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# Concomitant effect of boron molybdenum and nickel on physiological traits and nodulation of blackgram (Vigna mungo L. Hepper)

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#### Abstract

A field experiment was conducted on the concomitant effect of boron, molybdenum and nickel as basal physiological traits and nodulation of blackgram (*Vigna mungo* L. Hepper) at Agricultural college farm, Bapatla during *rabi*, 2017-18. It consisted of eight treatments of micronutrient application *viz.*, control (T<sub>1</sub>), B (T<sub>2</sub>), Mo (T<sub>3</sub>), Ni (T<sub>4</sub>), B+Mo (T<sub>5</sub>), B+Ni (T<sub>6</sub>), Mo+Ni (T<sub>7</sub>) and B+Mo+Ni (T<sub>8</sub>) with randomized block design concept and replicated thrice. The results revealed that application of B, Mo and Ni individually and in combination increased the CGR and RGR. Increase in NAR was observed with T<sub>3</sub> and T<sub>5</sub> to T<sub>8</sub>. Increase in internal CO<sub>2</sub> concentration and decrease in transpiration rate was obtained with all treatments T<sub>5</sub> to T<sub>8</sub>. Nodule number and nodule weight plant<sup>-1</sup> found increased with B, Mo and combination treatments T<sub>5</sub> to T<sub>8</sub>.

Keywords: Blackgram, boron, molybdenum, nickel, leaf area, crop growth rate

#### Introduction

Pulses, an indispensable source of protein, are one of the important groups of food crops in Indian agriculture after cereals and oilseeds. Blackgram (*Vigna mungo* L. Hepper) is the third important pulse crop grown throughout India and is known as urdbean, urd and urad. The most important constraint limiting crop yield in developing nations worldwide and especially among resource-poor farmers is soil fertility status. The overall strategy for escalating crop yields and sustaining them at a high level must include the management of soil nutrients including both macro and micro. Micronutrients are essential elements that are used by plants in small quantities to orchestrate a range of physiological functions.

Among these micro nutrients, Boron (B) is one of the most important trace elements, which is essential for normal life cycle of the plants and it plays important role in metabolic processes such as nucleic acid metabolism, carbohydrate biosynthesis, photosynthesis and protein metabolism (Pilbeam and Kirkby., 1983)<sup>[8]</sup>. Molybdenum is important for good foliage growth of higher plants. It is involved in the process of nitrogen fixation, nitrate reduction and nitrogen metabolism. *et al.* (2015)<sup>[6]</sup> reported that Mo application significantly increases the leaf area, fresh weight and dry weight of the plant. Significant effect of molybdenum and more nodules with molybdenum application was reported by Awomi *et al.* (2012)<sup>[2]</sup>.

Nickel shows effect on growth and physiological attributes in blackgram. Fresh and dry weights were increased with increased levels of Nickel was reported by Gautam *et al.* (2014)<sup>[3]</sup>. Hence, keeping all these aspects in view, the present investigation on blackgram crop was conducted.

