

**RESEARCH ARTICLE** 



# Optimizing morpho-physiological traits and yield potential in Indian mustard [*Brassica juncea* (L) Czern] through irrigation frequency, humic acid and sulphur application

Toko Manna & Anaytullah Siddique<sup>\*</sup>

Department of Agronomy, School of Agriculture, Lovely Professional University, Phagwara 144 411, Punjab, India

\*Email: anaytullahsiddique@gmail.com

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### Abstract

The depleting of water resources and poor nutrient availability during crop growth pose significant stress, adversely affecting the vegetative and reproductive growth of mustard crops. This stress extends its negative impact to the reproductive stage and on oil content. Consequently, morpho-physiological traits were targeted for evaluation under varying irrigation frequencies and the application of humic acid and sulphur, both individually and in combination, using a split-plot design. The results demonstrated that the treatment involving three irrigations (I3) and the combination of humic acid and sulphur (T3) were consistently the most effective and statistically significant at p = 0.05 for most parameters. The interaction effect of these treatments was observed specifically for absolute growth rate (AGR), crop growth rate (CGR), relative growth rate (RGR), total number of branches per plant, seed yield (kg/ha) and total chlorophyll content (mg/g). Furthermore, I3 exhibited the highest percentage increase over the control for parameters such as plant height (9.3%), dry matter accumulation (33.5%), number of leaves (65.24%), leaf area (24.97%), leaf area index (24.97%), absolute growth rate (36.01%), crop growth rate (36.01%), net assimilation rate (23.42%), relative growth rate (15.34%), total number of branches (10.85%), seed yield (56.9%) and total chlorophyll content (10.85%). Similarly, T3 recorded increases of 12.1%, 31.3%, 22.46%, 12.63%, 31.93%, 12.41%, 4.02%, 34.17%, 50.70% and 12.75% for these same parameters, respectively. This study highlights the significant potential of combining three irrigations at critical growth stages with the application of humic acid and sulphur to mitigate the adverse effects of water stress on morpho-physiological traits in Indian mustard (Brassica juncea (L) Czern). Such interventions demonstrate promise in improving growth, yield and physiological performance under induced water stress conditions.

#### **Keywords**

chlorophyll; humic acid; irrigation frequency; leaf area index; net assimilation rate; sulphur

### Introduction

Mustard (*Brassica juncea* (L) Czern) is a highly valuable oilseed crop worldwide (1). Currently, India contributes approximately 12-13% of global oilseed production, while Nepal leads with a contribution of 41.3%. Mustard is a significant source of edible oil, rich in essential fatty acids, vitamins and minerals vital for human health (2-3). Additionally, it contains S-amino acids, which are crucial for both human nutrition and animal feed (4).

Irrigation is a critical component during the crop growth period, as it fulfills the water requirements necessary for various metabolic activities. Insufficient irrigation, especially at critical growth stages, induces moisture stress in mustard crops. This stress triggers the production of reactive oxygen species (ROS) at the cellular level, which accelerates lipid peroxidation and biochemical alterations. These changes weaken the plant's defense mechanisms, resulting in stunted morphological growth and reduced crop yield (5). In contrast, optimal irrigation scheduling, combined with the application of humic acid and sulphur, can significantly improve morpho-physiological growth and yield potential (6). Additionally, soil nutrient depletion disrupts metabolic processes, leading to nutritional deficiencies that adversely affect plant performance, such as reduced oil content in mustard due to sulphur deficiency.

Humic acid, an organic substance, plays a pivotal role in enhancing soil properties. It improves soil structure, increases water retention, stimulates root growth and enhances nutrient availability and microbial activity (7, 8). Moreover, humic acid facilitates osmotic adjustment by increasing soluble protein and free proline levels and enhancing the activity of antioxidative enzymes in plants (9). Sulphur application, on the other hand, is essential for protein synthesis, chlorophyll formation, enzyme activation, improved nutrient uptake and glucosinolate production in mustard crops (10, 11).

The combined application of humic acid and sulphur has shown promising results in promoting morphological growth and biochemical responses in oilseed crops, including soybean, olive and flax, under various stress conditions (12-14). Together, humic acid and sulphur work synergistically to improve soil fertility, enhance plant growth and contribute to sustainable agricultural practices by fostering healthier and more resilient crops.

### **Materials and Methods**

A study was conducted to evaluate the impact of irrigation strategies in conjucation with the application of humic acid and sulphur to optimize the yield potential of Indian mustard (*Brassica juncea* (L) Czern). The research was carried out at the Research Farm of Lovely Professional University, during the *Rabi* session of 2022-23 and 2023-24. The pooled data from this study are comprehensively presented in the research paper.

