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# Phenology of Indian mustard (*B. juncea*) as affected by sowing dates and microbes

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## Abstract

The present study investigated the role of seeds dressed with microbial cultures sown at two dates on phenological traits in mustard varieties. Field experiment was conducted with three mustard varieties (Giriraj, PBR 357 and RH 725) sown at two dates with microbial treatments (Pusa Sanjeevani, Microbial formulation and control/no culture) in completely randomised block design (RBD) during two *rabi* seasons 2021-22 and 2022-23. Planting time and microbial treatments had a significant influence on phenology of the three varieties. Timely sown crop showed early initiation and completion of flowering and siliquae formation whereas, days for flowering duration, siliquing duration, reproductive phase and physiological maturity were extended over late-sown crops. Control with no microbial inoculum took more days for initiation i.e. 10.4 for flowering and 12.7 days for siliquing. However, days taken to completion of flowering and fruiting were more 8.6 and 9.2 days, respectively. Delayed planting resulted in shorter durations of flowering by 1.8 days and siliquing by 3.5 days leading to shortened reproductive phase with forced maturity w.r.t untreated control. Seed yield varied significantly under both sowing conditions. Under late planting, yield was reduced by 10.8% in control/ with no microbial inoculum, 5.5% with Microbial formulation and 5.4% with Pusa Sanjeevani. Seed yield had positive and significant correlation with phenological traits under both the sowing conditions.

**Keywords:** Heat Stress; Microbial Treatments; Sowing Conditions; Seed Yield

## 1. Introduction

Mustard (*Brassica juncea* L.), an important *rabi* season crop, belongs to the family Brassicaceae. India ranks as the third-largest producer of rapeseed and mustard globally, contributing 11.3% to the total production, following Canada and China (Jat et. al, 2019). In India, rapeseed and mustard are the third most significant sources of edible oil, after soybean and groundnut. According to the IPCC report, global temperatures are expected to rise by 1.5°C between 2021 and 2040 due to changing climatic conditions (Seneviratne et. al, 2023). Abiotic stress causes more than 50% of yield losses which affect both the quantity and quality of crop production (Salam et. al, 2023). Heat stress is the most predominant abiotic stress that severely impact the seed germination, growth, development, flowering, branching, seed weight and overall yield (Sharma et. al, 2022). Heat stress during flowering and grain filling stage accelerated the fruiting and siliquing development, leading to shorter reproductive phase and early maturity which ultimately affected the crop yield (Djanaguiraman et. al, 2020). Consequently, heat stress not only reduces crop yields but also compromises the nutritional and economic value of the harvested seeds.

Rapeseed-mustard holds crucial significance as an oilseed crop in India, cultivated for its productivity and its ability to withstand various stress conditions such as drought, heat, low light and salinity. Plants exhibit a range of morpho-physiological and biochemical changes and adopt various mitigation strategies to withstand abiotic stresses (Singh et. al, 2023). A promising and cost-effective approach to manage abiotic stresses involves utilizing beneficial microorganisms, such as plant growth-promoting rhizobacteria (PGPR), nitrogen-fixing rhizobia and arbuscular

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mycorrhizal fungi (AMF) to enhance tolerance to drought and heat stress (Alori et. al, 2017). Microorganisms have remarkable characteristics including their resilience to extreme conditions, widespread presence and genetic diversity. Their interactions with crop plants offer valuable opportunities and the development of innovative strategies for effectively using these microorganisms individually or as consortium holds great potential for enhancing production and sustainability (Sagar et. al, 2023). The study aims to explore the effect of microbial inoculations on phenological traits of mustard under heat stress.

## 2. Material and methods

### 2.1. Experimental site

The experiment was conducted in the fields of Oilseeds section, Department of Plant Breeding and Genetics, Punjab Agricultural University, Ludhiana during 2021-22 and 2022-23 (30° 54' N latitude and 75° 48' E longitude and at an altitude of 247 meters above the mean sea level)

### 2.2. Planting material and experimental details

Seeds of released mustard varieties (Giriraj, PBR 375 and RH 725) from different agroclimatic zones of India were procured from Oilseeds section, Department of Plant Breeding and Genetics, Punjab Agricultural University, Ludhiana and were treated with Pusa Sanjeevni from IARI, New Delhi and Microbial formulation from CSIR, Lucknow as per their guidelines and sown under two dates: in October as timely sown (TS) and in November as late sown (LS) (Fig. 1).

### 2.3. Inoculation of seeds

10% of jaggery solution was used as an adhesive for coating the microbial cultures onto seeds. The known volume of jaggery solution along with the microbial inoculum were applied to the seeds. The coated seeds were uniformly spread on the polythene sheet and dried in the shade.

Each inoculated variety was planted in five rows, with a row length of 3 m at 30 cm row spacing in randomised block design (RBD) with three replicates. All recommended package of practices were followed to raise a healthy crop. Phenological traits comprising flowering initiation (one fully developed flower was appeared in all plots for each treatment), flowering completion (100% flowering in all plots within each treatment), flowering duration (period from flower initiation to completion), siliquae initiation (at least one flower in each plot was developed to siliqua), siliquae completion (100% siliquing in all plants of plots), siliquae duration (period from siliquae initiation to completion), reproductive phase (period from flowering initiation to siliquae completion) and physiological maturity (aerial plant parts turned light yellow to brown, siliquae to lemon yellow colour and seeds from brown to brown-black) were recorded. Correlation and linear regression analysis were performed to work out the relationship between phenological traits and seed yield.