#### **Materials and Methods**

A field experiment was conducted to study the concomitant effect of boron, molybdenum and nickel as basal physiological traits and nodulation of blackgram (*Vigna mungo* L. Hepper) at Agricultural College Farm, Bapatla during *Rabi* season of 2017-18. The experiment consisted of 8 treatments *viz.*,  $T_1$ - no micronutrient application (control),  $T_2$ - Borax @ 2.5 kg ha<sup>-1</sup>,  $T_3$ - Ammonium molybdate @ 1.5 kg ha<sup>-1</sup>,  $T_4$ - Nickel chloride @1 kg ha<sup>-1</sup>,  $T_5$ - Borax @ 2.5 kg and Ammonium molybdate @ 1.5 kg ha<sup>-1</sup>,  $T_6$ - Borax @ 2.5 kg and Nickel chloride @1 kg ha<sup>-1</sup>,  $T_8$ - Borax @ 2.5 kg, Ammonium molybdate @ 1.5 kg and Nickel chloride @1 kg ha<sup>-1</sup>,  $T_8$ - Borax @ 2.5 kg, Ammonium molybdate @ 1.5 kg and Nickel chloride @1 kg ha<sup>-1</sup>,  $T_8$ - Borax @ 2.5 kg, and Nickel chloride @1 kg ha<sup>-1</sup>,  $T_8$ - Borax @ 2.5 kg, Ammonium molybdate @ 1.5 kg and Nickel chloride @1 kg ha<sup>-1</sup>,  $T_8$ - Borax @ 2.5 kg, Ammonium molybdate @ 1.6 kg and Nickel chloride @1 kg ha<sup>-1</sup>,  $T_8$ - Borax @ 2.5 kg, Ammonium molybdate @ 1.6 kg and Nickel chloride @1 kg ha<sup>-1</sup>,  $T_8$ - Borax @ 2.5 kg, Ammonium molybdate @ 1.6 kg and Nickel chloride @1 kg ha<sup>-1</sup>,  $T_8$ - Borax @ 2.5 kg, Ammonium molybdate @ 1.6 kg and Nickel chloride @1 kg ha<sup>-1</sup>,  $T_8$ - Borax @ 2.5 kg, Ammonium molybdate @ 1.6 kg and Nickel chloride @1 kg ha<sup>-1</sup>,  $T_8$ - Borax @ 2.5 kg, Ammonium molybdate @ 1.6 kg and Nickel chloride @1 kg ha<sup>-1</sup> and applied as basal. It was laid in randomized block design and replicated thrice. The soil was neutral in reaction, low in salinity and nitrogen, medium in available phosphorus and organic carbon and very high in potassium.

The standard packages of cultural practices were followed throughout crop growth period. The data on leaf area with leaf area meter; internal CO<sub>2</sub>, transpiration rate and stomatal conductance with IRGA; crop growth rate (CGR), relative growth rate (RGR), net assimilation rate (NAR), and nodule number and weight were recorded and analyzed as per the statistical procedures given by Panse and Sukhatme (1985)<sup>[7]</sup>.

# **Results and Discussion**

#### Leaf area

Application of micronutrients B, Mo and Ni significantly

influenced the leaf area in blackgram. At 20 DAE, maximum leaf area was resulted with  $T_8$  (B + Mo + Ni) (139.1 cm<sup>2</sup>) and it was on par with B+ Mo (T<sub>5</sub>) (136.8 cm<sup>2</sup>) and Mo +Ni (T<sub>7</sub>) (125.1 cm<sup>2</sup>), which was on par with T<sub>6</sub> (B+Ni) (116.9 cm<sup>2</sup>). The leaf area due to B, Mo and Ni application alone was found to be on par with control, which had the minimum (368.9 cm<sup>2</sup>) at 40 DAE and the maximum leaf area was noticed with T<sub>8</sub> (B+Mo+Ni) (534.8 cm<sup>2</sup>), followed by B+Mo (510.4 cm<sup>2</sup>), which was on par with Mo + Ni (500.4 cm<sup>2</sup>) and superior to B+Ni effect (441.6 cm<sup>2</sup>) (Table 1.).

Table 1: Effect of Boron mol	ybdenum and nickel or	n Leaf area and Crop	growth rate (CGR)

Treatments		Leaf area (cm <sup>2</sup> plant <sup>-1</sup> )				Crop growth rate (g m <sup>-2</sup> d <sup>-1</sup> )			
		40	60	At	20-40	40-60	60-		
	DAE	DAE	DAE	harvest	DAE	DAE	harvest		
T <sub>1</sub> : Control	91.8	368.9	332.6	304.7	6.90	10.63	2.49		
T <sub>2</sub> : Borax @ 2.5 Kg ha <sup>-1</sup>	108.0	395.6	371.7	356.8	8.33	12.76	3.27		
T <sub>3</sub> : Ammonium molybdate @ 1.5Kg ha <sup>-1</sup>	100.1	386.9	375.3	371.7	8.63	14.64	2.26		
T <sub>4</sub> : Ni Cl <sub>2</sub> 6H <sub>2</sub> O @1.0 Kg ha <sup>-1</sup>	93.6	376.9	343.5	325.3	8.01	12.82	2.69		
T <sub>5</sub> : Borax @ 2.5&Ammonium molybdate @ 1.5 Kg ha <sup>-1</sup>	136.8	510.4	480.5	431.7	10.34	14.95	3.75		
T <sub>6</sub> : Borax @ 2.5 & Ni Cl <sub>2</sub> . 6H <sub>2</sub> O @1.0 Kg ha <sup>-1</sup>	116.9	441.6	426.7	404.0	9.68	14.08	2.79		
T <sub>7</sub> : Ammonium molybdate @ 1.5 & Ni Cl <sub>2</sub> . 6H <sub>2</sub> O @ 1.0 Kg ha <sup>-1</sup>	125.1	500.4	454.3	415.7	9.88	14.23	3.03		
T8: Borax @ 2.5 & Ammonium molybdate @1.5 & Ni Cl2.6H2O @1.0 Kg	139.1	534.8	511.4	434.0	11.88	15.70	3.58		
ha <sup>-1</sup>	137.1	554.0	511.4	434.0	11.00	15.70	5.56		
SE (m) <u>+</u>	5.3	10.0	8.9	11.8	0.23	0.8	0.3		
CD (0.05)	16.3	30.2	27.2	36.0	0.7	2.6	0.7		
CV (%)	8.1	3.9	3.7	5.4	4.36	10.8	13		

