

Full Length Research Paper

Effects of foliar application of methanol on growth and yield of mungbean (*Vigna radiata* L.) in Rasht, Iran

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In order to study the effect of the time and concentration of methanol spraying on the growth and yield of mungbean (*Vigna radiata* L.), a factorial experiment in the form of randomized complete blocks was done with three replications in the research farm of the College of Agriculture of the Islamic Azad University, Rasht branch in the north of Iran in 2009. The concentration factor of spraying methanol was applied at four levels, that is control (without spraying (0)), 10, 20 and 30 volumetric percentages of methanol; while the time of spraying methanol was another factor used at three levels: in the morning (8:00 to 10:00 a.m.), at noon (13:00 to 15:00 p.m.) and in the evening (17:00 to 19:00 p.m.). Results showed that there was a significant difference between different methanol concentrations regarding number of seeds per pod, harvest index ($p < 0.01$) and seed yield per m^2 ($p < 0.05$). The largest numbers of seeds per pod and harvest index were in 30% methanol, while the highest seed yield was that of the 20% methanol with an average of 13.11 seeds, 38.22% and 55.97 g/m^2 , respectively. Moreover, spraying times had also had a significant difference in terms of the seed yield per m^2 and the harvest index at the level of 5%; the highest average values of seed yield and harvest index corresponding to spraying in the afternoon were 55.52 g/m^2 and 36.69%, respectively. The interaction of these two factors with none of the studied traits was not significant.

Key word: Mungbean, methanol spraying, concentration and time, pod yield, seed yield.

INTRODUCTION

Mungbean (*Vigna radiata* L.) is an annual plant of the legume family, Papilionacea subfamily, *Vigna* genus and *radiata* species. It is a short-day plant with different cultivars in terms of sensitivity. Mungbean is a warm season crop which grows in average temperatures of 20 to 40°C. Approximately, an area of 2.5 million hectares in world has been used for its cultivation from which 0.8 million tons of seeds are produced per annum. Mungbean seed is rich in proteins and its starch content is very important for nutritional uses.

Increasing the yield in the unit of surface is one of the most important issues that have attracted many researchers' attention. Photosynthesis is the substantial

process for the production of organic matter in plants. Usually, the amount of the produced dry matter has a direct relationship with the photosynthesis efficiency of the plant and also the way in which CO_2 fixation occurs in crops. Therefore, the acceleration of the photosynthesis rate could be useful for increasing the capacity of producing crops. Today, in order to achieve this goal, compounds such as methanol, ethanol, propanol, butanol and amino acids like glycine, glutamate and aspartate are used. One of the main advantages of these compounds is their preventing and reducing the effects of stresses induced, this is due to their photorespiration which ultimately results in increasing the production of organic matter in a plant along with increasing its growth and yield (Safarzadeh Vishekaei, 2007).

Non-electrolyte organic matters, that is, soluble sugars without electric charge such as alcohol, aldehydes and sugars are found as non-ionized water soluble forms.

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Table 1. Results of soil test.

Sand (%)	Silt (%)	Clay (%)	Organic material (%)	Nitrogen (%)	Absorbable potassium (ppm)	Absorbable phosphorus (ppm)	pH	EC (dS/m)
69	13	18	0.6	0.02	81	5	6.37	0.5

Moreover, due to their solubility in membrane-constituting substances, they can easily penetrate protoplasmic membranes. Through solving lipids, which exist in these membranes, they can pass them and enter the cell protoplasm. Methanol is 30 times more soluble in lipids than urea and it enters plant cells 300 times faster than the latter (Safarzadeh Vishekaei, 2007). Therefore, in an inactive way and through a simple emission from the membrane, methanol, ethanol and other alcohols are absorbed by plant cells (Murali et al., 1994). As a whole, compared with CO₂, methanol is a smaller molecule which can be used by C₃ plants for increasing photosynthesis (Li et al., 1995; Kotzabasis et al., 1999). Methanol spraying can delay the senescing of leaves through affecting ethylene-production stimuli in a plant, which increases the active photosynthetic period and the durability of the leaf surface (Heins, 1980).

