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Some characters appearance of unhulled rice of M6 of local rice mutant lines in South Kalimantan, Indonesia

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Abstract

Selection of mutant lines have been done during the last few years and have been generating some lines that shorter-lived and have higher yields than local rice varieties. The purpose of this study was to evaluate some characters appearance of unhulled rice of M6 of mutant lines. The experiment has been carried out in Sungai RangasHambuku Village, Martapura Barat District, Banjar Regency, South Kalimantan Province, Indonesia based on Randomized Block Design that consisted of 50 of M6 mutant lines and 3 controls with 2 replication. The variables analyzed were length and width of unhulled rice, ratio of length and width of unhulled rice, and amylose content of rice that followed by least significant difference (LSD) test if the analysis of variance significantly different. Result of this research showed that all lines classified as slender (length/width ratio of unhulled rice ranged from 3.73 - 4.99), but no lines that equivalent to slenderness of Cantik local variety (L/W = 5.58). Amylose content of rice of mutant lines ranged from a low - very high, namely 3.23% - 29.63%. The amylose content of lines were smaller, equal, or lower than Bestari, Inpara-2, and or Cantik.

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Introduction

According to Djaenudin (2008), tidal swamp land area reached 25.29 million ha. Tidal swamp land area in Indonesia spread in Sumatra, Kalimantan, Sulawesi and Papua. The development of tidal swamp land agriculture is a strategic step in answering the challenge of increasing agricultural production that is increasingly complex (Abdurachman and Ananto, 2000).

In general, rice breeding is to obtain a high yield, have resistance to environmental stress, have a good rice quality, and have the shorter plant age in order to support the sustainability of food security and self-sufficiency in rice. Short-lived and high-yielding varieties are an option in the development of tidal swamp land. However, the adoption of high-yielding rice varieties in South Kalimantan quite slow (Simanungkalit, 2004). Therefore, the development of improved varieties that are based on local varieties is important. Development of varieties based on local rice varieties aimed to obtaining new varieties that have local characteristics (adaptive, "pera", slender) but the age of the plant becomes shorter.

Mutation breeding in rice intended as a method for creating a wide genetic variability, so it is possible the selection of materials available to select mutants with properties that wanted (Riwidiharso and Susanto, 1996). There are a variability of local rice varieties in South Kalimantan (Wahdah and Langai, 2009). The yield of local rice varieties are stable, low input, and slender grain shape that is preferred by farmers and consumers (Sulaiman *et al*., 1995; Sulaiman, 1997). Local varieties in plant breeding is widely used to source good qualities such as flavor of rice (Daradjat *et al*., 2008). Increasing the genetic variability of the plant population can be done with mutation induction by gamma rays (Michi and Donini, 1983; Duncan *et al*., 1995 in Herison *et al*., 2008).

Preference of farmers to local varieties because of its ease of cultivation, lack of inputs, higher selling prices, and the characteristics of rice that preferred. Characteristics of rice that preferred in South Kalimantan are slender and "pera" (Wahdah and Langai, 2010). There are variability in some characters of South Kalimantan local rice (Wahdah and Langai, 2012).

There are variability of seed viability of some local varieties that irradiated with gamma rays (Wahdah and Langai, 2014). The selections of mutant lines that have done the last few years have been generated lines that are short-lived (Wahdah and Langai, 2016).

Plant age of M6 lines varies among lines and shorter than the Siam Unus, Pandak, Bayar Palas, Lemo Kwatik, and the Lakatan Gadur that according Khairullah *et al*. (2006) have the plant age 291, 305, 305, 272, and 295 days after seeding (DAS) respectively. Another disadvantage of local rice varieties are low yield. According Noorsyamsi *et al*. (1984) the average of yield of rice in the swamp area is 1.0 t ha^{-1}- 2.5 t ha^{-1}. Wirosoedarmo and Apriadi (2008) reported the results of local rice in South Sumatra, Indonesia are 1-2 t ha^{-1}.

