

Interactive Effect Of Different Level Of Potassium On Characters Related To Nitrogen Fixation In *Vigna Radiata* (L.) Wilczek under Water Stress

¹Nisha Kataria, ²Narender Singh

^{1,2}Plant Physiology and Biochemistry Laboratory, Department of Botany, Kurukshetra University, Kurukshetra-136119, Haryana, India

Abstract: *Vigna radiata* L. (Mungbean) is one of the most important pulse crops for protein supplement in a subtropical zone of the world, as it is the best alternative to meet the food needs of the large population of developing countries due to its nutritional superiority. The performance of two mungbean genotypes SML-668 and MH-318 to potassium fertilization 0.00, 1.54, 2.31 and 3.08 mmol dm⁻³ in addition to the existing level of 1.32 mmol dm⁻³ in the soil medium was investigated under water stress conditions on nitrogen fixation and its attributes. Water stress (Soil Moisture Contents 4.5 ± 0.5 %) was created by withholding irrigation at different growth stage. Water stress resulted in marked decrease in leghemoglobin and the activity of enzymes of nitrogen assimilation i.e. nitrate reductase (NRA) and nitrite reductase (NiRA) activity in both the genotypes. The nitrogen fixation were least affected when crop was stressed at the vegetative stage. Potassium treated plants maintained higher values for number of nodules, leghemoglobin content, nitrogenase, nitrate and nitrite reductase activity. The nitrogen fixation were least affected when crop was stressed at the vegetative stage. The reduction was relatively more in MH-318 genotype as compared to SML-668. This study provides direct evidence of the beneficial functions of potassium fertilization in mitigating the adverse effects of water stress on nitrogen fixation and its attributes.

Keywords: Leghemoglobin, Nitrogenase, Nitrate reductase, Potassium, *Vigna radiata* L., Water stress.

I. INTRODUCTION

The abiotic stresses negatively influence the yield of crops up to seventy percent [1], [2]. Water stress is the most prevalent abiotic stress that limits global plant growth and productivity more severely than that caused by any other environmental stresses [3], [4], [5]. Nitrogen fixation by leguminous plants is very sensitive to changes of chemical and physical conditions in soil. Deficit soil-moisture critically restricts N₂ fixation activity of nodule bacteria and thereby markedly reduces the yield. An efficient use of available water and better growth under limited water supply are desirable traits for crops under water deficit environments.

Vigna radiata (L.) Wilczek commonly known as mung bean is the third important grain legume crop in South East Asia and Africa, and a source food that has a high nutritive value [6]. It is not only a rich and economical source of protein, phosphorus, carbohydrate, minerals and provitamin A, but also commonly used as fodder and green manure. Mung bean contains bioactive components with antioxidant, antimicrobial and insecticidal properties [7], [8], [9]. During the past four decades, we have witnessed the doubling of human population and a concurrent doubling of food production. Plant nutrition has played a key role in the dramatic increase in demand for and supply of food. The scarcity of food containing high nutritive value is an increasing problem affecting millions of people living in poor and developing countries [10]. So, there is an urgent need for innovative technologies to address the growing challenges of malnutrition.

Status of mineral-nutrient in plants plays a critical role in increasing plant resistance to drought stress [11]. Potassium can stimulate increase in nitrogen fixation and its attributes by increasing availability of photosynthate to the nodules [12].

This shows the importance of potassium in legume nutrition. Potassium (K) has an important role in several physiological processes directly related to nodulation and N₂ fixation as well as carbohydrate transport [13] and incorporation of combined nitrogen into protein [14]. Potassium is involved in activating a wide range of enzyme system which regulates photosynthesis, water use efficiency and movement, nitrogen uptake and protein building [15].

Keeping the above in view, the present investigation was undertaken to evaluate the effects of potassium application on nitrogen fixation in Mungbean under water stress conditions.

II. MATERIALS AND METHODS

The present study was carried out for three consecutive years (2010-11, 2011-12 and 2012-13) during summer season under net house conditions at Botany Department, Kurukshetra University, Kurukshetra. Kurukshetra, stands in the plains of North Eastern part of Haryana State, in India, between latitude (29°-52' to 30°- 12') and longitude (76°-26' to 77°-04') with an altitude of 258.4 meters above sea level. The climate of the district is of pronounced character with a summer maximum temperature as high as 45°C and a winter temperature of as low as 3°C. The average yearly rainfall lies between 400-500 mm. The experiment was conducted in net house in order to avoid any birds attack on the plants and interruption by rainfall. The later was done by covering the net house structure with polyethylene plastic sheet at the time of rainfall.

A. Experimental design: Two genotypes of *Vigna radiata* i.e. SML-668 and MH-318 were selected for the present study.

(Figure 1)

The seeds were sterilized and inoculated with standard *Bradyrhizobium sp.* S-24 before sowing. The crop was raised in earthen pots (30 cm in diameter) lined with polythene bags and filled with 7.0 kg of dune sand. Five seeds were sown in each pot at uniform depth and distance. Thinning was done after one week of germination and only two plants of uniform size were retained in each pot. These pots were placed in the net house under natural conditions and the soil was saturated with water. Sowing was carried out at field capacity of soil. The experiment was laid out in factorial complete randomized design (CRD) with three replications.

