

Association and Path Coefficient Study in F₂ Population for Yield Attributing and Micronutrient Traits in Rice (*Oryza sativa* L.)

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Authors' contributions

This work was carried out in collaboration among all authors. Author AK designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Authors AK, NKS, RK, SKS, Nilanjaya, MKS and AT managed the analyses of the study. Author Banshidhar managed the literature searches. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/CJAST/2020/v39i1830794

Editor(s):

(1) Dr. Tushar Ranjan, Bihar Agricultural University, India.

(2) Dr. Md. Hossain Ali, Bangladesh Institute of Nuclear Agriculture (BINA), Bangladesh Agricultural University Campus, Bangladesh.

Reviewers:

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(2) Truong Ngoc Minh, Vietnam Academy of Science and Technology, Vietnam.

Complete Peer review History: <http://www.sdiarticle4.com/review-history/59300>

Original Research Article

Received 04 May 2020
Accepted 10 July 2020
Published 13 July 2020

ABSTRACT

In view to overcome the major problem of 'hidden hunger' mainly caused by micronutrient deficiency, breeding for micronutrient enriched staple food crops is important. In developing countries, iron and zinc deficiencies are reported to be major health risk factor causing a high mortality rate. So, for overcoming these nutritional deficiencies through genetic improvement, F₂ population of two rice crosses obtained by crossing diverse parents for micronutrients (mainly, Fe and Zn) were evaluated using randomised complete block design during *Kharif*, 2019 to study the relationship between different traits and to study the estimates of direct and indirect effect. Among the F₂ population of cross-I, grain yield per plant exhibited significant and positive association with seeds per panicle, tillers per plant, flag leaf area, harvest index, test weight, days to 50% flowering and days to maturity while negative and significant correlation with canopy temperature. For cross-

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II, grain yield per plant exhibited significant and positive association with seeds per panicle, tillers per plant, plant height, flag leaf area, SPAD value, harvest index, test weight and days to maturity while significant and negative correlation with grain Zn content, grain Fe content and canopy temperature. Hence, selection for the traits showing positive and significant association with grain yield in both the crosses will be rewarding. Highest positive direct effect on yield was shown by harvest index in cross-I. So, selection based on harvest index for grain yield per plant would be the most effective strategy for improvement of grain yield. No direct positive or negative effect of grain Fe and Zn content on yield was found.

Keywords: Correlation coefficient; path analysis; micronutrient; direct effect; indirect effect.

1. INTRODUCTION

Rice (*Oryza* spp.) is the staple food for more than half of the world's population, contributing over 20% of the total calorie intake of humans [1]. The leading producers of this cereal are China, India, and Indonesia which together account for over 50% of the world's total production [2]. Average daily intake of rice provides 20-80 per cent of dietary energy and 12-17 per cent dietary proteins for Asians [3]. The global production of rice is nearly 487.46 million tonnes over 161.10 million hectare area with the productivity of 3.02 tonnes per hectare (Statista, 2017-18). In India, rice is grown over an estimated area of 44.50 million hectares with the production of 110.15 million tonnes with its productivity being 3.90 tonnes per hectare [4]. It accounts about 43% of total food grain and 55% of cereals production in country. In Bihar, the total area under rice cultivation is nearly 3.23 million hectares with production of 6.80 million tonnes and its productivity is 2.10 tonnes per hectare [5].

Rice contains low iron (Fe) and zinc (Zn) levels, most of which is lost during grain processing like dehusking and polishing of grains. Populations with monotonous diets mainly consuming cereals are especially more prone to Fe and Zn deficiency, which affects around two billion peoples globally. Among cereals, rice is a suitable target for biofortification because anemia, caused by Fe-deficiency is a serious problem in developing countries where rice is a major staple crop. Nearly 50% of the Indian soils are deficient in Zn levels, particularly in those areas where rice-wheat system is followed and it affects 50% of rice, particularly grown under lowland conditions. Brown rice is rich in mineral value. However, rice is mostly consumed in the polished form which contains low mineral levels. In fact, the Fe content of endosperm is significantly lower than in other rice tissue, including leaves, stems, roots, husk, or the aleurone layer. 0.5% of total Fe content in aerial