The yields of the M6 mutant lines ranged from 3.83 t ha^{-1} – 6.03 t ha^{-1}, while the yield of Bestari, Inpara-2, and Cantik were 5.06 t ha^{-1}, 6.79 t ha^{-1} and 3.09 t ha^{-1}respectivily (Wahdah *et al*., 2016).

Pandan Putri is a rice mutant of Pandan Wangi whose age is almost 2 months shorter than Pandan Wangi with taste and aroma are similar to Pandan Wangi (Ismachin and Sobrizal, 2006). By this research, we expected there are some lines that short-lived, selender, and pera (high amylose content in the rice).

Therefore, it is necessary to evaluate some of the characteristics of unhulled rice namely the unhulled rice length, unhulled rice width, ratio of length/width of unhulled rice, and rice amylose content.

Materials and methods

Cultivation of rice was carried out for 5 months in Sungai Rangas Hambuku Village, Martapura Barat District, Banjar Regency, South Kalimantan Province, Indonesia.

Materials

The materials used in this experiment were 50 of M6 of mutant lines and three controls (Bestari and Inpara-2 as high yielding varieties and Cantik as a local variety), Phonska as N,P, K fertilizers, Urea as nitrogen fertilizer, insecticide, fungicide, moluscocide, and herbicide.

Experimental design

The research was conducted by Randomized Block Design with 2 replication. Factor to be tested was genotypes (50of M6 lines and 3 controls).

Cultivation techniques

Seeding has been done per row per treatment (50 of M6 mutant lines plus 3 controls). Transplanting seedling has been done when the seedlings were 25 days after sowingin each plot measuring 3 m × 4 m. Spacing has been used is 25 cm × 30 cm based on 2 : 1 of "legowo" planting system (two rows planted and one row unplanted). Fertilization of 300 kg Phonska ha^{-1} given at the age of 4 weeks after planting. Fertilization of 100 kg Urea ha^{-1} given twice (50 kg ha^{-1} at 2 weeks after planting and 50 kg ha^{-1} at 6 weeks after planting). Pests control have been done with pesticides as needed, while the weeds control is done manually and with herbicide according to recommended dosage. Harvesting is done if more than 90 percent of the panicles have matured.

The variables observed

The variables were observed were length, width, ratio of length and width of unhulled rice, and amylose content of rice.

Analysis of variances were conducted based on linear additive model , namely:

$Y_{ij} = \mu + B_i + L_j + €_{ij}$;

Y_{ij} = the appearance of a variable that affected by the i-th of blocks and the j-th of lines

μ = mean value

B_i= influence of i-th of blocks

Lj= influence of j-th of lines

€ ij = effect of random error

If the results of analysis of variance showed a significant effect at 5 % of alpha level, then followed by Least Significant Difference (LSD) test at 5 % of alpha level (Steel and Torrie, 1994).

Results and discussion

Based on variance analysis on Table 1, seen highly significant effect of genotype on unhulled rice length, ratio of length/width of unhulled rice, and rice amylose content, and significant effect on unhulled rice width. Results of Least Significant Different (LSD) test can be seen on Table 2. Recapitulation of percentage of different lines to each control by LSD test at 5% of alpha level for unhulled rice length, unhulled rice width, and the ratio of length/width of unhulledrice can be seen in Table 3. Whereas the maximum and minimum values of M6 of mutant lines and average values of characters of Bestari, Inpara-2, and Cantik can be seen in Table 4.

Length of unhulled rice

Based on Table 2, there are 2 lines (4 %) that have a length of unhulled rice longer than Bestari variety and as many as 48 lines (96 %) were equivalent. The lines that have alength of unhulled rice longer than Bestari variety were G-1 and G-8 lines, namely 11.09 mm and 10.15 mm respectively. The length of unhulled rice of Bestari variety was 9.54 mm. The length of unhulled rice of Bestari variety significantly longer than Inpara-2 and Cantik varieties, namely 8.75 mm and 7.44 mm respectively. All lines (100 %) of M6 have a length of unhulled rice longer than Inpara-2 and Cantik. The length of unhulled rice of Cantik variety was shortest among all lines, Bestari, and Inpara-2.

