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# Impact of high temperature at pod development stage on yield and quality of *Brassica juncea* cultivars under controlled conditions

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ABSTRACT

A study was conducted to assess the impact of short and long periods of high temperature in yield and quality of Indian mustard (*Brassica juncea* L. czery & Coss.) High temperature during reproductive phase affects the yield and quality of cool season crops to a greater extent than summer crops. *Brassica juncea* cultivars 'Pusa Bold' and 'Pusa Agrani' were exposed to high day temperature (35°C) at pod development stage for 5 and 10 days to assess their impact on yield parameters and seed quality. Longer exposure to stress caused drastic yield reduction in 'Pusa Bold' and 'Pusa Agrani' by 54% and 69% respectively. 'Pusa Bold' showed 23% increase in seed weight/pod at brief period of stress without any change in seed number whereas 'Pusa Agrani' was adversely affected. Significant decrease in soluble starch and increase in crude protein content were observed at longer exposure to high temperature in 'Pusa Agrani' that was at the expense of oil synthesis in the seeds. Fatty acid composition showed an increase in percentage of monosaturated fatty acid and decrease in polyunsaturated fatty acid under longer period of stress. At longer stress period thousand seed weight and seedling vigor index of both the cultivars were adversely affected. The differential response observed in the 2 cultivars of *B. juncea* under exposure to different periods of stress exhibited the adaptive mechanism in 'Pusa Bold' as it is a long duration cultivar and brief exposure to high temperature is compensated by maintaining net photosynthetic rate and by increased translocation to the developing seed. The shorter duration of seed development in 'Pusa Agrani' would accentuate the differences between the cultivars with varying rates of accumulation of seed reserves.

**Key words**: *Brassica juncea*, High temperature, Oil content, Per cent fatty acid Photosynthesis, Photosystem II efficiency, Seed quality, Vigor index, Yield

Concern over the climate change has motivated agriculture scientists worldwide to address the problem at different levels. Besides increase in level of atmospheric carbon dioxide, increase in incidence of short spells of high and low temperatures are the major effects of climate change. Recent studies have indicated the probability of 10–40% loss in crop production in India with increase in temperature by 2080–2100 (IPCC 2007). Physiological changes due to high temperature occur at all levels of structural organization that include changes in tissues and cell organelles, disorganization of cell membranes, impedance of photosynthesis via photochemical and biochemical reactions of the photosynthesis machinery (Wise *et al*. 2004). Plants respond to high temperatures through various adaptive mechanisms, such as synthesis of ion transporters, osmoprotectants, free radical scavengers, heat shock proteins, factors involved in signaling cascades and transcriptional control (Wang *et al*. 2004).

In northern India, yield reduction of irrigated mustard was comparatively less due to prevailing lower temperature in this region during the crop growth period (Boomiraj *et al*. 2010). Different phenological stages differ in their response to high temperature and this depends on species and genotype (Gunasekera *et al*. 2006). Plants stressed to high temperature at flowering stage show recovery from stress than plants stressed at pod development stage (Gan *et al.* 2004). This inability to recover is due to the detrimental effect of high temperature to seed development and results in reduced seed weight. Information on effect of temperature rise on seed yield is available but its effect on seed composition and oil quality is scarce. Oil is deposited late during seed development and high temperature exposure during that time results in a decrease in oil content. Fatty acid composition is also influenced with warmer conditions favouring production of saturated fatty acids, while cooler moist conditions favour

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the production of polyunsaturated fatty acids (Seiler 1983). Therefore, a study was conducted to assess the impact of short and long periods of high temperature on yield and quality of 2 Indian mustard (*Brassica juncea* L. czerng & Coss.) cultivars, 'Pusa Agrani' (short duration) and 'Pusa Bold' (long duration). The temperature exposure was given at pod development two stages, viz short exposure of 5 days and long exposure of 10 days. This would indicate the adaptive capacity of the cultivar to high temperature stress. Seed composition and quality were also determined to observe the impact of elevated temperature on seed quality.

