AN ANALYSIS OF GENETIC ASSOCIATION BETWEEN YIELD AND QUALITY TRAITS IN SEGREGATING POPULATION OF ORYZA SATIVA L.

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Abstract. Limited information has been reported on the genetic association of morphological and quality traits of Oryza sativa L. From a total of 50 genotypes, evaluated for genetic diversity using SSR markers, six diverse genotypes were selected for the generation of three crosses. Progenies of these crosses were grown to develop three F₂ populations. Morphological and quality traits were recorded and analysed for genotypic and phenotypic correlations followed by the path coefficient analysis from the genotypic correlation. Strongest association was observed for yield per plant and alkali spread value (ranging from -0.73 to -0.91 for different crosses). It was depicted from path coefficient analysis that length width ratio had a maximum direct effect on yield (ranging from 0.61 to 0.67 for different crosses), and also had a relatively high magnitude of the indirect effect to the yield via plant height, panicle length protein content and length width ratio. Although, minute differences were detected in the strength of association and direct/indirect effects for different populations, the direction of the association always remained the same. The present study showed that direct and indirect effects of traits should be considered for the selection of yield improvement instead of correlation, as a trait having high correlation may have a low direct effect and vice versa.

Keywords: Oryza sativa L., correlation, path coefficient analysis, qualitative traits, basmati rice

Introduction

Rice is the staple food for about half of the world population. Rice is a healthful cereal grain which owns unusual dietary requirements. In 2018-19 worlds’ rice production is expected to be 510 million tonnes while world’s consumption is also expected to be 503 million tonnes (FAO, 2018). Asia produced and consumed more than 90% of the world’s rice (Memon et al., 2015) as three billion Asians fulfil their 35-60% of caloric requirement through rice (Guyer et al., 1998). The cooking and eating qualities of rice are important in defining its economic value in export market and for user acceptance (Pingali, 1997). The Quality of rice grain is intricate characteristic comprising several components such as appearance, cooking and eating qualities (Rabiei et al., 2004). Physicochemical properties of rice are calculated on the basis of amylase content, protein contents and alkali spread value (Rohilla et al., 2000). Quality and yield of rice grain can be enhanced by understanding the genetic mechanism.
controlling these traits and association between these traits which is not very well elaborated in previous studies. Correlation studies reported in past have been conducted in homozygous populations but in this study we have conducted correlation studies in segregating populations.

Complex inheritance pattern of grain yield and quality traits necessitate their thorough understanding in order to strengthen rice breeding program. Studies of genetic correlation between yield and quality traits gave understanding about the extent and direction of association between these traits. However direct and indirect effect of different yield related traits are determined by path coefficient analysis. Genetic attributes could be ranked according to their contribution by the breeder (Dewey and Lu, 1959). Dewey and Lu (1959) developed a statistical technique for path analysis which helps in understanding the cause of association and provide information about cause and effect situation. Path analysis assists plant breeders in making selection as it dissects the correlation of different traits into direct and indirect effects (Dewey and Lu, 1959; Milligan et al., 1990; Samonte et al., 2005).

The main objective of this study was to (i) assess genetic variability and correlation among yield and quality traits of various segregating populations for selecting superior genotypes, (ii) to assess the direct and indirect contribution of yield components to yield, (iii) to assess the type of connection between quality and yield traits of rice.

Materials and methods

The present study was conducted in the experimental area of Department of Plant Breeding and Genetics, University College of Agriculture and Environmental Sciences, the Islamia University of Bahawalpur, Pakistan.

Development of plant material for genetic diversity studies

Thirty rice varieties were grown in nursery during stat of May 2013 up to three leaf stage. Of each genotype 4 cm leaf sample was collected and preserved in liquid nitrogen prior to DNA extraction by CTAB method (Ul-Allah et al., 2017). Twenty µl PCR reaction mixture included DNA 2 µl, 0.2 µl Taq polymerase enzyme, 1.5 µl of each primer, 2.5 µl of 2.5 mM dNTPs, 3 µl MgCl$_2$ and 2 µl PCR buffer along with 9.3 µl of ddH$_2$O. One hundred SSR markers (Table 1) were used for diversity studies. PCR product was studied on 1.5% agarose gel with Ethydium bromide as fluorescent dye. Banding pattern was scored in binary fashion for the calculation of similarity index which was deployed for the construction of dendrogram by using un-weighted pair group method with arithmetic mean (UPGMA).

Selection of diverse genotypes and development of populations

Based on dendrogram generated by UPGMA, six genetically diverse genotypes i.e., Basmati Pak, Basmati-198, Basmati-385, Super Basmati, Basmati-2000 and Shaheen Basmati were grown in field in end of May, 2013. Cross combinations were made among genotypes widely assorted in dendrogram in following fashion (Basmati Pak × HuoYou8166, Basmati-385 × HuoYou8305 and Basmati-198 × HuoJingXian) to generate F$_1$ seed. F$_2$ seed harvested from F$_1$ generation during September 2014 was sown in field during May 2015 adapting nursery sowing followed by field transplantation maintaining plant to plant and row to row distance of 30 cm. Soil was
sandy-loam with 50:30:20 sand, silt and clay proportion. Ten rows (each having fifteen plants) of each segregating population plus five rows of each parent were grown keeping single plant per hill in non-replicated fashion. All agronomic practices and crop protection measures were observed as per standard recommendations. Weeds, insect pests and diseases were controlled following standard recommendations.

