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Effects of Hydrangea (*Hydrangea macrophylla* L.) Leaf Extract Foliar Application on Growth and Yield of Mungbean (*Vigna radiata* L.)

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Abstract
Foliar fertilization is way of supplying supplementary doses of minor and major nutrients, plant hormones, and other beneficial substances to plants. A pot experiment was conducted at the Crop Farm, Eastern University Sri Lanka to identify the effects of folair application of *Hydrangea macrophylla* L. leaf extract (HmLE) on the growth and yield of mungbean (*Vigna radiata* L.) var. 'MI 5'. The experiment was arranged in Completely Randomized Design with 5 treatments and 4 replications. Once a week HmLE was applied at four different concentrations (T2: 10%, T3: 20%, T4: 50% and T5: 100%) and distilled water was applied as control (T1). The crop performance was recorded from two weeks after planting (WAP) to eight WAP. Analysis of variance was performed to compare effectiveness of treatments. Foliar application of HmLE had significant ($P<0.05\%$) effects on measured parameters of *Vigna radiata* over the control. Application of 20 % HmLE increased plant height (32.81%), root length (21.78%), leaf chlorophyll content (48.27%), leaf area (113%), Number of flowers per plant (50%), number of pods per plant (85.71%), Dry weight of shoot (132.93%), Number of effective nodules (224.32%) and Inter node length (72.59%) over the control. This could be due to availability of micro, macro and gibberellins in the HmLE. From the findings it could be concluded that, application of 20% HmLE could be recommended for mungbean (*Vigna radiata* L.) cultivation as an environment friendly practice to improve growth and yield.

Keywords: Foliar application, *Hydrangea macrophylla* L., Leaf extract, *Vigna radiate* L

1. Introduction
Increasing world population and their consumption requirements are placing extraordinary demands on agriculture. Several methods are practiced to increase food production. However around a billion of people are suffering from malnutrition and health problems because of modern agriculture practices. These practices tremendously degrading soil and aerial environment. Agricultural system should be improved by reducing soil degradation and protection of biodiversity. Therefore, use of natural substances to enhance crop productivity and soil fertility would reduce the usage of synthetic fertilizers and environmental pollution.

Hydrangea macrophylla L. is a flowering plant native to Japan. It belongs to family Hydrangeacea. Leaf extract of Hydrangea macrophylla has a natural substance which is called as Hydrangenol. It can be induced the gibberellin activity of plants [1]. Furthermore, the leaf extract of Hydrangea macrophylla has antifungal ability [2]. Hydrangea macrophylla liquid extract (HmLE) has several macro and micro nutrients. It could also be used to prepare the natural herbicides as well.

Mungbean (Vigna radiata (L.) Wilczek) is a popular crop in Sri Lanka and India. It's a major source of protein in human diet. It has ability to fix atmospheric nitrogen as well. Mungbean is a hardy, warm season annual crop. Mungbean has high source of nutrient including, Carbohydrate, Mn, K, Mg, folate, Cu, Zn, and various B vitamins. It is rich in digestible protein (approximately 25 - 28%). It is widely grown in tropical and sub tropical Asia because of its wider range of adaptability and drought tolerance feature. Mungbean is a cheap source of dietary protein with high levels of folate and iron compared to many other legumes [3].

Mungbean seeds are more palatable, nutritive, digestible and non-flatulent than other pulses grown in country. It contains 24.7% protein, 0.6% fat, 0.9% fiber and 3.7% ash (Ali *et al.*, 2010). relatively more essential amino acids such as Lysine and Tryptophan and thus, compliment the amino acids supplied by cereals and also they contain relatively small proportions of Sulphur containing amino acids such as Methionine and Cysteine and, also a useful source of Thiamine, Niacin and Calcium [3].

Some studies reported that folair application of nutrients would improve the growth and yield of mungbean. Hence HmLE could also be used as a foliar spray to increase the growth and yield of mungbean. It is an environmentally friendly option to increase the productivity of mungean. However optimum concentration of HmLE application for mungbean has not been identified yet. Hence this experiment was conducted with the objective of assessing the effects of different concentrations of Hydrangea macrophylla L. leaf extract (HmLE) on growth and yield of mungbean (Vigna radiata L.).