### **Experimental details**

The experiment was organised in a split-plot design (SPD) with sixteen possible combinations of irrigation regimes, humic acid and sulphur, along with four replications of the RLC-3 variety, which was collected from Punjab Agricultural University, Punjab, India. Different irrigation strategies *i.e.*  $I_0$ ,  $I_1$ ,  $I_2$  and  $I_3$ , were implemented in the main plot (each main plot covering 100m<sup>2</sup>), while chemical treatments, *i.e.*  $T_0$ ,  $T_1$ ,  $T_2$  and  $T_3$ , applied in the subplot (each subplot covering 25m<sup>2</sup>). Humic acid, sulphur, along with their combination (humic acid + sulphur) were used @ 4.86 and 73.57 kg acre<sup>-1</sup> respectively in the soil.

The experimental field had a sandy loam texture, comprising 62.85% sand, 19.1% silt and 12.6% clay, as determined by the International Pipette Method. Observations of morphological traits were recorded at regular intervals, spanning from 30 days to 90 days after sowing (DAS). Phenological traits were calculated during two growth periods: 30-60 DAS and 60-90 DAS.

### **Growth analysis**

To evaluate the efficacy of the treatments, morphological parameters such as plant height (cm), dry matter accumulation (g plant<sup>-1</sup>), number of leaves, leaf area (cm<sup>2</sup> plant<sup>-1</sup>) and total number of branches (plant<sup>-1</sup>) were recorded by averaging the measurement of five plants from each replication. Seed yield was recorded by sampling per square meter. Dry matter accumulation and leaf area were measured periodically through destructive sampling. The leaf area was assessed using a leaf area meter (MDL-1000 LICOR) and the data were used to compute phenological parameters such as Leaf Area Index (LAI), Absolute Growth Rate (AGR), Crop Growth Rate (NAR) as per the procedure given by (15-18).

The equations used for the computations are as follows:

LAI= Total leaf area (cm <sup>2</sup> ) / Total ground area (cm <sup>2</sup> )	(Eqn. 1)
$AGR = W_2 - W_1 / T_2 - T_1(g day^1)$	(Eqn. 2)
$CGR = (W_2-W_1) / (T_2-T_1)^* (1/Sample area) (g m^2 day)$	(Eqn. 3)
$RGR = (lnW_2 - lnW_1) / T_2 - T_1) (gg^1 day^1)$	(Eqn. 4)
$NAR = (W_2-W_1) / (T_2-T_1)^* (InA_2-InA_1) / (A_2-A_1) (g m^2 day)$	(Eqn. 5)
Whereas,	
$W_{\rm c}$ and $W_{\rm c}$ represent the total weights of dny matter (g)	at timos t

 $W_1$  and  $W_2$  represent the total weights of dry matter (g) at times  $t_1$  and  $t_2$  respectively

InW<sub>2</sub>-InW<sub>1</sub> represents the natural log of W<sub>2</sub> and W<sub>1</sub>

 $A_1$  and  $A_2$ =Leaf Area

 $T_1$  and  $T_2$  represent duration or interval

#### Total chlorophyll estimation

The total chlorophyll content was estimated (19). For this, 100 mg of leaf sample was homogenized with 10 ml of 80 % acetone. The homogenized sample was centrifuged at 5000 rpm and the aliquot was transferred to a 100 ml volumetric flask. The final volume was adjusted to 100 ml using 80% acetone. The optical density was measured at 663 and 645nm. The total chlorophyll was calculated using the following equation:

Total chlorophyll = 
$$\{20.2(D \, 645) + 8.02 (D \, 663)\} = \frac{V}{100 \times W}$$
 mg g<sup>-1</sup>  
(Eqn.6)

Whereas,

D= Intensity at which optical density was recorded

V= Final volume of the sample

W= Weight of leaf sample was taken for the estimation of total chlorophyll

Values 20.2 and 8.02 are constant factors.

### **Statistical Analysis**

The data collected from the experimental plots were subjected to statistical analysis to evaluate the significance of the treatments. The data were analyzed using a SPD for two-way analysis of variance (ANOVA). Additionally, the data were subjected to DMRT at a significance level of p=0.05%. Statistical analysis was performed using SPSS software (version 23) as per Field (2022).

### Results

# Plant height and dry matter accumulation and number of leaves

Plant height and dry matter accumulations were recorded at 60 and 90 DAS to assess the efficacy of treatments applied to Indian mustard. The data presented in Table 1 revealed the irrigation and nutrients treatments had a significant effect on the main plot and subplot parameters, along their interaction was found to be non-significant at (p=0.05%). Results from the main plot i.e. irrigation frequency revealed that as the number of irrigations increased from I<sub>0</sub> to I<sub>3</sub>, there was a corresponding improvements in plant height (PH), dry matter accumulation and the number of leaves plant<sup>-1</sup> by 13.6, 37 and 61.18%, respectively, at 90 DAS compared to the control .In the subplot treatments, T3 (a combination of humic acid and sulphur) was found to be the most effective compared to individual treatments (humic acid or sulphur alone). It resulted in increases of 9.7%, 29.8% and 23.98% in plant height, dry matter accumulation and the number of leaves per plant, respectively, over the control (Table 1).