parts of plant body was found in polished seeds (endosperm) of Tsukinohikari rice variety, despite 3.6% in brown seeds, 3.4% in husk, 2.4% in rachis and 90.6% in straw, respectively [6]. These Fe and Zn deficiencies can be addressed in several ways viz., nutritional diversification, food enrichment and biofortification. Supplementation or food fortification programs have not always been successful earlier because these are not cost effective. Crop Fe fertilization is also not very effective due to Fe soil insolubility. An alternative solution is Fe and Zn biofortification by developing cultivars that efficiently utilize, mobilize, uptake and translocate Fe and Zn to the edible parts like grains. While classical breeding approaches are effective in modifying the contents of inhibitors present in plant for Fe and Zn absorption, in recent past transgenic breeding have mainly focused to enhance Fe and Zn and its accumulation in grain. Thus, these breeding approaches of biofortification can save the life of millions of people in Asia, particularly in India and can solve the major problem of hidden hunger associated with micronutrient deficiency. The present article is an attempt to analyze the association of grain Fe and grain Zn content in rice with other morpho-physiological traits affecting grain yield along with direct and indirect effect of grain Fe and Zn content on grain yield. So that effective selection can be practiced for improving grain Fe and Zn content along with high yield.

Correlation coefficient is a statistical measure which is used to find out the nature, degree and direction of relationship between two or more variables or association between various characters including yield. This analysis measures the mutual relationship between various plant characters and determines the component characters on which selection can be based for improvement of economically important characters. But, selection based on correlation without considering the various types of the

interactions taking place between the component characters affecting yield may sometimes mislead. Moreover, it does not give any idea about the exact position of the relative importance of direct and indirect effect of the various component traits on yield. This extent of relative contribution of a particular character to any dependent component can be judged from path coefficient analysis. In path analysis, the correlation coefficient between two traits is separated into the components which measure the direct and indirect effects. Path coefficient is the component that measures the direct effects of a predictor variable upon its response variable, the second component being the indirect effects of a predictor variable on the response variable through another predictor variable [7]. It provides an exact idea about the relative importance of direct and indirect effects of each of the component traits contributing to yield.

Information on association of characters, direct and indirect effects contributed by each character towards yield will help breeders to formulate the effective criteria in selecting desirable genotypes in early segregating populations. In view of this, the present investigation was carried out with the objective to determine the association and direct and indirect effect of various morpho-physiological traits along with grain Fe and Zn content on yield by using F_2 populations of two crosses in rice.

2. MATERIALS AND METHODS

Two F_2 populations was generated from F_1 crosses of four diverse parents for micronutrient content which were selected and procured from Harvest Plus Rice Project at Department of Plant Breeding and Genetics, RPCAU, Pusa, Samastipur, Bihar. The parents used for crossing were TEVIRII, MTU 1010, KHUSISOI-RI-SAREKU and IR91175-27-1-3-1-3. In present study TEVIRII and KHUSISOI-RI-SAREKU were used as female parent having high grain Fe and Zn content, while MTU 1010 and IR91175-27-1-3-1-3 were used as male parent having low grain Fe and Zn content. These two F_2 population were evaluated for 14 traits including yield, yield contributing and micronutrient traits during *Kharif* 2019 in randomized complete block design with 3 replications for association and path coefficient studies. Data were recorded on individual plant basis on seventy plants per replication for all the traits. The estimation of micronutrients from brown rice [8] by XRF (X-Ray Fluorescence Spectrometry) was carried out at

HarvestPlus Division, ICRISAT, Hyderabad. Simple correlation coefficients were computed between pair of characters adopting the formula given by Al-Jibouri et al. [9] as well as Panse and Sukhatme [10]. Path coefficient was obtained according to the procedure as suggested by Dewey and Lu [7].

