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Original Article

Morphometric Analysis of Kernel Trait in Rice (*Oryza sativa* L.): In search of Correlation among Quality Traits from F_6 Breeding Lines of a cross between Pusa Basmati-1 and IR-72 rice varieties

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Abstract

The hybridization of Pusa Basmati 1 and IR 72 produced an aromatic, elongating, and non-aromatic, non-elongating mapping population for quality attributes in rice. In terms of other qualitative qualities, the parents also differed genetically, and their genetic distance was considerable. In this study 365 lines in the F6 population were examined for kernel dimensions, cooked kernel elongation ratio (CKER), alkali spreading value (ASV), and kernel dimensions both before and after cooking. The apparent amylose content of the sub-set of 110 F6 lines was also characterized. With the exception of kernel breadth and ASV, all features showed continuous and normal distribution, indicating quantitative inheritance. All of the characteristics, with the exception of ASV, showed transgressive segregation. High heritability findings suggest that the quality attributes in the F6 lines were genetically fixed. One-way analysis of variance (ANOVA) was used to evaluate four distinct hypotheses about the correlation of QTLs for quality attributes. The phenotypic variance explained by the QTLs was 45.68% for CKE-R, 38.63% for LAC, and 25.35% for L-BAC, indicating that the parent Pusa Basmati 1 was the source of the gene(s) controlling the inheritance of these traits.

Keywords:- Rice, *Oryza sativa* L., QTL analysis, cooked kernel elongation ratio (CKER), alkali spreading value (ASV).

Introduction

The most significant food source in the world is rice (Oryza sativa L.), and rice quality has a significant impact on market value and customer acceptance. Grain quality features and cooking and eating quality traits are two kinds of rice quality traits. The majority of the qualities have a quantitative basis. The challenges in attempting to improve these complex features have been exacerbated by the combination of polygenic inheritance and environmental factors. Quantitative inheritance exists for grain dimension features, such as length, breadth, and length-breadth ratio (McKenzie and Rutger, 1983; Takita, 1985; Chauhan and Chauhan, 1994; Geetha, 1998).

When cooked, certain types enlarge more than others. In high quality rice varieties, lengthwise expansion (also known as kernel elongation) without an increase in girth is seen to be a very desirable characteristic. When cooked, the basmati type aromatic rice varieties from India, Pakistan, Afghanistan, Iran, Bangladesh, and Burma all elongate by 100% (Khush *et al*., 1979). Additionally, the Japonica rice varieties significantly lengthen as they cook (Juliano and Perez, 1984). The starch-gelatinization temperature (GT) of these elongating rices is often low (70°C), the amylose content (AC) is typically low to moderate (25%), and the gel consistency (GC) is typically medium (41–60 mm; Juliano, 1979). Although cooked kernel elongation (CKE) is a very important component of quality

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which determines the price of rice in national and international markets, it has received little attention so far and only a few reports are available on the genetics and mapping of this trait.

Geetha (1998) noted that additive gene activity predominated in the case of CKE. So CKE has shown to be quite challenging to adequately phenotype. The waxy gene predominantly regulates the amylose content, a significant factor in rice starch quality. According to thorough genetic investigations, AC was determined by a single major locus with certain minor genes serving as modifiers (McKenzie and Rutger, 1983; Kumar and Khush, 1986a, 1986b, 1987, and 1988).

Cooking time is determined by the gelatinization temperature (GT), a physical characteristic of starch. The starch granules in hot water start to irreversibly swell within a temperature range (between 55°C and 79°C). GT and Alkali Spreading Value (ASV) have the opposite relationships. Rice with low and intermediate GT entirely or partially disintegrates in the alkali solution, but rice with high GT mostly escapes undamaged (Khush *et al*., 1979). It is unclear how ASV (or GT) is inherited. Early research (Stansel, 1965) came to the conclusion that 1-3 genes with many modifiers were involved; subsequently McKenzie and Rutger, in 1983 suggested quantitative inheritance.

In a review paper Rashmin M Dhingani and co-workers observed that it is now possible to analyze both the simply inherited and quantitative trait and identify individual genes controlling the traits of interest with the advancement of molecular marker analysis technology (Rashmin M Dhingani *et al.*, 2015). In order to identify molecular markers that are closely linked to the quantitative trait loci (QTL) for cooked kernel elongation, this study involved the development of a permanent mapping population using two parents that significantly differed in quality traits, particularly cooked kernel elongation (CKE) and aroma. This study's objective include QTL analysis for quality attributes using the mapping population's phenotypic data.

