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Agro-morphological variation of pigmented rice landraces from South Sulawesi grown in a temperate glasshouse of New Zealand

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Abstract. The present study is aimed to compare the agro-morphological traits of 15 pigmented rice landraces and two modern rice varieties grown in the same environment. The selected rice were cultivated in a greenhouse of Massey University, New Zealand using a randomised complete block design with five replications. Data collection included agro-morphological observation and measurement. Quantitative data were analysed with analysis of variance, followed with Tukey HSD. Result showed considerable variability in seedling vigour, lodging, plant height, panicle exertion, panicle thresh ability, spikelet fertility, and maturity among the studied pigmented rice landraces. Panicle exertion and spikelet fertility indicate plant cope toward environmental change, which was performed better by many landraces than modern varieties. Variability in morphological characteristics was also recorded among tested varieties. Utilisation of the trait diversity can be useful for future breeding programs, which can lead to this crops' improvement, including pest and disease resistance and grain quality research.

1. Introduction

Several research on the diversity of local pigmented rice across Indonesia have been carried out [1,2,3]. In particular, the physicochemical properties of pigmented rice from South Sulawesi have been revealed [4,5], confirming the potential of such varieties for nutraceutical products. Therefore, pigmented rice is considered as speciality rice, which deserves a better premium price in both the domestic and export market.

The pigmented rice landraces possess the immense potential of most valuable genes which can be utilised in the current breeding program to evolve varieties that have not only high yield potential and grain quality of nutritional contents but also resistant to biotic and abiotic stresses. Systematic study and characterisation of such germplasm is not only crucial for utilising the appropriate attribute-based donors but also essential for protecting the exotic rice.

Morphological rice characteristics are less documented in Indonesia [3]; hence the diversity of rice germplasms remain untapped. Plant morphologies are the primary basis for the classification of plants or an interpretation of plant adaptability. Moreover, the link between the morphologies of plants and



productivity has resulted in the concept of a desirable plant type (idiotype) [6]. Hence, there is a necessity to do the process of collection, exploitation and evaluation of untapped germplasms [7].

The study was aimed to compare the variability of 15 pigmented rice landraces and two modern varieties using agro-morphological characters and to provide useful information to facilitate the choice of genitors for further crop improvement.

2. Materials and methods

2.1 Seed materials

Seeds of 16 traditional pigmented rice varieties were collected from four regencies in South Sulawesi of Indonesia, namely: North Toraja, Tana Toraja, Enrekang, and Jeneponto (table 1). The seed collection is a part of local variety exploration program organised by a team from the Plant Biosciences & Reproduction Biotechnology Laboratory, Faculty of Agriculture, Hasanuddin University). Seed samples were collected during the timeframe of June - November. Local varieties are named according to farmers' information. In this research, two modern variety were used as comparison, *i.e.* Inpari 24 and Ciherang.

Table 1. List of pigmented rice landraces used in the present study and their source of origin.

Variety	Grain colour and type	Sub-district/Regency	Altitude (m asl)
Ko'bo	Black	Balusu, North Toraja	900
Lotong	Black	Nanggala, North Toraja	780
Ambo	Black	Balusu, North Toraja	903
Barri Rarang	Red	Bengkelekila, North Toraja	836
Lallodo	Black	Balusu North Toraja	905
Lea	Red	Sangalla, Tana Toraja	791
Lotong Tanduk	Black	Nanggala, North Toraja	908
Maminyak	Red	Baraka, Enrekang	727
Mandoti	Red	Salukanan, Enrekang	789
Balan	Red	Baraka, Enrekang	536
Kamida	Red	Curio, Enrekang	847
Jambu	Red	Banua, Enrekang	607
Le'leng	Black	Turatea, Jeneponto	115
Bakka Eja	Red	Turatea, Jeneponto	115
Punu Eja	Red	Turatea, Jeneponto	153
Inpari24*	Red	Purchased from Indonesian Rice Research Center, West Java	
Ciherang*	White	Purchased from Regional Crop Seed Center of Maros, South Sulawesi	

