering	< 75 days	1	Very early
	76-90 days	3	Early
	91-105 days	5	Medium
	106-120 days	7	Late
	> 120 days	9	Very late
Tiller	> 25	1	Very high
	20-25	3	Good
	10-19	5	Medium
	5-9	7	Low
	< 5	9	Very low
Panicle length	< 15 cm	1	Short
	16-25 cm	5	Medium
	> 25 cm	9	Long
80% Maturity	<100 days	1	Very early
	101-115 days	3	Early
	116-130 days	5	Medium
	131-145 days	7	Late
	> 145 days	9	Very late

Class frequencies of each phenotypic trait were used for diversity analysis according to the Shannon-Weaver diversity index:

$$H' = -\sum_{I=1}^{k} Pi \log_2 Pi$$

Where k is the number of phenotypic classes for a character and Pi the proportion of the total number of entries in the ith class. Each value of H' was standardized by dividing it by its maximum value ($\log_2 k$) to keep the values in the interval of 0 to 1, where the highest value indicates the maximum diversity (Sow *et al.*, 2014; Rabara *et al.*, 2014).

Results

Experimental condition

A total rainfall of 1442.5 mm was recorded, from June to November with a monthly cumulative mean of 381 mm from July to September. The acidity and salinity levels in the experimental field recorded before and every week after transplanting presented respectively a pH range between 6-7 and an electro-conductivity variation between 0.5 to 1 dS m^{-1}.

Agro-morphological and phenotypical variability

Significant variation with p-value < 0.001 and < 0.05 was observed for all the quantitative traits assessed (Table 4).

Table 4 - Descriptive statistics and analysis of variance of the recorded quantitative traits.

VARIABLE	MINIMUM	MAXIMUM	MEAN	SD	P-VALUE	HERITABILITY
50% flowering (days)	66	129	93	14.3	<0.001	0.92
80% Maturity (days)	92	157	116	14.5	<0.001	0.94
Plant height (cm)	51.7	151.5	109	20.8	<0.001	0.87
Tiller number m^{-2}	123.1	384.3	242	49.6	<0.001	0.37
Panicle length (cm)	11.5	30.6	20.4	2.9	<0.001	0.72
Panicle number m-^{2}	150	1025	514	180.2	<0.050	0.33
Grain yield (kg ha^{-1})	493	6395	3380	1076.4	<0.001	0.42

The accessions flowered between 66 to 129 days and matured between 92 to 157 days, while for plant height the range was from 51.7 to 152 cm. These differences observed in plant height, flowering and maturing period were also observed in tiller m^{-2} (123 to 384), panicle number m^{-2} (150 to 1025), grain yield (493 to 6395 kg ha^{-1}) and panicle length (11.5 to 30.6 cm). In the collection, 5% presented awn and the colour of the pericarp was mainly brown (68%), dark brown (18%), red (10%) and black (4%). Based on the shape and the colour of the pericarp, nineteen accessions were identified as *O. glaberrima*. They presented few or absence of ramifications and a coloured to red pericarp and were characterized by a flowering period varying between 66 and 122 days and a grain yield of 1399 to 3852 kg ha^{-1}.

The susceptibility to insect attack, particularly those ones causing deadhearts and whiteheads, was assessed. The phenotypic distribution of injured plant (deadhearts m^{-2} and whiteheads m^{-2}) are shown in figures 2 and 3. The most susceptible accession to insects causing deadhearts presented a mean of 32 injured plants m^{-2} compared to an overall mean of 9 damage plants m^{-2}. Referred to whiteheads symptoms, the overall mean was 20 plants m^{-2}, the most susceptible accession presenting a mean of 71 attacked panicles m^{-2}.