At 60 DAE, leaf area declined and it was less in control followed by Ni application. Maximum of it was noticed in  $T_8$  (B+Mo+Ni) (534.8 cm<sup>2</sup>). At harvest also, the effect of Ni application resulted in leaf area on par with control. The maximum leaf area noticed in  $T_8$  (B+Mo+Ni) was found on par with  $T_5$  (B+Mo),  $T_7$  (Mo+Ni) and  $T_6$  (B+Ni). Leaf area observed in  $T_3$  (Mo alone) showed parity with B+Ni and B alone.

The results obtained clearly indicated that application of B, Mo and Ni in blackgram crop enhances the leaf area and it was found high in combination treatments rather than in individual treatments. The increase in leaf area with B+Mo, B+Ni, Mo+Ni and B+Mo+Ni was 38.7, 19.7, 35.6 and 45.0 percent, respectively over control, while individual application of B and Mo increased it by 7.2 and 4.8 percent, respectively. The higher leaf area might be due to role of these nutrients in increasing the growth of the plant by supplementing the soil available nutrients and influencing various physiological process. The results obtained were in tune with the findings of Malik *et al.* (2015), Sritharan *et al.* (2015) and Sujeet, (2016) <sup>[6, 9, 10]</sup> in their findings.

#### Crop growth rate

The data calculated at three intervals of crop growth i.e. at 20-40 DAE, 40-60 DAE, and 60 DAE-harvest and computed on Crop Growth Rate (CGR) under the influence of micronutrients (B, Mo, Ni) were presented in Table 1. The CGR increased from 20-40 DAE to 40-60 DAE and then decreased, found high during 40-60 DAE. During 20-40 DAE, the higher CGR was recorded in T<sub>8</sub> (11.88). The next higher was noted in T<sub>5</sub> treatment (10.34) which was on parity with T<sub>7</sub> (9.88) and T<sub>6</sub> (9.68) treatments. In alone applications, the higher CGR value was showed by T<sub>3</sub> (Mo) which was on

parity with  $T_2$  and  $T_4(B, Ni)$  and lower CGR was reported with control.

During 40-60 DAE, the higher CGR reported with  $T_8$  treatment (15.70) showed parity with  $T_5$ ,  $T_3$ , $T_7$  and  $T_6$ . The lower CGR reported with control showed parity with  $T_4$  and  $T_2$ . During 60 DAE- harvest, the CGR varied from 2.49 to 3.58 g m<sup>-2</sup> d<sup>-1</sup>. The higher CGR value was recorded in  $T_5$  treatment which was on parity with  $T_8$ ,  $T_7$ ,  $T_4$  and control. The increase in CGR with  $T_2$ ,  $T_3$  &  $T_4$  was 20.0, 33.9 and 20.6 percent respectively, while that in  $T_5$ ,  $T_6$ ,  $T_7$  &  $T_8$  increased by 40.6, 32.4, 33.8 & 47.6 percent, respectively. Alone application of Mo also showed increased CGR greater than B and Ni alone and similar to combination treatments.