### 2.4. Statistical Analysis

Statistical significance ($p \leq 0.05$) was tested using the CPCS1 (2008) software for RBD. Pearson's correlation and linear regression graphs between phenological traits and seed yield were generated using Minitab statistical software 22.

### 2.5. Temperature variations

The climate in Ludhiana, Punjab, is subtropical and semi-arid and experiences hot and dry summers from April to June, humid summers from July to September and cold winters during December and January. During the summer, temperatures often exceed 38°C and occasionally spike to 40-45°C during periods of drought in May and June. During December and January, temperatures typically drop below 0.5°C, accompanied by occasional frosty spells. In timely sown crop maximum temperature varied from 34.8 to 42.0 °C in both cropping years, whereas late sown crop experienced 32.8 to 38.0 °C. Temperature variations existed during the two consecutive cropping season (Table 1).

**Table 1** Temperature variations at different growth stages of mustard during two crop season (2021-23)

| **Temperature (°C)** | | | | | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Year/stages** | **Vegetative stage** | | | **Flowering stage** | | | **Siliquing stage** | | | **Reproductive phase** | | | **Physiological maturity** | | | **Harvesting** | | |
| | **Timely sown** | | | | | | | | | | | | | | | | | |
| | Max | Min | Average | Max | Min | Average | Max | Min | Average | Max | Min | Average | Max | Min | Average | Max | Min | Average |
| 2021-22 | 20.8 | 6.4 | 13.6 | 14.5 | 8.0 | 11.3 | 17.1 | 8.0 | 12.6 | 16.1 | 8.2 | 12.2 | 30.8 | 15.4 | 23.1 | 42.0 | 24.2 | 33.1 |
| 2022-23 | 15.0 | 9.4 | 12.2 | 20.3 | 8.3 | 14.3 | 20.8 | 8.7 | 14.8 | 20.6 | 8.4 | 14.5 | 32.0 | 17.2 | 24.6 | 34.8 | 20.0 | 27.4 |
| | Late sown | | | | | | | | | | | | | | | | | |
| 2021-22 | 17.2 | 8.6 | 12.9 | 18.1 | 7.5 | 12.8 | 20.8 | 7.9 | 14.4 | 20.0 | 7.5 | 13.8 | 32.0 | 19.0 | 25.5 | 38.0 | 29.0 | 33.5 |
| 2022-23 | 16.0 | 9.0 | 12.5 | 20.9 | 7.2 | 14.1 | 22.1 | 8.6 | 15.4 | 21.7 | 7.7 | 14.7 | 28.6 | 14.0 | 21.3 | 32.8 | 20.8 | 26.8 |


**Figure 1** Timely and late sown crop at flowering stage

## 3. Results and discussion

### 3.1. Flowering Behaviour

Sowing dates and microbial treatments had significant effect on flowering behaviour comprising flowering initiation, completion and duration among the varieties as indicated by pooled mean of two-years (Table 2).

#### *3.1.1. Flowering initiation*

Days taken for flowering initiation was delayed in the treated varieties under late planting over normal sown crop. In the control/with no microbial treatment, initiation was early by 10.7 days in Giriraj, 9.6 days in PBR 357 and 10.8 days in RH 725. Varieties treated with Microbial formulation showed early initiation as compared to the control at two sowing dates. Flowering initiation was early by 8.4 days in Giriraj, 7.9 days in PBR 357 and 14.0 days in RH 725. However, with Pusa Sanjeevani the days taken were 9.1 (Giriraj), 9.8 (PBR 357) and 10.4 days (RH 725). Overall, initiation was delayed by 14 days with Microbial formulation and 10 days with Pusa Sanjeevani in RH 725 under late sown conditions over timely sown. On an average, days to flowering initiation declined gradually with Microbial formulation and Pusa Sanjeevani except for PBR 357.

#### *3.1.2. Flowering completion*

Flowering completion was delayed in the treated varieties under late planting over normal sown. In the control/with no microbial treatment, flowering completion was early by 8.5, 7.5 and 9.7 days in Giriraj, PBR 357 and RH 725, respectively. Varieties treated with Microbial formulation and Pusa Sanjeevani showed early completion as compared to the control at two sowing dates. Flowering completion was early by 5.6 days in Giriraj, 4.8 days in PBR 357 and 11.5 days in RH 725 treated with Microbial formulation. However, with Pusa Sanjeevani the days taken were 6.8 (Giriraj), 6.0 (PBR 357) and 7.6 (RH 725). Overall, completion was delayed by 11.5 days with Microbial formulation and 7.6 days with Pusa Sanjeevani in RH 725 under late sown conditions over timely sown. On an average, days to flowering completion declined gradually with Microbial formulation and Pusa Sanjeevani except for PBR 357.

#### *3.1.3. Flowering duration*

Flowering duration was longer in the varieties under normal planting than the late sown crop. In the control/uninoculated treatment, flowering duration (days) was more in normal sown varieties as compared when sown late by 2.1 days in Giriraj and PBR 357 and 1.1 days in RH 725. Varieties treated with microbial treatments showed longer duration as compared to the control at both sowing dates. However, with Microbial formulation, days taken for flowering duration were 2.8 days in Giriraj, 3.1 days in PBR 357 and 2.4 days in RH 725. Whereas, with Pusa Sanjeevani the days taken were 1.7 (Giriraj), 3.7 (PBR 357) and 2.8 days (RH 725). Overall, flowering duration was enhanced by 3.1 and 3.7 days in PBR 357 variety treated with Microbial formulation and Pusa Sanjeevani under late sown conditions over timely sown. On an average, days to flowering duration increased gradually with both the microbial inoculations.

