Journal of Applied Sciences

ISSN 1812-5654

Journal of Applied Sciences 8 (8): 1503-1510, 2008
ISSN 1812-5654

Inheritance of Gelatinization Temperature and Gel Consistency in Rice (*Oryza sativa* L.)

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Abstract: Gelatinization Temperature (GT) and Gel Consistency (GC) are important traits in determination of rice quality. In present study four rice cultivars namely Sang-e-Tarrom, Gerdeh, IR229 and IRRI2 have been utilized in hybridization as parents. In order to know about genetic characteristics and inheritance of the related traits, direct and reciprocal crosses have been conducted between parents Sang-e-Tarrom and Gerdeh and between IR229 and IRRI2. Inheritance of related traits have been studied in direct and reciprocal hybridizations for hard, intermediate and soft GC, low and high GT. For these evaluations P_1, P_2, F_1, F_2, BC_1, BC_2 generations and their reciprocal crosses have been utilized in present study. Results showed that gene hard GC dominates on intermediate and soft and also intermediate GT dominates on low. From direct and reciprocal crosses Gerdeh × Sang-e-Tarrom and IR229 × IRRI2 have been illustrated that GC and GT expressions are under monogenic control of one major gene corresponding with several modifier genes. Gene dosage effects play a tremendously important and effective role in segregation production among traits. Regarding to the obtained results selections can effectively be performed in later segregation generations for GC and in early generations for GT.

Key words: Rice, inheritance, cooking quality, gel consistency, gelatinization temperature, gene dosage effects

INTRODUCTION

Gel Consistency (GC) and Gelatinization Temperature (GT) are important rice quality traits (parameters) which have importance for evaluation of rice cooking quality. Gel consistency showing how stickiness the cooled paste of cooked rice flour is and used as a criterion for evaluation of cooked rice texture. Regarding the GC, which have been grouped based on continuous movement of the alkali gel during test using rice kernal powder, are divided into three groups namely soft, intermediate and hard with 16-32, 24-36 and 37-60 mm gel length, respectively (Yih-Chuan *et al*., 1985; Tomar and Nanda, 1987; Cagampang *et al*., 1973). Indeed, gel consistency and amylose content of rice and its viscosity profile or pasting profile have been studied since 1972 (Tomar and Nanda, 1987).

Studies showed that two rice cultivars with the same amylose content may be differentiated with gel consistency test and they have various cooking qualities. In such a case, cultivar with softer GC is preferable. Cooked rice cultivars with hard GC have changed to hard faster than those with soft GC. However, cultivars with soft GC cooked slowly and stay soft even after cooling; thus, consumers prefer cultivars having soft GC. Therefore, researchers have attempted to produce high yielding cultivars with soft GC (Khush *et al*., 1979).

Gelatinization temperature determines the cooking time of rice kernels (Heda and Reddy, 1986). This trait is of physico chemical importance and starch granules start to swell irreversibly in hot water. Gelatinization temperature ranges from 55 to 74°C. Cultivars with GT 55-69, 70-74 and 75-79°C are categorized as low, intermediate and high GT, respectively (Khush *et al*., 1979). Cooked kernels are transformed into hard and dry in high GT cultivars, whereas are sticky in low GT cultivars, such that from property we can directly determine the cooking quality of cultivars.

Heda and Ready (1986) mentioned AC and GT are two important traits in rice cooking quality which determine rice texture after cooking and also rice cooking time, respectively. Tan *et al*. (1999) expressed a closed relationship among three traits, such that, according to genetic analysis based on molecular markers only two types of parental relationships from these traits have been conveyed. They are high GT/soft GC/low AC or low GT/hard GC/high AC. Tang *et al*. (1998) studied inheritance of GC in crosses between indica and Japonica groups. Analyses have been performed based on single seeds harvested from one season over parents, F_1, RF_1, F_2 and RF_2 generations and differences existed among hard × soft, hard × intermediate and intermediate × soft GC indicated that GC is under control of a single major gene with several modifiers in similar loci and is affected by

