ISSN 2250-3153

Study the Response of Drought Stress Inducing by Mannitol in Germination to Seedling Stage of Mung Bean (*Vigna Radiata L.*) Variety MI5 and Variety Harsha

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Abstract- Mung bean (*Vigna radiata* L.) is one of the most important grain legumes cultivated in Sri Lanka. Drought is a major abiotic stress on Mung bean in the sub-humid, dry and intermediate zones of Sri Lanka. The present reveals the response of mung bean for the drought stress at germination to seedling stages. The major beneficiaries of the intervention are the poor, especially children and women, whose diets lacked much needed protein and iron. Supplementation with improved Mung bean recipes has effectively improved low body iron stores in children and women. Both urban and rural consumers now have access to improved quality mungbean at reasonable cost. The drought is a major environmental factor that determines the growth, the productivity of crops which is estimated to be one of the most serious yield reducing stresses in crops. Surface sterilized seeds were introduced to hormone free Murashige and Skoog (MS) basal medium with different concentrations of mannitol (0%, 10%, 20%, 30%, 40% and 50%). Seedlings heights, length of the roots and number of germinated seeds were recorded after 2^{nd} weeks of culture. Completely Randomized Design (CRD) with five replicates was used for study. Statistical analysis was performed with Duncan's multiple range test using SAS software (version 9.1.3). Results showed that highest seed germination (100%) from Mung bean variety MI 5 and Harsha in all mannitol concentrations. Reduction of seedling height and increasing the root length was observed while increasing the mannitol concentration in both varieties.

Index Terms- Drought stress, Mannitol, Mung bean, Seed germination

I. Introduction

Among the pulse crops, Mung bean has a special importance of intensive crop production due to its short growth period (Ahmed *et al*., 1978). The Mung bean (*Vigna radiata*), alternatively known as the Moong bean, Green gram, and Golden gram is a plant species in the legume family (Fabaceae).Native to the Indian subcontinent the Mung bean is mainly cultivated today in India, China, and Southeast Asia. It is also cultivated in hot, dry regions in Southern Europe and the Southern United States. It is used as an ingredient in both savory and sweet dishes in Sri Lanka about 150,000 acres of cultivation lands are in danger of getting destroyed due to the drought in certain parts of the country, especially in the North Central, North and North Western provinces. The main limitation in rice production is abiotic stress caused by salinity, drought, extreme temperature and submergence. To overcome this problem, we can use Mung bean in drought season.

Mannitol (also referred to as mannite or manna sugar) (Chaves and Oliveira, 2004) is a white, crystalline solid (Zidenga, 2006) with the chemical formula $C_6H_8(OH)_6$ which is a member of sugar alcohols, is an osmotic adjustment chemical to control osmotic potential in the culture media or nutrient solutions in order to induce water deficit conditions for protein expression or proteomics studies (Chaves and Oliveira, 2004). In plants, it is used to induce osmotic stress.

Mungbean is a major legume crop that supplements the largely cereal-based diets of the poor in Asia. When consumed together, cereals and legumes (also known as pulses) contribute significantly to a healthy and balanced diet. Cereals are deficient in the amino acid lysine, which legumes provide, whereas legumes are low in sulphur-rich amino acids, which cereals provide. High in protein and easy to digest, Mung bean consumed in combination with cereals can thus significantly increase the quality of protein in a meal. Mung bean also promises other health benefits for consumers, especially poor women and children, who are most vulnerable to the effects of poor nutrition and a lack of micronutrients in their diets. In this situation the main objective of this study were to evaluate the response of seed germination and seedling growth of Mung bean (*Vigna radiata*) variety MI 5 and variety Harsha to the drought conditions which was induced by Mannitol.

II. Materials and Methods

Plant materials and drought treatment

Seeds of Mung bean (*Vigna radiata* L.) variety MI 5 and Harsha cultivar were obtained from Department of Agriculture, Sri Lanka. First, seeds were rinsed with soap and washed with distilled water. Then seed coats were removed using forceps and sterilized seeds using 70% (v/v) ethanol for 3 min. Thereafter, the seeds were soaked in 20% (v/v) Chlorox solution (Sodium hypochlorite) for 20 min. Those seeds were rinsed in sterile distilled water for 3 times. They were dried using sterilde filter papers. Surface sterilized seeds were introduced to test tubes (2 seed per tube) containing 3ml of Murashige and Skoog (MS) basal medium with different mannitol concentrations (0%, 10%, 20%, 30%, 40% and 50%). The culture medium used for all the experiments was based on MS medium with 30 gL^{-1} sucrose and solidified by corn flour ($100gL^{-1}$). Medium was autoclaved for 21 minutes at 121°C after adjusting the pH to 5.8 (Dahanayake *et al*, 2012).