### 3.2. Fruiting Behaviour

Sowing dates and microbial treatments had significant effect on fruiting behaviour comprising siliquing initiation, completion and duration among the three varieties as indicated by pooled mean of two-years (Table 3).

#### *3.2.1. Siliquing initiation*

Siliquing initiation was delayed in the treated varieties under late planting over normal sown. In untreated control, initiation was early by 14.7 days in Giriraj, 9.6 days in PBR 357 and 13.7 days in RH 725. Varieties treated with Microbial formulation showed early initiation with treatments as compared to control at both the sowing dates. Siliquing initiation was early by 9.5, 6.0 and 15.8 days in Giriraj, PBR 357 and RH 725, respectively. However, with Pusa Sanjeevani the days taken were 12.8 (Giriraj), 7.8 (PBR 357) and 12.0 days (RH 725). Overall, initiation was delayed by 15.8 days with Microbial formulation in RH 725 and 12.8 days with Pusa Sanjeevani in Giriraj under late sown conditions over timely sown. On an average, early siliquae initiation was recorded with both inoculations except for Pusa Sanjeevani in PBR 357 with respect to respective control.

#### *3.2.2. Siliquing completion*

Delayed siliquing completion was witnessed in the treated varieties under late planting over normal one. In the control/with no microbial treatment, siliquing completion was early by 9.2 days in Giriraj, 7.3 days in PBR 357 and 11.1 days in RH 725. Varieties treated with Microbial formulation and Pusa Sanjeevani showed early completion as compared to the control at two sowing dates. Siliquing completion was early by 4.5, 3.8 and 9.5 days in Giriraj, PBR 357 and RH

725 varieties treated with Microbial formulation. However, with Pusa Sanjeevani the days taken were 7.0 (Giriraj), 5.0 (PBR 357) and 7.9 days (RH 725), respectively. Completion was delayed in RH 725 when planted late by 9.5 days with Microbial formulation and 7.9 days with Pusa Sanjeevani. Average of siliquae completion indicated that varieties treated with the two microbial inoculations showed early completion with respect to the control varieties.

*3.2.3. Siliquing duration*

Siliquing duration was longer in treated varieties in the normal sown than the late sown varieties. In the control/with no microbial treatment, siliquing duration was 5.5 days in Giriraj, 2.3 days in PBR 357 and 2.6 days in RH 725. Varieties treated with microbial treatments showed improved duration as compared to the control at both sowing dates. However, with Microbial formulation, siliquing duration (days) was 5.0 in Giriraj, 2.2 in PBR 357 and 6.3 in RH 725. Whereas, with Pusa Sanjeevani the days taken were 5.8 (Giriraj), 2.8 (PBR 357) and 4.1 days (RH 725). Overall, fruiting duration was extended by 6.3 days in RH 725 treated with Microbial formulation while 4.1 days in PBR 357 treated with Pusa Sanjeevani under late sown conditions over timely sown. Days for fruiting durations were enhanced by both the microbial inoculations in all the 3 varieties however Microbial formulation was more effective in variety PBR 357.

## 3.3. Reproductive phase and Physiological maturity

Significant variation existed for reproductive phase and days to maturity among the varieties with sowing dates and microbial treatments as indicated by pooled mean of two-years (Table 4).

*3.3.1. Reproductive phase*

Reproductive phase was shortened in all treated varieties under late sown as compared to timely sown. In the control/with no microbial treatment, reproductive phase was shortened by 2.9 days in Giriraj, 3.5 days in PBR 357 and 1.0 days in RH 725 when sown late. Microbial treatments in the varieties showed longer reproductive phase as compared to the control at both dates of sowing. However, with Microbial formulation, reproductive phase was enhanced by 3.9 days in Giriraj, 4.1 days in PBR 357 and 4.5 days in RH 725 while, with Pusa Sanjeevani reproductive phase improved by 1.7 (Giriraj), 4.7 (PBR 357) and 3.1 days (RH 725). Reproductive phase was more than 25 days in the 3 varieties under timely sown conditions while under late sown conditions reproductive phase was comparable in Giriraj and PBR 357. Reproductive phase did not vary in RH 725 at both the planting dates with no-inoculation/control. On an average, reproductive phase was improved with the microbial inoculations in all the varieties under present investigation by more than 4.5 days. Overall, with Microbial formulation improved reproductive phase in variety RH 725 and Pusa Sanjeevani in PBR 357 under late planting with respect to normal sown cultures.

*3.3.2. Physiological maturity*

In the control, varieties took more days to attain maturity while early maturity was recorded in the late planted varieties. In untreated control, days taken for attaining physiological maturity were lesser by 5.8 days in Giriraj, 8.2 days in PBR 357 and 11.3 days in RH 725. Microbial formulation treated varieties took lesser days i.e. 12.4 days in Giriraj, 15.8 days in PBR 357 and 14.0 days in RH 725, whereas, with Pusa Sanjeevani maturity was early by 5.0, 6.5 and 15.1 days in Giriraj, PBR 357 and RH 725 respectively with respect to timely sown inoculated varieties. Overall, Giriraj matured early by 12.4 days with Microbial formulation and 5.0 days with Pusa Sanjeevani under late sown conditions over timely sown. The average number of days to attain maturity however increased with microbial inoculations.