Table 1. List of primers used in present study

| Locus name | Chr | Forward primer sequence | Reverse primer sequence | Reference |
|------------|-----|-------------------------|--------------------------|-----------|
| RM1        | 1   | GCGAAAACACAATGCAAAAAAA | GCGTTGGTTGACACTGAC       | Panand et al., 1996 |
| RM2        | 7   | ACATGTCGACCTGCAACTG    | ATGTCCGGGAGTTGCTACG      | Panand et al., 1996 |
| RM3        | 6   | ACATTGACCGGCACTGAG     | GCCTCAGCCTCCACATCTTT     | Panand et al., 1996 |
| RM7        | 3   | TCTGTCGACCTGCAACTG     | TCCTCCATATTTCGTTTG       | Panand et al., 1996 |
| RM8        | 2   | CAGTGGCTGAAAAATACG     | GGCAAAACCTAACTCCTT       | Panand et al., 1996 |
| RM10       | 7   | TGGTTAAGGTTATGACAG     | CAGAATGGAGAAGTTGCTC      | Panand et al., 1996 |
| RM11       | 7   | TCTCCTTCTCCCGGATC      | ATAGCGGCGAGGGTTGAG       | Panand et al., 1996 |
| RM13       | 5   | TCCAAATGCGGCAAGAGAG    | GTGTTGGATGATTCACAG       | Panand et al., 1996 |
| RM16       | 3   | CAGTGCAGAGCACATATA     | AACACAGGAGTCGCAGC        | Panand et al., 1996 |
| RM17       | 12  | TGCCCTGTATTITCCGTCCT   | GGTGATCGCTTTCCCACTTC     | Panand et al., 1996 |
| RM18       | 7   | TTCCTCTACTGAGCTCTC     | GAGTCGCTGGCAGCTGAG       | Panand et al., 1996 |
| RM19       | 12  | CAAAACAGGAGACATGAG     | CTCAAGATGAGCAGCAAGA      | Panand et al., 1996 |
| RM21       | 11  | ACAGATTTCTCGGAGACAG    | GCTTCATGAGGAGGTTGAG      | Panand et al., 1996 |
| RM22       | 3   | GTGTTTGGAGGCCTTTACATTC | CTGTTGGCTTTCTTGCGT       | Panand et al., 1996 |
| RM26       | 5   | GAGTCGGACGAGCGAGA      | CTGCGAGGACGCAACTACA      | Chen et al., 1997 |
| RM27       | 2   | TTTCCTTCTCACCACCATCA   | TCTTTGACAGAGGGAAAGAGC    | Chen et al., 1997 |
| RM30       | 6   | GGTAGGACATGCTGACAG     | TCACCTACACAGACAGCAGC     | Chen et al., 1997 |
| RM31       | 5   | GATCAGGATATCGAGCTGAG   | AAGTCAGATACGCTCCTC       | Chen et al., 1997 |
| RM36       | 3   | CAACATGACACATGCGC      | GTACCTACAGACAGTGACC      | Chen et al., 1997 |
| RM38       | 8   | AGAGATCTCGGATCAGCAG    | TCAGGTTTATCCTCAGGAC       | Chen et al., 1997 |
| RM39       | 5   | GCCTTCCTGTCCTTCCTTC   | AATTCGAAGTGGTTGAGC       | Chen et al., 1997 |
| RM41       | 9    | AAGTCTGAGTTTGGCTCC    | AATTTCTAGGCTGAGG         | Chen et al., 1997 |
| RM42       | 8    | ATCTCAGGGATGACATGAG    | TTTGGATAGTTGAGGTTGAGC    | Chen et al., 1997 |
| RM44       | 4    | ACGGGGAGATCGGACAAAC    | TGCGGAAAAACCTACACCTC     | Chen et al., 1997 |
| RM47       | 7    | ACTCTCAGCTTCCATCCCA    | GTCGACGAGGTTGACATG       | Chen et al., 1997 |
| RM49       | 3    | TCCGAAGATTTGTTAAGGATCA | TTTGGACAGGATGAGG         | Chen et al., 1997 |
| RM50       | 6    | ACTGTACCGGCTGCAAGAG    | AAATTCAGATGAGGCTGAGC     | Chen et al., 1997 |
| RM55       | 7    | TCTCGTTCATGTCGTTCG     | ATCAGTACGATAGTTGCTC      | Chen et al., 1997 |
| RM56       | 3    | CCCTGCGGATGAGTAGAGAAG  | TCCGGTTTTTTATTAGG        | Chen et al., 1997 |
| RM60       | 3    | ATCTCAGTTCAGTGCTCC     | ATGTTGAGTGAGCTGATAC      | Chen et al., 1997 |
| RM70       | 7    | GTGAGCTTTCTTGTAGCTC    | GATGTAAAGATGATGCGC       | Chen et al., 1997 |
| RM72       | 2    | CTAGGAGCGGAAAAACAGAGTG | GGGTTGAGGAGGTTAATTAGT    | Temnykh et al., 2000 |