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these modifiers. Jenkings *et al.* (1979) concluded that inheritance pattern of GT is not so clear; however, it seems to have a simple inheritance pattern corresponding with one or two major genes. Heidelberg and Li (1999) during their study on hybrid of indica/Japonica observed that a single major effect and a minor effect gene control GT trait that both of them have found on chromosome 6 and by creating doubled haploids via the hybrid anther culture observed that for AC also have a single major and minor effect genes which existed on chromosomes 2 and 7, respectively. Tan *et al.* (1999) in their studies related to genetic analysis based on molecular markers for three traits using three populations of F_2, $F_{2:3}$ and F_9 which were obtained from combinations of parental inbred line Shanyou, found that all three traits have been controlled by only one gene locus that stayed on Wx area on short arm of chromosome 6. Mohamad *et al.* (2004) have considered inheritance of GT in three Malaysian landraces in F_2 generation and by analysis of F_2 generations seeds obtained from cross Mahsuri mutant × Mahsuri with GT (high × intermediate) showing two classes with ratio of 3 intermediate: 1 high. They found that this trait has been controlled by more than one locus. In cross between cultivars Mahsuri × 9192 with GT (intermediate × intermediate) only one of the created curves had not fitted with Mendelian feature. Therefore, results imply the complexity nature of this trait.

Because of triploid nature of endosperm and various roles of each parent (paternal and maternal effects) in endosperm of the first generation (F_1), it is expected that different expressions of trait have been observed in endosperm. The objective of present experiment was consideration of inheritance of two traits, GT and GC and determination of parental gene dosage effects on different levels of these traits.

MATERIALS AND METHODS

Four rice cultivars namely Sang-Tarrom, Gerdeh, IR229 and IRRI2 were used as parents for hybridizations. Selected parents had different levels of studied traits (Table 1).

In order to know inheritance of traits, an experiment was undertaken at research station of College of Agricultural Sciences and Natural Resources University of Mazandaran in 2005-2007. Direct and reciprocal crosses have been conducted between cultivar Sang-e-Tarrom × Gerdeh and between IR229 × IRRI2. The F_1 seeds were grown in flowering stage and often that half of plants were crossed by both parents and remained F_1 plants were selfed for obtaining F_2 seeds. Therefore, 10 different populations (generations) including first and second generations of hybrid seeds (F_1 and F_2), backcrossing with first and second parents (BC_1 and BC_2) and reciprocal crosses of all generations corresponding with parental seeds have been considered. Two months after harvesting, to measure related traits all seeds were transferred into laboratory and were analyzed. Number of seeds needed for parents and F_1 were 20-30 (each 10 seeds for one replication), for F_2 were 350-400 seeds and for each backcrossing were 40-80 seeds (Kumar and Khush, 1988; Mckenzie and Rutger, 1983). To measure GC and GT methods of (Cagampang *et al.*, 1973) and (Zaman *et al.*, 1986) in single seed system and (Little *et al.*, 1958) have been utilized, respectively.

Table 1: Origin, height and different levels of studied traits in parental cultivars

Cultivars	Origin	Height	GT	GC (mm)	AC (%)
Sang-Tarrom	Landrace (Iran)	Tall	3.30	33.8	22.89
Gerdeh	Landrace (Iran)	Tall	6.10	45.5	19.91
IR229	IRRI	Tall	6.39	19.3	27.16
IRRI2	IRRI	Short	7.00	33.2	24.85

GT = Gelatinization Temperature, GC = Gel Consistency, AC = Amylose Content, IRRI = International Rice Research Institute

Also, endosperm of parents and crosses have been used for determining these traits in two replications and obtained data were analyzed based on randomized complete block design. Reciprocal crosses have been used to determine gene dosage effects.

To compare means of crosses and to determine gene dosage effects SAS statistical software and Duncan's multiple ranges test were used. To calculate changes from each gene dosage in each trait, its amount in one of two parents that showed higher levels of trait in each cross supposed as 100 (3 gene dosages) and another parent with lowest amount of trait supposed as zero dosage and corresponding with two direct and reciprocal crosses of these two parents have been determined as a percentage of superior parent. Percentage of changes for each gene dosage has been calculated as declining higher dosage from lower dosage and to compare differences of gene dosage effects Duncan's test have been used.

RESULTS

Comparisons of different generations of Sang-Tarrom/Gerdeh cross for GC: Maternal parent (Sang-e-Tarrom) had intermediate GC (33.7 mm) and paternal parent (Gerdeh) had soft GC (45.5 mm), thus, they varied about 11.7 mm gel length from each other. Mean GC in F_1 of these parents with one gene dosage for soft GC was about 34.3 mm. However, mean GC in F_1 of their reciprocal cross with two gene dosages for soft GC was nearly 35.9 mm. It indicates dominance of intermediate GC on soft GC (Fig. 1).