**Table 2** Influence of sowing dates and cultures on flowering initiation, completion and duration in mustard varieties based on two years pooled data

| Cultures/ Varieties | Flowering initiation (days) | | | Flowering completion (days) | | | Flowering duration (days) | | |
|---|---|---|---|---|---|---|---|---|---|
| | No culture | | | No culture | | | No culture | | |
| | Timely sown | Late sown | Average | Timely sown | Late sown | Average | Timely sown | Late sown | Average |
| Giriraj | 68.3±4.5 | 79.0±1.3 | 73.7±2.9 | 86.7±5.7 | 95.2±1.2 | 90.9±3.4 | 18.3±1.5 | 16.2±0.3 | 17.2±0.8 |
| PBR 357 | 68.4±4.0 | 78.0±4.2 | 73.2±4.1 | 84.8±4.8 | 92.3±2.5 | 88.5±3.7 | 16.4±1.2 | 14.3±1.8 | 15.3±1.5 |
| RH 725 | 67.0±3.8 | 77.8±3.5 | 72.4±3.7 | 82.7±5.5 | 92.4±4.3 | 87.5±4.9 | 15.7±1.7 | 14.6±0.8 | 15.1±1.3 |
| Mean | 67.9±4.1 | 78.3±3.0 | | 84.7±5.3 | 93.3±2.7 | | 16.8±1.4 | 15.0±1.0 | |
| | **Microbial formulation** | | | **Microbial formulation** | | | **Microbial formulation** | | |
| Giriraj | 59.8±3.8 | 68.2±0.2 | 64.0±2.0 | 81.2±2.5 | 86.8±0.5 | 84.0±1.5 | 21.4±1.3 | 18.6±0.3 | 20.0±0.8 |
| PBR 357 | 58.4±2.0 | 66.3±5.0 | 62.5±3.5 | 78.5±0.3 | 83.3±4.3 | 80.9±2.3 | 20.1±2.3 | 17.0±0.7 | 18.5±1.5 |
| RH 725 | 57.0±1.7 | 71.0±0.5 | 64.0±1.1 | 76.2±1.5 | 87.7±1.0 | 81.9±1.3 | 19.1±3.2 | 16.7±1.5 | 17.9±2.3 |
| Mean | 58.3±2.5 | 68.5±1.9 | | 78.6±1.4 | 85.9±1.9 | | 20.2±2.3 | 17.4±0.8 | |
| | **Pusa Sanjeevani** | | | **Pusa Sanjeevani** | | | **Pusa Sanjeevani** | | |
| Giriraj | 57.9±2.3 | 67.0±4.8 | 62.5±3.6 | 79.0±3.8 | 85.8±3.7 | 82.4±3.8 | 21.1±1.5 | 19.4±1.2 | 20.2±1.3 |
| PBR 357 | 59.7±1.5 | 69.5±4.0 | 64.6±2.8 | 82.3±3.5 | 88.3±2.8 | 85.3±3.2 | 22.6±2.0 | 18.9±1.2 | 20.7±1.6 |
| RH 725 | 58.1±1.0 | 68.5±1.2 | 63.3±1.1 | 78.7±6.0 | 86.3±2.8 | 82.5±4.4 | 20.6±5.0 | 17.8±1.7 | 19.2±3.3 |
| Mean | 58.6±1.6 | 68.3±3.3 | | 80.0±4.4 | 86.8±3.1 | | 21.4±2.8 | 18.7±1.3 | |
| CD ($p \le 0.05$) | Y=0.265 V=0.325 C=0.325 Y×V=0.459 Y×C=0.459 V×C=0.562 E=0.265 Y×E=0.375 V×E=0.459 C×E=0.459 Y×V×C×E=1.125 | | | Y=0.294 V=0.359 C=0.359 Y×V=0.508 Y×C=NS V×C=0.623 E=0.294 Y×E=NS V×E=0.508 C×E=0.508 Y×V×C×E=1.246 | | | Y=0.125 V=0.268 C=0.268 Y×V= 0.379 Y×C=0.379 V×C=0.464 E=0.125 Y×E=0.309 V×E=0.379 C×E=0.379 Y×V×C×E=0.928 | | |

E= Environment, V= Varieties, C= Culture, Y= Year

**Table 3** Influence of sowing dates and cultures on siliquae initiation, completion and duration in mustard varieties based on two years pooled data

