

Estimation of Heritability and Genetic Advance for Morphological and Physiological Traits in Rice (*Oryza Sativa* L.) Under Heat Stress

Prita Sari Dewi^{1,2(\Box)}, Ida Widiyawati², Wilis Cahyani², and Kartika Ferrawati²

¹ Postgraduate Program Jenderal Soedirman University (Unsoed) Purwokerto Central Java, Purwokerto, Indonesia

prita.dewi@unsoed.ac.id

² Faculty of Agriculture Unsoed Purwokerto Central Java, Purwokerto, Indonesia

Abstract. The efficiency of plant breeding programs can be improved by determining genetic variability, heritability, and genetic advance. The experiment was carried out to study the genetic variability, broad-sense heritability, and genetic advance for some morphological and physiological traits in rice influenced by heat stress. A split plot design was used in this study with 16 rice varieties were arranged as sub plot with two heat stress treatments as the main plot. The temperature was set to 37-38 °C and 40-41 °C and the experiment was carried out with three replications. The observed morphological and physiological traits were tiller number, leaf number, plant height, root length, root fresh weight, root dry weight, shoot fresh weight, shoot dry weight, leaf area, proline content at the vegetative stage, number of sterile pollens, stomatal density, chlorophyl content, and 1000 seed weight. There was a high coefficient of variation for most of the traits observed in the experiment. The phenotypic variances of the observed variables were greater than those of the genotypic variances. This finding was in line with the trends of their phenotypic co-efficient of variances compared to genotypic co-efficient of variances. The broad-sense heritability values were high for all observed traits, except moderate for the number of sterile pollens. The higher the heritability, the better a trait can be inherited into the next progenies. The genetic advances were varied from low to high for all observed traits.

Keywords: "broad-sense heritability \cdot genetic advance \cdot genetic variability \cdot high temperature \cdot plant physiology."

1 Introduction

High temperature causing heat stress become problem in many agricultural lands in the world. It has been reported by the Central Bureau of Statistics that the highest temperature in Indonesia in 2021 was 38.4 °C in 2021 with the average temperature of 27.1 °C and positive anomaly of 0.4 °C. Rice plant can grow well from 25–35 °C for all stages of development [1]. Temperatures above 35 °C are reported to reduce vegetative growth [2] and interfere with generative growth resulted in low yield [3]. In addition, pollen is very

sensitive to high temperature stress causing pollen sterility. As a result, the fertilization process will be hampered and the yield will decrease [4].

One of the efforts to overcome this problem is breeding new rice varieties tolerant to high temperature stress. The initial step that needs to be done is screening rice germplasms for high temperature tolerance. After obtaining the genotype of rice plants tolerant of high temperature stress, crosses between the tolerant genotype and genotypes with other superior traits can be carried out as needed. Estimates of heritability values and genetic progress are needed to determine how much a trait can be passed down from one generation to the next [5]. Most of experiment related to genetic studies in plants is focused on genetic variability, heritability, and genetic advance for specific collections/landraces [6, 7]. However, estimation of heritability values and genetic advance in rice plants under abiotic stress was still limited. [8] reported that heritability for several physiological and yield contributing traits in wheat varieties under heat stress were moderate to high. Moreover, it was also reported that the genetic advance for these traits was moderate to high. The information regarding the genetic response of the rice varieties and genotypes under specific abiotic stress will help us to determine the mechanism of heat stress tolerance in rice plants and consider the specific traits to be used as morphological or physiological markers for selection. The heritability value can help plant breeders to determine the duration and selection method because it represents the proportion between genotypic and phenotypic variation that is inherited to the next generations.

This study was aimed to estimate the genetic variability, the broad-sense heritability, and the genetic advance of 16 rice varieties and accessions for some morphological and physiological traits in rice under high temperature stress.

2 Materials and Methods

2.1 Plant Materials

There were 16 rice varieties and accessions used in the experiment (Table 1). Some seeds were requested from Indonesian Centre for Rice Research (ICRR) and the remaining seeds were kindly provided from the breeders. These rice varieties and accessions are known for their superior characters including drought tolerant, salt-tolerant rice [9, 10], black rice [11, 12], upland rice, and commercial ones with excellent palatability, high yield, and wide adaptability.