Level of potassium: After germination (7 days after sowing), potassium was supplied in the soil in the form of muriate of potash in the following concentration in addition to the existing level (1.32 mmol dm⁻³) in the soil medium.

- a) 0
- b) 1.54 mmol
- c) 2.31 mmol
- d) 3.08 mmol

Level of stress: Water stress was created by withholding irrigation at different sampling stages:

Treatments	Soil Moisture Content (%)
a) Control	12.0 ± 0.5
b) Stress	4.5 ± 0.5

The pots were weighed daily and depletion in soil moisture content (SMC %) was maintained gravimetrically.

Sampling stages: The plants were sampled at following stages:

- i) Vegetative: 20±2 days after sowing (DAS)
- ii) Flowering: 35±2 DAS
- iii) 50% pod formation: 47±2 DAS

Each pot was supplied with equal quantity of nitrogen free nutrient solution (Wilson and Reisenauer, 1963) at a regular interval of 7-10 days.

Number of Nodules: The treated plants removed from soil and their nodule was separated from the root and counted.

Leghemoglobin Content: All the detached nodules from the roots of the sampled plants were mixed and their leghemoglobin content was estimated by the method of (Hartree, 1955), [16]

Nitrogenase activity: the Nitrogenase activity of nodules was measured by acetylene reduction assay (ARA) of (Hardy *et al.*, 1968), [17].

Nitrate and Nitrite Reductase activity: For determination of Nitrate reductase activity (NRA) the method of (Jaworski, 1971), [18] was adopted and the Nitrite reductase activity (NiRA) was estimated by the intact tissue assay of (Ferrari and Varner, 1971), [19].

Specific Nitrogenase Activity: Nodule Specific nitrogenase activity was determined by acetylene reduction assay (Nandwal *et al.*, 1991a), [20].

Statistical analysis: The data collected was analysed statistically by online Statistical Analysis (OPSTAT, CCS Haryana Agriculture University, Hisar). The significance of data obtained was judged from the critical difference at 5 % level of significance.

III. RESULTS

The number of nodules increased up to flowering and then decline in both the cultivars. A drastic reduction in number of nodules was observed under water stress condition. A significant increase in nodule number was observed under potassium treatment in both the cultivars (Table 1).

Water stress resulted in a significant decrease in the leg hemoglobin content of nodules in both the cultivars at all the sampling stages. The reduction was higher at pod formation stage (Table 2). Application of potassium resulted in a significant increase in the leg hemoglobin content also. Cultivar SML-668 exhibited higher leg hemoglobin content in comparison to *cv.* MH-318.

Nitrogenase activity gets inhibited under water stress at all stages in both the cultivars (Table 3). Maximum nitrogenase activity in control and stressed plants was observed at flowering stages and least at pod formation stage. Increased level of potassium resulted in increased nitrogenase activity under control in both the cultivars.

Specific nitrogenase activity gets inhibited under water stress. Maximum specific nitrogenase activity in control and stressed plants was observed at flowering stages and least at pod formation stage (Table 4).

Nitrate reductase activity (NRA) was highest at flowering stage in both the cultivars. Decrease in NRA activity was noted under water deficit condition (Table 5). The decrease was more at pod formation stage compared to other sampling stages. Potassium treatment significantly increased the NRA activity at all the stages.

Nitrite reductase activity (NiRA) reduced under water stress in both the cultivars (Table 6). Highest activity of nitrite reductase was observed at flowering stage. Increase in NiRA activity was observed under the treatment of Potassium in both the cultivars.

Cultivar SML-668 exhibited higher nitrogenase, NRA and NiRA activity over *cv.* MH-318 irrespective of sampling stages and potassium treatment.

IV. DISCUSSION

Drought greatly reduces nitrogen fixation, leading to low Nitrogen accumulation, dry matter production and yield [21], [22], [23]. Water stress reduces the Nitrogen uptake and application of potassium enhanced the uptake irrespective of levels of soil moisture. The present experiments revealed a decrease in number of nodules, leghemoglobin content, nitrogenase activity and nitrate and nitrite activity of nodules under water stress conditions in comparison to control. This effect of water stress could be ameliorated by potassium application (Fig. 2, 3, 4). These findings are in consonance with [12]. [24] also reported that applied potassium maintain sufficient rates of nitrogen fixation and N- turnover under water stress in *Vicia faba*. [25] reported that water stress caused reduction in nitrogen uptake in wheat. [26] also observed the similar response of water stress on rice.