| RM78       | 8    | CCGGGGATAAAACCATGAG    | GCTTCTATCTACAAATAGG      | Temnykh et al., 2000 |
| RM80       | 8    | TTGAAGGCGGTGAAAGAG     | CATCAACCTGCTCTTACCGC     | Chen et al., 1997 |
| RM82       | 7    | TGCTTCTTGTGATTTGCGC    | GCAGTGCTTGGAGATTAGC      | Chen et al., 1997 |
| RM83       | 12   | ACTGCGATGCAGGTTAGG     | CACCTAGAGAGCTGAGG        | Chen et al., 1997 |
| RM85       | 3    | CCAAGATGAAACCTGTAGG    | GCACAAGTGGAGCAGG         | Temnykh et al., 2000 |
| RM87       | 5    | CTCCTCGGATACAGCCTGAG  | GCGAAGTGAGAAGAAGAGAAG    | Temnykh et al., 2000 |
| RM88       | 8    | ACTTACGATGGTTGTGCTTC  | TAAATTGCAGCCTCCACTCC     | Temnykh et al., 2000 |
| RM201      | 9    | CTGTTTTTATTATTCCATGAC  | ATACTTTTATCTCAGGAT       | Chen et al., 1997 |
| RM202      | 11   | CAGATTGAGAGTGAATGCTC   | CAGAACAGGACGAGAGG         | Chen et al., 1997 |
| RM204      | 6    | GTGACGTTCGGTTGCTAGG    | GTATGACGCTGCTGCTAGC      | Chen et al., 1997 |
| RM205      | 9    | CTGTTGTCGAGGAGGAGGAG  | CTTGCAGTCCCTGGAGG         | Chen et al., 1997 |
| RM206      | 11   | CCACTGTCCTGAAAACTTCTCT | CTTGTCATGCTCTGAGG        | Chen et al., 1997 |
| RM207      | 2    | CCATGTTGGAGAGATGCTCA   | CACCTCACTCGTAAAGGCCC     | Chen et al., 1997 |
| RM209      | 11   | ATATGAGTTGCTGGCTGCG    | CAACAGTCTGCTCCTCCT       | Chen et al., 1997 |
| Locus name | Chr | Forward primer sequence | Reverse primer sequence | Reference |
|------------|-----|-------------------------|-------------------------|-----------|
| RM210      | 8   | TCACATTCGGTGGCATTG      | CGAGATGGTGTGTCATTG      | Chen et al., 1997 |
| RM214      | 7   | CAGATGATAGAAACCTCTCTCTC| AAGAACGTGACACTCAAA      | Chen et al., 1997 |
| RM215      | 9   | CAAATGAGGAGCAGCAAGAGAC| TGAGCACTTCCTCCTGTTAG    | Chen et al., 1997 |
| RM216      | 10  | GCATGCGGATGGTAAAG       | TGATAAAACACCACAGGCA     | Chen et al., 1997 |
| RM217      | 6   | ATCGACCAATGCTCCTC       | GGGTGTTGAAACAGGAC       | Chen et al., 1997 |
| RM219      | 9   | CTTCGAGATGTTAAGGCTC    | CATATGCGGATCCTGCTG      | Chen et al., 1997 |
| RM222      | 10  | CTTAAATGCGGACATGCGG    | CAAAGCTTGCAGGCAAAGAG    | Chen et al., 1997 |
| RM223      | 8   | GATGTGAGCTGGCTGAAAC    | GAAAGCAAGCTGCTGCAGTC    | Chen et al., 1997 |
| RM224      | 11  | ATCTGATGATTCACAGGAG    | TGCTTAAAGGAGCTCGG       | Chen et al., 1997 |
| RM225      | 6   | TGCCCTATATGCTGAGATG    | GAAAGTGACCTAGGAGGAG     | Chen et al., 1997 |
| RM228      | 10  | TGTCGCTATTAGCTCCTG     | GCTTGGCGCCCTCCTGACT     | Chen et al., 1997 |
| RM229      | 11  | CACTTCACAGCAAGACTGAC   | CGACGCTTCTGTTGAATTG     | Chen et al., 1997 |
| RM230      | 8   | GGCAAGCGCTTGAGTTGTC    | CACCGAGTACCTTCTCAAG     | Chen et al., 1997 |
| RM234      | 7   | ACAGATATACGAGATGCTG    | CAGCTGGAACAGACAGGAG     | Chen et al., 1997 |
| RM235      | 12  | GACAGCTCATGCGACAGAG    | TCACCTGTGACCTCCTTC      | Chen et al., 1997 |
| RM239      | 10  | TACAAAAATGCGGAGTCAC    | CATATGCGGACACCTGTC      | Chen et al., 1997 |
| RM241      | 4   | GACGCCAAATAAGATCTCTGA  | TGCAAGACGAGATTATAGT     | Chen et al., 1997 |
| RM242      | 9   | GGCACAATGCTGGTATGTC    | TATATGCAAGAGCTGAGG      | Chen et al., 1997 |
| RM244      | 10  | CCGACTGTTGCTCTTATCA    | CTGCTTGGCGGAGATCAG      | Chen et al., 1997 |