3. RESULTS AND DISCUSSION

Grain yield is a complex character, so for effective selection of this quantitative trait a detailed knowledge of strength and direction of association between grain yield, its attributing traits and quality traits like grain Iron and Zinc content is very important. A sound knowledge of this relationship among these traits can be further be exploited for selection of suitable breeding scheme. Present study of correlation for Cross-I (TEVIRII \times MTU 1010) revealed that the grain yield per plant was significantly and positively associated with seeds per panicle (0.216**), tillers per plant (0.767**), flag leaf area (0.352**), harvest index (0.915**), test weight (0.531**), days to 50% flowering (0.832**) and days to maturity (0.693**) while negatively and significantly correlated with canopy temperature (-0.839**). Thus, selection for yield contributing traits like, seeds per panicle, number of productive tillers per plant, flag leaf area, harvest index, test weight, days to 50% flowering and days to maturity will be highly effective in increasing the grain yield. The grain Zn content has positive and significant association with grain Fe content (0.567**). This significant and positive association between grain Zn and grain Fe may be due to the involvement of some common transporters which may be helping in accumulation of both Zn and Fe in the grain. The grain Fe content exhibited negative and significant relationship with seeds per panicle (-0.146*) and Plant height (-0.224**). Seed per plant has positive and significant association with tiller per plant (0.156*), harvest index (0.243**), days to 50% flowering (0.151*) and days to maturity (0.177**). Similarly, tiller per plant showed positive and significant association with flag leaf area (0.307*), harvest index (0.749**), test weight (0.308**), days to 50% flowering (0.748*) and days to maturity (0.660**). Flag leaf area showed positive and significant association with harvest index (0.364**), test weight (0.185**), days to 50% flowering (0.387*) and days to maturity (0.367**). Harvest index had positive and significant association with test weight (0.503**), days to 50% flowering (0.827*) and days to maturity (0.732**). Test weight

assumed significant and positive association with days to 50% flowering (0.471**) and days to maturity (0.397**). Days to 50% flowering exhibited positive and significant correlation with days to maturity (0.693**). These traits showed positive and significant association with grain yield per plant, thus, these traits can be used for indirect selection for yield. However, seed per plant showed negative and significant association with test weight (-0.352**), thus, selection for more seed per plant increases the yield but the seeds become less bold and lower in weight. Days to maturity showed positive and significant association with harvest index and test weight, thus it can be interpreted that late maturing lines have bolder seeds and higher economic yield.

For cross-II (KHUSISOI-RI-SAREKU × IR91175-27-1-3-1-3), study of correlation coefficient indicated that the grain yield per plant exhibited significant and positive association with seeds per panicle (0.452**), tillers per plant (0.584**), plant height (0.192**), flag leaf area (0.516**), SPAD value (0.668**), harvest index (0.316**), test weight (0.400**) and days to maturity (0.202**) while, significant and negative correlation with grain Zn content (-0.136*), grain Fe content (-0.179*) and canopy temperature (0.456**). So, selection considering seeds per panicle, tillers per plant, plant height, flag leaf area, SPAD value, harvest index, test weight and days to maturity will be effective in increasing grain yield. Grain zinc content have significant and positive correlation with grain Fe content (0.158**) and Days to 50% flowering (0.171*) while, significant and negative association with plant height (-0.207**). The grain Fe content have exhibited significant and negative correlation with SPAD value (-0.155*). Seeds per panicle had positive significant association with tillers per plant (0.156*), harvest index (0.243*), days to 50% flowering (0.151*) and days to maturity (0.177*). Tiller per plant has positive and significant association with flag leaf area (0.307**), harvest index (0.749*), test weight (0.308**), days to 50% flowering (0.151*) and days to maturity (0.177*). Flag leaf area has positive and significant correlation with harvest index (0.503**), test weight (0.185**), days to 50% flowering (0.387**) and days to maturity (0.367**). Harvest Index showed significant and positive correlation with test weight (0.503**), days to 50% flowering (0.827**) and days to maturity (0.732**). Test weight had significant and positive association with days to 50% flowering (0.471**) and days to maturity

(0.397**). Days to 50% flowering have positive and significant association with days to maturity (0.881**). These traits showed significant and positive correlation with yield, thus, these traits can be used for indirect selection to get higher yield. However, seeds per panicle has significant and negative correlation with canopy temperature (-0.262**) and test weight (-0.352**); tillers per plant has significant and negative correlation with canopy temperature (-0.707**); canopy temperature has significant and negative association with flag leaf area (-0.270**), harvest index (-0.842**), test weight (-0.427**), days to 50% flowering (-0.771**) and days to maturity (-0.693**). Similar findings were obtained by researchers [11,12,13,14].