### Leaf area and leaf area index (LAI)

The Leaf Area Index (LAI) represents the total leaf area per plant relative to the ground area covered, which reflects the morphological growth of the plant. Leaf area and LAI were analyzed throughout the crop growth period and statistical analysis indicated that both parameters were significant at p= 0.05 for most intervals, though their interaction was nonsignificant (Table 2). Data revealed a gradual increase in total leaf area (cm<sup>2</sup>) up to 60 DAS, with the highest values observed under I3 compared to other irrigation treatments. Improvements in leaf area and LAI due to I3 were recorded at 24.97% over the control at 60 DAS. Similarly, T3 (humic acid +

Table 1. Treatments on plant height, dry matter accumulation and number of leaves in India mustard [Brassica juncea (L) Czern]

Treatment	Plant height (cm)		Dry matter accum	ulation (g plant <sup>-1</sup> )	Number of leaves plant <sup>-1</sup>		
Main plot	60 DAS	90 DAS	60 DAS	90 DAS	60 DAS	90 DAS	
lo	111.0 <sup>b</sup>	155.7°	14.9 <sup>d</sup>	29.2 <sup>d</sup>	10.06 <sup>d</sup>	13.23 <sup>d</sup>	
l1	116.8ª [5.0%]	167.3 <sup>b</sup> [6.95%]	20.9 <sup>c</sup> [28.9%]	40.8 <sup>c</sup> [28.4%]	16.75° [39.97%]	17.34 <sup>c</sup> [23.78%]	
l <sub>2</sub>	122.6ª [ 9.5%]	180.1ª [13.6%]	21.6 <sup>b</sup> [31.1%]	43.0 <sup>b</sup> [32.0%]	21.64 <sup>b</sup> [53.53%]	27.05 <sup>b</sup> [51.14%]	
I <sub>3</sub>	122.3ª [9.3%]	180.2ª [13.6%]	22.3ª [33.5%]	46.4ª [37.0%]	28.93ª [65.24%]	34.04ª [61.18%]	
CD (p=0.05%)	8.3	9.6	0.28	0.50	1.94	1.67	
			Subplot				
T <sub>0</sub>	109.5 <sup>d</sup>	161.4 <sup>d</sup>	16.0 <sup>c</sup>	32.4 <sup>d</sup>	16.87 <sup>d</sup>	19.48 <sup>d</sup>	
$T_1$	120.6 <sup>b</sup> [9.2%]	173.8 <sup>b</sup> [7.2%]	21.4 <sup>b</sup> [25.2%]	21.4 <sup>b</sup> [25.2%] 43.0 <sup>b</sup> [24.5%]		24.24 <sup>b</sup> [19.61%]	
T <sub>2</sub>	118.1º [ 7.2%]	169.7° [4.9%]	19.0° [15.9%]	37.7° [14.0%]	18.44 <sup>c</sup> [8.52%]	22.29 <sup>c</sup> [12.60%]	
T <sub>3</sub>	124.6ª [ 12.1%]	178.5ª [9.7%]	23.3ª [31.3%]	46.2ª [29.8%]	21.76ª [22.46%]	25.63ª [23.98%]	
CD (p=0.05%)	3.0	3.1	0.30	0.45	1.22	1.28	
CD at (p=0.05%) MXS	NS	NS	NS	NS	NS	NS	

Note:

I<sub>0</sub> =No post sowing irrigation, I<sub>1</sub> =One post sowing irrigation, I<sub>2</sub> =Two post sowing irrigation, I<sub>3</sub> =Three post sowing irrigation (Vegetative + Flowering + Seed 1. filling stage)

2. T<sub>0</sub> = Control, T<sub>1</sub>= Humic acid, T<sub>2</sub> = Sulphur, T<sub>3</sub> = Humic acid + Sulphur

3. C. D=Critical Difference

4. Data presented in parenthesis represent the % increase over control

5. DAS= days after sowing

Table 2. Treatments impact on leaves area and LAI in Indian mustard [Brassica juncea (L) Czern]

