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Effect of Combination of Humic Acid and Chelated Potassium on Crop Production and Qualitative Traits of Mung bean (*Vigna radiate* L.) under Drought Stress Conditions

Morteza Jalizavi[1], Mani Mojaddam*[2]

1- Msc. Student, Department of Agronomy, Ahvaz Branch, Islamic Azad University, Ahvaz, Iran.
2- Associate Professor, Department of Agronomy, Ahvaz Branch, Islamic Azad University, Ahvaz, Iran.

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ABSTRACT

BACKGROUND: The management of fertilizers is one of the important factors that greatly affect the growth, development and yield of mung bean.

OBJECTIVES: This study was conducted to investigation effect of different irrigation levels and combination humic acid and chelated Potassium on quantitative and qualitative traits of Mung bean.

METHODS: This research was carried out via split plot experiment based on randomized complete blocks design (RCBD) with three replications along 2023 year. The main factor included drought stress (80 mm evaporation from class A evaporation pan or control, 120 mm evaporation from class A evaporation pan, 160 mm evaporation from class A evaporation pan) based on the daily evaporation reported from Ahvaz weather station, the closest station to the test site. Also biologic fertilizer (nonuse of biofertilizer or control, 4 $L.ha^{-1}$ humic acid, 4 $kg.ha^{-1}$ Chelated Potassium, Combination of 4 $L.ha^{-1}$ humic acid and 4 kg.ha-1 Chelated Potassium) belonged to sub plots.

RESULT: Result of analysis of variance revealed effect of drought stress and biologic fertilizer on all measured traits was significant but interaction effect of treatments (instead seed yield) was not significant. Mean comparison result of different level of drought stress indicated that maximum amount of all measured traits (instead protein content) was for 80 mm evaporation from class A evaporation pan. Also as for Duncan classification made with respect to different level of biologic fertilizer maximum and minimum amount of measured traits belonged to combination of 4 $L.ha^{-1}$ humic acid and 4 $kg.ha^{-1}$ Chelated Potassium and control.

CONCLUSION: Therefore, in general, it can be stated that in order to achieve the maximum crop production and qualitative components, planting mung bean with 80 mm from evaporation pan with combined application of humic acid and potassium chelate fertilizer in studied areas will be recommended.

