AGRO-MORPHOLOGICAL AND MOLECULAR CHARACTERIZATION OF RED RICE (*Oryza sativa* L.) GERMPLASM OF HIMACHAL PRADESH

THESIS

By

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(A-2020-30-043)

Submitted to

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PALAMPUR – 176 062 (H.P.) INDIA

in

Partial fulfilment of the requirements for the degree

of

MASTER OF SCIENCE IN AGRICULTURE
(DEPARTMENT OF GENETICS AND PLANT BREEDING)
(GENETICS AND PLANT BREEDING)

2022


Dedicated to My Advisor,
Revered Grandparents
(Amma, Baba),
Parents
&
Loving Brother!

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CERTIFICATE – I

This is to certify that the thesis entitled **"Agro-morphological and molecular characterization of red rice (*Oryza sativa* L.) germplasm of Himachal Pradesh"** submitted in partial fulfilment of the requirements for the award of the degree of **Master of Science (Agriculture)** in the discipline of **Genetics and Plant Breeding** of CSK Himachal Pradesh Krishi Vishvavidyalaya, Palampur is a bonafide research work carried out by **Ms. Rishita Kapoor (A-2020-30-043)** daughter of **Smt. Neelam Kapoor & Sh. Vijay Kapoor** under my supervision and that no part of this thesis has been submitted for any other degree or diploma.

The assistance and help received during the course of this investigation have been fully acknowledged.

Place : Palampur
Dated :14 October, 2022

(Dr. Neelam Bhardwaj)
Major Advisor

CERTIFICATE- II

This is to certify that the thesis entitled **"Agro-morphological and molecular characterization of red rice (*Oryza sativa* L.) germplasm of Himachal Pradesh"** submitted by **Ms. Rishita Kapoor (A-2020-30-043)** daughter of **Smt. Neelam Kapoor & Sh. Vijay Kapoor** to the CSK Himachal Pradesh Krishi Vishvavidyalaya, Palampur in partial fulfillment of the requirements for the degree of **Master of Science (Agriculture)** in the discipline of **Genetics and Plant Breeding** has been approved by the Advisory Committee after an oral examination of the student in collaboration with an External Examiner.

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"Presentation inspiration and motivation have always played a key role in the success of any venture"

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Place: Palampur
Dated: 14 October, 2022 ***(Rishita Kapoor)***

TABLE OF CONTENTS

LIST OF ABBREVIATIONS USED

S. No.	Abbreviation	Meaning
1.	%	Per cent
2.	&	And
3.	/	Per
4.	<	Less than
5.	>	More than
6.	µg	Microgram
7.	µl	Microlitre
8.	µM	Micromolar
9.	bp	Base pair
10.	cm	Centimeter
11.	conc.	Concentrated
12.	CTAB	Cetyl trimethyl ammonium bromide
13.	dATP	Deoxyadenosine triphosphate
14.	dCTP	Deoxycytosine triphosphate
15.	df	Degree of freedom
16.	dGTP	Deoxyguanosine triphosphate
17.	DNA	Deoxyribonucleic acid
18.	dNTP	Deoxynucleotide triphosphate
19.	dTTP	Deoxythymidine triphosphate
20.	E	East
21.	EDTA	Ethylene diamine tetra acetic acid
22.	et al.	Et alii (and others)
23.	etc.	Et cetera
24.	Fig.	Figure(s)
25.	g	Gram
26.	GCV	Genotypic coefficient of variation
27.	HCl	Hydrochloric acid
28.	hr	Hour
29.	i.e.	id est (that is)
30.	ISSR	Inter Simple Sequence Repeats
31.	kb	Kilobase
32.	KCl	Potassiun chloride
33.	kg	Kilogram
34.	l	Litre
35.	M	Molar

S. No.	Abbreviation	Meaning
36.	m^2	Square metre
37.	mg	Milligram
38.	$MgCl_2$	Magnesium chloride
39.	min	Minute(s)
40.	ml	Milliliter
41.	mM	Millimolar
42.	mM	Millimolar
43.	N	North
44.	NaCl	Sodium chloride
45.	ng	Nanogram
46.	^oC	Degree Celsius
47.	P	Page(s)
48.	PCA	Principal Component Analysis
49.	PCR	Polymerase Chain Reaction
50.	PCV	Phenotypic coefficient of variation
51.	pH	Puissance de hydrogen (ion conc.)
52.	PIC	Polymorphic Information Content
53.	ppm	Parts per million
54.	PVP	Polyvinyl pyrrolidone
55.	Q	Quintal
56.	rA	Additive genetic correlation
57.	RAPD	Random amplified polymorphic dna
58.	RNase	Ribonuclease
59.	rpm	Revolutions per minute
60.	sec	Second(s)
61.	SNP	Single Nucleotide Polymorphism
62.	SSR	Simple Sequence Repeats
63.	*Taq* polymerase	*Thermus aquaticus* DNA polymerase
64.	TE	Tris EDTA buffer
65.	Tris	Tris (hydroxy methyl) amino methane
66.	U	Units
67.	UBN	Uniform Blast Nursery
68.	UV	Ultraviolet
69.	V	Volts
70.	*viz.*,	Vi delicet (namely)

LIST OF TABLES

LIST OF FIGURES

LIST OF PLATES

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ABSTRACT

The present investigation entitled "Agro-morphological and molecular characterization of red rice (*Oryza sativa* L.) germplasm of Himachal Pradesh" was undertaken to identify and characterize the rice genotypes by assessing the nature of variation and extent of genetic diversity among the genotypes using morphological and molecular markers. The experimental material comprising 43 genotypes including two checks were evaluated in Randomized Block Design during *Kharif* 2021 with three replications. Data was recorded on grain yield per plant and various morphological traits along with reaction to leaf blast and neck blast. Genetic diversity among different genotypes was studied on the basis of morphological traits using Mahalanobis D^2-statistics and molecular analysis. The morphological analysis was done as per the standard statistical procedures and molecular analysis was done using NTSYS-pc, DARwin and STRUCTURE softwares. Analysis of variance revealed that mean sum of squares due to genotypes were significant for all the agro-morphological traits except flag leaf width indicating ample amount of genetic variability in the material under study. High PCV, GCV and high heritability coupled with high genetic advance was recorded for total tillers per plant, L:B ratio and grain yield per plant providing higher chance of selection for these traits. Grain yield per plant exhibited significant positive correlation with all the traits except flag leaf width and grain breadth. Path coefficient analysis revealed the high direct effect of biological yield per plant, harvest index, flag leaf length and spikelets per panicle on grain yield per plant. D^2-statistics grouped 43 rice genotypes into four clusters. Based on D^2 statistics highest inter-cluster distance was recorded between cluster III (Sukara, Desidhan and Sukara red) and cluster IV (Kalijhini-2, HPR-2913, Phulpatas-21 and HPR-2795). Hence, it has been well established that the genotypes belonging to these clusters are genetically diverse. More the genetically diverse parents used in the hybridization program, greater will be the chances of obtaining high heterotic hybrids. Cluster III had the highest cluster mean values for most of the traits. Twenty one SSR primers amplified 53 polymorphic alleles with the mean of 2.52 alleles per primer. Furthermore, 35 genotypes were common between D^2 analysis and molecular analysis showing the congruence of molecular markers with the morphological descriptors and hence, providing a powerful tool to characterize and identify the genotypes of rice. Three genotypes namely Desidhan, HPR-2913 and Sukara Red were found promising on the bases of their mean performance and resistance to leaf blast and neck blast, therefore, could be further used in rice breeding programme.

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1. INTRODUCTION

Rice (*Oryza sativa* L.) belongs to family poaceae. It is one of the world's most important food crops and is grown in 115 countries in different parts of the world providing staple food to more than half of the world's population. India has the largest area under rice in the world and ranks second in production next to China. As a major cereal crop, it is a diversified crop species due to its adaptation to a wide range of geographical, ecological and climatic regions (Yadav et al. 2013). The cultivated rice of Asia is supposed to have originated in the South and South East Asia. Since India forms a major part of this region, hence traditionally rich in the rice diversity including the wild progenitors of cultivated rice (Singh et al. 2001). According to USDA, the total area coverage of rice in the world is 164.82 million hectare with 507.46 million metric tons production and 4.64 metric tons/ha productivity. In India, the area under rice is 45 million hectare with 122.27 million metric tons production and 4.09 metric tons/ha productivity (Anonymous a 2021).

In Himachal Pradesh rice is an important cereal crop next to maize during wet season. The total area coverage of rice in the Himachal Pradesh is 75,000 hectare with 0.146 million metric tons production and 1.94 metric tons/ha productivity which accounts for 10.8% of area and 10.2% of production on total food grain basis and 22.2% of area and 18.8% of production on wet season crop basis in the state. The total area coverage of red rice in the state is 2,000 hectare approximately with 9926 q production and 8-10 q/ha productivity (Anonymous b 2021). It is cultivated in ten of the twelve districts of the state except Kinnaur and Lahaul & Spiti. Only Kangra and Mandi districts of the state account 71.2% of area and 69.7% of production.

A number of red grained varieties are cultivated in Kerala, Tamil Nadu, Himachal Pradesh and Karnataka. As in white rice, great diversity exists among the red rice. They are glutinous and non-glutinous; scented and non-scented; late and early maturing; short and long grained. Himalayan red rice has made its presence felt in the export market for its long slender character. Hence it is also called as RED PEARLS of Himalayas (Kaushik and Santiaguel 2014). Red rice is considered to be

highly nutritive and medicinal. It has a higher content of crude fiber (2.7%), crude protein (10.4%), minerals and antioxidants than white rice having higher nutrient density and lower glycaemic index which makes it comparatively superior than white rice. Red rice is also found to have a higher iron, magnesium, calcium and zinc content than white rice.

In Himachal Pradesh, the area under traditional rice varieties is considerably reducing due to various reasons. However, some traditional varieties are still grown in some isolated pockets because of their adaptability to stress situations like drought, quick germination, quality preference, early maturity and cold tolerance etc. These are found in mid and high hills of the state. In the mid hill traditional varieties like *Kalizhi*n*i*, *Madhumalti*, *Muskan*, *Achhoo*, *Chetru Basmati*, *Seond Basmati*, *Ram Juwain*, *Hatkoti Basmati* and *Panarsa* local are grown for their local preference for quality. In the high hills, traditional varieties are *Jattoo*, *Matali*, *Lal Dhan*, *Deval*, *Chohartu* and *Sukara Dhan* etc. are grown.

Assessment of genetic diversity is very important in rice breeding for selection, conservation and proper utilization of different land races (Mohammmadi et al. 2008). Morphological markers have been routinely used for characterization and estimation of genetic diversity. Since morphological traits are affected by environment, so there is a need to go for a highly reliable and precise method for assessment of genetic variability with no environmental effects. Assessment of genetic diversity with molecular markers overcomes this problem. They are devoid of environmental effects and provide a true representation of the entire genome. Several molecular marker types are available and each has their advantages and disadvantages. Simple sequence repeat (SSR) markers are now being extensively used in many crops which are clusters of short tandem repeated nucleotide bases distributed throughout the genome. They are widely used because of being highly reliable (reproducible), co-dominant in inheritance, multi-allelic, relatively simple, cheap to use, highly polymorphic and requiring small amount of DNA for scoring. In the light of above observations, the present study has been planned to characterize the red rice germplasm and evaluate it for yield and related traits for use in the future breeding programmes.

Keeping above factors in view study entitled, “Agro-morphological and molecular characterization of red rice (*Oryza sativa* L.) germplasm of Himachal Pradesh” has been undertaken with the following objectives:

Objectives:

To

- characterize red rice germplasm using morphological and SSR markers.
- identify the promising red rice genotypes for yield and component traits.

2. REVIEW OF LITERATURE

Characterization and quantification of genetic diversity has been a major goal in evolutionary biology. Characterization is a critical step to be carried out to identify accessions to find genetic relationships among genotypes. A flourishing plant breeding program heavily relies upon existence of variability in the base population for various traits and information on genetic control of concerned character is useful for effective execution of any breeding program. Systematic study and characterization of high quality germplasm is not only important for utilizing the appropriate attribute based donors, but also essential in the present era for protecting the unique rice. Different genetic parameters *viz.*, variability (mean, range, coefficient of variation), morphological diversity and molecular diversity are important and exhibit to study yield, its contributing traits and grain quality traits.

The literature pertaining to recent problem has been compiled under the following headings:-

2.1 Genetic variability, heritability and genetic advance

Any crop improvement depends on proper utilization of the available germplasm stocks. To provide information for any plant breeding initiatives, morphological characterization of germplasm is essential (Das and Ghosh 2011). Different land races can be distinguished from one another and can be recognized

with the use of characterization. Both qualitative and quantitative traits can be characterized, however quantitative traits are thought to be more crucial for identifying a specific plant variety, while qualitative traits are more genetically controlled and hence less dependent on the environment.

(a) Morphological variability

Jing et al. (2010) examined the yield, harvest index (HI) and grain quality of five rice genotypes produced from various germplasm at eight agro-ecological sites in the tropics and subtropics of Asia and found considerable genetic variability among genotypes.

Semwal et al. (2014) investigated the quatitative and quanlitative characteristics of 23 landraces, including five red rice cultivars. Characterization revealed that the seed coat came in a variety of colours including white, golden yellow, light brown and red. Awns were discovered in 17 landraces and both the maximum and minimum awn lengths were identified in red rice landraces. The red rice genotypes were found to have a maximum 1000 grain weight.

Sinha et al. (2015) investigated the grain morphological characteristics of 55 traditional rice varieties including grain size, shape and awns. It was discovered that the variation in rice grain length in the landraces tested ranged from 5.6 to 11.2 mm, while grain width varied from 1.8 to 4 mm. The L:B ratio ranged from 2.15 to 4.45 and the weight of 1000 grains was found to be between 10.1 and 33.6 g.

Kumar et al. (2016) studied 64 aromatic rice germplasm for their agro morphological characters including auricle colour, ligule, stigma colour and awns. The analysis of variance revealed that the wide range of variation existed among the germplasms and the germplasm lines. Three genotypes were recorded with pale red or red in colour while 55 genotypes were recorded with white kind of decorticated grain colour and 31 genotypes were found to have awns.

Puren (2017) assessed the productivity of red and black rice genotypes of Himachal Pradesh and found two red genotypes Bongal Dhan and HPR-2800 to have a better L:B ratio. The red rice genotypes Bongal Dhan, HPR-2800, ACC-19186,

Karad, Chohartu and Nagar Dhan were identified to have the best grain quality among all the examined genotypes.

Pachauri et al. (2017) assessed 124 rice germplasm lines including red rice germplasm lines for 19 morphological and 11 agronomical parameters. It was discovered that the days to 50 per cent flowering varied from 66 to 110 days while days to 75 per cent maturity varied from 86 to 130 days. The other characters which include effective tillers per plant and height of the plant were observed to be between 5 to 26 and 70 to 184 cm, respectively.

Rachappanavar (2017) investigated 30 different genotypes of rice and revealed that mean sum of squares due to genotypes were significant for all the morphological traits studied except for 1000 seed weight and grain width.

Singh et al. (2020) assessed the morphological characteristics of Himachal Pradesh red rice and discovered that plant height ranged from 83.55 to 140.40 cm, days to 50 per cent flowering ranged from 85 days to 108.50 days and total tillers per plant varied between 5.15 and 9.85. Days to 75 per cent maturity ranged from 121 days to 140 days.

Alagappan and Bhardwaj (2022) investigated 35 red rice germplasm lines of Himachal Pradesh in Rice and Wheat Research Centre Malan for 12 quantitative and six qualitative traits. The analysis of variance revealed that the wide variation existed among the germplasm lines for all the traits studied except flag leaf length, harvest index and grain breadth.

(b) Genetic variability and Heritability

The information on the genetic variability in respect of economic characters in a population is the primary need for a sound breeding approach. A precise assessment of the nature and extent of genetic diversity within the red rice germplasm lines is necessary not only for better knowledge of the pattern of varietal differentiation and evolution but also assisting the breeders in selecting materials for further genetic improvement of cultivars and management of red rice genetic resources. Phenotypic variability can be simply measured in a population and it is grouped into genetic and non-genetic (environmental) component. Genotypic variation is heritable from one

generation to another, which remains unaffected by environmental conditions. This type of variability that arises due to genetic component is more useful to a plant breeder for exploitation in selection or hybridization.

Heritability is the degree of resemblance between parents and the offsprings and specifies the proportion of the total variability that is due to genetic cause or the ratio of genetic variance to the total variance. Heritability in broad sense was proposed by (Hanson et al. 1956) as the ratio of genetic variance to the total variance, while the narrow sense heritability has been defined as the ratio of additive variance to the phenotypic variance (Lush 1949). The significance of each genotype is dependent on heritability estimates. While the heritability value suggests the relative effectiveness of selection based on a trait(s) phenotypic expression, genetic innovation is more useful in predicting the actual selection value (Johnson et al. 1955).

Genetic advance explains the degree of genetic gain obtained in a character under selection pressure. High genetic advance coupled with high heritability estimates offers the most suitable conditions for selection. Wright (1921) reported that heritability components comprised of additive and non-additive portion and selection mainly responds to the former one. However, high heritability alone does not necessarily mean high genetic gain. Therefore estimates of genetic advance along with heritability is important to have an idea of the effectiveness of selection. The genetic gain that can be expressed for a particular character through selection is the product of its heritability, phenotypic standard deviation and selection differential (Burton 1952).

Burton and DeVane (1958) suggested that genetic coefficient of variation together with heritability estimates would give reliable estimates of the extent of improvement from selection and further remarked that expected genetic gain provided practical knowledge for breeders.

Thongbam et al. (2010) analysed the genetic characteristics of indigenous cultivars of rice and discovered that all of the characters had a high degree of association between genotypic coefficient of variation (GCV) and phenotypic coefficient of variation (PCV) as well as high heritability, while grain length, grain breadth and L:B ratio exhibited high heritability with low genetic advance.

Sohrabi et al. (2012) studied 50 germplasm samples for 12 quantitative characteristics and discovered that plant yield and days to 50 per cent flowering showed high heritability coupled with high genetic advance. Plant height and the number of grains per panicles showed a lower phenotypic coefficient of variation (PCV) although days to 50 per cent flowering and days to 75 per cent maturity showed higher PCV values. For 1000 grain weight, plant height, days to 50 per cent flowering, spikelets per panicle and days to 75 per cent maturity the heritability was found to be high.

Thongbam et al. (2012) assessed the morphological and nutritional characteristics of indigenous rice cultivars and observed that all physical traits tested had high heritability with medium genetic advance.

Dutta et al. (2013) assessed the variability and genetic factors of 68 rice genotypes. All characters showed significant variations for 12 agronomically significant characters. Eight characters *viz.*, tillers per plant, days to 50 per cent flowering, harvest index, spikelets per panicle, spikelets per plant, spikelet density, panicles per plant and grain yield were found to have high genotypic and phenotypic coefficients of variation, high heritability (in the broad sense) and high genetic advance as a percentage of mean. Therefore, these traits were influenced by additive gene action and a successful selection program for agronomic improvement can be based on these traits.

Sanghera et al. (2013) investigated the genetic variability on grain yield and yield component characteristic for 14 red rice cultivars in the temperate area of Kashmir. Grain yield and panicle weight had large genotypic and phenotypic coefficients of variation. Number of grains per panicle and L:B ratio showed considerable genotypic and phenotypic coefficients of variation. Days to 50 per cent flowering, days to 75 per cent maturity, plant height, grain length, grain breadth and L:B ratio all showed high heritability along with high genetic advance. While 1000 grain weight and grains per panicle were found to have a moderate genetic advance.

Chakravorty and Gosh (2013) assembled 51 traditional rice varieties to characterize 28 agro-morphological variables. Significant variations were recorded in all the quantitative traits. The parameters L:B ratio and number of grains per panicle

were found to have moderate coefficients of variation, whereas plant height and grain length showed low coefficient of variation.

Dhurai et al. (2014) analysed the genetic variability among some quantitative and quality traits in rice (*Oryza sativa* L.) and discovered the greater estimations of PCV and GCV for the number of grains per panicle and yield per plant. This suggested that the genotypes selected for investigation had a diverse genetic background and there may be scope for genetic improvement through the use of direct selection for these characters.

Khare et al. (2014) recorded high heritability along with high to moderate phenotypic and genotypic coefficients of variation and also high to moderate genetic advance as a percentage of mean for grain yield per plant, plant height, test weight, fertile spikelets per panicle, total grains per panicle and number of effective tillers per plant.

Ahmad et al. (2015) investigated colored rice varieties and found that the phenotypic variance for all examined characteristics was higher than the genotypic variance. The highest genetic advancement in yield per plant was measured as a percentage of the mean. For days to 50 per cent flowering, days to 75 per cent maturity, plant height, 1000 grain weight, harvest index, yield per plant, grain length and L:B ratio high heritability was observed. The yield per plant and harvest index were found to have high heritability along with high genetic advancement.

Kumar (2015) examined 30 rice genotypes and found that values of phenotypic coefficient of variation (PCV) were higher than their respective genotypic coefficient of variation (GCV) indicating considerable influence of environment on the performance and expression of the characters of genotypes. High PCV and GCV was observed for seeds per panicle. Moderate PCV and GCV was observed for effective tillers, leaf length, leaf area index, days to panicle initiation, 1000 seed weight and grain width.

Tuhina-Khatun et al. (2015) examined 43 genotypes for 22 morphological parameters including 34 red rice genotypes. According to the study, all of the characteristics selected for analysis had larger phenotypic coefficients of variation (PCV) than genotypic coefficients of variation (GCV). Number of filled grains per

panicle characteristics had the greatest PCV followed by yield per plant. Significant GCV values were found for the number of filled grains per panicle, harvest index, total number of tillers per plant and biological yield per plant, but the lowest GCV values were found for the grain length, days to 75 per cent maturity and days to 50 per cent flowering. The number of filled grains per panicle, grain width and yield per plant were shown to had greater heritability.

Devi et al. (2016) assessed 27 rice genotypes for morphological features for the estimation of genetic diversity. Yield per plant had the highest PCV and GCV estimates while test weight, flag leaf length, plant height and effective tillers per plant had moderate PCV and GCV values. For days to 50 per cent flowering, grain length, grain breadth, panicle length and L:B ratio low PCV and GCV were reported.

Abebe et al. (2017) examined 36 rice genotypes for genetic variability, heritability and genetic advance for grain yield. Plant height, biological yield and grain yield were found to exhibit higher PCV and GCV values. Characters including plant height, biological yield and panicle length demonstrated the high heritability. High genetic advance as percent of means were simultaneously recorded for plant height, biological yield and grain yield.

Puren (2017) examined the morphological characters of outstanding red and brown rice cultivars as well as landraces. The PCV, GCV, genetic advance and heritability of the grain yield per plant, panicles per plant, spikelets per panicle and number of grain per panicle were all found to be high, while L:B ratio found to have high heritability coupled with moderate PCV and GCV.

Rachappanavar (2017) investigated 30 different genotypes of rice and revealed the presence of considerable amount of genetic variability in the material under study. PCV and GCV were found to be high for most of the traits. High heritability coupled with high genetic advance was observed for effective tillers per plant, seeds per panicle and seed yield per plot.

Srivastava et al. (2017) studied exotic 22 genotypes and observed high PCV and GCV for filled grains per panicle, spikelets per panicle and plant height. The high heritability was recorded for filled grains per panicle, spikelets per panicle, plant height, test weight, flag leaf length and yield per plant. Number of spikelets per

panicles, plant height, 1000 grain weight, flag leaf length and grain yield per plants also exhibited high heritability.

Bagudam et al. (2018) investigated genetic variability in 46 rice genotypes and revealed the presence of genetic variability among the genotypes. For plant height, number of tillers per plant, number of panicles, weight of panicles, number of grains, test weight, single plant yield, plot yield and biomass the estimated PCV were slightly higher than GCV. For days to 50 per cent flowering, panicle length and harvest index moderate PCV and GCV were recorded. For plant height, number of tillers per plants, test weight, yield per plant and biomass yield high GCV and PCV were recorded. For panicle length moderate genetic advance along with moderate heritability was observed. High heritability coupled with high genetic advance were recorded for days to 50 per cent flowering, plant height, number of tillers per plants, number of panicles, panicle length, panicle weight, grain number, test weight, single plant yield, plot yield, biological yield and harvest index.

Kishore et al. (2018) examined 20 rice genotypes for 13 quantitative traits. Plant height showed the widest range of phenotypic variations followed by fertile spikelets per panicle, biological yield per plant, days to 50 per cent flowering, days to 75 per cent maturity, test weight, flag leaf length, harvest index, effective tillers per plant and grain yield per plant. The spikelets per panicles, test weight, grain yield per plant, harvest index, biological yield per plant and flag leaf width showed the greatest difference between GCV and PCV.

Lakshmi et al. (2018) investigated the morphological traits of African rice germplasm and found that the values of GCV and PCV were higher for the number of productive tillers per plant and spikelets per panicle. The values of GCV and PCV were discovered to be low for grain breadth and L:B ratio but observed to be moderate for days to 50 per cent flowering, plant height, effective tillers, 1000 grain weight, grain yield per plant and grain length. Days to 50 per cent flowering, plant height and number of spikelets per panicle all showed high heritability along with high genetic advance.

Bhattachraya and Chakraborty (2019) evaluated 28 aromatic rice genotype and found significant mean squares due to genotypes for each of the 16 quantitative

features indicating the presence of genetic variability. The harvest index had the highest coefficient of variation followed by the number of productive tillers per plant. High to moderate genetic advance was observed for number of filled grains, days to 50 per cent flowering and grain yield per plant along with high to moderate heritability suggested that additive gene action predominated for the expression of these traits.

Kujur et al. (2019) compiled 261 farmers variety and examined these for 30 yield as well as quality traits. The maximum PCV and GCV were found for total number of spikelets per plant followed by number of fertile spikelets per plant, plant height and days to 75 per cent maturity. The traits like days to 50 per cent flowering, days to 75 per cent maturity, 1000 grain weight, grain yield per plant, biological yield per plant, number of fertile spikelets per panicle, number of tillers per plant, number of effective tillers per plant, plant height, harvest index, grain length, grain breadth, L:B ratio and flag leaf length exhibited high heritability coupled with high genetic advance. However, medium heritability with high genetic advance was found for flag leaf width.

Kumaresan and Manonmani (2019) evaluated 21 indigenous rice varieties of Tamilnadu for morphological characters and discovered that the traits plant height, number of productive tillers per plant and grain yield per plant had high GCV and PCV. The low GCV and PCV was found for days to 50 per cent flowering. All of the studied characters including days to 50 per cent flowering, plant height, number of panicles per plant, number of productive tillers per plant, length of panicles, 1000 grain weight and grain yield per plant had PCV values slightly higher than GCV. High heritability coupled with high genetic advance was recorded for plant height, number of panicles per plant, number of productive tillers, 1000 grain weight and grain yield per plant.

Kumar (2020) investigated 30 red rice genotypes and observed that high PCV and GCV were recorded for grain yield and the number of productive tillers per plant while the lowest GCV and PCV were found for days to 75 per cent maturity and panicle length. For days to 50 per cent flowering, plant height, number of tillers per plants, number of productive tillers per plants, number of grains per panicles, number

of filled grains per panicle, L:B ratio, 1000 grain weight and grain yield per plant high heritability along with high genetic advance was found.

Singh et al. (2020) assessed 30 local red rice germplasm of Himachal Pradesh samples for morphological traits such as days to 50 per cent flowering, days to 75 per cent maturity, plant height, total tillers per plants, panicle length and grain yield. Analysis of variance showed that the samples had a high genetic diversity. Plant height and grain yield were observed to have moderate GCV and PCV. For plant height high heritability along with moderate genetic advance was detected whereas for the other examined variables high heritability along with low genetic advance was recorded.

Widyayanti and Purwaningsih (2020) examined the physical characteristics of 11 native red rice varieties in Indonesia in order to detect and assess genetic diversity as well as heritability. The number of productive tillers per plants was found to have high coefficient of variation. Plant height, number of effective tillers per plant, number of grains and number of panicles were found to have high heritability as well as a wide coefficient of variation.

Akhtar et al. (2022) evaluated 58 aromatic rice genotypes on the basis of 12 agro-morphological traits. It was found that phenotypic coefficient of variation exhibited higher values but maintained close relation with genotypic coefficient of variation for all the traits. High genetic advance was recorded for the traits like plant height, panicle length, number of filled grains per panicle and 1000 grain weight.

Alagappan and Bhardwaj (2022) investigated 35 red rice germplasm lines of Himachal Pradesh at Rice and Wheat Research Centre Malan for 12 quantitative and six qualitative traits. They found that germplasm lines showed high heritability coupled with high genetic advance as percent of mean for biological yield per plant, 1000 grain weight and L:B ratio which indicated the presence of high additive gene action.

Satya et al. (2022) investigated 48 landraces of rice to estimate the genotypic and phenotypic variability, heritability and genetic advance. The ANOVA showed significant differences for each of the investigated attributes. A minor difference between the genotypic coefficient of variation (GCV) and the phenotypic coefficient

of variation (PCV) was observed. Except for days to 75 per cent maturity all of the characters under study showed high heritability along with high genetic advance as percent of mean indicating the primacy of additive gene action.

2.2. Correlation coefficient analysis

Studies on correlation are a way to gauge how closely two variables are related and associated. Due to the possibility of indirect selection, it is crucial in plant breeding. The plant breeder may benefit from knowing how the development of one character will result in changes in other characters that occur simultaneously by studying the correlation between various characters. It is crucial to comprehend the relationships between various qualities and grain yield since grain yield is one of the most crucial traits in any breeding program. However, grain yield is a complicated character to research because of the interactions it has with numerous other traits. In order to anticipate the best cross combinations, identify the features for the optimal plant type and possibility of indirect selection plant breeders must find simple correlation and the level of indirect impacts of attributes on grain yield.

Thongbam et al. (2010) assessed the data in indigenous cultivars and found that grain length was significant and positively correlated with L:B ratio, while L:B ratio was negatively correlated with grain breadth.

Sohrabi et al. (2012) studied 50 germplasm for 12 quantitative traits and correlated the result which showed that most of the traits had positive correlation. Traits including days to 50 per cent flowering, days to 75 per cent maturity, number of grains per panicle, yield per plant, panicle length and spikelet fertility were all found to be extremely significant and positively correlated with plant height. It was noted that there was a strong association between the plant yield and days to 50 per cent flowering, days to 75 per cent maturity, plant height, number of panicles and 1000 grain weight.