According to Silitonga *et al.* (2003) and Irawan and Purbasari (2008), the length of unhulled rice classified into 4 groups, ie very long (> 7.50 mm), long (6.61 mm - 7.50 mm), medium (5.51 mm - 6.61 mm) and short (<5,51 mm). Thus, although there are differences in the average of length of unhulled rice among genotypes (all lines and all controls) were tested, all genotypes were classified very long ie> 7.5 mm. Range of the length of unhulled rice of M6 lines in this experiment were 8.75 mm - 11.09 mm, while the length of unhulled rice of Bestari, Inpara-2, and

Cantik respectively 9.54 mm, 8.75 mm, and 7.44 mm. According Wahdah and Langai (2009), the length of unhulled rice of local varieties at South Kalimantan tidal swamp are long - very long, ie ranged from 7.14 mm - 9.98 mm, while according to Khairullah *et al.* (2006) relatively very long (7.7 mm-8.8 mm).

Table 1. Recapitulation of variance analysis of unhulled rice length, unhulled rice width, ratio of grain length: grain width, and rice amylose content.

Characters	df	F_{values}	$F_{0.05}$	$F_{0.01}$	CV (%)
Unhulled rice length (L)	52	5.89**	1.596	2.166	0.61
Unhulled rice width (W)	52	1.77*	1.596	2.166	0.01
L/W (√x + 10)	52	40.07**	1.596	2.166	0.24
Amylose content (√ X)	52	7.25**	1.596	2.166	7.08

** = highly significant, * = significant.

The significant difference of unhulledrice grain length between genotypeshas been reported by Vanaja and Babu (2006); Thayumanavan *et al.* (2010); Subbaiah *et al.*(2011); Dhanwani *et al.*(2013). The range of grain length of 56 rice genotypes tested were 7.35 mm – 10.11 mm (Vanaja and Babu, 2006) Yadav *et al.* (2007) reported that the range of grain length of 6 rice genotypeswere 5.85 mm – 8.25 mm.The significant difference in the analysis of variance is an indication of a broad genetic variability (Thayumanavan *et al.*, 2010). According to Dhanwani *et al.* (2013), the heritability of rice grain length is high.

Table 2. Mean of unhulled rice length, unhulled rice width, length/width ratio of unhulled rice, and rice amylose content of 53 genotypes (consist of 50 of M6 of mutant lines and 3 of controls).

Lines/Control	Unhulled rice length (mm)	Unhulled rice width (mm)	Length/width ratio of unhulled rice	Amylose content of rice(%)
G-1	9.68	3.55	3.73	10.28 (3.21)
G-2	11.09	3.46	3.94	6.68 (2.58)
G-3	9.88	3.46	3.87	8.06 (2.82)
G-4	9.64	3.49	3.80	10.12 (3.18)
G-5	9.88	3.46	3.87	3.23 (1.78)
G-6	9.70	3.47	3.85	3.61 (1.90)
G-7	9.84	3.47	3.86	22.90 (4.78)
G-8	10.12	3.47	3.87	23.96 (4.89)
G-9	9.91	3.46	3.88	4.88 (2.20)
G-10	9.87	3.46	3.88	12.03 (3.37)
G-11	9.51	3.46	3.85	11.93 (3.39)
G-12	9.64	3.46	3.85	4.29 (2.06)
G-13	9.54	3.47	3.84	7.47 (2.73)
G-14	9.92	3.46	3.88	6.15 (2.46)
G-15	9.76	3.46	3.86	6.73 (2.58)
G-16	9.94	3.46	3.87	7.16 (2.67)
G-17	9.76	3.47	3.85	4.61 (2.14)
G-18	9.72	3.47	3.85	11.03 (3.32)
G-19	9.59	3.47	3.84	7.63 (2.76)
G-20	9.73	3.43	3.94	8.43 (2.89)
G-21	9.61	3.47	3.84	9.75 (3.11)