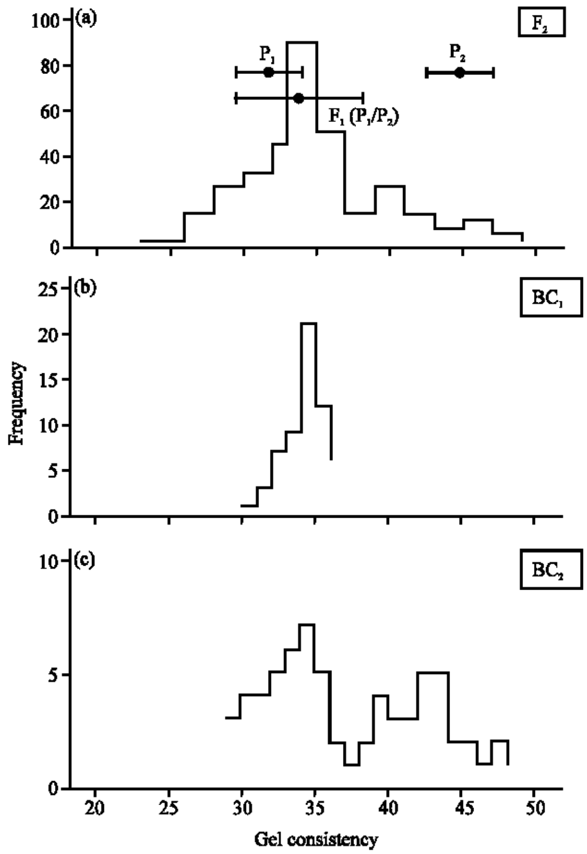

Fig. 1: Frequency distribution of seeds with different gel consistency from the cross Sang-e-tarom/Gerdeh

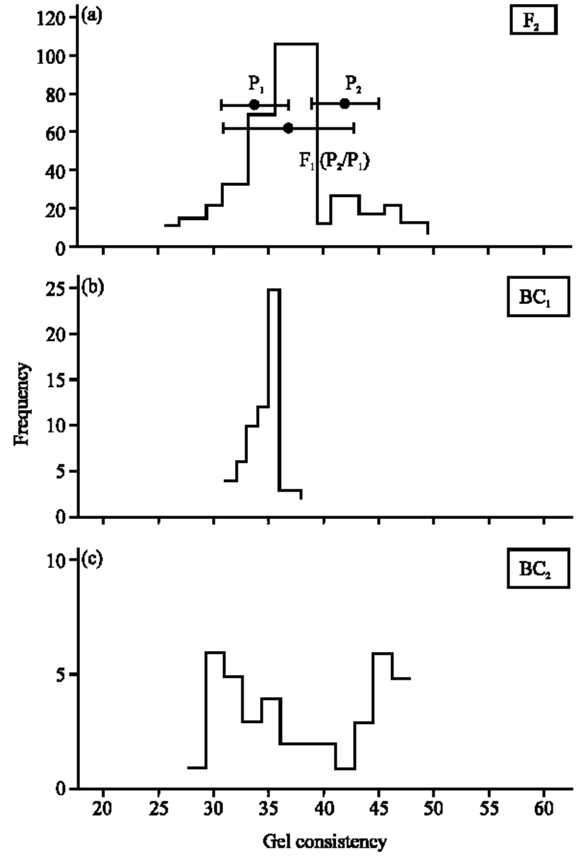

Fig. 2: Frequency distribution of seeds with different gel consistency from the cross Gerdeh/Sang-e-tarom

Gel consistency from 354 seeds of F_2 generation of this cross was stayed at two classes as 267 seeds in first class (23-35 mm) and 87 seeds in second class (37-49 mm), respectively. It implies a suitable 3:1 ratio (p = 0.5-0.75, χ^2 = 0.033) (Fig. 1). Figure 1b showed that with analysis of 61 seeds of BC_1 [Sang-Tarrom × (Sang-e-Tarrom/Gerdeh)], obtained curve categorizes GC into only one class. This result expresses that there is very little differences between zero and two gene dosages for soft GC. The curve obtained from analyze of 67 seeds of BC_2 [Gerdeh × (Sang-Tarrom/Gerdeh)] expresses two classes with 36 seeds in one class (29-36 mm) and 31 seeds in another class (37-48 mm). These data was suited with 3:1 ratio (p = 0.5-0.75, χ^2 = 0.033) (Fig. 1c).