There is increase in number in nodules with potassium application under water stress conditions. The decrease in nodule number under water stress conditions also observed by [27], [28], [29]. Moisture stress also delays nodule formation in leguminous crops [30]. Number of nodules reached their maximum at flowering stage in control as well as in stress, after which there was a reduction in nodule number. This reduction was due to severe senescence and decaying of nodule. The decrease in nodule number per plant occurs due to an arrest in the development of new nodules under the imposed stressful conditions. Nodule senescence occurs under prolonged period of water deficit [31], [32] hence decrease in number of nodule observed with the progression of water stress. Treatment with potassium increased the number of nodule of both the varieties of Mungbean (Table 1).

Water stress resulted in the marked reduction of leg hemoglobin content in both the cultivars (Table 2). Leghemoglobin exhibit the highest value at flowering stage and then declined subsequently at later stage of growth. The reduction was maximum at pod formation stage. The decrease in leghemoglobin content during water stress also observed by [33] in *Medicago sativa*. It was due to inhibition of protein synthesis and to an increased proteolytic activity in nodule cytosol rather than to a specific proteolysis of leghemoglobin. The severe stress condition resulted in considerable reduction in leghemoglobin content of the nodules [34].

Nitrogenase activity gets inhibited under water stress at all stages in both the cultivar (Table 3). Highest nitrogenase activity in control and stressed plants were observed at flowering stage and least at pod formation stage. The decline in total nitrogenase activity after flowering might have been either due to non-availability of photosynthates in nodules. Similar results also observed by [35] in soybean and in pigeon pea by [36]. Such inhibitory effect of water stress on nitrogen-fixation has reported by several authors [37], [38]. [37] reported that it may be soluble nitrogen content of nodules which influence the nitrogenase activity depending upon its utilization and accumulation in addition photosynthesis which are responsible for decline in nitrogenase activity under water stress condition. Application of potassium maintained higher nitrogenase activity under water stress condition in both the cultivars. The decrease in nitrogen fixation per plant was a consequence of both the lower amount of nodules and the decrease in the specific nitrogenase activity per gram of nodule. Nitrogenase activity was dependent largely on low reduction under severe drought [39]. Treatment with potassium also increases the specific nitrogenase activity at all sampling stages. A decrease in Specific nitrogenase activity under water stress was accompanied by a concomitant decrease in nodule respiration and was not due to less supply of photosynthates [40].

NRA and NiRA activity exhibit the highest value at flowering stage and declined subsequently at later stages of growth (Table 5, 6). Highest activity of nitrite reductase was observed at flowering stage. [41] reported that water stress may decrease the NRA activity either by inhibiting nitrate uptake or protein synthesis. Application of potassium resulted in the increased NO_3^- in control as well as under water deficit condition in both the cultivars. [42] declared that the potassium by increasing the nitrate reductase activity, leads to efficient formation of molecules with nitrogen in their structure, which is responsible for synthesis of proteins and enzyme. [43] also reported that potassium absorption increased the nitrate reductase activity. Increase in leaf SPAD values by potassium enriched plant is confirmed with the knowledge of nutrients that increases activity of nitrate reductase enzyme. [44] reported that potassium application promoted the activity of nitrate reductase enzyme and thus nitrates assimilation by the plant.

V. CONCLUSION

Therefore, it is concluded, that water stress adversely affected the nitrogen fixation and the related traits in contrast with increasing potassium consumption, negative effect of water stress on nitrogen fixation of Mungbean genotypes consequently improved. Activity of nitrogenase, nitrate reductase (NRA), nitrite reductase (NiRA), Leghemoglobin and number of nodules showed abrupt decrease under water stress conditions. Potassium proved effective in alleviating the deleterious effect of water stress by enhancing the activity of nitrogenase, NRA, NiRA and Leghemoglobin content of nodules in both the cultivars. Potassium improved the nodulation, nitrogen fixation and enzymes of nitrogen assimilation.

ACKNOWLEDGEMENT

The authors are grateful to the **International Potash Institute, Switzerland** for the financial support and Kurukshetra University, Kurukshetra, India for providing the necessary facilities.