Usually, increased methanol concentration in plant tissues regulates the metabolic rate of their activities (Hemming and Criddle, 1995; Ramberg et al., 2002; Downie et al., 2004) and affects the efficiency of carbon conversion and carbon conversion-related metabolic paths. Under low-concentration consecutive methanol application conditions, at first, the metabolic respiration is increased and later, it would decrease. Therefore, the effect of the methanol applied on plants is a function of time and spraying intensity (Hemming and Criddle, 1995; Gout et al., 2000).

In plants which suffer a shortage of water, methanol spraying of the aerial parts increases the chlorophyll concentration; while in plants that have enough water, this practice causes the concentration to have a slight reduction (Ramberg et al., 2002). Thus, methanol spraying results in increased production and reduces plants' water requirement in warm and dry conditions (Nemecek-Marshall et al., 1995; Fall and Benson, 1996). Consumption of methanol by some of the C₃ species during their growth period, where photorespiration is at a high level, increases plant turgescence. Usually, plants treated with methanol spraying are more tolerant of wilting (Nonomura and Benson, 1992). Metabolism of methanol and its conversion to sugars change the osmotic potential of the leaves. In addition, it increases the turgor pressure and the pores as well. In fact, keeping the pores open causes assimilation and the growth rate of a plant to increase which in itself would lead to early maturity and less water requirement (Nonomura and Benson, 1992; Makhdom et al., 2002).

Bhattacharya et al. (1985) studied the effects of ethanol, methanol and acetone on mungbean and found

that they increased the yield, accelerated the maturity and reduced the drought stress and the plant's water requirement. In the results of Li et al. (1995), it was reported that methanol had a positive effect of the seed yield, seed weight and the number of pods per plant in soy. By studying the role of methanol in stimulating growth, Ramberg et al. (2002) concluded that in plants which had water shortage, it increased their biomass, while methanol treatment in crops with sufficient water reduced their biomass. Results of a research conducted by Safarzadeh Vishekaei (2007) showed that spraying the aerial parts of peanut with a 20% methanol solution increased the leaf area index, crop growth rate, leaf area duration, pod growth rate, radiation use efficiency, pod and seed yields, 100-seeds weight, number of mature pods and the protein content of a peanut seed. Makhdom et al. (2002) examined the effect of spraying methanol on cotton and found that consuming methanol increases the dry matter, photosynthesis, pore conductivity and seed yield. Furthermore, it reduces cotton's water requirement and increases the leaf surface. The plant height and its growth increased about 50%. Thus, the present research was also conducted with the purpose of studying the possibility of using methanol foliar spraying to increase the growth and yield of mungbean in Rasht city in the north of Iran.

MATERIALS AND METHODS

In order to study the effect of the time and concentration of methanol foliar spraying on the growth and yield of mungbean in the 2009 crop year, an experiment was conducted in the Faculty of Agriculture of the Islamic Azad University, Rasht branch located 15 km Rasht city in the north of Iran (situated at 37°15'N and 49°53'E). Based on the Koppen classification, this region has a very humid climate with warm summers. Average annual precipitation level of the region is approximately 1250 mm and reaches about 430 mm during the cultivation season. Soil test results revealed that the soil texture is of the sandy-loam type (18% clay, 69% sand and 13% silt) with the pH and EC values being 6.37 and 0.5 dS/m, respectively. In addition to what we earlier-mentioned, the organic matters, absorbable potassium, absorbable phosphorus and nitrogen were 0.6%, 81ppm, 5 ppm and 0.02% in soil, respectively (Table 1).