Treatment	Le	aves area (cm² plan	t-1)	LAI			
Main plot	30 DAS	60 DAS	90 DAS	30 DAS	60 DAS	90 DAS	
lo	362.97 <sup>b</sup>	1448.01 <sup>b</sup>	842.91°	0.81ª	3.22 <sup>b</sup>	1.87°	
$I_1$	365.51ª [0.69%]	1820.96ª [20.48%]	1207.76 <sup>b</sup> [30.21%]	0.81ª [0.69%]	4.05ª [20.48%]	2.68 <sup>b</sup> [30.21%]	
I <sub>2</sub>	365.55° [0.70%]	1879.65ª [22.96%]	1302.13 <sup>ab</sup> [35.27%]	0.81ª [0.70%]	4.18ª [22.96%]	2.89 <sup>ab</sup> [35.27%]	
I <sub>3</sub>	368.63ª [1.53%]	1930.01ª [24.97%]	1327.26ª [36.49%]	0.82ªm [1.53%]	4.29ª [24.97%]	2.95ª [36.49%]	
CD (p=0.05%)	NS	114.06	112.75 NS		0.25	12.05	
			Subplot				
T <sub>0</sub>	284.25 <sup>d</sup>	1635.20 <sup>c</sup>	1021.28 <sup>d</sup>	0.63 <sup>d</sup>	3.63 <sup>c</sup>	2.27°	
T <sub>1</sub>	399.28 <sup>b</sup> [28.81%]	1804.77 <sup>b</sup> [9.40%]	1212.29 <sup>b</sup> [15.76%]	0.89 <sup>b</sup> [28.81%]	4.01 <sup>b</sup> [9.40%]	2.69 <sup>b</sup> [15.76%]	
T <sub>2</sub>	331.344 <sup>c</sup> [14.21%]	1767.03 <sup>b</sup> [7.46%]	1158.76º [11.86%]	0.75º [14.21%]	3.93 <sup>b</sup> [7.46%]	$2.58^{ m b}$ [11.86%]	
T <sub>3</sub>	447.76ª [36.52%]	1871.62ª [12.63%]	1287.71ª [20.69%]	0.99ª [36.52%]	4.16ª [12.63%]	2.86ª [20.69%]	
CD (p=0.05%)	) 11.29 49.64 52.78 0.03		0.03	0.11	0.12		
CD at (p=0.05%) MXS	NS	NS	NS	NS	NS	NS	

#### Note:

Io =No post sowing irrigation, I1 =One post sowing irrigation, I2 =Two post sowing irrigation, I3 =Three post sowing irrigation (Vegetative + Flowering + Seed 1. filling stage),

2.  $T_0$  = Control,  $T_1$ = Humic acid,  $T_2$  = Sulphur,  $T_3$  = Humic acid + Sulphur,

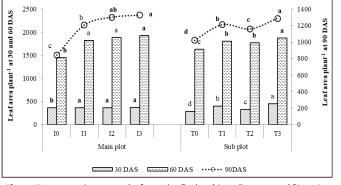
3. C. D=Critical Difference

Data presented in parenthesis represent the % increase over control 4.

DAS= days after sowing, LAI= leaf area index 5.

As per the alphabets of the Duncan, the same alphabet indicates nonsignificant difference while different alphabets indicate significant differences among 6. the treatments.

3



**Fig. 1.** Treatments impact on leaf area (cm<sup>2</sup>) plant<sup>1</sup> in Indian mustard [*Brassica juncea* (L) Czern].

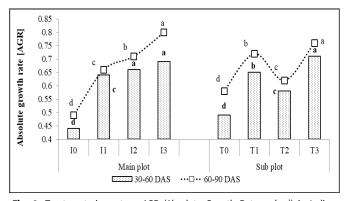
sulphur) showed the best performance among subplot treatments, resulting in a 12.63% increase in both leaf area and LAI over the control (Fig. 1 and 2).

### Phenological parameters (AGR, CGR, RGR and NAR)

Phenological parameters such as absolute growth rate (AGR), crop growth rate (CGR), relative growth rate (RGR) and net assimilation rate (NAR) were evaluated to determine the effectiveness of irrigation and chemical treatments. The data (Fig. 3-6) demonstrated statistically significant improvements at p= 0.05 for both factors. Growth parameters showed a consistent increase from I1 to I3 and from T1 to T3 compared to I0 and T0, respectively. Among irrigation frequencies, I3 recorded the highest values for AGR, CGR, RGR and NAR during 60-90 DAS, with respective improvements of 40.34%, 11.84% and 13.89% over the control. In subplot treatments, T3 achieved the maximum phenological growth, with respective increases of 28.23%, 7.16% and 13.75% over the control (Table 3).

### Total number of branches and seed yield

The total number of branches per plant and seed yield (kg ha<sup>-1</sup>) were measured to evaluate the effectiveness of treatments on



**Fig. 3.** Treatments impact on AGR (Absolute Growth Rate g day<sup>1</sup>) in Indian mustard [*Brassica juncea* (L) Czern].

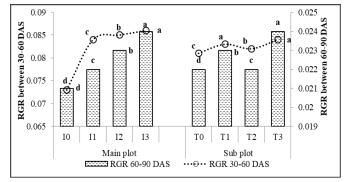


Fig. 5. Treatments impact on RGR (Relative Growth Rate g  $g^{\cdot 1}day^{\cdot 1})$  in Indian mustard [Brassica juncea (L) Czern].

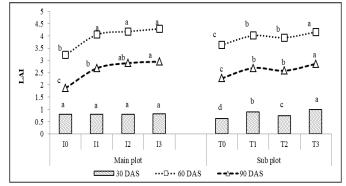
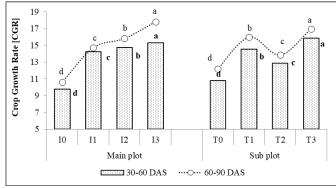


Fig. 2. Treatments impact on LAI (Leaf Area Index) in Indian mustard [Brassica juncea (L) Czern].