REFERENCES

- [1] G Kaur, S Kumar, H Nayyar, HD Upadhyaya, (2008) Cold stress injury during the pod-filling phase in chickpea (*Cicer arietinum* L.): Effects on quantitative and qualitative components of seeds. *J Agro Crop Sci.* 194 (6):457–464.
- [2] P Thakur, S Kumar, JA Malik, JD Berger, H Nayyar, (2010) Cold stress effects on reproductive development in grain crops: an overview. *Environ Exp Bot.* 67(3):429–443.
- [3] N Vorasoot, P Songsri, C Akkasaeng, S Jogloy, A Patanothai, (2003) Effect of water stress on yield and agronomic characters of peanut. *Songklanakarinn.* *J Sci Technol.* 25(3):283–288.
- [4] CA Jaleel, P Manivannan, A Wahid, M Farooq, R Somasundaram, R Panneerselvam, (2009). Drought stress in plants: a review on morphological characteristics and pigments composition. *Int J Agr Biol.* 11:100–105.
- [5] JS Boyer, (1982) *Plant Productivity Environ.* 218: 443-448.
- [6] P Vijayalakshmi, S Amirthaveni, RP Devadas, K Weinberger, SCS Tsou, S Shanmugasundaram, (2003) Enhanced bioavailability of iron from Mungbeans and its effects on health of school children. Technical Bulletin No. 30, 03-559. Asian vegetable research and development center, Shanhua, Taiwan.
- [7] LV Kaprelynts, SV Kisilev, EG Iorgachova, (2003) Soybean isoflavones and prospects of their therapeutic application. *Voprosy Pitaniya.* 72, 36–41.
- [8] T Madhujith, M Nacz, F Shahidi, (2004) Antioxidant activity of common beans (*Phaseolus vulgaris* L.). *J Food Lipids.* 11, 220–233.
- [9] MSA Ahmad, M Hussain, S Ijaz, AK Alvi, (2008) Photosynthetic performance of two mungbean (*Vigna radiata*) cultivars under lead and copper stress. *Inter J Agri Biol.* 10: 167–172.
- [10] F. Burchi, J. Fanzo, and E. Frison, (2011) The Role of Food and Nutrition System Approaches in Tackling Hidden Hunger, *Int J Environ Res Public Health.* 2011, 8, 358-373.
- [11] H Marschner, (1995) Mineral nutrition of higher plants. 2nd Edition. Academic press, San Deigo, California, USA, pp. 889.
- [12] K Mengel, MR Haghparast, K Koch, (1974) The effect of potassium on the fixation of molecular nitrogen by root nodules of *Vicia faba*. *Plant Physiol.* 54: 535-538.
- [13] M Becker, KH Diekmann, JK Ladha, SK De Datta, JCG Ottow (1991) Effect of NPK on growth and nitrogen fixation of *Sesbania rostrata* as a green manure for lowland rice (*Oryza sativa* L). *Plant Soil.* 132: 149–158.
- [14] S Feigenbaum, K Mengel, (1979) The effect of reduced light intensity and sub-optimal Potassium Supply on N₂ fixation and N turnover in *Rhizobium* infected lucerne. *Physiol Plant.* 45: 245–249.
- [15] HT Nguyen, AT Nguyen, BW Lee, J Schoenau, (2002) Effects of long-term fertilization for cassava production on soil nutrient availability as measured by ion exchange membrane probe and by corn and canola nutrient uptake. *Korean J Crop Sci.* 47: 108-115.
- [16] EF Hartree, (1955) Haematin compounds. In: *Modern methods of plant analysis* (eds.) Peach, K. and Tracay, M. V., Springer - Verlag, Berlin. Pp. 197-245.
- [17] RWF Hardy, RD Holsten, EK Jackson, RC Burns, (1968) The acetylene/ ethylene assay for nitrogen fixation. Laboratory and field evaluation. *Plant Physiol.* 43: 1185-1207.
- [18] EG Jaworski, (1971) Nitrate reductase assay in intact plant tissues. *Biochem Biophys Res Commun.* 43, pp. 1274-1279.
- [19] TE Ferrari, JE Varner, (1971) Intact tissue assay for nitrite reductase in barley aleurone layers. *Plant Physiol.* 47: 790-794.