| RM245      | 9   | ATGCAGCAGGATAAGATG     | CAGCTGGAACAGACAGGAG     | Chen et al., 1997 |
| RM246      | 1   | GAGCTTCATGACGACATTC    | CTGAGTCTGTGCGGACT       | Chen et al., 1997 |
| RM247      | 12  | TAGTGGCGTGATGTAAGC     | CATATGCGGAGAAGACAG      | Chen et al., 1997 |
| RM248      | 7   | TCTCTGTTAAACTCTGCTC    | GTAGCTTGCAGTCTGAGT      | Chen et al., 1997 |
| RM250      | 2   | GTTTCAAAACCAAGCTGATC   | GATGAAAGGCTCTCAAGCAG    | Chen et al., 1997 |
| RM253      | 6   | TCTCTCATGACGACATTC     | GCATTTGCTGACAGAGGAG     | Chen et al., 1997 |
| RM254      | 11  | AGCCCGGATAAATACCTCAC  | CTGAGGAGGATTTGATAGC     | Chen et al., 1997 |
| RM255      | 4   | GTGTGCGTGAGGGAGATGGT   | CGAAGACGCTGATCCTCAG     | Chen et al., 1997 |
| RM256      | 8   | GACAGGAGGATGGAAGAGCC   | GTTGTAGATGTCGACAGGAG    | Chen et al., 1997 |
| RM257      | 9   | CAGTCCCGCAAGCAGAGATCC | GAGTCCAGGCTGAGTACTG     | Chen et al., 1997 |
| RM258      | 10  | TGCTGTATGATGCTCAGC     | TGCGCTTTAAGGCTGTCG      | Chen et al., 1997 |
| RM259      | 1   | TGAGACTTAGGAGAGAGG     | CGTGTGACTGTCGACTG       | Chen et al., 1997 |
| RM260      | 11  | ACTTCACTATGACGCCAGAG   | GCAAACTCTTTCTACGTCAG    | Chen et al., 1997 |
| RM261      | 4   | CTATTCTTCTCTTGGTGC     | TGACATCGCGAATCTCC       | Chen et al., 1997 |
| RM262      | 2   | CATTCGCTCGCCTCAACT     | CAGACAAGGTTGCTTCG       | Chen et al., 1997 |
| RM263      | 2   | CCCAGGCTAGCTCAAGAC     | GCTACGTGTTAGCTACAG      | Chen et al., 1997 |
| RM264      | 8   | GTTCTGACCTCTGCTCCCTC   | GATCCGCTGATGACAGT       | Chen et al., 1997 |
| RM266      | 2   | TATGTATACCAAGACCTCT   | GTTGAAGCCAAATCTGCA      | Chen et al., 1997 |
| RM267      | 5   | TGCAAGCAGATAGAGAAAGAGTG| AGCAAGACGACACTTGAATG    | Chen et al., 1997 |
| RM269      | 10  | GAAAGGAGATAGGACAGAG    | GCAAAATGCGCTGAGTCTG     | Chen et al., 1997 |
| RM270      | 12  | GGCCGTGGTTCTAAATCAC   | TGCCGCTATACAGTGCGAG     | Chen et al., 1997 |
| RM271      | 10  | TCAGATCTCAATCTCCTC    | TCGGAGATGCTCAGAGGAGG    | Chen et al., 1997 |
| RM272      | 1   | AATTGCTGAGAGGGAGAGG    | CATGCAGCTGAGTCAGGAG     | Chen et al., 1997 |
| RM273      | 4   | GAAGCCGTGCGTAAGTACCC  | GTTTCTACATGACAGGAGC     | Chen et al., 1997 |
| RM274      | 5   | CCTCGCTTATGAGGAGCTG    | CTCTCTACATCCTACATGC     | Chen et al., 1997 |
| RM275      | 6   | GCAATGATGTCGACACTG    | CATTCGCAACTCTCATTACACT | Chen et al., 1997 |
| RM276      | 6   | CTTACAGCTTGACACCTCCTG | TCTCTCATACTGACAGTATC    | Chen et al., 1997 |
| RM278      | 9   | GTATTGACCTAACAATAATC  | TCAACTGACGACTCCTGTCG    | Chen et al., 1997 |
| RM279      | 2   | GGGGAGAGGAGGTCTTCTCTC | GGGTTGATTAAACTCTCGC     | Chen et al., 1997 |
| RM280      | 4   | ACAAGCTCCTCTTTGCGC    | TGGTCTGGACAGCAGGAG      | Chen et al., 1997 |
| RM281      | 8   | ACAAAGCTACCAGTGACAGACG| GTTCTCTACATCCTACATGC    | Chen et al., 1997 |
| RM282      | 3   | CGTGTGCGGAAAGGCTGCG   | CAGTCTGCTGAGTTCAGGAG    | Chen et al., 1997 |
| RM283      | 1   | GTCTCAGATGCTACCTCTG    | CGGCTAGAGATGCTTGTAG     | Chen et al., 1997 |
| RM284      | 8   | ATCTCTGATACCTCATTCC   | CCTTGACGTTGACAGAGAGC    | Chen et al., 1997 |
| RM285      | 9   | CTTGCGGCCAATAATGTCA   | GCGCGGAGCATGAGGAAAGG    | Chen et al., 1997 |
Morphological and quality traits evaluation