G-22	9.47	3.46	3.85	17.44	(4.18)
G-23	9.54	3.47	3.84	8.69	(2.77)
G-24	9.57	3.47	3.84	7.85	(2.79)
G-25	9.56	3.46	3.85	6.47	(2.53)
G-26	9.77	3.47	3.85	23.85	(4.88)
G-27	9.59	3.47	3.84	29.15	(5.38)
G-28	9.73	3.47	3.85	11.50	(3.36)
G-29	9.66	3.47	3.85	10.23	(3.18)
G-30	9.64	3.46	3.86	6.04	(2.46)
G-31	9.65	3.46	3.86	6.84	(2.60)
G-32	9.76	3.46	3.86	11.45	(3.37)
G-33	9.62	3.46	3.85	7.63	(2.75)
G-34	9.79	3.47	3.86	4.93	(2.20)
G-35	9.88	3.46	3.87	6.15	(2.41)
G-36	9.75	3.47	3.85	7.47	(2.73)
G-37	9.83	3.47	3.86	29.63	(5.44)
G-38	9.65	3.45	3.89	10.87	(3.29)
G-39	9.41	3.46	3.85	9.54	(3.09)
G-40	9.92	3.47	3.86	14.26	(3.71)
G-41	9.52	3.46	3.85	11.98	(3.45)
G-42	9.76	3.46	4.99	5.09	(2.25)
G-43	9.60	3.46	4.80	24.33	(4.92)
G-44	9.63	3.46	4.84	13.04	(3.60)
G-45	9.78	3.47	4.87	7.42	(2.55)
G-46	9.39	3.46	4.74	25.28	(5.03)
G-47	9.78	3.46	4.89	6.41	(2.53)
G-48	9.87	3.46	4.94	7.85	(2.80)
G-49	9.56	3.47	4.71	7.37	(2.71)
G-50	9.80	3.46	4.91	7.85	(2.80)
Bestari	9.54	3.47	4.74	6.31	(2.50)
Inpara-2	8.75	3.46	4.38	12.99	(3.59)
Cantik	7.44	3.37	5.58	9.65	(3.07)
$LSD_{0.05}$	0.49	0.04	0.20	7.08	
CV (%)	0.61	0.01	0.24	0.21	

Width of unhulled rice

Based on LSD test of the unhulled rice width (Table 2), there is 1 of lines (2%) have a width of unhulled rice wider than Bestari and Inpara-2 varieties, namely G-1 (3.55 mm), 98 % of lines were equivalent, and none of lines that has a narrower a width of unhulled rice. All lines have a width of unhulled rice wider than Cantik variety. The range of unhulled rice width of M6 of lines were 1.34 mm – 2.64 mm. The width of unhulled rice of Bestari, Inpara-2, and Cantik varieties were 3.47 mm, 3.46 mm, and 3.37 mm respectively (Table 4).

Putra *et al.* (2010), classifying the width of unhulled rice into 3 groups, ie narrow, if the width of unhulled rice is <1 mm, medium, if the width of unhulled rice ranging between 1 mm -2 mm, and wide, if the width of unhulled rice is > 2 mm. The width of unhulled rice of M6 mutant lines in this study ranging 3.43 – 3.55 (wide), while Bestari, Inpara-2, and Cantik as controls were 3.47 mm, 3.46 mm, and 3.37 mm respectively. Results of Ellya research (2013) on M3 mutant lines which are generation above the M6's mutant population in this research have width of unhulled rice which ranging between 2.18 mm -2.53 mm (wide).