Aditya and Bhartiya (2013) studied morphological features and analysed that plant height, days to 50 per cent flowering, days to 75 per cent maturity, flag leaf length, flag leaf width, grain length, L:B ratio were all positively and significantly correlated with grain yield. A significant and positive correlation between plant height

and days to 50 per cent flowering, days to 75 per cent maturity, flag leaf length, flag leaf width was also observed.

Chakravorty and Gosh (2013) investigated traditional rice varieties and correlated the data. They found a positive correlation between flag leaf length and flag leaf width. While grain weight and grain length were found to be significantly associated.

Sanghera et al. (2013) studied morphological traits of red rice at the genotypic and phenotypic levels and found significantly positive correlation between the number of tillers per plant and grain yield.

Sinha and Mishra (2013) examined 20 landraces for agro-morphological features including 20 qualitative and 13 quantitative characters. Phenotypic trait correlation analysis including 14 distinct quantitative agro-morphological features showed that days to 50 per cent flowering and days to 75 per cent maturity had the highest correlations. A highly significant association was discovered between grain length and grain breadth. L:B ratio was discovered to have a negative correlation with grain width.

Jambhulkar and Bose (2014) studied 22 upland genotypes of rice and recorded quantitative traits like days to 50 per cent flowering, plant height, total tiller per plant, grains per panicle, test grain weight and grain yield. They found that there is a significant and positive genotypic and phenotypic correlation between yield and plant height.

Ahmad et al. (2015) studied the agro-morphological traits for coloured rice and discovered a negative association between plant height and the number of tillers per plant. A positive correlation of the number of tillers and the harvest index with the grain yield per plant was observed.

Kumar (2015) examined the 57 genotypes to know about the correlation among yield and yield attributing traits at the experimental farms of the Department of Genetics and Plant Breeding, CSKHPKV, Palampur. In general the genotypic correlations were found to be higher than phenotypic ones. It was found that grain yield per plant had positive and significant associations with total tillers per plant,

effective tillers per plant, spikelet fertility, biological yield per plant and harvest index.

Sinha et al. (2015) analysed the morphological features and found that grain length and breadth were significantly positively correlated with 1000 grain weight whereas L:B ratio was significantly correlated with grain length and negatively correlated with grain width.

Rathore et al. (2016) investigated 76 populations of weedy rice from various agro-climatic zones in India. The number of grains per panicle was positively correlated with days to panicle emergence. There was a negative correlation between the number of tillers and 1000 grain weight. Days to panicle emergence and days to 75 per cent maturity were simultaneously discovered to have a significant and positive correlation. The findings demonstrated that in contrast to morphological variability in grain characteristics, all physiological factors had high correlations.

Kadidaa et al. (2017) identified the diversity of various local upland rice genotype based on agronomic features and production potential. The outcome revealed a significant positive correlation between the number of filled grains, 1000 grain weight, total number of tillers and productive tillers. Negative correlation was observed between plant height and grain yield per plant. Number of filled grains per panicle was found to be positively correlated with yield.

Puren (2017) conducted correlation studies on red and brown rice and discovered a significant positive correlation between grain yield and the number of panicles per plant, spikelets per panicle, grains per panicle, L:B ratio, number of tillers per plant and days to 50 per cent flowering at both the phenotypic and genotypic levels. Plant height, grain breadth and grain yield were all found to have a negative and significant correlation with grain yield per plant.

Archana et al. (2018) studied 38 genotypes for morphological and nutritional parameters and observed that grain yield was positively correlated with plant height, number of productive tillers per plant, number of effective spikelets per panicle, grain length, L:B ratio, 1000 grain weight and harvest index. Significant and negative correlations were observed between days to 50 per cent flowering, days to 75 per cent maturity and grain yield.

Bagudam et al. (2018) investigated 46 rice genotypes for morphological parameters such as days to 50 per cent flowering, plant height, total tillers per plant, 1000 grain weight, grain yield per plant, biological yield and harvest index. Grain yield per plant was observed to exhibit a positive and significant association with total tillers per plant, days to 50 per cent flowering, biological yield and effective tillers per plant when correlated with the yield and yield component traits that were taken into consideration. In addition, days to 50 per cent flowering showed a negative association with harvest index but a positive and significant correlation with plant height.

Kishore et al. (2018) analysed the evaluated morphological traits for correlation and found that the grain yield per plant was significant and positively associated with harvest index, plant height, biological yield per plant and test weight. While grain yield per plant was found to have a significant but negative association with effective tillers per plant. Test weight and biological yield demonstrated a positive and significant association with grain yield per plant.

Alagappan and Bhardwaj (2022) analysed 35 red rice germplasm lines of Himachal Pradesh at Rice and Wheat Research Centre Malan for 12 quantitative and six qualitative traits and found that the grain yield per plant was discovered to be significant and positively associated with biological yield per plant followed by flag leaf length, harvest index, days to 50 per cent flowering, plant height and spikelets per panicle.

Singh et al. (2020) investigated the data collected from the traits analysed in the red rice germplasm of Himachal Pradesh and discovered a strong positive correlation between total tillers per plant and grain yield. At both the genotypic and phenotypic levels the correlation between days to 50 per cent flowering and days to 75 per cent maturity, plant height and panicle length was found to be positive. At both the genotypic and phenotypic levels plant height was found to be strongly associated with panicle length although it was also positively correlated with tillers per plant at the genotypic level only. At both the genotypic and phenotypic levels total tillers per plant simultaneously displayed a positive correlation with panicle length.

Akhtar et al. (2022) evaluated 58 aromatic rice genotypes on the basis of 12 agro-morphological traits. A correlation study showed that grain yield per plant was positively and significantly correlated with tillers per plant, panicles per plant, number of filled grains per panicle, total number of filled grains per plant and 1000 grain weight.

2.3 Path coefficient analysis

The genetic makeup of the population under consideration determines the importance of plant breeding strategies for character development. Before character development was undertaken, it was necessary to understand about the direct and indirect selection because it was one of the processes that contributed to such success. Path co-efficient analysis is a useful tool for identifying the primary and secondary causes of associations between features. It also helps in the critical investigation of the particular forces at work in a given correlation and quantifies the relative weight that each causative element has. In contrast to the correlations which only reveal the relationship between two variables, path coefficient analysis divides the correlations to reveal the relationship between two variables. The main economic characteristic of rice grain yield is dependent on a number of interrelated component qualities. One component at a time, even a small modification, might cause the complex to become unstable. As a result, it is necessary to study these associated qualities to determine how they affect grain production both directly through component character and indirectly through other component features. Therefore, a key component of any breeding programs is the analysis of the direct and indirect impacts of yield and related traits on grain yield per plant from genotypic correlation and phenotypic correlation.

Aditya and Bhartiya (2013) conducted path coefficient analysis to find out the relationships between the traits to determine the character that contributed to grain yield. They discovered that the L:B ratio followed by grain width and tillers per plant had the strongest direct and positive impact on grain yield. Grain length, L:B ratio and 1000 grain weight showed strongest positive but indirect effects. Grain yield was shown to be positively impacted by plant height, days to 50 per cent flowering, days

to 75 per cent maturity, tillers per plant, flag leaf length, 1000 grain weight and grain length.

Sanghera et al. (2013) evaluated the morphological features of red rice through genotypic path analysis and found that panicle density had the highest positive influence followed by plant height and days to 50 per cent flowering.

Jambhulkar and Bose (2014) divided the data on examining the features of 22 upland genotypes and combined it using route analysis in order to determine the direct and indirect impacts of the variables that contributed to grain yield. They found that plant height had the highest direct influence on yield followed by grains per panicle, days to 50 per cent flowering and 1000 grain weight.

Chowdhury et al. (2016) investigated the association between grain yield and micronutrient content together with other yield attributing traits to assess the impact of the direct influence on yield and yield related traits. The experiment demonstrated that 1000 grain weight and the number of filled grains per panicle had a highest positive direct influence and these were also discovered to be yield attributing traits.

Puren (2017) examined the path analysis and found that at the phenotypic level panicles per plant had the highest positive direct effect followed by spikelets per panicle, grains per panicle, L:B ratio and grain yield per plant. However, it was discovered that panicles per plants, grains per panicle and spikelets per panicle all had a significant direct and beneficial impact on the amount of grain yield per plant. At the genotypic level panicles per plant, spikelets per panicle, L:B ratio and grains per panicle were determined to have the highest contributions to grain yield per plant.

Archana et al. (2018) investigated the direct and indirect impacts of yield and components traits on grain yield per plant using genotypic and phenotypic correlation analysis on morphological characters. According to the study conducted 1000 grain weight, the number of filled grains per panicle, harvest index, plant height, grain breadth and L:B ratio displayed the most favourable direct effects on grain yield per plant. They concluded that the 1000 grain weight, L:B ratio, number of filled spikelets per panicle and harvest index all had a strong positive connection with grain yield as well as a direct impact on grain yield.

Bagudam et al. (2018) studied coefficient analysis of rice and showed that effective tillers per plant had the most positive direct influence on grain yield followed by 1000 grain weight, harvest index and biological yield. The number of tillers per plant, plant height and days to 50 per cent flowering were found to have a negative direct impact on grain yield.

Kishore et al. (2018) analysed yield contributing traits and divided the analysed traits into groups to identify the traits that contributed to the yield in order to boost grain yield. The biological yield per plant was discovered to have the highest direct positive influence on grain yield followed by harvest index, days to 50 per cent flowering, flag leaf width, fertile spikelets per panicle and panicle length. Test weight, fertile spikelets per plant and flag leaf length were all indirectly impacted by biological yield per plant. Test weight demonstrated a good indirect effect due to effective tillers per plant. The harvest index, flag leaf width, plant height, panicle length, days to 75 per cent maturity, test weight and days to 50 per cent flowering were also contributed indirectly to yield per plant.

2.4 Genetic diversity

The choice of germplasm is an essential and crucial step in any plant breeding program such as improvement of existing varieties or to produce hybrids and can determine the success or failure of the program. The study of genetic divergence helps in the choice of genotypes to be used in breeding programs for the development of new populations. Genetic diversity is related to the degree of distance between populations in the set of genetic traits that differ between the populations.

Genetic diversity refers to the variations among the alleles of a gene and it may be examined at nucleotide level in the DNA sequence. Various classical and DNA tools are available to access genetic diversity at morphological level and can be expressed in the form of dendrogram and genetic distance (Singh et al. 2021). Genetically diverse genotypes are used as valuable source by the plant breeders for the development of new or improved crop varieties with desirable traits to cope up the biotic and abiotic stresses such as drought tolerance, salt tolerance, insect pest and disease resistance etc.

2.4.1 Morphological diversity

D^2 analysis is a very useful technique in quantifying the degree of divergence between inbred lines or any biological population at genotypic level. It is also helpful in assessment of relative contribution of different components to the total divergence at both intra and inter-cluster level. It measures the force of differentiation at intra and inter cluster levels. The varieties were grouped into number of clusters as per the standard procedure. D^2 statistic developed by (Mahalanobis 1936) provides a measure of magnitude of relative contribution of each component character to the total divergence. Mahalanobis D^2 statistic is more reliable in selection of potential parent for hybridization program to get the desirable recombinants in segregating generation. It is being extensively used to meet the specific breeding objectives.

The idea of principal component analysis was given by Karl Pearson. The PCA is used as a data reduction tool in exploratory data analysis and for making predictive models. Principal component analysis also decreases the number of variables responsible for the highest percentage of total variance of the experimental data. It allows the relationship between variables and observations to be studied, as well as recognizing the data structure (Pearson 1901).

Asish et al. (2010) investigated genetic divergence of fifty-one breeding lines/cultivars of rice studied for 14 physico-chemical quality characteristics of different duration groups *viz.*, early, mid-early, medium and long duration. The genotypes were grouped into thirteen clusters, out of which cluster III with seventeen varieties was the largest.

Padmaja et al. (2011) analysed the genetic divergence in 150 genotypes of rice by using Mahalanobis D^2 statistics and revealed the existence of considerable diversity among the genotypes. On the basis of D^2 values the genotypes were categorized into 13 clusters. Cluster XI was the largest consisting of 28 genotypes while cluster I contained only two genotypes. Cluster I showed maximum inter-cluster distance with cluster IX indicating that the genotypes from these clusters may be used as potential donors in hybridization program to obtain desirable recombinants.

Parikh et al. (2011) evaluated the 71 rice accessions to study the diversity pattern among the genotypes. The genotypes were grouped into eight clusters. The

mode of distribution of genotypes from different geographic regions into various clusters was at random indicating no association between geographical distribution of genotypes and genetic divergence. The inter-cluster distance indicating wider genetic diversity among the accessions of different groups.

Sharma et al. (2011) evaluated the genetic divergence using D^2 statistic in 63 rice genotypes and grouped them into eight different clusters, in which, cluster VII was the largest having 16 genotypes. Based on mean performance, genetic distances and clustering pattern the genotypes Pusa Basmati-1 and Taraori Basmati of cluster II; Sarju52, Narendra Usar-3 and Narendra-80 of cluster IV; Badshah Bhog, MTU-7029 and BPT5204 of cluster VI and Malviya-36, Kanakjeer and Super Basmati of cluster VIII proved to be promising genotypes for use in hybridization program.

Vennila et al. (2011) evaluated 41 rice genotypes for yield and yield attributing characters. The analysis of variance revealed significant differences among the genotypes for all the characters studied. Based on the genetic distance, all the 41 genotypes were grouped into 13 different clusters. The maximum inter-cluster distance was recorded between clusters III and XIII and the maximum intra-cluster distance was found in cluster XI followed by VI. The characters like number of grains per panicle, plant height, grain length and grain breadth contributed maximum towards genetic diversity.

Parikh et al. (2012) evaluated the 71 rice accessions from Madhya Pradesh and Chhattisgarh to study the diversity pattern among the genotypes. The genotypes were grouped into eight clusters. The highest inter-cluster distance was observed between cluster VI and VIII (6.51) followed by cluster II & VII (5.56) showing wide diversity among the groups. The highest intra-cluster distance was observed for the cluster VII and the lowest for the cluster V.

Chakravorty et al. (2013) assessed genetic divergence among 51 rice genotypes and grouped them into 11 clusters. Cluster II was found to be the largest comprising 16 genotypes followed by cluster III having eight genotypes and cluster I included seven genotypes. Cluster VI and XI had single genotype. The characters *viz.*, tiller diameter, tiller length and grain length contributed maximum towards genetic divergence. Principal component analysis revealed that six quantitative characters *viz.*,

leaf length, number of tillers per plant, tiller diameter, number of grains per panicle, L:B ratio and grain length significantly influenced the variation in these landraces.

Devi et al. (2015) evaluated 92 rice cultivars using Mahalanobis D^2 statistic. Based on cluster analysis, the genotypes were grouped into ten clusters of which clusters VII and IX are the largest clusters consisting of 16 genotypes each while cluster V was the smallest with only a single genotype. The maximum intra cluster distance (D = 14066.5) was found in cluster VI consisting of two traditional varieties AS 100 and Chittimutyalu. The most divergent clusters found were Clusters V and VI. Minimum inter cluster distance (D = 5144.43) was found between Clusters VIII and IX. 1000 grain weight and head rice recovery were found to be the most contributing traits towards genetic diversity.

Kumar (2015) estimated the diversity among 57 genotypes at the experimental farms of the Department of Genetics & Plant Breeding, CSKHPKV, Palampur. On the basis of D^2 values 57 genotypes were grouped into nine distinct overlapping clusters which suggested the presence of high degree of diversity. Cluster II and IV accommodated maximum 14 genotypes each. The inter cluster distances were found to be higher than intra cluster distances which indicated wider genetic divergence among the genotypes. Cluster mean values showed high variation for all the characters except total tillers per plant and effective tillers per plant.

Rachappanavar (2017) assessed the genetic diversity among 30 genotypes using D^2 statistic which results in the grouping of genotypes into six clusters indicating the presence of considerable genetic diversity among all the genotypes. It was found that the contribution of seed yield per plot was highest towards genetic divergence followed by grain length, days to panicle completion, days to pollen shed, seeds per panicle, grain width, plant height, 1000 seed weight, leaf length, panicle length, effective tillers, leaf width, leaf area index and days to panicle initiation.

Singh et al. (2018) collected 418 wild rice accessions and planted them in *Kharif* season of each year from 2011 to 2014 at Indian Agricultural Research Institute, New Delhi, India. The accessions were subjected to Principal component analysis which revealed continuous variation for the studied morphological traits in each ecotype group.

Yadav et al. (2019) investigated genetic diversity at the 24 most important blast resistance gene loci utilizing 28 gene-specific markers. Total 161 landraces from all over India were used. The principal coordinate analysis classified the landraces into two sub–populations.

Singh et al. (2020) analysed 30 red rice local germplasm collected from different areas of Himachal Pradesh to study genetic diversity. Generally the local germplasm was divided into two clusters- clusters I and II. Three lines *viz.*, Bayal-k, Bayal-1 and Bayal-p made up Cluster I whereas 27 lines made up Cluster II. Cluster II was further broken into two sub clusters - clusters 2A and 2B each had 6 and 21 lines respectively. Groupings I and II were distant clusters based on agro-morphological features, allowing lines from these clusters to be chosen for the hybridization program.

Akhtar et al. (2022) evaluated 58 aromatic rice genotypes on the basis of 12 agro-morphological traits to characterize and estimate genetic diversity. Based on Manhattan clustering, 58 genotypes were grouped into five distinct clusters. 24 genotypes were found in cluster III, 17 in cluster I, 9 in cluster II, 7 in Cluster IV and 1 genotype was found in Cluster V. The principal component analysis revealed that total number of filled grains per plant had a strong relation with grain yield.

Satya et al. (2022) investigated 48 landraces of rice to estimate the genetic divergence which were divided into ten different clusters using cluster analysis. The research aided in understanding how genetic diversity among genetic resources varied.

2.4.2 Molecular diversity

2.4.2.1 Molecular Markers Applications

Estimation of genetic diversity using molecular techniques is more reliable as it is based on highly polymorphic molecular markers which remain unaffected by the influence of environment (Singh et al. 2021).

Fisseha et al. (2013) analysed genetic diversity in 24 rice cultivars using 29 SSR markers and detected the 144 alleles. The number of alleles per loci ranged from 3 to 8 with a mean value of 4.966 alleles per locus.

Netravati et al. (2013) used 12 SSR markers to study the genetic diversity in 48 aromatic rice varieties. A total of 28 bands appeared. The number of alleles per locus ranged from 1 to 5 with an average of 2.08 alleles per locus.

Vinita et al. (2013) studied the genetic diversity in 41 rice genotypes using 24 microsatellite markers. The number of alleles per microsatellite locus ranged from 2 to 4 with an average value of 2.79 alleles per locus. Polymorphism information content values ranged from 0 to 0.66 with an average of 0.38. The dendrogram based on molecular marker analysis grouped the 41 rice cultivars into four diverse groups.

Shahriar et al. (2014 a) used three advanced breeding lines with 3 SSR markers to study the diversity analysis and detected a total of 29 alleles among the rice genotypes with an average of 9.67 alleles per locus. Polymorphism information content ranged from 0.47 to 0.88 with an average value of 0.71. A dendrogram was constructed based on total microsatellite polymorphism and 34 genotypes were grouped into four major clusters at 0.36.

Shahriar et al. (2014 b) used 19 SSR markers to study the diversity analysis in 24 rice genotypes and identified a total of 110 reproducible polymorphic alleles from the loci with an average of 5.79 alleles per locus (ranged from 3-12 alleles). The PIC values ranged from 0.37 (RM18) to 0.87 (RM493) with an average of 0.65. The UPGMA cluster revealed six main genetic groups at a cut off value of 32% of similarities comprising of three separate clusters.

Ramadan and Elmoghazy (2015) used 46 SSR markers to study the genetic diversity in seven rice genotypes for drought tolerance. The results indicated that among the total SSR markers used, 43 SSR loci were polymorphic and produced 127 alleles. The number of alleles per locus generated by each marker varied from 2 to 6 alleles with an average of 2.76 alleles per locus. The overall size of amplified fragments ranged from 93 to 487 bp. The PIC values ranged from 0.21 to 0.79 with an average of 0.46. Out of the used polymorphic SSR markers 19 markers were highly informative (PIC > 0.50), 21 markers were informative (0.50 < PIC < 0.25) and three markers were slightly informative (PIC < 0.25).

Salgotra et al. (2015) used 40 microsatellite markers to study genetic diversity in 141 basmati rice genotypes and detected 112 alleles with the maximum and

minimum PIC values of 0.63 and 0.17 for the primers RM206 and RM213 respectively.

Venkatesan and Bhat (2015) evaluated the genetic diversity and relationship among 40 aromatic rice through microsatellite marker (SSR) analysis using 24 primer pairs, of which 22 (91.6%) were polymorphic. In total 51 alleles were detected for 22 polymorphic primer pairs with an average of 2.3 alleles per locus. Polymorphism Information Content values ranged from 0.05 to 0.57 with an average of 0.33. Four SSR loci revealed PIC values higher than 0.50. 40 aromatic rice genotypes were grouped into two major clusters at simple matching (SM) coefficient value of 0.48.

Ashraf et al. (2016) estimated 16 rice genotypes found in Western Himalayas of Kashmir and Himachal Pradesh with a set of 24 SSR markers. A total of 68 alleles were detected and number of alleles per locus generated varied from 2 to 3. The PIC values varied from 0.36 to 0.86 with an average of 0.62 per locus and the genotypes got separated in six different clusters.

Thomas and Dominic (2016) assessed the genetic diversity among 25 coastal rice populations of five regions of Kerala using 18 microsatellite markers. A mean PIC value of 0.37 and an average of 3.5 alleles per loci were observed. Mean heterozygosity value of 0.29 and gene diversity value of 0.41 was attained.

Rachappanavar (2017) assessed the genetic diversity among 30 genotypes with a set of 48 SSR markers out of which 27 were polymorphic. Total 74 alleles were detected. The average number of alleles per locus detected was 2.74. PIC value ranged from 0.08 to 0.61 with an average of 0.42. A dendrogram was constructed using UPGMA method which grouped the genotypes into five clusters.

Bisht et al. (2018) analysed the genetic diversity of eight aromatic landrace rice populations from four eco-geographic regions using 17 SSR markers. 69 unique alleles were recorded. In terms of effective number of alleles, predicted heterozygosity and Shannon's information index Kala Joha from Assam was found to be most diversified. Maximum variation among populations inside groups was found by analysis of molecular variance (AMOVA), followed by maximum variation among groups (30.52%) and remaining variation (30.12%) within populations.

Singh et al. (2018) collected 418 wild rice accessions for genetic diversity with a set of 24 SSR markers and planted them in *Kharif* season at Indian Agricultural Research Institute, New Delhi, India. The 24 SSR markers showed total 96 alleles. Major allele frequency (A) ranged from 0.33 (HvSSR05-37) to 0.90 (HvSSR06-34), gene diversity (G) ranged from 0.17 (HvSSR06-34) to 0.75 (HvSSR07-33), heterozygosity (Ho) ranged from 0.0 (HvSSR05-07) to 0.74 (HvSSR07-33) and PIC ranged from 0.16 (HvSSR06-34) to 0.72 (HvSSR07-33). A dendrogram was generated based on Chord frequency distance by UPGMA method, which grouped the accessions into three major clusters.

Sood et al. (2018) investigated 47 landraces of rice that were collected from various agroclimatic zones in Himachal Pradesh along with three control varieties using molecular markers to determine their genetic relationships. 15 RAPD and 11 ISSR markers were detected with a higher amount of polymorphism, however the ISSR polymorphism percentage was larger than the RAPD. Cluster I contained 44 of the 50 landraces. The majority of the genotypes being grouped into one cluster revealed affinities between the genotypes showing their common ancestry. According to RAPD, the landraces "IC 3131155" and "Sukara" were the most divergent, however the ISSR results indicated that "Local Variety" and "Lalzhini" were the most diversified.

Dwivedi et al. (2019) studied the genetic diversity of 30 rice accessions both of basmati and non-basmati including two red rices collected from Rice and Wheat Research Centre, Malan. Analysis was performed with the set of 36 genome wide SSR markers. Five major clusters were formed as a result of molecular characterization. SSR markers detected a total of 83 alleles ranged from 2-4. The PIC value among SSR loci ranged from 0.062 to 0.664.

Yadav et al. (2019) investigated the genetic diversity at the 24 most important blast resistance gene loci utilizing 28 gene-specific markers. Total 161 landraces from all over India were used. The cluster analysis grouped entire 161 landraces into two major groups. The population structure analysis classified the landraces into two sub–populations. Analysis of molecular variance (AMOVA) exhibited maximum (93%) diversity within the population and least (7%) diversity between populations. Five

markers including K3957, Pikh, Pi2–i, RM212 and RM302 were strongly associated with blast disease.

Hazarika and Deka (2021) evaluated the genetic diversity of aromatic rice accession of Assam for 14 genotypes. Thirty two random SSR markers and one gene-based marker were included in the molecular genetic study out of which 18 informative SSR were used for genetic analysis based on their amplification and resulted in significant variation between the studied aromatic lines. PIC value ranged from 0.1326 to 0.5408.

2.5 Disease reaction to Blast

Puren (2017) examined red and black rice genotypes and discovered that Bongaldhan, Desidhan and IC 3131180 were resistant to leaf and neck blast in both synthetic epiphytotic conditions and actual field conditions. For disease resistance, the red rice genotypes Bongaldhan, Brighudhan, HPR 2800 and ACC 19186 were shown to be promising.

Mau et al. (2018) investigated blast resistance levels in Indonesian highland cultivars of black and red rice. Four races of blast were injected into 42 different rice genotypes and the severity of the illness was utilized to categorize the resistance to blast of the genotypes that were evaluated. The outcome showed that rice genotype by blast race had a highly significant interaction on either the duration of infection or the severity of the sickness. For the inoculation of the races 033 and 073 and the races 133 and 173 the examined rice genotypes were divided into six resistance levels and five resistance levels respectively. Three local genotypes showed a high level of blast resistance along with 11 genotypes showed resistance to a single race and 12 genotypes showed resistance to many races.

Yadav et al. (2019) investigated the total 161 landraces from all over India for their leaf blast resistance under natural condition in the Uniform Blast Nursery (UBN) at the experimental farm of ICAR–NRRI, Cuttack. In 2015 and 2016 two replications of the leaf blast screening were performed twice each over the two wet seasons. Based on the Standard Evaluation System (SES) of IRRI, the landraces were divided into three distinct groups: highly resistant (21), moderately resistant (70) and susceptible (70).

Janthasri and Parinthawong (2020) analysed landrace rice Dawk Pa-yawm Rai because of its high resistance to 18 blast isolates, moderate resistance to five blast isolates and susceptibility to two blast isolates. After identifying the genetic basis of blast resistance in the F_2 population, disease evaluation revealed that 524 F_2 plants were resistant to the disease and 206 F_2 plants were vulnerable. 3:1 division of susceptible and resistant individuals was shown to have a satisfactory match (R:S). The findings indicated that this may be a significant factor and valuable tool for mapping the blast resistance gene in the Dawk Pa-yawm Rai variety.

Mustikarini et al. (2020) analysed red rice in order to find an appropriate line of red rice that is resistant to blast illness. According to the findings, different red rice lines responded differently to the blast illness attack. Race blast 033 was resistant to the ten red rice lines whereas races 073, 133 and 173 were resistant to nine red rice lines, seven red rice lines and ten red rice lines respectively. The highest resistance was found in the red rice lines 19i-06-09-23-27, 19i-06-09-23-3 and 19i-06-30-17-17.

Alagappan and Bhardwaj (2022) investigated 35 red rice germplasm lines of Himachal Pradesh in Rice and Wheat Research Centre Malan for 12 quantitative and six qualitative traits. The genotypes IC-12180, HPR-2800, HPR2795, HPR2913 and HPR-2914 were found to be resistance to both leaf and neck blast under open field conditions.

3. MATERIALS AND METHODS

The present investigation entitled "Agro-morphological and molecular characterization of red rice (*Oryza sativa* L.) germplasm of Himachal Pradesh" was carried out during *Kharif*, 2021 at the experimental farm of Rice and Wheat Research Centre, Malan. Geographically, the farm is located at 32^0 1'N Latitude and 76 ^{0}20'E longitude, 5km from the famous Chamunda Temple in the lap of majestic Dhauladhar range of North Western Himalayas representing the mid hill zone (Zone-II) of Himachal Pradesh and is characterized by humid sub-temperate climate with high rainfall (2500 mm/annum). This place is 950 m above mean sea level. The molecular work was carried out in the Molecular Cytogenetics & Tissue Culture Lab of the Department of Genetics and Plant Breeding. The details of the materials used and methods employed in the present investigation are described in this chapter under the following headings:

3.1 Materials and layout of the design

3.2 Observations recorded

3.2.1 Agro-morphological analysis

3.2.2 Reaction to diseases

3.3 Statistical analysis

3.4 Molecular analysis

3.1 Materials and layout of the design

The experimental material for the present study comprised of 39 red rice and 2 white rice genotypes along with two checks namely HPR-2720 and HPR-2795. These genotypes and checks were assessed for 18 different agro-morphological characters which include twelve quantitative and 6 quality traits in Randomized Complete Block Design during *Kharif*, 2021. Each entry was raised in plot size of 3m x 0.4m with row to row and plant to plant spacing of 20 cm and 15 cm, respectively (having 2 rows per genotype) in 3 replications.

Recommended package of practices were followed for raising the crop. The details of genetic stocks are given in Table 3.1.

Table 3.1 List of germplasm lines of red rice evaluated under present study

Sr. No.	Genotypes	Sr. No.	Genotypes	Sr. No.	Genotypes
1	IC-12180	16	Varundhan	31	HPR-2913
2	Kaluna	17	Hatiali	32	HPR-2914
3	Ramjuwain	18	HPR-2800	33	Gocha
4	Chohartu	19	Naggardhan	34	New Chaina-21
5	Sukara	20	Roda Dhan	35	Old Chaina-21
6	Karad	21	Nailina	36	Phulpatas-21
7	Kalaina	22	Deval	37	Karad 21-1
8	IC-12164	23	Bhrigudhan	38	Karad 21-2
9	Acchoo	24	Kalijhini	39	Karad 21-3
10	Begmi	25	Matali	40	Karad 21-4
11	Bathidhan	26	Dodadhan	41	Jattu
12	HPR-2906	27	HPR-2902	**42**	**HPR-2720 (C)**
13	Desidhan	28	HPR-2904	**43**	**HPR-2795 (C)**
14	Kalijhini-2	29	HPR-2905		
15	Sukara red	30	HPR-2908		

3.2 Observations recorded

Field data were recorded on the basis of five randomly selected plants for each genotype in each replication except for days to 50% flowering and days to 75% maturity for which data was recorded on the plot basis. The data from the selected five plants was used to calculate the average.