* Modern variety (nationally released)

2.2 Timing and methods.

The study was conducted in Plant Growth Unit at Massey University, Turitea campus (40°22'40"S 175°36'48"E, elevation: 35 m above sea level) from December 2017 (summer) to July 2018 (winter). Rice plants for the preliminary research were cultivated in a controlled environment. The experiment was set up as a Randomised Block Design with five replicates. Potting mix media with slow released fertiliser was used. Drip irrigation was established in week 4, where each pot was placed in 9.6 L spout

bucket to achieve flooded/saturated condition. Fertilisation was done when leaf nitrogen status was assessed as low based on a Leaf Color Chart (score ≤ 3) (Rice Knowledge Bank, 2018). Peters® Professional Allrounder (N-P-K : 20-20-20+TE) at 1g/L was used as fertiliser, incorporating with drip irrigation (5 L per day).

2.3 Data collection and analysis.

The accessions were characterised according to Standard Evaluation System for Rice [8]. A total of seven agronomic traits data (table 2) and twelve of morphological characteristics (table 3) were collected. Simple statistical parameters *i.e.* mean and standard error were determined for the quantitative traits using statistical package SPSS 15.0. The data after compiling was statistically analysed using SPSS 15.0 and Tukey post hoc test (Honest Significance Difference) test was applied to test the significance of varietal differences. Pearson correlation was also used to investigate any association between parameters.

Table 2. Descriptions and scales/codes of agronomic traits to evaluate 15 pigmented rice landraces from South Sulawesi [8].

Descriptors	Observed phenotypic classes	Evaluation phase
Seedling /vegetative vigour (Vg)	1-extra weak;9-very weak vigorous;3-vigorous;5-normal;7-	Seedling vigour:2 Vegetative vigour:3
Tillering ability (Ti)	1-very high (>25);3-good(20-25);5-medium(10-19);7-low(5-9);9-very low (<5)	At growth stage 5
Plant height	1-semidwarf (lowland:<110cm;upland:<90cm);2-intermediate (lowland:110-130cm;upland(90-125cm);9-tall(lowland>130cm;upland:>125cm)	At growth stage 7-9
Panicle exertion	1-enclosed;3-partly exerted (panicle base is slightly beneath the collar of flag leaf blade);5-just exerted (panicle base coincides with the collar of the flag leaf blade);7-moderately well exerted (panicle base is above the collar of the flag leaf blade);9-well exerted(panicle base appears well above the collar of the flag leaf blade)	At growth stage 7-9 (near maturity)
Panicle threshability	1-difficult (less than 1%);3-moderately difficult (1-5%);intermediate (6-15%);7-loose (26-50%);9-easy(51-100%)	At growth stage 9
Spikelet fertility	1-highly fertile(>90%);3-fertile(75-89%);partly sterile(50-74%);highly sterile(<50% to trace);9-0%	At growth stage 9
Maturity	Number of days from seeding to grain ripening (85% of grains on panicle are mature)	At growth stage 9

Table 3. Morphological character descriptors used to evaluate 15 pigmented rice landraces from South Sulawesi.