| Cultures/ Varieties | Siliquing initiation (days) | | | Siliquing completion (days) | | | Siliquing duration (days) | | |
|---|---|---|---|---|---|---|---|---|---|
| | No culture | | | No culture | | | No culture | | |
| | Timely sown | Late sown | Average | Timely sown | Late sown | Average | Timely sown | Late sown | Average |
| Giriraj | 72.5±0.7 | 87.2±0.8 | 79.8±0.7 | 95.0±2.0 | 104.2±6.5 | 99.6±4.3 | 22.5±0.8 | 17.0±0.4 | 19.7±0.6 |
| PBR 357 | 74.9±0.6 | 84.5±0.7 | 79.7±0.6 | 95.5±3.3 | 102.8±9.8 | 99.1±6.6 | 20.6±0.3 | 18.3±1.0 | 19.4±0.6 |
| RH 725 | 73.4±0.9 | 87.1±0.7 | 80.2±0.8 | 92.7±6.7 | 103.8±11.2 | 98.2±8.9 | 19.3±0.3 | 16.7±0.4 | 18.0±0.3 |
| Mean | 73.6±0.7 | 86.3±0.7 | | 94.4±4.0 | 103.6±9.2 | | 20.8±0.5 | 17.3±0.6 | |
| | **Microbial formulation** | | | **Microbial formulation** | | | **Microbial formulation** | | |
| Giriraj | 65.9±0.7 | 75.4±0.7 | 70.6±0.7 | 89.5±3.3 | 94.0±7.5 | 91.7±5.4 | 23.6±0.7 | 18.6±1.0 | 21.2±0.8 |
| PBR 357 | 66.3±0.6 | 72.3±0.6 | 69.3±0.6 | 91.5±4.2 | 95.3±10 | 93.4±7.1 | 25.2±0.4 | 23.0±0.8 | 23.3±0.6 |
| RH 725 | 62.3±0.5 | 78.1±0.6 | 70.2±0.5 | 88.0±1.0 | 98.5±5.2 | 93.2±3.1 | 25.7±0.6 | 19.4±0.8 | 22.6±0.7 |
| Mean | 64.8±0.6 | 75.3±0.6 | | 89.7±2.8 | 95.9±7.6 | | 24.8±0.5 | 20.3±0.9 | |
| | **Pusa Sanjeevani** | | | **Pusa Sanjeevani** | | | **Pusa Sanjeevani** | | |
| Giriraj | 63.3±0.9 | 76.1±0.7 | 69.7±0.8 | 87.5±2.8 | 94.5±0.7 | 91.0±1.8 | 24.2±0.5 | 18.4±0.5 | 21.3±0.5 |
| PBR 357 | 68.2±0.5 | 76.0±0.5 | 72.1±0.5 | 91.3±1.7 | 96.3±5.8 | 93.8±3.8 | 23.1±0.8 | 20.3±0.2 | 21.7±0.5 |
| RH 725 | 64.5±0.9 | 76.5±0.7 | 70.5±0.8 | 88.3±4.3 | 96.2±6.5 | 92.2±5.4 | 24.6±0.6 | 19.7±0.6 | 22.1±0.6 |
| Mean | 65.3±0.8 | 76.2±0.6 | | 89.1±2.9 | 95.7±4.3 | | 24.0±0.6 | 19.5±0.4 | |
| CD (p≤0.05) | Y=0.279 V=0.342 C=0.342 Y×V=0.484 Y×C=0.484 V×C=0.593 E=0.279 Y×E=NS V×E=0.484 C×E=0.484 Y×V×C×E=1.185 | | | Y=0.311 V=0.380 C=0.381 Y×V= 0.538 Y×C=0.538 V×C=0.659 E=0.311 Y×E=NS V×E=0.538 C×E=0.538 Y×V×C×E=1.318 | | | Y=0.212 V=0.260 C=0.260 Y×V=NS Y×C=0.368 V×C=0.450 E=0.212 Y×E=NS V×E=0.368 C×E=0.368 Y×V×C×E=0.637 | | |

E= Environment, V= Varieties, C= Culture, Y= Year

**Table 4** Influence of sowing dates and cultures on reproductive phase, physiological maturity and seed yield in mustard varieties based on two years pooled data

| Cultures/ Varieties | Reproductive phase (days) | | | Physiological maturity (days) | | |
|---|---|---|---|---|---|---|
| | **No culture** | | | **No culture** | | |
| | **Timely sown** | **Late sown** | **Average** | **Timely sown** | **Late sown** | **Average** |
| Giriraj | 26.7±0.3 | 23.8±0.6 | 25.2±0.5 | 121.0±1.7 | 115.2±2.2 | 118.1±1.9 |
| PBR 357 | 27.1±0.8 | 23.6±0.6 | 25.3±0.7 | 126.5±1.8 | 118.3±1.0 | 122.4±1.4 |
| RH 725 | 25.6±0.5 | 26.0±1.0 | 25.8±0.7 | 128.8±2.2 | 117.5±6.2 | 123.2±4.2 |
| Mean | 26.5±0.5 | 24.5±0.7 | | 125.4±1.9 | 117.0±3.1 | |
| | **Microbial formulation** | | | | | |
| Giriraj | 29.7±0.7 | 25.8±0.7 | 27.7±0.7 | 137.2±4.3 | 124.8±7.5 | 131.0±5.9 |
| PBR 357 | 33.1±0.9 | 29.0±1.1 | 31.0±1.0 | 140.8±4.0 | 125.0±3.3 | 132.9±3.7 |
| RH 725 | 31.0±1.2 | 27.5±1.3 | 29.2±1.2 | 140.2±4.8 | 126.2±7.3 | 133.2±6.1 |
| Mean | 31.3±0.9 | 27.5±1.0 | | 139.4±4.4 | 125.3±6.1 | |
| | **Pusa Sanjeevani** | | | | | |
| Giriraj | 29.6±0.5 | 27.9±1.2 | 28.7±0.8 | 139.7±1.0 | 134.7±3.5 | 137.2±2.3 |
| PBR 357 | 31.6±0.9 | 26.9±0.8 | 29.2±0.8 | 143.5±3.0 | 137.0±5.2 | 140.2±4.1 |
| RH 725 | 30.3±1.2 | 27.2±0.3 | 28.7±0.8 | 142.0±6.7 | 126.9±13.2 | 134.5±9.9 |
| Mean | 30.5±0.9 | 27.3±0.8 | | 141.7±3.6 | 132.9±7.3 | |
| CD (p≤0.05) | Y=0.204 V=0.249 C=0.249 Y×V=0.353 Y×C=0.353 V×C=0.432 E=0.204 Y×E=0.288 V×E=0.353 C×E=0.353 Y×V×C×E=0.864 | | | Y=0.632 V=0.774 C=0.774 Y×V=1.095 Y×C=1.095 V×C=1.341 E=0.632 Y×E=NS V×E=1.095 C×E=1.095 Y×V×C×E=2.683 | | |