In analysis of reciprocal crosses of F_2, BC_1 and BC_2 populations (Gerdeh × Sang-e-Tarrom) similar to above cases have been observed (Fig. 2). In GC from 336 seeds of F_2 population in cross Gerdeh/Sang-e-Tarrom) two classes were observed that 255 seeds were in the first class (24-36 mm) and 81 seeds were in second class (37-49 mm) showing 3:1 ratio (p = 0.5-0.78, χ^2 = 0.143).

Curve drawn from analysis of BC_1 seeds showed one class with 31-36 mm, however, for BC_2 it showed two certain classes with 21 seeds in the first class (28-36 mm) with intermediate GC and 23 seeds in the second class with soft GC, so that, this result indicated dominance of intermediate GC on soft GC (Fig. 2).

Comparison of different generations of IR229/IRRI2 cross for GC: Maternal parent IR229 with GC of 19.3 mm and paternal parent IRRI2 with GC of 33.32 mm showed variation equal to 13.9 mm gel length. Mean GC of F_1 obtained from cross IR229/IRRI2 with one gene dosage for intermediate GC and from reciprocal cross IRRI2/IR229 with two gene dosages for intermediate GC were 21.4 and 22.5 mm, respectively (Fig. 4a). Range of 354 analyzed seeds in F_2 generation from cross IR229/IRRI2 showed 17-38 mm variation that 261 seeds with gel length of 17-23 mm were in one class and 87 seeds with gel length of 24-36 were in another class and only 3 seeds with 38 mm were also existed that this result is in accordance with ratio 3:1 (p = 0.5-0.75, χ^2 = 0.101) (Fig. 3a).

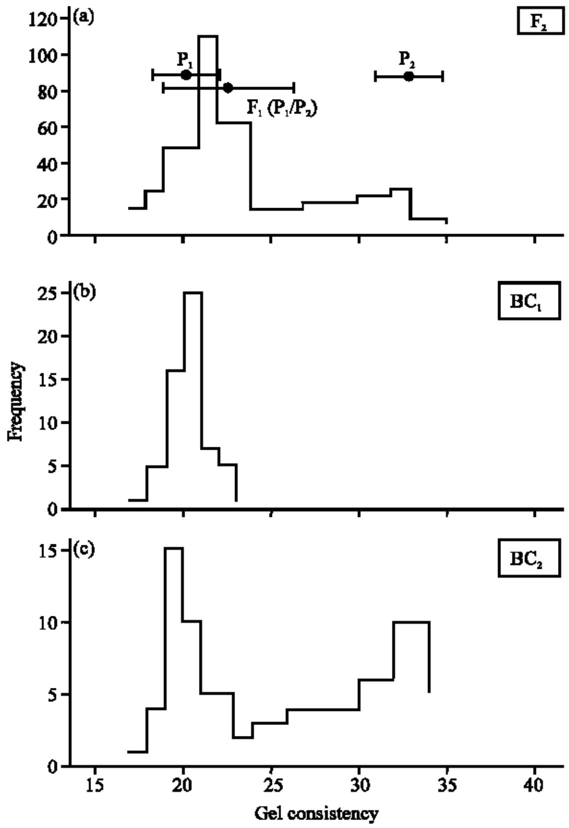

Fig. 3: Frequency distribution of seeds with different gel consistency from the cross IR229/IRRI2

