Influence of Seed Size on Germination and Early Seedling Growth in Indian Mustard (*Brassica juncea* L.)

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Indian mustard (*Brassica juncea* L.) accounts for about 66% of the total germplasm holdings of rapeseedmustard group of oilseed crops in the country. Conservation of good quality seeds of a germplasm accession is a prerequisite for its long-term viability and use in crop breeding programme. Seed size in Indian mustard influenced the loss of germination during storage. In the present investigation the effects of seed size on oil and protein content of seed, seed germination and seedling growth were studied during 2008-09 with 4 varieties and an advanced breeding line (Kranti, Pusabold, Pusa Jaikisan, Varuna, and BPR-549-9) of Indian mustard. Oil content varied from 36.4-38.9%, 39.0-42.6% and 39.0-43.8%, respectively, for small, medium and large group of seeds. Seed size had significant and positive correlation with root length (r= 0.655^*), dry weight (r= 0.894^{**}), root to shoot ratio (r= 0.879^{**}) and vigour index (r= 0.938^{**}). Seedlings dry weight, vigour index, root length and root to shoot ratio increased with an increase in seed size thereby suggesting that the seeds after grading should be conserved in the gene bank.

Key Words: Brassica juncea L., Seedling vigour index, Germination, Seed size, Germplasm, Conservation

Introduction

Rapid seedling establishment is an important requirement for successful crop production in dryland farming systems. Seed size, as a characteristic of seed quality, influences seedling growth and establishment. Seedling establishment and speed of emergence influence the time required for seedling to reach the autotrophic phase. Most investigators have reported a positive relationship between seedling vigor, improved stand establishment and higher productivity of cereal crops with plants originating from large seed compared to those grown from smaller seed. Amico et al. (1994) concluded that higher vigor that occurred in larger seed is due to the larger food reserves in these seeds. They also noted a positive linear relationship between seed weight and emergence in the field. Rapeseed-mustard is an important group of oilseed crops in India having more than 14, 000 germplasm accessions (Anonymous, 2009). Indian mustard (Brassica juncea L.), among these crops account for nearly 66 % of the total germplasm holdings in the country (Anonymous, 2007). Conservation of germplasm is an important aspect of crop genetic resource management programme. Kant and Tomar (1995) reported that seed size in Indian mustard also influenced seed germination. Furthermore, the rate of dry matter transfer and depletion of nutrients from the cotyledons is dependent on the size

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of the seed (Naidu and Narayanan, 1981) thus influencing seedling development. Therefore, conservation of good quality seeds of a germplasm accession is a prerequisite for its long-term viability, early seedling establishment, vigorous and healthy crop growth for higher yield and use in crop breeding programme. Grading of seeds is an effective method for eliminating seed of weaker seeds. In the present investigation an attempt was made to assess the effects of different grades of seeds on oil, protein content, seed germination and seedling growth of Indian mustard.

Materials and Methods

The present experiment was carried out during 2008-09 in laboratory with 4 varieties and an advanced breeding line of Indian mustard (Kranti, Pusabold, Pusa Jaikisan, Varuna and BPR-549-9). Grading of seeds was done by passing a random sample through a series of 3 sieves of different pore sizes and seeds were categorized in to three groups, *viz.*, small ($< 2 \text{ mm}^2$), medium (2 mm^2) and large (4 mm^2). The oil and protein content of different groups were analyzed using a pre-calibrated NIR Analyzer (Dicky John Instalab 600) while a random sample of 1000-seed was weighed with a digital electronic balance (Precisa). The pure seed fraction is poured onto the working table, mixed well, and divided with a spatula into the

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same number of equal parts as replicates. Each part is mixed well again and the replicates counted at random. The moist filter paper was lined in Petri dishes(9-cm diameter) and seeds were placed on top of the paper. The seeds in Petri-dishes were allowed to germinate in B.O.D incubator at a temperature of $25 \pm 1^{\circ}C$ and 55-60% relative humidity. The uniform temperature was maintained throughout the germination period. Seedlings were exposed to light for eight hours in every 24 hour as per rules (ISTA, 1985). At the first count (3rd day), the normal seedlings were removed and recorded and final count was taken on 7th day. The experiment was repeated thrice. Root dry weight was divided by shoot dry weight to work out the root to shoot ratio. Seedling vigour index was computed as the product of seedling dry matter and seed germination (Abdul-Baki and Anderson, 1973) and expressed in percent. Water imbibition of different groups was recorded on 20 seeds each during 2-12 hours of seed soaking and computed as follows:

$$IR = \frac{W_2 - W_1}{W_1} \times 100;$$

Where, $IR = W_1 = Initial$ weight of seeds and $W_2 =$ Weight of imbibed seeds.