E= Environment, V= Varieties, C= Culture, Y= Year

Variations in phenology is considered as one of the most significant and observable markers of how the plants respond to sowing dates with change in environment. Sharma and Sardana (2013), Priya (2020) and Dar (2023) recorded delayed flowering and siliquae initiation and completion in *B. juncea* under stress conditions. Kaur et. al, (2022) and Langangmeilu et. al, (2023) reported significant delay in all the phenophases with late planting in *B. juncea* under temperature stress. Flowering duration decreased significantly with delayed planting from 45 (6$^{th}$ October sowing) to 41 days (23$^{rd}$ October sowing) and 35 days (12$^{th}$ November sowing) in *B. juncea* (Devi and Sharma 2017). Elevated temperature promoted flowering in soybean as reported by No et. al, (2021). Likewise, shorter reproductive phase under heat stress has been reported by Kinay and Kayacetin (2023) in *Brassica*. A sudden rise in air temperatures during the reproductive stage shortened the seed filling period. Sharma (2020) reported negative influence of high temperature on photosynthetic efficiency, translocation and nutrients uptake which ultimately affected the growth phases and led to early or enforced maturity. Farhat et. al, (2023) observed that the number of days to flowering decreased in rapeseed plants treated with a consortium of Rhizobium species (*Bacillus subtilis* and *Pseudomonas aeruginosa*) as compared to untreated plants. Early-sown crops encountered sufficient soil moisture and warm temperatures during the vegetative and reproductive phases, resulting in improved development and higher yield in yellow mustard (*Sinapis alba*) than late sown crop (Anuroop et. al, 2022).

### 3.4. Seed yield

Significant variation existed for seed yield among the varieties with sowing dates and microbial treatments as indicated by pooled mean of two-years.

Seed yield was maximum under normal sown than late sown crop. In untreated control, seed yield declined by 7.3% in Giriraj, 12.9% in PBR 357 and 11.9% in RH 725. Microbial treated varieties improved seed yield than the untreated control. However, varieties treated with Microbial formulation suffer lesser yield decline of 6.3% in Giriraj, 5.4% in PBR 357 and 5.5% in RH 725. Similarly, yield was reduced by 4.9, 5.5 and 5.4% in Giriraj, PBR 357 and RH 725 varieties respectively treated with Pusa Sanjeevani (Figure 2). Overall, maximum seed yield decline was 6.3% in Giriraj variety treated with Microbial formulation and in PBR 357 and RH 725 varieties treated with Pusa Sanjeevani under late sown conditions over timely sown conditions. On an average, yield enhancement has been recorded with microbial inoculations as indicated by the lesser decline in seed yield w.r.t the control. Ali et. al, (2024) found that 1% rise in temperature resulted in 10% decline in seed yield in rice during the dry season. Kheir et. al, (2021) studied that heat stress reduced seed yield by 18.5% in canola. Similarly, high temperature stress reduced yields in many cultivated crops such as rice (Yaliang et. al, 2020), soybean (Nakagawa et. al, 2020) and wheat (Schittenhelm et. al, 2020). Also, increased salinity stress from 1.8 to 12 dS/m decreased grain yield by 54.6 % in *Eruca sativa* as reported by Shariatinia et. al, (2021). In contrast, wheat plants inoculated with *Enterobacter* SA187 experienced a 40% increase in seed yield (Park et. al, 2017). Ji et. al, (2022) also showed enhanced seed yield in wheat plants treated *Bacillus subtilis* HG-15 and *Bacillus velezensis* JC-K3 under low salt stress.