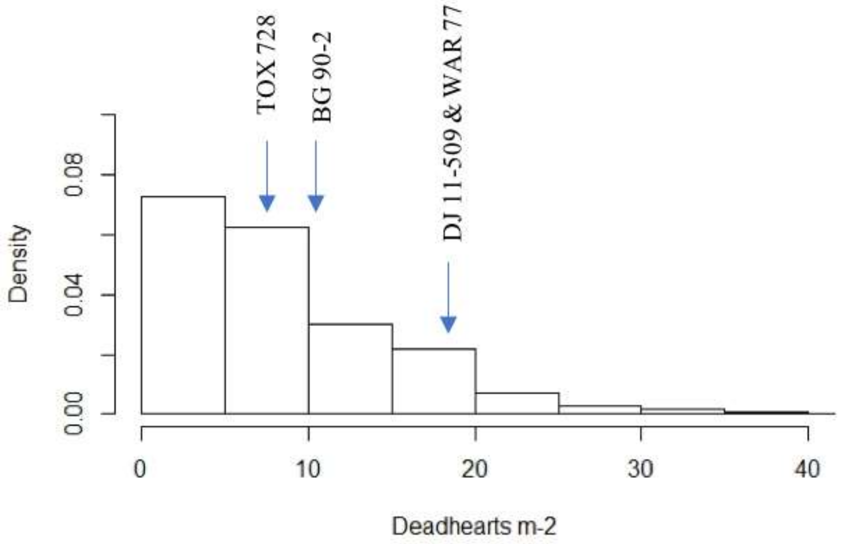

Figure 2 - Frequency class distribution of injured plant with deadhearts symptoms (control elitevarieties are indicated as a reference)

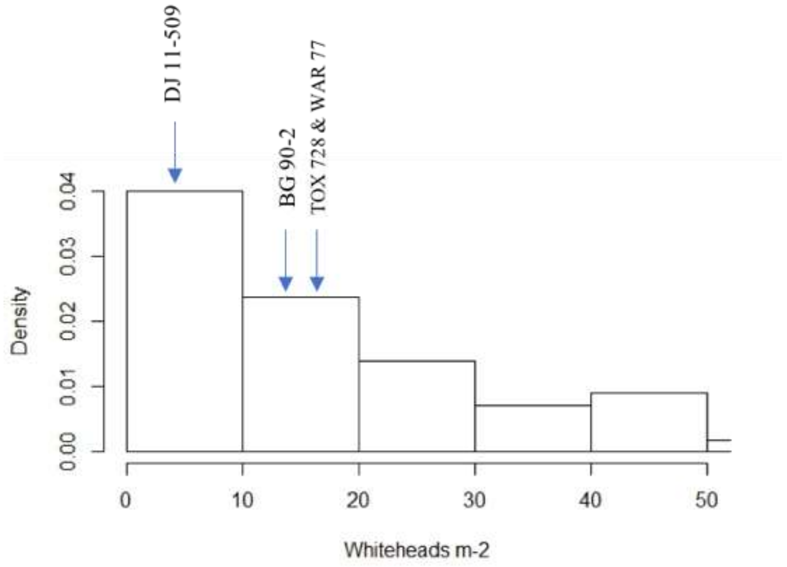

Figure 3 - Frequency class distribution of injured plant with whiteheads symptoms (control elite varieties are indicated as a reference)

Three main components explained 83.9% of the total variation observed in the collection (Table 5). The first component (PC1) counted for 48.5% of the total variation with plant height, days to 50% flowering, days to 80% maturity and panicle length as main contributors. PC2 and PC3 contributed respectively for 20.5% and

14.9% of the variation. Grain yield and panicle number were highly correlated to PC2 while PC3 was mostly and significantly associated to tiller m^{-2} (0.91).

Table 5 - Principal component correlation matrix, Eigen value and proportion of total variance of different variables

VARIABLE	PC1	PC2	PC3	PC4
Plant height (cm)	0.84	0.03	-0.25	-0.10
50% Flowering (day)	0.93	-0.04	0.15	0.24
Tiller m^{-2}	0.16	0.26	0.93	-0.20
Panicle length (cm)	0.84	0.16	-0.12	-0.14
Grain yield (kg ha^{-1})	0.25	0.83	-0.27	-0.25
80% Maturity (day)	0.93	-0.06	0.11	0.25
Panicle number m^{-2}	-0.39	0.80	0.03	0.39
Eigenvalue	3.40	1.44	1.04	0.41
Variability (%)	48.55	20.51	14.91	5.81
Cumulative %	48.55	69.06	83.98	89.78

The bi-plot representation of PC1 and PC2 accounting together for 69% of total variation (Figure 4) did not show a clear grouping. The structuration observed with the principal component analysis (PCA) was therefore not enough informative.