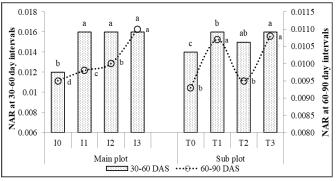
Indian mustard. Both parameters were found to be statistically significant at p= 0.05 for irrigation frequency and chemical treatments, with a significant interaction effect as well. Among irrigation frequencies, I3 recorded the highest number of branches per plant, with a 37.08% increase over the control. Similarly, T3 emerged as the most effective among chemical treatments, resulting in a 34.17% increase in branches over the control. In terms of seed yield, I3 and T3 achieved the highest gains, with improvements of 56.93% and 50.70%, respectively, over the control (Fig. 7).

#### Total chlorophyll content

Total chlorophyll content was measured to assess the impact of irrigation and chemical treatments. Both irrigation frequency and chemical treatments significantly influenced total chlorophyll content at p=0.05, with a significant interaction effect. Among irrigation frequencies, I3 resulted in the maximum increase in chlorophyll content over the control, with improvements of 10.85% and 14.83% at 60 DAS and 90 DAS, respectively. Similarly, T3 (humic acid + sulphur) exhibited the highest chlorophyll content gains, with increases of 12.75% and







**Fig. 6.** Treatments impact on NAR (Net Assimilation Rate g m<sup>2</sup> day<sup>1</sup>) in Indian mustard [*Brassica juncea* (L) Czern].

Table 3. Treatments impact on AGR, CGR, RGR and NAR in India mustard [Brassica juncea (L) Czern]

Treatment	AGR (g day <sup>-1</sup> )		CGR (g m <sup>2</sup> day <sup>-1</sup> )		RGR (g g <sup>-1</sup> day <sup>-1</sup> )		NAR (g m² day-1)	
Main plot	30-60 DAS	60-90 DAS	30-60 DAS	60-90 DAS	30-60 DAS	60-90 DAS	30-60 DAS	60-90 DAS
lo	0.44 <sup>d</sup>	0.49 <sup>d</sup>	9.79 <sup>d</sup>	10.62 <sup>d</sup>	0.073 <sup>d</sup>	0.021 <sup>d</sup>	0.012 <sup>b</sup>	0.0095 <sup>d</sup>
I <sub>1</sub>	0.64° [31.34%]	0.66° [27.81%]	14.26 <sup>c</sup> [31.34%]	14.72 <sup>c</sup> [27.81%]	0.084 <sup>c</sup> [13.55%]	0.022° [3.58%]	0.016ª [21.04%]	0.0098° [3.50%]
<b>I</b> <sub>2</sub>	0.66ª [33.63%]	0.71⁵ [32.87%]	14.75 <sup>b</sup> [33.63%]	15.82 <sup>b</sup> [32.87%]	0.085 <sup>b</sup> [14.44%]	0.023 <sup>b</sup> [6.32%]	0.016ª [22.09%]	0.0100 <sup>b</sup> [5.28%]
I <sub>3</sub>	0.69ª [36.01%]	0.80ª [40.34%]	15.29ª [36.01%]	17.81ª [40.34%]	0.086ª [15.34%]	0.024ª [11.84%]	0.016ª [23.42%]	0.0110ª [13.89%]
CD (p=0.05%)	0.0092	0.0115	0.21	0.26	0.0006	0.0003	0.00062	0.0009
			9	Subplot				
T <sub>0</sub>	0.49 <sup>d</sup>	0.58 <sup>d</sup>	10.80 <sup>d</sup>	12.18 <sup>d</sup>	0.0805 <sup>c</sup>	0.022 <sup>d</sup>	0.014 <sup>c</sup>	0.0093 <sup>b</sup>
T1	0.65 <sup>♭</sup> [25.68%]	0.72 <sup>b</sup> [23.83%]	14.54 <sup>b</sup> [25.68%]	15.98 <sup>b</sup> [23.83%]	0.083ª [3.09%]	0.023 <sup>b</sup> [4.74%]	0.016 <sup>b</sup> [10.42%]	0.0107ª [12.53%]
T <sub>2</sub>	0.58° [16.15%]	0.62° [12.06%]	12.88° [16.15%]	13.85 <sup>c</sup> [12.06%]	0.082 <sup>b</sup> [1.30%]	0.0224 <sup>c</sup> [1.69%]	0.015 <sup>ab</sup> [6.64%]	0.0095 <sup>b</sup> [2.13%]
T <sub>3</sub>	0.71ª [31.93%]	0.76ª [28.23%]	15.87ª [31.93%]	16.97ª [28.23%]	0.084ª [4.02%]	0.024ª [7.16%]	0.0159ª [12.41%]	0.0108ª [13.75%]
CD (p=0.05%)	0.0095	0.0112	0.21	0.25	0.0009	0.0004	0.00047	0.0004
CD at (p=0.05%) MXS	NS	0.022	NS	2.35	0.0019	0.0007	NS	NS