KEYWORDS: *Chlorophyll, Leaf area index, Pod length, Protein, Seed yield.*

***Corresponding Author:** Mani Mojaddam ✉ manimojaddam@yahoo.com

1. BACKGROUND

Fabaceae family is one of the most important sources of protein and energy. Food legumes, after maize, wheat and rice, are the most important crops used for feeding the world's people, especially in developing countries (Mohtashami *et al*., 2016a). By having 17-40% protein, legumes play an important role in the production of protein and calories needed by humans (Mohtashami *et al*., 2016b). The increasing need of society to food and irregular growth of the population and reduction of food sources is one of the most important issues that have attracted politicians, thinkers and researchers' attention. Iran will need food approximately 2 times more than the current figure until another 20 years with current consumption level. The population growth and economic and social development of the country in the past two decades has led to a dramatic increase in the consumption of protein, especially red meat. Accordingly, the increased production of protein substances, especially plant proteins which are more valuable sources in nutrition, is inevitable. Legumes, by having approximately 20% of protein and sometimes more, play an important role in providing the protein needed by human beings, especially in countries with low animal and agricultural products. Legumes in human nutrition can be considered as a good dietary supplement for cereals (Mahmoudi *et al*., 2016). Potassium mainly exists in three different forms in the soil, which includes usable, stabilized potassium and potassium found in soil minerals. Therefore, in order to supply potassium needed by the plant, soluble and exchangeable potassium should be provided by adding chemical fertilizers or by releasing stabilized potassium and weathering minerals containing potassium (such as mica and feldspar) (Tabatabaei, 2014). In a research investigating the effect of potassium sulfate fertilizer on mung bean, it was reported that the yield of pod increased with increasing the amount of potassium sulfate fertilizer from 0 to 50 kg.ha^{-1} under favorable irrigation conditions with a steep slope, which shows that the yield of pod increased to some extent with the increase of fertilizer. Potassium sulfate has increased and since then, with the increase of potassium sulfate fertilizer from 50 to 100 kg.ha^{-1}, the yield of pod has decreased rapidly (Ali *et al*., 2010). Researchers reported that the use of potassium sulfate in mung bean is directly related to growth, biomass and yield. The results showed that the yield response to different levels of potassium (0, 100, 120 kg.ha^{-1}) is different. The lowest yield (700 kg.ha^{-1}) was obtained with no potassium application (control treatment) and the highest yield (1096 kg.ha^{-1}) was obtained with the application of 120 kg.ha^{-1} potassium (Kumar *et al*., 2014). Asghar *et al*. (2006) by examining the effect of potassium sulfate (at three levels of 0, 150, 250 kg.ha^{-1}) on the yield of mung bean cultivars, they showed that the highest number of seeds per pod was obtained from the treatment of 250 kg.ha^{-1} of potassium fertilizer which was consistent with the results of this research. In arid and semi-arid areas of the world, water is the principal limiting factor of agricultural production primarily due to low and/or uneven distributions of annual rainfall (Keshavarz Afshar *et al*., 2014). Increasing the yield requires the use of proper agricultural management in each region and the knowledge of physiological relationships of plant with the agricultural systems (Hassani *et al*., 2015). Sadeghipour (2018) stated the effect of the irrigation cycle on the yield and its components was significant. The treatment without irrigation in the flowering stage reduced the number of pods in the plant, the number of seeds per pod and the seed yield, while the limited irrigation in the pod filling stage reduced the 1000-seed weight. Efficient cell development and growth of plant tissues, transport, storage of assimilates and other internal actions which are mostly based on physiological, biochemical and biophysical interactions, need sufficient potassium in the cell sap. Potassium plays a role in plant water status and overcomes soil moisture stress (Marscner, 1995). The role of potassium in reducing the effects of stresses such as drought, cold and high light intensity in plants has been reported (Waraich *et al*., 2012).

2. OBJECTIVES

This study was conducted to investigation effect of different irrigation levels and combination humic acid and chelated Potassium on quantitative and qualitative traits of Mung bean.

3. MATERIALS AND METHODS

3.1. Field and Treatments Information

This research was carried out via split plot experiment based on randomized complete blocks design with three replications along 2023 year. Place of research was located in Dashteazadegan region at longitude 48°10'E and latitude 31°33'N in Khuzestan province (Southwest of Iran). The main factor included drought stress (I_1: 80 mm evaporation from class A evaporation pan or control, I_2: 120 mm evaporation from class A evaporation pan I_3: 160 mm evaporation from class A evaporation pan) based on the daily evaporation reported from Ahvaz weather station, the closest station to the test site. Also biologic fertilizer (F_1: nonuse of biofertilizer or control, F_2: 4 $L.ha^{-1}$ humic acid, F_3: 4 $kg.ha^{-1}$ Chelated Potassium, F_4: Combination of 4 $L.ha^{-1}$ humic acid and 4 $kg.ha^{-1}$ Chelated Potassium) belonged to sub plots. This experiment had 36 plots. Each plot consisted of 6 lines with a distance of 50 cm and 5 meters length. The distance between the shrubs on every row was 15 cm.

3.2. Farm Management

Base fertilizers (50 $kg.ha^{-1}$ Nitrogen from urea, 80 $kg.ha^{-1}$ phosphorus from triple superphosphates and 80 $kg.ha^{-1}$ potassium from potassium sulfate) were added to the soil based on soil tests and the recommendations of the Iranian Soil and Water Research Institute at planting stage. 50 $kg.ha^{-1}$ Nitrogen from urea was added at stem elongation stage.