At maturity data was recorded on one hundred fifty randomly selected guarded plants from each population for plant height, panicle length, and yield per plant. Seed samples were collected separately for the estimation of Amylose content, Alkali Spread Value, protein content and grain length/width ratio. Chemical analysis of Protein contents were accomplished according to AACC method 2000.

Correlation analysis

Correlation coefficient among the characters under study was estimated according to the statistical techniques given by Gomez and Gomez (1984).

\[ r = \frac{\sum XY - \sum X \cdot \sum Y / n}{\sqrt{[\sum X^2 - (\sum X)^2 / n][\sum Y^2 - (\sum Y)^2 / n]}} \]

where n is number of observations, X is first variable, Y is second variable, while genotypic correlation was calculated according to formula

\[ r = \frac{CoVF_2 - CoVP}{\sqrt{[VF_2(X) - V_P(X)][VF_2 - CoVP(Y)]}} \]

Path analysis

Path coefficient analysis was performed according to the method given by Dewey and Lu (1959) for the yield and quality traits of Rice, keeping yield per plant as resultant variable and rest of the traits as causal variables.

Results

Genetic diversity studies

Thirty rice genotypes having wide range of phenotypic variability (Table 2) for all of the traits under study were subjected to Simple sequence repeat (SSR) based genetic diversity study (Fig. 1). Cluster analysis revealed the genetic relatedness among 30 genotypes of rice as shown in the dendrogram (Fig. 2). One hundred SSR markers equally distributed on twelve chromosomes were randomly selected from the genetic map of Rice which revealed polymorphism percentage of 11.3 to 77.5%. Distribution of these genotypes in the dendrogram shows the genetic variation among the genotypes. Cluster analysis grouped the genotypes into four clusters with reference to yield and yield related traits having smallest cluster (Cluster I) with two varieties, Huo Jing Xian and Huoyou8187 and the largest cluster (Cluster IV) having nine varieties, while second and third clusters possesses eight varieties each. Huojing xian, Huoyou-8305, Basmati-Pak, Basmati-198 and Basmati-385 on the bases of genetic divergence owing to variation in yield were selected for making three cross combinations.
Correlation analysis

Genetically diverse F2 population (Fig. 3) for all of the traits studied was obtained from above mentioned cross combinations. Populations from all three cross combinations are significantly different from each other for plant height, panicle length, yield per plant, alkali spread value, amylase content, L/W ratio and protein content. All the characters studied were significantly correlated with yield per plant with few exceptions (discussed below). Genotypic and phenotypic correlation coefficients of three cross combinations populations for various traits are presented in Table 3. Highest positive phenotypic correlation for plant height was observed with protein content values 0.61 in Basmati-198 × HuoJingXian, while it was minimum with L/W ratio 0.01 in the same cross. Phenotypic correlation of plant height was lowest with Alkali spread value -0.48 while genotypic correlation between plant height and amylase content was highest having numerical value -0.71. Highest positive phenotypic correlation for panicle length was with Yield per plant 0.73 while genotypic correlation between panicle length and ASV was -0.64. Panicle length show minimum genotypic correlation with protein content and L/W ratio having numerical values 0.04 and -0.05 respectively. Yield per plant being the most important character of plant showed strongest phenotypic and genotypic correlation with ASV -0.91 while the genotypic and phenotypic correlation between YPP vs panicle length 0.14 and YPP vs Plant height -0.54 respectively, were minimum. Genotypic correlation of ASV was strongest with YPP -0.91, but phenotypic correlation of ASV was weakest with panicle length -0.31. Alkali Spread Value showed strongest positive phenotypic correlation with amylase content 0.87. Length Width ratio show strongest positive phenotypic correlation with protein content 0.86 while strongest negative phenotypic correlation of L/W ratio was found with amyllose content -0.89. Contrary to this weakest genotypic correlation of L/W ratio was observed with panicle length -0.05. Protein content shows highest positive phenotypic correlation with L/W ratio i.e. 0.72 and strongest negative genotypic correlation with ASV -0.91. Strong negative (-0.73) phenotypic correlation was observed between protein content and ASV while weakest positive (0.04) genotypic correlation was found between protein content and panicle length.

Table 2. Mean values of various traits of parent, studied in this experiment

| S. No | VAR/TR     | PH     | PL     | YPP    | ASV     | Aamyl | L/W ratio | Protein |
|-------|------------|--------|--------|--------|---------|-------|-----------|---------|
| 1     | BASMATIPAK | 132.98 | 22.68  | 14.04a | 5.59b   | 25.67cd| 4.63cd    | 7.79cd  |
| 2     | M133       | 139.07 | 21.86a | 14.71a | 5.33ab  | 25.63cd| 4.65cd    | 7.75def |
| 3     | KASHMIRBAS | 136.63a| 21.97ab| 14.08ab| 5.51ab  | 25.79cd| 4.67cd    | 7.75def |
| 4     | BASMATI198 | 136.53a| 23.18ac| 14.23ab| 5.55b   | 25.67cd| 4.93f     | 7.83efg |
| 5     | BASMATI385 | 137.60a| 23.22a | 14.99ab| 5.66bc  | 25.06bc| 4.93f     | 7.68cd  |
| 6     | KS282      | 140.84a| 21.97ab| 14.63ac| 5.37ab  | 25.10bc| 4.87ef     | 7.90f   |
| 7     | NIABIRR19  | 128.03b| 22.68bc| 14.65ac| 5.42ab  | 25.26bc| 4.97efg    | 7.94f   |
| 8     | PAK23717   | 127.36a| 21.74ab| 12.12abc| 5.43a  | 24.88bc| 4.94ef     | 7.64ef  |
| 9     | IR36       | 116.89abc| 21.02a | 11.58abc| 6.25c | 26.44d | 4.74c     | 7.25ab  |
| 10    | MB385      | 116.48abc| 23.17abc| 10.71acd| 5.87cd | 27.05de| 4.67cd    | 7.21ab  |
| 11    | MB198      | 117.48abc| 22.71bc| 13.50aef| 5.27ab | 27.48e | 4.76d     | 7.19ab  |
| 12    | BAS2000    | 115.75abc| 23.47bc| 13.61ad | 5.27ab | 25.39bc| 4.57c     | 7.18ab  |
| 13    | M153       | 115.46abc| 24.27cd| 13.70abcd| 5.48ab| 26.38cd| 4.84d     | 7.15a   |
Ghouri et al.: An analysis of genetic association between yield and quality traits in segregating population of *Oryza sativa* L. - 5217 -