Descriptions	Observed phenotypic classes	Evaluation phase
Leaf length	Actual measurement in cm of the widest portion of the leaf blade just below the flag leaf	At growth stage 6
Leaf blade colour	1-light green;2-green;3-dark green;4-purple tips;5-purple margins;6:purple blotch;7:purple	At growth stage 4-6
Ligule length	Enter actual measurement of ligules measured in mm from the base of collar to the tip. Sample size 5	At growth stage 4-5
Ligule colour	0-absent;1:white;2:purple lines;3:purple	At growth stage 4-5
Ligule shape	0-absent;1-acute to acuminate;2-cleft;3-truncate	At growth stage 3-4
Auricle colour	0-absent;1-light green;2-purple	At growth stage 4-5
Culm internode colour	1-green;2-light gold;3-purple lines;4-purple	At growth stage 7-9
Panicle length	Enter actual measurement in cm from panicle base to tip	At growth stage 8
Awning	0-absent;1-short and partly awned;5-short and fully awned;7:long and partly awned;9-long and fully awned	At growth stage: 7-9
Awn colour	0-awnless;1-straw;2-gold;3-brown;4-red;5-purple;6-black	At growth stage: 6
Grain width	Enter the actual measurement of width in mm as the distance across the fertile lemma and the palea in the widest point	At growth stage:9
Grain length/width ratio	Ratio of length in mm as the distance from the base of the lowermost sterile lemma to the tip (apiculus) of the fertile lemma/palea per width	At growth stage:9

3. Results and discussion

3.1 Agronomic traits

The assessed pigmented rice landraces showed considerable variability for the studied agromorphological characteristics. Seven agronomic traits comparison among studied varieties are presented in table 4.

3.1.1 Maturity. Number of days to achieve the maturity of rice seeds were in the range from 157 cm in Jambu to 215 cm in each variety of Lallodo and Mandoti. Modern varieties reached their maturity longer than expected, indicating a response to the environmental difference from their original habitat. Based on observation, plants with early maturity relatively have intermediate to semi-dwarf height, such as Balan, Bakka Eja, Inpari 24 and Ciherang. However, Jambu has early maturity but fall into the tall group.

Generally, the maturity days reached in this trial were longer than reported maturity in references [9,10] which under five months if grown in Toraja regions for some varieties like

Lallodo, Ambo, Mandoti and Lotong. Environmental factors such as light reception and temperature are some of the key factors.

3.1.2 Plant height. Plant height can be seen in three classes, namely, intermediate (Balan), semi-dwarf (Bakka Eja, Inpari, Ciherang) and tall type for the remaining tested varieties. 5.8% intermediate height, 17.6% semi-dwarf, 76.5% tall in height. Traditional rice varieties typically have tall stature whereas modern varieties/early maturing types that tend to be semi-dwarf. In general, most tested varieties grew higher than their common height in their origin of habitat. On the other hand, decreased height, earlier maturity, and longer panicles were recorded from upland rice of South Sulawesi under lowland cultivation [11]. Semi-dwarf rice plant is a primary target for improving the lodging resistance [12], but the drawback is, the canopy for photosynthesis and number of biomass are limited, and then decrease the grain yield [13]

3.1.3 Panicle exertion. There are four groups of panicle exertion, which are dominated by well exerted (58.8%), whereas partly exerted was shown in two modern varieties. Most traditional varieties such as Kobo, Lotong, Ambo, Ra'rang, Lotong Tanduk, Maminyak, Kamida, Jambu, Leleng and Punu Eja showed well-exserted panicles. In contrast, modern varieties have partly exerted panicles.

Panicle exertion is one of the important agronomic traits since it can be an indication of genetic defect caused by environmental or pest -disease attack [14]. Incomplete panicle exertion indicates cold injury at the reproductive stage, which lead to damaged grain yield as disease incidence raises [15]. The long and well-exserted panicles are the typical characters of upland rice panicles, as supported by [16], that upland traditional rice are adapted to harvesting and storage by panicles which are long and well exerted.

3.1.4 Threshability. Six traditional varieties and two modern varieties were observed to be in the easy category of threshability. Grain characteristics of the ease threshability group are awnless. In contrast, awned grains have intermediate to moderately difficult of threshability.

The result confirmed the threshability of awned genotypes is more difficult than awnless. Some farmers in Toraja prefer to have moderate-intermediate threshability of panicles to minimise losses during post-harvest, as they transport their stacks of panicles manually and store the panicles with the stalks, without shattering [17].