- [20] AS Nandwal, S Bharti, MS Kuhad, IS Shroan, (1991) (a) Water relations and gaseous exchange studies in pigeonpea under depleting soil water potential. *Plant Physiol Biochem.* 29: 75-78.
- [21] AL Chapman, RC Muchow, (1985) Nitrogen accumulated and partitioned at maturity by grain legumes grown under different water regimes in a semi-arid tropical environment. *Field crops Res.* 11: 69-79.
- [22] JD Devries, JM Bennett, SL Albrecht, KJ Boote, (1989) Water relations, nitrogenase activity and root development of three grain legumes in response to soil water deficits. *Field Crops Res.* 21: 215-226.
- [23] M De Silva, LC Purcell, CA King, (1996) Soybean petiole ureide response to water deficits and decreased transpiration. *Crop Sci.* 36: 611-616.
- [24] JI Sprent, (1979) Nitrogen fixation and agriculture. In: *The Biology of nitrogen fixing organisms.* (ed.) sprent, I.J. Mcgraw-Hill, New York. Pp. 101.
- [25] AL Patel, J Singh, (1998) Nutrient uptake and distribution in aerial parts of wheat under water stress at different growth stages. *Ann Agricultural Biol.* 3: 5-8.
- [26] KK Baruah, SS Bhuiyan, TJ Ghosh, AK Pathak, (1998) Response of rice (*Oryza sativa* L.) genotypes to moisture stress imposed at seedling stage. *Indian J Plant Physiol.* 3: 181-184.
- [27] DL Smith, M Dijak, DJ Hume, (1988) The effect of water deficit on N₂ (C₂H₄) fixation by white bean and soybean. *Can J Plant Sci.* 68: 957-967.
- [28] MJ Delgado, F Ligerio, C Lluch, (1994) Effects of salt stress on growth and nitrogen fixation by pea, faba-bean, common bean and soybean plants. *Soil Biol Biochem.* 26: 371-376.
- [29] R Sangakkara, UA Hartwig, J Nosberger, (1996) Growth and symbiotic nitrogen fixation of *Vicia faba* and *Phaseolus vulgaris* as affected by fertiliser potassium and temperature. *J Sci Food Agric.* 70: 315-320.
- [30] GHS Reddi, TY Reddy, (1995) Irrigation efficiencies and water use efficiency. *Efficient use of Irrigation Water.* Kalyani Publishers, New Delhi. Pp. 199-218.
- [31] A Djekoun, D Planchon, (1991) Water status effect on dinitrogen fixation and photosynthesis in soybean. *Agron J.* 83: 316-322.
- [32] R Hooda, (1987) Partitioning and utilization of photosynthate for dry matter production and nitrogen fixation under water stress in chickpea (*Cicer arietinum* L.) Ph.D Thesis, H.A.U. Hisar (Haryana).
- [33] M Becana, P Aparicio-Tejo, J Pena, J Aguirreolea, M Sanchez-Diaz, (1986) N₂ fixation (C₂H₂- Reducing Activity) and Leghemoglobin content during nitrate- and Water-Stress-Induced Senescence of *Medicago sativa* Root Nodules. *J Exp Bot.* 37: 597-605.
- [34] K Swaraj, AF Topunov, L Golubeva, WL Kretovich, (1984) Effect of water stress on enzymatic reduction of leghemoglobin and nitrogen fixation in soybean nodules. *Advances in frontier Aras of plant biochemistry,* (eds).
- [35] AJ Ciha, WA Brun, (1978) Effect of pod removal on non-structural carbohydrate concentration in soybean tissue. *Crop Sci.* 18: 773-776.
- [36] YP Luthra, IS Sheoran, R Singh, (1983) Ontogenetic interactions between photosynthesis and symbiotic nitrogen fixation in pigeonpea. *Ann Appl Biol.* 103: 549-556.
- [37] AS Nandwal, S Bharti, MS Kuhad, IS Singh, (1991) (b) Nitrogen status of pigeon pea (*Cajanus cajan*) under water deficit. *Indian J Exp Biol* 29: 879-880.
- [38] MA Mukane, VD Chavan, BB Desai, (1993) Effect of water stress on metabolic alterations in Pigeonpea (*Cajanus cajan* (L.) *Millspaugh*) genotypes. *Legume Res.* 16: 45-50.
- [39] S Pimratch, S Jogloy, N Vorasoot, B Toomsan, T Kesmala, A Patanothai, CC Holbrook, (2008) Effect of drought stress on traits related to N₂ fixation in eleven Peanut (*Arachis hypogaea* L.) genotypes differing in degree of resistance to drought. *Asian J Plant Sci.* 7: 334-342.

- [40] JL Durand, JE Sheehy, FR Minchin, (1987) Nitrogenase activity, photosynthesis and nodule water potential in soybean plants experiencing water deprivation. J Exp Bot. 38: 311-321.
- [41] HS Srivastava, (1980) Regulation of nitrate reductase in higher plants. Phytochemistry. 19: 725-773.
- [42] F Mohammad, U Naseem, (2006) Effect of K application on leaf carbonic anhydrase and nitrate reductase activities, Photosynthetic Characteristics, NPK and NO₃ contents, growth and yield of mustard. Photosynthetica. 44: 471-473.
- [43] S Umar, (2006) Alleviation of adverse effects of water stress on yield of sorghum, mustard and groundnut by potassium application. Pak J Bot. 38: 1373-1380.
- [44] A Dobermann, (2004) Crop potassium nutrition implications for fertilizer recommendations. Department of Agronomy and Horticulture, University of Nebraska, Lincoln, NE, pp 1-12.

APPENDIX - A

Table: 1. Interaction of water stress and applied potassium on nodules number/plant of Mungbean cultivars at different growth stage