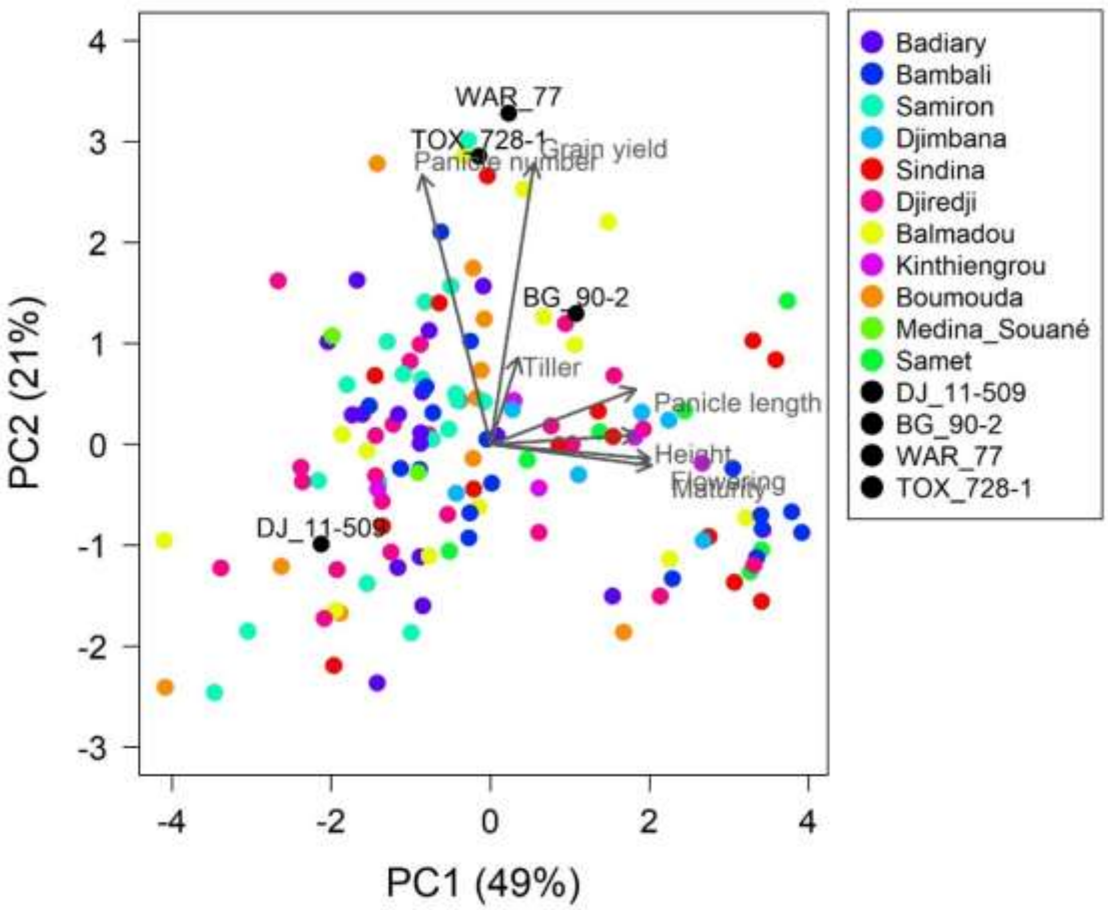

Figure 4 - Bi-plot representation by valley of the first two principal components (reference varieties are indicated with black dots)

Cluster analysis

Cluster analysis separated the 136 accessions and the four controls into three main clusters (Figure 5). Among the traits used for clustering, grain yield (p-value < 0.0001) and panicle number m^{-2} (p-value = 0.0004) were significantly the most discriminant.