Analysis of variance was conducted considering the data in factorial completely randomized design (Gomez and Gomez, 1984).

Results and Discussion

Analysis of variance revealed significant genotypic differences for all the characters except seed germination while seed size significantly affected all the characters investigated except seed germination and protein content. Water imbibition, 1000-seed weight, protein content, seedling dry weight and vigour index were significantly affected by interaction of variety x seed size.

Oil and Protein Content

Different groups of seeds had significantly different 1000-seed weight. Varuna showed maximum seed weight (2.94 g) within small seed group. The seed weight was highest for the genotype Pusa Jaikisan in the medium and BPR 549-9 in the large seed group (Table 1). Seed weight varied from 1.49 (Kranti) -2.94g (Varuna); 3.40 (Kranti) - 4.73g (Pusa Jaikisan) and 4.93 (Kranti)-6.54g (BPR-549-9), respectively, for small, medium and large seed group (Table 1). Oil content (%) also showed substantial variation among the genotypes and the groups.

The genotype Kranti and BPR 549-9 showed maximum (39.0%) oil content for smaller seeds while the genotype BPR-549-9 showed maximum oil content for medium (42.6%) and large (43.8%) seeds. Within the group oil content ranged from 36.4-39.0%, 39.0-42.6 and 39.0-43.8%, respectively, for small, medium and large seed group. The results suggested that higher oil content of large seeds might result in good seedling vigour as the seedling growth depends on the seed reserve during the initial growth stage as also suggested by (Naidu and Narayana 1981) in groundnut. Protein content (%) of the seeds did not differ significantly among the groups but genotypes had significant differences. The mean protein content was 20.1% for small, 20.2% for medium and 20.5% for large seeds (Table 1). Varuna and Pusa Jaikisan showed fairly consistent and higher protein content in all the groups of seeds. Since Indian mustard contains food reserve as oil, predominantly fatty acids, hence, limited variability existed for protein content and it seemed to be independent of seed size.

Water Imbibition and Germination

Water imbibition gradually increased from 2 to 12 hrs and was the highest in large seeds irrespective of the genotypes, ranging from 56.5% (Kranti) to 109.7% (Pusa Jaikisan) after 12 hours of seed soaking (Fig.1). The imbibition was 25.8(BPR 549-9)-62.3% (Pusa Jaikisan), 28.7 (BPR 549-9)-71.3% (Pusa Jaikisan), 37.0 (BPR 549-9)-88.4% (Pusa Jaikisan), 45.6 (Kranti)-105.0% (Pusa Jaikisan) and 48.0(Kranti)-107.9% (BPR 549-9), respectively, at 2, 4, 6, 8 and 10 hours of seed soaking. Among the groups, the range was 56.5 (Kranti)-93.1% (BPR 549-9), 78.5 (Pusa bold)-102.5% (BPR 549-9) and 89.1 (Pusa bold)-109.7% (Pusa Jaikisan, BPR 549-9), respectively, for small, medium and large seeds. The imbibition was 27.6 % higher in large seeds as compared to small seeds and the highest increase was recorded for the variety Kranti (58.9 %) at 12 hours of seed soaking. Pusa Jaikisan followed by BPR 549-9 showed the highest imbibition. The increased imbibition was also reflected in enhanced germination. The highest germination recorded for BPR 549-9 might be due to its better imbibition. The findings of the present investigation are in agreement with those earlier reports (Anonymous, 2008). There was no significant difference among the genotypes as well as different size of seeds for germination. The genotype BPR-548-9 showed maximum germination in all the groups varying from 94.5 to 97.2%. All the genotypes had > 90 % germination irrespective of the

Table 1. Effect of seed size on seed weight, oil, protein content, seedling growth and vigour index of Indian mustard