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| S. No | VAR/TR   | PH     | PL   | YPP      | ASV     | Aamyl | L/W ratio | Protein |
|-------|----------|--------|------|----------|---------|-------|-----------|---------|
| 14    | N185     | 115.36 | 22.36 | 13.65    | 6.74    | 26.49 | 4.26      | 7.13    |
| 15    | LH18     | 115.36 | 22.58 | 13.50    | 6.32    | 26.47 | 4.38      | 7.10    |
| 16    | 521      | 115.94 | 23.46 | 13.45    | 6.38    | 24.55 | 4.27      | 7.08    |
| 17    | A1       | 113.14 | 22.73 | 13.20    | 5.27    | 24.49 | 4.39      | 7.43    |
| 18    | SUPPERBAS| 113.18 | 23.46 | 13.10    | 5.95    | 25.63 | 4.71      | 7.24    |
| 19    | X625     | 113.27 | 24.24 | 12.74    | 5.27    | 25.48 | 4.75      | 7.29    |
| 20    | MK134    | 111.28 | 23.66 | 12.64    | 6.46    | 23.47 | 4.57      | 7.35    |
| 21    | SWAT1    | 110.27 | 22.68 | 12.70    | 6.25    | 23.84 | 4.26      | 7.36    |
| 22    | SHADAB   | 109.27 | 23.74 | 11.74    | 7.46    | 24.57 | 4.27      | 7.25    |
| 23    | SARSHAR  | 108.13 | 22.47 | 10.72    | 5.47    | 24.65 | 4.37      | 7.28    |
| 24    | KSK103   | 108.17 | 23.58 | 10.64    | 5.61    | 23.48 | 4.28      | 7.55    |
| 25    | X625     | 108.34 | 23.58 | 10.54    | 5.82    | 24.59 | 4.76      | 7.45    |
| 26    | BOYOU566 | 107.48 | 24.57 | 9.78     | 5.96    | 24.65 | 4.56      | 7.39    |
| 27    | BOYOU58305| 107.47| 22.16 | 9.54     | 6.57    | 25.58 | 4.36      | 7.85    |
| 28    | HUOYOU8305| 106.37| 23.28 | 8.49     | 5.68    | 23.52 | 4.57      | 7.73    |
| 29    | HUOYOU8187| 106.48| 24.27 | 8.43     | 4.90    | 24.48 | 4.95      | 7.63    |
| 30    | HJK      | 105.37 | 23.57 | 7.21     | 5.27    | 23.44 | 4.37      | 7.46    |

PH: plant height, PL: penical length, YPP: yield per plant, ASV: Alkali spread value, L/W ratio: length width ratio, Protein: protein content. *a,b,c,d* values with similar alphabets are statistically same

**Figure 1.** Gel electrophoresis image of RM-229 with rice genotypes in present study
Figure 2. UPGMA cluster analysis showing the diversity and relatedness among the 30 rice genotypes studied

Figure 3. Cumulative range of variability in F₂ population of all three crosses for various traits in present study
Table 3. Phenotypic and genotypic correlation among different yield and quality traits of rice in three different F2 populations

| Variables | Basmati Pak × HuoYou8166 | Basmati-385 × HuoYou8305 | Basmati-198 × HuoYou8305 |
|-----------|---------------------------|---------------------------|---------------------------|
|           | PH  | PL  | YPP | ASV  | Aamyl | L/W Ratio | Protein |
| PH        | 1.00 | 0.12 | -0.70** | -0.69** | -0.71** | 0.05 | 0.49* |
| PL        | 0.19 | 1.00 | 0.16 | -0.35 | -0.27 | -0.26 | 0.05 |
| YPP       | -0.68 | 0.67** | 1.00 | -0.91** | -0.86** | 0.56** | -0.11 |
| ASV       | -0.67 ** | -0.31 | -0.87** | 1.00 | 0.75** | -0.83** | -0.85** |
| Aamyl     | -0.66** | -0.23 | -0.82** | 0.92** | 1.00 | -0.57** | -0.89** |
| L/W Ratio | 0.09 | -0.21 | 0.82* | -0.79** | -0.50* | 1.00 | 0.62** |
| Protein   | 0.56** | 0.15 | -0.15 | -0.83** | -0.84** | 0.86** | 1.00 |

| Variables | Basmati-385 × HuoYou8305 | Basmati-198 × HuoYou8305 |
|-----------|---------------------------|---------------------------|
|           | PH  | PL  | YPP | ASV  | Aamyl | L/W Ratio | Protein |
| PH        | 0.42* | 1.00 | 0.14 | -0.64** | -0.58** | -0.05 | 0.04 |
| PL        | -0.56** | 0.53** | 1.00 | -0.83** | -0.72** | 0.73** | -0.15 |
| YPP       | -0.59 ** | -0.43* | -0.79** | 1.00 | 0.79** | -0.69** | -0.91** |
| ASV       | -0.54* | -0.25 | -0.73** | 0.87** | 1.00 | -0.85** | -0.87** |
| Aamyl     | -0.54* | 0.24 | 0.81** | -0.67** | -0.82** | 1.00 | 0.68** |
| L/W Ratio | 0.10 | 0.13 | -0.17 | -0.86** | -0.83** | 0.81** | 1.00 |
| Protein   | 0.59** | 0.15 | -0.17 | -0.86** | -0.83** | 0.81** | 1.00 |