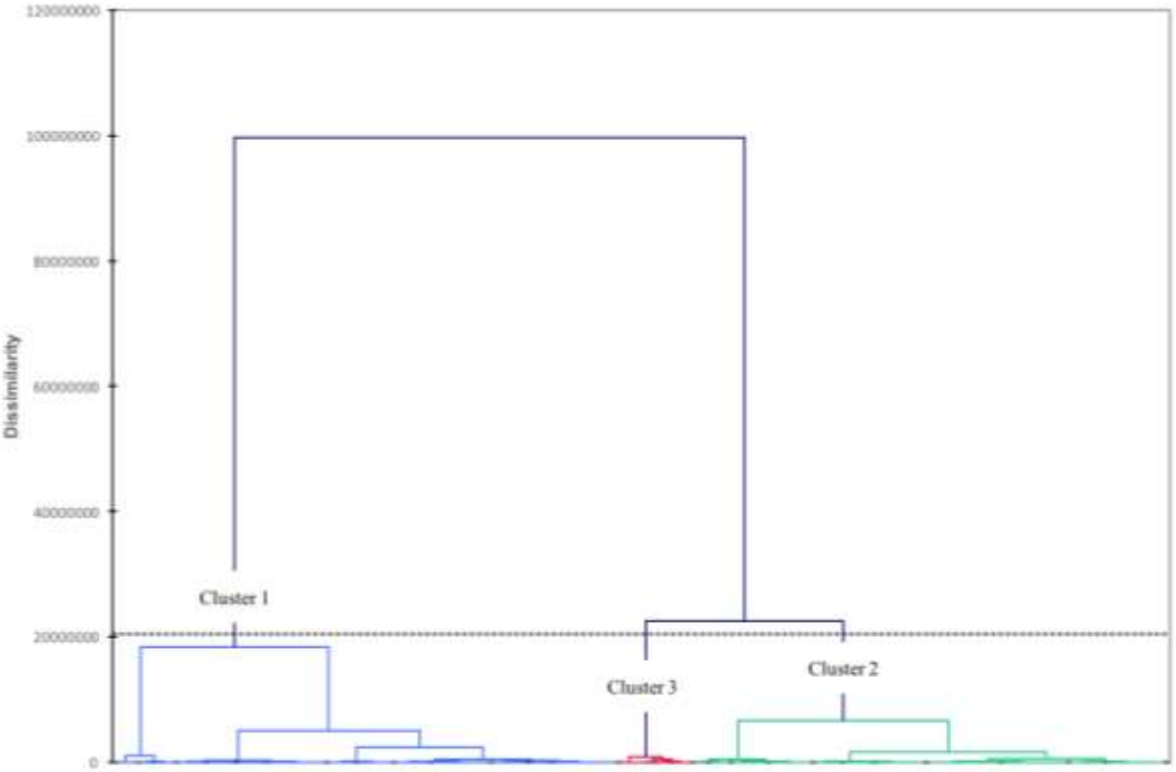

Figure 5 - Cluster view of tested accessions based on the Agglomerative Hierarchical Clustering (ACH) analysis performed using seven quantitative traits assessed on germplasm collection.

Table 6 summarizes the average characteristics of each cluster.

C3 cluster included the ten highest performing genotypes representing the 7% of the collection (Table 7). It was characterized by an average of 5583 kg ha^{-1} in grain yield, 621 panicles per m^{2} and 22 cm in panicle length. Three of four improved varieties were grouped together in cluster C3, notably BG 90-2 (5594 kg ha^{-1}), WAR 77 (5989 kg ha^{-1}) and TOX 728-1 (6394 kg ha^{-1}). Local cultivars named Chinois, Tiebounding, Kaourou, Kayoura and Kousoube showed a mean performance statistically similar to the three control varieties. Plant height in this cluster varied between 71.9 and 142.7 cm, panicle length from 19 to 30.6 cm and grain yield from 5138 to 6394.6 kg ha^{-1}. Thus, based on their characteristics, the accessions in C3 cluster, could show higher performances when grown in lowland ecologies.