The increase of CGR in blackgram with B, Mo and Ni application might be due to the involvement to these nutrients in plant metabolism as structural and functional constituents of enzymes, which further promotes plant water status, chlorophyll synthesis increase in translocation of photosynthetic assimilates and utilization of major and minor nutrients, increase in size and growth rate of organs, accumulation of dry matter and CO<sub>2</sub> assimilation. Mahilane and Singh (2018) <sup>[5]</sup>, Sritharan *et al.* (2015) <sup>[9]</sup> and Wasaya *et al.* (2017) <sup>[11]</sup> reported that B and Mo application increases CGR in blackgram.

#### Net Assimilation Rate

The data pertaining to net assimilation rate (NAR) at different growth stages of blcackgram as affected by basal application of B, Mo and Ni was presented in table 2. It was calculated at 3 intervals of crop i.e during 20-40 DAE, 40-60 DAE and 60 DAE to harvest, high values were recorded during the period of 40-60 DAE. The NAR values increased from 20-40DAE to 40-60DAE and then decreased.

	<b>Relative Gro</b>	$(mg g^{-1} d^{-1}))$	Net Assimilation Rate (mg cm <sup>-2</sup> d <sup>-1</sup> )			
Treatments	20-40 DAE	40-60	60 DAE -	20-40	40-60	60 DAE -at
		DAE	at Harvest	DAE	DAE	Harvest
T1: Control	56.9	42.2	6.3	1.220	1.898	0.446
T2: Borax @ 2.5 Kg ha <sup>-1</sup>	74.3	42.2	7.4	1.490	2.148	0.626
T3: Ammonium molybdate @ 1.5Kg ha <sup>-1</sup>	79.3	45.1	4.4	1.538	2.589	0.417
T4: Ni Cl <sub>2</sub> . 6H <sub>2</sub> O @1.0 Kg ha <sup>-1</sup>	68.8	43.6	5.7	1.421	2.076	0.487
T5: Borax @ 2.5&Ammonium molybdate @ 1.5 Kg ha <sup>-1</sup>	91.2	41.0	6.8	1.930	2.904	0.710
T6: Borax @ 2.5 & Ni Cl <sub>2</sub> . 6H <sub>2</sub> O @ 1.0 Kg ha <sup>-1</sup>	87.1	40.9	5.2	1.765	2.521	0.516
T7: Ammonium molybdate @ 1.5 & Ni Cl <sub>2</sub> 6H <sub>2</sub> O @ 1.0 Kg ha <sup>-1</sup>	88.8	40.7	5.6	1.839	2.700	0.570
T8: Borax @ 2.5 & Ammonium molybdate @ 1.5 & Ni Cl <sub>2.6</sub> H <sub>2</sub> O @1.0 Kg ha <sup>-1</sup> .	100.5	38.7	5.9	2.235	3.029	0.669
SE(m) <u>+</u>	1.6	0.8	0.1	0.04	0.15	0.05
CD(0.05)	4.9	2.4	0.4	0.13	0.545	0.15
CV (%)	3.4	3.4	4.0	4.51	10.3	10.5

Table 2: Effect of Boron Molybdenum and Nickel on Relative growth rate (RGR) and Net assimilation rate (NAR)

During 20-40DAE, in  $T_8$  treatment (2.235) significantly higher value was observed and lower NAR was observed in control. The next higher NAR was recorded in  $T_5$  treatment (1.930), which showed parity with  $T_2$  treatment and  $T_7$  was in parity with  $T_6$  treatment (1.765). In alone Mo alone showed higher values than remaining alone applications. During 40-60 DAE, the higher NAR was reported with the combined application of B+Mo+Ni ( $T_8$ -3.029) which showed parity with  $T_5$  (2.904), T7(2.7), T3(2.589) and  $T_6$  (2.521). The lower NAR was observed in control, which showed parity with alone application of B and Ni i.e.  $T_2$  and  $T_4$  treatments.

During 60 DAE to harvest, higher value of NAR was observed in  $T_5$  treatment (0.710) which showed parity with  $T_8$  (0.669),  $T_2$  (0.626) and  $T_7$  (0.570). The lower NAR value was recorded in control which showed parity with  $T_5$ ,  $T_3$  and  $T_4$  based on the above results, alone treatments  $T_2$  and  $T_4$  did not show increase in NAR over control. The treatments  $T_5$  to  $T_8$  and  $T_3$  increased the NAR by 1.5, 1.3, 1.4, 1.6 and 1.4 folds over the control. The increase in NAR is due to increase in leaf area, chlorophyll, photosynthesis in young leaves and dry matter production with micronutrient treatments. Sritharan *et al.* (2015) <sup>[9]</sup> reported increase in NAR of blackgram with application of B and Mo along with other nutrients and plant growth regulator.