Lower diagonal represents phenotypic correlation and upper diagonal represent genotypic correlation. Bold numerical values are diagonals. PH: plant height, PL: panicle length, YPP: yield per plant, ASV: Alkali spread value, L/W ratio: length width ratio, Protein: protein content. *Significant at the 5% level. **Significant at the 1% level

Path co-efficient analysis

Genetic correlation of different quality traits and yield components was further dissected to investigate the direct and indirect effects of each trait that it has on grain yield (Table 4). Direct effects explain the contribution of individual trait with grain yield while indirect effect explains the influence of a character on yield in association of other characters.

Direct effects

Direct effect is the contribution of the trait that directly determined the yield. The traits that have positive direct effect, with increase in them the yield will also increases. While increase in the characters having negative effect toward yield will results in decrease of yield. In the present population the results show that all the traits show positive direct effect in the yield except ASV. Highest positive direct effect was showed by L/W ratio (0.67) followed by panicle length (0.41), amyllose content (0.36) while lowest positive direct effect was contributed by plant height (0.22). Alkali spread value show negative direct effect with maximum value (-0.49) and minimum value (-0.21).
Table 4. Direct and indirect effect matrix. Dependent variable is YPP. The last column shows genotypic correlations of independent variables with YPP among different yield and quality traits of rice in three different F2 populations

| Variables       | PH   | PL   | ASV   | Aamyl | L/W ratio | Protein | YPP   |
|-----------------|------|------|-------|-------|-----------|---------|-------|
| Basmati Pak ×   |
| HuoYou8166     |      |      |       |       |           |         |       |
| PH              | 0.19 | 0.07 | -0.33 | -0.31 | -0.03     | -0.29   | -0.70 |
| PL              | 0.04 | 0.32 | -0.18 | 0.01  | 0.04      | -0.07   | 0.16  |
| ASV             | 0.12 | -0.27| -0.49 | 0.19  | 0.28      | -0.74   | -0.91 |
| Aamyl           | 0.10 | 0.07 | -0.82 | 0.33  | -0.73     | 0.19    | -0.86 |
| L/W ratio       | -0.01| -0.03| -0.34 | 0.56  | 0.61      | -0.23   | 0.56  |
| Protein         | -0.02| 0.02 | -0.18 | 0.32  | -0.15     | -0.1    | -0.11 |
| Basmati-385 ×   |
| HuoYou8305      |      |      |       |       |           |         |       |
| PH              | 0.22 | 0.09 | -0.27 | -0.31 | -0.02     | -0.32   | -0.61 |
| PL              | 0.04 | 0.41 | -0.24 | 0.02  | 0.05      | -0.05   | 0.14  |
| ASV             | 0.11 | -0.15| -0.21 | 0.51  | -0.57     | -0.52   | -0.83 |
| Aamyl           | 0.13 | 0.06 | -0.73 | 0.32  | -0.68     | 0.22    | -0.72 |
| L/W ratio       | 0.03 | -0.04| -0.32 | 0.73  | 0.67      | -0.35   | 0.73  |
| Protein         | 0.06 | 0.08 | -0.15 | 0.21  | -0.22     | -0.13   | -0.15 |
| Basmati-198 ×   |
| HuoJingXian     |      |      |       |       |           |         |       |
| PH              | 0.20 | 0.08 | -0.35 | -0.28 | -0.04     | -0.26   | -0.61 |
| PL              | 0.03 | 0.36 | -0.20 | 0.02  | 0.06      | -0.04   | 0.23  |
| ASV             | 0.18 | -0.56| -0.44 | 0.27  | 0.36      | -0.63   | -0.91 |
| Aamyl           | 0.09 | 0.05 | -0.78 | 0.36  | -0.84     | 0.27    | -0.85 |
| L/W ratio       | -0.03| -0.04| -0.43 | 0.57  | 0.65      | -0.24   | 0.48  |
| Protein         | -0.01| 0.05 | -0.12 | 0.26  | -0.14     | -0.16   | -0.12 |

PH: plant height, PL: penical length, YPP: yield per plant, ASV: Alkali spread value, L/W ratio: length width ratio, Protein: protein contents

Indirect effects

Plant height has positive indirect effect through panicle length which ranges 0.07-0.09. Negative indirect effect of plant height was present for all other traits which was minimum (-0.02) for L/W ratio while negative indirect effect was maximum (-0.35) for ASV. Panicle length has positive indirect effect with plant height, amylose content and L/W ratio which was highest for L/W ratio having value 0.06 while panicle length has minimum indirect positive effect with amylose content (0.01). Negative indirect effect of panicle length observed with ASV and protein content, being maximum for ASV (-0.24) and minimum for protein content (-0.04). Negative indirect effect of ASV was resulted with panicle length ranges (-0.15 to -0.56) and protein content ranges (-0.52 to -0.72). ASV showed positive indirect effect with plant height, amylose content and L/W ratio which is highest for L/W ratio (0.57) and lowest for (0.11). Amylose content has all the positive indirect effects except ASV and L/W ratio. Direct positive effect of amylose content was maximum through protein content (0.27) and was minimum through panicle length (0.05). Negative indirect effect was highest through L/W ratio (-0.84) while minimum was also through L/W ratio (-0.68). L/W ratio has all the negative indirect effect except through amylose content, the maximum negative indirect effect contributed through ASV (-0.43) while minimum through plant height (-0.01). Positive indirect effect of protein content presented through panicle length and amylose content.
which ranges (0.02-0.32). Negative indirect effect of Protein content ranges (-0.02 to -0.22) that are contributed through plant height, ASV and L/W ratio.