C2 and C1 clusters reached respectively a grain yield of 3967 kg ha^{-1} and 2501 kg ha^{-1}. They grouped 63 and 67 accessions representing respectively 45% and 48% of the total collection. Accessions in C1, like DJ 11-509, adapted to upland ecology, were mostly characterized (70%) by a relatively short growing cycle with less than 100 days to 50% flowering and a plant height varying from 51 to 119 cm. C2 cluster included only local genotypes and were characterized by a plant height arising from 69 to 151 cm, a flowering and maturity period comprised between 76 and 127 and 103 and 156 days respectively.

Table 6 - Characteristics of the identified groups based on the hierarchical ascendant cluster analysis using seven quantitative traits

Cluster	Sample size (N)	PH (cm)	50% Flowering (days)	Tiller m^{-2}	Panicle length (cm)	Grain yield (kg ha^{-1})	80% Maturity (days)	Panicle number m^{-2}
C1	67	105.5	91.4	243.4	19.9	2500.9	115.2	471.1
C2	63	112.3	94.6	235.9	20.7	3966.7	117.2	536.5
C3	10	110.9	94.7	270.8	21.9	5582.9	118.4	621.3

Table 7 - Agronomical characteristics of the highest performing accessions and control varieties (in bold character) in cluster C3

Species	Designation	Origin / Valley of collection	50% Flowering (days)	80% Maturity (days)	Plant height (cm)	Tiller m^{-2}	Panicle Length (cm)	Panicle number m^{-2}	Grain yield (kg ha^{-1})
O. sativa	Tox 728-1	ISRA/CRA Djibelor	87.4	112	100.8	217.1	22.5	690.9	6394.6
O. sativa	WAR 77	ISRA/CRA Djibelor	94.6	117	113.8	305.7	20.5	775.2	5989.3
O. sativa	Chinois	Boumouda	83.5	111	71.9	231.9	20.6	761.9	5800.2
O. sativa	Tiebounding	Balmadou	99.4	120	87.6	354.0	21.9	623.8	5649.3
O. sativa	BG 90-2	ISRA/CRA Djibelor	96.9	118	119.0	234.3	22.5	470.9	5594.8
O. sativa	Kouloukouba	Balmadou	100.3	129	134.1	259.8	21.4	661.5	5494.7
O. sativa	Tiebounding	Sindina	100.1	121	84.9	332.0	20.6	727.6	5302.3
O. sativa	Kaourou	Sindina	77.5	101	126.5	262.2	19.2	512.7	5298.6
O. sativa	Kayoura	Same	108.5	132	142.7	252.3	30.6	470.9	5166.8
O. sativa	Kousobe	Djiredji	99.2	122	128.1	258.6	19.6	517.6	5138.1
SED			5.83	4.77	10.78	58.03	2.16	159.63	1141.78
LSD			11.52	9.43	21.32	114.73	4.27	315.72	2257.79

LSD least significant difference, SED standard error of difference

Diversity indices of the phenotypic variability based on the geographical zones

The Shannon diversity indices based on the frequency proportions (Table 8) of the traits and their classes was calculated for individual and pooled characters following the different geographical zones.

Pooled characters over geographical zone (Table 9) presented an average diversity index ranging between 0.25 for the valley of Badiary and 0.68 for the valley of Sindina. The lowest mean diversity index for the individual characters was noticed in panicle length (0.28) and the highest in plant height (0.77). The overall mean diversity index for all the entries was estimated at 0.52.