3.2.1 Agro-morphological analysis

A. Quantitative traits:

Table 3.2 List of quantitative traits recorded in the study

S. No.	Trait	Observation Recorded
1.	**Days to 50% flowering**	The number of days were recorded from the date of sowing to the 50% flowering of the plants on plot basis in a row of genotype in each replication.
2.	**Days to 75% maturity**	The number of days were recorded from the date of sowing to the date of 75% maturity on plot basis in each genotype in each replication.
3.	**Plant height (cm)**	The average height of the selected plants were measured in cm at maturity from the ground level to the tip of the main panicle, excluding awns, if any.
4.	**Flag leaf length (cm)**	Flag leaf is the uppermost leaf, its length was measured in cm.
5.	**Flag leaf width (cm)**	Width of the flag leaf was measured in cm.
6.	**Total tillers per plant**	The total number of tillers per plant at maturity were recorded.
7.	**Effective tillers per plant**	Effective tillers are those who actually contribute to the grain yield. The effective tillers per plant (tillers bearing spike) were recorded at the time of maturity.
8.	**Spikelets per panicle**	Spikelets are the basic inflorescence unit of rice. Total number of spikelets on the main panicle of the sampled plants were recorded at maturity and their average number was worked out.

9.	**1000-seed weight (g)**	A random sample of thousand well filled grains/replication from the bulk produce of each genotype were recorded after threshing and weighed in gram.
10.	**Biological yield per plant (g)**	Sun dried weight of the total biomass above ground level was recorded from all the five **s**elected plants of each genotype after harvesting and averaged.
11.	**Harvest index (%)**	Harvest index was calculated by dividing seed yield per plant by biological yield per plant expressed as: $\frac{\text{Grain yield per plant}}{\text{Biological yield per plant}} \times 100$
12.	**Grain yield per plant (g)**	Panicles harvested from each plant were dried, hand threshed, grain cleaned, dried and weighed in grams.

B. Quality traits

Each entry was rated in each replication for the following quality traits (Table 3.3).

Table 3.3 List of quality traits recorded in the study

S. No.	Trait	Observation Recorded
1.	**Seed coat color**	Outer coat of rice grain (lemma and palea) were removed to check the seed coat colour.
2.	**Grain length [L] (mm)**	Length of five de-husked grains of each genotype from the bulk produce of each replication was recorded in mm using digital vernier calliper.
3.	**Grain breadth [B] (mm)**	Five de-husked grains of each genotype, whose length was earlier recorded used for recording grain breadth in mm using digital vernier calliper.
4.	**L:B ratio**	L: B ratio was calculated by dividing the grain length by its breadth as recorded above.
5.	**Lodging susceptibility (%)**	Lodging susceptibility was calculated on plot basis in each genotype in each replication.

		Lodging susceptibility was classified as: • 50% lodging • 25% lodging • No lodging
6.	**Awns**	Grains were checked for presence of awns and classified as: • Awned • Awnless

3.2.2 Evaluation of germplasm:

Rice germplasm consisting of 43 red rice genotypes was screened for leaf and neck blast as per SES 0-9 scale (IRRI, 2014).

Leaf blast evaluation:

Screening of genotypes for leaf blast resistance was done under natural epiphytotic conditions at RWRC, Malan, during *Kharif*, 2021 (Table 3.4) and their disease reaction was recorded from five plants per genotype on 0-9 scale using Standard Evaluation System (SES) for rice (IRRI, 2014).

Table 3.4 Evaluation of rice genotypes for leaf blast resistance

Score	Description	Disease reaction
0	No lesions observed	Immune
1	Small brown specks of pin-point size or larger brown specks without sporulating center	Resistant
3	Lesion type is the same as in scale 2, but significant number of lesions are on the upper leaves	Moderately Resistant
5	Typical blast lesions infecting 4-10% of the leaf area	Moderately Susceptible
7	Typical blast lesions infecting 26-50% of the leaf area	Susceptible
9	More than 75% leaf area affected	Highly Susceptible

Neck blast evaluation:

Evaluation of genotypes for neck blast resistance was also done under natural epiphytotic conditions at RWRC, Malan, during *Kharif*, 2021 (Table 3.5). The disease reaction of genotypes was recorded from five plants per genotype on 0-9 scale using Standard Evaluation System (SES) for rice (IRRI, 2014).

Table 3.5 Evaluation of rice genotypes for neck blast resistance

Score	Description	Disease reaction
0	No incidence	Immune
1	Less than 5% infected panicles	Resistant
3	5-10% infected panicles	Moderately Resistant
5	11-25% infected panicles	Moderately Susceptible
7	26-50% infected panicles	Susceptible
9	More than 50% infected panicles	Highly Susceptible

3.3 Statistical analysis

The observations recorded as above for the various yield and yield contributing characters were subjected to the following statistical analysis.

3.3.1 Analysis of variance

The data for different characters was analyzed as per Panse and Sukhatme (1989):

$$Y_{ij} = m + g_i + r_j + e_{ij}$$

Where,

Y_{ij} = phenotypic observation of i^{th} genotype grown in j^{th} replication

m = general population mean

g_i = effect of i^{th} genotype

r_j = effect of j^{th} replication

e_{ij} = error associated with i^{th} genotype in the j^{th} replication

On the basis of this model the analysis of variance was done as follows:

Table 3.6 Analysis of variance

Source of variation	Degree of freedom	Sum of squares	Mean sum of squares	F ratio	Expected MS
Replications(r)	(r-1)	Sr	Mr=Sr/(r-1)	Mr/Me	$\sigma^2e+g\sigma^2r$
Genotypes (g)	(g-1)	Sg	Mg=Sg/(g-1)	Mg/Me	$\sigma^2e+r\sigma^2g$
Error (e)	(r-1) (g-1)	Se	Me=Se/(r-1)(g-1)	-	σ^2e
Total	(rg-1)	-			

Where,

r = number of replications

g = number of genotypes

σ^2e = error variance = Me

σ^2g = variance due to genotypes = (Mg-Me)/r

σ^2r = variance due to replications = (Mr-Me)/g

σ^2p = phenotypic variance = $\sigma^2g + \sigma^2e$

The standard error of mean SE (m) (±), standard error of difference SE (d) (±) and critical difference (CD at 5%) for comparing the means of any two lines were computed as follows:

Standard error (SE) of mean

$$SE(m) = \pm \sqrt{\frac{Me}{r}}$$

Standard error (SE) of difference

$$SE(d) = \pm \sqrt{\frac{2Me}{r}}$$

Critical difference (CD)

CD = SE (d) ×'t'

Where,

SE (d) ± = Standard error of difference

't' = tabulated value of 't' at 5% level of significance at error degree of freedom.

Coefficient of variation (CV)

The coefficient of variation was calculated as per the following formulae:

$$CV\ (\%) = \frac{\sqrt{M_e}}{Grand\ mean} \times 100$$

The calculated 'F' values were compared with the tabulated 'F' values at 5% level of significance. If the calculated 'F' value was higher than the tabulated, it was considered to be significant. All the characters which showed significant differences among genotypes were further subjected to the analysis for the different parameters.

3.3.2 Estimation of parameters of variability

The genotypic, phenotypic and environmental coefficients of variation were estimated following Burton and De Vane (1953):

$$\text{Genotypic coefficient of variation (GCV \%)} = \frac{\sqrt{\sigma^2_g}}{\overline{X}} \times 100$$

$$\text{Phenotypic coefficient of variation (PCV \%)} = \frac{\sqrt{\sigma^2_p}}{\overline{X}} \times 100$$

$$\text{Environmental coefficient of variation (ECV \%)} = \frac{\sqrt{\sigma^2_e}}{\overline{X}} \times 100$$

Where,

$\sqrt{\sigma^2_g}$ = genotypic standard deviation

$\sqrt{\sigma^2_p}$ = phenotypic standard deviation

$\sqrt{\sigma^2_e}$ = environmental standard deviation

$\overline{x}$ = population mean

3.3.3 Heritability (%)

Heritability in broad sense (h^2_{bs}) was calculated as per the following formula given by Burton and De Vane (1953) and Johnson et al. (1955).

$$\text{Heritability (broad sense) } (h^2_{bs}) = \frac{\sigma^2_g}{\sigma^2_p} \times 100$$

Where,

h^2_{bs} = heritability

σ^2_g = genotypic variance and

σ^2_p = phenotypic variance

3.3.4 Genetic advance (%)

The expected genetic advance (GA) resulting from the selection of 5% superior individuals was calculated as per Burton and De Vane (1953) and Johnson et al. (1955).

$$GA = K \times \sigma_p \times h^2_{bs}$$

Where,

K = 2.06 (selection differential at 5% selection intensity)

h^2_{bs} = heritability (broad sense) $\left(\frac{\sigma^2 g}{\sigma^2 p}\right)$

σ_p = phenotypic standard deviation

$$\text{Genetic advance as percentage of mean (GA \%)} = \frac{\text{Expected GA}}{\text{Grand Mean}} \times 100$$

For categorizing the magnitude of different parameters, the following limits were used:

PCV and GCV	> 20%	High
	10% - 20%	Moderate
	< 10%	Low
Heritability (h^2_{bs})	>80%	High
	50% - 80%	Moderate
	< 50%	Low
Genetic advance (GA)	> 20%	High
	10 % - 20%	Moderate
	< 10%	Low

3.3.5 Estimation of correlation coefficients:

For computing phenotypic, genotypic and environmental correlation coefficients, analysis of co-variance was carried out in all possible pairs of combinations of the characters following Al-Jibouri et al. (1958) and Dewey and Lu (1959).

Table 3.7 Analysis of co-variance

Source of variation	df	Mean sum of product	Expected mean sum of product
Replications(r)	(r-1)	Mr_{xy}	$\sigma e_{xy} + g.\sigma r_{xy}$
Genotypes(g)	(g-1)	Mg_{xy}	$\sigma\, e_{xy} + r.\sigma g_{xy}$
Error (e)	(r-1)(g-1)	Me_{xy}	σe_{xy}

Where,

R = No. of replications

G = No. of genotypes

σe_{xy} = Environmental co-variance of character x and character y

σg_{xy} = Genotypic co- variance of character x and character y

σp_{xy} = Phenotypic co- variance of character x and character y

The genotypic, phenotypic and error co-variances were calculated as follows:

Genotypic co-variances (σg_{xy}) = Mg_{xy} - Me_{xy} / r

Phenotypic co-variances (σp_{xy}) = σg_{xy} + σe_{xy}

Environmental co-variances (σe_{xy}) = Me_{xy}

Phenotypic coefficient of correlation (rp_{xy})

$$rp_{xy} = \frac{\sigma p_{xy}}{(\sigma^2 px \times \sigma^2 py)^{1/2}}$$

Where,

σp_{xy} = phenotypic covariance between characters x and y

$\sigma^2 p_x$ = phenotypic variance of character x

$\sigma^2 p_y$ = phenotypic variance of character y

Genotypic coefficient of correlation (rg_{xy})

$$(rg_{xy}) = \frac{\sigma g_{ym}}{(\sigma^2 g_x x \, \sigma^2 g_y)^{1/2}}$$

Where,

σg_{xy} = genotypic co-variance between character x and y

$\sigma^2 g_x$ = genotypic variance of character x

$\sigma^2 g_y$ = genotypic variance of character y

Environmental coefficient of correlation (re_{xy})

$$(re_{xy}) = \frac{\sigma e_{xy}}{(\sigma^2 e_x \times \sigma^2 e_y)^{1/2}}$$

Where,

σe_{xy} = environmental co-variance between character x and y

$\sigma^2 e_x$ = environmental variance of character x

$\sigma^2 e_y$ = environmental variance of character y

Test of significance

The significance of phenotypic coefficient of correlation at (g–2) degrees of freedom and environmental coefficient of correlation at [(r–1) (g–1) –1] degrees of freedom, where r and g stand for number of replication and number of genotypes, respectively, were tested at 5% level of significance against the table values of correlation coefficient (Fisher and Yates 1963).

To test the significance of genotypic coefficient of correlation, the F value was calculated using:

$$F = [(g–2)\ r^2]/\ (1–r^2)$$

and compared with the F distribution at 1 and (g–2) degrees of freedom, where g and r stand for number of genotypes and genotypic coefficient of correlation, respectively (Mead and Curnow 1983).

3.3.6 Path coefficient analysis

Path coefficient is a standardized partial regression coefficient, which permits the partitioning of the correlation coefficients into direct and indirect effects. The genotypic correlation coefficients and phenotypic correlation coefficients were used in finding out their direct and indirect contribution towards yield/plant as proposed by Wright (1921). The path coefficient analysis of important component quality traits with grain yield was carried out by following Dewey and Lu (1959) as under:

$Py_1 + Py_2.r_{12} + Py_3.r_{13} + \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots + Py_n.r_{1n} = ry_1$

$Py_1.r_{12} + Py_2 + Py_3.r_{23} + \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots + Py_n.r_{2n} = ry_2$

$Py_1.r_{13} + Py_2.r_{23} + Py_3 + \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots + Py_n.r_{3n} = ry_3$

:

:

:

:

$Py_1.r_{1n} + Py_2.r_{2n} + Py_3.r_{3n} + \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots + Py_n.r_{(n-1)n} = ry_n$

Where,

Py_1, Py_2, Py_3 ……..., Py_n are the direct path effects of 1, 2, 3, ……..., n variables on the dependent variable "y"

r_{12}, r_{13}, ……..., $r_{(n-1)\,n}$ are the coefficients of correlation between various independent variables and ry_1, ry_2, ry_3, ……, ryn are the correlation coefficients of independent variables with dependent variable "y"

The variation in the dependent variables which remained undetermined by including the other variables was assumed to be due to the variables (s) not included in the present investigation. The degree of the determination (P^2xR) of such variables was calculated as follows:

Residual effect (P x R) = $\sqrt{(1-R^2)}$

Where,

$R^2 = py_1.ry_1 + py_2.ry_2 +$ ---------- $+ py_n.ry_n$

or

$$R^2 = \sum_{i=1}^{n} p_{iy}.r_{iy}$$

Where,

R^2 is the square multiple correlation coefficient and is the amount of variation in yield that can be accounted by the yield component characters included in the present investigation. For performing path analysis adjusted treatment means have been used.

3.3.7 Estimation of genetic diversity

A measure of group distance based on multiple characters was given by Mahalanobis (1936).

With x_1, x_2, x_3,……., x_p as the multiple measurements available on each individual and d_1, d_2, d_3, ……,d_p as $x_1^{-1} - x_1^{-2}$, $x_2^{-1} - x_2^{-2}$, ……,$x_p^{-1} - x_p^{-2}$, respectively, being the difference in the means of two populations, Mahalanobis D^2-analysis is defined as follows:

$$pD^2 = b_1d_1 + b_2d_2 + \ldots\ldots\ldots\ldots b_pd_p$$

Here, the b_1 values are to be estimated such that the ratio of variance between the populations to the variance within the populations is maximized. In terms of variances and covariances, the D^2 value is obtained as follows:

$$pD^2 = W^{ij} (x_i^{-1} - x_i^{-2}) (x_j^{-1} - x_j^{-2})$$

where,

W_{ij} is the inverse of estimated variance covariance matric.

I. Test of significance

Using (V) statistics which, in turn, utilizes Wilk's criteria, simultaneous test of difference mean values of a number of correlated variables/characters at 'pq' d.f. (where p = number of variables/characters and q = number of germplasm-1) done as suggested by Rao (1952).

Testing of significance of D^2 values

The D^2 values obtained for a pair of population was taken as the calculated value of χ^2 and was taken against the tabulated value of χ^2at 'p' d.f., where p is the number of characters.

II. Grouping of genotypes into various clusters

Using D^2 values, different genotypes were grouped into various clusters following Toucher's method as suggested by Rao (1952).

III. Average intra and inter cluster distance

Average intra-cluster $D^2 = \Sigma D_i^2 / n$

where,

ΣD_i^2 = sum of all distances between all possible combinations (n) of the genotypes included in the cluster.

Average inter-cluster distance $D^2 = \Sigma D_{ij}^2 / n_i \ldots\ldots n_j$

where,

ΣD_{ij}^2 = sum of all distances between all possible combinations ($n_i.n_j$) of the genotypes between the clusters.

n_i = number of genotypes in i^{th} cluster

n_j = number of genotypes in j^{th} cluster

IV. Cluster mean

Character means of red rice genotypes falling under different clusters in individuals as well as combined over environments were also calculated.

V. Contribution of individual towards divergence

In all combinations each character was ranked on the basis of $d_i = Y_i^j - Y_i^k$ values. Rank 1 was given to all the highest mean difference and rank 'p' to the lowest mean difference, where 'p' is the total number of characters. The contribution of individual character to the divergence has been worked out in terms of 'n' number of times it appeared first.

VI. Principal Component analysis (PCA)

The main goal of PCA is to reduce the dimensionality in a set of correlated attributes into a smaller set of uncorrelated attributes that explain the majority of variation in the original attributes and was calculated using the statistical software StatistiXL version 1.10

3.4 Molecular analysis

Simple Sequence Repeats (SSR) assay

Forty three red rice genotypes including two checks used in this study, were subjected to SSR assay as the following procedure:

3.4.1 Extraction of plant genomic DNA

For molecular analysis, genomic DNA was isolated from young leaf tissue (0.5-1 g) of each line using CTAB method (Murray and Thompson, 1980). The leaf tissues were rinsed in deionized water, dried on tissue paper discs and ground to fine powder in liquid nitrogen in autoclaved pre-cooled pestles and mortars. The ground tissue was transferred to a separate 2 ml eppendorf tubes containing 800 µl of extraction buffer (2% CTAB, 100 mM Tris, 20 mM EDTA, 1.4 mM NaCl and 1% PVP, pH 8.0) maintained at 60^0C in water bath and mixed vigorously. The mixture was incubated at 60^0C for 1 hour with occasional mixing. An equal volume of chloroform : isoamyl alcohol (24:1) was added to the tubes followed by gentle mixing. The mixture was centrifuged at 10,000 rpm for 10 minutes at 4^0C. The aqueous phase was then transferred to a fresh tube, followed by addition of 500 µl of pre-chilled isopropanol. The contents of the tubes were gently mixed and the mixture was incubated at -20^0C for 1 hour. DNA was precipitated by centrifugation at 10,000 rpm for 10 minutes using centrifuge (Sigma 2 16K).

The supernatant was drained and the resulting pellet was washed twice with 1 ml of 70% chilled ethanol. The pellet was dried in a stream of sterile air in a laminar air flow cabinet for 3-4 h. Dried DNA pellet was dissolved in 1 ml TE buffer (10 mM Tris-HCl, 0.1 mM EDTA, pH 8.0). The dissolved DNA was treated with 1 µl of RNase (10 mg/ml). The quantity and quality of DNA was estimated through electrophoresis using 0.8% agarose gel (HIMEDIA).

3.4.1.1 Purification of DNA

For purification of extracted DNA, 100 µl of phenol : chloroform : isoamyl alcohol (25:24:1) was added to the tubes followed by gentle mixing. The mixture was centrifuged at 10,000 rpm for 10 minutes at 4^0C. The aqueous phase was transferred to fresh tube, followed by addition of 200 µl of pre-chilled isopropanol. The contents of the tubes were mixed gently and the mixture was incubated at -20^0C for 30 min. DNA was precipitated by centrifugation at 10,000 rpm for 10 minutes using centrifuge.

The supernatant was drained and the resulting pellet was washed twice with 70% chilled ethanol. The pellet was dried in a stream of sterile air in a laminar airflow cabinet. Dried DNA pellet was dissolved in 80 µl TAE buffer. The quantity and quality of DNA was estimated through electrophoresis using 0.8% agarose gel (HIMEDIA).

3.4.2 Primers used

A total of 96 SSR primers were screened for their amplification given in Table 3.8, of which 21 primers were selected for use in the present study on the basis of polymorphism and reproducible amplification products.

Table 3.8 List of rice SSR primer sequences used in the present study

S. No.	SSR markers	PRIMER SEQUENCES (5′3′)	
1	RM 21	F: ACAGTATTCCGTAGGCACGG	R: GCTCCATGAGGGTGGTAGAG
2	RM 55	F: CCGTCGCCGTAGTAGAGAAG	R: TCCCGGTTATTTTAAGGCG
3	RM 84	F: TAAGGGTCCATCCACAAGATG	R: TTGCAAATGCAGCTAGAGTAC
4	RM 167	F: GATCCAGCGTGAGGAACACGT	R: AGTCCGACCACAAGGTGCGTTGTC
5	RM 130	F: TGTTGCTTGCCCTCACGCGAAG	R: GGTCGCGTGCTTGGTTTGGTTC
6	RM 206	F: CCCATGCGTTTAACTATTCT	R: CGTTCCATCGATCCGTATGG
7	RM 151	F: GGCTGCTCATCAGCTGCATGCG	R: TCGGCAGTGGTAGAGTTTGATCTGC
8	RM 224	F: ATCGATCGATCTTCACGAGG	R: TGCTATAAAAGGCATTCGGG
9	RM 219	F: CGTCGGATGATGTAAAGCCT	R: CATATCGGCATTCGCCTG
10	RM 216	F: GCATGGCCGATGGTAAAG	R: TGTATAAAACCACACGGCCA
11	RM 229	F: CACTCACACGAACGACTGAC	R: CGCAGGTTCTTGTGAAATGT
12	RM 235	F: AGAAGCTAGGGCTAACGAAC	R: TCACCTGGTCAGCCTCTTTC
13	RM 153	F: GCCTCGAGCATCATCATCAG	R: ATCAACCTGCACTTGCCTGG
14	RM 257	F: CAGTTCCGAGCAAGAGTACTC	R: GGATCGGACGTGGCATATG
15	RM 211	F: CCGATCTCATCAACCAACTG	R: CTTCACGAGGATCTCAAAGG
16	RM 265	F: CGAGTTCGTCCAAGTGAGC	R: CATCCACCATTCCACCAATC
17	RM 272	F: AATTGGTAGAGAGGGGAGAG	R: ACATGCCATTAGAGTCAGGC
18	RM 240	F: CCTTAATGGGTAGTGTGCAC	R: TGTAACCATTCCTTCCATCC
19	RM 138	F: AGCGCAACAACCAATCCATCCG	R: AAGAAGCTGCCTTTGACGCTATGG
20	RM 208	F: TCTGCAAGCCTTGTCTGATG	R: TAAGTCGATCATTGTGTGGACC
21	RM 159	F: GGGGCACTGGCAAGGGTGAAGG	R: CTTGTGCTTCTCTCTCTCTCTCTCTC
22	RM 254	F: AGCCCCGAATAAATCCACCT	R: CTGGAGGAGCATTTGGTAGC
23	RM 334	F: GTTCAGTGTTCAGTGCCACC	R: GACTTTGATCTTTGGTGGACG
24	RM 270	F: GGCCGTTGGTTCTAAAATC	R: TGCGCAGTATCATCGGCGAG
25	RM 220	F: GGAAGGTAACTGTTTCCAAC	R: GAAATGCTTCCCACATGTCT

26	RM 236	F: GCGCTGGTGGAAAATGAG	R: GGCATCCCTCTTTGATTCCTC
27	RM 248	F: TCCTTGTGAAATCTGGTCCC	R: GTAGCCTAGCATGGTGCATG
28	RM 253	F: TCCTTCAAGAGTGCAAAACC	R: GCATTGTCATGTCGAAGCC
29	RM 421	F: AGCTCAGGTGAAACATCCAC	R: ATCCAGAATCCATTGACCCC
30	RM 148	F: ATACAACATTAGGGATGAGGCTGG	R: TCCTTAAAGGTGGTGCAATGCGAG
31	RM 226	F: AGCTAAGGTCTGGGAGAAACC	R: AAGTAGGATGGGGCACAAGCTC
32	RM 184	F: ATCCCATTCGCCAAAACCGGCC	R: TGACACTTGGAGAGCGGTGTGG
33	RM 204	F: GTGACTGACTTGGTCATAGGG	R: GCTAGCCATGCTCTCGTACC
34	RM 335	F: GTACACACCCACATCGAGAAG	R: GCTCTATGCGAGTATCCATGG
35	RM 338	F: CACAGGAGCAGGAGAAGAGC	R: GGCAAACCGATCACTCAGTC
36	RM 114	F: CAGGGACGAATCGTCGCCGGAG	R: TTGGCCCCCTTGAGGTTGTCGG
37	RM 1	F: GCGAAAACACAATGCAAAAA	R: GCGTTGGTTGGACCTGAC
38	RM 17	F: TGCCCTGTTATTTTCTTCTCTC	R: GGTGATCCTTTCCCATTTCA
39	RM 452	F: CTGATCGAGAGCGTTAAGGG	R: GGGATCAAACCACGTTTCTG
40	RM 211	F: CCGATCTCATCAACCAACTG	R: CTTCACGAGGATCTCAAAGG
41	RM 180	F: CTACATCGGCTTAGGTGTAGCAACACG	R: ACTTGCTCTACTTGTGGTGAGGGACTG
42	RM 212	F: CCACTTTCAGCTACTACCAG	R: CACCCATTTGTCTCTCATTATG
43	RM 215	F: CAAAATGGAGCAGCAAGAGC	R: TGAGCACCTCCTTCTCTGTAG
44	RM 222	F: CTTAAATGGGCCATATGCG	R: CAAAGCTTCCGGCCAAAAG
45	RM 228	F: CTGGCCATTAGTCCTTGG	R: GCTTGCGGCTCTGCTTAC
46	RM 85	F: CCAAAGATGAAACCTGGATTG	R: GCACAAGGTGAGCAGTCC
47	RM 127	F: GTGGGATAGCTGCGTCGCGTCG	R: AGGCCAGGGTGTTGGCATGCTG
48	RM 230	F: GCCAGACCGTGGATGTTC	R: CACCGCAGTCACTTTTCAAG
49	RM 252	F: TTCGCTGACGTGATAGGTTG	R: ATGACTTGATCCCGAGAACG
50	RM 276	F: CTCAACGTTGACACCTCGTG	R: TCCTCCATCGAGCAGTATCA
51	RM 280	F: ACACGATCCACTTTGCGC	R: TGTGTCTTGAGCAGCCAGG
52	RM 13	F: TCCAACATGGCAAGAGAGAG	R: GGTGGCATTCGATTCCAG
53	RM 14	F: CCGAGGAGAGGAGTTCGAC	R: GTGCCAATTTCCTCGAAAAA
54	RM 413	F: GGCGATTCTTGGATGAAGAG	R: TCCCCACCAATCTTGTCTTC
55	RM 586	F: ACCTCGCGTTATTAGGTACCC	R: GAGATACGCCAACGAGATACC
56	RM 11	F: TCTCCTCTTCCCCCGATC	R: ATAGCGGGCGAGGCTTAG
57	RM 134	F: ACAAGGCCGCGAGAGGATTCCG	R: GCTCTCCGGTGGCTCCGATTGG
58	RM 25	F: GGAAAGAATGATCTTTTCATGG	R: CTACCATCAAAACCAATGTTC
59	RM 205	F: CTGGTTCTGTATGGGAGCAG	R: CTGGCCCTTCACGTTTCAGTG
60	RM 244	F: CCGACTGTTCGTCCTTATCA	R: CTGCTCTCGGGTGAACGT
61	RM 289	F: TTCCATGGCACACAAGCC	R: CTGTGCACGAACTTCCAAAG
62	RM 463	F: TTCCCCTCCTTTTATGGTGC	R: TGTTCTCCTCAGTCACTGCG
63	RM 142	F: CTCGCTATCGCCATCGCCATCG	R: TCGAGCCATCGCTGGATGGAGG
64	RM 144	F: TGCCCTGGCGCAAATTTGATCC	R:GCTAGAGGAGATCAGATGGTAG
65	RM 166	F: GGTCCTGGGTCAATAATTGGGT	R: TTGCTGCATGATCCTAAACCGG
66	RM 168	F: TGCTGCTTGCCTGCTTCCTTT	R: GAAACGAATCAATCCACGGC
67	RM 177	F: CCCTCTTAGACAGAGGCCAGAGGG	R: GTAGCCGAAGATGAGGCCGCCG

68	RM 179	F: CCCCATTAGTCCACTCCACCACC	R: CCAATCAGCCTCATGCCTCCCC
69	RM 185	F: AGTTGTTGGGAGGGAGAAAGGCC	R: AGGAGGCGACGGCGATGTCCTC
70	RM 213	F: ATCTGTTTGCAGGGGACAAG	R: AGGTCTAGACGATGTCGTGA
71	RM 246	F: GAGCTCCATCAGCCATTCAG	R: CTGAGTGCTGCTGCGACT
72	RM 247	F: TAGTGCCGATCGATGTAACG	R: CATATGGTTTTGACAAAGCG
73	RM 250	F: GGTTCAAACCAAGCTGATCA	R: GATGAAGGCCTTCCACGCAG
74	RM 259	F: TGGAGTTTGAGAGGAGGG	R: CTTGTTGCATGGTGCCATGT
75	RM 260	F: ACTCCACTATGACCCAGAG	R: GAACAATCCCTTCTACGATCG
76	RM 263	F: CCCAGGCTAGCTCATGAACC	R: GCTACGTTTGAGCTACCACG
77	RM 277	F: CGGTCAAATCATCACCTGAC	R: CAAGGCTTGCAAGGGAAG
78	RM 287	F: TTCCCTGTTAAGAGAGAAATC	R: GTGTATTTGGTGAAAGCAAC
79	RM 303	F: GCATGGCCAAATATTAAAGG	R: GGTTGGAAATAGAAGTTCGGT
80	RM 304	F: TCAAACCGGCACATATAAGAC	R: GATAGGGAGCTGAAGGAGATG
81	RM 312	F: GTATGCATATTTGATAAGAG	R: AAGTCACCGAGTTTACCTTC
82	RM 331	F: GAACCAGAGGACAAAAATGC	R: CATCATACATTTGCAGCCAG
83	RM 332	F: GCGAAGGCGAAGGTGAAG	R: CATGAGTGATCTCACTCACCC
84	RM 340	F: GGTAAATGGACAATCCTATGGC	R: GACAAATATAAGGGCAGTGTGC
85	RM 350	F: TGATCGTCGCGATTCCCGGC	R: CCCCACCCTGCGCCTCTCCC
86	RM 428	F: AACAGATGGCATCGTCTTCC	R: CGCTGCATCCACTACTGTTG
87	RM 441	F: ACACCAGAGAGAGAGAGAGAGAG	R: TCTGCAACGGCTGATAGATG
88	RM 451	F: GATCCCCTCCGTCAAACAC	R: CCCTTCTCCTTTCCTCAACC
89	RM 462	F: ACGGCCCATATAAAAGCCTC	R: AAGATGGCGGAGTAGCTCAG
90	RM 465	F: GTGCCTCCATCATCATCATC	R: TAGGACAAGCGAAGAAACCG
91	RM 482	F: TCTGAAAGCCTGACTCATCG	R: GTCAATTGCAGTGCCCTTTC
92	RM 495	F: AATCCAAGGTGCAGAGATGG	R: CAACGATGACGAACACAACC
93	RM 498	F: AATCTGGGCCTGCTCTTTTC	R: TCCTAGGGTGAAGAAAGGGG
94	RM 511	F: CTTCGATCCGGTGACGAC	R: AACGAAAGCGAAGCTGTCTC
95	RM 512	F: CTGCCTTTCTTACCCCCTTC	R: AACCCCTCGCTGGATTCTAG
96	RM 541	F: TATAACCGACCTCAGTGCCC	R: CCTTACTCCCATGCCATGAG

- **PCR amplification of DNA**

For the amplification of genomic DNA, a reaction mixture of 12.5 µl volume was prepared using 7.15 µl of sterilized distilled water, 1.0 µl template DNA (25 ng/µl), 0.5 µl of forward and 0.5 µl of reverse primer (5 µM), 1.0 µl $MgCl_2$ (25 mM), 1.25 µl 10X PCR buffer (10 mM Tris-HCl, 50 mM KCl, pH 8.3), 1.0 µl dNTP mix (0.2 mM each of dATP, dGTP, dCTP and dTTP) and 0.1 µl *Taq* polymerase (5 U/µl). The amplifications were carried out in S1000^{TM} Thermal Cycler (BIO-RAD) using protocol given in Table 3.9.