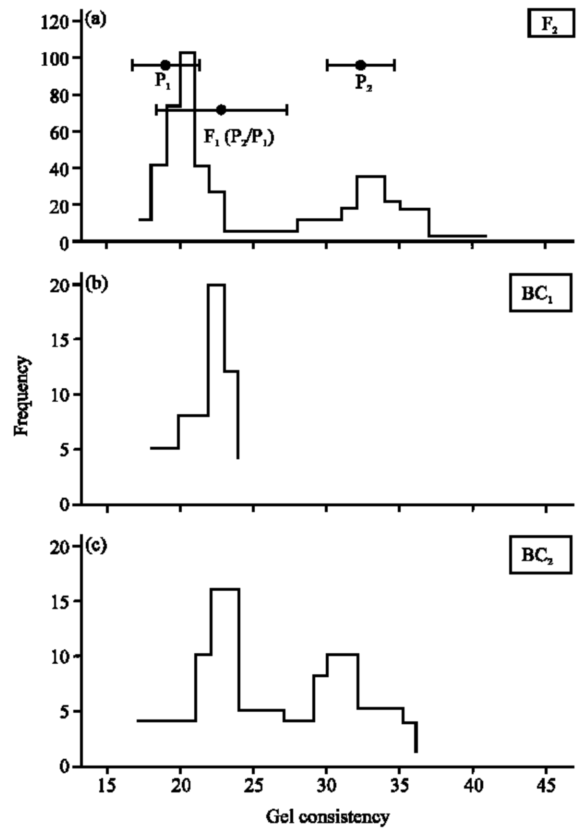

Fig. 4: Frequency distribution of seeds with different gel consistency from the cross IRRI2/IR229

Curve of GC drawn from analyzing of 59 seeds of BC_1 [IR229 × (IR229/IRRI2)] produced one class with 17-25 mm gel length that expresses little differences between zero and two gene dosages for intermediate GC (Fig. 3b).

Curve drawn from analyzing of seeds of BC_2 [IRRI2 × (IR229/IRRI2)] showed two classes that 37 seeds were in the first class with 17-23 gel length and 35 seeds were in the second class with 24-35 gel length indicating 1:1 ratio ($\chi^2 = 0.014$) (Fig. 3c). Above results imply that in analyses of F_2, BC_1 and BC_2 populations hard GC was dominated on intermediate GC.

Analyses of reciprocal cross (IRRI/IR229) showed similar results for direct cross populations (Fig. 4). In analyzing of 414 seeds of F_2 population extracted from cross IRRI2/IR229 306 seeds with gel length of 17-23 mm were in the first class and 108 seeds with gel length of 24-37 mm were in the second class and only 2 seeds were observed having soft gel that was suited with ratio 3:1 ($p = 0.5$-0.75, $\chi^2 = 0.26$).

Comparison of different generations of Sang-e-Tarrom/Gerdeh cross for GT: Maternal parent Sang-Tarrom had intermediate to high GT (3.3) and paternal parent Gerdeh had low GT (6.1). Mean GT of F1 obtained from Gerdeh/sang-Tarrom cross was 3.7 and in F_1 of its reciprocal cross was 4.8 that showed significant differences between direct and reciprocal crosses indicating gene dosage effects. Both F_1 progenies were in intermediate GT.

Results obtained from GT of 306 seeds of F_2 generation in Sang-e-Tarrom/Gerdeh cross showed two classes that 234 seeds with intermediate GT and 72 seeds with low GT were in agreement with 3:1 ratio ($\chi^2 = 0.35$) (Fig. 5a). Figure 5a also showed that from 48 analyzed seeds of BC_1 46 seeds showed intermediate GT and only 2 seeds showed low GT. Although there is significant difference between zero and two gene dosages, because of showing intermediate GT in both cases, both of them is stayed only into one class. Curve drawn from analyzing 50 seeds of BC_2 showed two classes. Because of significant difference observed between one and three gene dosages, 30 seeds were included in one class with intermediate GT and 20 seeds in another class with low GT that indicates a 3:1 ratio ($p = 0.5$-0.75, $\chi^2 = 2$) (Fig. 5c).

Aggressive segregations have been observed in F_2, BC_1 and BC_2 for both parents included in present

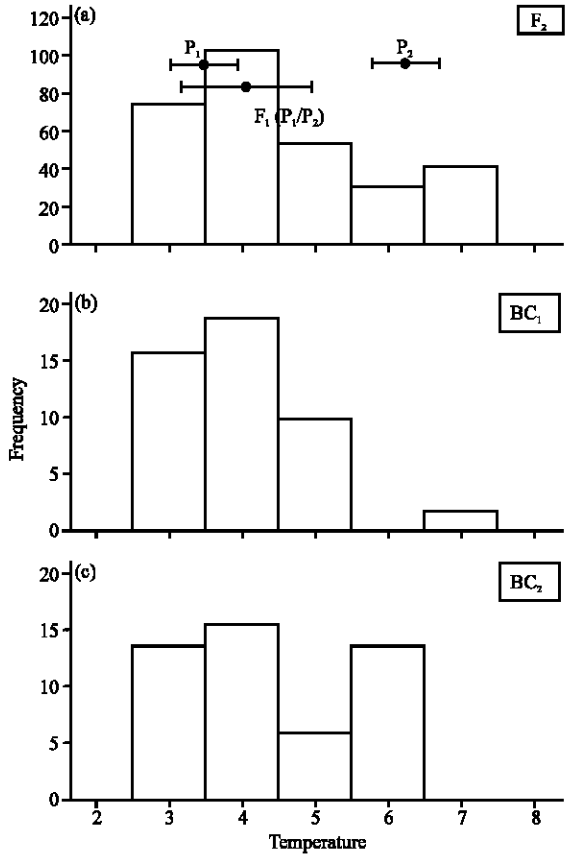

Fig. 5: Frequency distribution of seeds with different gelatinization temperature from the cross Sang-e-tarom/Gerdeh