# **Relative Growth Rate**

The data pertaining to Relative Growth Rate (RGR) was recorded at an interval of 20 days from 20 DAE and the means were presented in table 2. The RGR showed a gradual decrease from 20-40 DAE to 60 DAE- Harvest. Higher RGR was recorded at 20-40 DAE followed by 40-60 DAE. The RGR recorded during the period of 20-40 DAE, was significantly higher in  $T_8$  treatment (100.5). The next higher value was recorded in  $T_5$  (91.2) which showed parity with  $T_7(88.8)$  and  $T_6(87.1)$ . In alone application the higher RGR value was recorded in  $T_3(79.3)$  followed by  $T_2(74.3)$  and  $T_4(68.8)$  and lowest was observed in control.

During 40-60 DAE, the significantly higher RGR was observed in  $T_3$  treatment (Mo) and lower RGR was in  $T_8$ compared to control, which was in parity with remaining treatments. During 60 DAE to harvest, B and B+Mo treated plants had higher RGR and the remaining had lower RGR than control. These results indicated that RGR decreased continuously from 40 DAE, the increase in RGR in  $T_2$  to  $T_4$ during 20-40 DAE was 1.3, 1.4 and 1.2 folds respectively, and in combination treatments  $T_5$  to  $T_8$ , RGR increased by 1.6, 1.5, 1.6 and 1.8 folds respectively. This increase is due to increase in leaf area, chlorophyll content and photosynthesis all these factors reflected the increase in RGR and total biomass.

#### Internal CO<sub>2</sub>

The data with respect to effect of micronutrients on internal  $CO_2$  concentration in leaves of blackgram were presented in Table 3. The data revealed that B, Mo and Ni application significantly influenced the internal  $CO_2$  concentration in blackgram leaves over control.

Treatments		Internal CO <sub>2</sub>			Stomatal conductance (m mol m <sup>-2</sup> s <sup>-1</sup> )			Transpiration rate (m mol m <sup>-2</sup> s <sup>-1</sup> )		
		40 DAE	60	20	40 DA E	60 DAE	20 DAE	40 DAE	60 DAE	
	DAE	DAE	DAE		DAE		DAE	DAE	DAE	
T1: Control	285.3	358.5	317.4	115.8	146.0	127.6	3.15	3.78	3.56	
T2: Borax @ 2.5 Kg ha <sup>-1</sup>	316.7	404.2	383.2	131.7	166.9	142.5	2.37	2.80	2.52	
T3: Ammonium molybdate @ 1.5Kg ha <sup>-1</sup>	304.3	374.3	318.0	139.3	169.1	145.9	2.10	3.02	2.80	
T4: Ni Cl <sub>2</sub> 6H <sub>2</sub> O @1.0 Kg ha <sup>-1</sup>	317.7	384.2	354.6	128.0	159.7	136.1	2.45	2.83	2.66	
T5: Borax @ 2.5&Ammonium molybdate @ 1.5 Kg ha <sup>-1</sup>	475.2	594.3	577.8	170.8	222.9	185.5	1.61	2.27	2.02	
T6: Borax @ 2.5 & Ni Cl <sub>2</sub> 6H <sub>2</sub> O @ 1.0 Kg ha <sup>-1</sup>	490.8	537.6	530.5	149.5	183.0	161.0	1.91	2.48	2.33	
T7: Ammonium molybdate @ 1.5 & Ni Cl <sub>2</sub> .6H <sub>2</sub> O @ 1.0 Kg ha <sup>-1</sup>	502.0	553.0	549.0	164.5	195.2	173.6	1.99	2.56	2.42	
T8: Borax @ 2.5 & Ammonium molybdate @ 1.5 & Ni Cl <sub>2.6</sub> H <sub>2</sub> O @ 1.0 Kg ha <sup>-1</sup> .	540.3	664.3	650.3	213.0	285.9	240.3	1.43	1.87	1.73	
SE(m) <u>+</u>	3.3	15.5	7.8	2.7	9.2	8.6	0.05	0.06	0.13	
CD(0.05)	10.0	47.4	23.7	8.2	28.2	26.4	0.15	0.17	0.39	
CV (%)		5.5	2.9	3.0	8.3	9.1	4.08	3.51	8.69	