3.1.5 Seedling vigour. Seedling vegetative vigour fell into three groups: weak (Kobo, Lotong, Lotong Tanduk), normal (Lea, Kamida) and the remainder as vigorous (table 4). In general, most tested varieties, including modern varieties had acceptable seedling vigour.

3.1.6 Spikelet fertility. Our result only have two groups of spikelet fertility: partly sterile and highly sterile. Some traditional varieties perform better than the modern ones. Unexpected sterility of panicles may resulted from microclimate discrepancy in pot trial compared to the origin. Moreover there was disruption from the heat fan which caused dried panicle during anthesis.

The insufficient spikelet fertility in this research might also resulted from cold stress and low solar radiation during reproductive phase. According to [18], during reproductive period, low temperature-induced spikelet sterility, could be associated with the physiological response mechanisms in the pollen, anther, and stigma.

Table 4. Agronomic traits of pigmented rice landrace of South Sulawesi and two modern rice varieties for comparison

Variety	Maturity (days)	Plant height (cm)	Panicle exertion	Threshability	Seedling vegetative vigour	Spikelet fertility	Tillering ability
Kobo	195	Tall	Well exerted	Moderately difficult	Weak	Partly sterile	Medium
Lotong	202	Tall	Well exerted	Moderately difficult	Weak	Highly sterile	Medium Very high
Ambo	190	Tall	Well exerted	Intermediate	Vigorous	Partly sterile	
Ra'rang	195	Tall	Well exerted	Easy	Vigorous	Highly sterile	Good Very high
Lallodo	215	Tall	Moderately well	Intermediate	Vigorous	Highly sterile	
Lea	202	Tall	Moderately well	Easy	Normal	Highly sterile	Good Very high
L. Tanduk	195	Tall	Well exerted	Intermediate	Weak	Highly sterile	
Maminyak	190	Tall	Well exerted	Moderately difficult	Vigorous	Partly sterile	Good Very high
Mandoti	215	Tall	Moderately well	Moderately difficult	Vigorous	Highly sterile	Very high
Balan	170	Intermediate	Just exerted	Easy	Vigorous	Highly sterile	Very high
Kamida	198	Tall	Well exerted	Easy	Normal	Highly sterile	Very high
Jambu	157	Tall	Well exerted	Intermediate	Vigorous	Highly sterile	Very high
Le'leng	198	Tall	Well exerted	Intermediate	Vigorous	Partly sterile	Very high
Bakka Eja	160	Semidwarf	Just exerted	Easy	Vigorous	Highly sterile	Very high
Punu Eja	198	Tall	Well exerted	Easy	Vigorous	Partly sterile	Very high
Inpari 24	160	Semidwarf	Partly exerted	Easy	Vigorous	Highly sterile	Very high
Ciherang	160	Semidwarf	Partly exerted	Easy	Vigorous	Highly sterile	Very high

3.1.7 Tillering ability. The tillering ability of all varieties in this study appears to be medium (10-19 tillers per plant) to very high (more than 25 tillers/plant). The ideal planting space is 25 cm to allow the appropriate growth of tillers and minimise disease in humid condition. Most of the varieties showed a very high tillering ability while few varieties namely Maminyak, Lea and Ra'rang had good tillering ability and only Kobo as well as Lotong had medium tillering ability.

Similarly [10] reported significant difference in traits such as maturity, height, tiller number, seed weight, and productivity among 29 rice landraces from Toraja region. A few of them have coloured grain, such as Pare Lea, Pare Lallodo, and Pare Ambo. However, the tiller number reported by [9] was 3-4 times higher (17-30) than tiller number reported in [10]. These two different studies were undertaken in different locations but in the Toraja region hence variability in result is noted.

3.2 Morphological characteristics

3.2.1 Quantitative measurements. Ra'rang has the longest ligule length among the landraces, which similar to modern Inpari 24. Jambu has the shortest ligule. correlation coefficient of ligule length and grain length/width ratio was 0.115 Which interpreted as little if any correlation [19]. Conversely, a little negative correlations were found between ligule length and leaf length, panicle length, and grain width.