#### Note:

I<sub>0</sub> =No post sowing irrigation, I<sub>1</sub> =One post sowing irrigation, I<sub>2</sub> =Two post sowing irrigation, I<sub>3</sub> =Three post sowing irrigation (Vegetative + Flowering + Seed filling stage),

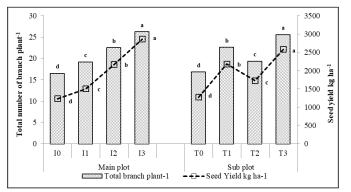
2.  $T_0$  = Control,  $T_1$ = Humic acid,  $T_2$  = Sulphur,  $T_3$  = Humic acid + Sulphur,

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4. Data presented in parenthesis represent the % increase over control

5. DAS= days after sowing, AGR= Absolute growth rate, CGR= Crop growth rate, RGR= Relative growth rate, NAR= Net assimilation rate

6. As per the alphabets of the Duncan, the same alphabet indicates nonsignificant difference while different alphabets indicate

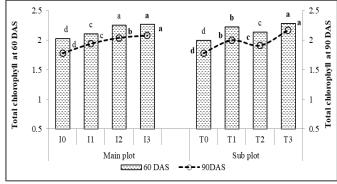


**Fig. 7.** Treatments impact on the total number of branches and seed yield kg ha<sup>-1</sup> in Indian mustard [*Brassica juncea* (L) Czern].

18.17% at the respective intervals. The data also indicated a slightly higher chlorophyll content at 60 DAS compared to 90 DAS for all treatment combinations (Fig. 8).

### Discussion

Limited rainfall and poor nutrient status are major factors contributing to the yield constraints in mustard cultivation. Adequate soil moisture ensures better plant establishment, while subsequent morpho-phenological growth, yield and yield attributes require both moisture and nutritional support to achieve optimal levels. The data presented in Table 1 highlights the significance of the treatments applied, where the combination of I3 (three levels of irrigation frequency) and T3 (humic acid and sulphur) resulted in statistically significant improvements in morphological traits such as plant height (cm), dry matter accumulation (g plant<sup>-1</sup>), number of leaves (plant<sup>-1</sup>) and leaf area (cm<sup>2</sup> plant<sup>-1</sup>) (Fig. 1). A similar findings reported enhanced morphological growth with three irrigations applied at critical growth stages (20). Additionally, studies demonstrated the beneficial effects of humic acid and sulphur, whether applied



**Fig. 8.** Treatments impact on total chlorophyll (mg g<sup>-1</sup>) in Indian mustard [*Brassica juncea* (L) Czern].

individually or in combination, on morphological growth (21, 22).

Optimal growth of phenological parameters such as Leaf Area Index (LAI), Crop Growth Rate (CGR), Relative Growth Rate (RGR) and Net Assimilation Rate (NAR) is dependent on several factors, including leaf area and dry matter accumulation. The data from Tables 2 and 3 indicate significant improvements in these parameters under the influence of irrigation frequency and chemical treatments, with the I3 and T3 combination emerging as the most effective treatment (Figs. 2-7). These findings align with the studies that mentions the combination of three irrigations with humic acid and sulphur as one of the most effective treatments for modulating morpho-phenological growth in mustard (23). Similar results were also reported (24-26).

Furthermore, the applied treatments not only improved plant height, dry matter accumulation and LAI but also ensured optimal leaf area and total chlorophyll content. These factors collectively contribute to carbohydrate production through photosynthesis (Fig. 8) and support vegetative growth, as shown by phenological traits like AGR, CGR, RGR and NAR, which were significantly better in the treatment groups compared to the control sets of IO and TO (Fig. 3-6).

The application of optimum irrigation in combination with humic acid and sulphur plays a vital role in bridging the gap between soil and plant by facilitating the supply of moisture and nutrients (27, 28). The role of humic acid in improving soil properties, such as aggregation, aeration, water holding capacity (WHC) and ion availability, is well established. These improvements collectively enhance water and nutrient uptake, which are crucial for biomass accumulation in plants (29, 30). Similarly, sulphur has been recognized for its ability to improve oil content in oilseed crops, particularly mustard. It interacts with plant metabolites to mitigate moisture stress, fulfilling the sulphur requirements while facilitating the synthesis of numerous sulphur-containing compounds. These compounds enhance the plant's ability to improve yield attributes (31-34).

### Conclusion

This study focused on analyzing the efficacy of irrigation scheduling and chemical treatments applied in Indian mustard to optimize morpho-phenological growth, yield attribute and seed yield. The findings of the study suggested that individual and combined effects of three irrigations at a critical stage of Indian mustard (I<sub>3</sub>) and combinations of humic acid and sulphur  $(T_3)$  expressed their potential to modulated morphophenological growth such as PH (cm), dry matter accumulation (g plant<sup>-1</sup>), leaf area (cm<sup>2</sup> plant<sup>-1</sup>), LAI, AGR (g day<sup>-1</sup>) CGR (g m<sup>2</sup>  $day^{-1}$ ), RGR (g g<sup>-1</sup> day<sup>-1</sup>) and NAR (g m<sup>2</sup> day<sup>-1</sup>), subsequently, it also influences the total number of the branch (plant<sup>-1</sup>), chlorophyll content (mg g<sup>-1</sup>) thereby the highest seed yield of Indian mustard was achieved. Moreover, both the chemicals humic acid and sulphur act synergistically at the biochemical level and coordinate with metabolites to regulate the supply of moisture and nutrients to the plants. Therefore, it may be suggested that the use of three irrigation in combination with humic acid and sulphur might be auxiliary to boost the yield of Indian mustard by optimizing the morpho-phenological traits.