The light-disk harrow was used to mix the soil and the fertilizer after soil fertilization. The furrower was used to make furrows at a distance of 50 cm. The zinc and manganese Nano-chelate were used in the furrows (with 4cm depth) before planting. The furrows were covered with soil. The seeds were planted 2 cm above the fertilizer. Potassium fertilizer based on 35% potassium chelate was purchased from Sepehr Parmis Technology Company and humic acid containing 12% fulvic acid and 68% humic acid was purchased and used from Arman Sabz Adina Company. The application of humic acid and potassium fertilizer was done twice during crop growing season at before flowering and before podding stages. The application of humic acid and potassium chelate according advised amounts based on the treatment with irrigation water was used in the field. Weeds were controlled by manual weeding. Physical and chemical properties of the soil are mentioned in table 1.

Table 1. Physiochemical characteristics of field soil

Soil depth (cm)	OC (%)	SP (%)	P (ppm)	K (ppm)	pH	EC ($ds.m^{-1}$)	Soil texture
0-15	0.47	47	10.1	210	7.1	4.42	Clay loam
15-30	0.32	45	9.8	198	7.0	4.21	Clay loam

3.3. Measured Traits

After removing the border of plots, collect all pods in three, four and five planting lines (an area equal to two square meters) and after separating the seeds from the pods, the seeds are heated at 75 degrees for 48 hours was put in oven. Finally, the seed yield was determined based on grams per square meter. The height of 10 plants was randomly measured from the crown to the end of the stem and their average was considered as the plant height for each plot. Nitrogen content of the seeds was measured using with the Kjeldahl method (included nitrogen digestion, distillation and titration). The seed protein content was obtained by multiplying the seed nitrogen percent by 6.25. Protein yield was achieved by multiplying protein content by seed yield (Linn and Martin, 1999). To determine the leaf area of the linear relationship S= K. L.W was used in which S, L and W were the leaf area, L and W respectively, the maximum length and width of each leaf and K= 0.75 correction coefficient. The leaf area index was calculated from leaf area ratio to ground level. Chlorophyll index was measured with a Spad meter device, model (spad-502) available in the laboratory of Islamic Azad University of Ahvaz, with an average of 10 leaves from the middle of the plant and from three parts of the leaf at 9:30-10 the morning during flowering from Each plot.

3.4. Statistical Analysis

Analysis of variance and mean comparisons were done via SAS (Ver.8) software and Duncan multiple range test at 5% probability level.

4. RESULT AND DISCUSSION

4.1. Plant height

According result of analysis of variance effect of drought stress and biologic fertilizer on plant height was significant at 1% probability level but interaction effect of treatments was not significant (Table 2). Mean comparison result of different level of drought stress indicated that maximum plant height (54.07 cm) was noted for 80 mm evaporation from class A evaporation pan and minimum of that (43.25 cm) belonged to 160 mm evaporation from class A evaporation pan (Table 3).

Table 2. The results of analysis of variance of measured traits

S.O.V	df	Plant height	Pod length	Leaf area index	Chlorophyll index	Seed yield	Protein content
Replication	2	3.01^{ns}	0.24^{ns}	0.011^{ns}	10.17^{ns}	845.3^{ns}	0.08^{ns}
Drought stress (I)	2	388.70**	92.35**	0.618*	146.51*	45608.9**	24.10*
Error I	4	14.6	0.972	0.071	19.76	337.11	6.15
Bio fertilizer (F)	3	453.02**	66.19**	1.105**	115.31*	27609.2**	48.27*
I × F	6	3.92^{ns}	0.041^{ns}	0.012^{ns}	4.24^{ns}	8834.05**	0.33^{ns}
Error II	18	11.68	0.781	0.053	17.93	280.6	4.19
CV (%)	-	7.04	9.11	10.42	7.81	9.24	9.50

$^{ns,\ *\ and\ **}$: no significant, significant at 5% and 1% of probability level, respectively.