Materials and methods:

Two rice lines Pusa Basmati-1 (aromatic, elongating) and IR-72 (non-aromatic, non-elongating) were chosen as parents and a mapping population segregating for quality traits was developed. This investigation started when the population was at the F_6 stage.

Quality Characterization of the Segregating Populations

A total of 365 distinct plants from the F6 population were examined for grain and kernel dimensions (GL, GB, GL-B, LBC, BBC, and L-BBC). For kernel dimensions, before and after cooking, cooked kernel elongation, and alkali spreading value (LBC, BBC, L-BBC, LAC, BAC, L-BAC, CKER, and ASV) these 365 single plants from the F6 population were phenotyped. The subset of 110 F6 lines' apparent amylose content (AC) was calculated. Using the technique outlined in the Standard Evaluation System (SES, IRRI, 1996), the ASV and the apparent AC were calculated.

On a piece of black chart paper measuring 7 x 4 cm, 5.5 x 2.5 cm of double-sided tape were used to connect a total of 10 well-developed grains per plant and de-husked grains, or kernels (pericarp removed by polishing with sand paper, Million flint paper, grade-0, Million abrasives Pvt. Ltd., India). Under an Olympus dissecting microscope, the length and width of each were measured using a stage-micrometer and an eye-piece graticule (standardised using a stage micrometre). For each of the three replications, the averages for grain length, grain breadth, kernel length, and kernel breadth were measured. By dividing each grain's and kernel's length by their respective breadth, the respective length-breadth ratio for each replication was computed, and the average was determined.

In a microwave oven (LG, model Intellogrill) that had been preheated at 1000 C for 5 minutes, a total of 20 dehusked and polished rice kernels were cooked for 15 minutes. Five whole cooked kernels were examined under a microscope to measure their length and breadth as previously mentioned. The three replications' average values were computed. LAC and BAC were divided to determine the L-BAC. Both a percentage and a ratio are used to express cooked kernel elongation. The ratio and percentage elongation are calculated by dividing the average cooked kernel length by the average uncooked kernel length (Table I).

Analysis of the Data on Quality Traits Obtained from the Segregating Populations

The statistical programme Statistica 5.0 was used to calculate the mean, range, and standard deviation for the F6 (365 lines) populations. Using the software programme SPSS 10.0 (SPSS Inc.

USA), the frequency distribution of the attributes for the population was tested using the F6. Kolmogorov-Smirnhov and 2 tests. Heritability and variance were computed in the F6 population using SPSS 10.0.

QTL Analysis

One-way analysis of variance (ANOVA): ANOVA analysis were conducted using software SPSS 10.0.

Results

Characteristics of quality traits

The Tables I and II show the range, mean, and standard deviation of quality traits for the F6 population (cooking-related elongation represented as a ratio, alkali spreading value, and kernel dimensions before and after cooking), as well as the variance and heritability (h2) for the same quality traits for F6 lines. Cooked kernel elongation was expressed as a ratio since percentage data need to undergo an arc-sine transformation for statistical analysis, but it was also expressed as a percentage in accordance with accepted standards for clarity's sake.

Table I: The range, mean and standard deviation of quality traits for F_6 population (kernel dimensions before and after cooking, CKER and ASV of 500 lines)

	Traits	Range		Mean	Standard deviation
	Kernel length-breadth ratio before cooking	3.11	5.99	4.203	0.468
F_6 population	Kernel length before cooking	5.41	9.04	7.10	0.713
	Kernel breadth before cooking	1.36	2.11	1.66	0.145
	Kernel length-breadth ratio before cooking	3.102	5.56	4.283	0.510
	Kernel length after cooking	7.32	17.66	11.876	1.792
	Kernel breadth after cooking	1.78	3.91	2.816	0.292
	Kernel length-breadth ratio after cooking	2.29	7.69	4.268	0.817
	Cooked kernel elongation ratio	1.31	2.47	1.813	0.206
	Alkali spreading value	2.00	7.00	4.398	1.665

Table II. The variance and heritability (h^2) for quality traits in the F_6 population from the cross Pusa Basmati 1-IR 72.