South Kalimantan local rice varieties generally have a width of unhulled rice ranged between 1.7 mm - 1.9 mm (Khairullah *et al*., 2006). Wahdah and Langai (2009) reported that the average width of unhulled rice of local varieties in tidal swamp area of South Kalimantan was 1.23 mm. Thus, there are some lines of M6 mutant have width of unhulled rice equivalent to rice local varieties, but there are also wider than the local varieties.

Table 3. The percentage of lines that significantly different from each control by LSD test at 5% of alpha level for unhulled rice length, unhulled rice width, and ratio of unhulled rice length/width, and rice amylose content.

Compared with controls		Unhulled rice length	Unhulled rice width	Length and width ratio of unhulled rice	Rice amylose content
Bestari	<	0	0	0	14
	=	96	0	0	20
	>	4	100	100	66
Inpara-2	<	0	0	0	76
	=	98	98	0	8
	>	2	2	100	16
Cantik	<	80	80	100	54
	=	16	0	0	10
	>	4	20	0	36

Thesignifi cant difference of rice grain breadth (width) also reported by Vanaja and Babu (2006); Subbaiah *et al.*(2011); Dhanwani *et al.* (2013). Vanaja and Babu (2006) reported the grain breadth range of 56 genotypes were 2.56 mm – 3.76 mm while Yadav *et al.* (2007) reported that the range of grain breadth of 6 rice genotypes were 1.65 mm – 2.93 mm. The heritability of rice grain breadth was high (Dhanwani *et al.*,2013).

Length and width ratio of unhulledrice

Length and width (breadth) ratio is a measure of slenderness of unhulled rice. Based on the recapitulation in Table 3, there are 41 lines (82%) having a ratio of length and width of unhulled rice are smaller than Bestari variety, 8 lines (16%) are equivalent, and 2 lines (4%) are smaller than Bestari. There are 40 lines (80%) more slender and there are 10 lines (20%) more rounded than Inpara-2 variety. The Cantik variety most slender than all lines of M6 but all lines (100%) are slender.

The range of slenderness of the unhulled rice were 3.90-5.53, whereas Bestari, Inpara-2, and the Cantik varieties were 4.73, 4.37, and 5.52repectivily.

The ratio of length/width of unhulled rice (L/W or L/B) is a measure of slenderness of unhulled rice. According Sajak *et al.* (2012); Irawan and Purbasari (2008), slenderness categorized as slender if the L/W is > 3, medium if L/W ranging 2.1 - 3.0 and rounded if is < 2.1. Based on the classification, then all lines and all the controls are classified as slender (L/W> 3).

There are significant difference of L/B ratio of rice grain (Vanaja and Babu, 2006; Subbaiah *et al.* 2011; Dhanwani *et al.*, 2013). The range of grain length/breadth ratio of 56 rice genotypes tested were 1.95 - 3.85 (Vanajadan Babu, 2006); 1.99 – 4.39 in 6 rice genotypes (Yadav *et al.*, 2007). The change of L/B ratio of milled rice has been reported by Bughio *et al.* (2007), namely from 3.50 into 4.08 (17 %) in Shadap mutant, 2.61 into 3.46 (33 %) in Shua-92 mutant, 3.07 into 3.24 (17 %) in Khushboo-95, and 3.07 into 3.24 (6 %) in Sarsharmutant. Bari *et al.* (1981) reported that there are a number of lines mutant derived from IR-6 and IR-8 varieties that have L/B ratio greater than its parent. According to Dhanwani *et al.* (2013), the heritability of L/B ratio is high.

The weight of 1000 grains of unhulled rice represents the size of grain. According Sajak *et al.* (2012), weight of 1000 grains of unhulled rice classified as heavy if > 30 g, moderate if it is between 25-30 g, and light if < 25 g. Wahdah *et al.* (2016) reported that the weight of 1000 grains of unhulled rice of M6 between 21.7 g - 29.0 g (light - moderate). Related to the size of rice, Jumberi *et al.* (2008) suggested that two local varieties are very popular among the farmers of South Kalimantan are Siam Saba and Siam Mutiara each has an average weight of 1000 grains of unhulled rice were 17.6 g - 17.9 g and 17.6 g - 17.7 g. Based on the weight of 1000 grains of unhulled rice and L/W ratio, it is possible to selection grains that light (the weight of 1000 grains <25 g) and or slim (P/L > 3).