Table 3: Effect of Boron Molybdenum and Nickel on Internal CO2, Stomatal conductance and Transpiration rate

At 20 DAE, among the treatments  $T_2$  to  $T_8$ , higher internal  $CO_2$  concentration was noted in  $T_8$ , the lower was noted in Mo application alone followed by B and Ni alone, which were on par with each other. At 40 DAE,  $T_8$  recorded the maximum internal  $CO_2$ . The next in the order was  $T_5$ , followed by  $T_7$  that showed parity with  $T_6$ . Minimum internal  $CO_2$  concentration was recorded in control which was on par with  $T_2$ ,  $T_3$  and  $T_4$ . At 60 DAE, among all the treatments,  $T_8$  recorded higher internal  $CO_2$  concentration followed by  $T_5$ . The treatment  $T_6$  and  $T_7$  recorded the value on par with each other. Lower internal  $CO_2$  concentration was recorded in control which was on par with  $T_3$ . Application of B alone resulted in more internal  $CO_2$  concentration than Ni application alone.

It can be concluded Application of these nutrients in combination resulted in higher values of internal CO<sub>2</sub> concentration than individual application. At 20 DAE, increase in internal CO<sub>2</sub> concentration was 1.9 folds in T<sub>8</sub>, 1.8 in T<sub>7</sub>, 1.7 in T<sub>5</sub> and T<sub>6</sub> and 1.1 folds in T<sub>2</sub>, T<sub>3</sub> and T<sub>4</sub>.Same trend was noted with T<sub>8</sub> and T<sub>5</sub> at 40 DAE, while in T<sub>7</sub> and T<sub>6</sub> internal CO<sub>2</sub> concentration increased by 1.5 folds. At 60 DAE, individual application of B and Ni increased it by 1.2 and 1.1 folds respectively and combination treatments increased it by 1.7 to 2.0 folds. This increase in internal CO<sub>2</sub> concentration could be due to the increase in plant water status, turgidity of cells and increase in stomatal conductance caused by the supplementation of B, Mo and Ni as observed in the present investigation.

#### Stomatal conductance

The data regarding to the effect of micronutrients (B, Mo and Ni) on stomatal conductance of blackgram leaves recorded at 20 days interval from 20 DAE were presented in Table 3. At 20 DAE, all micronutrient treatments showed significantly higher stomatal conductance than control. Among the treatments,  $T_2$  to  $T_8$ , B+Mo+Ni treated plants recorded higher stomatal conductance (213.0), next in the order were B+Mo, Mo+Ni and B+Ni. The lower stomatal conductance was recorded by Ni treated plants, which showed parity with B application. Mo applied plants had stomatal conductance greater than Ni treated plants, on par with boron treated plants.

At 40 DAE, stomatal conductance observed in plants treated with, B, Mo and Ni alone showed parity with control. Maximum stomatal conductance was found in  $T_8$ , next in order were  $T_5$ ,  $T_7$  and  $T_6$ . At 60 DAE, also stomatal conductance observed in plants treated with B, Mo and Ni alone showed parity with control. The maximum was observed in  $T_8$  followed by  $T_5$ ,  $T_7$  and  $T_6$ , which were on par with each other.

In summary, these results envisaged that B, Mo and Ni application has the positive impact on stomatal conductance in leaves of blackgram. The treatments  $T_2$ ,  $T_3$  and  $T_4$  enhanced this parameter at 20 DAE only. B, Mo and Ni application in combination had remarkable effect on stomatal conductance (1.3, 1.3 to 1.4, 1.5 and 1.8 to 2.0 folds

respectively in  $T_6$ ,  $T_7$ ,  $T_5$  &  $T_8$  respectivey). Compared to the Mo+Ni and B+Ni treatments, greater enhancement in stomatal conductance was attained with B+Mo and B+Mo+Ni. In plants that received the B, Mo and Ni, leaves maintained the higher RWC, which would result in increased turgidity of cells including guard cells and this might have facilitated the increase in stomatal conductance. Higher nutrient uptake and photosynthetic rate might have resulted in increased turgidity of guard cells and thus higher stomatal conductance.