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# **Authors' Contributions**

AS was responsible for conceptualizing the research, gathering relevant literature and preparing the original draft of the manuscript. TM contributed to the conceptualization, developed the research layout, took part in writing and editing and provided overall supervision throughout the study.

### **Compliance with Ethical Standards**

**Conflict of interest:** Authors do not have any conflict of interests to declare.

Ethical issues: None

#### References

- Shah SH, Parrey ZA, Islam S, Tyagi A, Ahmad A, Mohammad F. Exogenously applied sulphur improves growth, photosynthetic efficiency, enzymatic activities, mineral nutrient contents, yield and quality of *Brassica juncea* L. Sustainability. 2022;14:14441. https:// doi.org/10.3390/su142114441
- Jat RS, Singh VV, Sharma P, Rai PK. Oilseed *Brassica* in India: Demand, supply, policy perspective and future potential. OCL. 2019;26:8. https://doi.org/10.1051/ocl/2019005
- Kohli SK, Bali S, Tejpal R, Bhalla V, Verma V, Bhardwaj R, et al. *In-situ* localization and biochemical analysis of bio-molecules reveal Pbstress amelioration in *Brassica juncea* L. by co-application of 24epibrassinolide and salicylic acid. Sci Rep. 2019;9:3524. https:// doi.org/10.1038/s41598-019-39712-2
- Shah SH, Islam S, Parrey ZA, Mohammad F. Role of exogenously applied plant growth regulators in growth and development of edible oilseed crops under variable environmental conditions: A review. J Soil Sci Plant Nut. 2021;21:3284-308. https:// link.springer.com/article/10.1007/s42729-021-00606-w
- Singh R, Singh P, Singh H, Raghubanshi AS. Impact of sole and combined application of biochar, organic and chemical fertilizers on wheat crop yield and water productivity in a dry tropical agroecosystem. Biochar. 2019;1:229-35. https://doi.org/10.1007/s42773-019-00013-6
- Tamboli YA, Yadav JS, Kumar P, Dahiya R, Kumar A. Growth and phenological responses of Indian mustard (*Brassica juncea* L.) to different irrigation levels, varieties and antitranspirants. J Pharm Innov. 2021;10:920-26. https://www.thepharmajournal.com/ archives/2021/vol10issue7/PartL/10-6-272-683.pdf
- Ampong K, Thilakaranthna MS, Gorim LY. Understanding the role of humic acids on crop performance and soil health. Front Agron. 2022;4:848621. https://doi.org/10.3389/fagro.2022.848621
- Wu Y, Li S, Chen G. Impact of humic acids on phosphorus retention and transport. J Soil Sci Plant Nut. 2020;20:2431-39. https:// doi.org/10.1007/s42729-020-00308-9
- 9. Rathore SS, Shekhawat K, Kandpal BK, Premi OP, Singh SP, Chand G. Sulphur management for increased productivity of Indian mustard: A review. Ann Plant Soil Res. 2015;17:1-12.
- Maurya MK, Sarma A, Behera P, Karan N, Shukla VK, Kumar, A, et al. A critical review on sulphur application in rapeseed-mustard to enhancing productivity and oil quality. Int J Plant Soil Sci. 2023;35:661-71. https://journalijpss.com/index.php/IJPSS/ article/view/4026
- 11. Sohail A, Ahmed U, Khan RA, Khan MHA, Ikramullah, Iqbal A, et al. Application of humic acid and foliar sulfur positively affect yield and related attributes of soybean under the agroclimatic region of Peshawar Pakistan. Pure Appl Biol. 2024;14:8-20.
- 12. Danyaei A, Hassanpour S, Baghaee MA, Dabbagh M, Babarabie M. The effect of sulfur-containing humic acid on yield and nutrient uptake in olive fruit. Open J Ecol. 2017;7:279-88.
- Ahmad H, Khan SJ, Riaz R, Kafeel M. Application of humic acid and sulphur on the vigour and viability of harvested soybean seeds with active compounds. J Adv Nut Sci Tech. 2023;3:92-99. https:// anst.scione.com/newfiles/anst.scione.com/42/ANST-16-23.pdf
- Bakry AB, Sadak MS, El-Karamany MF. Effect of humic acid and sulfur on growth, some biochemical constituents, yield and yield attributes of flax grown under newly reclaimed sandy soils. ARPN J Agric Biol Sci. 2015;10:247-59. https://www.arpnjournals.com/jabs/ research\_papers/rp\_2015/jabs\_0715\_737.pdf
- Watson DJ. Comparative physiological studies on the growth of field crops: I. Variation in net assimilation rate and leaf area between species and varieties and within and between years. Ann Bot. 1947;11:41-76. https://www.jstor.org/stable/42907002