Table 3.9 PCR conditions used for amplification of *Oryza sativa* genomic DNA

Primer Type	Steps	Temperature and time	Cycles
SSR	Initial denaturation	94^0C for 5 minutes	1
	Denaturation	94^0C for 30 seconds	
	Annealing	57^0C for 30 seconds	35
	Extension	72^0C for 1 minute	
	Final extension	72^0C for 5 minutes	
	Storage	4^0C for ∞	1

The amplification products were electrophoresed in 3% agarose gel (HIMEDIA) and stained with ethidium bromide (0.5µg/ml). The gels were visualized and photographed using the Gel-Documentation Unit.

3.4.4 Analysis of SSR profiles

The amplified DNA of 43 red rice germplasm lines generated SSR marker profiles. The presence or absence of each SSR band of a particular molecular weight was scored manually. A binary data matrix with '1' and '0' indicating the presence and absence of particular molecular weight, respectively was generated separately for each primer. The binary data were used to generate a similarity matrix using Jaccard's coefficient, $J_{ij} = C_{ij}/(n_i + n_j - c_{ij})$, where '$C_{ij}$' is the number of positive matches between two genotypes, while n_i and n_j is the total number of band in genotype i and j, respectively in SIMQUAL programme of NTSYS-pc package (Rohlf 1993; 1998).

Genetic distances (GD) were calculated as $GD = 1–[C_{ij}/(n_i + n_j - C_{ij})]$. The data were subsequently used to construct a dendrogram using the unweighted

pair group method with arithmetrical averages (UPGMA) in SAHN program of NTSYS-pc package.

I. Polymorphic information content (PIC)

The PIC values provide an estimate of the discriminatory power of the locus or loci by taking into account not only the number of alleles that are expressed, but also the relative frequency of those alleles. It measures the informativeness of a given DNA marker and these were calculated according to Anderson et al. (1993).

$$\mathrm{PIC}i = 1 - \sum_{i=1}^{k} P_i^2$$

Where, k is the total number of alleles detected by a given marker locus and P_i is the frequency of the i^{th} allele in the set of genotypes investigated. PIC value ranges from 0 (monomorphic) to 1 (very highly discriminative, with many alleles each in equal and low frequency).

II. Parameters of genetic variation

Various parameters of genetic variation *viz.*, observed number of alleles, effective number of alleles, Nei's (1973) gene diversity, genetic diversity over all populations, genetic diversity within population, coefficient of gene differentiation (proportion of genetic diversity between populations), polymorphic loci and gene flow were estimated using POPGENE (Yeh and Boyle, 1997).

III. Clustering Analysis

Binary data was used to calculate a genetic dissimilarity matrix using the Jaccard dissimilarity index (d_{ij}) between pairs of accessions (units). Jaccard's similarity indexes (Jaccard, 1901) were calculated with data provided by the observation of the presence (1) or absence (0) of bands between pairs of accessions using the formula $D_{(ij)} = a/(a+b+c)$ where, a is the number of fragments shared by accessions.

A STRUCTURE version 2.3.4 (Pritchard et al. 2000) was used to infer the genetic structure so as to obtain an estimate of the likely number of population genetic clusters (K). The number of clusters of the population (K) were identified by performing 6 iterations and setting the value of k from 1-25 with a burn-in period of 100,000 and 100,000 number of Markov Chain Monte Carlo (MCMC) repeats after burn-in. Maximal value of LnP (D) and the posterior probability of data as per Evanno et al. (2005) was obtained using STRUCTURE HARVESTER (Earl and vonHoldt 2012).

4. RESULTS AND DISCUSSION

Rice is the principle cereal food crop grown most extensively in the tropical and sub-tropical regions of the world. In India, though cultivated on large area, the productivity of rice crop has been low and need the development of more high yielding varieties adapted to different seasons and agronomic conditions. Now most of the plant breeders recognize the importance of utilizing genetic diversity in breeding programmes to meet the continuously expanding needs of varietal improvement. The assemblage, evaluation and preservation of the entire germplasm is essential to reward breeding efforts.

The characterization of germplasm accessions established distinctiveness among rice genotypes, which is essential in the present era for protecting the unique rice cultivars. Since morphological traits are affected by environment, so there is a need to go for a highly reliable and precise method for assessment of genetic variability with no environmental effects. Assessment of genetic diversity with molecular markers overcomes this problem.

The present investigation, therefore, was taken up with 41 rice genotypes for their agronomical characterization, assessment of variability, genetic divergence and molecular characterization at CSKHPKV Rice and Wheat Research Centre, Malan during *Kharif*, 2021 and the experiment results on various aspects obtained from present investigation have been described in the following heads:

4.1 Analysis of variance for the experimental design

4.2 Mean performance for various characters in rice genotypes

4.3 Disease reaction

4.4 Parameters of variability

4.5 Correlation coefficient analysis

4.6 Path analysis

4.7 Genetic diversity studies through morphological markers

4.8 Principle component analysis (PCA)

4.9 Genetic diversity studies through molecular markers

4.1 Analysis of variance for the experimental design

The result of analysis of variance for all the characters studied has been presented in Table 4.1. Perusal of the result revealed the presence of significant difference among the genotypes at 5 per cent level of significance for days to 50 per cent flowering, days to 75 per cent maturity, plant height, flag leaf length, total tillers per plant, effective tillers per plant, spikelet per panicle, 1000 grain weight, grain length, grain breadth, L:B ratio, biological yield per plant, harvest index and grain yield per plant justifying the presence of sufficient genetic variability in the material under study. Differences were found non-significant for flag leaf width.

Similar results were also reported by Rachappanavar (2017) and Alagappan and Bhardwaj (2022). They revealed significant values for days to 50 per cent flowering, days to 75 per cent maturity, plant height, flag leaf length, total tillers per plant, effective tillers per plant, spikelet per panicle, 1000 grain weight, grain length, grain breadth, L:B ratio, biological yield per plant, harvest index and grain yield per plant.

Table 4.1 Analysis of variance of rice genotypes for yield and related traits

S.No.		Mean sum of square		
	Source of variation	**Replication**	**Treatment**	**Error**
	df	**2**	**42**	**84**
1	Days to 50% flowering	8.81	198.42*	2.98
2	Days to 75% maturity	5.61	53.14*	3.42
3	Plant height	81.13	431.87*	43.15
4	Flag leaf length	29.08	128.56*	13.47
5	Flag leaf width	0.17	0.21	0.06
6	Total tillers per plant	0.15	9.06*	0.20
7	Effective tillers per plant	0.05	6.83*	0.17
8	Spikelet per panicle	0.54	654.71*	1.94
9	1000 grain weight	0.57	44.05*	0.88
10	Grain length	0.03	2.02*	0.03
11	Grain breadth	0.004	0.44*	0.01
12	L:B ratio	0.01	1.13*	0.02
13	Biological yield per plant	44.60	171.42*	20.22
14	Harvest index	44.86	81.68*	15.32
15	Grain yield per plant	2.57	49.02*	2.95

*Significance at 5%

Plate 4.1 Field view of rice genotypes at early growth stage

4.2 Mean performance for various characters in rice genotypes

In the present study 39 red rice and 2 white rice genotypes along with two checks *viz.*, HPR-2720 and HPR-2795 were evaluated for yield and its contributing traits during *Kharif*, 2021. The mean performance of these genotypes is presented in Appendix I.

4.2.1 Quantitative traits

4.2.1.1 Days to 50 per cent flowering

The estimate of mean values indicated that the days to 50 per cent flowering ranged from **71 - 102 days** with a mean value of **88.60 days**. Deval was the earliest flowering genotype taking 71 days to mature while Karad 21-3 was the most late maturing genotype.

Out of 41 genotypes, 30 genotypes *viz.*, Deval, Nailina, HPR-2906, Dodadhan, Varundhan, Bathidhan, IC-12164, HPR-2908, New Chaina-21, Phulpatas-21, Gocha, Jattu, Bhrigudhan, Begmi, Matali, HPR-2902, Chohartu, Old Chaina-21, Hatiali, Roda dhan, HPR-2904, IC-12180, HPR-2913, Ramjuwain, Kaluna, HPR-2914, HPR-2905, HPR-2800, Karad 21-2 and Karad 21-4 were early flowering genotypes with respect to the best check HPR-2795. However, seven genotypes *viz.*, Kalijhini-2, Kalaina, Sukara, Kalijhini, Naggardhan, Sukara red and Karad 21-1 were observed to be statistically at par with the best check.

Plate 4.2 Field view of experiment at flowering stage

4.2.1.2 Days to 75 per cent maturity

Days to 75 per cent maturity ranged from **108 to 123 days** with an average duration of **115 days.** The genotype Bhrigudhan was the earliest maturing genotype among all.

Out of the 41 genotypes, 28 genotypes *viz.*, Bhrigudhan, Begmi, HPR-2906, Deval, Jattu, Dodadhan, Nailina, Varundhan, IC-12164, Bathidhan, Matali, Kalijhini, HPR-2902, Phulpatas-21, Chohartu, HPR-2908, Gocha, IC-12180, Old Chaina-21, New Chaina-21, Ramjuwain, Naggardhan, Hatiali, HPR-2800, Roda Dhan, Kaluna,

HPR-2913 and HPR-2904 were found significantly early maturing in comparison to the best check HPR-2795. However, 12 genotypes *viz.*, HPR-2914, Sukara, HPR-2905, Karad 21-3, Karad 21-1, Kalaina, Karad 21-2, Karad 21-4, Kalijhini-2, Acchoo, Sukara red and Desidhan were statistically at par with the HPR-2795 and one genotype Karad was significantly late than the best check for days to 75 per cent maturity.

Plate 4.3 Field view of the experiment at maturity stage

4.2.1.3 Plant Height (cm)

Plant height among the genotypes was ranged from **101.37cm to 150.67cm** with mean height of **129.22cm**. Among all the genotypes IC-12164 was found to be the shortest among all genotypes. However, genotypes HPR-2902 was observed with highest plant height.

Out of 41 genotypes, nine genotypes *viz.*, IC-12164, Nailina, HPR-2908, New Chaina-21, Sukara, Karad 21-1, Karad, Jattu and HPR-2800 were significantly dwarf in plant height in comparison to best check HPR-2720, while 28 genotypes *viz.*, Matali, Karad 21-2, Old Chaina-21, Begmi, Ramjuwain, Phulpatas-21, Kalijhini, Gocha, Hatiali, Karad 21-3, Bathidhan, Deval, Naggardhan, Kalaina, Roda dhan,

HPR-2914, Karad 21-4, Bhrigudhan, Sukara red, Dodadhan, HPR-2905, Kalijhini-2, Varundhan, Desidhan, Chohartu, Kaluna, HPR-2904 and IC-12180 were statistically at par with HPR-2720.

4.2.1.4 Flag leaf length (cm)

The mean values for flag leaf length ranged from **24.03 cm to 62.53 cm** with general mean **36.13 cm.** Genotype Desidhan had highest flag leaf length, while, HPR-2908 was recorded with lowest flag leaf length.

Among 41 genotypes, two genotypes *viz.*, Desidhan and HPR-2913 were significantly superior to the best check HPR-2795 whereas 22 genotypes *viz.*, Hatiali, Karad 21-2, HPR-2800, Sukara, Phulpatas-21, Acchoo, Kalaina, Naggardhan, Kalijhini, HPR-2906, Matali, Varundhan, New Chaina-21, HPR-2914, Sukara red, Kalijhini-2, HPR-2904, Karad 21-4, HPR-2902, HPR-2905, Jattu and Chohartu were found statistically at par with the best check HPR-2795.

4.2.1.5 Flag leaf width (cm)

The mean values for flag leaf width ranged from **0.95 cm to 2.07 cm** with general mean **1.54 cm.** The highest flag leaf length was observed in Acchoo and lowest in genotype Ramjuwain. None of the genotypes were found significantly superior for flag leaf width to the best check HPR-2795.

4.2.1.6 Total tillers per plant

Total tillers per plant among the genotypes ranged between **5 to 12** with an average value of **8.28**. Among all the genotypes, Desidhan exhibited highest total tillers per plant and Acchoo exhibited lowest total tillers per plant.

Out of 41 genotypes, 14 genotypes *viz.*, Hatiali, Karad 21-2, Old Chaina-21, Naggardhan, Varundhan, Karad, IC-12180, New Chaina-21, Bathidhan, Phulpatas-21, Sukara, Sukara red, HPR-2913 and Desidhan were found to be significantly superior to the best check HPR-2795, while 17 genotypes *viz.*, HPR-2904, Karad 21-3, Kalaina, HPR-2908, HPR-2902, Karad 21-4, IC-12164, Begmi, Kalijhini-2, Nailina,

Ramjuwain, Dodadhan, HPR-2906, HPR-2914, Gocha, Matali and Jattu were statistically at par with best check HPR-2795.

4.2.1.7 Effective tillers per plant

Effective tillers per plant among the genotypes studied were observed in the range of **4 to 11** with mean value of **7.51**. The highest number of effective tillers per plant were reported in genotype Desidhan while lowest number of effective tillers per plant were recorded in the genotype Acchoo.

Among 41 genotypes, eight genotypes *viz.*, Old Chaina-21, Bathidhan, Sukara, Phulpatas-21, New Chaina-21, Sukara red, HPR-2913 and Desidhan were significantly superior as compared to the best check HPR-2795. Nineteen genotypes *viz.*, Karad 21-4, Nailina, HPR-2902, Matali, Jattu, Dodadhan, IC-12164, Gocha, Begmi, HPR-2906, Karad, IC-12180, Karad 21-2, Ramjuwain, Kalijhini-2, Hatiali, HPR-2914, Naggardhan and Varundhan were statistically at par with the best check HPR-2795.

4.2.1.8 Spikelets per panicle

The mean value of spikelets per panicle ranged from **35 to 90** with a general mean of **60.12**. The highest number of spikelets per panicle were observed in HPR-2913 and the lowest number of spikelets per panicle were observed in Karad.

Among 41 genotypes, none of the genotype was significantly superior or statistically at par as compared with the best check HPR-2795.

4.2.1.9 1000 grain weight (g)

The mean value for this trait was 23.50 g, with a range of **16.83 g to 35.15 g.** Among all the genotypes studied, Karad 21-4 exhibited highest 1000 grain weight. However, lowest 1000 grain weight was reported in IC-12180.

Among 41 genotypes, none of the genotype was significantly superior or statistically at par with the best check HPR-2795.

4.2.1.10 Biological yield per plant (g)

Biological yield per plant varied from **31.15 g to 62.25 g** with an average value of **39.22 g**. Among all the genotypes studied, Desidhan had highest biological yield per plant. However, lowest biological yield per plant was reported in the genotype Nailina.

Among 41 genotypes, none of the genotype was significantly superior to the best check HPR-2795 while, four genotypes *viz.*, Desidhan, HPR-2913, Sukara and Sukara red were statistically at par with HPR-2795.

4.2.1.11 Harvest index (%)

Harvest index varied from **22.64% to 46.39%** with the mean value of **31.45%**. Highest harvest index was reported in the genotype HPR-2913 whereas, lowest harvest index (%) was reported in the genotype Deval.

Among 41 genotypes, none of the genotype was significantly superior to the best check HPR-2720. However, seven genotypes *viz.,* HPR-2913, Sukara, HPR-2914, New Chaina-21, Varundhan, Desidhan and Phulpatas-21 were statistically at par with the best check HPR-2720.

4.2.1.12 Grain yield per plant (g)

Grain yield per plant ranged from **7.17 g to 24.51 g** with mean value of **12.45 g**. Among all the genotypes taken in the present study, genotype Desidhan exhibited highest grain yield per plant. On the other hand, genotype Deval exhibited the lowest grain yield per plant.

Among all the 41 genotypes only one genotype Desidhan was significantly superior as compared to the best check HPR-2795. Two genotypes Sukara red and HPR-2913 were statistically at par with the second check HPR-2795 for this trait.

4.2.2 Quality traits

4.2.2.1 Grain length (mm)

Grain length ranged from **3.17 mm to 6.46 mm** with the mean value of **4.27 mm**. Among all the genotypes, Kalijhini-2 had the longest grain and Deval had shortest grain.

Out of 41 genotypes taken in present study, none of the genotypes were significantly superior as compared to the best check HPR-2795 while, three genotypes *viz.*, Phulpatas-21, HPR-2913 and Kalijhini-2 were statistically at par with HPR-2795 for grain length.

4.2.2.2 Grain Breadth (mm)

Grain breadth ranged between **1.35 mm to 3.13 mm** with the mean value of **2.11 mm**. Genotype Dodadhan had highest grain breadth among all the genotype. While, genotype Begmi exhibited lowest grain breadth.

Two genotypes Begmi and Acchoo were significantly superior as compared with the best check HPR-2720. However, seven genotypes *viz.*, HPR-2800, HPR-2908, Kalijhini-2, HPR-2905, HPR-2906, Kalaina and Phulpatas-21 were statistically at par with the best check HPR-2720 for this particular trait.

4.2.2.3 L:B ratio

L:B ratio ranged between **1.16 to 4.10** with the mean value of **2.10**. Among the studied genotypes, highest grain L:B ratio was recorded in Kalijhini-2 and lowest L:B ratio was reported in Roda dhan.

Two genotypes *viz.*, Kalijhini-2 and Phulpatas-21 were significantly superior as compared with the best check HPR-2795.

4.2.2.4 Lodging susceptibility (%)

Effect of lodging in traditional landraces is always a major drawback but in present study 24 traditional red rice genotypes were recorded to be lodging resistant.

However, 11 genotypes exhibited only 25% lodging and eight genotypes exhibited 50% lodging (Table 4.2).

Table 4.2 Classification of rice genotypes on the basis of lodging susceptibility

S.no	Lodging effect	No. of genotype	Genotypes
1	50%	8	Kaluna, Ramjuwain, Chohartu, Sukara, Karad, Kalaina, IC-12164, Acchoo
2	25%	11	Bathidhan, HPR-2906, Varundhan, Naggardhan, Roda Dhan, Nailina, Deval, Bhrigudhan, Kalijhini, Matali, IC-12180
3	No lodging	24	Begmi, Desidhan, Kalijhini-2, Sukara red, Hatiali, HPR-2800, Dodadhan, HPR-2902, HPR-2904, HPR-2905, HPR-2908, HPR-2913, HPR-2914, GOCHA, New Chaina-21, Old Chaina-21, Phulpatas-21, Karad 21-1, Karad 21-2, Karad 21-3, Karad 21-4, Jattu, HPR-2720 (C), HPR-2795 (C)

4.2.2.5 Awns

Characteristics of presence of awns in red rice genotypes were reported in 21 genotypes while 22 genotypes were found to be awnless (Table 4.3).

Table 4.3 Classification of rice genotypes on the basis of awn characteristic

S.no	Awns	No. of genotype	Genotypes
1	Awned	21	IC-12180, Kaluna, Ramjuwain, Sukara, Karad, Kalaina, IC-12164, Bathidhan, HPR-2906, Desidhan, Sukara red, Hatiali, Matali, HPR-2913, HPR-2914, Gocha, Karad 21-2, Karad 21-3, Jattu, HPR-2795 (C), Chohartu
2	Awn-less	22	Acchoo, Begmi, Kalijhini-2, Varundhan, HPR-2800, Naggardhan, Roda Dhan, Nailina, Deval, Bhrigudhan, Kalijhini, Dodadhan, HPR-2902, HPR-2904, HPR-2905, HPR-2908, New Chaina-21, Old Chaina-21, Phulpatas-21, Karad 21-1, Karad 21-4, HPR-2720 (C)

4.2.2.6 Seed coat colour

Colour of seed coat in present study was found to be dark red coloured in 36 genotypes. However, five genotypes were reported to be light red coloured (Table 4.4).

Table 4.4 Classification of rice genotypes on the basis of seed coat colour

S.no	Seed coat colour	No. of genotypes	Genotypes
1	Dark red	36	Acchoo, Begmi, Kalijhini-2, Varundhan, HPR-2800, Naggardhan, Roda Dhan, Deval, Bhrigudhan, Dodadhan, HPR-2902, HPR-2904, HPR-2905, HPR-2908, New Chaina-21, Old Chaina-21, Karad 21-1, Karad 21-4, IC-12180, Kaluna, , Sukara, Kalaina, IC-12164, Bathidhan, HPR-2906, Desidhan, Sukara red, Hatiali, Matali, HPR-2913, Gocha, Karad 21-2, Karad 21-3, Chohartu, HPR-2795 (C), HPR-2720 (C)
2	Light red	5	Karad, Nailina, Kalijhini, HPR-2914, Jattu
3	White	2	Ramjuwain, Phulpatas-21

Mean performance of the genotypes revealed that Desidhan was superior to the best check HPR-2795 for grain yield per plant. However, two genotypes *viz.*, Desidhan and HPR-2913 were found superior to the second check HPR-2720 for this trait. Overall, Desidhan, HPR-2913 and Sukara red were found to be superior for most of the traits *viz.*, days to 75 per cent maturity, flag leaf length, total tillers per plant and effective tillers per plant along with the grain yield and hence can be used in hybridization programme. In addition, Phulpatas-21, Old Chaina-21, Bathidhan, Sukara, New Chaina-21, Hatiali, Bathidhan, Varundhan and IC-12180 were also found promising genotypes on the bases of their mean performance for most of the traits (Table 4.5)

Table 4.5 Promising genotypes on the basis of mean performance for seed yield and other related traits

S. No.	Traits	Best check	No. of promising genotypes	Details of genotypes
1.	Days to 50% flowering	HPR-2795	30	Deval, Nailina, HPR-2906, Dodadhan, Varundhan, Bathidhan, IC-12164, HPR-2908, New Chaina-21, Phulpatas-21, Gocha, Jattu, Bhrigudhan, Begmi, Matali, HPR-2902, Chohartu, Old Chaina-21, Hatiali, Roda dhan, HPR-2904, IC-12180, HPR-2913, Ramjuwain, Kaluna, HPR-2914, HPR-2905, HPR-2800, Karad 21-2, Karad 21-4
2.	Days to 75% maturity	HPR-2795	29	Bhrigudhan, Begmi, HPR-2906, Deval, Jattu, Dodadhan, Nailina, Varundhan, IC-12164, Bathidhan, Matali, Kalijhini, HPR-2902, Phulpatas-21, Chohartu, HPR-2908, Gocha, IC-12180, Old Chaina-21, New

				Chaina-21, Ramjuwain, Naggardhan, Hatiali, HPR-2800, Roda dhan, Kaluna, HPR-2913, HPR-2904, HPR-2914
3.	Plant height (cm)	HPR-2720	9	IC-12164, Nailina, HPR-2908, New Chaina-21, Sukara, Karad 21-1, Karad, Jattu, HPR-2800
4.	Flag leaf length (cm)	HPR-2795	2	HPR-2913, Desidhan
5.	Total tillers per plant	HPR-2795	14	Hatiali, Karad 21-2, Old Chaina-21, Naggardhan, Varundhan, Karad, IC-12180, New Chaina-21, Bathidhan, Phulpatas-21, Sukara, Sukara red, HPR-2913, Desidhan
6.	Effective tillers per plant	HPR-2795	8	Old Chaina-21, Bathidhan, Sukara, Phulpatas-21, New Chaina-21, Sukara red, HPR-2913, Desidhan
7.	Spikelets per panicle	HPR-2795	-	-
8.	1000-grain weight (g)	HPR-2795	-	-
9.	Grain length [L] (mm)	HPR-2795	-	-
10.	Grain breadth [L] (mm)	HPR-2720	2	Begmi, Acchoo
11.	L:B ratio	HPR-2795	2	Phulpatas-21, Kalijhini-2
12.	Biological yield per plant (g)	HPR-2795	-	-
13.	Harvest index (%)	HPR-2720	-	-
14.	Grain yield per plant (g)	HPR-2795	1	Desidhan

4.3 Disease reaction

4.3 Disease reaction

4.3.1 Leaf blast

Response of genotypes to leaf blast (*Pyricularia oryzae*) was observed under open field condition at RWRC Malan and the data are presented in Table 4.6. It was observed that 37.20 per cent of the genotypes were susceptible to the disease while, 41.86 per cent had moderate susceptibility, 11.62 per cent moderately resistant while 9.30 per cent genotypes were found resistant to the disease.

Results revealed that four genotypes *viz.*, IC-12180, HPR-2800, HPR-2795 and HPR-2913 showed resistant reaction to the disease under open field conditions, whereas five genotypes *viz.*, HPR-2914, HPR-2720, Desidhan, Sukaraand Sukara Red exhibited moderately resistant reaction. Eighteen genotypes were found to be moderately susceptible and 16 genotypes were found susceptible to the disease. The Resistant genotypes can be used as donor parents for future breeding programme of rice.

Table 4.6 Evaluation of rice genotypes to leaf blast resistance

Score	Disease reaction	No. of genotypes	Genotypes
0	Immune	-	-
1	Resistant	4	IC-12180, HPR-2800, HPR-2795, HPR-2913
3	Moderately Resistant	5	HPR-2914, HPR-2720, Desidhan, Sukara, Sukara Red
5	Moderately Susceptible	18	IC-12164, Kalaina, Bathidhan, DodaDhan, Ramjuwain, Karad, Varun Dhan, Chohartu, Varundhan, Kalijhini, Begmi, HPR-2902, HPR-2908, HPR-2904, HPR-2905, New Chaina-21, Old Chaina-21, Phulpatas-21
7	Susceptible	16	Kaluna, Acchoo, Gocha, Kalijhini-2, NaggarDhan, Nailina, Hatiali, Matali, HPR-2906, RodaDhan, Deval, BhriguDhan, Karad 21-1, Karad 21-2, Karad 21-3, Karad 21-4
9	Highly susceptible	-	-

4.3.2 Neck blast

Response of genotypes to neck blast (*Pyricularia oryzae*) was also observed under open field conditions. Among all the genotypes, it was observed that 34.88 per cent of the genotypes were susceptible to the disease while, 48.84 per cent had moderate susceptibility 6.98 per cent moderately resistance while 9.30per cent genotypes were found resistant to the disease.

Results revealed that four genotypes *viz.*, IC-12180, HPR-2800, HPR-2795 and HPR-2914 showed resistant reaction to neck blast whereas three genotypes *viz.*, HPR-2720, HPR-2913and Desidhan showed moderately resistant reaction. Twenty

two genotypes were found to be moderately susceptible while 15 were found susceptible to the disease (Table 4.7).

Table 4.7 Evaluation of rice genotypes to neck blast resistance

Score	Disease reaction	No. of genotypes	Genotypes
0	Immune	-	-
1	Resistant	4	IC-12180, HPR-2800, HPR-2795, HPR-2914
3	Moderately Resistant	3	HPR-2720, HPR-2913, Desidhan
5	Moderately Susceptible	21	IC-12164, BathiDhan, DodaDhan, Begmi, Ramjuwain, Kalaina, BhriguDhan, Deval, Chohartu, Sukara Red, Kalijhini, HPR-2902, HPR-2908, HPR-2904, HPR-2905, Sukara, Varun Dhan, New Chaina-21, Old Chaina-21, Phulpatas-21, Jattu
7	Susceptible	15	Kaluna, RodaDhan, Acchoo, Karad, Gocha, Kalijhini-2, Hatiali, NaggarDhan, Nailina, Matali, HPR-2906, Karad 21-1, Karad 21-2, Karad 21-3, Karad 21-4
9	Highly susceptible	-	-

Overall, IC-12180, HPR-2800, HPR-2795, HPR-2913, Desidhan and HPR-2720 exhibited disease reaction varying between resistant and moderately resistant both to leaf and neck blast. Hence, these can be includedasdonors in breeding for blast resistance.

The results are in agreement with the findings of several workers. Puren (2017) found similar results and reported HPR-2800 as a promising genotype. Alagappanand Bhardwaj (2022) also reported similar findings in red rice germplasm.

4.4 Parameters of variability

Effectiveness of any selection programme depends upon the existence of genetic variability within the population. The variability at phenotypic level arises due to genotypic as well as through environmental influences thus contributing towards the development of phenotypes. The estimated values of parameters of variability *viz.*, phenotypic coefficient of variation (PCV) and genotypic coefficient of variation (GCV) along with the broad sense heritability (h^2bs) and expected genetic advance as

percent of mean for different traits are presented in the Table 4.8. A wide range of genetic variability was observed for all the characters studied.

4.4.1 Coefficients of variation

The degree of variability in a crop species is of vital importance because it provides the basis for determining the total variation present in a population that result from genotypic and environmental effects. The knowledge of phenotypic coefficient of variation (PCV) and genotypic coefficient of variation (GCV) is helpful in predicting the amount of variation present in the given germplasm which helps in formulating an efficient breeding programme. An assessment of variability parameters revealed presence of some variation among the genotypes under study.

The estimates of phenotypic coefficient of variation (PCV) and genotypic coefficient of variation (GCV) were classified as low (<10%), Moderate (10-20%) and high (>20%). Based on the classification, high PCV estimates were observed for the traits flag leaf width, total tillers per plant, effective tillers per plant, spikelets per panicle, L:B ratio, biological yield per plant and grain yield per plant. On the other hand, moderate PCV was reported in plant height, flag leaf length, 1000 grain weight, grain length, grain breadth and harvest index while, lower PCV was reported in days to 50 per cent flowering and days to 75 per cent maturity.