This research was done as a two-factor factorial experiment in a basic plan of randomized complete blocks in three replications. The first factor was the time of methanol application in three levels [spraying in the morning (8:00 to 10:00 a.m.), at noon (13:00 to 15:00 p.m.) and in the evening (17:00 to 19:00 p.m.) and the second factor, that is methanol use was considered at four levels (0, 10, 20 and 30% methanol). To each one of these methanol application practices, 2 g/l glycine and 1 g/l tetrahydrofolate were added as catalysts. Also, to improve and increase the viscosity of

methanol solution, 1 g/l between 80 was used. The experiment was conducted in 4×2 m plots, each of which having four cultivation rows. Also, distances between plots of each replication and between replications were 0.5 m and about 1 m, respectively. The distance between rows was 50 cm, while the distance between every two plants on the rows was 20 cm. Mungbean seeds used in this experiment were of the indigenous type. Plot irrigation was done every six days and weeding was carried out both mechanically and manually. Methanol spraying was done twice during the growing season with 10-day intervals. The first spraying of the plants was done during early pod formation and continued until solution drops flowed on the plants.

The studied characteristics were pod yield in the unit of surface, seed yield in the unit of surface, number of pods per m^2 , 100-seeds weight, number of seeds per pod, harvest index (HI), pod length and plant height at the time of harvesting. To calculate the 100-seeds weight, seeds from mature pods of each plot were weighed by a scale. Also, ten pods were randomly selected from each plot and their seeds were counted. The average of ten counts gave the number of seeds per pod. Then, ten pods were randomly selected from each plot with the length of each pod measured. The average of ten obtained lengths gave the pods' length at the time of harvest. In addition, ten plants were randomly selected from each plot with the height of each plant measured and when these ten heights were averaged, the plants' height at the time of harvest was obtained. When the plants reached the harvest stage, five plants from each plot were selected with their biological yield and economic yield (pods) parts separately dried in the oven and then weighed. Weights obtained from each plant were put in the following formula and thus, the harvest index (HI) was calculated. In order to do the analysis of variance and mean comparison was used from SPSS 16.

$$\text{Harvest Index} = \frac{\text{Economic Yield (Pods)}}{\text{Biological Yield (Total)}} \times 100$$

RESULTS AND DISCUSSION

Pod length

The effect of methanol concentration and its spraying time on the pod length of mungbean was not significant (Table 2). With consideration of the fact that this trait is more genetically affected and less influenced by the environment, this result seemed logical.

Plant height

The effect of methanol concentration and spraying on mungbean height was not significant (Table 2). Since the first spraying was done during early pod formation stages and because mungbean has a limited growth, more growth could not be expected. So this finding contradicts those obtained by Mirakhori et al. (2009) in their study on soybean. However, it should be noted that methanol spraying in that experiment was done before soybean's pod formation stage.

Number of pods per m^2

The effect of the time and concentration of methanol

spraying on the number of pods was not significant (Table 2), which was consistent with the results of Murali et al. (1994) and Sunderman and Sweeney (1997). However, in the study of Li et al. (1995), methanol spraying had a positive effect on the number of pods in peanut and soybean.

Number of seeds per pod

In this study, the effect of methanol foliar spraying on the number of seeds per pod was significant at the probability level of 1% (Table 2) with the largest ($M = 13.11$) and least number of seeds per pod being those of 30% methanol and the control treatments (Table 3). Therefore, the results of the present research contradicted those of Sunderman and Sweeney (1997). The spraying time and the interaction between time and spraying concentration did not have a significant effect on the number of seeds (Table 2).

100-seeds weight

In this research, the time and concentration of spraying methanol on the leaves did not have any significant effect on the 100-seeds weight of mungbean (Table 2) which was consistent with the results of Rajala et al. (1998) who studied the effect of methanol on spring cereals, peas and summer forage rape seed and also those of Sunderman and Sweeney (1997) who studied this effect on soybean. Furthermore, the 100-seeds weight is a genetic trait and is less affected in different experiments. Moreover, Mirakhori et al. (2009) and Li et al. (1995) concluded that methanol spraying had a positive effect on the 100-seeds weight of soybean.