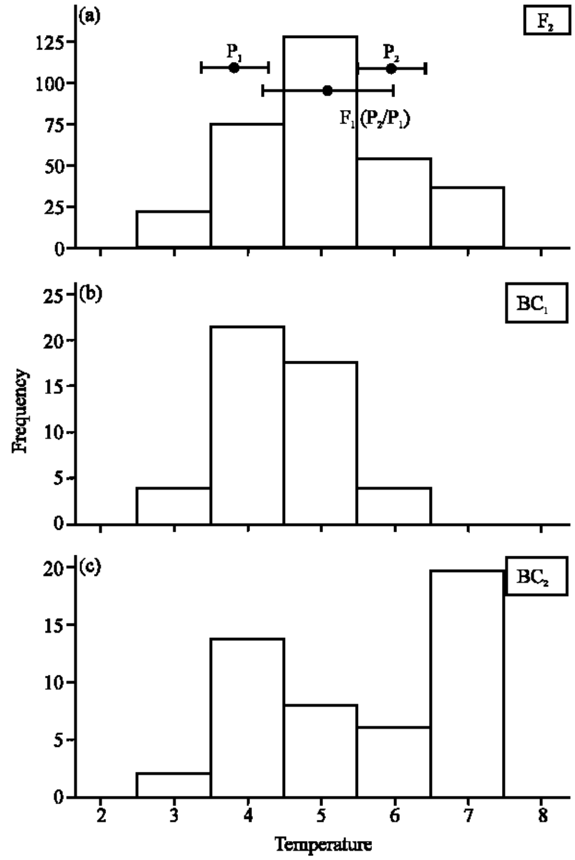

Fig. 6: Frequency distribution of seeds with different gelatinization temperature from the cross Gerdeh/Sang-e-tarom

experiment. Figure 6 demonstrated that from analyzing of F_2, BC_1 and BC_2 populations in reciprocal cross (Gerdeh/Sang-Tarrom) have also obtained similar results. From analyzing 288 seeds in F_2 generation obtained from Gerdeh/Sang-Tarrom cross, 255 seeds showed intermediate GT and 63 seeds showed low GT that was in agreement with 3:1 ratio (Fig. 6a).

In analyzing of 46 seeds of BC_1 in Sang-Tarrom × (Gerdeh/Sang-Tarrom) 44 seeds were included in one class with intermediate GT and only 2 seeds in another class with low GT that maximum frequency observed for intermediate GT (Fig. 6b). Curve drawn from analyzing 50 seeds showed two classes with 24 and 26 seeds, respectively. These classes were suited with 1:1 ratio ($p = 0.5\text{-}0.78$, $\chi^2 = 0.08$).

Results imply that the trait GT is controlled by a major gene. (Mckenzie and Rutger, 1983) by crossing between an intermediate GT and a low GT cultivar found that GT is controlled by one major gene with several modifier effects.

Comparisons of different generations of IR229/IRRI2 cross for GT: Both parents have low GT and therefore their F_1 means in direct and reciprocal crosses are stayed nearly in mid-parent position. Also, all generations showed low GT. In analyzing of 204 seeds from F_2 generation of IR229/IRRI2 and their reciprocal crosses one class have observed that only 12 seeds had GT of 5 (Fig. 7, 8). Therefore, due to close similarity of parents and F_1s, none other class has been observed in F_2 and backcrosses whether in direct or in reciprocal crosses.

Gene dosage effects: Means of GC and GT in parents and hybrids and variations due to different gene dosages invarious crosses are shown in Table 2 and 3.

Determination of gene dosage effects on controlling GC: Crossing between intermediate (Sang-e-Tarrom) and soft (Gerdeh) gel consistency genotypes.