Culture condition: Seeds were inoculated in prepared culture medium. Cultures were incubated for 2 weeks in the culture room with light intensity of 1000 µmol/m2/sec and at 25±1°C and 70-80% relative humidity with a 16/8 hrs light/dark photoperiod.

Experimental design: The numbers of seeds germinated were recorded after 2nd week. End of the 2nd week, length of the root and height of the seedling. All experiments reported here were done five replicates for each mannitol treatment. Statistical analysis was performed with Duncan's multiple range test using SAS software (version 9.1.3). The germination percentage was performed using the following formulae;

$$\text{Germination percentage (\%)} = [\frac{\text{Germinate seeds}}{\text{no. of total seeds}} \text{ X } 100]$$

III. Results and Discussion

Seed germination percentage: All the seeds were given 100% germination in all different mannitol concentrations. Germination percentages of Mung bean variety MI 5 and Harsha were not reduced when increasing of mannitol concentration. But the survival percentage of seedlings (rice) cultivated in salt-induced osmotic stress was greatly reduced, and to a higher degree than those under mannitol-induced stress (Nishimura *et al.*, 2011).

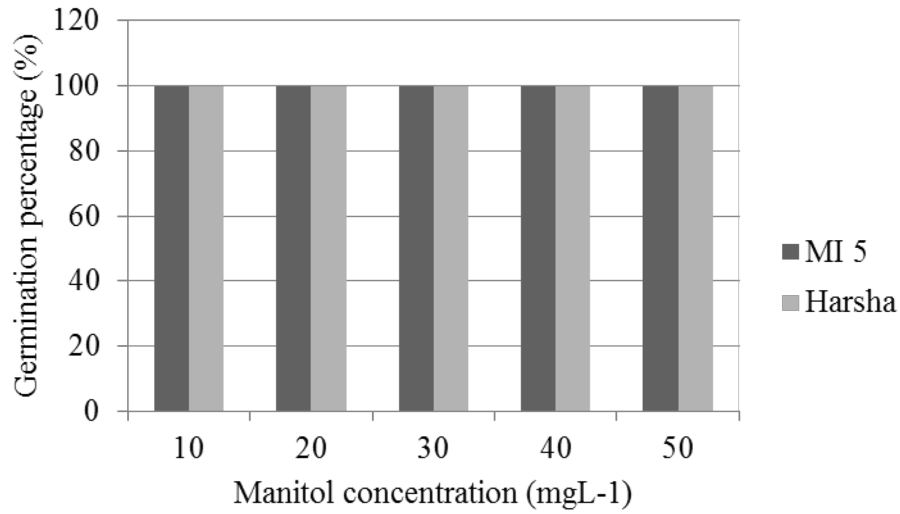

Figure 1: Effect of mannitol on seed germination percentage

Seedling height was influenced by different mannitol percentages: Mung bean variety MI 5 and variety Harsha seedling height was reduced significantly when increasing the mannitol concentration (Figure 2).The data of mean height of seedlings of table 1 shows that height is significant with each mannitol concentrations. Shoot length was higher in seeds treated with water, mannitol and lower concentration of K_2HPO_4 and KNO_3 as compared to seedlings grown from no treated Chickpea seed (Nighat, Sumaira and Farhat, 2006).

Mean Root length as influenced by different mannitol percentage: The data (Table 2) reveal that all length are significant from each mannitol concentration and Both Mung bean cultivars root lengths increasing along the high mannitol percentage shows in figure 2. From current findings; root length and dry root mass increase with mannitol in rice is confirming the previous results of many scientists practiced in different crops (Nighat *et al.*, 2006; Nishimura *et al.*, 2011; Hoekstra *et al.*, 2001).