Though, by the study of correlation coefficients we get information on various aspects of inter-relationship among different traits, these study do not reveal any information on cause and effect of association between two traits. Path coefficient analysis allows separation of the direct effect and their indirect effects through other attributes by partitioning the correlations [15]. Hence, the path coefficient analysis [7] was done to know the direct and indirect effects of yield components on yield. Path analysis helps us to know the stable associations among the traits for use in future selections by plant breeders [16]. In the present study, grain yield per plant was considered as the dependent variable and phenotypic correlation of this character with other traits were partitioned into their corresponding direct and indirect effects through path coefficient analysis. The study indicated that for F₂ population of cross-I (TEVIRII X MTU 1010) only harvest index have exhibited high positive direct effect on grain yield per plant. The indirect effects among the other traits indicated that tillers per plant (0.3642), days to 50% flowering (0.4022) and days to maturity (0.3557) have high positive indirect effect *via* harvest index on grain yield per plant. Therefore, for the improvement of grain yield, indirect selection would be recommended along with these traits. Canopy temperature (-0.4095) have high negative indirect effect *via* harvest index on grain yield per plant. For the F₂ population of cross-II (KHUSISOI-RI-SAREKU X IR91175-27-1-3-1-3), SPAD value (0.3975) exhibited high direct effect on yield. Number of tillers per plant (0.2094) and canopy temperature (-0.2046) have expressed moderate positive and negative direct effect, respectively. None of the other traits have exhibited high indirect effect *via* other component traits on grain yield per plant.

Table 1. Phenotypic correlation coefficient in F₂ population of cross-I (TEVIRII × MTU 1010) for different traits

	Zn content	Fe content	SPP	TPP	PH	PL	CT	FLA	SPAD	HI	TW	DFP	DM	GYPP
Zn content	1.000													
Fe CONTENT	0.567**	1.000												
SPP	-0.033	-0.146*	1.000											
TPP	-0.108	-0.034	0.156*	1.000										
PH	-0.120	-0.224**	0.045	-0.029	1.000									
PL	0.008	0.014	-0.044	-0.041	0.117	1.000								
CT	0.069	0.096	-0.262**	-0.707**	0.007	-0.008	1.000							
FLA	0.050	-0.006	0.112	0.307**	-0.025	-0.061	-0.270**	1.000						
SPAD	-0.012	-0.060	0.118	0.105	0.066	-0.069	-0.101	-0.014	1.000					
HI	-0.067	-0.113	0.243**	0.749**	0.038	-0.001	-0.842**	0.364**	0.112	1.000				
TW	0.070	-0.049	-0.352**	0.308**	-0.050	0.014	-0.427**	0.185**	0.010	0.503**	1.000			
DFP	-0.026	0.007	0.151*	0.748**	0.006	-0.016	-0.771**	0.387**	0.186**	0.827**	0.471**	1.000		
DM	0.002	0.027	0.177**	0.660**	0.009	-0.013	-0.693**	0.367**	0.160*	0.732**	0.397**	0.881**	1.000	
GYPP	-0.069	-0.097	0.216**	0.767**	-0.006	-0.030	-0.839**	0.352**	0.133	0.915**	0.531**	0.832**	0.693**	1.000

Table 2. Phenotypic correlation coefficient in F₂ population of cross-II (KHUSISOI-RI-SAREKU × IR91175-27-1-3-1-3) for different traits