1000-seed weight (g)			Oil content (%)			Protein content (%)			Germination (%)			Shoot length (cm)		
Small	Medium	Large	Small	Medium	Large	Small	Medium	Large	Small	Medium	Large	Small	Medium	Large
1.75	4.46	6.54	39.0	42.6	43.8	18.3	19.6	19.8	94.5	96.0	97.2	3.70	2.70	2.20
1.49	3.40	4.93	39.0	41.7	41.7	20.1	20.3	20.5	92.1	93.3	95.0	3.73	3.07	2.90
2.47	4.68	6.34	38.3	40.8	41.3	19.3	19.5	19.9	93.5	96.6	94.7	4.20	4.10	3.60
2.62	4.97	6.52	36.4	39.0	39.0	22.3	21.2	21.1	92.7	91.1	95.2	4.30	3.63	3.10
2.94	4.10	5.91	38.8	40.1	400	20.3	20.7	21.3	90.8	92.2	94.7	4.20	3.90	3.00
	0.24			0.77			0.56			NS			0.56	
	0.19			0.59			NS			NS			0.44	
	0.42			NSa			0.97			NS			NS	
	Small 1.75 1.49 2.47 2.62	Small Medium 1.75 4.46 1.49 3.40 2.47 4.68 2.62 4.97 2.94 4.10 0.24 0.19	Small Medium Large 1.75 4.46 6.54 1.49 3.40 4.93 2.47 4.68 6.34 2.62 4.97 6.52 2.94 4.10 5.91 0.24 0.19	Small Medium Large Small 1.75 4.46 6.54 39.0 1.49 3.40 4.93 39.0 2.47 4.68 6.34 38.3 2.62 4.97 6.52 36.4 2.94 4.10 5.91 38.8 0.24 0.19 19 10	Small Medium Large Small Medium 1.75 4.46 6.54 39.0 42.6 1.49 3.40 4.93 39.0 41.7 2.47 4.68 6.34 38.3 40.8 2.62 4.97 6.52 36.4 39.0 2.94 4.10 5.91 38.8 40.1 0.24 0.77 0.19 0.59	Small Medium Large Small Medium Large 1.75 4.46 6.54 39.0 42.6 43.8 1.49 3.40 4.93 39.0 41.7 41.7 2.47 4.68 6.34 38.3 40.8 41.3 2.62 4.97 6.52 36.4 39.0 39.0 2.94 4.10 5.91 38.8 40.1 400 0.24 0.77 0.19 0.59 0.59	Small Medium Large Small Medium Large Small 1.75 4.46 6.54 39.0 42.6 43.8 18.3 1.49 3.40 4.93 39.0 41.7 41.7 20.1 2.47 4.68 6.34 38.3 40.8 41.3 19.3 2.62 4.97 6.52 36.4 39.0 39.0 22.3 2.94 4.10 5.91 38.8 40.1 400 20.3 0.24 0.77 0.19 0.59 0.59 0.59	Small Medium Large Small Medium Large Small Medium 1.75 4.46 6.54 39.0 42.6 43.8 18.3 19.6 1.49 3.40 4.93 39.0 41.7 41.7 20.1 20.3 2.47 4.68 6.34 38.3 40.8 41.3 19.3 19.5 2.62 4.97 6.52 36.4 39.0 39.0 22.3 21.2 2.94 4.10 5.91 38.8 40.1 400 20.3 20.7 0.24 0.77 0.56 0.19 0.59 NS	Small Medium Large Small Medium Large Small Medium Large 1.75 4.46 6.54 39.0 42.6 43.8 18.3 19.6 19.8 1.49 3.40 4.93 39.0 41.7 41.7 20.1 20.3 20.5 2.47 4.68 6.34 38.3 40.8 41.3 19.3 19.5 19.9 2.62 4.97 6.52 36.4 39.0 39.0 22.3 21.2 21.1 2.94 4.10 5.91 38.8 40.1 400 20.3 20.7 21.3 0.24 0.77 0.56 0.19 0.59 NS 14.3	Small Medium Large	Small Medium Large Small Medium Large Small Medium Large Small Medium 1.75 4.46 6.54 39.0 42.6 43.8 18.3 19.6 19.8 94.5 96.0 1.49 3.40 4.93 39.0 41.7 41.7 20.1 20.3 20.5 92.1 93.3 2.47 4.68 6.34 38.3 40.8 41.3 19.3 19.5 19.9 93.5 96.6 2.62 4.97 6.52 36.4 39.0 39.0 22.3 21.2 21.1 92.7 91.1 2.94 4.10 5.91 38.8 40.1 400 20.3 20.7 21.3 90.8 92.2 0.24 0.77 0.56 NS NS NS 0.19 0.59 NS NS NS	Small Medium Large Small Med	Small Medium Large	Small Medium Large Small Medium 1.49 3.40 4.93 39.0 41.7 41.7 20.1 20.3 20.5 92.1 93.3 95.0 3.73 3.07 2.47 4.68 6.34 38.3 40.8 41.3 19.3 19.5 19.9 93.5 96.6 94.7 4.20 4.10