- Wareing PF, Phillips IDJ. Growth and differentiation in plants (Vol. 3). Oxford: Pergamon Press. 1981. https://doi.org/10.1086/406701
- 17. Watson DJ. The physiological basis of variation in yield. Adv Agron. 1952;4:101-45. https://doi.org/10.1016/S0065-2113(08)60307-7
- Willams RF. The physiology of plant growth with special reference to the concept of net assimilation rate. Ann Bot. 1946;10:41-72. https://doi.org/10.1093/oxfordjournals.aob.a083119
- Arnon DI. Copper enzymes in isolated chloroplasts. Polyphenol oxidase in *Beta vulgaris*. Plant Physiol. 1949;24:1-15. https:// doi.org/10.1104/pp.24.1.1
- Ahamed KU, Alamin M, Monir MR, Fatima S, Nahar K. Effect of sowing time and irrigation frequency on growth and yield of mustard (*Brassica napus* L.). Int J Adv Agric Sci Tech. 2019;4:01-11.
- 21. Akter C, Islam MM, Parvin S, Shafiqul, M. Response of different mustard varieties to humic acid. J Bio Agric Res. 2023;30:2541-50. https://doi.org/10.18801/jbar.300223.307
- Narayan OP, Kumar P, Yadav B, Dua M, Johri AK. Sulfur nutrition and its role in plant growth and development. Plant Signal Behav. 2023;18:2030082. https://doi.org/10.1080/15592324.2022.2030082
- 23. Verma OP, Singh S, Parihar SS. Growth performance of ethiopian mustard (*Brassica carinata*) with different irrigation, nitrogen and sulphur levels. J Water Manage. 2009;17:126-34.
- Maurya SK, Kalhapure A, Singh N, Kumar A, Yadav P, Kumar M, et al. Growth and yield response of different Indian mustard [*Brassica juncea* (L.)] varieties to irrigation scheduling. Biol Forum. 2021;14:434-39. https://doi.org/10.56093/ijas.v94i4.144818
- Imran A, Amanullah A, Al Tawaha AR. Humic acid and sulfur integration enhance growth and yield-related traits of *Brassica napus* L. J Plant Nut. 2023;10:2303-14. https://doi.org/10.1080/01904167.2022.2128820
- 26. Alizadeh S, Roozbahani A, Rad AHS, Seyedhadi MH. Foliar application

of humic acids improves seed yield and oil quality of rapeseed (*Brassica napus* L.) genotypes at well-time and late planting dates. J Soil Sci Plant Nut. 2022;22:549-59. https://doi.org/10.1007/s42729-021-00670-2

- Hossain MB, Alam MS, Ripon MA. Effect of irrigation and sowing method on yield and yield attributes of mustard. Rajshahi University J Life Earth Agric Sci. 2013;41:65-70. https://doi.org/10.3329/rujleas.v41i0.21626
- Sharma DK, Singh KN. Effect of irrigation on growth, yield and evapotranspiration of mustard (*Brassica juncea*) in partially reclaimed sodic soils. Agric Water Manage. 1993;23:225v232. https://doi.org/10.1016/0378-3774(93)90030
- Mudhoo A, Ramasamy DL, Bhatnagar A, Usman M, Sillanpaa M. An analysis of the versatility and effectiveness of composts for sequestering heavy metal ions, dyes and xenobiotics from soils and aqueous milieus. Ecotoxicol Environ Saf. 2020;197:110587. https:// doi.org/10.1016/j.ecoenv.2020.110587
- 30. Bitew Y, Alemayehu M. Impact of crop production inputs on soil health: A review. Asian J Plant Sci. 2017;16:109-31.
- Ahmad M, Waraich EA, Skalicky M, Hussain S, Zulfiqar U, Anjum MZ, et al. Adaptation strategies to improve the resistance of oilseed crops to heat stress under a changing climate: An overview. Front Plant Sci. 2021;12:767150. https://doi.org/10.3389/fpls.2021.767150
- Ingenbleek Y, Kimura H. Nutritional essentiality of sulfur in health and disease. Nut Rev. 2013;71:413-32. https://doi.org/10.1111/nure.12050
- Singh S, Singh V, Layek S. Influence of sulphur and zinc levels on growth, yield and quality of soybean (*Glycine max* L.). Int J Plant Soil Sci. 2017;2:1-7. https://journalijpss.com/index.php/IJPSS/article/view/193
- Singh MK, Sirothia P, Singh J, Upadhyay PK. Effect of sulphur levels on mustard crops. Int J Curr Microbiol Appl Sci. 2018;10:481v490. https:// www.ijcmas.com/7-10-2018/Manoj20Kumar20Singh,20et20al.pdf