Pod yield per m^2

In the present research, the effect of methanol spraying time and concentration was not significant on mungbean's pod yield (Table 2), which was consistent with the results of these studies (Murali et al., 1994; Sunderman and Sweeney, 1997; Rajala et al., 1998). However, Mirakhori et al. (2009) and Safarzadeh Vishekaei (2007) reported that methanol increases the pod yield in peanut and soybean.

Seed yield per m^2

The effect of the time and concentration of methanol spraying on the seed yield was significant at the probability level of 5% (Table 2). Results from mean comparisons showed that the highest seed yield ($M = 55.97 \text{ gr/m}^2$) was that of the 20% methanol concentration. Also, the highest seeds yield ($M = 55.52 \text{ gr/m}^2$) was that

Table 2. Analysis of variance for the effects of concentration and time of methanol application on growth and yield of mungbean (*Vigna radiata* L.).

Source of variation	df	Mean squares							
		Pod length	Plant height	100-seeds weight	Pod yield per m ²	Number of seeds per pod	Number of pods per m ²	Seed yield per m ²	Harvest index
Block	2	1.7*	1482.58* *	1.27 ^{ns}	2138.36**	0.69 ^{ns}	4911.02**	643.48**	9.49
Concentration of methanol	3	0.19 ^{ns}	124.69 ^{ns}	0.58 ^{ns}	219.71 ^{ns}	1.31**	399 ^{ns}	115.67*	225.48**
Time of application	2	0.46 ^{ns}	80.33 ^{ns}	1.39 ^{ns}	62.7 ^{ns}	0.06 ^{ns}	199.69 ^{ns}	105.77*	159.44*
Concentration × Time	6	0.34 ^{ns}	84.22 ^{ns}	0.96 ^{ns}	97.48 ^{ns}	0.19 ^{ns}	234.58 ^{ns}	9.59 ^{ns}	21.33 ^{ns}
Error	22	0.29	56.88	0.44	105.92	0.21	150.3	25.67	28.46
C.V (%)		4.3	9.45	10.2	10.62	3.68	4.84	9.66	16.3

^{ns}Non significant, *significant at P<0.05 and **significant at P<0.01.

Table 3. Mean comparison effect of methanol on growth and yield of mungbean (*Vigna radiata* L.) by Duncan's multiple range test (DMRT).

Treatments	Pod length (cm)	Plant height (cm)	100-seeds weight (g)	Pod yield (g/m ²)	Number of seed per pod	Number of pod per m ²	Seed yield (g/m ²)	Harvest index (%)
Concentration of methanol								
M ₁ = 0	12.48a	75.22b	6.66a	92.23b	12.28c	132.78b	48.55c	26.31c
M ₂ = 10%	12.80a	79.55ab	6.26a	93.66ab	12.45bc	133.33b	50.28bc	31.86b
M ₃ = 20%	12.78.a	79.88ab	6.48a	103.16a	12.90ab	146.33a	55.97a	34.48ab
M ₄ = 30%	12.69a	84.33a	6.86a	98.25ab	13.11a	142.00ab	54.96ab	38.22a
Time of application								
T ₁ = Morning (8-10 am)	12.69a	82.41a	6.68ab	94.25a	12.70a	134.08a	49.60b	29.52b
T ₂ = Midday (1-3 pm)	12.88a	79.58a	6.18b	97.61a	12.60a	139.75a	52.20ab	31.94b
T ₃ = Afternoon (5-7 pm)	12.49a	77.25a	6.83a	98.61a	12.75a	142.00a	55.52a	36.69a
Interaction								
M ₁ T ₁	12.35b	80.00bc	7.41ab	83.46b	12.33b	117.33b	44.00d	23.40d
M ₂ T ₁	12.60ab	85.00ab	6.71bc	88.23ab	12.60ab	127.00ab	48.51cd	30.89abcd
M ₃ T ₁	13.11ab	80.33bc	6.13bc	102.15ab	13.00ab	145.67a	52.80abcd	29.54bcd
M ₄ T ₁	12.70ab	84.33ab	6.48bc	103.17ab	12.90ab	146.33a	53.08abcd	34.27abc
M ₁ T ₂	12.74ab	79.33bc	5.69c	94.93ab	12.33b	138.00ab	49.58bcd	24.06d
M ₂ T ₂	13.48a	80.00bc	5.91c	99.53ab	12.10b	142.67a	51.19abcd	27.70cd
M ₃ T ₂	12.61ab	79.00bc	6.61bc	102.63ab	13.00ab	142.33a	55.37abc	35.57abc
M ₄ T ₂	12.71ab	80.00bc	6.51bc	93.36ab	13.00ab	136.00ab	52.67abcd	40.45a
M ₁ T ₃	12.36b	66.33c	6.87bc	98.30ab	12.20b	143.00a	52.09abcd	31.49abcd

