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Genetic resources of black rice (*Oryza sativa* L.) in West Java Indonesian for potential breeding material

Tita Kartika Dewi[1*], Tati Nurmala[2], Ruminta[3], M. Djali[4]
[1] Department of Agrotechnology, Faculty of Agribusiness and Agricultural Engineering, Subang University, Indonesia
[2, 3] Department of Agronomy, Faculty of Agriculture, Padjadjaran University, Indonesia
[4] Department of Food Industry Technology, Faculty of Agricultural Industrial Technology, Padjadjaran University, Indonesia

Abstract
Black rice has a lot of superiorities such as superior of specific location, anthocyanin content, and diseases and bugs resistant. Characterization aims to investigate the description of black rice plant characters that until today have been identified in detail, especially in West Java. Materials used in this research are several local cultivars of Black Rice from various areas in West Java [Tasik; Garut; Bandung (Soreang and Arcamanik); Subang (Cibeusi, Cipunagara, and Blanakan); Indramayu; and Bogor]. The observation was performed to identify the morphology characters namely leaves, stems, tassel, and grain. The data of morphology and agronomy characters are displayed based on its observation and assessment following Standard Evaluation System (SES) for Rice. The result showed the variation of morphological character of several cultivar of West Java Black Rice. From clustering process, there was found that West Java Black Rice can be categorized into four groups or clusters.

Keywords: agronomy, character, morphology, West Java

1. Introduction
West Java has countless natural resources, one of them is the existence of various species of local rice. Among those plant species there are several varieties of local rice plants, such as Black Rice with the excellent natures that cannot be found in other varieties [1]. This diversity of local West Java rice is a valuable basic capital to develop the agriculture in the food corps sector to support the national food self-sufficiency program. Additionally, this local West Java black rice becomes one of the germplasm resources of diversity of rice genetic in Indonesia and is expected to support the development of functional foodstuff.
The existence of local rice depends only on what have been done by local community and its genetic resource has not been conserved properly to protect the varieties from extinction and genetic erosion [2]. Furthermore, the utilization of those local rice varieties in the program of rice plant breeding in order to produce a new excellent variety has not been optimized [3].
The great number of rice cultivar and variety creates difficulty in differentiate it. Therefore, the categorization of that variety and cultivar is needed using numeric taxonomy. In the numeric taxonomy it is usually executed using phonetic approach [4, 5]. The used approach can be in the form of morphological characters, anatomy, chemical, sitology, isozyme, or DNA [6, 7]. However, the morphological characters and anatomy are the two most chosen characters in taxonomy research. The morphological characters that are usually used as the differentiators are the plant height, the number of productive buds, the stem color, the leaf color and surface, the number of grains in one tassel, the color and shape of grain.[8] In addition, disposal character can differentiate rice cultivar. [9]
Every cultivar of local rice may have similarity and difference on its characters. The existence of whether similarity or difference can be utilized to reveal how close the genetic relationship among rice cultivars. The more identical the closer the genetic relationship. On the contrary, the more different the more distant the genetic relationship. The same character categorization is the foundation of classification. The local black rice cultivar identification in Indonesia is extremely necessary to understand the specific character so that we can benefit from the potential that is possessed and eliminate the unwanted character for the variety betterment program. Characterization is the activity in order to identify the critical nature that can have economic value or something can distinguish from related varieties. Data collection about this cultivar is incredibly crucial to gain various information about that cultivar. The description regarding one cultivar can ease the search of information about the cultivar when it is needed as the genetic resource in the process of breeding. The objective of this research is to characterize the West Java Local Black Rice.

2. Materials and Methods
2.1 Plant Preparation
The experiment has been conducted for six months from February to August 2017 to identify and characterize 31 accessions of local black rice from several districts in West Java namely from Tasik, Garut, Soreang-Bandung, Arcamanik-Bandung, Cibeusi-Subang, Cipunagara-Subang, Blanakan-Subang, Indramayu and Bogor. The selected seeds of 31 accessions of local black rice were immersed for 24 hours and then they were growth in the germinator for 14

days. After 14 days, theye were transplanted to plastic pot containing rice field soil and manure (1:3/v:v) with flooding water.

2.2 Plant Characterization

Characterization was performed both in the time of generative and vegetative. The data in the both morphology and agronomy characters are displayed based on the assessment and observation following Standard Evaluation (SES) for Rice. From 48 characters registered in the guidance, it is selected 22 observed agronomy and morphology characters that are: agronomy characters (plant height, loss, flock, plant age, number of productive buds, number of contained grain per tassel, 100 grains weight and yield) and morphology characters (stem corner, leg color, leaf ear color, leaf tongue color, leaf sheet color, leaf surface, leaf corner, leaf length, leaf width, flag leaf corner, tassel length, grain shape, and grain color).