As for Duncan classification made with respect to different level of biologic fertilizer maximum and minimum amount of plant height belonged to combination of humic acid and chelated Potassium (56.01 cm) and control (42.30 cm) (Table 3). Golgul *et al*. (2023) reported under drought stress, there is a competition between aerial (stem) and ground (root) parts to obtain food and in this competition, a larger share of photosynthetic materials is allocated to the root and as a result, less photosynthetic materials are allocated to the stem, which is one of the aerial parts of the plant as a result, the height of the plant may decrease. Zyada *et al*. (2020) reported that humic acid foliar application increased plant height. Other researchers such as Tripura *et al*. (2017) and Kuntyastuti *et al*. (2019) have also pointed out the positive role of humic acid and potassium chelate fertilizer in increasing plant height, which was consistent with the results of this research.

4.2. Pod length

Result of analysis of variance revealed effect of drought stress and biologic fertilizer on pod length was significant at 1% probability level, but interaction effect of treatments was not significant (Table 2). According result of mean comparison maximum of pod length (12 cm) was obtained for 80 mm evaporation from class A evaporation pan and minimum of that (7.78 cm) was for 160 mm evaporation from class A evaporation pan (Table 3). Skandarnejad *et al*. (2017) reported that the reduction of pod length is the reason that drought stress has reduced cell divisions and cell elongation, and the vegetative growth of the plant has been reduced, so the biological yield of the plant has decreased. Sadeghipour and Aghaei (2014) reported an increase in the number of seeds per pods under non-stress conditions than to stress situation was 25.64%. That matter was related to more photosynthesis and greater transfer of photosynthetic material to the seeds. It seems crop affected drought stress during flowering stages due to the short flowering period and sterility of some of their flowers due to pollen germination and lack of proper fertilization led to reduce the number of pods and seeds compared to normal conditions (Najarian *et al.*, 2016).

Table 3. Mean comparison different level of drought stress and biologic fertilizer on studied traits

Treatment	Plant height (cm)	Pod length (cm)	Leaf area index	Chlorophyll index	Seed yield (g.m^{-2})	Protein content (%)
Drought stress						
80 mm evaporation from class A evaporation pan	54.07a	12.0a	2.86a	57.43a	245.71a	19.50c
120 mm evaporation from class A evaporation pan	48.12b	9.32b	2.10b	54.20b	169.11b	21.34b
160 mm evaporation from class A evaporation pan	43.25c	7.78c	1.67c	51.0c	128.60c	24.65a
Biologic fertilizer						
Nonuse of biofertilizer or control	42.30c	8.22c	1.70c	49.11c	143.58c	20.01c
4 L.ha^{-1} humic acid	49.11b	9.03bc	2.0b	55.30b	180.04b	21.76b
4 kg.ha^{-1} Chelated Potassium	46.5b	10.0b	2.24ab	54.19b	175.26b	22.0ab
Combination of 4 L.ha^{-1} humic acid and 4 kg.ha^{-1} Chelated Potassium	56.01a	11.55a	2.90a	58.24a	225.68a	23.55a

*Mean which have at least once common letter are not significant different at the 5% level using (DMRT).

Evaluation means comparison result indicated in different level of biologic fertilizer the maximum pod length (11.55 cm) was noted for combination of humic acid and chelated Potassium and minimum of that (8.22 cm) belonged to control treatment (Table 3). By investigating and studying different levels of potassium sulfate in mung bean plant, it was reported that the number of pods plant, seed and biological yield were significantly affected by potassium. The lowest number of pods and the number of seeds per pod were obtained from the treatment without potassium (control) (Abbas *et al*., 2011). According to the findings of Maqbool *et al*. (2018), the reduction of pod length in drought stress conditions can be due to the plant's sensitivity to drought stress in the reproductive growth stage, which is consistent with the results of this research.