The characters varied differently among valleys and not all of them had all the descriptor class represented. Plant height, days to 50% flowering and days to 80% maturity were the most variable characters. Based on rice descriptors, 48% of the germplasm collection were semi dwarf (< 110 cm), 38% intermediate (111-130 cm) and 15% considered as tall (> 130 cm). Considering the period of flowering, 48% of the collection were early (76-90 days), 24% medium (91-105 days), 22% late (106-120 days) and 2% considered as very late (> 120 days). The extra early (< 75 days) genotypes represented only 4% of the collection. As for the flowering date, the early maturing accessions represented 54% of the collection. The collection was also characterized by a low (52%) to medium (48%) tillering ability and intermediate panicle length (16-25 cm) for 89% of the genotypes.

Table 8 - Frequency proportion of the different classes of the traits based on the geographical distributions

Valley	N. of accessions	Flowering (days)					Maturity (days)				
		1	3	5	7	9	1	3	5	7	9
Badiary	15	0.0	86.7	13.3	0.0	0.0	0.0	100.0	0.0	0.0	0.0
Balmadou	14	7.1	35.7	42.9	14.3	0.0	7.1	42.9	35.7	14.3	0.0
Bambali	17	0.0	35.3	23.5	29.4	11.8	0.0	52.9	5.9	23.5	17.6
Boumouda	9	0.0	44.4	44.4	11.1	0.0	0.0	44.4	44.4	11.1	0.0
Djimbana	6	0.0	16.7	33.3	50.0	0.0	0.0	33.3	16.7	50.0	0.0
Djiredji	15	13.3	53.3	33.3	0.0	0.0	20.0	53.3	26.7	0.0	0.0
Kinthiengrou	6	0.0	33.3	16.7	50.0	0.0	0.0	33.3	16.7	50.0	0.0
Medina	4	0.0	75.0	0.0	25.0	0.0	0.0	75.0	0.0	25.0	0.0
Same	8	0.0	12.5	25.0	62.5	0.0	0.0	25.0	12.5	62.5	0.0
Samirong	20	10.0	75.0	10.0	5.0	0.0	10.0	80.0	10.0	0.0	0.0
Sindina	14	0.0	28.6	21.4	42.9	7.1	0.0	28.6	21.4	42.9	7.1
Tingtingkom	8	0.0	37.5	25.0	37.5	0.0	12.5	37.5	12.5	37.5	0.0
Total	136	3.7	47.8	24.3	22.1	2.2	5.1	54.4	16.9	20.6	2.9

Valley	N. of accessions	Tiller (N m^{-2})					Panicle length (cm)			Plant height (cm)		
		1	3	5	7	9	1	5	9	1	5	9
Badiary	15	0.0	0.0	33.3	66.7	0.0	0.0	100.0	0.0	53.3	46.7	0.0
Balmadou	14	0.0	0.0	71.4	28.6	0.0	7.1	85.7	7.1	50.0	21.4	28.6
Bambali	17	0.0	0.0	41.2	58.8	0.0	0.0	82.4	17.6	29.4	47.1	23.5
Boumouda	9	0.0	0.0	55.6	44.4	0.0	11.1	88.9	0.0	66.7	33.3	0.0
Djimbana	6	0.0	0.0	50.0	50.0	0.0	0.0	100.0	0.0	16.7	66.7	16.7
Djiredji	15	0.0	0.0	53.3	46.7	0.0	0.0	100.0	0.0	80.0	20.0	0.0
Kinthiengrou	6	0.0	0.0	66.7	33.3	0.0	0.0	83.3	16.7	16.7	50.0	33.3
Medina	4	0.0	0.0	75.0	25.0	0.0	0.0	100.0	0.0	50.0	50.0	0.0
Same	8	0.0	0.0	62.5	37.5	0.0	0.0	62.5	37.5	12.5	50.0	37.5
Samirong	20	0.0	0.0	50.0	50.0	0.0	15.0	85.0	0.0	60.0	40.0	0.0
Sindina	14	0.0	0.0	50.0	50.0	0.0	0.0	78.6	21.4	42.9	21.4	35.7
Tingtingkom	8	0.0	0.0	75.0	25.0	0.0	0.0	87.5	12.5	62.5	25.0	12.5
Total	136	0.0	0.0	52.2	47.8	0.0	3.7	89.0	7.4	47.8	37.5	14.7