2.2 Heat Stress Experiment

The experiment was carried out in the screenhouse of Center for Crop and Horticultural Seeds (Balai Benih Tanaman Pangan dan Hortikultura) Purwokerto Central Java Indonesia. A split plot design was applied with heat stress as the main plot and genotype as the sub plot with three replicates. Temperature treatment was consisted of the temperature between 37–38 °C and above 40–41 °C. The plants were grown in the polybags following cultural practices and plant protection recommended by Ministry of Agriculture Republic of Indonesia. The temperature for T2 and T3 was achieved by covering the screenhouse with vinyl plastic. Observation of the temperature and humidity were

Variety	Superior trait	Source
Inpari 18	Tolerant to drought	Indonesian Center for Rice Research (ICRR)
Inpari 19	Tolerant to drought	ICRR
Inpari 20	Tolerant to drought	ICRR
Inpago Lipigo 4	Tolerant to drought	ICRR
Inpago 8	Tolerant to drought	ICRR
Inpari 34 Agritan	Tolerant to salinity	ICRR
Inpari 35	Tolerant to salinity	ICRR
Inpari Unsoed 79 Agritan	Tolerant to salinity	ICRR
Jeliteng	A variety of black rice	ICRR
РН 3	An accession of black rice	Perjuangan University West Java
PH 5	An accession of black rice	Perjuangan University West Java
Inpago Unsoed 1	A lowland rice variety with aromatic and fluffy rice type	Jenderal Soedirman University
IR 64	High yield and wide adaptability	ICRR
Parimas	Tolerant to drought and aluminium	Jenderal Soedirman University
Ciherang	High yield and wide adaptability	ICRR
Situbagendit	Tolerant to drought	ICRR

Table 1.	Rice	genotypes	used	in	this	study
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carried out three times a day. First observation was done in the morning (6 am), in the midday (12 am), and in the evening (6 pm) for 19 weeks. The observed variables were tiller number (tiller), leaf number (leaf), plant height (cm), root number (root), root dry weight (g), root fresh weight (g), shoot dry weight (g), shoot fresh weight (g), leaf area (cm²), proline content at the vegetative stage (μ gml⁻¹), number of sterile pollens (pollen), stomatal density (stomata/mm²), chlorophyl content (mgl⁻¹), and weight of 1000 seeds (g).

2.3 Data Analysis

The data recorded using the experiment were then used for statistical analysis by using analysis of variance (ANOVA) for the split plot design using IBM SPSS 17.0. Means from the result of ANOVA analysis were used to calculate some genetic parameters such as variance of genetics, variance of phenotype, variance of environment, genotypic coefficient of variation (GCV), phenotypic coefficient of variation (PCV), and the genetic advance. The phenotypic variation for all observed variables were divided into genetic factors, environmental factors, and interaction between genetics and environmental factors which were estimated following [13].

$$\sigma^2 p = \sigma^2 t + \frac{\sigma^2 t x g}{g} + \frac{\sigma^2 e}{rg}$$
(1)

where $\sigma^2 e$, $\sigma^2 txg$, and $\sigma^2 p$ are variance of environment, variance of genotype, and variance of phenotype, respectively. MSe, MSt, MStxg, r, and g are the mean squares of error, the mean squares of temperature treatment, the mean squares of interaction between temperature treatment and genotype, number of replications, and number of rice accessions or genotypes used in the experiment, respectively.

$$\% PCV = \frac{\sqrt{\sigma^2 p}}{\bar{x}} x100 \tag{2}$$

$$\% GCV = \frac{\sqrt{\sigma^2 txg}}{\bar{x}} x100 \tag{3}$$

The criteria for dividing genotypic and phenotypic coefficient of variation were according to [9]. CGV and PCV were categorized as narrow if the value is 0-10%, moderate if the value is > 10%, and wide if the value is > 20%.

$$h^2 b = \frac{\sigma^2 t}{\sigma^2 t + \frac{\sigma^2 t g}{g} + \frac{\sigma^2 e}{rg}}$$
(4)

Broad-sense heritability was calculated according to [14]. Genetic advance (GA) and genetic advance in per cent (GA%) were calculated following [15].

$$GA = h^2 b.i.\sigma p \tag{5}$$

where h_b^2 is broad-sense heritability, i is selection differential, with the value of 1.76 at 10% level of selection intensity, and σp is phenotypic standard deviation. Broad-sense heritability was categorized as low ($h^2b < 20\%$), moderate ($20\% \le h^2b \le 50\%$), and high ($h^2b > 50\%$) following [16].