The estimates of genotypic coefficient of variation (GCV) reflects the total amount of genotypic variability that was transmitted from parents to the progeny. GCV estimates were observed high for the traits total tillers per plant, L:B ratio and grain yield per plant. Moderate GCV was reported for flag leaf length, flag leaf width, effective tillers per plant, 1000 grain weight, grain length, grain breadth, biological yield per plant and harvest index while, lower PCV was reported in days to 50 per cent flowering, days to 75 per cent maturity and plant height.

In the present study, phenotypic coefficient of variation (PCV) were found to be slightly higher than their respective genotypic coefficient of variation (GCV) for all the traits studied (Table 4.7), which indicated that the apparent variation is not only due to genotypes but also due to considerable influence of environment on the performance of genotypes.

Table 4.8 Estimates of parameter of variability for morphological, yield and yield contributing traits in rice genotypes

S.No.	Traits	Grand Mean ± SE (m)	Range	PCV %	GCV %	h^2bs (%)	GA as % mean
1	Days to 50% flowering	88.56 ± 1.00	71.00 - 102.33	9.32	9.11	95.63	18.36
2	Days to 75% maturity	115.16 ± 1.07	108.33 - 123.33	3.88	3.54	82.89	6.63
3	Plant height	129.22 ± 3.79	101.37 - 153.33	10.17	8.81	75.02	15.72
4	Flag leaf length	36.13 ± 2.12	24.03 - 62.53	19.93	17.14	74.01	30.38
5	Flag leaf width	1.54 ± 0.14	0.95 - 2.07	21.21	14.83	48.90	21.36
6	Total tillers per plant	8.25 ± 0.26	4.63 - 11.93	21.63	20.95	93.82	41.80
7	Effective tillers per plant	7.57 ± 0.24	4.23 - 10.87	21.82	18.21	69.65	31.23
8	Spikelet per panicle	60.09 ± 0.80	34.67 - 86.70	26.78	22.52	70.71	38.95
9	1000 grain weight	23.50 ± 0.54	16.83 - 35.15	16.63	16.14	94.23	32.28
10	Grain length	4.27 ± 0.09	3.17 - 6.42	19.46	19.09	96.25	38.58
11	Grain breadth	2.11 ± 0.06	1.35 - 3.13	18.68	17.93	92.15	35.46
12	L:B ratio	2.10 ± 0.09	1.16 - 4.10	29.78	28.87	93.95	57.64
13	Biological yield per plant	39.22 ± 2.60	31.15 - 62.25	21.43	18.10	71.37	31.50
14	Harvest index	31.45 ± 2.26	22.64 - 46.40	19.46	14.96	59.09	23.68
15	Grain yield per plant	12.45 ± 0.99	7.17 - 24.51	34.36	31.47	83.87	59.37

SE: Standard error
PCV: Phenotypic coefficient of variation
GCV: Genotypic coefficient of variation
ECV: Environmental coefficient of variation
h^2bs: Heritability in broad sense
GA: Genetic advance (%) of mean

Similar results of higher phenotypic coefficient of variation (PCV) in comparison to genotypic coefficient of variation (GCV) in red rice were also reported by Alagappan and Bhardwaj (2022), Puren (2017) and Rachappanavar (2017). For the quantitative characters, Singh et al. (2020) and Alagappan and Bhardwaj (2022) reported similar finding for days to 50 per cent flowering and days to 75 per cent maturity. While, result on flag leaf length, flag leaf width and harvest index were found to be in accordance with Devi et al. (2016). Kujur et al. (2019) supported the report on effective tillers per plant, spikelets per panicle, biological yield per plant and grain yield per plant. Bagudam et al. (2018) supported the results for harvest index and total tillers per plant.

4.4.2 Heritability in broad sense (h^2bs)

When a major portion of variability is due to heritable variation it could be measured in terms of degrees in which it is transmitted to the progeny, the term referred to as heritability. The ratio of genetic variance to the total variance is heritability in broad sense. Burton and DeVane (1958) has suggested that a genetic coefficient of variability together with the heritability estimates would give more reliable indication of the expected genetic gain by selection. Heritability in broad sense is of tremendous significance to the breeders as its magnitude indicates reliability with which a genotype can be recognized by its phenotypic expression (Lush 1949). The information on heritability estimates is useful in studying the inheritance of quantitative traits as well as for planning breeding programmes with desired degree of expected genetic progress.

The estimates of heritability in broad sense were classified as low (<50 per cent), Moderate (50-80%) and high (>80%). The present study revealed that heritability in broad sense was high for days to 50 per cent flowering, days to 75 per cent maturity, L:B ratio and grain yield per plant. Moderate heritability was observed for plant height, flag leaf length, effective tillers per plant, spikelets per panicle, biological yield per plant and harvest index. Low heritability was observed for Flag leaf width (Table 4.8).

The results are supported by the findings of Thongbam et al. (2012). Thongbam et al. (2010) observed high heritability for grain length, grain breadth and L:B ratio. Sohrabi et al. (2012) supported the findings for 1000 grain weight, days to 50 per cent flowering and days to 75 per cent maturity, while the result of grain breadth and grain yield per plant were in accordance with the findings reported by Tuhina-Khatun et al. (2015).

4.4.3 Genetic advance (GA)

The estimates of heritability on any traits alone fail to indicate the response to selection. Thus, estimates appear to be more useful when accompanied by estimates of genetic advance (Johnson et al. 1955). Thus, genetic advance has an added edge over heritability as a guiding factor for breeders in various selection programmes.

Expected genetic advance as per cent of mean indicates the mode of gene action in the expression of a trait, which helps in choosing an appropriate breeding method. For predicting reliable estimates of additive and non-additive effects, heritability should be considered in conjugation with genetic advance (Burton and DeVane 1958; Johnson et al. 1955). The results of genetic advance for various traits are given in Table 4.8.

Genetic advance expressed as percentage of mean were classified as low (<10%), Moderate (10-20%) and high (>20%). Genetic advance expressed as percentage of mean was high for most of the traits *viz.,* flag leaf length, flag leaf width, total tillers per plant, effective tillers per plant, spikelets per panicle, 1000 grain weight, grain length, grain breadth, L:B ratio, biological yield per plant, harvest index and grain yield per plant indicating that these traits were under the control of additive gene action. Moderate genetic advance as percentage of mean was obtained for days to 50 per cent flowering and plant height. It was low for days to 75 per cent maturity indicating that these traits were under the control of non-additive gene action. The results were supported by Thongbam et al. (2012), Ahmad et al. (2015), Akhtar et al. (2022) and Singh et al. (2020).

Based on the overall results for heritability and genetic advance, high heritability (>80%) coupled with high genetic advance as percent of mean (>20%) was observed for total tillers per plant, 1000 grain weight, grain length, grain breadth, L:B ratio and grain yield per plant indicated the presence of high additive gene action thus, selection for such traits may be beneficial in future breeding programme.

High heritability (>80%) along with moderate genetic advance (10-20%) was observed for days to 50 per cent flowering, which suggested that, combination of careful and restricted selection might be effective for the improvement of these traits in breeding programme.

High heritability (>80%) coupled with low genetic advance (<10%) was observed for days to 75 per cent maturity indicated the role of non-additive gene action in the inheritance of these traits, which revealed the importance of dominance

and epistatic effects in the inheritance of these traits and selection would be less effective.

Dutta et al. (2013) recorded high heritability coupled with high genetic advance for total tillers per plant and grain yield per plant. Results of Sanghara et al. (2013) were found to be in accordance with the present study for grain length, grain breadth and L:B ratio. However, results on days to 75 per cent maturity were in accordance with Singh et al. (2020).

4.5 Correlation coefficient analysis

Study on association of various yield attributing traits is essential for accumulating the optimum contribution of such traits to yield. Seed yield is within great influence of environmental conditions, has complex mode of inheritance and low heritability (Bocanski et al. 2010). On the contrary most of the yield components are less complex and because of this by using some other trait which is highly correlated with seed yield and has higher heritability, should make the selection of the best progenies more reliable (Vasic et al. 2001; Bekavac et al. 2007; 2008). Study of yield components and their inter-relationship along with yield and their direct and indirect contributions towards yield is of immense importance. The phenotype of a plant is the result of interaction of a large number of factors. Hence, the final yield is the sum total of effects of several component factors. Therefore, it is important to know the extent and nature of inter-relationship between seed yield and its contributing traits and also among themselves, to identify traits for increasing the efficiency of both direct and indirect selection for yield improvement.

In the present study (Table 4.9 and Table 4.10), the estimates of phenotypic correlation coefficient as well as genotypic correlation coefficient were computed for different yield and its component traits to determine the traits on which selection can be emphasized. The results obtained are discussed as under:

The magnitude of genotypic correlation was more than the phenotypic ones depicting strong inherent relationship between the traits. The grain yield per plant positively and significantly correlated with days to 50 per cent flowering, days to 75 per cent maturity, plant height, flag leaf length, total tillers per plant, effective tillers

per plant, spikelet per panicle, 1000 grain weight, grain length, L:B ratio, biological yield per plant and harvest index at phenotypic level as well as at genotypic level indicating that selection through these traits would be effective.

Days to 50 per cent flowering showed significant and positive correlation with plant height at genotypic level only while it was significant and positively correlated with days to 75 per cent maturity, flag leaf length, L:B ratio and biological yield per plant at both phenotypic as well as genotypic levels. On the other hand, it was recorded to be significantly and negatively correlated with grain breadth at both phenotypic as well as genotypic levels. Days to 75 per cent maturity exhibited significant and positive correlation with flag leaf length, grain length, L:B ratio and biological yield per plant at both phenotypic as well as genotypic levels. It was significantly and positively correlated with plant height and total tillers per plant while, significantly but negatively correlated with grain breadth at genotypic level only.

Significant positive association was observed for plant height with harvest index at genotypic level only, while it was significant and positively associated with flag leaf length, flag leaf width, grain length, L:B ratio, biological yield per plant at both phenotypic as well as genotypic levels. Flag leaf length showed significant and positive correlation with 1000 grain weight and L:B ratio at genotypic level only, while it showed significant and positive correlation with total tillers per plant, effective tillers per plant, grain length, biological yield per plant and harvest index at both phenotypic and genotypic levels. Flag leaf width exhibited significant and negative correlation with biological yield per plant at genotypic level only, while significant and negative correlation with total tillers per plant, effective tillers per plant and grain breadth at both phenotypic and genotypic levels.

Total tillers per plant was significant and positively correlated with effective tillers per plant, grain length, L:B ratio, biological yield per plant and harvest index at both phenotypic as well as genotypic levels. Similarly, effective tillers per plant showed significant and positive correlation with spikelets per panicle, grain length,

TABLE 4.9 Estimates of phenotypic correlation coefficients among various yield, yield components and grain quality traits in rice genotypes

	Days to 75% maturity	Plant height	Flag leaf length	Flag leaf width	Total tillers per plant	Effective tillers per plant	Spikelet per panicle	1000 grain weight	Grain length	Grain breadth	L:B ratio	Biological yield per plant	Harvest index	Grain yield per plant
Days to 50% flowering	0.799*	0.158	0.292*	-0.049	0.035	0.017	-0.011	-0.078	0.086	-0.268 *	0.203*	0.357 *	0.030	0.276 *
Days to 75% maturity		0.116	0.300*	-0.008	0.159	0.149	0.109	0.019	0.186*	-0.164	0.212*	0.415 *	0.136	0.370 *
Plant height			0.445 *	0.224*	-0.100	-0.063	-0.030	0.124	0.220*	-0.086	0.210*	0.192*	0.125	0.224*
Flag leaf length				0.087	0.204*	0.227 *	0.060	0.131	0.216*	-0.068	0.153	0.340 *	0.348 *	0.470 *
Flag leaf width					-0.293 *	-0.295 *	0.100	-0.072	-0.077	-0.174*	0.066	-0.145	-0.043	-0.114
Total tillers per plant						0.942 *	0.144	0.040	0.566 *	0.062	0.269 *	0.585 *	0.494 *	0.672 *
Effective tillers per plant							0.194*	0.096	0.585 *	0.050	0.291 *	0.563 *	0.506 *	0.664 *
Spikelet per panicle								0.266 *	0.270 *	0.023	0.159	-0.044	0.308 *	0.176*
1000 grain weight									0.253 *	0.124	0.089	0.077	0.284 *	0.236 *
Grain length										-0.130	0.748 *	0.536 *	0.554 *	0.676 *
Grain breadth											-0.722 *	-0.154	-0.002	-0.100
L:B ratio												0.410 *	0.324 *	0.456 *
Biological yield per plant													0.247 *	0.814 *
Harvest index														0.755 *

***Significance at P≤0.05**

TABLE 4.10 Estimates of genotypic correlation coefficients among various yield, yield components and grain quality traits in rice genotypes

	Days to 75% maturity	Plant height	Flag leaf length	Flag leaf width	Total tillers per plant	Effective tillers per plant	Spikelet per panicle	1000 grain weight	Grain length	Grain breadth	L:B ratio	Biological yield per plant	Harvest index	Grain yield per plant
Days to 50% flowering	0.859*	0.204*	0.345*	-0.084	0.042	0.016	-0.012	-0.084	0.084	-0.285*	0.212*	0.438*	0.030	0.308*
Days to 75% maturity		0.215*	0.363*	-0.016	0.180*	0.170	0.114	0.009	0.210*	-0.186*	0.241*	0.577*	0.153	0.447*
Plant height			0.528*	0.388*	-0.104	-0.080	-0.039	0.148	0.249*	-0.100	0.243*	0.223*	0.264*	0.298*
Flag leaf length				0.035	0.238*	0.269*	0.070	0.174*	0.253*	-0.079	0.182*	0.450*	0.487*	0.563*
Flag leaf width					-0.423*	-0.426*	0.148	-0.108	-0.112	-0.188*	0.048	-0.207*	0.025	-0.112
Total tillers per plant						0.984*	0.152	0.043	0.583*	0.079	0.270*	0.697*	0.599*	0.713*
Effective tillers per plant							0.202*	0.108	0.615*	0.052	0.312*	0.703*	0.655*	0.747*
Spikelet per panicle								0.275*	0.277*	0.025	0.164	-0.039	0.394*	0.197*
1000 grain weight									0.269*	0.120	0.108	0.111	0.373*	0.269*
Grain length										-0.129	0.755*	0.626*	0.658*	0.705*
Grain breadth											-0.714*	-0.176*	-0.008	-0.106
L:B ratio												0.477*	0.390*	0.478*
Biological yield per plant													0.568*	0.906*
Harvest index														0.857*

***Significance at P≤0.05**

L:B ratio, biological yield per plant and harvest index at both phenotypic as well as genotypic levels. Spikelets per panicle was significant and positively correlated with 1000 grain weight, grain length and harvest index at both phenotypic as well as genotypic levels. Thousand grain weight exhibited significant and positive correlation with grain length and harvest index at both phenotypic as well as genotypic levels. Significant positive association was observed for grain length with L:B ratio, biological yield per plant and harvest index at both phenotypic as well as genotypic levels. Significant negative association was observed for grain breadth with L:B ratio at phenotypic level and biological yield per plant at genotypic level. L:B ratio showed significant positive association with harvest index and biological yield per plant. Biological yield per plant in turn showed significant positive association with harvest index.

The results are supported by the findings of Aditya and Bhartiya (2013), Jambhulkar and Bose (2014) and Kumar (2015). Kishore et al. (2018) also observed positive correlation of grain yield with harvest index, biological yield per plant and plant height, while positive association between grain yield per plant with plant height and grain length was also reported by Archana et al. (2018). Thongbam et al. (2010) and Sinha et al. (2015) reported that grain length was significant and positively correlated with L:B ratio while L:B ratio was negatively correlated with grain breadth. Sinha and Mishra (2013) reported high correlation between days to 50 per cent flowering and days to 75 per cent maturity. Sanghera et al. (2013) reported significantly positive correlation between the number of tillers per plant and grain yield. Ahmad et al. (2015) reported negative association between plant height and the number of tillers per plant. Bagudam et al. (2018) showed positive correlation between days to 50 per cent flowering and grain yield per plant as reported in present study. Positive correlation between spikelets per panicle and grain yield was reported by Puren (2017).

4.6 Path Analysis

The correlation coefficients are quite helpful in determining the components of a complex trait like seed yield but an exact picture of the relative importance of direct

and indirect influence of each component trait which is not provided by such studies as these estimates provide nature and magnitude but not its cause. Path coefficient (Wright 1921; Dewey and Lu 1959) under such circumstances plays an important role in partitioning the correlations into direct and indirect effects of a specific causal factor. Correlation gives only the relation between two variables whereas path analysis allows to separate the direct effect and their indirect effects through other attributes by partitioning the correlations. When a dependent character like yield is to be improved, which is governed by many independent traits through direct or indirect effects of other traits, then sometimes even character showing significant correlation with the yield may not be considered for improvement as its correlation with yield may be due to the indirect effects of this trait through other traits. Thus, it is always more appropriate to split the correlation value into direct and indirect effects through path coefficient analysis. Path coefficient analysis provides better means for selection by resolving the correlation coefficient of yield and its components into direct and indirect effects. The present investigation was therefore, aimed to estimate the direct and indirect effects of different traits on seed yield per plant and the values are presented in Table 4.11 and Table 4.12.

4.6.1 Estimates of direct effect

A residual effect of 0.00760 and 0.00149 at both phenotypic as well as genotypic level respectively, showed high contribution towards variability in grain yield per plant by the character chosen for study.

At both phenotypic as well as genotypic level 12 traits *viz.*, days to 50 per cent flowering, days to 75 per cent maturity, plant height, flag leaf width, total tillers per plant, effective tillers per plant, spikelets per panicle, 1000 grain weight, grain length, L:B ratio, biological yield per plant and harvest index exhibited significant positive correlation with grain yield per plant.

At phenotypic level, biological yield per plant (0.670) had the highest positive direct effect on grain yield per plant followed by harvest index (0.577), flag leaf length (0.038), spikelet per panicle (0.032), days to 50 per cent flowering (0.026),

total tillers per plant (0.026), 1000 grain weight (0.014), grain breadth (0.012), plant height (0.009) and L:B ratio (0.009). Thus they can be considered for direct selection for high grain yield.

At genotypic level, biological yield per plant (0.695) followed by harvest index (0.486), spikelets per panicle (0.062), flag leaf length (0.039), days to 50 per cent flowering (0.026), grain breadth (0.023), plant height (0.017) and L:B ratio (0.011). Thus they can be considered for direct selection for high grain yield.

Similar positive effect on grain yield with harvest index and days to 50 per cent flowering were reported by Kishore et al. (2018). On the other hand, positive direct effect of grain breadth, L:B ratio on grain yield was reported by Archana et al. (2018). Aditya and Bhartiya (2013) observed positive impact of days to 50 per cent flowering, days to 75 per cent maturity, plant height, tillers per plant, flag leaf length, 1000 grain weight and grain length on grain yield.

4.6.2 Estimation of indirect effect

The positive and significant correlation of days to 50 per cent flowering, days to 75 per cent maturity and plant height was the result of high indirect effect via biological yield per plant whereas significant positive correlation of flag leaf length, total tillers per plant, effective tillers per plant effect, grain lenghth and L:B ratio was contributed by high indirect effect via biological yield per plan and harvest index. These findings are supported by Aditya and Bhartiya (2013) who observed strongest positive and indirect effects of grain length, L:B ratio and 1000 grain weight on grain yield.

The lower residual effect (0.00760 and 0.00149) at phenotypic as well as genotypic levels revealed that the characters chosen in path analysis were adequate and appropriate and indicated that most of the variation found in dependent trait was well explained by the contributing traits. Based on path analysis, it was concluded that biological yield per plant and harvest index were observed as best selection indices because of their high direct contribution towards grain yield per plant.

TABLE 4.11 Estimates of direct and indirect effects on grain yield at phenotypic level for different traits

Traits	Days to 50% flowering	Days to 75% maturity	Plant height	Flag leaf length	Flag leaf width	Total tillers per plant	Effective tillers per plant	Spikelet per panicle	1000 grain weight	Grain length	Grain breadth	L:B ratio	Biological yield per plant	Harvest index	Grain yield per plant
Days to 50% flowering	**0.026**	-0.015	0.001	0.011	0.000	0.001	-0.001	0.000	-0.001	-0.002	-0.003	0.002	0.239	0.017	0.276*
Days to 75% maturity	0.021	**-0.019**	0.001	0.012	0.000	0.004	-0.005	0.004	0.000	-0.005	-0.002	0.002	0.278	0.078	0.370*
Plant height	0.004	-0.002	**0.009**	0.017	0.000	-0.003	0.002	-0.001	0.002	-0.005	-0.001	0.002	0.128	0.072	0.224*
Flag leaf length	0.008	-0.006	0.004	**0.038**	0.000	0.005	-0.007	0.002	0.002	-0.005	-0.001	0.001	0.228	0.201	0.470*
Flag leaf width	-0.001	0.000	0.002	0.003	**-0.001**	-0.008	0.009	0.003	-0.001	0.002	-0.002	0.001	-0.097	-0.025	-0.114
Total tillers per plant	0.001	-0.003	-0.001	0.008	0.000	**0.026**	-0.030	0.005	0.001	-0.014	0.001	0.003	0.392	0.285	0.672*
Effective tillers per plant	0.000	-0.003	-0.001	0.009	0.000	0.024	**-0.032**	0.006	0.001	-0.015	0.001	0.003	0.377	0.292	0.664*
Spikelet per panicle	0.000	-0.002	0.000	0.002	0.000	0.004	-0.006	**0.032**	0.004	-0.007	0.000	0.002	-0.029	0.178	0.176*
1000 grain weight	-0.002	0.000	0.001	0.005	0.000	0.001	-0.003	0.009	**0.014**	-0.006	0.002	0.001	0.052	0.164	0.236*
Grain length	0.002	-0.004	0.002	0.008	0.000	0.015	-0.019	0.009	0.003	**-0.025**	-0.002	0.007	0.359	0.320	0.676*
Grain breadth	-0.007	0.003	-0.001	-0.003	0.000	0.002	-0.002	0.001	0.002	0.003	**0.012**	-0.007	-0.103	-0.001	-0.100
L:B ratio	0.005	-0.004	0.002	0.006	0.000	0.007	-0.009	0.005	0.001	-0.019	-0.009	**0.009**	0.275	0.187	0.456*
Biological yield per plant	0.009	-0.008	0.002	0.013	0.000	0.015	-0.018	-0.001	0.001	-0.013	-0.002	0.004	**0.670**	0.142	0.814*
Harvest index	0.001	-0.003	0.001	0.013	0.000	0.013	-0.016	0.010	0.004	-0.014	0.000	0.003	0.165	**0.577**	0.755*

RESIDUAL EFFECT: 0.00760

***Significance at P≤0.05**

TABLE 4.12 Estimates of direct and indirect effects on grain yield at genotypic level for different traits

Traits	Days to 50% flowering	Days to 75% maturity	Plant height	Flag leaf length	Flag leaf width	Total tillers per plant	Effective tillers per plant	Spikelet per panicle	1000 grain weight	Grain length	Grain breadth	L:B ratio	Biological yield per plant	Harvest index	Grain yield per plant
Days to 50% flowering	**0.026**	-0.047	0.003	0.013	0.002	-0.001	-0.001	-0.001	0.000	-0.004	-0.006	0.002	0.305	0.015	0.308*
Days to 75% maturity	0.023	**-0.055**	0.004	0.014	0.000	-0.005	-0.006	0.007	0.000	-0.009	-0.004	0.003	0.401	0.074	0.447*
Plant height	0.005	-0.012	**0.017**	0.020	-0.009	0.003	0.003	-0.002	-0.001	-0.011	-0.002	0.003	0.155	0.128	0.298*
Flag leaf length	0.009	-0.020	0.009	**0.039**	-0.001	-0.007	-0.009	0.004	-0.001	-0.011	-0.002	0.002	0.313	0.237	0.563*
Flag leaf width	-0.002	0.001	0.007	0.001	**-0.023**	0.012	0.014	0.009	0.000	0.005	-0.004	0.001	-0.144	0.012	-0.112
Total tillers per plant	0.001	-0.010	-0.002	0.009	0.010	**-0.028**	-0.032	0.009	0.000	-0.025	0.002	0.003	0.484	0.291	0.713*
Effective tillers per plant	0.000	-0.009	-0.001	0.010	0.010	-0.028	**-0.032**	0.013	0.000	-0.026	0.001	0.003	0.489	0.318	0.747*
Spikelet per panicle	0.000	-0.006	-0.001	0.003	-0.003	-0.004	-0.007	**0.062**	-0.001	-0.012	0.001	0.002	-0.027	0.191	0.197*
1000 grain weight	-0.002	0.000	0.003	0.007	0.003	-0.001	-0.004	0.017	**-0.004**	-0.011	0.003	0.001	0.077	0.181	0.269*
Grain length	0.002	-0.012	0.004	0.010	0.003	-0.016	-0.020	0.017	-0.001	**-0.042**	-0.003	0.008	0.435	0.320	0.705*
Grain breadth	-0.008	0.010	-0.002	-0.003	0.004	-0.002	-0.002	0.002	0.000	0.005	**0.023**	-0.008	-0.122	-0.004	-0.106
L:B ratio	0.006	-0.013	0.004	0.007	-0.001	-0.008	-0.010	0.010	0.000	-0.032	-0.016	**0.011**	0.332	0.189	0.478*
Biological yield per plant	0.012	-0.032	0.004	0.017	0.005	-0.020	-0.023	-0.002	0.000	-0.027	-0.004	0.005	**0.695**	0.276	0.906*
Harvest index	0.001	-0.008	0.005	0.019	-0.001	-0.017	-0.021	0.024	-0.001	-0.028	0.000	0.004	0.395	**0.486**	0.857*

RESIDUAL EFFECT: 0.00149

***Significance at P≤0.05**

4.7 Genetic diversity studies through morphological markers

The selection of suitable diverse parents for hybridization programme is an important aspect of any crop improvement programme for getting desired recombinants. Genetic diversity present in the available inbred lines provides immense value on crop improvement for character(s) of interest. For selecting the parents for hybridization, which are required to be divergent enough for the character(s) of interest, estimation of the genetic distance is most important.

Mahalanobis D^2-analysis, which gives clear idea about the diverse nature of the population, is a powerful tool for estimating genetic diversity among different genotypes and to identify the parents for hybridization to obtain desirable recombinants. Evaluation of genetic diversity helps in reducing the number of breeding lines from the large germplasm and the progenies derived from diverse parents are expected to show a broad spectrum of genetic variability and provide better scope to isolate superior recombinants.

4.7.1 Test of significance

The technique of multivariate analysis was used for grouping of genotypes into clusters. Simultaneous test of significance based on Wilk's criterion and D^2 values was obtained for each pair of populations. Results of this simultaneous test of significance were observed to be significant indicating the presence of sufficient genetic diversity among genotypes.

4.7.2 Grouping of genotypes into clusters

It is the task of grouping a set of genotypes in which accessions falling in the same group are more similar to each other than to those in other groups or clusters which is very helpful in diversity analysis. In the present investigation with non-hierarchical Euclidean cluster analysis, 43 genotypes of red rice which were selected for morphological as well molecular characterization are grouped into three major clusters. All the clusters are polygenotypic based on genetic divergence (Fig. 4.1 and Fig. 4.2). Maximum number of genotypes are placed in cluster II (19 genotypes) followed by cluster I (17 genotypes), cluster IV (4 genotypes) and cluster III (3 genotypes). Genotypes falling under each cluster are presented in Table 4.13.