	LBC	BBC	L-BBC	LAC	BAC	L-BAC	CKER
Variance							
σ^2G	5.059	0.210	2.595	32.012	0.863	6.645	0.660
σ^2E	0.062	0.004	0.020	0.625	0.015	0.049	0.007
σ^2P	5.121	0.214	2.615	32.637	0.878	6.694	0.667
h^2 (σ^2G-σ^2P)	98.77	97.77	99.23	98.08	98.22	99.26	98.93

LBC-kernel length before cooking, BBC-kernel breadth before cooking, L-BBC-kernel length-breadth ratio before cooking, LAC-kernel length after cooking, BAC-kernel breadth after cooking, L-BAC-kernel length-breadth ratio after cooking, CKER-cooked kernel elongation ratio, σ^2G-genotypic variance, σ^2E-variance due environment, σ^2P-phenotypic variance, h^2 (σ^2G- σ^2P)-heritability.

Except for the correlation found between CKER and L-BBC, which was negligible, other character combinations had highly significant correlations, whether positive or negative (at the 0.01 level) as shown in Table III, IV and V).

Table III. Distribution of quality traits in different segregating populations F_6

Ppn	Trait	Kolmogorov-Smirnov Test		χ^2 Test	df	'p' value	Type of Distribution
		'd' value	'p' value				
F6	Kernel length before cooking	0.021	NS	18.912	13	0.128	Normal
	Kernel breadth before cooking	0.057	<0.01	61.143	13	0.000	
	Kernel length-breadth ratio before cooking	0.030	NS	15.941	14	0.317	Normal
	Kernel length after cooking	0.043	NS	27.607	11	0.003	Normal
	Kernel breadth after cooking	0.028	NS	24.327	8	0.003	Normal
	Kernel length-breadth ratio after cooking	0.047	NS	29.538	9	0.0005	Normal
	Cooked kernel elongation ratio	0.026	NS	18.063	11	0.080	Normal
	Alkali spreading value	0.132	<0.01	805.863	17	0.000	

NS-non significant, Ppn- population

Table IV. Correlation coefficients between kernel dimensions, before and after cooking, cooked kernel elongation ratio of the F_6 population derived from the cross Pusa Basmati 1-IR 72

	LBC	BBC	L-BBC	LAC	BAC	L-BAC	CKER
LBC	1.000						
BBC	0.222**	1.000					
L-BBC	0.676**	-0.560**	1.000				
LAC	0.331**	-0.171**	0.425**	1.000			
BAC	-0.041**	0.338**	-0.290**	0.043**	1.000		
L-BAC	0.298**	-0.338**	0.523**	0.814**	-0.528**	1.000	
CKER	-0.297**	-0.306**	-0.005	0.798**	0.066**	0.632**	1.000

**Correlations are significant at 0.01 level. * - Correlations are significant at 0.05 level; LBC-kernel length before cooking, BBC-kernel breadth before cooking, L-BBC-kernel length-breadth ratio before cooking, LAC-kernel length after cooking, BAC-kernel breadth after cooking, L-BAC-kernel length-breadth ratio after cooking, CKER-cooked kernel elongation ratio.

Table V Correlation coefficients between kernel dimensions, before and after cooking, cooked kernel elongation ratio, apparent amylose content and ASV of the genotyped 113 F_6 population derived from the cross Pusa Basmati- 1 X IR-72.

	LBC	BBC	L-BBC	LAC	BAC	L-BAC	CKER	AC	ASV
LBC	1.000								
BBC	-0.050	1.000							
L-BBC	0.777**	-0.662**	1.000						
LAC	0.402**	-0.244**	0.471**	1.000					
BAC	-0.268**	0.317**	-0.403**	-0.230*	1.000				
L-BAC	0.434**	-0.340**	0.556**	0.897**	-0.618**	1.000			
CKER	-0.077	-0.215*	0.096	0.878**	-0.112	0.746**	1.000		
AC	0.093	0.080	0.012	-0.068	0.074	-0.093	-0.139	1.000	
ASV	-0.246**	-0.144	-0.091	0.116	-0.111	0.136	0.256**	0.060	1.000

**Correlations are significant at 0.01 level. * - Correlations are significant at 0.05 level, LBC-kernel length before cooking, BBC-kernel breadth before cooking, L-BBC-kernel length-breadth ratio before cooking, LAC-kernel length after cooking, BAC-kernel breadth after cooking, L-BAC-kernel length-breadth ratio after cooking, CKER-cooked kernel elongation ratio, AC-apparent amylose content, ASV-alkali spreading value.