Table 9 - Shannon-Weaver Diversity indices of individual character and over geographical zones

VALLEY	50% FLOWERING H’	80% MATURITY H'	PLANT HEIGHT H’	TILLER H’	PANICLE LENGTH H’	MEAN H’ ± SD
Badiary	0.24	0.00	0.63	0.40	0.00	0.25 ± 0.27
Balmadou	0.74	0.74	0.94	0.37	0.46	0.65 ± 0.23
Bambali	0.82	0.71	0.96	0.42	0.42	0.67 ± 0.24
Boumouda	0.60	0.59	0.58	0.43	0.32	0.50 ± 0.12
Djimbana	0.63	0.63	0.79	0.43	0.00	0.50 ± 0.30
Djiredji	0.60	0.63	0.46	0.43	0.00	0.42 ± 0.25
Kinthiengrou	0.63	0.63	0.92	0.40	0.41	0.60 ± 0.21
Medina	0.35	0.34	0.63	0.35	0.00	0.33 ± 0.22
Same	0.56	0.56	0.89	0.41	0.60	0.60 ± 0.18
Samirong	0.51	0.40	0.61	0.43	0.38	0.47 ± 0.09
Sindina	0.77	0.77	0.97	0.43	0.47	0.68 ± 0.23
Tingtingkom	0.67	0.78	0.82	0.35	0.34	0.59 ± 0.23
Average	0.59	0.57	0.77	0.40	0.28	0.52 ± 0.21

H’ Shannon-Weaver diversity index, SD standard deviation

Discussion

The assessment of phenotypic variability of rice landraces and farmers’ traditional cultivars adapted to local conditions is an important aspect giving information on their characteristics which could be useful for plant breeding and improvement of elite varieties (Sow *et al.*, 2013; Sanni *et al.*, 2016).

This study was carried out to contribute to the preservation and the valorisation of local rice germplasm from Casamance region. It showed that rice landraces cultivated in Casamance are a useful reservoir of genetic diversity. They were already used in the past for improving introduced elite varieties. Thus, in the late sixties and seventies, some dwarf varieties belonging to subspecies *indica* (Taichung Native1, IR8 and I Kong Pao) were crossed with farmers’ traditional ecotypes from Casamance to improve their adaptability to upland and lowland local ecologies. These breeding programs led to the development and release of some popular varieties named Collection of Djibelor, represented by the DJ series (DJ 684-D, DJ 346-D, DJ 11-509, DJ 12-519 etc.) and the Collection of Sefa (Se 302G, Se 314G etc.) (ISRA-Djibelor, 1978; 1980). Moreover, accessions of *O. glaberrima* (Agnoun *et al.*, 2012), originated from this region were identified for resistance to several biotic and abiotic constraints and also used as parent in many breeding program (Semagn and Ndjiondjop, 2007; Sahrawat and Sika, 2002; Cherif *et al.*,2009; Agnoun *et al.,* 2012).

The 136 accessions evaluated in this study were collected in 29 villages in the Middle Casamance region which counts for about 36.000 ha of valleys suitable for rainfed rice cultivation (ANSD/SRSD, 2015). The agro-morphological characters assessed were mainly based on local farmers’ preferred traits including plant height, growth cycle referring to flowering and maturing periods, panicle length and number, tillering ability, grain yield and resistance to insect attack. Taking into consideration local farmers’ needs and preferences would help to prioritize and guide efficient breeding programs and facilitate further adoption of the new performing genetic material adapted to local ecologies (Ngailo *et al.*, 2015).

Experimental conditions of a pH around 6 and an electro-conductivity of 1 dS m^{-1} did not affect the performance of tested genotypes. In fact, rice plant development is estimated to be normal under pH conditions ranging from 4 and to 8 (Montcho *et al*., 2013) and an electro-conductivity less than 3 dS m^{-1} (Bimpong *et al*., 2014).