2. Materials and methods
A pot experiment was conducted in the Crop Farm, Eastern University Sri Lanka (latitude of 70043'N and the longitude of 810 42 E at an elevation of 7.8m above mean sea level) to find out the effects of Hydrangea macrophylla leaf extract (HmLE) application on growth and yield of Vigna radiate L. variety 'MI 5'. The experiment was arranged in Completely Randomized Design with 5 treatments and 4 replications. Once a week HmLE was applied at different concentrations

(T2: 10%, T3: 20%, T4: 50% and T5: 100%) and distilled water was applied as control (T1). The experiment was carried out from June to September. The experiment was arranged in Completely Randomized Design with 5 treatments and 4 replications.

2.1 Preparation of liquid extract of Hydrangea macrophylla

Hydrangea macrophylla leaves were collected and shade dried for 3-4 days. Then it was grinded by using laboratory grinder. After that powder was taken and sieved. Then 5g of powder was dissolved in 100ml distilled water. Then solution was autoclave at 121˚C, 15Ibs/sq inch for 20 minutes. Thereafter, liquid extract was filtered by using double layered cheese cloth. It was allowed to cool at 4˚C. Then liquid extract was centrifuged at 5000 rpm for 15 minutes. This supernatant was called as 100% Hydrangea leaf extract [4].

2.2 Preparation of Pot

The pots were prepared by using PVC pipes. The pots were cut in longitudinally and fixed again by bonding agents for the purpose of taking the sample for analyzing without any damage to root system. Bottom of pots were covered by filter paper with l mm pores size nylon net. Volume of PVC pipe was 5.2cm inner diameter and 30cm height. Each pot was filled with soil mixture of topsoil, red soil and compost at the rate of 1:1:1. and Department of Agriculture (Sri Lanka) recommended basal fertilizer. Mungbean plants were planted in each pots (Fig.1). Once a week HmLE was applied at different concentrations (T2: 10%, T3: 20%, T4: 50% and T5: 100%) and distilled water was applied as control (T1) as per treatment structure. All the other management practices were followed uniformly for all treatments. Their performances of plants were recorded from two weeks after planting (WAP) up to eight WAP.

Fig 1: Experimental arrangement

2.3 Physico-chemical analyses of Hydrangea Leaf extract

Physio-chemical properties of HmLE were analyzed at Laboratory, CIC Agribusiness center, Pelawehera, Sri Lanka.

2.4 Measurements

Vegetative growth parameters viz. plant height, leaf area, chlorophyll content (SPAD), internode length, number of flowers per plant and root length were measure thirty days after planting. Yield parameters viz. number of pods/plant, number of seeds/pod, 100 seeds weight and plant biomass were measured at the time of harvesting.

2.5 Statistical analysis

Analysis of variance (ANOVA) was performed by using StatistiXL 2.0 (statistiXL Nedlands, Western Australia). Mean comparison was performed within treatments using Tukey's Studentized Test at 5% significant level for precise and easy interpretation of results of this experiment.

3. Results and discussion

3.1Plant height (cm)

Maximum average plant height was recorded in plants at T3(20% HmLE) throughout the experiment while the minimum was recorded in T1 (control). It clearly indicated that application of 20% concentration of HmLE had significantly ($p<0.05$) increased average plant height by (32.81%) compared to control. Khosa *et al.* [5] reported that presence of macro nutrient in foliar sprays increase plant height. Increase in plant could also be due to presence of nitrogen (N) and potassium in HmLE. Srikrishnah *et al.* [6] opined that, nitrogen has the potential to increase plant height of Dracaena varieties at optimum level. Potacium could also influence plant height. This statement was in agreement with Dkhil *et al.* [7]. Kazemi [8] reported that application of Zn and Fe containing foliar spray showed significant effect on vegetative growth of plants. Therefore, it could be stated that, N, Zn and Fe in the foliar spary of HmLE increased the plant height of mungbean. IAA might be presence in HmLE. It may also cause increase in plant height. IAA has the potential to increase plant height [9] [10].