Genotype	Root length (cm)			Dry weight / 5 seedlings (mg)			Root to shoot ratio			Vigour index (%)		
	Small	Medium	Large	Small	Medium	Large	Small	Medium	Large	Small	Medium	Large
BPR 549-9	12.30	13.20	14.20	12.30	20.47	29.23	3.37	4.97	6.67	1163.2	1971.2	2884.5
Kranti	10.20	10.30	11.20	11.53	17.60	18.67	2.73	3.43	3.87	1058.9	1643.0	1782.9
Pusa bold	11.27	12.90	13.80	11.83	18.87	28.10	2.80	3.13	3.83	1108.7	1828.1	2661.6
Pusa Jaikisan	10.00	10.30	11.70	15.63	20.83	29.60	2.33	3.57	3.90	1451.2	1899.6	2820.5
Varuna CD (P = 0.05)	9.57	10.20	11.60	11.13	16.50	22.43	2.70	3.40	3.93	1466.7	1520.6	2123.6
CD(P = 0.05)												
Genotypes (G)		1.44			1.48			0.77			179.5	
Groups (g)		1.11			1.14			0.60			139.0	
G x g interaction		NS			2.56			NS			310.9	

NSa: Non-significant

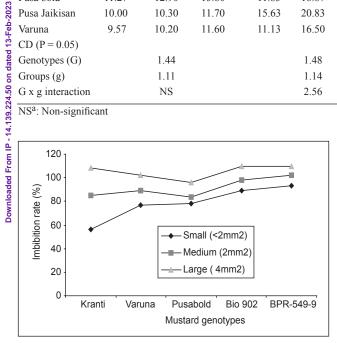


Fig. 1. Effect of seed size on water imbibition of different mustard genotypes

group. Dubey et al. (1989) and Kant and Tomar (1995) also reported reduced germination up to 8% of small seeds in Indian mustard

Seedling Growth

The seed size and shoot length appeared to have an inverse relationship as the shoot length was significantly reduced with increased seed size. The decrease in shoot length was about 25% for large seeds over that of small seed. In the small size group, all the varieties had similar shoot length (Table 1). The varieties Pusa Bold, Pusa Jaikisan and Varuna in the medium group and all the varieties under large seed group exhibited significant differences in shoot length. Root length increased by about 9.3 and 14% for the medium and large seeds over that of small seeds. Genotype BPR 549-9 had the maximum root length in all the three groups varying from 12.3-14.2 cm. Genotypes Pusa bold and BPR 549-9 had significantly longer roots than the other varieties in small, medium and large seed groups (Table 1). The increase in root length with the seed size suggested that the large seeds might result in better crop establishment in Indian mustard. Kant and Tomar (1995) also reported significant increase in root and non-significant differences in shoot length in Indian mustard. However, Dubey et al. (1989) reported significant increase in root and shoot length of large seeds over small seeds. Seedling dry weight differed significantly among the groups and showed an increase of 39.7 and 89.8% for medium and large over the small seeds, respectively. There was further increase of 35.9% in seedling dry weight of large

seeds over the medium ones. Pusa Jaikisan followed by BPR 549-9 had significantly higher seedling dry weight than other genotypes in all the three groups (Table 1). The increase in dry weight of the seedling could be due to increase in root length as well as more dry matter accumulation in shoot and roots per unit length. Increase in seed size was also reflected in enhanced root to shoot ratio. The mean root to shoot ratio was 2.79, 3.70 and 4.44, respectively, for small, medium and large seeds. Genotype BPR 549-9 had the highest root to shoot ratio in early seedling development might result in better crop establishment.

Seedling Vigour Index

Since seedling vigour index was the product of germination and seedling dry weight, hence followed the similar pattern of change as that of seedling dry weight. It differed significantly among the groups and showed an increase of 41.8 and 96.4% for medium and large over the small seeds, respectively. Seedling vigor of large seeds was higher by 38.5% over that the medium seeds. Seedling vigour was significantly higher for the genotype BPR 549-9 followed by Pusa Jaikisan than other genotypes in large seeds (Table 1). The increase in seedling vigour was primarily due to more seedling dry matter with increase in seed size, as the germination did not differ significantly among the genotypes and group of seeds.

Correlation analysis revealed that seed size had no relationship with seed germination, oil and protein content. But positive and significant association of seed size was observed with root length(r=0.655*), dry weight (r=0.894**), root to shoot ratio (r=0.879**) and vigour index (r=0.938**). Sedighi *et al.* (2011) also observed similar trend of correlation in safflower. The results of the present investigation revealed that although seed germination was not affected significantly by seed size but root length, seedling dry weight, root to shoot ratio and seedling vigour index enhanced with increase in seed size which would result in early and fast emergence of the crop especially under limited moisture conditions. Therefore, simple technique of seed grading could be valuable for the long-term conservation of the germplasm. However, there is a need to further study the effects of storage on germination and seedling development of different grades of seeds.

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