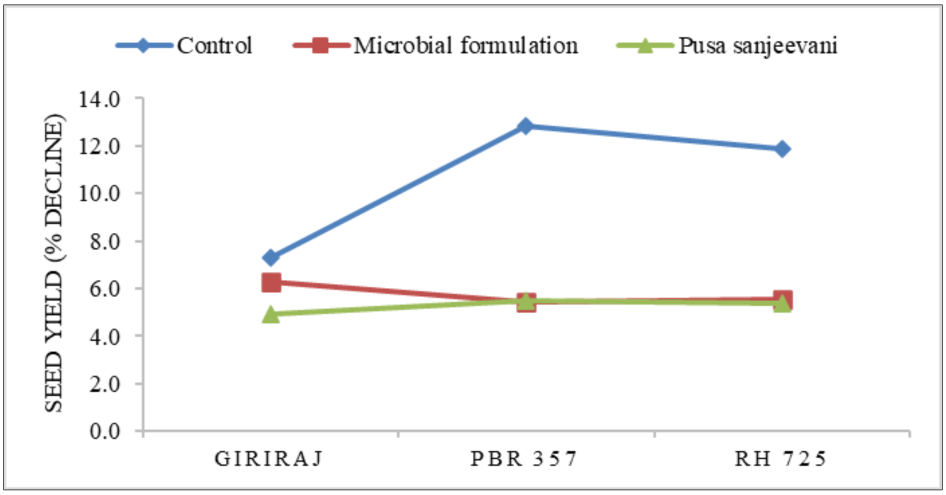
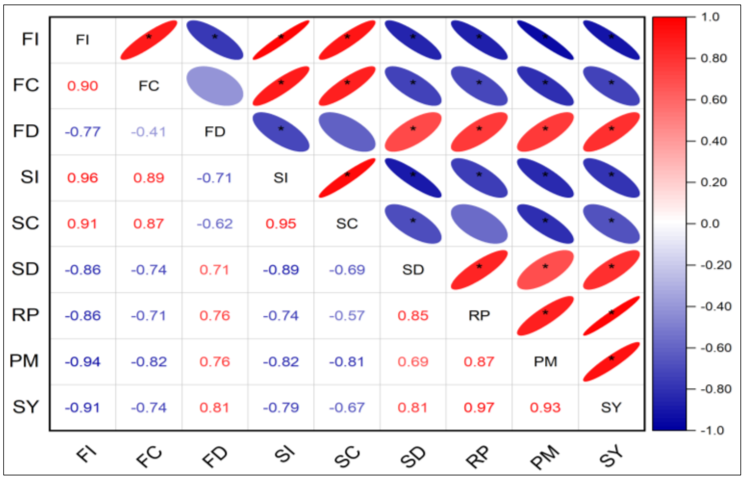


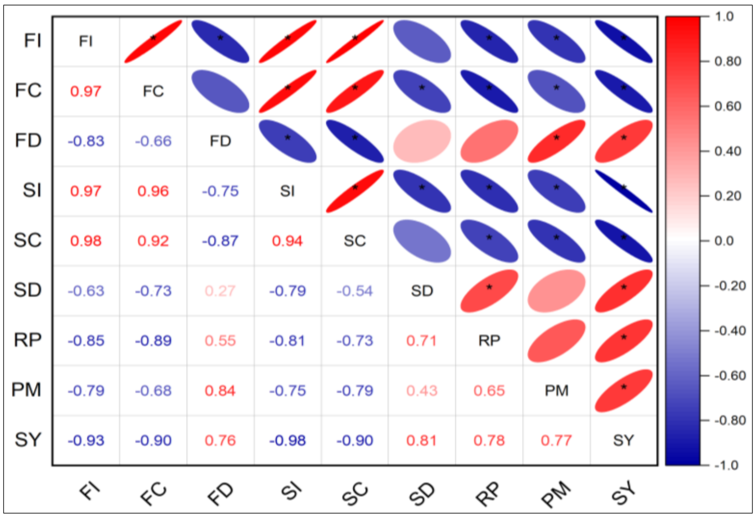

**Figure 2** Seed yield (% decline) in three varieties under microbial culture treatments

### 3.5. Pearson's Correlation and linear regression analysis

Significant relationship between all phenological traits and seed yield was reported under both normal and late sown conditions. Seed yield had positive and significant correlation with flowering duration (r=0.81** and r=0.76*), siliquing duration (r=0.81** and r=0.81**) reproductive phase (r=0.97** and r=0.78*) and physiological maturity (r=0.93** and r=0.77*) and negative but significant relationship of yield was with flowering initiation (r= -0.91** and r= -0.93**), flowering completion (r= -0.74* and r= -0.90**), siliquae initiation (r= -0.79* and r= -0.98**) and siliquae completion (r= -0.67* and r= -0.90**) under both timely and late sowing conditions (Fig. 3). Linear regression analysis of phenological traits with seed yield indicated differential association among them. High $R^2$ values for reproductive phase and physiological maturity exhibited a strong positive relationship with seed yield, particularly in timely sown crop indicating significant influence on seed yield. Flowering and siliquing duration showed a moderate relationship with seed yield under normal and late sown conditions. However, flowering and siliquing completion in timely sown crops had weaker correlations, indicating their less influence critical in determining seed yield (Table 5 and Fig. 4).




Timely sown




Late sown

*Significant at 5%, **Significant at 1%, FI- flowering initiation, FC- Flowering completion, FD- Flowering duration, SI- Siliquae initiation, SC- Siliquae completion, SD- Siliquae duration, RP- Reproductive phase, M- Physiological maturity, SY- Seed yield