Discussion

For the purpose of creating a mapping population segregating for quality attributes, the aromatic-non-aromatic combination of Pusa Basmati-1 X IR-72 cross was chosen based on preliminary agromorphological evaluation of several genotypes and crosses. The majority of the qualities desired in high-quality aromatic rice were integrated in one genotype by the aromatic parent Pusa Basmati-1 (an EB), including extra-long, slender grains and kernels, the greatest LAC (16.04 mm), a high CKER (1.99), and a high ASV. This genotype has a questionably high apparent amylose content (25.63). However, according to Siddiq *et al.* (1994) and Sengupta and Choudhury (1995), this genotype exhibited late flowering (95 days) and was vulnerable to the majority of insects, pests, and diseases.

Breeding trials commonly use the non-aromatic parent, IR-72, a semi-dwarf HYV with various resistances created by IRRI, as a global check. In contrast to Pusa Basmati-1, IR- 72 had early flowering, medium-length, slender grains, medium-length kernels, low ASV (2.08), low CKER (1.51), and either high GT and high AC (27.92%) or low LAC (9.59mm). Additionally, neither parent was photoperiod sensitive, allowing for quick generational change. The transgressive segregation for all quality traits in the segregating generations (F2 and F6), with the exception of ASV, further supported the genetic separation between the cross's parents. However, the parents were genetically compatible enough to yield a promising population from which aromatic, elongating, medium high yielding recombinants may be recovered by selection.

So with the exception of BBC and ASV, all traits were continuously and normally distributed, indicating quantitative inheritance, according to the Kolmogorov-Smirnov and 2 tests. While the ASV distribution was bimodal, the kernel breadth distribution was close to normal. All of the characteristics, with the exception of ASV, showed transgressive segregation. The F6 lines were genetically fixed for the quality traits and eligible for QTL analysis, according to the heritability values, which ranged from 97.77 to 99.26 as shown in Table III, IV and V.

In a similar investigation the inheritance pattern of kernel length and cooked kernel elongation in the cross Taroari Basmati x RAU 3036, and Pusa Basmati 1121 x Chhatri was studied. From the data from the F_2 lines it was concluded that kernel length and kernel elongation ratio was controlled by one or two major genes and those were influenced by few modifier genes (Kumar *et al*, 2018).

In the F6 population, CKER had strong positive correlations with LAC and L-BAC, positive correlations with BAC albeit with moderate coefficients, and negative correlations with LBC and BBC. LBC had significant and strong correlations with L-BBC, BBC, LAC, and L-BAC, but negative correlations with CKER and BAC. L-BBC, LAC, L-BAC, and CKER all had negative correlations with BBC, while BAC and LAC had positive correlations. L-BBC had a strong positive correlation with LAC and L-BAC but a weak negative correlation with BAC. L-BAC, CKER, and BAC were all favorably linked with LAC (low coefficient value), as well as with each other. BAC and L-BAC had a strong negative correlation, although CKER and BAC had a positive correlation with a modest coefficient value.

Conclusion

In the F6 population, LBC had a negative and insignificant association with both BBC and CKER, but a positive correlation with LBC and BBC. One of the two qualities, AC, was not substantially connected with any other trait in the sample population while the other, ASV, was favourably correlated with CKER and negatively correlated with LBC.

All of the characteristics, with the exception of ASV, showed transgressive segregation. High heritability findings suggest that the quality attributes in the F6 lines were genetically fixed. One-way analysis of variance (ANOVA) was used to evaluate four distinct hypotheses about the correlation of QTLs for quality attributes. The phenotypic variance explained by the QTLs was 45.68% for CKE-R, 38.63% for LAC, and 25.35% for L-BAC, indicating that the parent Pusa Basmati 1 was the source of the gene(s) controlling the inheritance of these traits. It can be concluded that kernel length and kernel elongation ratio is controlled by one or two major genes and those are influenced by few modifier genes. But there is no consensus yet on the nature of inheritance of kernel length and kernel elongation.