Sampling Stages	K (mM)	Varieties						C.D. at 5% level
		SML-668			MH-318			
		Control	Stress	Mean	Control	Stress	Mean	
Vegetative Stage	0	18.01	12.52 (30.4)*	15.26	9.33	5.33 (42.8)*	7.33	V = 1.99 S = 1.98 K = 2.81 V×S = 2.83 V×K = N/A S×K = N/A V×S×K = N/A
	1.54	22.36 (24.1)	13.60 (8.6)	17.98	10.89 (16.7)	5.62 (5.4)	8.25	
	2.31	26.68 (48.1)	14.13 (12.8)	20.40	12.77 (36.8)	5.80 (8.8)	9.28	
	3.08	30.24 (67.9)	14.48 (15.6)	22.36	14.14 (51.5)	5.94 (11.4)	10.04	
	Mean	24.32	13.68		11.78	5.67		
Flowering Stage	0	30.61	17.34 (43.3)*	23.97	15.26	8.02 (47.4)*	11.64	V = 2.52 S = 2.52 K = 3.57 V×S = 3.57 V×K = N/A S×K = 5.05 V×S×K = N/A
	1.54	40.48 (32.2)	19.74 (13.8)	30.11	19.63 (28.6)	8.78 (9.4)	14.20	
	2.31	50.69 (65.5)	20.89 (20.4)	35.79	23.87 (56.4)	9.09 (13.3)	16.48	
	3.08	55.42 (81.0)	21.38 (23.2)	38.40	26.50 (73.6)	9.42 (17.4)	17.96	
	Mean	44.30	19.83		21.31	8.82		
Pod formation stage	0	24.30	15.30 (37.0)*	19.80	12.30	6.85 (44.3)*	9.57	V = 1.99 S = 1.99 K = 2.82 V×S = 2.82 V×K = N/A S×K = 3.99 V×S×K = N/A
	1.54	31.17 (28.2)	16.90 (10.4)	24.03	14.82 (20.4)	7.35 (7.2)	11.08	
	2.31	38.06 (56.6)	17.65 (15.3)	27.85	18.14 (47.4)	7.62 (11.2)	12.88	
	3.08	42.33 (74.1)	18.15 (18.6)	30.24	19.79 (60.8)	7.78 (13.5)	13.78	
	Mean	33.96	17.00		16.26	7.40		

Values represent means, n=3;

Values in parentheses are: (i) * Percent decrease under stress and (ii) Percent increase with K

Table: 2. Interaction of water stress and applied potassium on leghemoglobin content (mg g⁻¹ FW) of nodules of Mungbean cultivars at different growth stage

Sampling Stages	K (mM)	Varieties						C.D. at 5% level
		SML-668			MH-318			
		Control	Stress	Mean	Control	Stress	Mean	
Vegetative Stage	0	1.53	1.07 (30.0)*	1.30	1.42	0.93 (34.5)*	1.17	V = 0.064 S = 0.064 K = 0.090 V×S = 0.090 V×K = N/A S×K = 0.127 V×S×K = N/A
	1.54	1.60 (4.5)	1.10 (2.8)	1.35	1.47 (3.5)	0.95 (2.1)	1.21	
	2.31	1.89 (23.5)	1.19 (11.2)	1.54	1.70 (19.7)	1.02 (9.6)	1.36	
	3.08	1.95 (27.4)	1.22 (14.0)	1.58	1.74 (22.5)	1.04 (11.8)	1.39	
	Mean	1.74	1.14		1.58	0.986		
Flowering Stage	0	2.30	1.57 (31.7)*	1.93	2.11	1.36 (35.5)*	1.73	V = 0.064 S = 0.064 K = 0.090 V×S = N/A V×K = N/A S×K = 0.128 V×S×K = N/A
	1.54	2.52 (9.5)	1.65 (5.1)	2.08	2.23 (5.6)	1.41 (3.6)	1.82	
	2.31	2.98 (29.5)	1.91 (21.6)	2.44	2.56 (21.3)	1.57 (15.4)	2.06	
	3.08	3.17 (37.8)	2.00 (27.3)	2.58	2.66 (26.0)	1.64 (20.5)	2.15	
	Mean	2.74	1.78		2.39	1.49		
Pod formation stage	0	1.05	0.52 (50.4)*	0.785	0.90	0.42 (53.3)*	0.66	V = 0.059 S = 0.059 K = 0.083 V×S = N/A V×K = N/A S×K = N/A V×S×K = N/A
	1.54	1.12 (6.6)	0.54 (3.8)	0.833	0.93 (3.3)	0.43 (2.3)	0.68	
	2.31	1.24 (18.1)	0.57 (9.6)	0.906	1.04 (15.5)	0.45 (7.1)	0.74 5	
	3.08	1.28 (21.9)	0.58 (11.5)	0.93	1.06 (17.7)	0.46 (9.5)	0.76	
	Mean	1.17	0.554		0.983	0.44		

Values represent means, n=3;

Values in parentheses are: (i) * Percent decrease under stress and (ii) Percent increase with K

Table: 3. Interaction of water stress and applied potassium on total nitrogenase activity (μ mol C_2H_4 evolved plant⁻¹ h⁻¹) of nodules of Mungbean cultivars at different growth stage