**Figure 3** Correlation of phenological attributes with seed yield across sowing dates

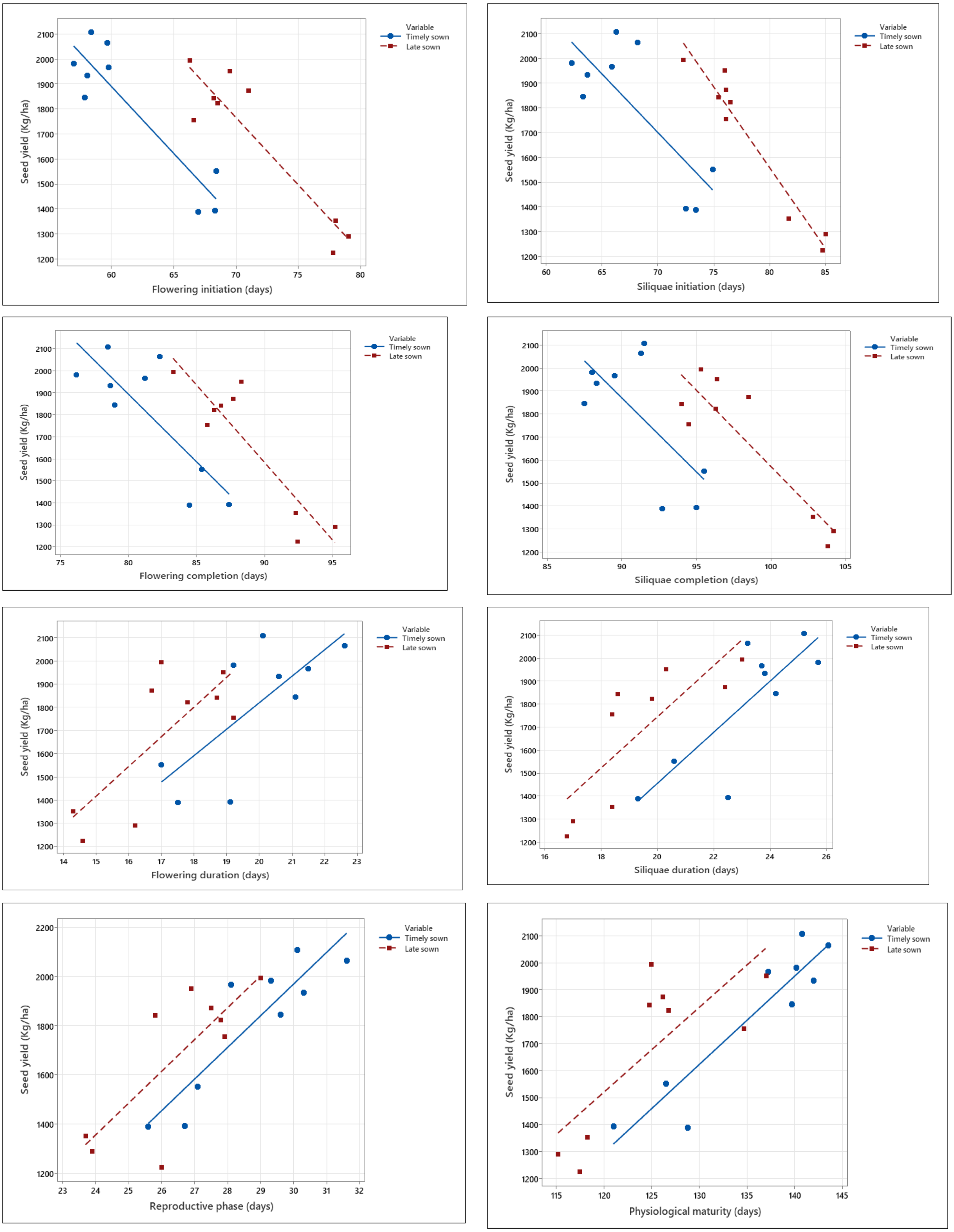



**Figure 4** Relationship of phenological traits with seed yield under timely and late sown conditions

Our results are consistent with findings of Rameeh (2014) whereas positive relationship of seed yield with days to maturity was reported. Priya (2020) reported that flowering and siliquae initiation and completion had strong relationship with yield under timely and late sown conditions. Likewise, Bazzaz et. al, (2020) stated that high temperature due to late sowing strongly affected flowering duration and maturity of mustard which ultimately affects the seed yield. Sah et. al, (2020) showed weak relationship between flowering and siliquing traits under irrigated condition, while stronger relationship existed under moisture stress. Kaur et. al, (2022) also showed a weak relationship of flowering completion with seed yield under timely sown crop over late sown crop.

**Table 5** Relationship of phenological traits with seed yield

| **Traits** | | **Timely sown** | | **Late sown** | |
|---|---|---|---|---|---|
| | | **y=mx+c** | **$R^2$** | **y=mx+c** | **$R^2$** |
| Flower | Initiation | -53.617x + 5108.8 | 0.827 | -54.006x + 5549 | 0.855 |
| | Completion | -62.553x + 6879.3 | 0.549 | -70.555x + 7935.8 | 0.801 |
| | Duration | 98.721x - 121.32 | 0.667 | 128.24x - 506.63 | 0.582 |
| Siliquae | Initiation | -48.446x + 5094.9 | 0.628 | -52.655x + 5840.7 | 0.962 |
| | Completion | -64.382x + 7665.7 | 0.454 | -66.472x + 8221.5 | 0.819 |
| | Duration | 111.24x - 768.6 | 0.662 | 111.56x - 486.29 | 0.652 |
| Reproductive phase | | 110.92x - 1461 | 0.936 | 129.41x - 1750.1 | 0.608 |
| Physiological maturity | | 32.798x - 2640 | 0.861 | 31.492x - 2259.4 | 0.600 |

## 4. Conclusion

The study highlighted that timely sown crop encountered sufficient soil moisture and warm temperature conditions leading to improved development and higher yield in mustard varieties. Timely sown crop exhibited early flowering and fruiting behaviour along with longer flowering and siliquing durations and reproductive phase than late sown crop. In contrast, delayed sowing experienced environmental stress, causing accelerated maturation but often incomplete development and reduced yield. Moreover, microbial treatments consistently outperformed the untreated control in all phenological traits, thereby enhancing crop performance, growth and overall productivity.

## Compliance with ethical standards

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*Disclosure of conflict of interest*

The authors declared that no conflict of interests exist.

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