# Transpiration rate

Data with respect to the effect of micronutrients on transpiration rate of blackgram leaves were presented in Table 3. At 20 DAE, plants treated with B+Mo+Ni had minimum transpiration rate followed by T<sub>5</sub>, T<sub>7</sub>, T<sub>6</sub> and T<sub>3</sub> in the order. The treatments T<sub>6</sub>, T<sub>7</sub> and T<sub>3</sub> were on par with each other. Maximum transpiration rate was recorded in control plants. Followed by T<sub>2</sub> and T<sub>4</sub> which were on par with each other. At 40 DAE, among all the treatments, T<sub>8</sub> recorded lower transpiration rate followed by T<sub>5</sub>, T<sub>6</sub> &T<sub>7</sub> in the order. The treatment T<sub>6</sub> and T<sub>7</sub> were on par with each other. Higher transpiration rate was recorded in control followed by T<sub>3</sub>, T<sub>4</sub> & T<sub>2</sub> in the order. The treatments T<sub>2</sub> and T<sub>4</sub> were on par with each other.

At 60 DAE, lower transpiration rate was recorded in  $T_8$  followed by  $T_5$ ,  $T_6$ ,  $T_7 \& T_2$  in the order. The treatment  $T_6$ ,  $T_7$  and  $T_2$  were on par with each other. Higher transpiration rate was observed in control followed by  $T_3$  and  $T_4$  which were on par. To summarize, transpiration rate was found high at 40 DAE compared to that at 20 and 60 DAE. Transpiration rate declined in plants with combined application of micronutrients compared to individual application and control. Individual application decreased the transpiration rate by 1.3 to 1.5 folds at all stages. Where as the combination treatments decreased the transpiration rate by 1.6-2.2, 1.5-2.0 and 1.5-2.1 folds, respectively at 20, 40 and 60 DAE.

# Nodule number plant<sup>-1</sup>:

Application of B, Mo and Ni significantly influenced the number of nodules plant<sup>-1</sup> (Table 4). Combined application showed more effect than alone application. At 20 DAE, nodules  $plant^{-1}$  were less in control, found on par with T<sub>2</sub> and T<sub>4</sub> application alone. Nodule formation was more in the plants treated with three nutrients, significantly superior to rest of the treatments. The treatments  $T_5$  and  $T_7$  stood second in exhibiting the impact followed by  $T_6$  and  $T_3$  in the order. At 40 DAE, nodule formation was maximum in T<sub>8</sub>. The next higher value was noticed in T<sub>7</sub>, T<sub>5</sub> and T<sub>6</sub>, which were on par with each other. The minimum was noticed in control which showed parity with nickel application. Among rest of the treatments, the influence of Mo&Ni, B&Mo and B& Ni was on par with each other and greater (27.0, 26.6 & 26.0, respectively) than Mo (24.6) and B(23.6). Parity was observed between T<sub>2</sub> & T<sub>3</sub> and T<sub>3</sub> & T<sub>6</sub>.

1 reatments	Number			Weight of the Nodules (mg plant <sup>-1</sup> )			
		<b>40 DAE</b>	60 DAE	20 DAE	40 DAE	60 DAE	
T1: Control	4.7	21.7	5.0	6.3	93.8	8.0	
T2: Borax @ 2.5 Kg ha <sup>-1</sup>	5.0	23.7	6.0	7.0	98.6	9.0	
T3: Ammonium molybdate @ 1.5Kg ha <sup>-1</sup>	5.7	24.7	7.7	7.3	100.6	10.5	
T4: Ni Cl <sub>2</sub> . 6H <sub>2</sub> O @1.0 Kg ha <sup>-1</sup>	5.3	22.3	6.0	6.5	94.5	8.8	
T5: Borax @ 2.5&Ammonium molybdate @ 1.5 Kg ha <sup>-1</sup>	7.7	26.7	9.3	8.0	103.1	14.0	
T6: Borax @ 2.5 & Ni Cl <sub>2</sub> .6H <sub>2</sub> O @ 1.0 Kg ha <sup>-1</sup>	6.7	26.0	8.3	7.0	105.1	13.1	
T7: Ammonium molybdate @ 1.5 & Ni Cl <sub>2</sub> 6H <sub>2</sub> O @ 1.0 Kg ha <sup>-1</sup>	7.3	27.0	8.7	8.4	101.8	13.7	
T8: Borax @ 2.5 & Ammonium molybdate @ 1.5 & Ni Cl <sub>2</sub> 6H <sub>2</sub> O @ 1.0 Kg ha <sup>-1</sup> .	8.7	30.0	11.0	10.0	107.	15.0	
SE(m) <u>+</u>	0.3	0.6	0.5	0.3	0.9	0.4	
CD(0.05)	0.9	1.7	1.5	0.8	2.7	1.2	
CV (%)		3.9	11.1	6.2	1.5	5.8	