The agro-morphological characterization showed a significant variation among accessions for all the traits assessed. Local landraces were mainly semi dwarf to intermediate type and early maturing. The collection was also characterized by a low to moderate tillering ability and medium panicle length. Some accessions showed similar yield performance than improved upland (DJ 11-509) or lowland varieties (WAR 77, DJ 684-D, BG 90-2). However, in addition to their characteristic of tall and late maturing, rice landraces are generally known as low yielding genotypes (Sow *et al*., 2013; Sanni *et al*., 2016). This can suggest that the collection might include old introduced improved varieties considered over time as local or traditional cultivars. Nevertheless, the presence of such variability indicates that there is a continuous manipulation shaping local genepool and providing an opportunity for *in situ* conservation of local biotypes and maintenance of agro-ecological environments (Manzelli *et al*., 2005). The late flowering accessions represented 22% of the collection and was mainly intermediate to tall plant type. A satisfactory level of resistance to insect attack was noticed for most of accessions.

Phenotypical variation was explained by three principal components accounting for 84% of the total variation. The distribution of the accessions appeared to be associated with the ecologies where they were collected as demonstrated by the ranking position of control varieties. Indeed, based on their agro-morphological characteristics, 48% of the accessions seem to be more adapted to upland ecology, 45% to hydromorphic conditions while 7% presented the characteristics of typical lowland varieties. As noticed during the collect, most of the accessions follow farmers' predefined ecologies. This distribution agrees with Sow et al. (2013) arguing that farmers assign rice accessions to their appropriate ecologies.

Shannon-Weaver diversity indices of pooled characters over geographical zones showed a relatively low variation in some valleys, notably in Badiary, Medina Souané and Djiredji. Sindina, Balmadou and Bambali valleys had the highest diversity index. An average overall mean diversity of 0.52 was observed in the germplasm collection which could be explained by the fact that farmers may share germplasm within and among villages and valleys. The involvement of farmers to work with NGOs and research institutes results however in the introduction of new agricultural practices and technologies including improved high yielding varieties which could explain the relative high diversity observed in some valleys. On the other hand, farmers, to meet their needs, act as main force in the dynamic of crop populations providing opportunities for hybridization among interfertile landraces and/or exchange of genetic material, thus actively contributing to landrace adaptation and evolution (Manzelli *et al*, 2005). Moreover, the introduction of improved and promising varieties has not always provided the desired results in terms of diffusion and adoption in small scale farming systems due, often, to their no-adaptation to farmers' needs, preferences and conditions (Roling *et al*., 2004).

The accessions classified as *O. glaberrima* were identified based on some slight morphological characteristics as described by Linares, 2002, Agnoun *et al*., 2012. Further characterization including the use of molecular markers are therefore needed to confirm the accessions identified as *O. glaberrima*. The number of accessions classified as *O. glaberrima* were fare lower than the ones of *O. sativa*. This result was expected because, as many countries in West Africa, the introduction of improved Asian rice varieties lead to the regression of the cultivation of glaberrima rice (Linares, 2002; Teeken *et al*., 2012). The tradition of cultivating

African rice is however preserved by some farmers particularly in the Diola ethnicity both for its taste and use in ritual ceremonies (Linares, 2002; Agnoun *et al.*, 2012).

Conclusion

To our knowledge, this study is the first attempt in estimating phenotypical variability on traditional rice germplasm from Middle Casamance. It aims at recognizing the importance of agricultural biodiversity as a strategic resource in subsistence agricultures.

The findings from the agro-morphological characterization provide a baseline about the diversity observed in the collection in relation with the geographical zones. The phenotypic data recorded on key traits based on farmers' preferences and perceptions can allow for an efficient and sustainable use of this genetic diversity. The definition of farmer-oriented *in situ* conservation and improvement programs could lead to stabilize and increase local rice production. Specifically, the possibility to know and exploit local rice landraces can be particularly useful in developing varieties which meet farmer's needs and adapted to heterogeneous environmental conditions.

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