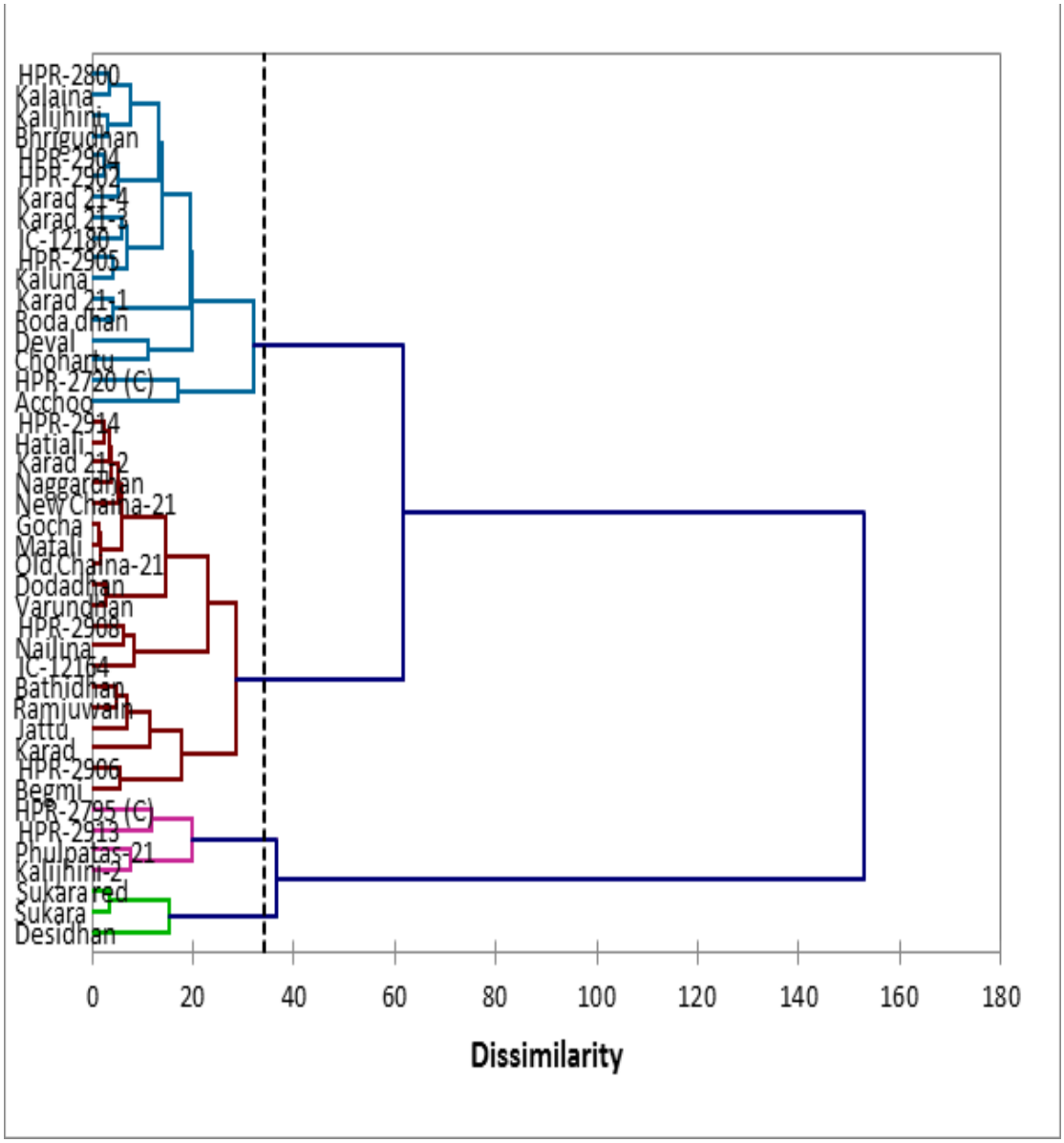

Fig. 4.1 Dendrogram of rice genotypes generated using Mahalanobis D^2-cluster analysis

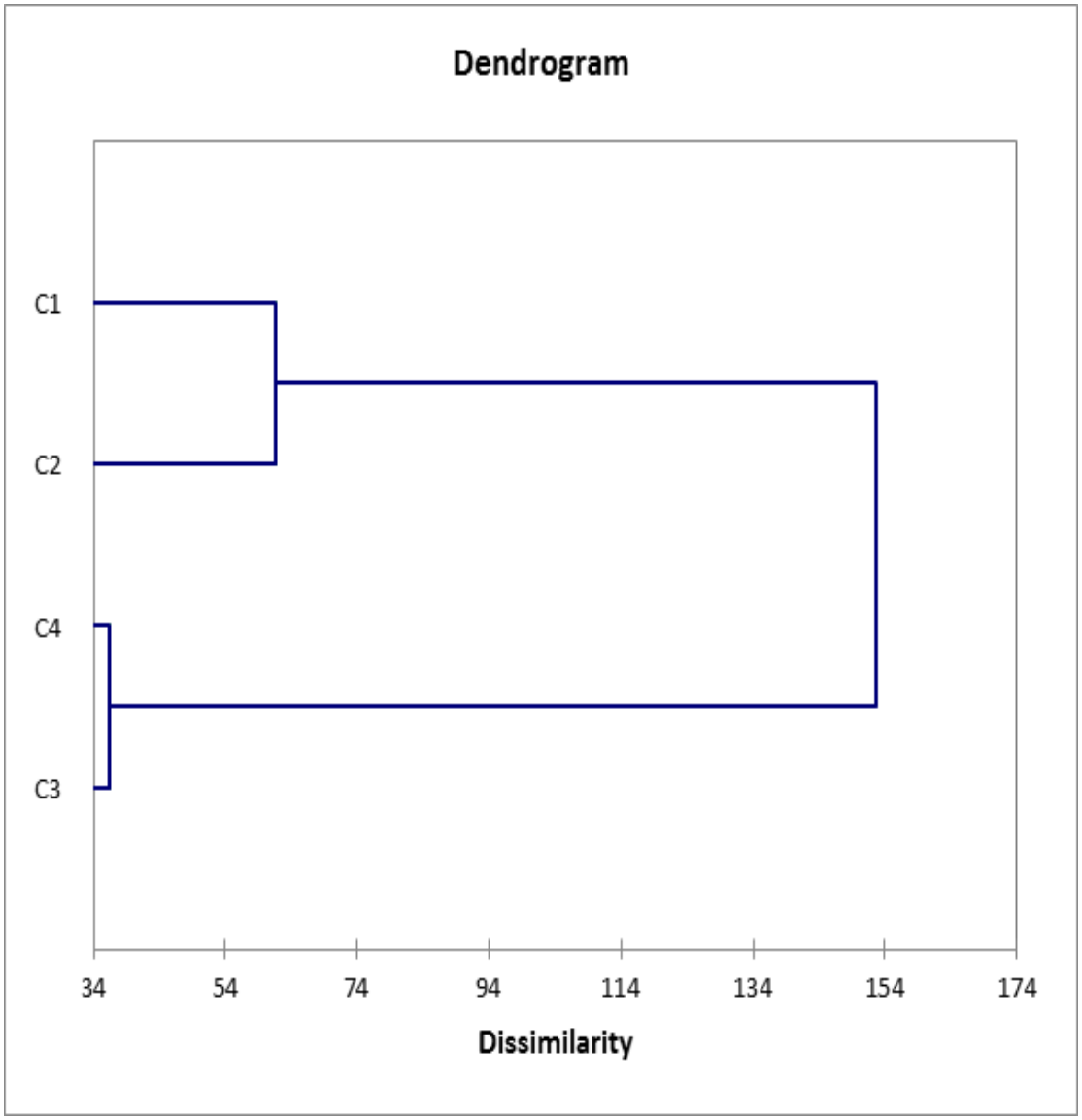

Fig. 4.2 Dendrogram of rice genotypes showing clusters

Table 4.13 Grouping of rice genotypes into different clusters on the basis of Mahalanobis D^2-analysis

Clusters	Total number of genotypes	Genotypes
Cluster I	17	IC-12180, Kaluna, Chohartu, Kalaina, Acchoo, HPR-2800, Roda Dhan, Deval, Bhrigudhan, Kalijhini, HPR-2902, HPR-2904, HPR-2905, Karad 21-1, Karad 21-3, Karad 21-4, HPR-2720 (C)
Cluster II	19	Ramjuwain, Karad, IC-12164, Begmi, Bathidhan, HPR-2906, Varundhan, Hatiali, Naggardhan, Nailina, Matali, Dodadhan, HPR-2908, HPR-2914, Gocha, New Chaina-21, Old Chaina-21, Karad 21-2, Jattu
Cluster III	3	Sukara, Desidhan, Sukara red
Cluster IV	4	Kalijhini-2, HPR-2913, Phulpatas-21, HPR-2795 (C)

Similar studies were taken up by Chakravorty et al. (2013) who assessed genetic divergence among 51 rice genotypes and grouped them into 11 clusters using D^2 analysis. The results of the present study are in agreement with the findings of Parikh et al. (2011), Devi et al. (2015), Kumar (2015), Rachappanavar (2017), Akhtar et al. (2022) and Satya et al. (2022).

Genetic diversity is generally associated with geographical diversity, but the former is not necessarily directly related with geographical distribution, the genotypes within the same clusters originated from different geographical regions of the world. This indicated that the geographical distribution and genetic divergence did not follow the same trend which might be due to continuous exchange of genetic material among the countries of the world.

4.7.3 Average intra and inter-cluster distances

The average intra and inter-cluster distances are presented in (Table 4.14). The highest intra-cluster distance is observed for cluster II (4.57) and the minimum intra-cluster distance is observed for cluster III (4.23). The highest inter-cluster distance is

observed between clusters IV and III (5.66) and least for cluster I and II (3.86) indicating that the genotypes belonging to these clusters were comparatively less diverse. High intra-cluster distances revealed that genotypes within the same cluster were quite diverse; hence the selection of parents within the cluster would be effective.

Table 4.14 Average intra and inter-cluster distance

	Cluster I	**Cluster II**	**Cluster III**	**Cluster IV**
Cluster I	**18.64 (4.31)**	14.90 (3.86)	29.99 (5.48)	29.53 (5.43)
Cluster II		**20.88 (4.57)**	31.70 (5.63)	29.95 (5.47)
Cluster III			**17.86 (4.23)**	32.04 (5.66)
Cluster IV				**19.31 (4.39)**

Bold values are intra cluster distance
Data in parenthesis are $\sqrt{D^2}$ value

The results are in accordance with the findings of Parikh et al. (2011) and Padmaja et al. (2011). Similar result are reported by Vennila et al. (2011), Chakravorty et al. (2013) and Devi et al. (2015).

4.7.4 Cluster means

The cluster means of red rice genotypes falling under different clusters are presented in (Table 4.15). The average cluster mean values for different traits revealed that the genotypes included in cluster III showed highest values for most of the traits *viz.*, days to 50 per cent flowering, days to 75 per cent maturity, flag leaf length, total tillers per plant, effective tillers per plant, biological yield per plant and grain yield per plant followed by cluster IV which showed highest values for plant height, spikelets per panicle, 1000 grain weight, grain length, L:B ratio and harvest index. Cluster II showed highest value for grain breadth, while cluster I showed maximum value for flag leaf width.

Cluster I showed minimum values for most of the traits *viz.*, total tillers per plant, effective tillers per plant, grain length, L:B ratio, biological yield per plant, harvest index and grain yield per plant followed by cluster II which showed minimum values for days to 50 per cent flowering, days to 75 per cent maturity, plant height and flag leaf length. Cluster III showed minimum values for flag leaf width, spikelets per panicle and 1000 grain weight, while Cluster IV showed minimum value for grain breadth.

Table 4.15 Cluster means of four clusters for different traits of rice genotypes

Traits	Cluster I	Cluster II	Cluster III	Cluster IV	Mean	Min	Max
Days to 50% flowering	91.12	84.42	98.00	90.25	90.95	84.42*	98.00**
Days to 75% maturity	115.80	113.23	121.11	117.08	116.81	113.23*	121.11**
Plant height	133.84	122.95	129.17	139.38	131.34	122.95*	139.38**
Flag leaf length	36.63	33.32	45.23	40.55	38.93	33.32*	45.23**
Flag leaf width	1.67	1.45	1.43	1.53	1.52	1.43*	1.67**
Total tillers per plant	6.77	8.66	11.52	9.67	9.16	6.77*	11.52**
Effective tillers per plant	6.24	7.96	10.03	8.98	8.30	6.24*	10.03**
Spikelet per panicle	56.67	60.48	52.84	78.27	62.06	52.84*	78.27**
1000 grain weight	23.09	23.31	20.87	28.09	23.84	20.87*	28.09**
Grain length	3.67	4.29	4.72	6.39	4.77	3.67*	6.39**
Grain breadth	2.08	2.20	2.03	1.89	2.05	1.89*	2.20**
L:B ratio	1.86	2.01	2.33	3.43	2.41	1.86*	3.43**
Biological yield per plant	35.60	37.79	57.75	47.46	44.65	35.60*	57.75**
Harvest index	28.48	31.87	36.33	38.40	33.77	28.48*	38.40**
Grain yield per plant	10.16	11.91	21.01	18.36	15.36	10.16*	21.01**

* Minimum ** Maximum

Among four clusters, cluster III showed the highest cluster mean values for most of the traits, suggesting that genotypes falling in cluster III can be selected directly on the basis of these traits and used in hybridization program. It has been well established that more the genetically diverse parents used in the hybridization program, greater will be the chances of obtaining high heterotic hybrids. Similar studies were also done by Sharma et al. (2011), Chakravorty et al. (2013) and Kumar (2015).

4.7.5 Contribution of individual character towards divergence

The relative per cent contribution of the individual trait to the genetic divergence among red rice genotypes is presented in (Table 4.16). The maximum contribution towards the genetic divergence is exhibited by spikelets per panicle followed by plant height, days to 50 per cent flowering and biological yield per plant.

Table 4.16 Relative contribution of individual trait towards divergence among rice genotypes

Trait	Contribution (%)
Days to 50% flowering	11.35
Days to 75% maturity	5.88
Plant height	16.75
Flag leaf length	9.14
Flag leaf width	0.37
Total tillers per plant	2.43
Effective tillers per plant	2.11
Spikelet per panicle	20.62
1000 grain weight	5.35
Grain length	1.15
Grain breadth	0.54
L:B ratio	0.86
Biological yield per plant	10.55
Harvest index	7.28
Grain yield per plant	5.64

Divergence among red rice genotypes is presented in (Table 4.16). The maximum contribution towards the genetic divergence is exhibited by spikelets per panicle followed by plant height, days to 50 per cent flowering and biological yield per plant.

The results are supported by the earlier findings of Vennila et al. (2011) who revealed that plant height contributed maximum towards genetic divergence. Also, the results are in accordance with the Asish et al. (2010), Devi et al. (2015) and Rachappanavar (2017).

On the basis of cluster analysis it can be concluded that in the population under study, high variability was observed among the genotypes of different clusters for different traits studied.

4.8 Principal component analysis (PCA)

Principal component analysis helps in identifying the most relevant characters that can be used as descriptors by explaining as much of total variation in the original set of variables as possible with a few components as possible and reducing the dimension of the problem. Principal components are significant for eigen values greater than or equal to 1.0. Out of total 15 principal components, 5 principal components are having their eigen values greater than 1.0 which is significant. As a result a total of 79.67% variation was explained by the first five significant principal component.

Eigen values and proportion of accounted variance for each variable have also been shown in (Table 4.17). The first principal component (PC1) was the most important and explained 35.62% of the total variance which was mainly contributed by grain yield per plant, biological yield per plant, grain length, effective tillers per plant and total tillers per plant, the principal component (PC2) contributed 15.30% of

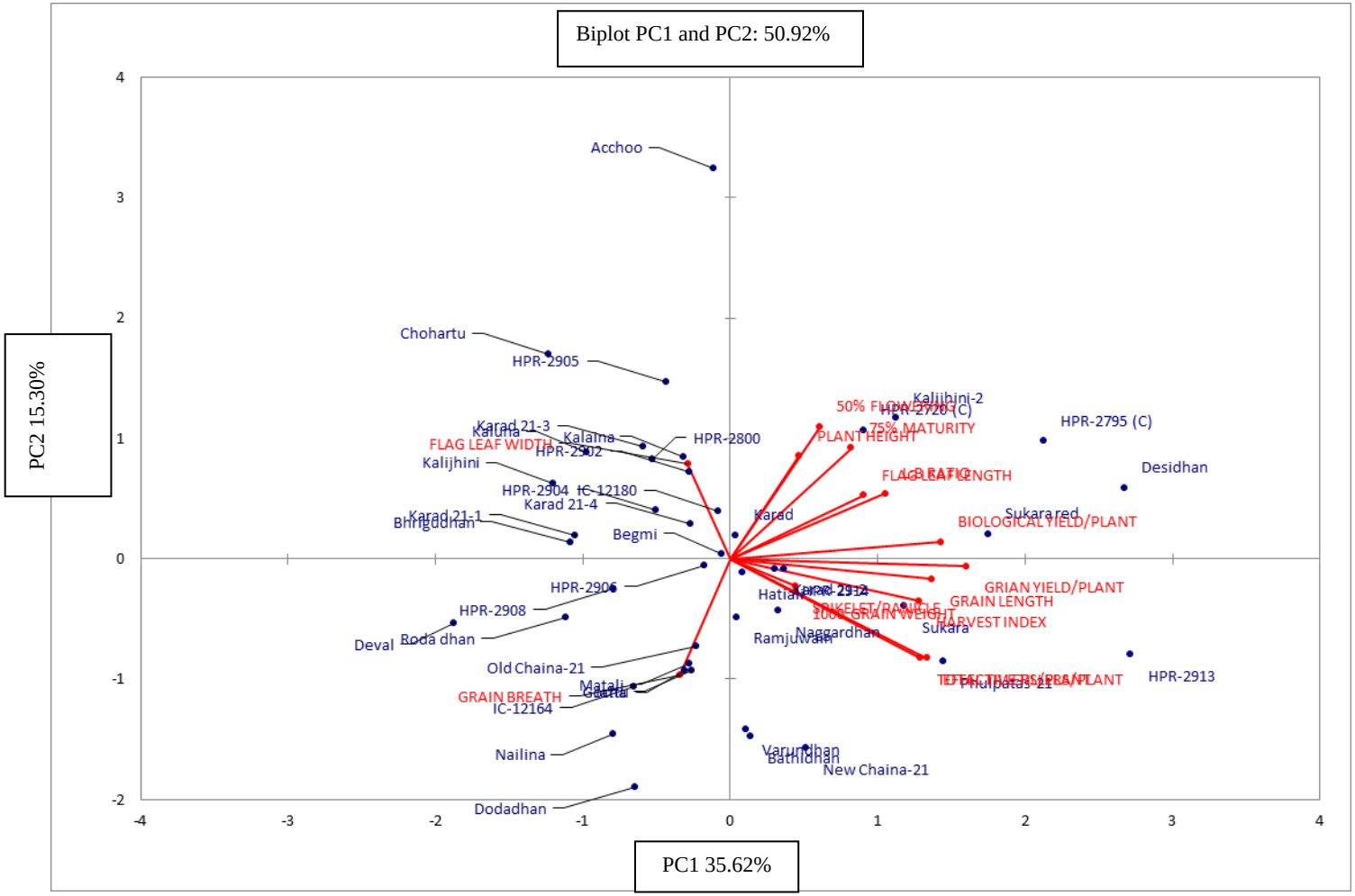

Fig. 4.3 Biplot of different variables and genotypes on PC1 and PC2

Table 4.17 Eigen vectors for five components of different traits in 43 rice genotypes

Variable	Eigenvectors				
	PC1	**PC2**	**PC3**	**PC4**	**PC5**
Eigen Value	5.34	2.30	1.71	1.49	1.11
Variation (%)	35.62	15.30	11.41	9.93	7.43
Cumulative (%)	35.62	50.92	62.32	72.26	79.69
Days to 50% flowering	0.155	0.429	-0.379	0.131	0.292
Days to 75% maturity	0.209	0.359	-0.328	0.179	0.354
Plant height	0.119	0.333	0.292	0.297	-0.390
Flag leaf length	0.230	0.205	0.068	0.397	-0.232
Flag leaf width	-0.074	0.305	0.413	0.032	-0.090
Total tillers per plant	0.330	-0.322	-0.185	-0.069	-0.086
Effective tillers per plant	0.341	-0.323	-0.135	-0.066	-0.064
Spikelet per panicle	0.112	-0.089	0.341	0.050	0.664
1000 grain weight	0.112	-0.108	0.343	0.258	0.304
Grain length	0.349	-0.067	0.197	-0.197	-0.009
Grain breadth	-0.088	-0.378	-0.068	0.551	-0.008
L:B ratio	0.269	0.212	0.202	-0.508	0.008
Biological yield per plant	0.364	0.055	-0.221	0.001	-0.156
Harvest index	0.327	-0.137	0.263	0.124	0.015
Grain yield per plant	0.410	-0.024	0.008	0.099	-0.080

the total variance through days to 50 per cent flowering, days to 75 per cent maturity, plant height and flag leaf width. The principal component (PC3) explained 11.41% of the total variance which was mainly contributed by flag leaf width, spikelets per panicle and 1000 grain weight. PC4 contributed 9.93% of the total variance through grain breadth and flag leaf length. PC5 contributed 7.43% of the total variance through spikelets per panicle, days to 75 per cent maturity and 1000 grain weight.

Considering the eigen vectors grain yield per plant, days to 50 per cent flowering, flag leaf width, grain breadth and spikelets per panicle are the major sources of diversity among these red rice genotypes.

The loading plot is drawn for PC1 and PC2 also indicated the importance of different morphological traits and genotypes for explaining the variance among accessions and understanding genotypes relationship (Fig. 4.3). Scattered diagram revealed that maximum genotypes were unique as they fall in different corners of biplot. The results are in agreement with the findings of Chakravorty et al. (2013), Singh et al. (2018), Yadav et al. (2019) and Akhtar et al. (2022).

4.9 Genetic diversity studies through molecular markers

The D^2-statistics provides a powerful conventional tool to quantify the variation in morph metric traits and also to measure the intra and inter group distances to isolate divergent genotypes. Generally, the composition of the clusters differs over environments and locations indicating that D^2-statistics is influenced by the environmental variation resulting in inconsistent clustering of some genotypes. However, genome characterization based on DNA markers is least influenced by the environment and is more reliable.

The development of molecular markers provides a means of assessing genetic diversity at the DNA level. Presently simple sequence repeat (SSR) markers have been widely used in cereal crops for diversity analysis. In particular, SSR markers are potentially useful for large-scale DNA fingerprinting of rice genotypes due to a high level of polymorphism detected, automated analysis systems and high accuracy and repeatability.

4.9.1 Marker informativeness

In the present study, a total of 96 SSR primers were screened for PCR amplification of 43 red rice genotypes. Out of 96 SSRs, 21 primers showed polymorphism (Table 4.18, Plate 4.4 and 4.5).

Table 4.18 Number of scorable and polymorphic SSR bands along with PIC value and number of alleles by 21 primers

S.No.	Primer	PIC value	Total number of allele	Effective number of alleles
1	**RM 235**	0.416	3	2.02
2	**RM 167**	0.361	2	1.90
3	**RM 130**	0.511	3	2.49
4	**RM 151**	0.361	2	1.90
5	**RM 216**	0.552	3	2.65
6	**RM 229**	0.335	2	1.74
7	**RM 153**	0.219	2	1.33
8	**RM 257**	0.270	3	1.43
9	**RM 220**	0.375	2	2.00
10	**RM 272**	0.273	2	1.48
11	**RM 240**	0.480	3	2.16
12	**RM 148**	0.309	2	1.61
13	**RM 134**	0.219	2	1.33
14	**RM 334**	0.399	3	1.65
15	**RM 421**	0.588	3	2.96
16	**RM 248**	0.110	2	1.12
17	**RM 204**	0.518	3	2.46
18	**RM 413**	0.270	3	2.35
19	**RM 441**	0.401	3	1.68
20	**RM 451**	0.474	3	2.18
21	**RM 541**	0.315	2	1.66
	TOTAL	**7.756**	**53**	**40.1**
	MEAN	**0.369**	**2.52**	**1.91**

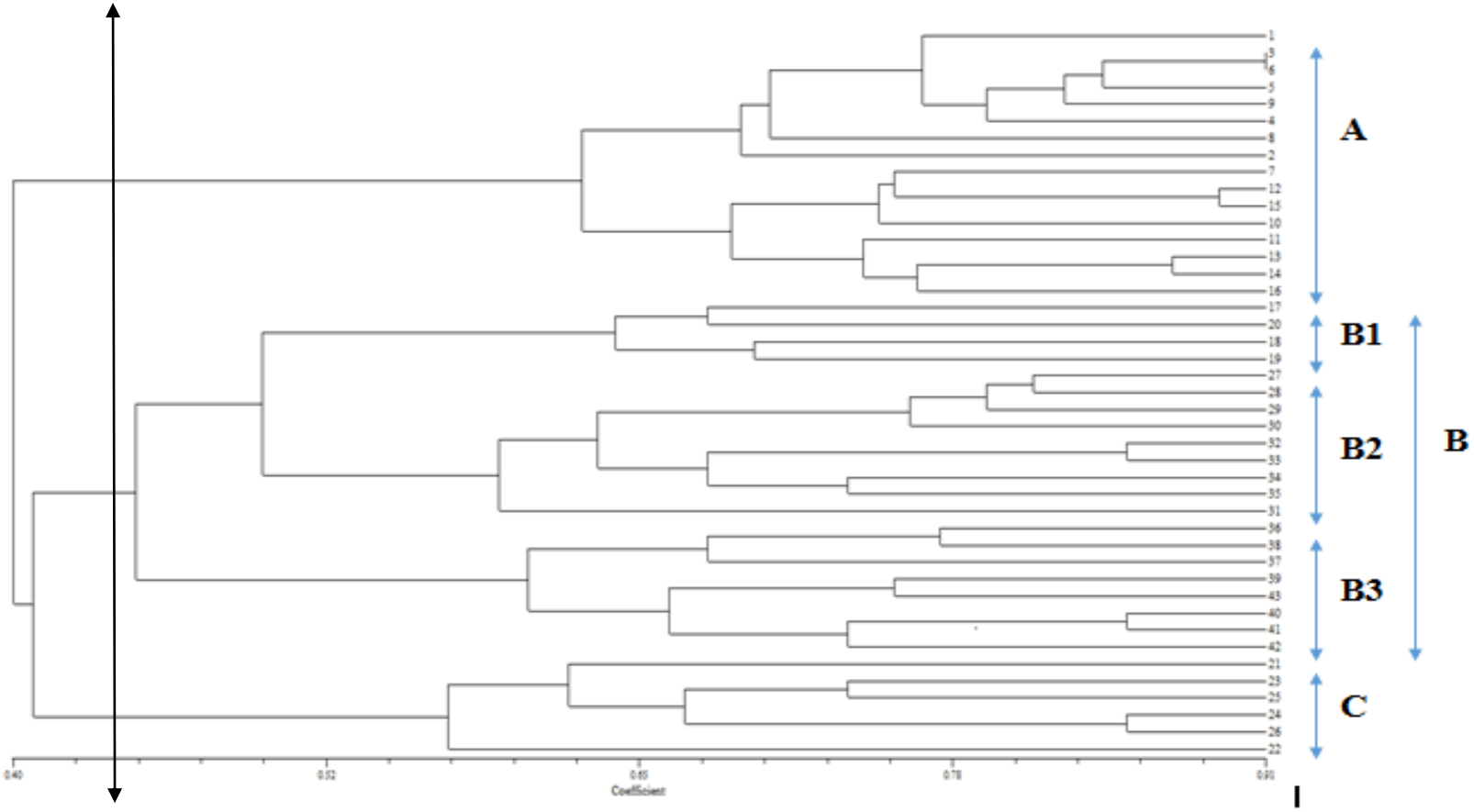

Fig. 4.4 Dendrogram depicting genetic relationships among rice genotypes constructed by NTSYS–PC (version 2.02) using UPGMA method

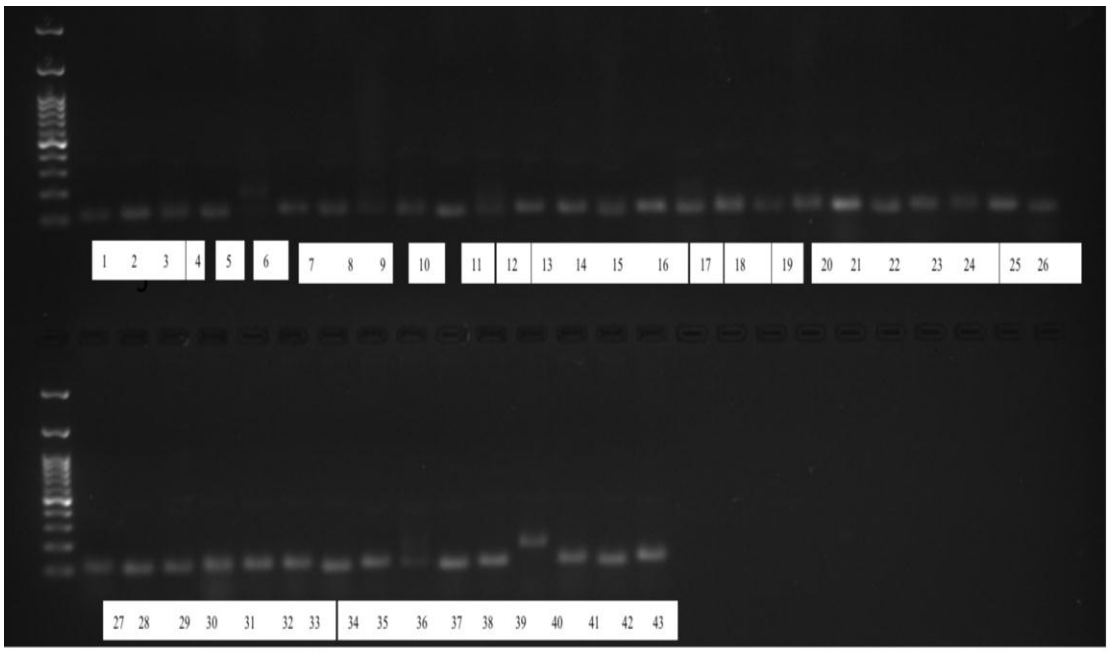

Plate 4.4 SSR profile in red rice genotypes revealed using primer *RM 204*, M=1000 bp DNA ladder

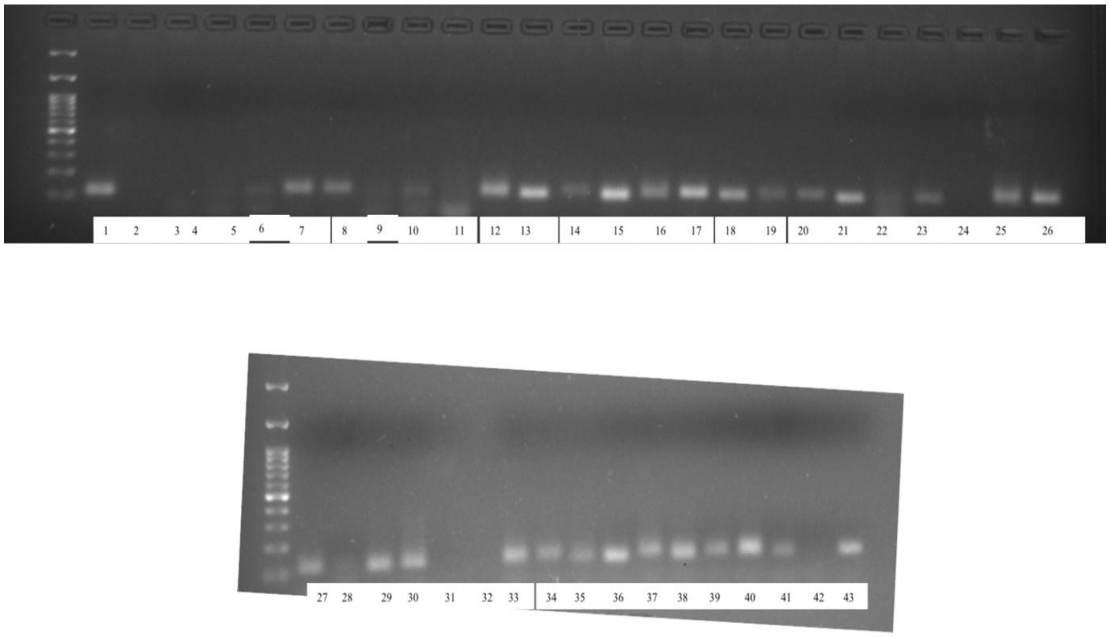

Plate 4.5 SSR profile in red rice genotypes revealed using primer *RM 229*, M=1000 bp DNA ladder

Polymorphic information content, a parameter associated with the discriminating power of markers, ranged from 0.110 (*RM248*) to 0.588 (*RM421*) with a mean of 0.369 per primer. Highest PIC was showed by the primer *RM421* (0.588) followed by *RM216* (0.522), *RM204* (0.518) and *RM130* (0.511) (Table 4.17).

This corresponded well with the average PIC value of 0.38 obtained by Vinita et al. (2013) who used microsatellite markers to study the genetic relatedness in 41 rice genotypes. This showed that the present study was in accordance with earlier observation.