Table 4: Effect of Boron, Molybdenum and Nickel on Nodule number and weight (mg plant<sup>-1</sup>) in Blackgram

At 60 DAE, minimum number was found in control, which showed parity with application of B and Ni alone. Maximum number of it was found in application of three nutrients, followed by  $T_5$  combination that showed parity with  $T_7$ ,  $T_6$ , which were on par with T<sub>3.</sub> Application of B increased the nodule number by 8.8 and Mo by 13.3 percent, where as the combination  $(T_5 to T_8)$  treatments increased the nodule number by 22.5, 19.8, 24.4 and 38.2 percent, respectively. More number of nodules in Mo applied plants either independently or in combination with B and Ni could be due to the role of three nutrients in the process of nitrogen fixation and nodule tissue formation, that ultimately results in better nodule count and these findings were in conformity with many researchers, who reported an increase in the nodulation with these nutrients. Alam et al. (2017), Awomi et al. (2012) and Khan et al. (2014)<sup>[1, 2, 4]</sup> were also found same results.

# Weight of Nodules (mg plant<sup>-1</sup>)

The influence of micronutrients basal application on weight of nodules was found significant Table 4. At 20 DAE, weight of nodules plant<sup>-1</sup> in control was less (6.3 mg), showed parity with  $T_2$ ,  $T_4$  &  $T_6$  and differed significantly with other treatments. It was paramount in plants treated with B+Mo+Ni. The next high value was noticed in  $T_7$  and  $T_6$  application. The difference in nodule weight between the treatments  $T_3$  and  $T_5$  was not significant.

At 40 DAE, the treatments resulted in nodule weight varying from 93.8 to 107.0 mg plant<sup>-1</sup>. It was minimum in control and nickel applied plants. Maximum weight was noticed in  $T_8$ followed by  $T_6$  which showed parity with  $T_5$ . Significant variation was not observed in nodule weight recorded in  $T_5$ ,  $T_7$  and  $T_3$ . Similarly no difference was noticed between  $T_3$ and  $T_2$ . At 60 DAE, maximum weight of nodules was found in  $T_8$ , followed by  $T_5$ , which was on par with  $T_7$  and  $T_6$ . Minimum weight was found in control and it did not differ with  $T_2$  and  $T_4$ . Molybdenum ( $T_3$ ) application showed moderate weight.

In summary, these results clearly conformed that B, Mo and Ni application in blackgram crop enhanced the nodule weight. Application of these nutrients in combination increased the nodule number there by increased nodule weight, size and greater impact than its individual application. The increase in nodule weight with  $T_2$  and  $T_3$  over control was 5.0 and 7.2 percent respectively, while that in  $T_5$ ,  $T_6$ ,  $T_7$  and  $T_8$  increased by 10.0, 12.0, 8.5 and 14.0 percent respectively. Mo and B as it promotes the merestamatic and vascular development of root nodules (Rathi., 2016) structural constituent of enzymes nitrogenase and nitrate reductase exhibit spectacular effects on dry and fresh weight of nodules and it directly reflects the

increase in the capacity of nitrogen fixation and with increased nodule number and size the fresh and dry weight of nodules also increased as per findings of present investigation and these results supports the findings of Awomi *et al.*(2012) <sup>[2]</sup> and Khan *et al.*(2014) <sup>[4]</sup>

# Conclusion

From the present study it can be informed that, individual application of B, Mo and Ni had less increase in physiological effects like Leaf area, CGR, RGR, NAR, Internal CO<sub>2</sub>, transpiration rate, stomatal conductance and number of nodules and weight of nodule plant<sup>-1</sup> when compared with same nutrients applied as combination. The increase of all traits was significantly increased and observed more significant difference between the treatments when the nutrients applied combinedly.

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