3.2 Chlorophyll content (SPAD)

The maximum average chlorophyll content was recorded in Plants at T3(20% HmLE) while the minimum average plant chlorophyll content was recorded in T1 (control). There was a clear evidence that, application of 20% HmLE had significantly ($p<0.05$) increased average chlorophyll content by (48.27%) compared to control plant. The HmLE contains several macro and micro nutrients and it could act as a biostimulant as well. N and Mg are essential for chlorophyll synthesis. These are found in HmLE extract. It could be the reason for higher chlorophyll content of plants at T3. Khosa *et al.* [5] pointed out that micro and macro nutrient found in foliar extract, enhanced the chlorophyll content. Presence of Zn and Fe also increased plant chlorophyll content. The results were in agreement with Arif *et al.* [11].

3.3 Leaf area (cm2)

The present experiment revealed that foliar application of HmLE with 20% concentration was increased average leaf area per plant (113%) compared with T1 (control). Maximum average leaf area per plant was recorded in T3 (20%) while minimum leaf area per plant was shown in T1 (control). It was clearly indicated that presence of micro and macronutrient in foliar extracts, increased the leaf area per plant [5]. Results were in agreement with Nadeem *et al.* [12] who observed significantly higher leaf area by the

application of micronutrients to wheat Further, it was reported that Zn and Fe were involved to increase leaf area of plants [7].

3.4 Shoot biomass (g)

Highest shoot dry weight was recorded at T3 (132.93%) comparison with T1 (control) plant. It clearly showed that foliar application of HMLE on Vigna radiata was increased the dry matter accumulation of shoots. Srikrishnah *et al*. [13] observed that, biomass production were in accordance with the trend of variances for LA in Dracaena. Sutharsan *et al*. [4] identified that an increase in leaf area lead to an increase in dry matter accumulation of crops. This might be due to the micro and macronutrients Further, Asad and Rafique [14], stated that application of micro nutrients increased wheat dry matter content. Application of Fe resulted in better dry matter content of soybean [15].

3.5 Root biomass (g)

The maximum average root dry weight was recorded in T3 (20%) followed by T4, T5, and T2 while the minimum average of root dry weight was recorded in T1 (control). The root image was shown in Fig. 2. It could be due to presence of higher amount of phosphorus in HmLE which promote the root development. Root growth could be influenced by gibberellins activity inducing effect of Hydrangea leaf extract. These results are in conformity with the report of Asen *et al*. [1]. Gibberellin activity increased the root growth [16]. It also might be affected to increased dry matter content of Vigna radiata. It was reported that foliar application with micro and macro nutrients such as Zn and Fe significantly influenced dry weight of plant [8].

3.6 Number of flowers per plant

Plants grown at T3 (20% Hydrangea leaf extract) produced highest number of flowers while lowest number of flowers per plant was recorded in T1 (control). It showed that foliar application of HmLE has the potential to increase the number of flowers in mungbean.50% increment in number of flower was observed at T3 in comparison to control plants (T1). The presence of higher amount of Potassium stimulated flower initiation and also gibberellins activity inducing ability was increased the flower initiation. These results are in agreement with Asen *et al*. + and Bottini *et al*. [16]. Presence of Zn, Cu, Mn and Fe also might be affected to increase number of flowers. The results were in agreement with the findings of Kazemi [8] and Dkhil *et al*. [7]

3.7 Number of pods per plant

The maximum average pods per plant was recorded in T3 (20%) HmLE while minimum number of pods per plant was recorded in T1 (control). Application of 20% Hydrangea leaf extract had significantly ($p<0.05$) increased average number of pods per plant by (85.71%) comparison with T1 (control) plants. These results confirm with the results of Asen *et al*., [1]. It showed that, the gibberellins inducing ability increased the number of pods per plant and also these results are confirmed with the results of Khosa *et al*. [5]. It was confirmed that presence of micro and macro nutrient such as Zn, Cu, Mn and Fe was affected to influencing number of pods. Ample supply of N and P through foliar sprays play vital role in for pollen viability because of increase fertilization. Therefore, increased pod setting and number of pods [17].