4.3. Leaf area index (LAI)

According result of analysis of variance effect of drought stress and biologic fertilizer on LAI was significant at 5% and 1% probability level, respectively but interaction effect of treatments was not significant (Table 2). Assessment means comparison result indicated in different level of drought stress the maximum LAI (2.86) was noted for 80 mm evaporation from class A evaporation pan and minimum of that (1.67) belonged to 160 mm evaporation from class A evaporation pan treatment (Table 3). The results of the research of Salehi *et al*. (2017) and Shokouhfar and Abofatilehnezhad (2013) confirm that drought stress has a significant effect on the leaf area index, and with the increase of drought stress, the LAI decreased significantly, which is consistent with the results of this study. Compare different level of manganese Nano-chelate showed that the maximum and the minimum amount of LAI belonged to combination of humic acid and chelated Potassium (2.9) and control (1.7) treatments (Table 3). Zyada *et al*. (2020) stated that the increase in potassium absorption causes a positive effect on photosynthesis, increasing the growth rate and the leaf area index of the plant. The increase in leaf area index with the application of humic acid and potassium chelate was also reported by Sadaf and Tahir (2017) and Eryigit and Husamalddin (2023), which was consistent with the results of this research.

4.4. Chlorophyll index

Result of analysis of variance revealed effect of drought stress and biologic fertilizer on chlorophyll index was significant at 5% probability level, but interaction effect of treatments was not significant (Table 2). Mean comparison result of different level of drought stress indicated that maximum chlorophyll index (57.43) was noted for 80 mm evaporation from class A evaporation pan and minimum of that (51) belonged to 160 mm evaporation from class A evaporation pan (Table 3). As for Duncan classification made with respect to different level of biologic fertilizer maximum and minimum amount of chlorophyll index belonged to combination of humic acid and chelated Potassium (58.24) and control (49.11) (Table 3). It seems that the higher chlorophyll index in fertilizer treatments is due to the fact that potassium activates many enzymes, which are necessary for the synthesis of chlorophyll and the formation of carbohydrates. Studies by Ganjipour (2007) confirmed these results. Abbas *et al*. (2011) by investigating and studying different levels of potassium sulfate in mung bean plant, reported that the chlorophyll index and seed yield were significantly increased by potassium consumption, which was consistent with the results of this research. Miri *et al*. (2024) evaluated the effect of drought stress on mung bean plant traits and stated that water stress reduced seed yield, relative moisture content and total chlorophyll content of mung bean plant. Aghdasi *et al*. (2018) studied the effect of water deficit stress on some physiological traits and the quantitative and qualitative yield of mung bean and reported that amount of chlorophyll b decreased in both stress levels in the vegetative and reproductive stages under the influence of drought stress compared to the control. Also the effect of drought stress on the reproductive treatment (32% reduction compared to the control) was more than the vegetative treatment.

4.5. Seed yield

According result of analysis of variance effect of drought stress, biologic fertilizer and interaction effect of treatments on seed yield was significant at 1% probability level (Table 2). According result of mean comparison maximum of seed yield (245.71 $g.m^{-2}$) was obtained for 80 mm evaporation from class A evaporation pan and minimum of that (128.60 $g.m^{-2}$) was for 160 mm evaporation from class A evaporation pan (Table 3). Evaluation means comparison result indicated in different level of biologic fertilizer the maximum seed yield (225.68 $g.m^{-2}$) was noted for combination of humic acid and chelated Potassium and minimum of that (143.58 $g.m^{-2}$) belonged to control treatment (Table 3). Evaluation means comparison result of interaction effect of treatments indicated maximum seed yield (251.03 $g.m^{-2}$) was noted for 80 mm evaporation from class A evaporation pan and combination of humic acid and chelated Potassium and lowest one (119.22 $g.m^{-2}$) belonged to 160 mm evaporation from class A evaporation pan and nonuse of biofertilizer (Fig.1).