Sampling Stages	K (mM)	Varieties						C.D. at 5% level
		SML-668			MH-318			
		Control	Stress	Mean	Control	Stress	Mean	
Vegetative Stage	0	2.87	0.50 (82.5)*	1.68	2.26	0.35 (84.5)*	1.30	V = 0.062 S = 0.062 K = 0.088 V×S = 0.088 V×K = 0.125 S×K = 0.125 V×S×K = 0.176
	1.54	3.73 (29.9)	0.57 (14.0)	2.15	2.81 (24.3)	0.39 (11.4)	1.60	
	2.31	4.64 (61.6)	0.76 (52.0)	2.70	3.51 (55.3)	0.49 (40.0)	2.00	
	3.08	4.99 (73.8)	0.82 (64.0)	2.90	3.69 (63.2)	0.52 (48.5)	2.10	
	Mean	4.05	0.66		3.06	0.43		
Flowering Stage	0	10.64	1.38 (87.0)*	6.01	9.23	0.82 (91.1)*	5.02	V = 0.069 S = 0.069 K = 0.098 V×S = 0.098 V×K = 0.138 S×K = 0.138 V×S×K = 0.195
	1.54	12.36 (16.1)	1.47 (8.7)	6.91	10.38 (12.4)	0.87 (6.1)	5.62	
	2.31	14.94 (40.4)	1.73 (25.3)	8.33	12.25 (32.7)	1.00 (21.9)	6.62	
	3.08	16.18 (52.0)	1.81 (31.1)	8.99	12.86 (39.3)	1.05 (28.0)	6.95	
	Mean	13.53	1.59		11.18	0.93		
Pod formation stage	0	1.58	0.29 (81.6)*	0.93	1.39	0.17 (87.7)*	0.78	V = 0.058 S = 0.058 K = 0.082 V×S = 0.082 V×K = N/A S×K = 0.116 V×S×K = N/A
	1.54	1.79 (13.2)	0.31 (6.9)	1.05	1.52 (9.3)	0.18 (5.8)	0.85	
	2.31	2.14 (35.4)	0.33 (13.7)	1.23	1.79 (28.7)	0.19 (11.7)	0.99	
	3.08	2.24 (41.7)	0.34 (17.2)	1.29	1.88 (35.2)	0.20 (14.7)	1.04	
	Mean	1.93	0.31		1.64	0.18		

Values represent means, n=3;

Values in parentheses are: (i) * Percent decrease under stress and (ii) Percent increase with K

Table: 4. Interaction of water stress and applied potassium on specific nitrogenase activity ($\mu\text{ mol C}_2\text{H}_4\text{ g}^{-1}\text{ nodule dry wt. h}^{-1}$) of Mungbean cultivars at different growth stage

Sampling Stages	K (mM)	Varieties						C.D. at 5% level
		SML-668			MH-318			
		Control	Stress	Mean	Control	Stress	Mean	
Vegetative Stage	0	40.30	5.17 (87.1)*	22.73	32.07	3.45 (89.2)*	17.76	V = 0.063 S = 0.063 K = 0.089 V×S = 0.089 V×K = 0.126 S×K = 0.126 V×S×K = 0.178
	1.54	43.55 (8.0)	5.33 (3.0)	24.44	34.01 (6.0)	3.53 (2.3)	18.77	
	2.31	51.54 (27.8)	5.67 (9.6)	28.60	38.29 (19.4)	3.71 (7.5)	21.00	
	3.08	53.44 (32.6)	5.93 (14.7)	26.82	39.36 (22.7)	3.82 (10.7)	21.59	
	Mean	47.20	4.09		35.93	3.62		
Flowering Stage	0	49.12	6.42 (86.9)*	27.77	40.01	5.18 (87.0)*	22.59	V = 0.065 S = 0.065 K = 0.091 V×S = 0.091 V×K = 0.129 S×K = 0.129 V×S×K = 0.183
	1.54	60.48 (23.1)	6.78 (5.6)	33.63	47.66 (19.1)	5.40 (4.2)	26.53	
	2.31	68.49 (39.4)	7.94 (23.6)	38.21	51.44 (28.5)	6.25 (20.6)	28.84	
	3.08	70.07 (42.6)	8.58 (33.6)	39.32	52.86 (32.1)	6.69 (29.1)	29.77	
	Mean	62.04	7.43		47.99	5.88		
Pod formation stage	0	13.29	1.80 (86.4)*	7.50	11.33	1.31 (88.4)*	6.32	V = 0.062 S = 0.062 K = 0.088 V×S = 0.088 V×K = 0.125 S×K = 0.125 V×S×K = 0.176
	1.54	14.16 (6.5)	1.85 (2.7)	8.00	11.85 (4.5)	1.33 (1.5)	6.59	
	2.31	16.56 (24.6)	1.94 (7.7)	9.25	13.77 (21.5)	1.37 (4.5)	7.57	
	3.08	16.99 (27.8)	1.99 (10.5)	9.49	14.16 (24.9)	1.44 (9.9)	7.80	
	Mean	15.25	1.89		12.77	1.36		

Values represent means, n=3;

Values in parentheses are: (i) * Percent decrease under stress and (ii) Percent increase with K

Table: 5. Interaction of water stress and applied potassium on nitrate reductase activity ($\mu\text{g No}_2$ produced g^{-1} fresh wt. h^{-1}) of nodules of Mungbean cultivars at different growth stage