3.8 Internode length (cm)

Foliar application of HmLE with lower concentration (20%) on Vigna radiata was increased average internode length. The maximum average internode length was recorded in T3 (20%) Hydrangea leaf extract while minimum internode length was recorded in T1 (control). Inter node length was increased by (72.59%) in T3 (20%) comparison with T1 (control) plant. This was due to gibberellins ability inducing effect. These results are conformity with the report of Asen *et al*. [1]. Gibberellins has the ability to induce internode length of plants [16].

3.9 Number of effective nodules

Foliar application of HmLE with lower concentration (20%) on Vigna radiata was increased average number of effective nodules. The maximum average number of effective nodules was recorded in T3 (20%) Hydrangea leaf extract while minimum number of effective nodules were recorded in T1 (control). This might be due to presence of micro and macronutrient in Hydrangea leaf extract and gibberellins inducing ability [1]. Milev [18] reported that Phosphorus and potassium in foliar fertilizers have significant effect on the process of fixation of atmospheric nitrogen. Further, Ohara [19] opined that application of Fe enhanced number of nodules and Fe improved rhizobia nutrients, nodulation, nodule activity and biological nitrogen fixation.

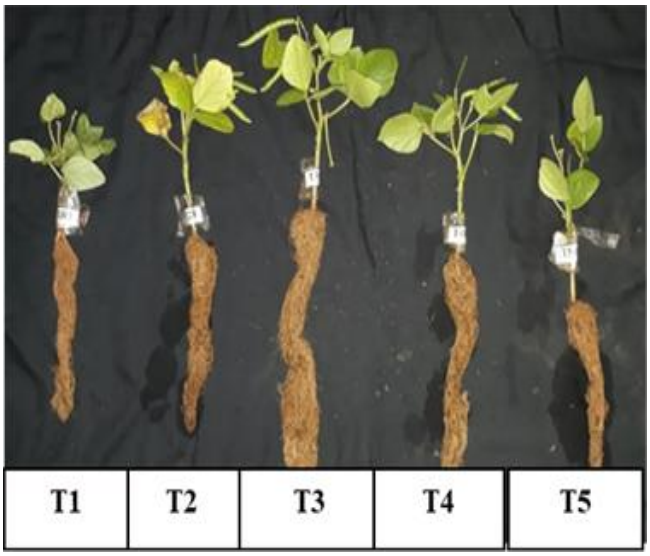

Fig 2: Plants belong to different treatments at maturity

3.10 100 seeds weight (g)

There was a significant differences ($p<0.05$) between 100 seeds weight with T1 and T2, T3, T4, T5. The maximum average 100 seeds weight per plant was recorded in T3 (20%) HmL E while minimum 100 seeds weight per plant was recorded in T1 (control). This might be due to the availability of micronutrient such as zinc and manganese in HmLE. Sultana *et al*. [20] reported that availability of Zinc and manganese in foliar application increased 100 seeds weight. Further, Dkhil *et al*. [7] reported that Zn, Cu and Mn increased weight of seeds.

Overall, all the results are consistency to those of Asen *et al*. [1] who determined that gibberellins activity inducing ability of Hydrangea macrophylla leaf extract was increased the number of pods per plat, number of flowers per plant, root

growth and also internode length. These results are in agreement with the report of Bottini *et al*., [16] who reported that Gibberellin induced all above parameters. Presence of micro and macro nutrients of HmLE induced vegetative and reproductive growth parameters of *Vigna radiata*.

Table 1: Effects of Hydrangea macrophylla L. leaf extract on vegetative performances of mungbean (Vigna radiate L.) Var.MI 5.