2.3 Clustering of Morphology Characteristics

To reveal the variability and the genetic relationship of black rice germplasm as the product of inventory process, clustering analysis was conducted to analyze the morphology characteristic. Clustering analysis was performed in complete linkage using distance coefficient Euclidean.

2.4 Statistical Analysis

The data were analyzed and displayed visually in the form of dendrogram of genetic relationship among black rice accessions using SPSS Var 16 data program.

3. Results

Based on the investigation, there were found 31 accessions of black rice germplasm that are distributed in different location. Nine accessionwere found in Subang, namely Cibeusi, Blanakan-1, Blanakan-2, Blanakan-3, Blanakan-4, Cipunagara-1, Cipunagara-2, Cipunagara-3 and Cipunagara-4; six accessions were found in Bandung, namely Arcamanik-1, Arcamanik-2, Arcamanik-3, Arcamanik-4, Soreang-1 and Soreang-4; five accession were found in Bogor, namely Bogor-1, Bogor-2, Bogor-3, Bogor-4 and Bogor-5; six accessions were found in Indramayu, namely Indramayu-1, Indramayu-2, Indramayu-2, Indramayu-3, Indramayu-4, Indramayu-5, and Indramayu-6; one accession was found in Tasikmalaya, namely Tasik-1;and four accession were found in Garut namely Garut-1, Garut-2, Garut-3 and Garut (Table 1).

Table 1: Clustering of West Java Black Rice Lines using the Method Between Group Linkage (Agglomeration Schedule)

S	Cluster Combined		Co	Stage Cluster First Appears		Next Stage
	C-1	C-2		C-1	C-2	
1	2	20	0.968	0	0	16
2	8	18	1.022	0	0	10
3	13	14	1.040	0	0	20
4	21	22	1.051	0	0	9
5	29	30	2.092	0	0	17
6	6	16	2.232	0	0	10
7	27	28	2.577	0	0	19
8	19	23	2.959	0	0	16
9	7	21	3.339	0	4	14
10	6	8	3.986	6	2	15
11	4	5	4.413	0	0	13
12	12	24	4.625	0	0	20
13	4	25	5.020	11	0	21
14	7	9	5.674	9	0	18
15	6	15	6.654	10	0	18
16	2	19	6.973	1	8	19
17	29	31	7.319	5	0	22
18	6	7	8.233	15	14	21
19	2	27	8.583	16	7	22
20	12	13	9.531	12	3	25
21	4	6	10.046	13	18	23
22	2	29	10.935	19	17	24
23	4	26	11.932	21	0	24
24	2	4	16.415	22	23	26
25	11	12	19.695	0	20	26
26	2	11	24.977	24	25	27
27	2	3	27.907	26	0	28
28	2	10	37.600	27	0	29
29	2	17	47.218	28	0	30
30	1	2	135.772	0	29	0

(S: Stage; C: Cluster; Co: Coefficients), Annotation: Clustering analysis result through Between Group Linkage Method using SPSS Var. 16 Statistic Program.

Based on the clustering analysis it is found that there are several groups of objects that is following the characteristic

similarity possessed by the plants. The objects that have similar closeness are placed in the same cluster, so that the produced clusters have high internal homogeneity and external heterogeneity. The result of cluster analysis on the strain of black rice in West Java through the process of Clustering Method Between Group Linkage is displayed in Figure 1.