Sampling Stages	K (mM)	Varieties						C.D. at 5% level
		SML-668			MH-318			
		Control	Stress	Mean	Control	Stress	Mean	
Vegetative Stage	0	63.07	33.05 (47.6)*	48.06	57.05	27.09 (52.5)*	42.07	V = 0.073 S = 0.073 K = 0.103 V×S = 0.103 V×K = 0.145 S×K = 0.145 V×S×K = 0.205
	1.54	80.23 (27.2)	36.29 (9.8)	58.26	71.79 (25.8)	29.05 (7.2)	50.42	
	2.31	93.05 (47.5)	37.94 (14.8)	65.49	81.07 (42.1)	30.24 (11.6)	55.66	
	3.08	96.09 (52.3)	38.88 (17.6)	67.48	84.10 (47.4)	31.28 (15.4)	57.69	
	Mean	83.11	36.54		73.50	29.41		
Flowering Stage	0	82.04	37.10 (54.7)*	59.57	73.05	32.12 (56.0)*	52.58	V = 0.071 S = 0.071 K = 0.100 V×S = 0.100 V×K = 0.142 S×K = 0.142 V×S×K = 0.201
	1.54	111.7 (36.1)	44.23 (19.2)	77.97	96.52 (32.1)	37.08 (15.4)	66.80	
	2.31	134.0 (63.3)	48.08 (29.6)	91.06	116.5 (59.4)	39.64 (23.4)	77.57	
	3.08	136.0 (65.7)	49.76 (34.1)	92.92	119.0 (62.9)	40.75 (26.8)	79.92	
	Mean	115.9	44.79		101.2	37.15		
Pod formation stage	0	78.01	28.97 (62.8)*	53.49	70.09	25.22 (64.0)*	47.65	V = 0.079 S = 0.079 K = 0.111 V×S = 0.111 V×K = 0.158 S×K = 0.158 V×S×K = 0.223
	1.54	103.3 (32.4)	32.86 (13.4)	68.10	89.05 (27.0)	27.85 (10.4)	58.45	
	2.31	125.0 (60.2)	34.66 (19.6)	79.86	108.0 (54.0)	29.31 (16.2)	68.70	
	3.08	126.0 (61.5)	35.75 (23.4)	80.90	109.0 (55.5)	30.58 (21.2)	69.82	
	Mean	108.1	33.06		94.07	28.24		

Values represent means, n=3;

Values in parentheses are: (i) * Percent decrease under stress and (ii) Percent increase with K

Table: 6. Interaction of water stress and applied potassium on nitrite reductase activity ($\mu\text{g No}_2$ reduced g^{-1} fresh wt h^{-1}) of nodules of Mungbean cultivars at different growth stage

Sampling Stages	K (mM)	Varieties						C.D. at 5% level
		SML-668			MH-318			
		Control	Stress	Mean	Control	Stress	Mean	
Vegetative Stage	0	17.82	10.07 (43.4)*	13.95	16.76	9.16 (45.3)*	12.96	V = 0.052 S = 0.052 K = 0.074 V×S = 0.074 V×K = 0.104 S×K = 0.104 V×S×K = 0.147
	1.54	21.43 (20.2)	10.91 (8.3)	16.17	19.23 (14.7)	9.79 (6.8)	14.51	
	2.31	27.54 (54.5)	12.24 (21.5)	19.89	24.11 (43.8)	10.85 (18.4)	17.48	
	3.08	28.63 (60.6)	12.54 (24.5)	20.59	24.66 (47.1)	11.13 (21.5)	17.90	
	Mean	23.85	11.44		21.19	10.23		
Flowering Stage	0	23.62	11.17 (52.7)*	17.40	21.82	9.86 (54.8)*	15.84	V = 0.060 S = 0.060 K = 0.085 V×S = 0.085 V×K = 0.120 S×K = 0.120 V×S×K = 0.170
	1.54	29.42 (24.5)	12.72 (13.8)	21.07	26.02 (19.2)	10.79 (9.4)	18.40	
	2.31	37.43 (58.4)	13.98 (25.1)	25.71	32.25 (47.8)	12.01 (21.8)	22.13	
	3.08	38.26 (61.9)	14.35 (28.4)	26.31	33.36 (52.8)	12.16 (23.3)	22.76	
	Mean	32.18	13.05		28.36	11.20		
Pod formation stage	0	21.11	8.99 (57.4)*	15.05	20.50	8.03 (60.8)*	14.26	V = 0.065 S = 0.065 K = 0.092 V×S = 0.092 V×K = 0.129 S×K = 0.129 V×S×K = 0.183
	1.54	25.79 (22.1)	9.91 (10.2)	17.85	23.83 (16.2)	8.73 (8.7)	16.28	
	2.31	32.96 (56.1)	11.10 (23.4)	22.03	29.83 (45.5)	9.63 (19.9)	19.73	
	3.08	33.97 (60.9)	11.34 (26.1)	22.65	30.59 (49.2)	9.91 (23.4)	20.25	
	Mean	28.45	10.33		26.19	9.07		

Values represent means, n=3;

Values in parentheses are: (i) * Percent decrease under stress and (ii) Percent increase with K

APPENDIX – B



Fig.1. (a): Experimental site at Botany Department, Kurukshetra University; (b): Effect of different concentration of potassium on root growth; (c-d): Effect of different concentration of potassium on plant growth of two cultivars i.e. MH-318 and SML-668 respectively.