Table 4. The maximum and minimum values of characters of lines, average values of Bestari, Inpara-2.

Characters	Maximum values	Minimum values	Bestari	Inpara-2	Cantik
Unhulled rice length (mm)	11.09	7.44	9.54	8.75	7.44
Unhulled rice width (mm)	2.64	1.34	2.02	2.00	1.22
L/W of unhulled rice	5.53	3.90	4.73	4.37	5.52
Rice amylose content (%)	29.63	3.23	6.31	12.99	9.65

Amylose content of rice

There are 33 lines (66%) that have amylose content in rice greater than Bestari, 7 lines (14%) are lower, and 10 lines (20%) are equivalent. Compared with Inpara-2, there are 38 lines (76%) have a lower amylose content, 4 lines (8%) are equivalent, and 8 lines (16%) are higher. When compared with the Cantik local variety, then there are 27 lines (54%) are having a lower amylose content, 5 lines (10%) are equivalent, and 18 lines (36%) are higher.

The classification of rice based on the indices of amylose according Allidawati and Bambang (1989), namely sticky rice with amylose content is very low (<10%), fluffier rice(10-20%), fluffier medium rice (20-24%), and high amylose content ("pera"), namely > 25%, while Silitonga *et al.* (2003) classified the amylose content of rice into 6 groups, namely glutinous (waxy) if the amylose content <3.0%, very low if it is in the range 3.1% - 10.0%, low if 10.1% - 15.0%, medium if 15.1% - 20.1%, height if between 20.1% - 25.0%, and very high if it is in the range of 25.1 % - 30.1%.

The range of rice amylose content of M6 lines in this experiment was 3.23% - 29.63% (very low -very high), while Bestari, Inpara-2, and Cantik were 6.31% (very low), 12.99% (low), and 9.65% (very low) respectively.

Comparative of amylose content of mutant lines with its parent was reported by Bughio *et al.* (2007), namely from 30.10 in parent into 30.70 in Shadap mutant (2 %), 26.2 parent into 26.7 in Shua-92 mutant (2 %) , 19.81 in parent into 20.80 (5 %) in Khushboo-95 mutant (5 %), and 27.2 parent into 26.2 in Sarshar mutant (4 %). Vanaja and Babu (2006) reported that 56 genotype tested showed significant differences of rice amylose content. The range of amylose content of rice genotypes tested were 19.46- 36.8 %.

Rice texture of Bestari classified as fluffier rice (Indonesian Rice Research Institute, 2008a) as well as Inpara-2 (Indonesian Rice Research Institute, 2008b). Hairmansis *et al.* (2013) reported that the amylose content of rice which come from plant breeding selection results which directed to high amylose content of rice in his research have amylose content of 19.25% -28.63%. Inpara-2 in the description of variety had higher level of amylose in this experiment, namely 22.06% (Indonesian Rice Research Institute, 2008b). Somantri (1983) in Aliawati (2003) state that the ranges of amylose content of rice are between 1%-37%. Thus can be selected lines that pera or fluffier for tested in M7, caused the range of rice amylose content in this experiment was 3.23% - 29.63% (very low -very high).

Conclusion

None of M6 mutant lines which slenderness equivalent to Cantik local variety (L/W = 5.58), but all lines classified as slender (the range of L/W ratio of unhulled rice were 3.73 - 4.99).There are some lines that have amylose content of rice were smaller, equal, or lower than Bestari, Inpara-2, and or Cantik varieties. The amylose content of rice of M6 mutant lines ranged from low to very high (3.23% - 29.63%).

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