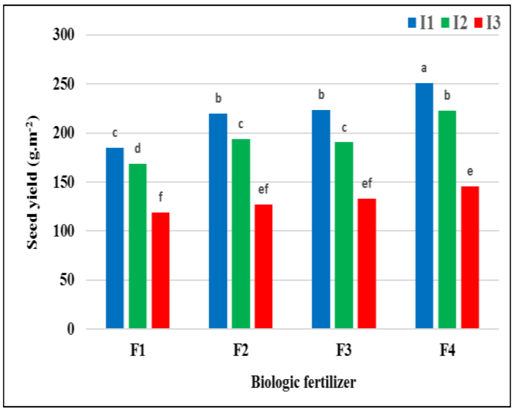

Fig.1. Interaction effect of drought stress and biologic fertilizer on seed yield. I_1: 80 mm evaporation from class A evaporation pan or control, I_2: 120 mm evaporation from class A evaporation pan I_3: 160 mm evaporation from class A evaporation pan. F_1: nonuse of biofertilizer or control, F_2: 4 $L.ha^{-1}$ humic acid, F_3: 4 $kg.ha^{-1}$ Chelated Potassium, F_4: Combination of 4 $L.ha^{-1}$ humic acid and 4 $kg.ha^{-1}$ Chelated Potassium (Means with similar letters in each column are not significantly different by Duncan's test at 5% probability level)

Abbas *et al*. (2011) by study on the application different levels of potassium sulfate in Mung bean, it was reported that the number of pods per plant, number of seeds per pod and seed yield were significantly affected by potassium. The minimum number of pods and number of seeds per pod were obtained from control treatment. Shirvanipour *et al.* (2014) reported the effect of potash fertilizer on biological yield was significant. So the highest amount of biological yield (198.4 $g.m^{-2}$) was obtained from use 100 $kg.ha^{-1}$ of potash and the lowest one (117.1 $g.m^{-2}$) was obtained from non-application of potash fertilizer. Fayad *et al*. (2019) by assess the application of humic acid on cowpea crop production, stated that the use of organic fertilizers such as humic acid increased the total chlorophyll, number of pods per plant, number of seeds per pod, the seed weight and the seed yield.

4.6. Protein content

Result of analysis of variance revealed effect of drought stress and biologic fertilizer and chelated Potassium on protein content was significant at 5% probability level, but interaction effect of treatments was not significant (Table 2). Assessment means comparison result indicated in different level of drought stress the maximum protein content (24.65%) was noted for 160 mm evaporation from class A evaporation pan and minimum of that (19.50%) belonged to control treatment (Table 3). Compare different level of manganese Nano-chelate showed that the maximum and the minimum amount of protein content belonged to combination of humic acid and chelated Potassium (23.55%) and control (20.01%) treatments (Table 3). Under drought stress, due to the shortening of grain filling period, the photosynthetic material transfer to the seed is reduced and starch content storage decreases, so that matter led to decreases the seed size and increases the percentage of protein. Also in stress conditions, the crop by increases the amount of soluble proteins by producing stress-tolerant proteins (De-Mejia *et al*., 2003). Daniel and Triboi (2008) reported drought stress led to increased seed protein percentage than to normal conditions. Because of the decrease in photosynthetic material transport led to reduce the ratio of starch endosperm volume to total seed volume. In this study, it seems that increasing seed protein percentage in use vermicompost treatment may be due to mineralization process of nitrogen and release of nitrogen in soil and appropriate moisture.

5. CONCLUSION

Therefore, in general, it can be stated that in order to achieve the maximum crop production and qualitative components, planting mung bean with 80 mm from evaporation pan with combined application of humic acid and potassium chelate fertilizer in studied areas will be recommended.

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FOOTNOTES

AUTHORS' CONTRIBUTION: All authors are equally involved.

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