The effect of the first dosage resulted from cross Sang-e-Tarrom/Gerdeh increased to 66% and showed that

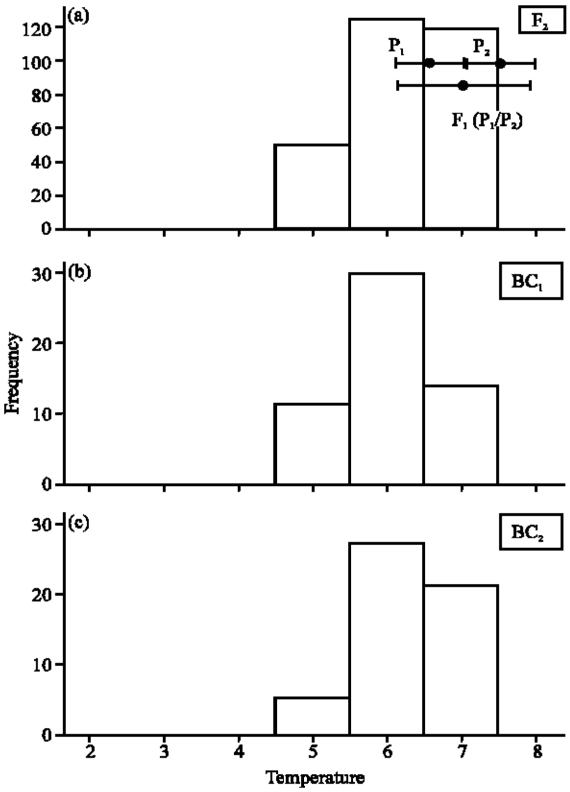

Fig. 7: Frequency distribution of seeds with different gelatinization temperature from the cross IR229/IRRI2

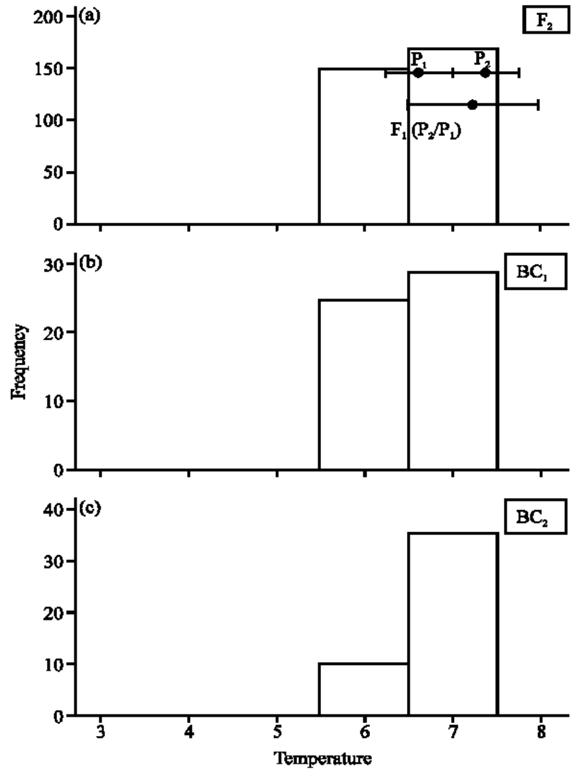

Fig. 8: Frequency distribution of seeds with different gelatinization temperature from the cross IRRI2/IR229

Table 2: Means of GC and GT in parents and hybrids (F_1, RF_1) in rice

Characters	Cross	P_1	F_1	RF_1	P_2
GT	Sang-Tarrom/Gerdeh	3.3	3.7	4.8	6.1
	IR229/IRRI2	6.3	6.5	6.8	7.0
GC	Sang-Tarrom/Gerdeh	33.8	34.2	35.8	45.5
	IR229/IRRI2	19.3	21.3	22.8	33.2

there is no significant difference with zero dosage. The second dosage has not shown any significant difference from the first dosage. However, there is no significant difference between F_1, RF_1 for gel consistency. However, the third dosage showed significant difference over the second dosage.

Crossing between hard and intermediate gel consistency genotypes: One and second dosage of the intermediate gel consistency gene in cross IR229/IRRI-2 did not show significant difference with zero dosage. In cross IR229/IRRI-2, only third dosages increased the gel consistency. However, all the three dosages have significantly enhanced the gel consistency.

Determination of gene dosage effects on controlling GT: Cross between high and low gelatinization temperature genotypes.