Table 1: Effect of different mannitol concentrations on mean height of the seedlings and mean length of the roots in Mung bean variety MI 5 and variety Harsha

Mannitol Percentage	Mean height of the seedling (cm)		Mean length of the root (cm)	
	MI 5	Harsha	MI 5	Harsha
0%	15.70^{a}	14.88^{a}	1.73^{f}	2.20^{c}
10%	14.64^{b}	11.84^{b}	2.40^{e}	2.68^{bc}
20%	14.38^{c}	11.50^{b}	2.75^{d}	2.94^{ab}

30%	14.14[d]	11.08[b]	3.12[c]	3.25[ab]
40%	13.42[e]	10.70[b]	3.37[b]	3.27[ab]
50%	12.00[f]	10.27[c]	3.53[a]	3.38[a]

Column values followed by the same letter are not significantly different as determined by Duncan's multiple range test (P=0.05).Values in same column with same letter denoted non- significant difference

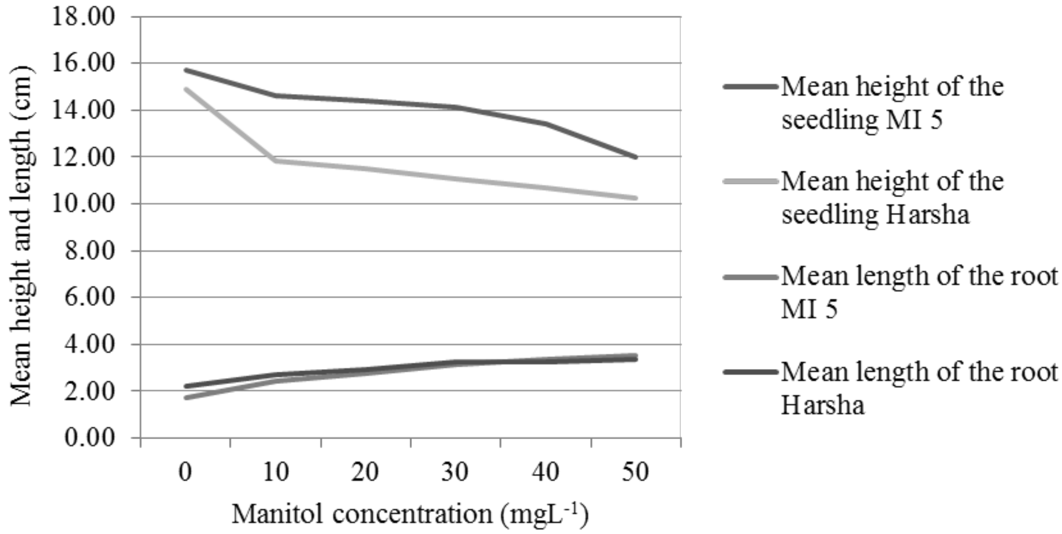

Figure 2: Effect of different mannitol concentrations on mean seedlings height and mean length of the roots in Mung bean variety MI 5 and variety Harsha

In particular, it is predicted that water deficit will continue to be a major abiotic factor affecting global crop yields. One third of the world's population resides in water stressed regions, and with elevated CO_2 levels in the atmosphere and climatic changes predicted in the future, drought could become more frequent and severe. In tolerant plants, there are many defense mechanisms such as osmoregulation, ion homeostasis, antioxidant and hormonal systems, helping plants to stay alive and development prior to their reproductive stages (Hasegawa, 2000). The seedlings of drought-tolerant cultivars showed lesser reduction of both growth characteristics and better stability of photosynthetic pigments than those of drought-sensitive cultivars. This could suggest that seedlings of drought-tolerant cultivars may have better adaptive responsibility such as the controlling stomatal pore and the stability of organelles within the cell (Setter and Flannigan, 2001). Above results focus on drought stress in seedlings and it triggers many common reactions in plants and leads to cellular dehydration which causes osmotic stress and removal of water from the cytoplasm into the extracellular space. The degree of plant growth inhibition due to osmotic stress is dependent on the stress exposed times, the particular tissues and the plant species (Smirnoff, 1998). In Mung bean water stress significantly effects on root system, shoot bio mass production and final yield (Ranawake *et al.*, 2011). From former details mentioned in above shows Mung bean variety MI 5 and Harsha has ability to adopt and to tolerance above conditions on drought by current findings.

IV. Conclusion

Drought tolerance response of Mung bean was showed that increasing in mannitol percentages were significantly affect for mean seedlings height which were decreased while mean length of the roots were increased. Above facts reveal that Mug bean variety MI 5 and Harsha respond to drought tolerance in seedling stage.

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