The most fundamental factor in the diversity of grain colours is the genetic factor that expresses the grain colour of the variety. Result of bioactive content assessment of pigmented rice landrace of South Sulawesi also confirmed the high anthocyanin content of black rice, and considerable amount of phenolic content in red rice [5].

Maminyak has the longest leaf (78.80 cm), whilst Ciherang, Balan, and Inpari have the shortest leaf length. has the shortest leaf length (45 cm; 48 cm and 51 cm respectively). Moderate positive correlation ($r=0.577$ $N=80$ $p<0.01$) was found between leaf length and plant height.

The longest panicle length was found in Lotong (36.5 cm), which significantly differ from Rarang, Lallodo, Lea, Lotong Tanduk, Mandoti, Balan, Kamida, Leleng, Bakka Eja, Punu Eja and two modern varieties (Ciherang and Inpari 24) (Table 5). As expected, the shortest panicle was found in Ciherang. Panicle length has low positive correlation with leaf length ($r=0.488$ $N=80$ $p<0.001$) (Table 5). Multiple genes controlled panicle length, a typical quantitative trait, and may be affected by environmental conditions significantly. In addition, breeding varieties with large panicle and the ideal plant architecture traits are significant for increasing the grain yield of rice [20].

Table 5. Measurement of morphological characteristics of pigmented rice landrace of South Sulawesi and two modern rice varieties for comparison.

Variety	Ligule length	Leaf length	Panicle length	Grain's length/width ratio	Grain width
Kobo	14.20±0.66edc	63.00±2.19gfedcb	32.00±2.30ba	2.78±0.07gf	2.90±0.13cba
Lotong	17.20±1.24cba	76.10±3.26cba	36.50±0.87a	3.14±0.08fedc	3.00±0.05ba
Ambo	13.80±1.24edc	63.40±2.71fedcb	29.60±0.53cdba	2.64±0.04g	3.00±0.06ba
Ra'rang	20.40±1.63a	68.40±3.16edcba	21.80±2.89hgfed	2.92±0.12gfed	2.84±0.08cba
Lallodo	14.80±0.50edcb	72.60±1.89dcba	21.80±1.98hgfed	3.32±0.11dc	2.72±0.09dcba
Lea	13.00±1.38edc	70.40±2.25dcba	23.40±1.78hgfedc	2.84±0.05gfe	2.86±0.07cba

L. Tanduk	11.50±0.50ed	63.00±2.00gfedcb	23.00±2.00hgfedc	2.77±0.05	3.25±0.05
Maminyak	13.60±0.93edc	78.80±2.96a	29.20±1.59edcba	2.89±0.15gfed	2.76±0.08dcba
Mandoti	12.00±0.71edc	76.00±3.41cba	25.40±0.98gfedcb	3.11±0.09fedc	2.86±0.02cba
Balan	15.60±0.51dcba	48.20±2.44hg	20.20±1.36hgf	3.19±0.05fedc	2.66±0.05dc
Kamida	11.00±0.45ed	59.20±0.86hgfed	27.80±1.66fedcb	3.09±0.10gfedc	2.70±0.03dcb
Jambu	10.00±0.55e	77.40±2.58ba	30.04±1.16cba	3.25±0.06edc	2.82±0.04cba
Le'leng	12.60±0.81edc	59.40±2.01hgfed	25.20±0.73gfedcb	2.91±0.02gfed	3.04±0.06a
Bakka Eja	12.60±0.81edc	53.90±1.35hgfe	21.20±1.01hgfe	3.85±0.08ba	2.48±0.06ed
Punu Eja	12.00±0.95edc	62.00±4.12gfedc	22.40±1.69hgfedc	2.99±0.12gfed	3.00±0.10ba
Inpari 24	20.20±1.20ba	51.00±1.87hgf	18.93±1.22hg	4.13±0.08a	2.24±0.05e
Ciherang	14.00±1.10edc	45.40±5.57h	15.40±0.51h	3.52±0.10cb	2.18±0.04e

Values followed the same letter are not different ($P < 0.05$).