	Zn content	Fe content	SPP	TPP	PH	PL	CT	FLA	SPAD	HI	TW	DFP	DM	GYPP
Zn content	1.000													
Fe CONTENT	0.158**	1.000												
SPP	-0.045	-0.070	1.000											
TPP	0.001	-0.130	0.274**	1.000										
PH	-0.207**	-0.087	0.128	0.100	1.000									
PL	-0.092	0.079	0.002	0.052	0.017	1.000								
CT	-0.019	-0.017	-0.129	-0.268**	0.161*	-0.020	1.000							
FLA	-0.047	-0.117	0.315**	0.343**	-0.008	-0.027	-0.269**	1.000						
SPAD	-0.052	-0.155*	0.293**	0.534**	0.057	0.021	-0.335**	0.376**	1.000					
HI	-0.069	-0.016	0.097	0.1679*	0.043	0.027	-0.200**	0.222**	0.256**	1.000				
TW	-0.022	0.018	0.279**	0.206**	0.118	0.052	-0.265**	0.215**	0.251**	0.077	1.000			
DFP	0.171*	-0.011	0.026	0.047	-0.106	-0.012	-0.123	0.050	0.096	0.069	0.030	1.000		
DM	-0.083	-0.012	0.005	0.022	0.002	0.057	-0.059	0.025	0.095	0.147*	0.089	0.018	1.000	
GYPP	-0.136*	-0.179**	0.452**	0.584**	0.192**	-0.006	-0.456**	0.516**	0.668**	0.316**	0.400**	0.068	0.202**	1.000

Table 3. Path coefficient analysis of different traits for cross-I (TEVIRII × MTU 1010)

	Zn content	Fe content	SPP	TPP	PH	PL	CT	FLA	SPAD	HI	TW	DFF	DM
Zn content	-0.0151	-0.0085	0.0005	0.0016	0.0018	-0.0001	-0.0010	-0.0008	0.0002	0.0010	-0.0011	0.0004	0.0000
Fe CONTENT	0.0030	0.0053	-0.0008	-0.0002	-0.0012	0.0001	0.0005	0.0000	-0.0003	-0.0006	-0.0003	0.0000	0.0001
SPP	-0.0024	-0.0105	0.0714	0.0112	0.0032	-0.0031	-0.0187	0.0080	0.0084	0.0174	-0.0252	0.0108	0.0127
TPP	-0.0153	-0.0049	0.0221	0.1415	-0.0040	-0.0057	-0.1001	0.0434	0.0149	0.1061	0.0436	0.1059	0.0935
PH	0.0018	0.0033	-0.0007	0.0004	-0.0148	-0.0017	-0.0001	0.0004	-0.0010	-0.0006	0.0007	-0.0001	-0.0001
PL	-0.0001	-0.0003	0.0008	0.0007	-0.0022	-0.0185	0.0001	0.0011	0.0013	0.0000	-0.0003	0.0003	0.0002
CT	-0.0108	-0.0151	0.0412	0.1111	-0.0011	0.0013	-0.1571	0.0425	0.0159	0.1324	0.0671	0.1211	0.1090
FLA	0.0007	-0.0001	0.0016	0.0044	-0.0004	-0.0009	-0.0038	0.0142	-0.0002	0.0052	0.0026	0.0055	0.0052
SPAD	-0.0002	-0.0009	0.0017	0.0016	0.0010	-0.0010	-0.0015	-0.0002	0.0148	0.0017	0.0001	0.0027	0.0024
HI	-0.0327	-0.0549	0.1183	0.3642	0.0185	-0.0003	-0.4095	0.1770	0.0546	0.4858	0.2447	0.4022	0.3557
TW	0.0098	-0.0068	-0.0493	0.0431	-0.0070	0.0020	-0.0597	0.0259	0.0014	0.0704	0.1397	0.0658	0.0555
DFF	-0.0077	0.0020	0.0445	0.2198	0.0017	-0.0048	-0.2265	0.1137	0.0546	0.2431	0.1385	0.2937	0.2588
DM	-0.0004	-0.0054	-0.0354	-0.1317	-0.0017	0.0026	0.1383	-0.0733	-0.0320	-0.1460	-0.0793	-0.1757	-0.1994
GYPP	-0.0694	-0.0968	0.2160	0.7676	-0.0062	-0.0302	-0.8390	0.3520	0.1326	0.9159	0.5311	0.8327	0.6937

R SQUARE = 0.8893, RESIDUAL EFFECT = 0.3327

Table 4. Path coefficient analysis of different traits for cross-II (KHUSISOI-RI-SAREKU × IR91175-27-1-3-1-3)