The present results are also in agreement with previous observation made by several workers *viz.,* Shahriar et al. (2014a), Shahriar et al. (2014b), Ramadan and Elmoghazy(2015), Salgotra et al. (2015), Venkatesan and Bhat (2015), Ashraf et al. (2016) and Rachappanavar (2017) where an average PIC valve of 0.71, 0.65, 0.46, 0.63, 0.33, 0.62 and 0.42 was recorded, respectively. Dwivedi et al. (2019) and Hazarika and Deka (2021) also supported the present investigation who found PIC value ranged from 0.062 to 0.664 and 0.133 to 0.541 respectively.

In the present study, observed number of alleles ranged between 2 to 3 with the mean value of 2.52. Effective number of alleles ranged between 1.12 to 2.96 with the mean value of 1.91 (Table 4.18).

The results are supported with the findings of Fisseha et al. (2013), Netravati et al. (2013), Vinita et al. (2013), Shahriar et al. (2014b), Ramadan and Elmoghazy (2015), Salgotra et al. (2015), Venkatesan and Bhat (2015), Ashraf et al. (2016), Thomas and Dominic (2016) and Rachappanavar (2017). Alleles were detected with an average of 4.97, 2.08, 2.79, 9.67, 2.76, 2.8, 2.3, 2.6, 3.5 and 2.74 respectively, from diverse collection of rice accessions, cultivated rice varieties and hybrids, basmati and other scented rice varieties, Indian rice germplasm and from different parts of other countries respectively. This discrepancy might be related to the genotypes used and selection of SSR primers with scorable alleles.

4.9.2 Diversity analysis

Based on polymorphism exhibited by SSR markers, dendrogram was constructed using Jaccard's similarity coefficient using UPGMA method of NTSYS - PC package (v.2.02), the genotypes were grouped into two main clusters (Fig. 4.4).

On the basis of SSR data, 43 red rice genotypes were divided into three clusters. Cluster B contains maximum number of genotypes i.e., 21, followed by cluster A (16 genotypes) and cluster C (6 genotypes). Cluster B was further sub-divided into three sub clusters (Table 4.19).

The cluster A has 16 genotypes *viz.*, IC-12164, IC-12180, Kaluna, Karad, Chohartu, Ramjuwain, Acchoo, Kalaina, Sukara, HPR-2906, Desidhan, Begmi, Kalijhini-2, Sukara red, Bathidhan and Varundhan. Within the three sub-clusters of cluster B the sub-cluster B2 has maximum (9) genotypes *viz.*, HPR-2902, HPR-2904, HPR-2905, HPR-2908, HPR-2913, HPR-2914, Gocha, HPR-2720 (C) and HPR-2795 (C) followed by sub-cluster B3 which has (8) inbred lines *viz.*, New Chaina-21, Old Chaina-21, Phulpatas-21, Karad 21-1, Karad 21-2, Karad 21-3, Karad 21-4 and Jattu.

Table 4.19 Grouping of rice genotypes into different clusters on the basis of SSR data

Clusters	Sub Clusters	No. of Genotypes	Genotypes
A		16	IC-12164, IC-12180, Kaluna, Karad, Chohartu, Ramjuwain, Acchoo, Kalaina, Sukara, HPR-2906, Desidhan, Begmi, Kalijhini-2, Sukara red, Bathidhan, Varundhan
B (21)	B1	4	Hatiali, HPR-2800, Naggardhan, Roda Dhan
	B2	9	HPR-2902, HPR-2904, HPR-2905, HPR-2908, HPR-2913, HPR-2914, Gocha, HPR-2720 (C), HPR-2795 (C)
	B3	8	New Chaina-21, Old Chaina-21, Phulpatas-21, Karad 21-1, Karad 21-2, Karad 21-3, Karad 21-4, Jattu
C		6	Nailina, Deval, Bhrigudhan, Kalijhini, Matali, Dodadhan

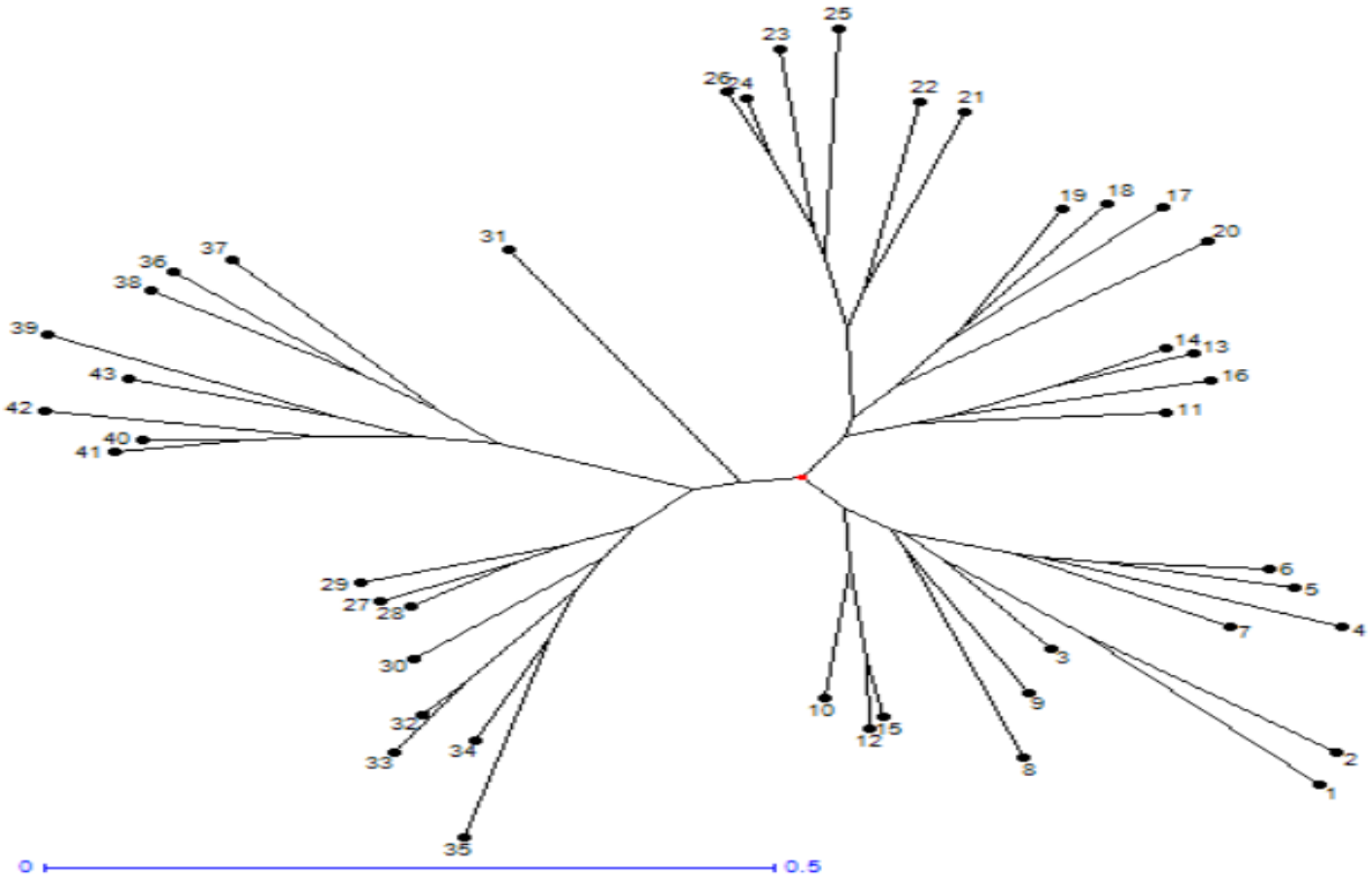

Fig. 4.5 Neighbor-joining tree using SSR markers generated by DARwin software

Sub-cluster B1 has (4) genotypes *viz.,* Hatiali, HPR-2800, Naggardhan and Roda Dhan.

Using SSR data, neighbour-joining tree was computed using the DARwin software (v.5.0) (Perrier and Jacquemoud 2006) (Fig. 4.5). Branch robustness was tested using 1000 bootstraps. Here, also SSR analysis resulted in almost similar grouping of the test population with the Jaccard's similarity coefficient using UPGMA method. Three branches emerged from the central point showing that the genotypes are divided into three clusters. This is in confirmation with the results of Vinita et al. (2013), Rachappanavar (2017) and Singh et al. (2018).

4.9.3 Population Structure

Detailed knowledge of genetic relatedness among individuals in an association panel is a key factor to avoid spurious associations. Population structure (Q matrix), estimated using STRUCTURE (Falush et al. 2003) and expressed as membership probabilities, is one way to correct spurious associations due to genetic relatedness. It is often difficult to estimate the true number of population (k). Generally, k is taken to be the value with the highest estimated LnP (D) value returned by STRUCTURE (Pritchard et al. 2000). However, in real situations, few data sets confirm precisely to the STRUCTURE model and the LnP (D) value keeps increasing when k reaches the true number. Evanno et al. (2005) suggested an ad hoc method, Δk (Delta), the second-order rate of change of the likelihood function with respect to k performed well in indicating the real number.

In this study, 43 red rice genotypes were subjected to STRUCTURE analysis for estimation of population structure using a panel of 21 SSR markers. For estimation of the exact population substructure (K), ten independent Ks (from K=2 to K=10 where, K is kinship matrix) were initiated. The STRUCTURE analysis divided the population into two groups which was the most biologically meaningful population structure because of the rapid rate of change in LnP (D) values between successive k, but the differentiations at K=2 were almost consistent with pedigree knowledge with few exceptions. Thus, the pedigree information was used to guide the division of P1 and P2 groups combining with the cluster membership (Fig. 4.6).

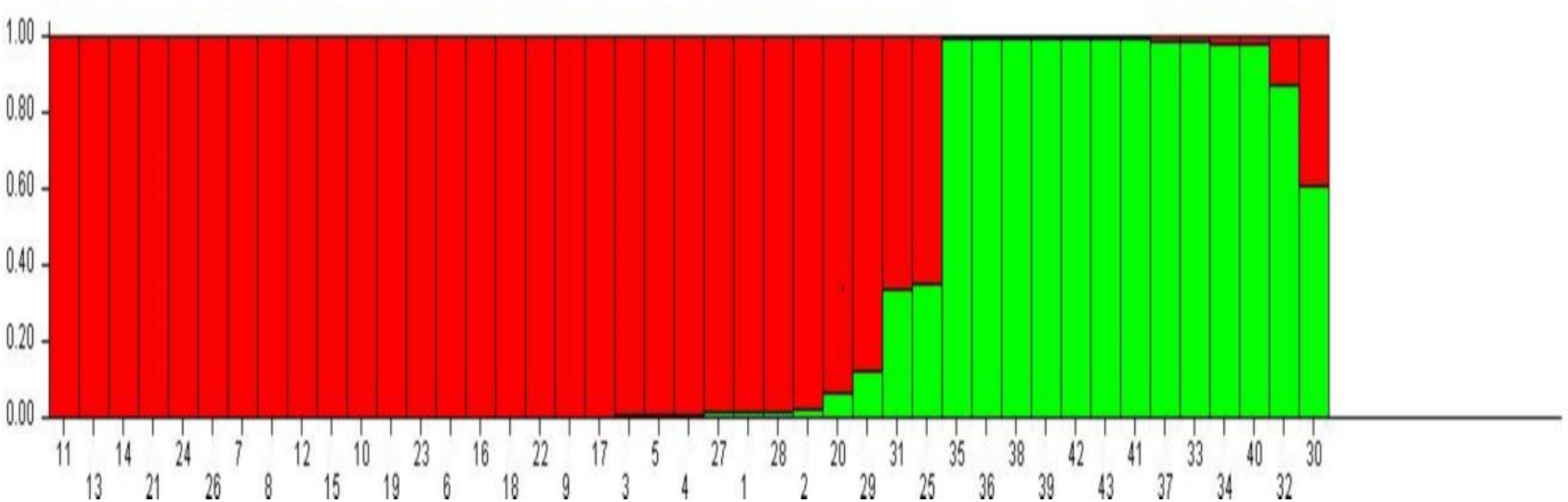

Fig. 4.6: Population Structure of 43 rice genotypes using STRUCTURE software program (K=2)

However, there are two main disadvantages of this panel used for association mapping. First, the sample size of this panel was very small and it had relatively low power of association mapping. It shows that this panel is suitable for the traits controlled by major QTLs and needs to be extended for further investigating the genetic basis of interesting traits controlled by genes with moderate or even minor effects. Secondly, structure analysis using this panel would increase false negative association and reduce the power for traits strongly correlated with population structure. This was in confirmation with the results of Yadav et al. (2019).

4.9.4 Comparison of genetic diversity through morphometric and molecular markers

Characterization of diversity in majority of plant species is primarily based on morphological evaluation which is vulnerable to intrinsic and extrinsic factors. It is desirable to compare any molecular characterization with morphological markers to understand the degree of correspondence between the two.

D^2 analysis of morphological data divided rice genotypes into four separate clusters, of which cluster II had 19 genotypes followed by cluster I (17 genotypes), cluster IV (4 genotypes) and cluster III (3 genotypes) whereas, molecular data grouped rice genotypes into three clusters of which cluster B had maximum 21 genotypes followed by cluster A having 16 genotypes and cluster C having 6 genotypes.

When comparing molecular clusters (SSR) individually with morphometric clusters, the similarity between clusters was found (Table 4.20). Molecular cluster A was compared with D^2 cluster II and III having seven and two genotypes in common respectively. When molecular cluster B was compared with D^2 cluster I, cluster II and cluster IV, nine, eight and three genotypes were found to be common respectively. When molecular cluster C was compared with D^2 cluster I and cluster II, three genotypes were found to be common in each cluster. Therefore, based on D^2 clustering and molecular clustering, total 36 genotypes were found to be in common.

Table 4.20 Comparison of clustering patter using D^2 analysis and SSR data

SSR data molecular			D^2 morphological data				
Cluster No.	**No. of genotypes in each cluster**	**Genotypes**	**Cluster No.**	**No. of genotypes in each cluster**	**Genotypes**	**Common genotypes**	**No of Common genotypes**
A	16	IC-12164, IC-12180, Kaluna, Karad, Chohartu, Ramjuwain, Acchoo, Kalaina, Sukara, HPR-2906, Desidhan, Begmi, Kalijhini-2, Sukara red, Bathidhan, Varundhan	II	19	Ramjuwain, Karad, IC-12164, Begmi, Bathidhan, HPR-2906, Varundhan, Hatiali, Naggardhan, Nailina, Matali, Dodadhan, HPR-2908, HPR-2914, Gocha, New Chaina-21, Old Chaina-21, Karad 21-2, Jattu	Ramjuwain, Karad, IC-12164, Begmi, Bathidhan, HPR-2906, Varundhan,	7
			III	3	Sukara, Desidhan, Sukara red	Sukara, Sukara red	2
B	21	Hatiali, HPR-2800, Naggardhan, Roda Dhan HPR-2902, HPR-2904, HPR-2905, HPR-2908, HPR-2913, HPR-2914, Gocha, HPR-2720 (C), HPR-2795 ©, New Chaina-21, Old Chaina-21, Phulpatas-21, Karad 21-1, Karad 21-2, Karad 21-3, Karad 21-4, Jattu	I	17	IC-12180, Kaluna, Chohartu, Kalaina, Acchoo, HPR-2800, Roda Dhan, Deval, Bhrigudhan, Kalijhini, HPR-2902, HPR-2904, HPR-2905, Karad 21-1, Karad 21-3, Karad 21-4, HPR-2720 (C)	HPR-2800, Roda Dhan, HPR-2902, HPR-2904, HPR-2905, Karad 21-1, Karad 21-3, Karad 21-4, HPR-2720	9
			II	19	Ramjuwain, Karad, IC-12164, Begmi, Bathidhan, HPR-2906, Varundhan, Hatiali, Naggardhan, Nailina, Matali, Dodadhan, HPR-2908, HPR-2914, Gocha, New Chaina-21, Old Chaina-21, Karad 21-2, Jattu	Karad 21-2, Hatiali, Naggardhan, HPR-2908, HPR-2914, Gocha, New Chaina-21, New Chaina-2	8
			IV	4	Kalijhini-2, HPR-2913, Phulpatas-21, HPR-2795 (C)	HPR-2913, Phulpatas-21, HPR-2795 (C)	3
C	6	Nailina, Deval, Bhrigudhan, Kalijhini, Matali, Dodadhan	I	17	IC-12180, Kaluna, Chohartu, Kalaina, Acchoo, HPR-2800, Roda Dhan, Deval, Bhrigudhan, Kalijhini, HPR-2902, HPR-2904, HPR-2905, Karad 21-1, Karad 21-3, Karad 21-4, HPR-2720 (C)	Deval, Bhrigudhan, Kalijhini	3
			II	19	Ramjuwain, Karad, IC-12164, Begmi, Bathidhan, HPR-2906, Varundhan, Hatiali, Naggardhan, Nailina, Matali, Dodadhan, HPR-2908, HPR-2914, Gocha, New Chaina-21, Old Chaina-21, Karad 21-2, Jattu	Nailina, Matali, Dodadhan	3

Adequate genetic diversity among different genotypes of red rice was revealed using both D^2-statistic and SSR analysis. Based on morphological and molecular evidence, there was parallelism between components of the clusters. The present study revealed that although genetic diversity analysis on the basis of agro-morphological traits is an important tool to identify genotypes showing similarities or dissimilarities, but, oftenly it is highly influenced by the environment. Whereas the structural/molecular level approach efficiently eliminates the blurring effect and discloses similarities among genotypes. By characterizing cultivars following molecular analysis, it was possible to research the degree of diversity within and between genotypes and to create an index of genetic similarity between genotypes. On the basis of these observations, SSR markers have been identified as a powerful tool over agro-morphological genetic diversity studies.

5. SUMMARY AND CONCLUSIONS

The present investigation entitled "Agro-morphological and molecular characterization of red rice (*Oryza sativa* L.) germplasm of Himachal Pradesh" was undertaken to assess the genetic diversity and characterization of red rice germplasm using morphological and molecular markers and to identify the potential germplasm lines for grain yield and its related traits.

The experimental material of the present study comprised of 41 red rice genotypes along with two checks namely HPR-2720 and HPR-2795. These genotypes were evaluated for various quantitative and quality traits including disease reaction to leaf and neck blast under natural field conditions.

Total eighteen agro-morphological and quality characters were studied including days to 50 per cent flowering, days to 75 per cent maturity, plant height (cm), flag leaf length (cm), flag leaf width (cm), total tillers per plant, effective tillers per plant, spikelets per panicle, 1000-seed weight (g), biological yield per plant (g), harvest index (%), grain yield per plant (g), seed coat color, grain length [L] (mm), grain breadth [B] (mm), L:B ratio, lodging susceptibility (%) and awns.

Mean performance of the genotypes revealed that Desidhan was superior to the best check HPR-2795 for grain yield per plant. However, two genotypes *viz.*, Desidhan and HPR-2913 were found superior to the second check HPR-2720. Based upon overall performance of genotypes Desidhan HPR-2913 and Sukara red were found to be superior for most of the traits *viz.,* days to 75 per cent maturity, flag leaf length, total tillers per plant, effective tillers per plant and grain yield and hence can be used in hybridization programme.

Four genotypes *viz.,* IC-12180, HPR-2800, HPR-2795 and HPR-2913 showed resistance to the leaf blast while, five genotypes *viz.,* HPR-2914, HPR-2720, Desidhan, Sukaraand Sukara Red showed moderately resistant reaction. However, four genotypes *viz.,* IC-12180, HPR-2800, HPR-2795 and HPR-2914 showed resistance to the neck blast while, three genotypes viz., HPR-2720, HPR-2913 and

Desidhan were found to be moderately resistant. Overall, IC-12180, HPR-2800, HPR-2795, HPR-2913, Desidhan and HPR-2720 were observed to be highly promising against both leaf and neck blast. Hence, these genotypes can be used as donors in breeding programme for blast resistance.

Perusal of the data on analysis of variance (ANOVA) revealed the presence of significant difference among the genotypes for all the traits studied except flag leaf width justifying the presence of sufficient genetic variability. Hence, presenting scope for improvement of these traits through hybridization and selection.

Analysis of parameter of variability showed that phenotypic coefficient of variation (PCV) were higher than their respective genotypic coefficient of variation (GCV) for all the traits studied which indicated the effect of environment on the expression of traits. High PCV and GCV (>20%) was observed for total tillers per plant, spikelets per panicle, L:B ratio and grain yield per plant.

High heritability was reported for days to 50 per cent flowering, days to 75 per cent maturity, total tillers per plant, 1000 grain weight, grain length, grain breadth, L:B ratio and grain yield per plant. Overall high heritability coupled with high genetic advance was observed for total tillers per plant, 1000 grain weight, grain length, grain breadth, L:B ratio and grain yield per plant indicating additive gene action of these traits. High heritability coupled with low genetic advance was observed for days to 75 per cent maturity indicating the role of non-additive gene action and selection for these traits would be less effective.

Correlation studies revealed that grain yield per plant had significant positive correlation with days to 50 per cent flowering, days to 75 per cent maturity, plant height, flag leaf length, total tillers per plant, effective tillers per plant, spikelets per panicle, 1000 grain weight, grain length, L:B ratio, biological yield per plant and harvest index implying that selection for higher yield through these traits would be effective.

Path studies revealed that biological yield per plant had the highest positive effect on grain yield per plant followed by harvest index, flag leaf length, spikelet per

panicle, days to 50 per cent flowering and total tillers per plant revealing these are the important traits for direct selection for yield.

Genetic diversity studies using Mahalanobis D^2-analysis was conducted for 43 genotypes, which grouped these genotypes into four clusters indicating the presence of considerable genetic diversity among all the genotypes. Maximum genotypes were placed in cluster II which included 19 genotypes (Ramjuwain, Karad, IC-12164, Begmi, Bathidhan, HPR-2906, Varundhan, Hatiali, Naggardhan, Nailina, Matali, Dodadhan, HPR-2908, HPR-2914, Gocha, New Chaina-21, Old Chaina-21, Karad 21-2 and Jattu) followed by cluster I consisting 17 (IC-12180, Kaluna, Chohartu, Kalaina, Acchoo, HPR-2800, Roda Dhan, Deval, Bhrigudhan, Kalijhini, HPR-2902, HPR-2904, HPR-2905, Karad 21-1, Karad 21-3, Karad 21-4 and HPR-2720) genotypes, cluster IV containing 4 genotypes (Kalijhini-2, HPR-2913, Phulpatas-21 and HPR-2795) and cluster III containing 3 genotypes (Sukara, Desidhan and Sukara red).

The intra cluster distance was maximum in cluster II (4.57) and minimum in cluster III (4.23), whereas, highest inter-cluster distance (5.66) was recorded between cluster III (Sukara, Desidhan and Sukara red) and cluster IV (Kalijhini-2, HPR-2913, Phulpatas-21 and HPR-2795). Hence, it has been well established that the genotypes belonging to these clusters are genetically diverse. More the genetically diverse parents used in the hybridization program, greater will be the chances of obtaining high heterotic hybrids.

Among four clusters, cluster III showed the highest cluster mean values for most of the traits suggesting that genotypes falling in cluster III (Sukara, Desidhan and Sukara red) can be selected directly on the basis of these traits and be used in hybridization program.

Principal component analysis revealed that the first principal component (PC1) was observed to be the highest contributing (relative contribution %) to the total variation, which was mainly through the biological yield per plant, grain length, effective tillers per plant and total tillers per plant.

Based on polymorphism exhibited by SSR markers, dendrogram was constructed using Jaccard's similarity coefficient and the genotypes were grouped into three major clusters. Cluster I comprised of 16 genotypes (IC-12164, IC-12180, Kaluna, Karad, Chohartu, Ramjuwain, Acchoo, Kalaina, Sukara, HPR-2906, Desidhan, Begmi, Kalijhini-2, Sukara red, Bathidhan and Varundhan), 21 genotypes were placed in cluster II (Hatiali, HPR-2800, Naggardhan, Roda Dhan, HPR-2902, HPR-2904, HPR-2905, HPR-2908, HPR-2913, HPR-2914, Gocha, HPR-2720 (C), HPR-2795 ©, New Chaina-21, Old Chaina-21, Phulpatas-21, Karad 21-1, Karad 21-2, Karad 21-3, Karad 21-4 and Jattu) and cluster III comprised of 6 genotypes (Nailina, Deval, Bhrigudhan, Kalijhini, Matali and Dodadhan).

A total of 35 genotypes were found common while comparing molecular and morpho-metric clusters, thereby exhibiting the congruence between morphological and SSR data.

Conclusions

- Desidhan HPR-2913 and Sukara red were found promising genotypes on the bases of their mean performance for most of the traits.

- Based on the disease reaction, genotypes namely IC-12180, HPR-2800, HPR-2795, HPR-2913, Desidhan and HPR-2720 were observed to be highly promising genotypes against both leaf and neck **blast. Hence** can be included as donors in breeding for blast resistance.

- Sufficient genetic variability and significant differences were observed among the genotypes for all the traits except flag leaf width suggesting prevalence of wide range of genetic variation and scope of selection for these traits among the genotypes.

- The estimate of PCV was higher than corresponding GCV for all the characters studied which indicated that the apparent variation is not only due to genotypes but also due to the influence of environment.

- High PCV and GCV values were recorded for total tillers per plant, spikelets per panicle, L:B ratio and grain yield per plant. So, these traits provide the higher chances for selection.

- High heritability coupled with high genetic advance were recorded for total tillers per plant, 1000 grain weight, grain length, grain breadth, L:B ratio and grain yield per plant indicated that these characters are governed by additive genes and selection will be rewarding for improvement of such traits.

- Traits namely days to 50 per cent flowering, days to 75 per cent maturity, plant height, flag leaf length, total tillers per plant, effective tillers per plant, spikelets per panicle, 1000 grain weight, grain length, L:B ratio, biological yield per plant and harvest index showed significant positive correlation with grain yield per plant.
- Highest direct effect was found for biological yield per plant followed by harvest index, flag leaf length, spikelet per panicle, days to 50 per cent flowering and total tillers per plant indicating that selection would be effective for these traits.
- Principal component analysis revealed that out of 15 PCs, 5 PCs were having eigen values more than 1 and showed 79.69% of total variation. PC1 and PC2 showed about 50.91% variability among all the traits.

- Genetic diversity studies using Mahalanobis D^2-statistic grouped 43 red rice genotypes into 4 clusters where highest intra-cluster distance was observed for Cluster II while maximum inter-cluster distance was observed between Cluster IV (Kalijhini-2, HPR-2913, Phulpatas-21 and HPR-2795) and Cluster III (Sukara, Desidhan and Sukara red) suggesting the possibilities of having a diverse segregating population if parents are chosen from these clusters for hybridization programme.

- Cluster III showed the highest cluster mean values for most of the traits suggesting that genotypes falling in cluster III can be selected directly on the basis of these traits and used in hybridization program.

- Molecular diversity studies using SSR markers according to NTSYS-PC (version 2.02) grouped 43 red rice genotypes into 3 clusters and STRUCTURE software divided them into two populations.

- Upon comparison of genetic diversity and molecular diversity studies a high level of correspondence of results was observed as most of the genotypes were common among the clusters. Genetic diversity based on D^2 revealed that 35 genotypes showed correspondance of results with molecular diversity analysis.