## MATERIALS AND METHODS

Seeds of mustard (*B. juncea*) cultivars, 'Pusa Agrani' and 'Pusa Bold' were sown in pots filled with 10 kg soil of 3:1 mixture of sandy loam to farmyard manure at Indian Agricultural Research Institute, New Delhi, India (28° 4′, 77° 9′E and elevation 228 m above mean sea level). NPK equivalent to 60:30:30 kg/ha was supplied in 4 equal split doses. The plants were watered at weekly interval and thinned to 3 plants/pot at 3-leaf stage. There were 5 replications/treatment. The pots were transferred at pod stage to the growth chamber maintained at high temperature. The high temperature (35°C) was given as 2 treatments, viz 5 and 10 days exposure. The temperature increase in the growth chamber was achieved by ramping the temperature at 3°C from 23–35°C over 4 hr and maintaining at 35°C for 3 hr and then ramping back to 23°C for the remaining 5 hr. Night temperature was maintained constant at 20°C for 12 hr. Day/night cycles for temperature and photo period were 12 hr day and 12 hr night. The control plants were kept under ambient temperature conditions of 25/20°C in another growth chamber. All plants were maintained at 70±5% relative humidity and photosynthetically active radiation (PAR) of 300 µmol/m$^2$/sec. After the treatment the treated plants were removed from the chamber along with the control plants and kept in greenhouse till maturity.

Net photosynthetic rate was taken on the youngest fully expanded leaf using portable IRGA (LI 6400F, LICOR, USA) a day after removal of the plants from the growth chambers. The efficiency of photosystem II (ΦPS II) was measured by the same instrument in light adapted leaves by giving a light pulse of 500 µmol/m$^2$/s for 0.8s. All the observations were taken on 3 randomly selected plants from 3 pots of control and stress. The experiment was terminated when the pods reached maturity (brown to tan colour). Observations war taken at harvest included pods/plant, pod length, seeds/pod, seed weight/pod, and yield/plant. Dry matter was determined after drying the leaves and stems for 72 hr at 60°C in an oven. Seed filling rate/day was calculated from the yield and days to maturity from pod initiation stage. Seed and nutritional quality was determined in the harvested seed to assess the effect of high temperature on quality of the produce. Seed oil content was measured using 16 MHz pulsed Nuclear Magnetic Resonance Spectrometer. Fatty acid composition was analyzed using gas chromatography by transesterification of oil to fatty acid methyl esters (Clarus 500, Perkin Elmer, USA). Esters of fatty acids were identified by comparing the retention time with standard esters. Each fatty acid was expressed as a per cent of the total fatty acids. Total soluble sugars and starch were measured by anthrone method (Mc Cready *et al.* 1950). Nitrogen was estimated by modified Kjeldahl method using a N autoanalyser and crude protein percentage was calculated by N×5.3. Seed quality was evaluated by germination ability and vigor of the seedling. Seed germination test was conducted by placing 50 seeds in 3 replicates between paper towels. Seedling vigour index was calculated as percentage of germination×total seedling dry weight (Abdul- Baki and Anderson 1973). Statistical analysis was performed using MSTAT C software package. Differences between means were evaluated using LSD test at $P \leq 0.05$.

## RESULTS AND DISCUSSION

Severity of yield loss was higher in plants exposed to 10 days of high temperature with 'Pusa Bold' showing 54% and 'Pusa Agrani' 69% reduction compared to the control plants whereas exposure for a short period of 5 days reduced seed yield by 14 and 31% in 'Pusa Bold' and 'Pusa Agrani' respectively (Table 1). Yield loss of 77% was observed in mustard plants exposed to high temperature during pod development stage but the plants had the ability to recover when stress was given at a stage earlier than this (Gan *et al*. 2004). The maturity dates of plants stressed for 10 days were advanced by 9 days in 'Pusa Agrani' and 7 days in 'Pusa Bold'. The plants stressed for 5 days matured only two days earlier than the control plants in both the cultivars. Dry matter production and plant height reduced significantly in both the cultivars under long period of high temperature (Table 1). Lower biomass with shorter and thinner stems was also observed in *B. napus* plants grown under elevated temperature (Qaderi *et al.* 2006). In general, yield reduction due to heat stress at the pod development stage may result from reduced pods/plant, seeds/pod and seed weight/pod indicating that seed setting and filling of the latter formed seeds are affected. In our experiment, exposure to high temperature for 10 days reduced pod number by 34 and 47%, whereas the reduction in seeds/pod was 19 and 27% in 'Pusa Bold' and in 'Pusa Agrani' respectively (Table 1). The latter formed flowers were subjected to high temperatures for a long period of 10 days within the growth chamber that may have resulted in floral sterility and thus reduction in pod number/plant. Our findings corroborate with the results of Morrison and Stewart (2002). A short period of stress enhanced pod production in 'Pusa agrani' by 8% compared to control plants (Table 1). The ability of plants to produce more pods on branches after removal from high temperature was responsible for the increase in total number of pods. However, the increase in pods/plant in