Treatments	Plant height(cm)	Root length (cm)	Chlorophyll content (SPAD	Leaf Area (cm^2)	Internode length (cm)	No of flowers/ plant
T1(Control)	19.2±0.7^{c}	27.7±0.70^{c}	29.0±1.8^{c}	88.806±2.479^{b}	1.463±0.171^{b}	12 ± 0.6^{b}
T2:10%	20.6±0.7^{c}	28.0±1.06^{bc}	37.8±1.3^{b}	104.969±5.96^{b}	1.513±0.151^{b}	12 ± 0.5^{b}
T3:20%	25.5±0.8^{a}	33.8±0.48^{a}	43.0±0.9^{a}	189.754±2.71^{a}	2.525±0.088^{a}	18 ± 1.1 ^{a}
T4:50%	23.4±0.7^{b}	31.3±0.43^{ab}	37.2±1.4^{b}	96.170±4.429^{b}	1.688±0.172^{b}	15 ± 1.06^{a}
T5:100%	22.5±0.5^{b}	29.2±1.22b^{c}	34.5±1.0^{b}	93.901±3.827^{b}	1.688±0.116^{b}	15 ± 1.0 ^{a}
F	*	*	*	*	*	*

Value represents mean± standard error of four replicates. Mean values in a column having the dissimilar letter/letters indicate significant differences at 5% level of significance (Tukey's Studentized Test). *=Significant at 5% probability level.

Table 2: Effects of *Hydrangea macrophylla* L. leaf extract on reproductive performances of mungbean (*Vigna radiata* L.) Var. MI 5.

Treatment	No of pod/plant	Number of seeds/pod	100 seeds weight (g)	No of effective nodules	Shoot biomass (g)	Root biomass (g)
T1:Control	7.0±0.2^{c}	5.75±0.16^{d}	5.250±0.171^{c}	36.750±3.599^{d}	0.498±0.095^{b}	0.488±0.088^{c}
T2:10%	7.3±0.1^{c}	7.00±0.37^{c}	5.450±0.176b^{c}	59.875±4.919^{bc}	0.620±0.070^{b}	0.761±0.142^{bc}
T3:20%	13.5±0.5^{b}	10.00±0.2^{a}	8.425±0.628^{a}	120.000±7.533^{a}	1.160±0.123^{a}	1.978±0.267^{a}
T4:50%	9.2±0.3^{b}	8.75±0.16^{b}	6.975±0.232^{b}	79.750±5.411^{b}	0.804±0.099a^{b}	1.169±0.063^{b}
T5:100%	8.0±0.0^{bc}	8.12±0.12^{b}	6.300±0.208^{bc}	52.750±4.386^{cd}	0.688±0.079^{b}	0.991±0.080^{bc}
F	*	*	*	*	*	*

Value represents mean± standard error of four replicates. Mean values in a column having the dissimilar letter/letters indicate significant differences at 5% level of significance (Tukey's Studentized Test). *=Significant at 5% probability level.

4. Conclusion

In this experiment, plants grown at T3 (application of 20% HmLE at once a week) showed significantly better performances in growth and yield parameters. It might be due to gibberellin activity inducing ability and presence of micro and macronutrient presentin Hydrangea leaf extract. Further HmLE could also act as biostimulant to increase growth and yield of mungbean plants. From the findings it could be concluded that, application of 20% HmLE could be recommended for mungbean (*Vigna radiata* L.) cultivation as an environment friendly option to improve growth and yield.

Table 3: Chemical properties of Hydrangea macrophylla liquid extract

Variables	Unit	Value
Colour	-	Green
PH	-	4.98
EC	uS/cm	4070
TDS	g/l	2.11
Salinity	ppt	2.1
Nitrogen	ppm	400
Phosphorous	ppm	69
Potassium	ppm	982
Magnisium	ppm	148
Calcium	ppm	136
Iron	ppm	2.1
Manganese	ppm	2.7
Zinc	ppm	1.0
Copper	ppm	10.0

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