Table 3: Variations of GC and GT of rice kernels due to each gene dosage

		Gene dosage			
Characters	Cross	0	1	2	3
GT	Sang-Tarrom/Gerdeh	54.10	6.55^{ns}	18.03*	21.31*
	IR229/IRRI2	90.00	2.86^{ns}	4.28^{ns}	2.86^{ns}
GC	Sang-Tarrom/Gerdeh	74.50	0.66^{ns}	3.74^{ns}	21.10**
	IR229/IRRI2	58.13	6.32^{ns}	4.22^{ns}	32.23**

*, **Significant at 5 and 1% levels, respectively, ns = not significant

The average gelatinization temperature of F_1 generation resulted from cross Sang-e-Tarrom/Gerdeh is equivalent with maternal gelatinization temperature indicating no signification difference between zero and one dosage genes, but in reciprocal cross (gerdeh/Sang-e-Tarrom) the average gelatinization temperature F_1 generation was 4.8 mainly because of parental replacement and the second dosage, however, it showed a significant decreases. The third dosage showed a significant increase over the second dosage.

Cross between low and low temperature gelatinization parental genotypes: With respect to these results, due to existence of no significant variation in direct and reciprocal crosses in all generations F_1 generation has not shown significant difference between one, two and three gene dosages.

Therefore, information on dosage effects in all crosses is useful for better understanding the segregation patterns for these traits in different generations. Occurrence of different classes on the basis of dosage effects would be helpful for explaining the mode of inheritance of these important traits.

DISCUSSION

In study of the quality traits like gel consistency and gelatinization temperature, we focus on endosperm as one of the most important parts of seed. Endosperm is a triploid tissue having one dosage of paternal alleles and two dosages of the maternal.

The aim of gel consistency study was selection of soft gel consistency (with gel length of 37 to 60) which improves rice high cooking quality (Khush *et al*., 1979). In current study, effects of parental genetic analysis with hard, intermediate and soft gel consistency showed that differences between these three groups were controlled by a basic gene. Hard gel consistency has dominated on intermediate and intermediate gel consistency has dominated on soft gel.

Tang *et al.* (1998) in their analysis based on a single-grain of parents, F_1, RF_1, BC_1, BC_2, F_2 generations in hard and soft, hard and intermediate and intermediate and soft crosses showed that hard gel consistency has dominated on intermediate and soft controlled by a major gene with multiple alleles in similar locus and modifiers. Tang and Khush (1993) in analysis of a 6×6 diallel cross concluded that hard gel consistency has dominated on soft gel consistency. This trait has also influenced by a major gene and some minor genes. The hardening of gel consistency in all hybrids due to crossing testers showing hard gel consistency by maternal parents indicating soft and intermediate gel consistency reported by numerous investigators emphasizes that hard gel consistency has dominated on intermediate gel consistency (Juliano, 1971; Kaw and Delacruz, 1990; Tang and Khush, 1993; Tomar, 1987). Transgressive segregants were observed in all frequency distributions of crosses for gel consistency in the second generation and backcrosses. Transgressive segregants were observed in two crosses (Sang-e-Tarrom/Gerdeh) and (IR229/IRRI-2) in F_2 and BC_2 generations for both parents but in BC_1 was observed only for dominant parent. Also, in RBC_1 was seen a slight deviation relate to recessive parent. Tang and Khush (1993) concluded that transgressive segregants in some crosses and their separations are important. (Kumar and Khush, 1988) concluded trangressive segregants are due to a major gene. In crosses of Sang-e-Tarrom/Gerdeh and IR229/IRRI2 and their reciprocals intermediate and hard gel consistency was dominated, respectively. Because of importance of soft gel consistency in breeding programs, in different generations' desired conclusion can not be presentable, we propose selection can be done in later generations.

The purpose of improvement of gelatinization temperature is access to intermediate GT which have an adequate cooking quality. The results of this study showed that rice with appropriate gelatinization temperature can be easily selected and early generation selection may be effective. In generation F_2 and BC_2 in Gerdeh/Sang-e-Tarrom cross showed transgressive segregants in both parents but in BC_1 there is higher tendency toward dominant parent. (Kumar and Khush, 1986) reported that transgressive segregants is because of small polygenes or modifiers functions on gene dosage effects. Some investigators showed that inheritance of the gelatinization temperature is monogenic and some other showed it as polygenic (Heu and Choe, 1973; IRRI, 1976; Puri and Siddiq, 1980).

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