Abbreviations used:

AC-apparent amylose content;

ASV-alkali spreading value;

EB-evolved basmati

GL-grain length;

GB-grain breadth;

GL-B-grain length-breadth ratio;

LBC-kernel length before cooking;

BBC-kernel breadth before cooking;

L-BBC- kernel length-breadth ratio before cooking;

LAC-kernel length after cooking;

BAC-kernel breadth after cooking;

L-BAC-kernel length-breadth ratio after cooking;

CKE-cooked kernel elongation;

CKER-cooked kernel elongation ratio;

GT-gelatinization temperature;

QTL-quantitative trait loci;

PVE- phenotypic variation explained;

Conflict of Interest: The first author has developed all the rice lines, as Research Associate, PMCG Section, Bose Institute. There is no conflict of interest with anybody or organization.

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References

Chauhan, J. S., & Chauhan, V. S. (1994). Genetic analysis of grain dimensions and weight and their association with grain yield in rainfed rice (Oryza sativa). *Indian Journal of Agricultural Sciences (India)*.

Geetha S.1998. Combining ability analysis of quality characters of rice grains. Agric. Sci. Digest. 18: 1-3.

Juliano, B. O. (1979). The chemical basis of rice grain quality. *Chemical aspects of rice grain quality*, *69*, 90.

Juliano, B. O., & Perez, C. M. (1984). Results of a collaborative test on the measurement of grain elongation of milled rice during cooking. *Journal of Cereal Science*, *2*(4), 281-292. https://doi.org/10.1016/S0733-5210(84)80016-8

Kumar, V., Rastogi, N. K., Patel, N. B., & Mandal, S. N. (2018). Inheritance of kernel length and kernel elongation in basmati rice (Oryza sativa L.). *Electronic Journal of Plant Breeding*, *9*(2), 429-432. https://doi.org/10.5958/0975-928X.2018.00053.4

Khush, G. S., Paule, C. M., & De la Cruz, N. M. (1978). Rice grain quality evaluation and improvement at IRRI. In *Proceedings of the workshop on chemical aspects of rice grain quality*. Los Banos, Philippines, International Rice Research Institute, 1979..

Kumar, I., & Khush, G. S. (1986a). Genetics of amylose content in rice (Oryza sativa L). *Journal of Genetics*, *65*, 1-11. https://doi.org/10.1007/BF02923530

Kumar, I., & Khush, G. S. (1986b). Gene dosage effects of amylose content in rice endosperm. *The Japanese Journal of Genetics*, *61*(6), 559-568. https://doi.org/10.1266/jjg.61.559

Kumar, I., & Khush, G. S. (1987). Genetic Analysis of Different Amylose Levels in Rice 1. *Crop Science*, *27*(6), 1167-1172. https://doi.org/10.2135/cropsci1987.0011183X002700060016x

Kumar, I., & Khush, G. S. (1988). Inheritance of amylose content in rice (Oryza sativa L.). *Euphytica*, *38*, 261-269. https://doi.org/10.1007/BF00023529

McKenzie, K. S., & Rutger, J. N. (1983). Genetic analysis of amylose content, alkali spreading score, and grain dimensions in rice 1. *Crop Science*, *23*(2), 306-313. https://doi.org/10.2135/cropsci1983.0011183X002300020031x

Dhingani, R. M., Umrania, V. V., Tomar, R. S., Parakhia, M. V., & Golakiya, B. (2015). Introduction to QTL mapping in plants. *Ann Plant Sci*, *4*(04), 1072-1079..

Sengupta K. and Choudhury S. 1995. In “Varietal Characteristics for Identification-1. Rice” (Sengupta K. and Choudhury S., Eds.). pp.36. Directorate of Agriculture, West Bengal

Siddiq E. A, Muralidharan K, Sobha Rani N, Pillai K. G, Reddy J. R. K and Pasalu I. C. 1994. Basmati Rice. Directorate of Rice Research, ICAR, India.

SES. 1996. International Network for Genetic Evaluation of Rice. (1996). *Standard evaluation system for rice*. IRRI, International Rice Research Institute.

Stansel, J. W. (1965). The Influence of Heredity and Environment on Endosperm Characteristics of rice (Oryza sativa) (Doctoral dissertation, Purdue University).

Takita, T. (1985). Inheritance of grain size and the relationship between grain size and other characters in rice. *Bull. Natl. Agric. Res. Cent.*, *3*, 55-71.