Discussion

The study was conducted to add in the existing knowledge of relationship between yield and quality traits. For the improvement of complex quantitative traits, the knowledge of inter relationship for yield contributing and quality traits are important for a breeder to evolve high yielding cultivars with improved quality traits. Negative correlation of plant height with paddy yield show higher photosynthetic accumulation in the vegetative parts due to tallness rather then their accumulation in the reproductive parts (Zahid et al., 2006; Akinwale et al., 2011). Furthermore, dwarf genes have also been reported to have pleiotropic effect on yield (Zahid et al., 2006; Parsad et al., 2001; Tahir et al., 1988). However, the results were contradictory to Ranawake and Amarasinghe (2014) who reported that increase in plant height results in increased rate of photosynthesis in plant which transfer more photosynthates to grains. The contrary results may be attributed to genetic differences and environmental factors. Negative correlation between panicle length and plant height was also observed by Jayasudha and Sharma (2010) and Eradasappa et al. (2007), this may be as a result of compromise between reproductive and vegetative growth. Increase amylose content of grain will increase grain yield in rice. Positive correlation between amylose content and protein shows that increase in one character will lead to increase the in others traits and vice versa Anandan et al. (2011). For the improvement of yield direct selection through plant height will result in the reduction of grain yield per plant due its negative direct effect (Naseem et al., 2014; Hairmais et al., 2010; Akhtar et al., 2011). Panicle length has positive direct effect on yield so increase in panicle length will enhance yield. All the genetic coefficients of correlation were less than the phenotypic coefficient of correlation in present investigation which indicates the effect of environment in the observed correlation. Panicle length was positively correlated with grain yield which showed that improvement of this character will led to improved varieties with higher yield. Plant height and quality traits like alkali spread value, amylose content and protein content decrease with the increase of yield so higher yielding varieties will produce lower amylose, ASV content and protein contents (Sarkar et al., 2007; Sabesan et al., 2009). Protein contents have negative correlation with grain yield, but this correlation was negative, therefore, selection for higher yield may not have significant impact on protein contents. Here, it is important to notice that amylose content and plant height have negative correlation with grain yield of rice but when its correlation is divided into direct and indirect effects, direct effect of these traits was in positive direction. Similar findings have also been shown in cotton case of seeds per locule and lint percentage (Ul-Allah et al., 2017). In such conditions, selection become complicated and indirectly effecting traits must be considered for an effective selection. It is concluded that plant height and panicle length are the key yield influencing traits, while protein content and amylose content are the basic quality traits which also influence yield.

This reflect that with the increase in panicle length, number of spikelets may increase that result in increase of paddy yield (Ashfaq et al., 2012; Ramkrishnan et al., 2006). Increase amylose content of grain will increase grain yield in rice. Positive correlation
between amylose content and protein shows that increase in one character will leads to increase the others (Anandan et al., 2011).

Path coefficient analysis simplify the complex multiple effect of different traits on resultant traits (Ul-Allah et al., 2017) as it breaks the correlation into direct and indirect effects. Main purpose of the analysis is to identify that how different traits effect each other and modify the effect of other trait to the final yield. Plant height has positive direct effect on yield per plant as with the increase in plant height, plant produce photosynthates but due to accumulation of photosynthetic assimilate in vegetative parts, magnitude of its direct effect is low and indirect effect of plant height through various other traits lead to overall negative correlation. Plant height has indirect effect with panicle length toward the increment of yield as due to increase of plant height, panicle length also increases, which leads to higher yield. Similar correlation far panicle and plant height was identified by Jayasudha and Sharma (2010) and Eradasappa et al. (2007). Amylose content, ASV, length to width ratio and protein content have overall negative correlation with yield per plant as more photosynthates will accumulates in the vegetative parts and does not moves towards sink (Tahir et al., 1988; Zahid et al., 2006). Similar result was reported by Parsad et al. (2001). More the length of panicle more grain will produce which leads to increased grain yield (Immanue et al., 2011).

The negative direct effect of alkali spread value shows that it directly decreases the yield and also indirectly along with L/B ratio decreases the yield. Length to width ratio has positive direct effect on the yield while overall correlation with grain yield was positive, which means that with the increase in L/B ratio grain yield will increase. Nandan et al. (2010) suggested that the difference was due to different genotypes and environmental conditions. Protein content has positive direct effect on the yield and negative indirect effect via plant height, ASV and L/W ratio. While combined effect of protein with all other traits on the yield is positive.

Conclusion

It is concluded from this study that traits having positive correlation with yield may have negative direct effect and vice versa. Therefore, selection of traits for yield improvement should be made carefully by looking at direct effect of specific trait to yield as one trait with smaller direct effect may have higher correlation due to higher indirect effect of other traits. Information created from this research will help plant breeder for effective selection of traits in developing high yielding rice cultivars. High yield rice cultivars will contribute to sustainable profit of the farmers by getting higher yield without any extra inputs.

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