Table 1 Effect of different period of high temperature at pod development stage on growth, yield and seed composition of *Brassica juncea* cultivars

| Crop | Plant height (cm) | Biomass (g) | Yield/ plant (g) | Pod length (cm) | Pods/ plant | Seeds/ pod | Seed weight/ pod (mg) | 1000 seed weight (g) | Oil (%) | Starch (%) | Sugar (%) | Crude protein (%) |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| *'Pusa Agrani'* | | | | | | | | | | | | |
| Ambient | 99.2 | 8.59 | 2.79 | 5.93 | 62.7 | 11.0 | 436.7 | 2.08 | 37.4 | 9.8 | 1.55 | 20.8 |
| 5 day stress | 97.5 | 8.26 | 1.92 | 5.13 | 67.7 | 10.0 | 290.0 | 1.62 | 36.1 | 7.5 | 1.22 | 22.9 |
| 10 day stress | 87.5 | 5.97 | 0.86 | 5.97 | 33.0 | 8.0 | 240.0 | 1.4 | 32.7 | 7.8 | 1.28 | 25.9 |
| *'Pusa Bold'* | | | | | | | | | | | | |
| Ambient | 125.1 | 10.90 | 2.99 | 7.36 | 64.0 | 12.0 | 466.7 | 1.66 | 34.5 | 6.4 | 0.89 | 17.1 |
| 5 day stress | 109.8 | 6.39 | 2.56 | 7.11 | 43.4 | 12.3 | 573.3 | 2.01 | 37.5 | 7.4 | 1.38 | 18.5 |
| 10 day stress | 107.4 | 5.73 | 1.38 | 6.70 | 42.2 | 9.7 | 326.7 | 1.39 | 34.2 | 5.5 | 0.67 | 20.3 |
| LSD | | | | | | | | | | | | |
| ($P$<0.05) | 7.15 | NS | 0.35 | 0.90 | NS | 0.99 | 101.2 | NS | NS | 1.3 | 0.4 | 1.1 |
| Variety (V) | 8.76 | 1.65 | 0.43 | NS | 11.8 | 1.22 | 123.9 | 0.22 | NS | 1.6 | 0.5 | 1.4 |
| Treatment (T) | NS | 2.34 | NS | NS | 16.6 | 1.73 | NS | 0.31 | 1.7 | 2.2 | 0.8 | 2.0 |
| V×T | | | | | | | | | | | | |

'Pusa Agrani' was not reflected in seed weight/plant as a shorter period of stress reduced seed weight/pod (Table 1). *B. juncea* gave significantly higher pods/plant when stressed at 28°/18 °C and 35°/18 °C (Gan *et al*. 2004) that may contribute to improved yield under stress conditions. Seed filling rate showed that 'Pusa Bold' had a higher rate of grain filling (44.9 mg/plant/day) than 'Pusa Agrani' (36.9 mg/plant/day) at short exposure to high temperature. There was a 23% increase in seed weight/pod without a change in seed number in 'Pusa Bold' (Table 1). This increased mobilization efficiency of reserves may act as a potential strategy by 'Pusa Bold' to improve grain filling and yield (23% increase in seed weight/pod and 21% in 1000-seed weight) under shorter exposure to high temperature. The grain filling rate was reduced to 26.5 mg/plant/day in 'Pusa Bold' and 19.1 mg/plant/day in 'Pusa Agrani' under long period of stress resulting in pods with fewer seeds of lesser weight.