LITERATURE CITED

Abebe T, Alamerew S and Tulu L. 2017. Genetic variability, heritability and genetic advance for yield and its related traits in rainfed lowland rice (*Oryza sativa* L.) genotypes at Fogera and Pawe, Ethiopia. *Advances in Crop Science and Technology* 5: 272

Aditya JP and Bhartiya A. 2013. Genetic variability, correlation and path analysis for quantitative characters in rainfed upland rice of Uttarakhand hills. *Journal of Rice Research* 6: 24-34

Ahmad F, Hanafi MM, Hakim MA, Rafii MY, Arolu IW and Akmar Abdullah SN. 2015. Genetic divergence and heritability of 42 coloured upland rice genotypes (*Oryza sativa* L.) as revealed by microsatellites marker and agro-morphological traits. *Public Library of Science One* 10
DOI: 10.1371/journal.pone.0138246

Akhtar R, Iqbal A and Dasgupta T. 2022. Genetic diversity analysis of aromatic rice (*Oryza sativa* L.) germplasm based on agro-morphological characterization. *Oryza* 59: 141-149

Alagappan P and Bhardwaj N. 2022. Morphological characterization of red rice germplasm of Himachal Pradesh and identification of potential genotype for yield and biotic stress tolerance. *Journal of Cereal Research* 14: 63-75

Al-Jibouri HA, Miller PA and Robinson HF. 1958. Genotypic and environment variances and covariances in upland cotton cross of interspecific origin. *Agronomy Journal* 50: 633-636

Anderson JA, Churchill GA, Autrique JE, Tanksley SD and Sorrells ME. 1993. Optimizing parental selection for genetic linkage maps. *Genome* 36: 181-186

Anonymous a. 2021. Food and Agricultural organization of the United Nations. http://www. FAOstat. fao.org

Anonymous b. 2021. Statistical outline of Himachal Pradesh. Directorate of Economics and Statistics, Government of Himachal Pradesh, India

Archana RS, Rani MS, Vardhan KV and Fareeda G. 2018. Correlation and path coefficient analysis for grain yield, yield components and nutritional traits in rice (*Oryza sativa* L.). *International Journal of Chemical Studies* 6: 189-195

Ashraf H, Husaini AM, Ashraf Bhat M, Parray GA, Khan S and Ganai NA. 2016. SSR based genetic diversity of pigmented and aromatic rice (*Oryza sativa* L.) genotypes of the western Himalayan region of India. *Physiology and Molecular Biology of Plants* 22: 547-555

Asish K, Binodh R, Kalaiyarasi and Thiyagarajan K. 2010. Genetic divergence of rice varieties and hybrids for quality traits. *Oryza* 47: 91-95

Bagudam R, Eswari KB, Badri J and Rao PR. 2018. Correlation and path analysis for yield and its component traits in NPT core set of rice (*Oryza sativa* L.). *International Journal of Current Microbiology and Applied Sciences* 7: 97-108

Bagudam R, Eswari KB, Badri J and Rao PR. 2018. Variability, heritability and genetic advance for yield and its component traits in NPT core set of rice (*Oryza sativa* L.). *Electronic Journal of Plant Breeding* 9: 1545-1551

Bekavac G, Purar B, Stojakovic M, Jockovic D, Ivanovic M and Nastasic A. 2007. Genetic analysis of stay-green trait in broad-based maize populations. *Cereal Research Communications* 35: 31-41

Bekavac G, Purar B, Stojakovic M and Jockovic D. 2008. Relationship between line per se and testcross performance for agronomic traits in two broad-based populations of maize. *Euphytica* 162: 363-369

Bhattachraya S and Chakraborty NR. 2019. Assessment of genetic variability, correlation and path association for yield and yield components in aromatic non-basmati rice. *Journal of Pharmacognosy and Phytochemistry* 8: 1907-1914

Bisht IS, Pandey A, Yadav MC, Singh AK, Pandravada SR and Rana JC. 2018. Population structure of some indigenous aromatic rice (*Oryza sativa* L.) landraces of India. *Indian Journal of Biotechnology* 17: 110-117

Bocanski A, Sreckov Z, Natasic I, Alovic and Vukosavjev M. 2010. Correlation and path coefficient analysis of morphological traits of maize (*Zea mays* L.). *Research Journal of Agricultural Sciences* 42: 292-296

Burton GW. 1952. Qualitative inheritance in grasses. Vol. 1. In: Proceedings of the 6th International Grassland Congress, Pennsylvania State College. P 17-23

Burton GW and DeVane EH. 1953. Estimating heritability in tall Fescue (*Festuca arundinacea*) from replicated colonal material. *Agronomy Journal* 45: 478-481

Burton GW and DeVane EH. 1958. Estimating heritability in tall Fescue (*Festuca arundinacea*) from replicated colonal material. *Agronomy Journal* 45: 310-314

Chakravorty A and Ghosh PD. 2013. Estimation of variability, diversity and correlation studies with respect to different agro-morphological traits in traditional rice (*Oryza sativa* L.) of West Bengal, India. *Oryza* 50: 127-131

Chakravorty A, Ghosh PD and Sahu PK. 2013. Multivariate analysis of phenotypic diversity of landraces of rice of West Bengal. *American Journal of Experimental Agriculture* 3: 110-123

Chowdhury BD, Nath A and Dasgupta T. 2016. Evaluation of some popular rice genotypes with special emphasis on zinc, iron and protein content. *International Journal of Life Sciences* 51: 84-92

Das S and Ghosh A. 2011. Characterization of rice germplasm of West Bengal. *Oryza* 47: 201-205

Devi GR, Babu VR, Padmavathi G and Sunitha T. 2015. Genetic diversity in grain quality traits of rice genotypes. *Journal of Rice Research* 8: 28-31

Devi KR, Parimala K, Venkanna V, Lingaiah N, Hari Y and Chandra BS. 2016. Estimation of variability for grain yield and quality traits in rice (*Oryza sativa* L.). *International Journal of Pure and Applied Bioscience* 4: 250-255

Dewey DR and Lu KH. 1959. A Correlation and path coefficient analysis of components of crested wheat grass seed production. *Agronomy Journal* 51: 510-515

Dhurai SY, Bhati PK and Saroj SK. 2014. Studies on genetic variability for yield and quality characters in rice (*Oryza sativa* L.) under integrated fertilizer management. *The Bioscan* 9: 745-748

Dutta P, Dutta PN and Borua PK. 2013. Diversity analysis of some selected rice genotypes through SSR-based molecular markers, morphological traits as selection indices in rice. A statistical view. *University Journal of Agriculture Research* 1: 85-96

Dwivedi A, Rathour R, Basandrai D and Sarial AK. 2019. Molecular genetic diversity analysis using SSR markers of basmati rice (*Oryza sativa* L.) genotypes of northern hill region, India. *Journal of Cereal Research* 11: 224-230

Earl DA and vonHoldt BM. 2012. STRUCTURE HARVESTER: a website and program for visualizing STRUCTURE output and implementing the Evanno method. *Conservation Genetics Resources* 4: 359-361

Evanno G, Regnaut S and Goudet J. 2005. Detecting the number of clusters of individuals using the software STRUCTURE: a simulation study. *Molecular Ecology* 14: 2611-2620

Falush D, Stephens M and Pritchard JK. 2003. Inference of population structure using multilocus genotype data: linked loci and correlated allele frequencies. *Genetics* 164: 1567-87

Fisher RA and Yates Y. 1963. Statistical tables for biological, agricultural and medical research. Oliver and Boyd, Edinburgh, London P 155

Fisseha W, Tanee S, Chalermpol P and Prapa S. 2013. Genetic diversity analysis of rice cultivars from various origins using simple sequence repeat (SSR) markers. *African Journal of Biotechnology* 12: 4074-4081

Hanson CH, Robinson HF and Comstock RE. 1956. Biometrical studies of yield in segregating populations of Korean lespedeza. *Agronomy Journal* 48: 268-272

Hazarika D and Deka SD. 2021. Variation for grain morphology, molecular diversity and aroma analysis in speciality rice of Assam. *Journal of Cereal Research* 13: 84-91

IRRI. 2014. Standard Evaluation System (SES) for Rice, International Rice Research Institute, Los Banos, Philippines, 5th edition

Jaccard P. 1901. Etude comparative de la distribution florale dans une portion des Alpes et des Jura. Bulletin de la Societe vaudoise des sciences naturelles 37: 547–579

Jambhulkar NN and Bose LK. 2014. Genetic variability and association of yield attributing traits with grain yield in upland rice. *Genetika* 46: 831-838

Janthasri S and Parinthawong N. 2020. Blast resistance evaluation and genetic inheritance of gene controlling leaf blast resistance in Dawk Pa-yawmRai variety (GS23774). *International Journal of Agricultural Technology* 16: 49-54

Jing Q, Spiertz JHJ, Hengsdijk H, Keulen HV, Cao W and Dai T. 2010. Adaptation and performance of rice genotypes in tropical and subtropical environments NJAS Wageningen. *Journal of Life Sciences* 57: 149-157

Johnson HW, Robinson HF and Comstock RE. 1955. Estimates of genetic and environmental variability in soybeans. *Agronomy Journal* 47: 314-318

Joshi RK and Behera L. 2006. Identification and differentiation of indigenous non-basmati aromatic rice genotypes of India using microsatellite markers. *African Journal of Biotechnology* 6: 348-354

Kadidaa B, Sadimantara GR and Safuan LO. 2017. Genetic diversity of local upland rice (*Oryza sativa* L.) genotypes based on agronomic traits and yield potential in marginal land of North Burton Indonesia. *Asian Journal of Crop Science* 9: 109-117

Kaushik and Santiaguel AF. 2014. Red Pearls of the Himalayas. *Rice Today* 13: 18-19

Khare R, Singh AK, Eram S and Singh PK. 2014. Genetic variability, association and diversity analysis in upland rice (*Oryza sativa* L.). *SAARC Journal of Agriculture* 12: 40-51

Kishore C, Kumar A, Pal AK, Kumar V, Prasad BD and Kumar A. 2018. Character association and path analysis for yield components in traditional rice (*Oryza sativa* L.) genotypes. *International Journal of Current Microbiology and Applied Sciences* 7: 283-291

Kujur MJ, Koutu GK, Krishnan RS and Singh Y. 2019. Genetic variability of agro-morphological traits in traditional varieties of rice (*Oryza sativa* L.) from Madhya Pradesh, India. *International Journal of Chemical Studies* 6: 1693-1700

Kumar V. 2015. Genetic diversity and character association studies for some economic traits in rice (*Oryza sativa* L.). *The Bioscan* 10: 899-904

Kumar V, Rastogi NK, Sarawgi AK, Chandraker P, Singh PK and Jena BK. 2016. Agro-morphological and quality characterization of indigenous and exotic aromatic rice (*Oryza sativa* L.) germplasm. *Journal of Applied and Natural Science* 8: 314-320

Kumar B. 2020. An appraisal of genetic variability and diversity among traditional red rice landraces. *International Journal of Ecology and Environmental Sciences* 2: 177-181

Kumaresan D and Manonmani S. 2019. Assessment of genetic variability for qualitative and quantitative traits in local rice cultivars of Gudalur Valley of the Nilgiris. *Indian Journal of Pure Applied Biosciences* 7: 237-245

Lakshmi VI, Gireesh C, Sreedhar M, Vanisri S, Basavaraj PS, Muralidhara B, Anantha MS, Padmavathi G, Fiyaz AR, Jyothi B and Suvarna C. 2018.

Characterization of African rice germplasm for morphological and yield attributing traits. *International Journal of Current Microbiology and Applied Sciences* 7: 1288-1303

Lush JL. 1949. Heritability of quantitative characters in farm animals. In: Proceedings International Congress Genetics Heriditas. P 356-357

Mahalanobis PC. 1936. On the generalized distance in statistics. *Proceedings National Academy of Science* 2: 49-55

Mau YS, Ndiwa AS, Markus JE, Oematan SS, Nasution A, Handoko DD and Makbul K. 2018. Blast resistance levels of red and black upland rice local cultivars from Indonesia. *Asian Journal of Crop Science* 10: 53-65

Mc Couch SR, Chen X, Panaud O, Temnykh S, Xu Y, Cho YG, Huang N, Ishii T and Blair M. 1997. Microsatellite marker development, mapping and applications in rice genetics and breeding. *Plant Molecular Biology* 35: 89–99

Mead R and Curnow RN. 1983. Statistical Methods in Agriculture and Experimental Biology, Chapman and Hall, New York p 488

Mohammmadi NG, Arzani A, Rezai AM, Singh RK and Gregerio GB. 2008. Assessment of rice genotypes for salt tolerance using microsatellite markers associated with the saltol QTL. *African Journal of Biotechnology* **7**:730-736

Murray MG and Thompson WF. 1980. Rapid isolation of high molecular weight plant DNA. *Nucleic Acids Research* 8: 4321-4325

Mustikarini ED, Lestari T, Prayoga GI, Santi R and Dewi S. 2020. Selection of red rice (*Oryza sativa* L.) resistant blast disease. In: IOP Conference Series: Earth and Environmental Science Vol. 599. P 012065

Nei M. 1973. Analysis of gene diversity in subdivided populations. Proceedings of the National Academy of Sciences of the United States of America 70: 3321-3323

Netravati M, Samal KC, Bastia DN and Rout GR. 2013. Genetic diversity analysis in aromatic rice genotypes using microsatellite based simple sequence repeats (SSR) marker. *African Journal of Biotechnology* 12: 4238–4250

Pachauri AK, Sarawgi AK, Bhandarkar S and Ojha GC. 2017. Agro-morphological characterization and morphological based genetic diversity analysis of rice (*Oryza sativa* L.) germplasm. *Journal of Pharmacognosy and Phytochemistry* 6: 75-80

Padmaja D, Radhika K, Rao LVS and Padma V. 2011. Correlation and path analysis in rice germplasm. *Oryza* 48: 69-72

Panse VG and Sukhatme PV. 1989. Statistical methods for agricultural workers. New Delhi: Indian Council of Agricultural Research

Parikh M, Motiramani NK, Rastogi NK and Sharma B. 2011. Characterization and assessment of variability in aromatic rice germplasm. *Programme of Agriculture* 11: 343-347

Parikh M, Motiramani NK, Rastogi NK and Sharma B. 2012. Agro-morphological characterization and assessment of variability in aromatic rice germplasm. *Bangladesh Journal of Agricultural Research* 37: 1-8

Pearson, K., 1901. Principal components analysis. *The London, Edinburgh and Dublin Philosophical Magazine and Journal of Science* 6: 559

Perrier X and Jacquemoud CJP. 2006. DARwin software

Pritchard JK, Stephens M and Donnelly P. 2000. Inference of population structure using multilocus genotype data. *Genetics* 155: 945-959

Puren H. 2017. Characterization of landraces, elite cultivars of red and brown rice and their derivatives for morphometric and nutrient parameters. Ph D Thesis, p 179. Department of Genetics and Plant Breeding, CSK Himachal Pradesh Krishi Vishwavidyalaya, Palampur, India

Rachappanavar V. 2017. Morphological and molecular characterization of traditional and improved germplasm of paddy cultivated in Himachal Pradesh. M.Sc. Thesis, p 172. Department of Genetics and Plant Breeding, CSK Himachal Pradesh Krishi Vishwavidyalaya, Palampur, India

Ramadan EA and Elmoghazy AM. 2015. Molecular markers based genetic diversity analysis for drought tolerance in rice using SSR markers. *International Journal of Scientific Research in Agricultural Sciences* 2: 137-146

Rao CR. 1952. Advance Statistical Methods in Biometrical Research. John Wiley and Sons Inc. New York Edinburgh

Rathore M, Singh R, Kumar B and Chauhan BS. 2016. Characterization of functional trait diversity among Indian cultivated and weedy rice populations. *Scientific Reports* 6: 1-9

Rohlf FJ. 1993. NTSYS-pc: Numerical Taxonomy and Multivariate Analysis System, version 1.80 Exeter Software. New York, USA

Rohlf. 1998. NTSYS-pc: Numerical Taxonomy and Multivariate Analysis System, version 2.02 User Guide. Applied Biostatistics Inc., Setauket, New York. pp 37

Salgotra, R.K., Gupta, B.B., Bhat, J.A. and Sharma, S., 2015. Genetic diversity and population structure of Basmati rice (*Oryza sativa* L.) germplasm collected from North Western Himalayas using trait linked SSR markers. *Public Library of Science One* 10
DOI: https://doi.org/10.1371/journal.pone.0131858

Sanghera GS, Kashyap SC and Parray GA. 2013. Genetic variation for grain yield and related traits in temperate red rice (*Oryza sativa L.*) ecotypes. *Notulae Scientia Biologicae* 5: 400-406

Satya, Rao P and LVS. 2022. Phenotypic assessment of rice landraces for genetic variability and diversity studies under heat stress. *Oryza* 59: 31-38

Semwal DP, Pandey A, Bhandari DC, Dhariwal OP and Sharma K. 2014. Variability study in seed morphology and uses of indigenous rice landraces (*Oryza sativa* L.) collected from West Bengal, India. *Australian Journal of Crop Science* 8: 460

Shahriar MH, Robin AHK, Begum SN and Hoque A. 2014a. Diversity analysis of some selected rice genotypes through SSR based molecular markers. *Journal of Bangladesh Agricultural University* 12: 307–311

Shahriar MH, Robin AHK and Hoque A. 2014b. Diversity assessment of yield, yield contributing traits and earliness of advanced T-aman rice (*Oryza sativa* L.) lines. *Journal of Bioscience and Agriculture Research* 1: 101-111

Sharma SK, Nandan R, Singh SK, Sharma AK, Kumar S, Sharma PK, Singh MK and Kumar V. 2011. Genetic divergence in rice (*Oryza sativa* L.) genotypes under irrigated condition. *Indian Journal of Agriculture* 11: 321-325

Singh BN, Dhua SR, Sahu RK, Patra BC and Marndi BC. 2001. Status of rice germplasm-Its collection and conservation in India. *Indian Journal of Plant Genetic Resource* 14: 105-106

Singh B, Singh N, Mishra S, Tripathi K, Singh BP, Rai V, Singh AK and Singh NK. 2018. Morphological and molecular data reveal three distinct populations of Indian wild rice *Oryza rufipogon* Griff. species complex. *Frontiers in Plant Science* 9: 123

Singh HP, Basandrai D, Sharma M and Basandrai AK. 2020. Evaluation of red rice (*Oryza sativa* L.) local germplasm collected from Himachal Pradesh. *Himachal Journal of Agricultural Research* 46: 22-28

Singh HP, Raigar OP and Chahota RK. 2021. Estimation of genetic diversity and its exploitation in plant breeding. *The Botanical Review*: 1-23

Sinha AK and Mishra PK. 2013. Agro-morphological characterization and morphology based genetic diversity analysis of landraces of rice variety (*Oryza sativa* L.) of Bankura district of West Bengal. *International Journal of Current Research* 5: 2764-2769

Sinha AK, Mallick GK and Mishra PK. 2015. Grain morphological diversity of traditional rice varieties (*Oryza sativa* L.), in lateritic region of West Bengal. *International Journal of Conservation Science* 6: 162-168

Sohrabi M, Rafii MY, Hanafi MM, Siti Nor Akmar A and Latif MA. 2012. Genetic diversity of upland rice germplasm in Malaysia based on quantitative traits. *The Scientific World Journal* 2012: 9

Sood R, Thakur S, Bhardwaj N and Kaushik RP. 2018. Genetic diversity analysis of rice landraces of NW Himalayas using RAPD and ISSR markers. International *Journal of Current Microbiology and Applied Sciences* 7
DOI: https://doi.org/10.20546/ijcmas.2018.704

Srivastava N, Babu GS, Singh ON, Verma R, Pathak SK, Behra M, Jena D and Chanda M. 2017. Genetic variation, heritability and diversity analysis of exotic upland rice (*Oryza sativa* L.) germplasms based on quantitative traits. *The Pharma Innovation Journal* 6: 316-320

Thomas J and Dominic V. 2016. Assessment of genetic diversity and relationship of coastal salt tolerant rice accessions of Kerala (South India) using microsatellite markers. *Journal of Breeding and Genetics* 47: 35-42

Thongbam PD, Durai AA, Singh TA, Taorem BD, Gupta S, Mitra J, Pattanayak A, Dhiman KR, Bhadana VP, Hore DK and Ngachan SV. 2010. Grain and food quality traits of some indigenous medicinal rice cultivars of Manipur, India. *International Journal of Food Properties* 13: 1244-1255

Thongbam PD, Raychaudhury M, Durai A, Das SP, Ramesh T, Ramya KT, Fiyaz RA and Ngachan SV. 2012. Studies on grain and food quality traits of some indigenous rice cultivars of North-Eastern hill region of India. *Journal of Agricultural Science* 4: 259

Tuhina-Khatun M, Hanafi MM, Rafii Yusop M, Wong MY, Salleh FM and Ferdous J. 2015. Genetic variation, heritability and diversity analysis of upland rice (*Oryza*

sativa L.) genotypes based on quantitative traits. *BioMed Research International* 2015: 7

Vasic N, Ivanovic M, Peternelli L, Dockovic J, Stojakovic M and Bocanski J. 2001. Genetic relationships between grain yield and yield components in a synthetic population and their implications in selection. *Acta Agronomica Hungarica* 49: 337-342

Venkatesan K and Bhat KV. 2015. Microsatellite marker-based molecular characterization of small and medium-grained aromatic rice germplasm of Odisha, India. *SABRAO Journal of Breeding and Genetics* 47: 248-259

Vennila S, Anbuselvam Y and Palaniraja K. 2011. D^2 analysis of rice germplasm for some quantitative and quality traits. *Electronic Journal of Plant Breeding* 2: 392-403

Vinita P, Nilay T, Prashant V, Nagendra KS and Sanjay S. 2013. Molecular and morphological characterization of Indian farmers rice varieties (*Oryza sativa* L.). *Australian Journal of Crop Sciences* 7: 923-932

Widyayanti S and Purwaningsih H. 2020. Genetic diversity of local red rice cultivars collections of Yogyakarta AIAT, Indonesia based on morphological character. In: IOP Conference Series: Earth and Environmental Science Vol. 482, p 012040

Wright S. 1921. Correlation and causation. *Journal of Agricultural Research* 20: 557-585

Wright S. 1921. The method of path coefficient. *Annals of Mathematical Statistics* 5: 160-169

Yadav MK, Aravindan S, Ngangkham U, Raghu S, Prabhukarthikeyan SR, Keerthana U, Marndi BC, Adak T, Munda S, Deshmukh R and Pramesh D. 2019. Blast resistance in Indian rice landraces: genetic dissection by gene specific markers. *Public Library of Science One* 14
DOI: https://doi.org/10.1371/journal.pone.0211061

Yadav S, Singh A, Singh MR, Goel N, Vinod KK, Mohapatra T and Singh AK. 2013. Assessment of genetic diversity in Indian rice germplasm (*Oryza sativa* L.) use of random versus trait-linked microsatellite markers. *Journal of Genetics* 92: 545–557

Yeh FC and Boyle TBJ. 1997. POPGENE Microsoft windows-based software for population genetic analysis. A joint project development by Francis C. Yeh. University of Alberta and Tim Boyle, Center for International Forestry Research, Bogor, Indonesia

Appendix 1: Mean performance of rice genotypes for grain yield and related traits

	Days to 50% flowering	Days to 75% maturity	Plant height (cm)	Flag leaf length (cm)	Flag leaf width (cm)	Total tillers per plant	Effective tillers per plant	Spikelet per panicle	1000 grain weight (g)	Grain length (mm)	Grain breath (mm)	L:B ratio	Biological yield per plant (g)	Harvest index (%)	Grain yield per plant (g)
IC-12180	90.00	113.00	142.47	31.80	1.99	10.00	8.00	64.00	16.83	4.37	2.10	2.08	41.80	27.93	11.63
Kaluna	91.00	116.00	140.53	32.77	1.67	6.00	6.00	66.00	18.10	3.49	2.16	1.61	34.62	26.77	9.26
Ramjuwain	90.00	114.00	123.73	28.80	0.95	8.00	8.00	41.00	18.61	4.56	1.96	2.33	38.92	33.69	13.12
Chohartu	86.00	113.00	140.41	45.43	2.03	6.00	5.00	41.00	18.72	3.80	2.22	1.71	32.20	32.06	10.31
Sukara	96.00	118.00	114.13	35.13	1.48	11.00	9.00	55.00	19.19	4.72	2.03	2.33	51.32	35.80	18.26
Karad	100.00	123.00	119.40	29.03	1.20	9.00	8.00	35.00	19.78	4.36	2.34	1.87	45.80	25.32	11.57
Kalaina	94.00	119.00	129.50	35.77	1.20	8.00	7.00	51.00	20.18	3.67	1.73	2.13	41.94	23.62	9.87
IC-12164	82.00	112.00	101.37	28.80	1.50	8.00	8.00	53.00	20.80	4.15	2.35	1.77	46.51	23.92	11.18
Acchoo	101.00	121.00	150.00	35.53	2.07	5.00	4.00	43.00	21.66	4.54	1.41	3.22	38.89	32.03	12.46
Begmi	86.00	109.00	123.50	34.50	1.30	8.00	8.00	54.00	23.02	4.23	1.35	3.13	39.30	29.14	11.33
Bathidhan	81.00	112.00	127.93	29.47	1.30	10.00	9.00	36.00	23.56	4.67	2.55	1.83	42.48	32.71	13.77
HPR-2906	73.00	109.00	146.50	36.33	1.60	8.00	8.00	41.00	24.58	4.34	1.70	2.56	43.13	26.62	11.34
Desidhan	101.00	123.00	140.30	62.53	1.40	12.00	11.00	57.00	20.39	4.72	2.11	2.23	62.25	39.29	24.51
Kalijhini-2	93.00	120.00	134.20	38.37	1.50	8.00	8.00	70.00	23.89	6.42	1.57	4.10	42.54	29.09	12.50
Sukara red	97.00	122.00	133.07	38.03	1.40	11.00	10.00	46.00	23.04	4.73	1.96	2.42	59.67	33.90	20.27
Varundhan	74.00	110.00	137.40	36.77	1.50	9.00	8.00	61.00	23.89	4.57	2.59	1.76	36.99	37.08	13.76
Hatiali	87.00	115.00	127.20	34.60	1.96	9.00	8.00	72.00	24.44	4.46	2.16	2.06	36.39	33.67	12.29
HPR-2800	92.00	116.00	121.47	34.80	1.63	7.00	7.00	54.00	22.18	3.61	1.49	2.42	33.83	28.08	9.49
Naggardhan	97.00	115.00	129.23	35.90	1.40	9.00	8.00	61.00	23.27	4.58	2.51	1.82	40.53	33.58	13.66
Roda dhan	87.00	116.00	130.27	32.07	1.43	7.00	6.00	71.00	24.48	3.46	2.98	1.16	33.21	26.95	8.91
Nailina	72.00	110.00	101.53	29.17	1.87	8.00	7.00	64.00	24.49	4.00	2.19	1.82	31.15	34.29	10.68
Deval	71.00	109.00	128.60	31.57	1.77	6.00	5.00	47.00	25.15	3.17	2.52	1.26	32.07	22.64	7.17
Bhrigudhan	85.00	108.00	132.87	29.56	1.57	6.00	6.00	49.00	24.52	3.50	1.85	1.89	33.97	28.14	9.44
Kalijhini	96.00	113.00	125.27	36.13	1.40	6.00	5.00	58.00	24.77	3.31	2.02	1.64	31.97	24.37	7.71
Matali	86.00	112.00	122.13	36.61	1.37	9.00	7.00	76.00	24.93	4.05	2.30	1.76	32.91	32.85	10.84
Dodadhan	74.00	110.00	133.67	31.82	1.40	8.00	8.00	64.00	24.81	4.15	3.13	1.32	32.41	34.19	11.08
HPR-2902	86.00	113.00	150.47	43.33	1.93	8.00	7.00	49.00	25.61	3.95	2.06	1.92	35.59	30.05	10.56
HPR-2904	87.00	116.00	142.40	40.47	1.70	7.00	7.00	64.00	25.63	3.63	2.25	1.61	31.40	30.61	9.56
HPR-2905	91.00	118.00	133.87	44.07	1.87	7.00	7.00	79.00	17.32	3.51	1.69	2.20	33.19	28.61	9.46

	Days to 50% flowering	Days to 75% maturity	Plant height (cm)	Flag leaf length (cm)	Flag leaf width (cm)	Total tillers per plant	Effective tillers per plant	Spikelet per panicle	1000 grain weight (g)	Grain length (mm)	Grain breath (mm)	L:B ratio	Biological yield per plant (g)	Harvest index (%)	Grain yield per plant (g)
HPR-2908	82.00	113.00	108.87	24.03	1.73	8.00	7.00	84.00	17.36	3.65	1.55	2.35	31.91	30.81	9.75
HPR-2913	90.00	116.00	147.73	48.03	1.30	12.00	11.00	87.00	26.38	6.40	2.21	2.89	50.48	46.39	23.30
HPR-2914	91.00	117.00	130.47	37.93	1.53	8.00	8.00	82.00	21.42	4.56	2.16	2.11	35.49	36.81	13.02
Gocha	85.00	113.00	126.20	29.39	1.23	9.00	8.00	65.00	25.68	4.19	2.22	1.90	35.99	31.81	11.32
New Chaina-21	82.00	114.00	111.53	36.96	1.61	10.00	9.00	78.00	26.87	4.70	2.46	1.91	39.19	37.08	14.42
Old Chaina-21	87.00	114.00	123.13	33.60	1.47	9.00	9.00	70.00	22.63	3.83	2.12	1.81	35.59	29.74	10.52
Phulpatas-21	83.00	113.00	124.93	35.32	1.50	11.00	9.00	66.00	26.95	6.26	1.78	3.52	41.18	39.92	16.35
Karad 21-1	98.00	119.00	116.23	34.50	1.20	6.00	5.00	77.00	27.53	3.38	2.49	1.36	33.21	23.52	7.71
Karad 21-2	92.00	119.00	122.20	34.60	1.43	9.00	8.00	75.00	27.63	4.51	2.00	2.25	38.42	31.05	11.82
Karad 21-3	102.00	118.00	127.60	34.07	1.89	8.00	7.00	45.00	19.34	3.73	2.36	1.58	36.27	27.57	9.93
Karad 21-4	93.00	119.00	130.73	40.67	1.43	8.00	7.00	41.00	27.94	3.81	2.34	1.63	35.28	29.67	10.41
Jattu	85.00	109.00	120.00	44.80	1.10	9.00	7.00	39.00	25.19	4.05	2.23	1.82	34.93	31.25	10.79
HPR-2720 (C)	**98.00**	**121.00**	**132.67**	**40.20**	**1.63**	**7.00**	**7.00**	**64.00**	**32.53**	**3.43**	**1.63**	**2.12**	**45.80**	**41.47**	**18.83**
HPR-2795 (C)	**96.00**	**120.00**	**150.67**	**40.49**	**1.83**	**8.00**	**8.00**	**90.00**	**35.15**	**6.46**	**2.02**	**3.21**	**55.63**	**38.20**	**21.29**
Grand mean	**88.60**	**115.12**	**129.22**	**36.13**	**1.54**	**8.28**	**7.51**	**60.12**	**23.50**	**4.27**	**2.11**	**2.10**	**39.22**	**31.45**	**12.45**
Range lowest	71.00	108.00	101.37	24.03	0.95	5.00	4.00	35.00	16.83	3.17	1.35	1.16	31.15	22.64	7.17
Range highest	102.00	123.00	150.67	62.53	2.07	12.00	11.00	90.00	35.15	6.46	3.13	4.10 v	2.25	46.39	24.51
CD (5%)	**2.81**	**3.01**	**10.69**	**5.97**	**0.38**	**0.72**	**0.67**	**2.26**	**1.53**	**0.26**	**0.18**	**0.25**	**7.31**	**6.37**	**2.80**
CV (%)	**1.95**	**1.61**	**5.08**	**10.16**	**15.16**	**5.38**	**5.46**	**2.32**	**3.99**	**3.77**	**5.23**	**7.33**	**11.47**	**12.45**	**13.80**
Best check	**HPR-2795**	**HPR-2795**	**HPR-2720**	**HPR-2795**	**HPR-2795**	**HPR-2795**	**HPR-2795**	**HPR-2795**	**HPR-2795**	**HPR-2795**	**HPR-2720**	**HPR-2795**	**HPR-2795**	**HPR-2720**	**HPR-2795**

Brief Biodata of student

Name : Rishita Kapoor

Mother's Name : Mrs. Neelam Kapoor

Father's Name : Mr. Vijay Kapoor

Date of Birth : 1st August, 1998

Permanent Address: V.P.O. Gopalpur, Teh. Sarkaghat, Distt. Mandi (H.P.)-175007

Academic Qualification

Examination passed	Year	School/Board/ University	Marks (%)	Division	Major Subjects
10th	2013	HPBOSE Dharamshala	89.6	First	English, Mathematics, Hindi, Social science, Science, Sanskrit, Information Technology
12th	2015	HPBOSE Dharamshala	87.4	First	English, Biology, Physics, Chemistry, Computer Science
B.Sc. (hons.) (Agriculture)	2020	CSK HPKV Palampur	80.70	First	All Agriculture and Allied subjects
M.Sc. Agri. (Genetics and Plant Breeding)	2022	CSK HPKV Palampur	87.1	First	Major Discipline: Genetics and Plant Breeding Minor Discipline: Plant Pathology

Scholarships/Gold Medals/Any Other Distinction

2016-2020	College Merit Scholarship, B.Sc.(Hons) Agriculture
2020-2022	College Merit Scholarship, M.Sc. Agriculture
2022	First Prize in Inter-University Declamation Competition on National Mathematics Day, Second Prize in Inter-University Power Point Presentation Competition on National Science Day, Second Prize in Inter-University Tug of war Competition
Others	Participated in zonal students elocution XV agriculture science congress (October, 2021). Participation certificate for **zonal agrivision convention** at Jammu (April, 2022), virtual training program on QTL analysis and genome-wide association studies (Feb, 2022) and Training certificate for **Mission SAHASI** during Oct-Nov 2018 at Dharamshala