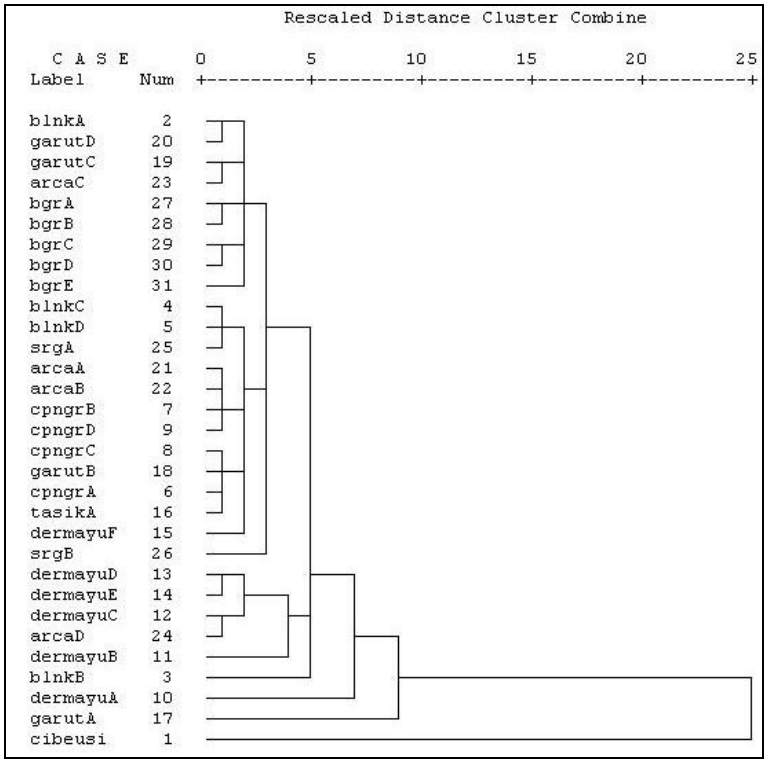

Fig 1: Dendrogram The Linkage among Groups

From the Euclidean distance among assessed variables (Table 1), thereare categorized hierarchically into two stages. Stage 1: was form one cluster, consisted of two strains, namely strain 2 (Blanakan-1) and strain 20 (Garut-4) with the Euclidean distance (coefficient) of 0.968. Then in the next stage was seen the number of 16. In the line of 16 (stage 16), there was observed of starin 2 (Blanakan-1) that was formed the clusted with the strine of 8 (Cipunagara-3). Stage 2: there was formed a cluster of strain 8 (Cipunagara-3) and strain 18 (Graut-2) and continued to the next stage of 10. It was happened until the last stage. Based on the agglomeration process, the number of cluster and the members that are formed as shows in Table 2.

Table 2: Cluster Membership

Case	4 - C	3 - C	2 - C
1: cibeusi	1	1	1
2: blanakan-1	2	2	2
3: blanakan-2	2	2	2
4: blanakan-3	2	2	2
5: blanakan-4	2	2	2
6: cipunagara-1	2	2	2
7: cipunagara-2	2	2	2
8: cipunagara-3	2	2	2
9: cipunagara-4	2	2	2
10: indramayu-1	3	2	2
11: indramayu-2	2	2	2
12: indramayu-3	2	2	2
13: indramayu-4	2	2	2
14: indramayu-5	2	2	2
15: indramayu-6	2	2	2
16: tasik-1	2	2	2
17: garut-1	4	3	2
18: garut-2	2	2	2
19: garut-3	2	2	2

20: garut-4	2	2	2
21: Arcamanik-1	2	2	2
22: Arcamanik-2	2	2	2
23: Arcamanik-3	2	2	2
24: Arcamanik-4	2	2	2
25: soreang-1	2	2	2
26: soreang-2	2	2	2
27: bogor-1	2	2	2
28: bogor-2	2	2	2
29: bogor-3	2	2	2
30: bogor-4	2	2	2
31: bogor-5	2	2	2

(C: Cluster), Annotation: Clustering analysis result through Between Group Linkage Method using SPSS Var. 16 Statistic Program.

4. Discussion

Black rice characteristics and clustering was conducting to find out the description of the characteristics of the local black rice plants in West Java and assess the similarity between the collection of black rice accessions from West Java. There are found that a quite large variation of morphological characteristic among investigated germplasms of black rice, such as the grain color, grain-end color, and color of rice husk. The grain color character was varied from yellow, purple, yellowish-brown, to brownish-black; the grain-end color was varied from straw yellow, yellow, brown, and black, also the identical thing was found in the color of rice husk which is varied from whitish-black, black with white rice base, and reddish-black. The morphological characters of grain can be used to differentiate the characteristics among local rice such as shape, size, surface, surface color, condition of surface-end, tail in surface-end (the existence, length and color), length of stem, color of stem, and grain loss. Grist showed that there are three types of grain shape namely rounded, medium, and thin.[10] The characters of shape, size, surface, and loss of grain can be utilized to differentiate among local rice cultivars in West Java. Meanwhile, grain shape, color, and loss of grain can be used to differentiate superior rice [8].

Morphological diversity among black rice germplasm is the influence of environment and genetic factor. Sitompul and Guritno state that characteristic appearance of plants is controlled by its genetic nature under the influence of environmental factors [11]. Every cultivar of investigated local rice may have similarity and difference on its characters. The existence of whether similarity or difference can be utilized to reveal how close the genetic relationship among rice cultivars. The more identical the closer the genetic relationship. On the contrary, the more different the more distant the genetic relationship. The same character categorization is the foundation of classification.

5. Conclusions

Taking into account the aforementioned characterization / clustering, description of properties of West Java Black Rice plants can be categorized into four groups / clusters that are:

a. Cluster 1 with Cibeusi strain as its member.
b. Cluster 2 with Blanakan-1,2,3,4, Cipunagara 1,2,3,4, Indramayu-1,2,3,4,5,6, Tasik-1, Garut-2,3,4, Arcamanik-1,2,3,4, Soreang-1, 2, Bogor-1,2,3,4 dan Bogor-5 strains as it members.
c. Cluster 3 with Indramayu-1 strain as its member.
d. Cluster 4 with Garut-1 strain as its member.

6. Acknowledgments

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