The genetic advance in percent of mean (GA % mean) was estimated following [15].

$$GA\%mean = \frac{GA}{\bar{x}}x100\tag{6}$$

GA was categorized as low (GA < 7%), moderate (7% \leq GA \leq 14%), and high (GA > 14%) according to [17].

3 Results and Discussion

Table 2 showed that the range of genotypic and phenotypic coefficient of variance was 0.92–17.75 and 12.12–93.17, respectively. Phenotypic variance is higher than that of

Observed variables	σ ² txg	$\sigma^2 p$	σ ² e	GCV		PCV	
				Value	Category	Value	Category
Tiller number (tiller)	0.02	71.65	7.32	0.92	Ν	60.03	W
Leaf number (leaf)	7.76	110.67	9.20	13.59	М	62.57	W
Plant height (cm)	26.83	217.62	77.18	5.15	Ν	14.68	М
Root number (root)	5.85	65.18	42.01	4.55	Ν	15.18	М
Root dry weight (g)	7.28	114.48	71.70	9.25	Ν	36.66	W
Root fresh weight (g)	5.50	1091.50	351.21	2.10	Ν	29.60	W
Shoot dry weight (g)	6.81	175.77	78.60	5.71	Ν	29.00	W
Shoot fresh weight (g)	147.22	4330.86	2065.09	5.39	Ν	29.21	W
Leaf area (cm ²)	6.31	516.00	32.11	6.64	Ν	60.09	W
Proline content at the vegetative stage (μgml^{-1})	0.48	0.26	1.20	14.74	М	18.20	М
Number of sterile pollens (pollen)	10395.47	147318.69	216266.17	12.67	М	47.68	W
Stomatal density (stomata/mm ²)	36.19	3781.49	2936.96	9.11	N	93.17	W
Chlorophyl content (mgl ⁻¹)	50.13	181.28	79.60	17.75	М	33.74	W
Weight of 1000 seeds (g)	0.17	9.18	2.82	1.65	N	12.12	М

Table 2. Genetic and phenotypic variations for the observed variables

Remarks: $\sigma^2 txg = genotypic$ variance, $\sigma^2 p = phenotypic$ variance, $\sigma^2 e = environmental variance, CGV = genotypic coefficient of variation, PCV = phenotypic coefficient of variation, N = narrow, M = moderate, and W = wide.$

the genotypic variance, followed by the phenotypic coefficient of variance (PCV) value which is greater than the genotypic coefficient of variance (GCV) value for all observed characters. Similar findings were reported by [8] for PCV of plant height, heading date, maturity date, canopy temperature at both vegetative and grain filling stages, chlorophyl content, spike per meter square, spikelet per spike, grains per spike, 1000 grain wight, and harvest index is greater than GCV in wheat genotypes under heat stress condition. When the level of genetic variation is narrow then this shows that the individual is in the population is relatively uniform. Therefore, selection for trait improvement is less effective [18]. On the other hand, the wider the genetic variation, the greater the chance for successful selection for the targeted traits. Based on the results of the analysis in this study, the small difference in variance between phenotypic and genotypic values was indicated by proline content at the vegetative stage. Therefore, the proline content at the vegetative stage is an important trait to select the genotypes which are tolerant to

heat stress. Proline has been known as the osmoprotectants for rice plant under abiotic stresses including drought, heat, salinity, and both drought and heat stresses [19].