Significant reduction in net photosynthetic rate (Pn) was observed under prolonged period of heat stress in both the cultivars (Fig 1). 'Pusa Agrani' showed reduced Pn even after a short period of stress with 8% decline compared to control (Fig 1). High temperature decreased the efficiency of photosystem II compared to ambient temperature in 'Pusa Bold' (Fig 2). Photostem II is highly thermolabile and its efficiency is greatly reduced or partially stopped under high temperature (Qaderi *et al*. 2006). Despite low photostem II efficiency net photosynthetic rate was maintained in 'Pusa Bold' at short period of stress. On the other hand, 'Pusa Agrani' was affected more in carbon fixation capacity at high temperature. This may be attributed to Rubisco deactivation under increased temperature (Crafts-Brandner and Savucci 2002). This shows that the short duration variety 'Pusa agrani' is more sensitive to elevated temperature that causes irreversible losses by limiting photosynthates to the developing seed, whereas the long duration cultivar 'Pusa Bold' compensates the loss by increasing its seed filling rate.

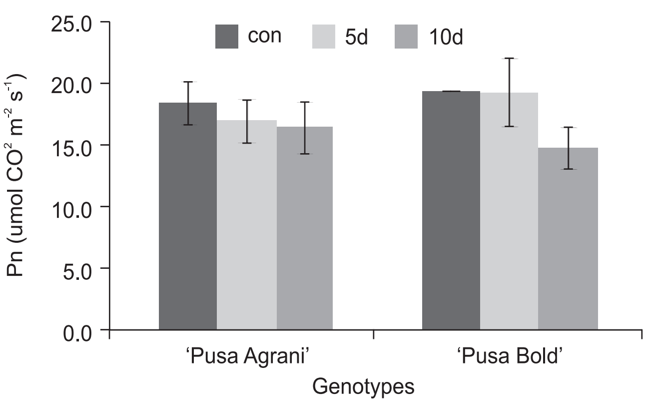


Fig 1 Net photosynthetic rate in *B. juncea* cultivars grown under high temperature for different periods of time. LSD ($P$< 0.05) Varieties=NS, Treatment=2.34, V×T= NS

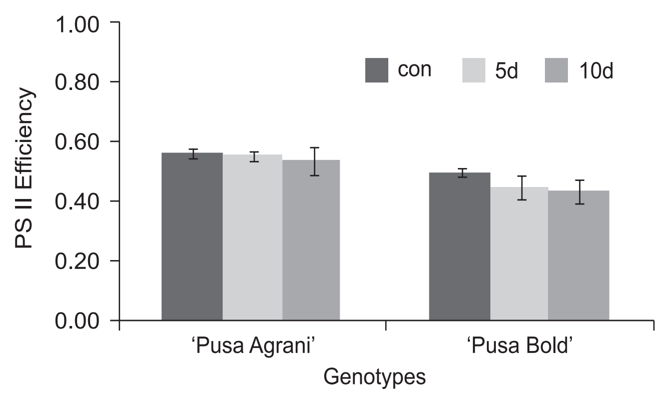


Fig 2 Efficiency of photosystem II in *B. juncea* cultivars grown under high temperature for different periods of time. LSD ($P$< 0.05) Varieties=0.032, Treatment=NS, V×T= NS

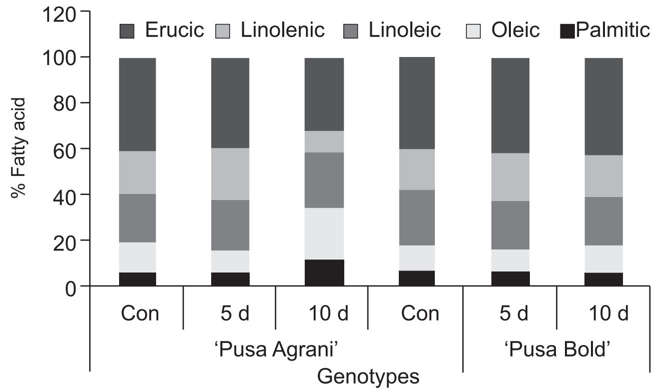


Fig 3 Effect of high temperature for different periods on fatty acid composition of *Brassica* cultivars. Each fatty acid is represented as percentage of total fatty acids