	Zn content	Fe content	SPP	TPP	PH	PL	CT	FLA	SPAD	HI	TW	DFF	DM
Zn content	-0.0591	-0.0094	0.0026	0.0000	0.0122	0.0055	0.0011	0.0028	0.0030	0.0041	0.0013	-0.0102	0.0049
Fe CONTENT	-0.0084	-0.0530	0.0037	0.0069	0.0046	-0.0042	0.0009	0.0062	0.0082	0.0008	-0.0010	0.0006	0.0006
SPP	-0.0073	-0.0113	0.1632	0.0448	0.0208	0.0003	-0.0211	0.0515	0.0480	0.0159	0.0457	0.0042	0.0008
TPP	0.0001	-0.0273	0.0575	0.2094	0.0209	0.0108	-0.0562	0.0719	0.0120	0.0352	0.0432	0.0098	0.0046
PH	-0.0279	-0.0118	0.0172	0.0135	0.1349	0.0022	0.0217	-0.0011	0.0077	0.0058	0.0160	-0.0143	0.0002
PL	0.0038	-0.0033	-0.0001	-0.0021	-0.0007	-0.0414	0.0008	0.0011	-0.0009	-0.0011	-0.0021	0.0005	-0.0024
CT	0.0038	0.0034	0.0264	0.0549	-0.0329	0.0041	-0.2046	0.0552	0.0686	0.0411	0.0544	0.0251	0.0121
FLA	-0.0080	-0.0201	0.0543	0.0591	-0.0014	-0.0046	-0.0464	0.1721	0.0648	0.0383	0.0371	0.0086	0.0043
SPAD	-0.0153	-0.0461	0.0874	0.1590	0.0170	0.0064	-0.0997	0.1120	0.3975	0.0762	0.0749	0.0286	0.0282
HI	-0.0049	-0.0011	0.0070	0.0121	0.0031	0.0019	-0.0144	0.0160	0.0184	0.0718	0.0055	0.0049	0.0106
TW	-0.0025	0.0021	0.0317	0.0233	0.0134	0.0058	-0.0301	0.0244	0.0285	0.0087	0.1131	0.0034	0.0101
DFF	0.0008	-0.0001	0.0001	0.0002	-0.0005	-0.0001	-0.0006	0.0002	0.0005	0.0003	0.0001	0.0049	0.0001
DM	-0.0106	-0.0015	0.0007	0.0028	0.0002	0.0073	-0.0075	0.0032	0.0121	0.0188	0.0114	0.0023	0.1275
GYPP	-0.1355	-0.1794	0.4517	0.5839	0.1916	-0.0059	-0.456	0.5156	0.6683	0.3159	0.3996	0.0684	0.2017

R SQUARE = 0.7145, RESIDUAL EFFECT = 0.5343

Similar results for path coefficient were recorded by several researchers [17,18,19,20]. Effect of grain Fe and Zn content on yield was found negligible in both crosses. The results of path coefficient for grain iron and grain zinc content were in accordance with the earlier findings [21, 22,23].

In the present study, the estimated residual effect was 0.3327 and 0.5343 in cross-I and cross II, respectively, indicating that about 46-77% of the variability in grain yield was contributed by the characters included in the study. The reason seems to be some other factors which have not been considered here, need to be included in this analysis to fully account for the variation in yield.

4. CONCLUSION

Among the F₂ population of cross-I, grain yield per plant exhibited significant and positive association with seeds per panicle, tillers per plant, flag leaf area, harvest index, test weight, days to 50% flowering and days to maturity while negative and significant correlation with canopy temperature. For cross-II, grain yield per plant exhibited significant and positive association with seeds per panicle, tillers per plant, plant height, flag leaf area, SPAD value, harvest index, test weight and days to maturity, while, significant and negative correlation with grain Zn content, grain Fe content and canopy temperature. Path coefficient study indicated that harvest index in cross-I and value of SPAD in cross-II have exhibited high positive direct effect on grain yield per plant. Grain Fe and Zn content exhibited negligible direct effect on yield in both the crosses. The traits showing significant positive correlation and high positive direct effect contribute directly towards high grain yield. Hence, direct selection of harvest index along with indirect selection for tillers per plant, days to 50% flowering and days to maturity in cross-I and for value of SPAD in cross-II for yield improvement will be rewarding.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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