It is necessary to determine the heritability estimation value of a character to predict the progress of a selection, whether the appearance of the character is heavily influenced by genetic or environmental factors [20]. Based on the analysis of the estimation of variance and heritability values, a plant character that has a low genetic diversity value does not necessarily have a low heritability value and vice versa. There are several characters of the outcome components in this study that have low PCV or CGV values but have high heritability categories. This is because the heritability value is formed by the ratio between genotypic variance to the total phenotypic variance [16, 21]. This is in line with the experiments of heat stress in wheat by [8] for all the vegetative and generative traits which had low CGV but showed high heritability. [22] also reported that all the observed traits exhibited high heritability although their GCV were varied between low to wide (Table 3 and 4).

The genetic advance was low for proline content at the vegetative stage and weight of 1000 seeds, moderate for root number, and the remaining observed variables were high. Characters with high genetic progress values indicate that these characters are supported by genetic factors, so that they can facilitate selection progress. Most of these studies had high and low values of genetic advancement. If the value of the genetic advance for a trait is high, it means that there is a great opportunity for improvement of the trait during selection. On the other hand, if the value of genetic advance is low, then

Observed variables	Population means	Broad-sense heritability	Category
Tiller number (tiller)	14.10	0.97	Н
Leaf number (leaf)	20.50	0.97	Н
Plant height (cm)	100.50	0.82	Н
Root number (root)	53.19	0.74	Н
Root dry weight (g)	29.19	0.82	Н
Root fresh weight (g)	111.61	0.90	Н
Shoot dry weight (g)	45.72	0.87	Н
Shoot fresh weight (g)	225.29	0.86	Н
Leaf area (cm ²)	37.80	0.99	Н
Proline content at the vegetative stage (μgml^{-1})	2.81	0.75	Н
Number of sterile pollens	805.00	0.48	М
Stomatal density (stomata/mm ²)	66.00	0.75	Н
Chlorophyl content (mgl ⁻¹)	39.90	0.72	Н
Weight of 1000 seeds (g)	25.01	0.89	Н

Table 3. Estimation of broad-sense heritability for the observed variables

Remarks: M = moderate and H = high.

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Observed variables	Genetic Advance (GA)	GA%	Category
Tiller number (tiller)	14.39	102.08	Н
Leaf number (leaf)	56.83	277.22	Н
Plant height (cm)	21.29	21.19	Н
Root number (root)	10.52	19.78	М
Root dry weight (g)	15.50	53.10	Н
Root fresh weight (g)	52.06	46.64	Н
Shoot dry weight (g)	20.31	44.42	Н
Shoot fresh weight (g)	99.38	44.11	Н
Leaf area (cm ²)	39.39	104.22	Н
Proline content at the vegetative stage (μgml^{-1})	1.31	46.53	L
Number of sterile pollens	321.13	39.89	Н
Stomatal density (stomata/mm ²)	80.73	122.31	Н
Chlorophyl content (mgl ⁻¹)	16.95	42.48	Н
Weight of 1000 seeds (g)	4.74	18.95	L

Table 4. Estimation of genetic advance for the observed variables

Remarks: H = high, M = moderate, and L = low.

the selection for this trait can be carried out after the population reached some extent of homozygosity which means need longer time because selection can only be taken in the late generation. Moreover, the selection can also be ended because the improvement of the trait is relatively low [23]. This indicates that the observed characters with low genetic advance based on the tested genotypes are regulated by the action of non-additive genes. Low genetic advance was also reported in the study of [24], including harvest age, number of productive tillers, weight of grain per panicle, number of grains per panicle, and weight of 1000 seeds.

This experiment showed that there is high value for the broad-sense heritability and the genetic advance for most all the observed traits. This is indicated that selection in rice breeding program tolerant for heat stress can be done in the early generations using several recommended selection methods such as mass selection, pure line selection, or pedigree [25].

4 Conclusions

The genotypic coefficient of variation was low to moderate while phenotypic coefficient of variation was moderate to wide for the observed variables. The broad-sense heritability was moderate except low for the number of sterile pollens. The genetic advance was low for proline content at the vegetative stage and weight of 1000 seeds, moderate for root number, and the remaining observed variables were high.

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