Table 3. Contd.

M ₂ T ₃	12.32b	73.66bc	6.16bc	93.23ab	12.66ab	130.33ab	51.13abcd	37.00abc
M ₃ T ₃	12.62ab	80.33bc	6.70bc	104.70a	12.70ab	151.00a	59.73a	38.32ab
M ₄ T ₃	12.66ab	88.66a	7.58a	98.23ab	13.43a	143.67a	59.13ab	39.95a

In each column, means with the similar letters are not significantly different at 5% level of probability using DMRT.

of the evening spraying (Table 3). Results from the present research in terms of the effect of methanol on the seed yield were consistent with those of these studies (Mirakhori et al., 2009; Li et al., 1995; Safarzadeh Vishekai, 2007), while contradictory results was obtained compared with those of this research by these studies (Rajala et al., 1998; Sunderman and Sweeney, 1997; Murali et al., 1994).

Harvest index

The effect of the time and concentration of methanol spraying on the harvest index was significant at probability levels of 5 and 1%, respectively (Table 2). It seemed that methanol mostly affected the allocation of most of the dry matter to the reproductive organs which of course requires more research. The harvest index gave the best results for 30% methanol consumption (M = 38.22%). Also, evening spraying resulted in the highest harvest index (M = 36.69%) (Table 3). Interaction between the concentration, time and the harvest index of mungbean was not significant (Table 2). Mirakhori et al. (2009) and Rajala et al. (1998) stated that methanol did not have any effect on the harvest index.

According to Murali et al. (1994) differences between the results of various researches might be due to different methanol concentrations and the experiment environment's climatic conditions. Therefore, the suitable concentration for spraying the studied plant should be taken into account. Also, the growth stage in which spraying is done is of great significance. As Nonomura and Benson (1992) concluded in their study that methanol mostly improves the growth of plants in dry conditions and is actually a factor for reducing drought stress, perhaps it is among the reasons for methanol's not being effective in some of the climatic conditions of Guilan province in the north of Iran, which is to some extent far from dry conditions and stress and this issue can show the importance of the climatic conditions of the experiment's environment quite well.

Conclusion

As a whole, obtained results showed that spraying methanol significantly affected the number of seeds per pod, seed yield and the harvest index. The largest

number of seeds was produced by 30% methanol concentration, while the highest seed yield was achieved by 20% methanol treatment and evening spraying. The highest harvest index was observed in 30% methanol concentration and evening spraying. Methanol spraying did not affect other traits, which seemed to be due to the fact that the studied traits were more affected by genetics than by the experiment's environment. Among these traits, 100-seeds weight and the pod length at the harvest can be mentioned. As discussed previously in terms of plant height at the time of harvesting, since mungbean has a limited growth, it cannot be expected to grow again after the reproduction stage.

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