3.2.2 Qualitative observations. Morphological characteristics of tested varieties observed under glass house conditions are presented in table 6. An important observed feature is the extent of pigmentation found in various plant parts including auricles, ligules, awns, culm internodes, and leaf blades among different varieties. While light green was the typical colour of auricles in most varieties tested in this experiment, only Ambo and Lotong Tanduk have the purple colour in the auricles. Awned seeds found in 52.9% of tested varieties. One of Wild rice species' distinct feature in morphology is a long awn [21]. Shorter awn or awnless grain in domesticated wheats facilitate harvesting, handling and storage [22].

The awn color of awned seed types ranged from straw (Kobo, Lotong, Lallodo, Maminyak, Mandoti, Jambu, Le'leng) to red (Ambo and Lotong Tanduk). Each variety of Ambo as well as Lotong Tanduk also shared similarity in culm internode color (purple), ligule color (purple), and leaf blade colour (purple in the margin), along with kernel colour of black. The culm internode colour of Ambo, L. Tanduk, Kamida were purple, while Punu Eja has purple line for their culm internode colour. Most varieties (88.2%) have white ligule colour while the ligule shape was cleft in all varieties. Plants with purple ligule colour (Ambo and Lotong Tanduk) also have purple margin leaf blade colour, and the remaining varieties are in green colour of leaf blade (table 6). Compared with black rice found in Central Java, namely Matesih, Klaten and Cempo, all of them have white ligule in cleft shape, green leaf blade color and white auricle [23].

Table 6. Morphological characteristics observation of pigmented rice landrace of South Sulawesi and two modern rice varieties for comparison

Variety	Auricle color	Awn color	Awning	Culm internode color	Ligule color	Ligule shape	Leaf blade color
Kobo	Light green	Straw	Long and Well	Green	White	cleft	Green
Lotong	Light green	Straw	Long and Well	Green	White	cleft	Green
Ambo	Purple	Red	Long and Well	Purple	Purple	cleft	Purple margin
Ra'rang	Light green	-	Awnless	Green	White	cleft	Green
Lallodo	Light green	Straw	Short and partly	Green	White	cleft	Green
Lea	Light green	-	Awnless	Green	White	cleft	Green
Lotong Tanduk	Purple	Red	Short and partly	Purple	Purple	cleft	Purple margin
Maminyak	Light green	Straw	Long and Well	Green	White	cleft	Green
Mandoti	Light green	Straw	Long and Well	Green	White	cleft	Green
Balan	Light green	-	Awnless	Green	White	cleft	Green
Kamida	Light green	-	Awnless	Purple	White	cleft	Green
Jambu	Light green	Straw	Long and Well	Green	White	cleft	Green
Le'leng	Light green	Straw	Long and Well	Green	White	cleft	Green
Bakka Eja	Light green	-	Awnless	Green	White	cleft	Green
Punu Eja	Light green	-	Awnless	Purple line	White	cleft	Green
Inpari 24	Light green	-	Awnless	Green	White	cleft	Green
Ciherang	Light green	-	Awnless	Green	White	cleft	Green

4. Conclusions

The discovery of genetic differences plays a key role in the development of varietal conditions that may contribute to food security. In this study, variability is seen in some agronomic traits and morphological characteristics of pigmented rice landraces.

Some pigmented rice landraces performed better than modern varieties under glasshouse environment of temperate climate. The better performance of pigmented rice landrace indicate the capability in coping towards different environment from their origin of habitat.

Pigmented rice plant idotype can be engineered from selecting desired agronomic traits, ideal architecture, as well as superior physicochemical properties. Farmers perspective and their requirement should be taken into consideration as well, since farmers are the key factor for preserving the germplasm in situ by retaining the varieties intergeneration.

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