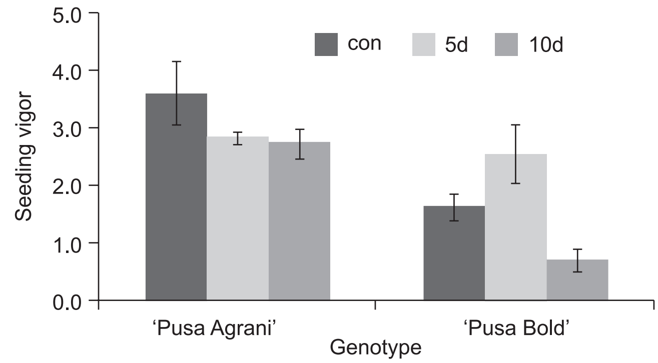


Fig 4 Effect of high temperature for different periods on seedling vigor in *Brassica* cultivars. LSD ($P< 0.05$) Varieties=0.359, Treatment=0.439, V×T= 0.621.

Analysis of the composition of seed harvested from the different treatments showed that the decrease in carbohydrate levels with increase in temperature (Table 1) is due to limitation of assimilate availability or transfer into seed under elevated temperature. At brief period of stress 1% increase in starch percentage was observed in 'Pusa Bold' (Table 1). Studies on biochemical mechanisms underlying the effect of temperature on starch deposition in other crops have shown the inactivation of soluble starch synthase (Chaitanya *et al*. 2001). In our studies also, the temperature instability of soluble starer synthase might have resulted in decrease of starch accumulation under long period of stress. Crude protein content increased by 3% in 'Pusa Bold' and 5% in 'Pusa Agrani' under long period of stress (Table 1). Under heat and drought stress, the natural reaction of the crop is to store less carbohydrate and more protein. In wheat, due to reduced photosynthesis and grain growth at high temperature, rate of protein synthesis is promoted and protein concentration increases (Blumenthal *et al*. 1991). An increase in protein concentration of 5% in 'Pusa Agrani' was accompanied by 5% decrease in oil content when exposed to longer period of high temperature (Table 1). The shorter duration variety 'P. agrani' showed greater increase in the protein content that is at the cost of oil synthesis in the seed. Adverse affects of high temperature on oil concentration during reproductive period of canola in Mediterranean type environments has been cited by many workers (Gunasekera *et al.* 2006). Quality of oil was also affected with elevated temperature with an increase in monounsaturated fatty acid (C18:1) and decrease in polyunsaturated fatty acid (C18: 3) in 'Pusa Agrani' under long period of stress. Erucic acid percentage also decreased in this cultivar (Fig 3). This pattern of increase in percentage of monounsaturated fatty acid (oleic acid) and decrease in polyunsaturated fatty acids (linoelenic acid) due to increase in temperature has been observed in mustard (Triboi-Blondel and Renard 1999). High temperature stimulated the biosynthesis of C18:1 and inhibited its desaturation in both the *B. juncea* cultivars exposed to long duration of high temperature. This suggests that C 18:2 desaturase is affected more at high temperature than other desaturase enzymes. The lower level of erucic acid in 'Pusa Agrani' may result from the effect of high temperature on enzyme elongase, that catalyses the elongation of fatty acid chain from 18:1 to 22:1. High temperature for a shorter period increased the level of polyunsaturates without affecting saturates.

Germination of harvested seeds was unaffected by temperature stress treatments (data not shown). Seedling vigor on dry weight basis showed a reduction in 'Pusa Agrani' but there was an improvement in 'Pusa Bold' under short period of stress. This could be a result of higher accumulation of photosynthates that are metabolized in the germinating seed and that later help in establishment of healthier seedlings. There was a drastic reduction in vigor index by 55% in 'Pusa Bold' and 22% in Pusa Agrani compared to control under longer period of stress (Fig 4) due to poor seed development and storage reserves.

Thus, this study showed the varied impact of short and long period of exposure of high temperature on *B. juncea* cultivars. 'Pusa Bold' a long duration cultivar responded positively to high temperature given for a brief period as seed weight/pod increased due to lesser competition between the pods and branches. Seed quality parameters like thousand seed weight, oil content and seedling vigor also improved. In the short duration cultivar 'Pusa Agrani' lower yields at short period of high temperature were attributed to reduced filling as a result of increased competition between pods for photoassimilates. Seed number/pod was reduced that shows that high temperature affects seed setting as well as seed filling in this cultivar. At longer period of high temperature stress both the cultivars showed similar response and were adversely affected in yield components and seed quality traits.


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